THE EFFECTS OF DENTAL IMPAIRMENT ON THE BIOLOGY AND BEHAVIORAL ECOLOGY OF WILD RING-TAILED LEMURS (*LEMUR CATTA*) AT THE BEZÀ MAHAFALY SPECIAL RESERVE, MADAGASCAR.

by

JAMES BURNS MILLETTE III

B.S., Emory University, 2003

M.A., University of Colorado Boulder, 2007

A thesis submitted to the

Faculty of the Graduate School of the

University of Colorado in partial fulfillment

of the requirement for the degree of

Doctor of Philosophy

Department of Anthropology

2016

This thesis entitled:
The Effects of Dental Impairment on the Biology and Behavioral Ecology of Wild Ring-tailed Lemurs (*Lemur catta*) at the Bezà Mahafaly Special Reserve, Madagascar
written by James Burns Millette III
has been approved for the Department of Anthropology

_____
Dr. Michelle L. Sauther

_____
Dr. Matt Sponheimer

_____
Dr. Gerardo Gutiérrez

_____
Dr. Joanna Lambert

_____
Dr. Frank P. Cuozzo

Date _____

The final copy of this thesis has been examined by the signatories, and we find that both the content and the form meet acceptable presentation standards of scholarly work in the above mentioned discipline.

IACUC protocol #: 1102.08

ABSTRACT:

Millette III, James Burns (Ph.D. Anthropology)

The Effects of Dental Impairment on the Biology and Behavioral Ecology of Wild Ring-tailed Lemurs (*Lemur catta*) at the Bezà Mahafaly Special Reserve, Madagascar.

Thesis directed by Professor Michelle L. Sauther

Ring-tailed lemurs (*Lemur catta*) of the Bezà Mahafaly Special Reserve (BMSR) demonstrate exceptionally high rates of dental wear and subsequent tooth loss due to tamarind fruit consumption. How such wear and loss affects their behavior, nutrition and health is not yet fully understood. This study examines how dental impairment impacts ring-tailed lemur biology and behavioral ecology through a broad dental-ecological framework involving use of topographic analyses of tooth form, food processing data collection, and fecal measures of nutritional status. This study also presents visual measures of coat and body condition applicable to dental studies. Increasing topographically-measured dental impairment was associated with longer tamarind fruit feeding times and increased use of alternative tamarind fruit processing techniques (e.g., use of open fruit, manual processing and toothrow use). Increasing impairment was also associated with longer bouts for variety of other food types and species, although several foods were associated with shorter bouts, suggesting that food mechanical properties play a role in the expression of behaviors relative to dental impairment. Individuals with tooth loss and wear demonstrate larger particles within their fecal material, suggesting they possess a reduced capacity to digest and absorb ingested foods. This hypothesis was supported by fecal-nutritional analyses indicating that impairment often reduces individual capacity to ferment structural carbohydrates and/or results in seasonally increased residual fecal protein. Dental

impairment was also associated with higher fecal ash content, suggesting that tooth wear at this site may also be a function of dietary silicates in addition to that caused by tamarind fruit feeding. Measures of coat and body condition indicate that lemurs show increased coat quality following the onset of the wet season when resource availability increases, and that body size appears to be related to reproductive status. As these are measures that appear to be linked to energetic status, their use may also be beneficial in future studies of dental impairment. Overall, data from this study indicate that dental impairment has real, significant and typically negative impacts on BMSR ring-tailed lemurs in terms of their feeding behaviors, ability to process foods and their capacity to utilize ingested resources.

DEDICATION:

This dissertation is dedicated to all those individuals who have supported and contributed to this project: friends, teachers, family and lemurs alike.

ACKNOWLEDGEMENTS:

Although only one name is listed on the title page of this dissertation, I am not the only individual who participated in this project on the impacts of dental impairment on ring-tailed lemur behavior and biology. From its onset, this project has involved collaboration with a multitude of individuals, each who have contributed significantly to its design, execution and completion. I therefore wish to acknowledge and thank all of those who have seen this project through from its inception to its final form in this dissertation.

I would first like to acknowledge the efforts of my graduate advisor and dissertation committee chair, Dr. Michelle Sauther. This work would not have been possible without her long-term research on ring-tailed lemurs at the Bezà Mahafaly Special Reserve (BMSR), and which has been conducted for over 25 years. Her work has not only contributed significantly to our understanding of ring-tailed lemurs, but has formed the framework on which this dissertation was designed and completed. Dr. Sauther has also been an excellent advisor during my time as a graduate student at the University of Colorado. She has not only assisted greatly in the development of this project in terms of its theoretical grounding, but has contributed to the securing of funding needed for this project and has guided this study's field and laboratory elements. She has also aided greatly in the interpretation of data generated by this study and has also provided valuable insight during the writing of this dissertation. In addition, Dr. Sauther has been an excellent mentor outside of this project, and has contributed significantly to my development as a scholar. She has not only included me within her own research, but has also

allowed me to contribute to development and execution of numerous projects and publications during my time as a graduate student.

I would also like to thank Dr. Frank Cuozzo for his assistance with my work at BMSR generally, and for his contributions to the development and execution of this project. The dental data on which this study was conducted relied heavily on both impressions and dental morphological data collected by Dr. Cuozzo at BMSR. Likewise, Dr. Cuozzo has assisted extensively with the design of this study, the securing of sufficient funding for this work and with securing access to laboratory resources necessary for the completion of dental topographic analysis. Dr. Cuozzo has also been an exceptional mentor and advisor during my graduate career. Despite being an "outside" committee member, he has participated and contributed greatly to every aspect of this dissertation project. He has also assisted in my development as a scholar during the course of my graduate career and with the publication of several documents resulting from my research during this time.

I am also extremely indebted to all individuals who assisted with the field element of this research project, conducted at BMSR from June 2012 to March 2013. I must first thank Professor Joelisoa Ratsirarson, Dr. Jeannin Ranoivanasy and Mrs. Mia Razafimahefa of ESSA-Forêts at the University of Antananarivo. Without their assistance securing permits, travel documentation and graduate student assistants, I would not have been able to conduct research in Madagascar generally, or at BMSR specifically. I also firmly appreciate the mentoring and historical knowledge about BMSR provided by Dr. Ratsirarson during his several visits to the site during my time spent there. I would also like to extend thanks to Dr. Ibrahim Antho Jacky Youssouf of the University of Toliara. Dr. Youssouf played an instrumental role in facilitating my research once I had arrived in Madagascar's southwest. He also played a major role in

arranging basic aspects of this project, such as transportation, lodging and facilitating the aid of student researchers. I also thank Dr. Youssouf for the opportunity to discuss my research with his undergraduate students at the University of Toliara. In addition to these senior researchers, I must also acknowledge the wonderful Malagasy student assistants who supported my work. These include Mr. Percy Yvon Rakoto of the University of Antananarivo and Mr. Naina Nicolas Rasolonjatovo of the University of Toliara. These individuals not only assisted with the collection of data necessary for the completion of this project, but were also instrumental in providing a link to the local community. Without their help, this project simply would not have been possible.

I also would like to extend my thanks to the members of Madagascar National Parks (MNP) who staff and maintain the Bezà Mahafaly Special Reserve. This includes the director of the reserve, Mr. Andry Serge Randrianandrasana and those working under his direction. As such, I would like to extend my thanks to Monja, Lala, Delaprairie, Vavy, Samsoa, and all those other individuals who worked at the reserve during my time there. It is only with their hard work that research is possible at BMSR, or that the reserve functions in the excellent manner that it does.

It is also important to acknowledge the assistance of the BMSR ecological monitoring team. I thank Efiteria, Edouard, Elahavelo and Enafa for their tireless efforts to assist with data collection. They spent countless hours collecting biological samples, census data and tracking lemur groups. I also thank Mr. Miandrisoa Razafindribe, the reserve's Chief of Research during my time there, for facilitating my work and arranging communications with ESSA-Forêts in Antananarivo. These individuals have also contributed greatly to the activities of the reserve overall and the maintenance of both animal and plant populations found there. The importance of their work cannot be overstated.

In addition to fieldwork, I also conducted a variety of laboratory analyses following my return from BMSR. I thank Dr. Peter Ungar for allowing my use of his scanning equipment at the Paleoanthropology Laboratory of University of Arkansas, Fayetteville. I also thank his students Carrie Healey and Sarah Livengood for their assistance in producing dental casts, conducting GIS topographic analysis and for scheduling access to the Paleoanthropology Laboratory. I also thank the members of Dr. Matt Sponheimer's nutritional laboratory. This is a research product, in whole or in part, of the Nutritional and Isotopic Ecology Lab (NIEL) at CU Boulder. In particular I thank Jennifer Leichliter and Oliver Paine for their assistance with training and their expertise using nutritional lab equipment. I am also very grateful for the help of Dr. Matt Sponheimer, who not only provided access to his lab, but who also provided excellent mentoring while conducting nutritional analyses. I also thank Ms. Hannah Padda for her assistance processing and assaying lemur nutritional samples. Without the help of these individuals, data which helps to better understand the impacts of dental wear on behavior and nutrition simply could not have been collected.

This project would not have been a success without sufficient financial support from a variety of sources. I am greatly indebted to the L.S.B. Leakey Foundation for their initial support of this project, and which allowed for this work to commence. I am also appreciative for the support of the National Science Foundation, which provided the majority of financial resources necessary for the field and laboratory elements of this project. Financial support for this project was also graciously provided by the University of Colorado, Department of Anthropology, as well as by Dr. Scott Ferris. I also would like to thank the CU Museum of Natural History for providing support from the William Burt Fund, as well as the CU Graduate School for providing

funding through the Beverly Sears Grant. These sources of funding were critical for supporting preliminary data collection which was carried out prior to this dissertation project.

I also extend thanks and gratitude to a number of individuals who played key individual roles with respect to this work's completion. First, I would like to recognize our veterinarian Dr. Cora Singleton, whose great knowledge of animal care allowed for the safe capture and release of all study subjects while at BMSR. I also thank Drs. James Loudon and Dana Whitelaw for initially introducing me to BMSR and the surrounding communities during the early portion of my research career there. I also appreciate support provided by Dr. Dennis Van Gerven and Dr. Bert Covert, particularly during the initial development of this project. I am also appreciative of Dr. Joanna Lambert who joined my dissertation committee with short notice and with great enthusiasm. I also would like to acknowledge the assistance of Dr. Heather Williams, who graciously read and provided feedback on endless drafts associated with this project. She also served as a valuable liaison to those in the USA during my fieldwork in Madagascar, and has supported my research tirelessly and without complaint. I can also not fail to mention her assistance with weighing and measuring of lemur fecal material at 2am. I also thank my family (Jim Millette, Susan Millette and Francis Jones) for their support during my time as a graduate student. I finally would like to acknowledge those other individuals who have assisted my development as an anthropologist and researcher in one way or another through the years. These include (but are not limited to), Dr. Jonathan O'Brien, Dr. Paul Sandberg, Dr. Darna Dufour, Dr. Gerardo Gutierrez, Dr. Krista Fish, Valerie McBride, Lesa Morris, Dr. Ben Freed, Dr. Agustin Fuentes, and Dr. Patricia Whitten.

Again, I thank everyone involved with this project for their efforts. This document, and the results presented within, would not have been possible without their assistance.

CONTENTS

LIST OF TABLES

Tables

LIST OF FIGURES

Figure

CHAPTER I**:**

BACKGROUND AND INTRODUCTION

The goal of this dissertation is to examine associations between dental impairment and the behavior, nutrition, food processing capacity, and health status of ring-tailed lemurs (*Lemur catta*) from the Bezà Mahafaly Special Reserve, Madagascar (BMSR) using a broad dental-ecological framework (see Cuozzo and Sauther, 2012). In doing so, I integrate a variety of behavioral, dietary, nutritional and health datasets with dental-morphological information to provide a broad framework with which to better understand the impacts of dental impairment among *L. catta*. The population of ring-tailed lemurs living at BMSR is furthermore an excellent study sample with which to understand these areas of interest for nonhuman primates broadly, as these animals demonstrate exceptionally high rates of significant tooth wear and subsequent wear-related tooth loss due to the use of tamarind fruit (*Tamarindus indica*), a commonly consumed food which is also a key fallback resource during the resource-depleted dry season (Cuozzo and Sauther, 2004, Cuozzo and Sauther, 2006a,b; Yamashita, 2008b; Sauther and Cuozzo, 2009; Yamashita et al., 2012, 2015a,b). Levels of tooth wear and loss at BMSR [up to 81% tooth loss in multiple individuals over a 10 year study period (e.g., Orange 170, Cuozzo and Sauther, 2004, 2006a; Black 226, this study)] reach those which are assumed to result in dental senescence (e.g., the functional loss of dental function; see Ungar, 2005; King et al., 2005) and which potentially reduces the individual's ability to process, consume and utilize food resources. Such an inability to process and utilize foods is likewise assumed to negatively impact individual

health, nutrition, reproduction and survival (see Gipps and Sanson, 1984; Lanyon and Sanson, 1986; Logan and Sanson, 2003a,b,c; Lucas, 2004; King et al., 2005, etc.). The effects of significant dental impairment have furthermore been interpreted to result the death of the individual. For example, Lucas (2004) notes that wild shrews often starve to death once their teeth have been worn to the gumline, while elephants have been observed to starve once a loss of occlusal contact between opposing positions has been achieved.

It has, however, been observed that BMSR *L. catta* survive and reproduce in comparatively good health over a period of at least several years despite possessing exceptionally high levels of tooth wear and loss, potentially through compensatory alterations to behavioral patterns (Cuozzo and Sauther, 2004, 2006a; Junge and Sauther, 2006; Millette et al., 2009). While sources and patterns of dental impairment are now well documented for BMSR *L. catta*, how such impairment impacts individual behavior, health, nutrition and ecology are not yet fully understood, nor is the process by which ring-tailed lemurs may compensate against the negative effects of dental impairment. In this dissertation, I seek to contribute to our understanding of the effects of dental impairment by examining how tooth wear and loss are related to aspects of food processing behavior (including compensatory behaviors), fecally-measured nutritional status, and food processing capacity as measured though fecal particle size. In addition, I also examine measures useful for understanding health status and its relationship to dental impairment through quantification of ring-tailed lemur coat and body condition.

In this initial chapter, I provide contextual background information for interpreting data presented in subsequent chapters. I first present information concerning aspects of dental functional morphology and adaptation, including a discussion focusing on the potential impacts of high-level dental wear (i.e., “gross” dental wear or “macrowear”). I then discuss methods used

for assessing worn tooth function, concentrating on those used to assess tooth wear among BMSR *Lemur catta* during this study (e.g., GIS-based dental topographic analysis). I then provide contextual information about dental-ecological research lead by Drs. Michelle Sauther and Frank Cuozzo on dental wear and tooth loss for BMSR *Lemur catta* and preliminary data regarding potential behavioral compensation for tooth loss among these animals (see Millette, 2007; Millette et al., 2009). I also provide additional contextual information concerning aspects of general ecology and resource availability at BMSR. Finally, I discuss the overall framework used by this project to assess aspects of dental impairment among BMSR *L. catta.*

Understanding Primate Dental Morphology, Function and Adaptation.

Dental anatomy and morphology are critically linked to the acquisition and mechanical breakdown of food items (Tattersall et al., 1988; Lucas, 2004). As with most mammals, primates reduce foods into particles through food processing and mastication. This process represents a key initial step towards the subsequent digestion and utilization of ingested food items. Lucas (2004) notes several mechanisms by which the initial processing and mastication of food items enhances digestion. First, the effectiveness of digestive enzyme action and microbial fermentation upon foods is proportional to the surface area of items consumed. Reducing foods into particles increases the surface area per unit of volume ingested. Smaller-sized food particles demonstrate increased surface area in comparison to larger-sized particles. Individuals capable of efficient comminution of foods may therefore demonstrate increased digestive efficiency in comparison to individuals who do not effectively reduce food items. Second, mastication provides access to food items which would remain indigestible without preliminary breakdown. For example, seeds remain inaccessible if their protective coating is not fractured and pass

directly though the digestive system without effective digestion or utilization by the consumer. Third, the reduction of foods permits consumption of items that cannot be otherwise swallowed due to size or shape. Processing and mastication may therefore allow individuals to access food items that they would otherwise be incapable of utilizing. As a result of these benefits, the ability to effectively reduce food items into easily swallowed and digested particles is often presumed to be essential to individual health and survival [(Gipps and Sanson, 1984; Lucas, 2004; King et al., 2005) see discussion below].

Molar teeth are the primary structures with which foods are reduced to particles during mastication. Researchers have long noted adaptive, functional relationships between primate molar morphology and the structural and mechanical properties of foods consumed (see Kay, 1975; Kay et al., 1978; Seligsohn, 1977; Yamashita, 1998a). Given the importance of food breakdown to digestion and fermentation, it is not surprising a variety of dental adaptations suited to reducing foods of differing properties have evolved across the primate order. Kay (1975) suggests that primate diets consist primarily of three principal food types: 1) plant materials high in structural carbohydrates such as leaves, bark and pith, 2) plant materials high in non-structural carbohydrates such as fruit and sap, and 3) animal foods. Commonly divided into the heuristic categories of “leaves,” “fruit,” and, “insects,” these food types each require different processing techniques in terms of shearing, crushing, and grinding which are in turn facilitated by tooth morphology (see also Kay and Hiiemae, 1974). Kay et al. (1978) suggest that shearing crest length and total molar basin areas may be used to assess the relative importance of shearing, crushing, and grinding in breaking down the diet of a given primate. Folivorous and insectivorous primates possess higher and longer shearing crests, and often enhanced grinding and crushing surfaces in comparison to frugivores (Kay, 1975). Shearing adaptations, in

particular, allow folivores to cut through leaf cellulose and effectively reduce leaf material into fine digestible fragments, and also allow insectivores to puncture and masticate the tough chitin exoskeleton of invertebrate prey. Although distinguishing between folivores and insectivores is difficult based on shearing capability alone, these dietary strategies may be effectively separated by body size, with folivores an order mass larger than insectivores as a result of energetic constraints (Kay, 1975; Kay et al., 1978; Fleagle, 1999). In contrast, more frugivorous taxa demonstrate smaller teeth in relationship to body size with reduced shearing crests. The teeth of frugivores therefore facilitate breakdown of foods by maximizing contact area between relatively blunt teeth and food items (Fleagle, 1999).

Boyer (2008) notes that assessments of diet and tooth form have often been conducted in a predominantly qualitative fashion. As noted above, primate taxa with relatively low-crowned teeth are thought to consume brittle foods requiring crushing, those with teeth with long, reciprocally bladed teeth were suited towards the consumption of leaves and insects, while those with tall cusps have diets consisting of insects with hard exoskeletons (Gregory, 1922; Szalay, 1968; Gingerich, 1974a,b; Godinot and Mahbouti, 1992 in Boyer 2008; Kay 1975; Strait 1993a,b; Evans and Sanson, 1998; Boyer 2008). In a number of classic studies, Kay and colleagues (e.g. Kay, 1975, 1977, 1978; Kay et al., 1978; Kay and Hylander, 1978; Kay and Covert, 1984) demonstrated that relationships between diet and dental form may be effectively quantified in living primates (and by extension, extinct primate taxa). Although these researchers developed a number of measures useful for linking morphology to diet (e.g. molar tooth size scaling, multivariate analysis of tooth features), the most commonly used and applied is that of “shearing quotient” (SQ) which has been successfully used to assess diet in a multitude of primate taxa, both living and extinct. Shearing quotient is defined for a given tooth as the

residual from a regression of the total length of shearing surfaces to that tooth's mesiodistal length. A high residual value therefore corresponds to relatively long shearing crest lengths and, in living primate taxa, a diet which is typically folivorous or insectivorous. Taxa with reduced shearing quotient residuals demonstrate omnivorous, frugivorous or graminivorous diets (see review in Boyer, 2008). Using a method similar to shearing quotient, Strait (1993b) found that insectivores with relatively "hard" diets demonstrate less shearing capacity than do those with relatively "softer" diets. Similar patterns have also been found for non-primate taxa with shearing quotient values also relating to diet among marsupials and chiropterans (Strait, 1993a; Hogue, 2004; Boyer, 2008).

Although this measure successfully links morphology to diet, the use of shearing quotients is limited due to variation between taxonomic groups in shearing crest length in relation to dietary profiles. For example, folivorous strepsirrhines demonstrate shorter crest lengths than do folivorous hominoids, although both demonstrate longer crests than do omnivorous relatives. Failure to generate residuals using a taxonomically proper regression may result in gross miscalculation of diet. For example, if *Avahi* (a highly folivorous strepsirrhine) is assessed using the hominoid-specific regression, it is misclassified as a frugivore (Kirk and Simons, 2001; Ungar and Kay, 1995; Boyer 2008). Given that shearing quotient must be determined using a regression based on a relevant taxonomic scale, its calculation and use is potentially limited to relatively close-related species. Shearing quotient is also limited in terms of its use with teeth where key landmarks (e.g., cusps and crests) necessary to calculate SQ have been ablated through wear (Ungar and M'Kirera, 2003; Evans, 2005; Boyer, 2008). These potential limitations of shearing quotient, however, are readily addressable using computer-intensive three-dimensional methods (see below).

Focusing on a more recent set of literature based upon sensory ecology and food item mechanical properties, Lucas (2004) argues that during the process of mastication dental geometry-induced shearing, grinding, and crushing are of secondary importance to the physical properties of food items. Food breakdown is instead dependent upon the ability of animals to produce and propagate the runaway growth of cracks in ingested items (Lucas, 2004; Cuozzo and Yamashita, 2006). The manner in which crack formation occurs relies primarily upon the physical properties of food itself, which is related to the food species' defenses against predation and/or damage. Food species demonstrate two basic strategies of mechanical defense: 1) stress-limited defenses and 2) displacement-limited defenses. Borrowing analytical techniques from materials science, these modes of mechanical defense may be described and quantified in terms of the relationship between the measures of elastic modulus and toughness. Elastic modulus (E) refers to the stiffness of a material and its resistance to bending. Toughness (R) reflects the ability of a material to arrest cracks once they have been initiated (Lucas, 2004; Cuozzo and Yamashita, 2006). Stress-limited defenses may be represented by the mathematical function "$\sqrt{(E*R)}$" and these properties are attempts to prevent cracks from starting. Plants with stress-limited defenses feature structures requiring predators to produce high forces to initiate cracking. Stress-limited foods are thus brittle, "hard" items that prevent initial crack formation by a consumer but shatter quickly once cracks have been started. Such plants rely on a defensive strategy by which a predator is rendered incapable of starting the initial breakdown down of a potential food item and is therefore unable to consume or effectively utilize that food item. In contrast, displacement-limited, "toughness-based" defenses are defined by the function "$\sqrt{(R/E)}$" and rely upon structures which arrest cracks once formed. Although consumers may start an initial crack within a potential food item, they cannot deform such foods to the point of

fragmentation. Further breakdown is thus precluded, thereby disallowing consumption. Thin, flat foods such as leaves, in turn, rely strictly on their toughness (*R*) to resist crack propagation and fragmentation (Ashby, 1989; 1999 in Lucas, 2004; Lucas 2004; Cuozzo and Yamashita, 2006).

Following this view, the physical properties of food items represent a key selective pressure in the evolution and development of primate dental morphology. Tooth form reflects adaptations necessary to create runaway crack propagation in foods with particular physical properties and mechanical defensive strategies. Stress-limited foods (e.g. seeds) are best defeated utilizing blunt cusps or wedged teeth that propagate cracks without incurring damage to the tooth itself. Likewise, "soft" fruits are best reduced by blunt teeth as these break open cell walls more effectively than do sharp, bladed teeth. Displacement-limited foods do not easily shatter and tend to prevent crack propagation. As a result, bladed teeth are required to start and continue crack propagation of "tough" items such as herbaceous or woody vegetation, or thin materials such as leaves. Similarly, bladed dentition is also utilized to breakdown tough insect exoskeletons (Lucas, 2004). While such patterns as these are frequently visible, linking distinct dental features to discrete categories of food items is often difficult, as morphology reflects mechanical properties which may be heterogeneous within food categories. Although both conceptualizations of dental morphology have a place when discussing gross wear, food property-based approaches may prove more adequately suited to this topic. Food property-based approaches moreover readily allow assessment of dental function despite ablation of cusp and crest surfaces as these often focus primarily upon overall tooth form rather than the quantification of distinct tooth features. In addition, tooth wear reflects damage incurred through mechanical interactions with ingested foods. Comprehending the physical properties of food items may therefore aid in understanding patterns of gross wear formation.

While these measures do appear to distinguish broad dietary categories, primates often do not fit into such discrete conceptions of feeding behavior and often consume heterogeneous diets (e.g., they do not consume strictly “fruit”, “leaves” or “insects”). Most primates are flexible in their dietary patterns, adjusting their diets to variations in food availability, food chemical defenses and nutritional content, and often consume food items from multiple trophic levels. As such, most primates may best be considered as omnivores rather than strict “folivores,” “frugivores” or “insectivores.” (Lambert and Rothman, 2015). Moreover, it is often difficult to discretely discern a given primate’s dietary patterns from dental morphology alone. For example, although it has been argued that large incisors are associated with the consumption of large fruits, among 4 Sumatran primate species Ungar (1996) found no relationship between incisor size and the consumption of fruit, food item size or habitual incisor use. Similarly, although chimpanzees possess dental-morphological and physiological adaptations to frugivory, they will often consume foods with high levels of pith and terrestrial herbaceous vegetation during periods of fruit inavailablity (Lambert and Rothman, 2006). Similarly, Asian colobines have been observed to demonstrate low rates of folivory despite apparent adaptations to leaf consumption (Marshall 2004; Yeager and Kool, 2000 in Marshall and Wrangham, 2007; Marshall and Wrangham, 2007). As such, dental morphology may be difficult to link to general dietary patterns, and a number of researchers have suggested that dietary adaptations may reflect foods consumed as secondary resources (see below).

As many primates consume mechanically and structurally heterogeneous diets, it is also important to consider to *which* food items in such a diet may correspond to a given species’ tooth morphology. It is furthermore important to note that *when* these foods are consumed may also impact tooth form. What is key here is considering whether the dentition is adapted towards the

consumption of foods which comprise the majority of the diet (e.g. Kay, 1975), or if dental morphology more accurately reflects adaptations necessary to break down food items which are consumed during critical bottlenecks in resource availability (Lambert et al., 2004; Ungar, 2007). Fallback food items, defined as "foods whose use is significantly negatively correlated with the abundance of preferred foods" (Marshall et al., 2009 in Sauther and Cuozzo, 2009) may represent a key force in the evolution of a species' dental morphology. A seminal example of this effect is reported in Lambert et al. (2004) with respect to relative molar enamel thickness in sympatric *Cercopithecus ascanius* (red-tailed guenons, thin enamel) and *Lophocebus albigena* (grey-cheeked mangabeys, thick enamel) in relation to the hardness of fallback foods consumed by these species. Thick enamel is often associated with hard-object feeding of foods such as seeds or unripe fruit foods (Kay, 1981; Dumont, 1995; Teaford et al., 1996; Lambert et al., 2004). Although *L. albigena* did consume absolutely harder foods than did *C. ascanius,* no differences were noted between the measured hardness of these species' overall diets across a period of several years. Significant dietary overlap was also noted between these species total diets. Both findings suggest that differences in morphology were not indicative of overall dietary choices. However, when limited to a single year (1997), *L. albigena* demonstrated significantly more hard-object feeding than did *C. ascanius,* primarily in the form of eating exceptionally hard *D. abyssinica* seeds and the consumption of bark. Lambert et al. (2004), suggest that differences in enamel thickness likely does not reflect overall dietary differences between these species, rather enamel thickness reflects species differences in diet during resource-depleted periods (but see McGraw et al., 2012). Thick enamel in *L. albigena*, they argue, has been selected to provide benefits during periods when preferred foods are unavailable. Ungar (2007) also highlights similar trends which may explain differences between gorilla and chimpanzee dental

morphology and fallback food choice. While chimpanzee molars demonstrate relatively low crowns and appear to be oriented towards a fruit-based diet, gorilla molar morphology is indicative of tougher food items such as leaves and fibrous plant parts. Nevertheless, both taxa exhibit extensive dietary overlap in areas where they live sympatrically (Williamson et al., 1990; Tutin and Fernandez, 1993). When preferred foods are scarce, however, gorillas tend to fall back more on leaves and fibrous foods than do chimpanzees. Morphological differences in the teeth of these species therefore likely reflect not overall dietary choice, but primarily fallback food choice. Similarly, Yamashita, 1998a found that among five Malagasy lemurs, dental morphology was most closely associated not with the most frequently consumed foods, but rather reflected the consumption of those foods with the most challenging mechanical properties, although this pattern did not hold for all measures calculated [(e.g., crest lengths) see also discussion in Cuozzo and Yamashita, 2006]. When considering form-function relationships between primate dental morphology and diet, it is thus important to consider not only the impact of the overall diet upon such associations, but also to consider foods which are available during potential selective bottlenecks resulting from reduced resource availability as well as the physical properties of such foods.

Gross Dental Wear: Sources, Adaptation, and Impacts.

Tooth wear results in the alteration of tooth morphology through physical and/or chemical processes, and is a commonly observed among both extant and fossil nonhuman primates (Ungar, 1998; Ungar and M'Kirera, 2003; Dennis et al., 2004; Lucas, 2004; King et al., 2005). Dental wear moreover may be thought of as "foodprint" (Ungar, 2014) which reflects the behavior, biology and ecology of a given species or individual, either living or extinct. As such,

dental wear ranges in severity from microscopic pits and scratches (microwear) through alterations in tooth morphology visible to the unaided eye [e.g., gross wear (Lucas, 2004)]. Gross dental wear is reflected in the gradual ablation of the occlusal surfaces through reduction of cusp and crest height and may result in the exposure of dentin through worn enamel. Extensive gross tooth wear may furthermore result in the reduction of the occlusal surface to a point where only a ring of enamel remains surrounds a flat pond of dentin. Alternatively, in extreme cases wear may result in the total ablation of the tooth crown to the root (King et al., 2005; Cuozzo and Sauther, 2006a,b). While chemical action due to acidic foods (e.g. erosion) may result in gross wear, scanning electron microscope studies suggest that wear is a primarily a function of mechanical processes resulting in formation of microscopic pits and scratches (Teaford and Glander, 1996; Lucas, 2004). Lucas suggests that such wear occurs through two mechanisms: 1) contact between occluding tooth surfaces (tooth-tooth wear, i.e., attrition) and 2) contact between teeth and food items (tooth-food wear, i.e., abrasion). Although tooth-tooth contact results in distinct wear facet patterning, and is used by many primates in canine sharpening (see Ryan, 1979; Janis, 1984), Lucas (2004) argues that wear results primarily from mastication of food items containing minute particulate matter. Microscopic pits result from compression of foods containing particulates of greater hardness than enamel such as plant phytoliths or quartz particles (Ungar et al., 1995; Lucas 2004; but see Sanson et al., 2007). Scratches, in turn, are produced by sliding particles across the tooth's surface, and require a lesser amount of force and particle hardness to induce wear. In instances where dentin has become exposed, similar pitting and scratching occurs, although lower forces are required to initiate wear. Gross dental wear is therefore a developmental process reflecting primarily accumulated microwear of both enamel and dentin, of which the latter wears at a faster rate once the enamel has been breached (Teaford and

Glander, 1996; Lucas, 2004). Food items such as woody plants, insect cuticles, hard fruits, or bone may cause wear directly by virtue of their hard or tough physical properties. Furthermore, tooth damage may also result from repeated consumption of hard items or exposure to high magnitude forces capable of fracturing the tooth (Lucas 1991; Schofield et al., 2002, Van Valkenburgh et al., 1990; Lucas, 2004; Cuozzo and Sauther, 2004). Experimental models based on resin-filled glass domes, Lucas et al. (2008; see also Constantino et al., 2009) suggest that feeding on hard objects may cause fractures in enamel to begin at the Dentin-Enamel Junction (DEJ) and proceed through the overlying enamel due to the differential hardness between hard enamel and comparatively soft dentin. These researchers also note that differences between enamel and dentin hardness may result in cracks (abfractions) around the tooth margin for species that consume relatively soft foods. Additionally, these authors argue that thick enamel and the presence of enamel rod decussation in the microstructure of enamel commonly seen in hard-object feeders may reflect adaptations intended to stop cracks from growing from the DEJ into the enamel, thereby preserving tooth function. These authors also hypothesize that the presence of cingula often found in “lower” primates may reflect adaptations meant to stem the growth of abfractions among primates with comparatively soft diets. These data moreover highlight that, in addition to facilitating food breakdown, tooth form and structure likely reflect features necessary to prevent tooth breakage during mastication. Such data are also significant to this study as Yamashita et al. (2015a) indicate that dental wear and tooth loss at BMSR may result from such DEJ-related fractures resulting from interactions between tooth form and diet at this site, and additionally note that individuals consuming more mechanically challenging diets demonstrate higher amounts of topographically measured dental wear (see below, see also Chapter 4 for a complete discussion of this paper).

Given the connection between tooth form and food breakdown, severe tooth wear may impair or prevent the reduction and subsequent digestion, fermentation and absorption of key food items. This is presumed to negatively impact individual health, life history, survival, and reproduction [(Gipps and Sanson, 1984; Lanyon and Sanson, 1986; Logan and Sanson, 2002a,b; Lucas, 2004; King et al., 2005) see below]. Natural selection, however, acts not only on unworn tooth form, but should also select for maintenance of mechanical efficiency despite wear. Much as unworn teeth are "designed" to overcome the structural and physical properties of food items, teeth should maintain morphological features suited for food reduction across the process of wear (Rensberger, 1973; Kay, 1981; Teaford, 1983; Janis and Fortelius, 1988; Ungar and M'Kirera, 2003; Dennis et al., 2004). As species-specific patterns of diet are reflected in unworn tooth morphology, features exposed by wear should also be suited to processing foods possessing physical properties commonly found within a species' dietary niche (Ungar and M'Kirera, 2003; King et al., 2005; Glowacka et al, 2016).

While researchers have long noted relationships between unworn tooth morphology, function and diet (e.g., Kay, 1975; Seligsohn, 1977; Kay et al., 1978), the lack of research focusing on dental wear and its influences on behavior, health and life history results, in part, from a paucity of effective methods for inferring and quantifying the impact of wear on dental function. Traditional methods for assessing dental functional morphology have relied primarily upon the quantification of distinct morphological features (e.g. crests and cusp morphology) and/or ratios between tooth features [(e.g. Shearing Quotient) M'Kirera and Ungar, 2003; Evans, 2005]. While such methods reliably distinguish primarily folivorous and insectivorous species from largely frugivorous species these utilize morphological landmarks typically obliterated by tooth wear (Kay 1978; Kay and Hylander, 1978; Kay and Covert, 1984; M'Kirera and Ungar,

2003). As a result, the function of worn teeth cannot be readily or easily assessed using traditional shearing quotient-based techniques. Such issues may, however, be overcome through use of computer intensive three-dimensional methods for assessing dental function that do not rely on standard morphological markers (see below). Such methods for understanding dental function are utilized by this study to examine the effects of dental impairment on food processing behavior (Chapter 4).

Three-Dimensional Methods for Assessing the Impact of Wear on Tooth Function.

Over approximately the past 15 years, a number of researchers have applied computer-intensive, three-dimensional methods for modeling the relationship of mammalian dental morphology and diet, as well as the impact of wear upon primate dental function. In addition to several other systems [e.g. Virtual Reality Modeling Language (VRML) or Computer Aided Drafting (CAD) systems], Geographic Information Systems (GIS) technology has been used to effectively examine worn teeth, and to study the maintenance and ultimate decline of tooth function through the course of wear (Zuccotti et al., 1998; Jernvall and Selanne, 1999; Ungar and Williamson, 2001; M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Ungar, 2004; Dennis et al., 2004; King et al., 2005; Bunn and Ungar, 2009; Head, 2011; Klukkert et al., 2012a; Cuozzo et al., 2014; Yamashita et al., 2015a; Glowacka et al., 2016). Although GIS technology was originally derived to model and analyze the Earth's surface, dental morphology may be characterized and assessed by obtaining a digital elevation model (DEM) of the tooth's occlusal surface using a laser scanner or other device capable of recording three-dimensional objects (e.g. confocal microscope or piezoelectric scanner) and entering the resulting elevation data as a landscape (Ungar and Williamson, 2001; M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003;

Ungar, 2004; King et al., 2005; Klukkert et al., 2012a,b; Cuozzo et al., 2014; Yamashita et al., 2015a). GIS tool packages may then be used to quantitatively characterize the topography of the entire occlusal surface. Such occlusal topographic models permit evaluation of functional morphology without specific landmarks, thus permitting a dimensionless characterization of worn teeth (M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004; King et al., 2005). Specifically, the measures of occlusal topographic *slope*, *relief* and *angularity* have demonstrated usefulness for assessing the functionality of worn teeth. *Slope* refers to the average change in elevation across the tooth's surface, and generally decreases with escalating wear. Slope may provide (along with some indication of diet) a measure of tooth wear, particularly when wear is also standardized for a given specimen using a complimentary technique [(e.g. Scott scores) M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004; Ungar and Bunn, 2008; Bunn and Ungar 2009]. Occlusal *relief* is defined as the ratio of occlusal surface area to planometric area, and serves as a measure broadly analogous to a "shearing quotient" applicable to worn teeth without morphological landmarks. Relief measurements thus provide insight into diet in a manner similar to shearing quotient (e.g. taxa with higher relief consume folivorous or insectivorous diets). Boyer (2008) also notes that relief index (although calculated using micro-CT techniques) permits assessment of morphologically divergent taxa while being relatively insensitive to taxonomic distinctions. Moreover, relief may be effectively and accurately calculated despite significant wear of features necessary to calculate shearing quotients. Relief indices often decline with wear, and may provide some indication of overall wear state (Ungar and M'Kirera, 2003, but see Boyer, 2008). Finally, *angularity* is defined as the rate of change in slope across the occlusal surface, and reflects the tooth's overall "jaggedness." Tooth angularity is thought to produce multi-directional forces upon food items facilitating their

breakdown. (Ungar and Williamson, 2000; M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003, but see Bunn and Ungar, 2009). Additionally, angularity scores tend to be maintained through the course of wear, only declining with extreme wear where the occlusal surface of the tooth has been fully removed. Angularity measures may provide insight about when wear reduces the occlusal surface of a tooth to the point which it no longer functions (Ungar and M'Kirera, 2003; Dennis et al., 2004, but see Klukkert et al., 2012a).

Three-dimensional methods have proven effective for examining dental functional morphology and characterizing development of tooth wear and its impacts on dental function in nonhuman primates. A series of GIS-based studies by Peter Ungar and Francis M'Kirera (M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003) comparing variably worn chimpanzee (*Pan troglodytes*) and gorilla (*Gorilla gorilla gorilla*) lower second molars demonstrate that topographic measures of slope, relief and angularity differentiate species-level differences in diet. Both taxa demonstrate reduced relief and slope with increasing wear, with gorillas showing relatively higher mean occlusal slope, relief and angularity across all degrees of wear. These authors suggest that these data are consistent with *G. g. gorilla*'s consumption of tougher fallback foods (see Tutin et al., 1991; Remis, 1997) and which require longer tooth-tooth contact times and steeper planes of contact for shearing and slicing. Lower values for each measure are indicative of flatter crushing and grinding surfaces consistent with and suited to the chimpanzee's fruit-based diet (M'Kirera and Ungar, 2003). Angularity scores remained constant across both taxa, and may highlight a means through which teeth maintain their function through the course of wear (Ungar and M'Kirera, 2003). In this case, angularity is maintained through the formation of enamel-edged pits resulting from differential wear between harder enamel and softer dentin once dentin has been exposed through the tooth's crown. These features

furthermore provide new food breakdown sites, which are well suited to shearing along newly exposed enamel (see also King et al., 2005; Glowacka et al., 2016). These authors suggest that occlusal surface angularity is crucial to preserving molar function and represents a measure that may be used to assess worn tooth functionality. Ungar and M'Kirera's findings therefore supply evidence that selection has produced teeth which wear in a manner facilitating the breakdown of foods with specific physical properties.

Dennis et al. (2004) similarly suggest that angularity may serve as a measure of molar functionality through a 7-year longitudinal study of gross wear among 14 wild mantled howler monkeys (*Alouatta palliata*) living within two microhabitats of differing food availability. These researchers found that while slope and relief continuously decreased from samples conducted at the study's onset and across those conducted two, four and seven years later, angularity scores only decreased after seven years. Such decreases in angularity suggest that dental functionality declines with severe wear, and that this measure may serve as an effective measure of the onset of dental senescence. For each individual, average values for slope, angularity and relief were calculated for the m2 occlusal surface from molds produced at the onset of the study and subsequently at 2, 4 and 7 years. As individuals aged, for all years slope and occlusal relief decreased. Due to formation of sharp shearing edges where enamel wear exposed dentin, angularity remained constant through year 4 – suggesting maintenance of dental function. However, decreases in angularity by year 7 indicate that functionality may decline with severe wear. Dennis et al. (2004) also noted no significant differences between microhabitats in patterns of wear development, suggesting diet is not the primary factor in gross wear patterning in this species. In this case, these researchers argue that patterning of gross wear likely follows a species-specific trajectory as determined by natural selection.

It is notable, however, that among primarily folivorous mountain gorillas (*Gorilla gorilla beringei*), Glowacka et al. (2016) reported that molar angularity was observed to decrease along with other measures of wear (e.g., relief index) as animals aged. However, these researchers did note that the greatest reductions in angularity were among the oldest individuals and for individuals with the greatest amount of wear but for which ages were unknown. They likewise reported that such reductions in angularity may represent individuals who are approaching dental senescence due to ablation of the occlusal surface. Such findings are similar to those of Klukkert et al. (2012a), who found that molar angularity decreased with wear among chimpanzees, but which may have been associated with individuals who were approaching a senescent state. Glowacka et al. (2016) further suggest that despite reductions in angularity, dental function was likely maintained through the creation of compensatory shearing crests produced during the course of wear and despite reductions of angularity. Thus, while angularity measure may serve as a measure of dental function in a number of species, reductions in angularity have been observed to occur with wear and may be independent of other factors which maintain dental function such as shearing crest length. Such maintenance of shearing crests and food processing performance through wear has been noted among both sifakas and geladas (King et al., 2005, Venkataraman et al., 2014), suggesting that such mechanisms may preserve functionality prior to dental senescence in these species. Few, if any, data from wild nonhuman primates are available to assess whether such changes in angularity impact aspects of health, nutrition, behavior or food processing capacity, and I know of no studies that have examined or published data on these areas of interest to date. As such, the association of this measure with the maintenance of dental functionality remains a hypothesis which has not yet been fully tested.

Dental topographic analysis has proven effective for discerning taxonomic patterns of dental wear, tooth form and use among non-great ape primates (such as reported by Ungar and M'Kirera, 2003 and Glowacka et al., 2016). For example, Ulhaas et al. (2004) add to the discussion of three-dimensional modeling of wear and taxonomic differences in tooth form with a study comparing variably-worn cercopithecine and colobine molars. In this study, both relief index and slope were determined for vervet and red-colobus molars using a system analogous to GIS but based on the Polyworks software package and CAD (Computer Aided Drafting) software. In addition, relative differences in wear were standardized using the ratio of exposed dentine to the tooth's total occlusal surface. These researchers note findings similar to those presented by Ungar and M'Kirera (2003) in that taxonomic and functional distinctions are present between colobine and cercopithecine teeth, despite possessing a similar bilophodont morphotype. Overall, colobines demonstrated higher, sharper crests with steep-sloped cusps of high relief. In contrast, cercopithecines featured comparatively more rounded cusps of reduced slope and relief and with less-pronounced crests. In terms of relief, colobines demonstrate higher occlusal relief through all wear stages, although both demonstrate a loss of relief through time. These authors note that comparatively high relief is maintained among colobines, even at relatively late wear stages, while cercopithecines are of relatively lower relief through the course of wear. These authors also argue that the maintenance of relief in colobines is indicative of shearing adaptations and correspond to a relatively folivorous diet. Taxonomic differences in wear patterning and dentin exposure are also visible. Although both samples show extensive wear on buccal cusps, on lingual cusps colobines demonstrate later exposure of dentine than do cercopithecines. In colobines dentine basins of lingual and buccal cusps fuse to form elongated areas of dentine exposure on the mesial side of the lophs. Combined with relatively steep cusps

with orientations suggestive of lateral movement during mastication, the authors further highlight the importance of leaves in the colobine diet and dentition. In contrast, for vervets the orientation of exposed dentine tends to face towards to central fovea of the molar until wear exposes dentine throughout the occlusal surface. The authors suggest that this is indicative of a mortar-and-pestle-like action between the molar basins and corresponding antagonistic cusps, and is consistent with fruit and seed-eating commonly observed among *C. aethiops.* These authors demonstrate that three-dimensional modeling may be applied to distinguishing tooth form and function on a greater taxonomic scale than the Ungar and M'Kirera's (2003) study, which was limited to hominoids. Nevertheless, these results suggest that these distinctions in form and function between taxa are maintained through the course of wear.

Findings similar to those reported by Ulhaas et al. (2004) have also been reported by Bunn and Ungar (2009) for lower first and second molars across a range of cercopithecine (*Cercopithecus campbelli* and *Cercocebus torquatus*) and colobine (*Colobus polykomos* and *Procolobus badius*) species. Using GIS techniques (e.g., those of Ungar and M'Kirera, 2003; Dennis et al., 2004; Cuozzo et al., 2014, etc.), these researchers found that for both m1 and m2 slope and relief values differed between folivorous colobines and frugivorous cercopithecine taxa. Interestingly, with the exception of the m2s of *P. badius and C. polykomos*, there were no significant differences for occlusal slope or relief between species or within broad dietary characterizations (e.g. frugivore vs. folivore). The authors suggest that this reflects the coarseness of these dietary characterizations and/or variations in the fracture mechanics of foods consumed. In contrast to predictions generated by Ungar and M'Kirera (2003) that more folivorous species should demonstrate greater angularity than frugivorous species, frugivorous *C. campbelli* demonstrated higher than expected angularity. Such predictions do, however, hold true

for differences between the colobine taxa and *C. torquatus*. With respect to wear, these authors found cercopithecine and colobine molars wear in a manner different than previously reported for hominoids and platyrrhines. While cercopithecine and colobine m2s and colobine m1s demonstrated reductions in slope and relief (e.g. became flatter) with wear as expected, cercopithecine m1s did not. The authors suggest that these results may stem from the fact that cercopithecine m1s start out relatively low to begin with, and may also demonstrate thicker enamel that is less susceptible to wear. As a result, these authors furthermore advise that m1 and m2 should be considered separately (e.g., m1s should not be compared to m2s directly) when discussing potential relationships between diet and tooth form. Although angularity has been suggested as an indicator of tooth function, in this study differences were noted between wear stages for m2s of all taxa, and m1 of two of the species. These differences, however, may result from the onset of dental senescence as almost all variation in angularity results from low angularity scores present in extensively worn specimens, as is expected if this measure is a valid indication of the loss of dental function (see above).

Although molars wear as to maintain functionality, severe and extreme wear may lead to total ablation of occlusal morphology. Severe wear thereby results in reduced masticatory efficiency and the onset dental senescence, the "…wearing out and end of functional life of the teeth." (Ungar, 2005:16533). Dental senescence likely results in negative effects in terms of health, survival and reproduction, due to reduced rates of energetic conversion for food items ingested. Severe wear has been reported in a number of wild primate populations, although its effects on health and evolutionary fitness have been little studied. Teaford and Glander (1996) found howling monkeys at Hacienda La Pacifica, Costa Rica demonstrate wear-related reduction of shearing facets in older individuals, which may result in the less efficient breakdown of food

items in comparison to prime-aged adults, although this hypothesis was not tested directly by these researchers. In a separate study conducted in this population of howling monkeys, Dennis et al. (2004) suggested that age-related changes in angularity and loss of functional efficiency coincided with extreme wear (see above).

King et al. (2005) highlight that GIS measures may prove useful not only for examining form-function relationships between tooth and diet and characterizing patterns of wear, but may also be of use for inferring the impact of dental wear and senescence on life history when combined with relevant individual life history and ecological data. Although these researchers measured shearing blade length rather than total surface relief or angularity in female *Propithecus edwardsi*, individuals maintained dental functionality through the course of wear by exposing auxiliary shearing crests until wear eventually removed all occlusal enamel. As much as Dennis et al. (2004) observed, molars maintained functionality through the appearance of compensatory shearing blades at sites of dentin exposure accompanying the reduction of crown height. However, around 18 years of age such compensatory shearing blades are lost as progressive wear reduces the occlusal surface, "…to a shallow dentine bowl surrounded by a low-relief enamel band" (King et al., 2005:16581). Despite dental senescence, many females continue to survive and reproduce for another 10 years. However, rates of infant survival among senescent females are reduced during periods of low rainfall. As *P. edwardsi* obtains most water through dietary sources, the authors suggest that dentally senescent mothers produce milk of both low nutritional and water content. Although milk content was not measured directly during this study, infants appeared to die primarily of dehydration. King et al. (2005), therefore provide intriguing evidence that tooth wear could have an impact on reproductive output and fitness. Thus the findings of King et al. (2005) provide evidence that use of GIS-based techniques used

in this study may be effectively combined with behavioral data to assess the potential impacts of dental wear and impairment. Overall, topographic methods, when combined with field observations, therefore indicate that although dental functionality may be maintained to a point, the onset of dental senescence may have important implications for life history outcomes in nonhuman primates.

Tooth Wear and Loss among BMSR *L. catta.*

As I have previously discussed elsewhere (see Millette, 2007; Millette et al., 2009), ring-tailed lemurs living in and around Parcel 1 of the Bezà Mahafaly Special Reserve demonstrate patterns of tooth wear and tooth loss which are well-suited to exploring the ontogeny and impacts of dental impairment in nonhuman primates. This population demonstrates exceptionally high rates of diet-related tooth wear and subsequent antemortem tooth loss in comparison to other nonhuman primate populations for which such data are available (Sauther et al., 2002; Cuozzo and Sauther, 2004, 2006a; Sauther and Cuozzo, 2009). In a study of 83 living BMSR *L. catta* Cuozzo and Sauther reported that 26.5% of individuals demonstrated the complete loss (e.g., total ablation of the crown) of at least one tooth as a function of dental wear (Cuozzo and Sauther, 2006a). Likewise, these researchers highlighted that BMSR *L. catta* demonstrate the highest rate of tooth loss reported for any wild nonhuman primate population, with 6.4% of all positions (n = 192/2988) being scored as absent (see Cuozzo and Sauther, 2006a). In addition, the loss of multiple teeth is also common among this population, with 10.8% of individuals demonstrating greater than 30% loss, and 4.6% showing greater than 50% tooth loss. Extreme amounts of tooth loss have also been reported, with a number of individuals being reported as functionally edentulous. The highest reported amount of tooth loss in living

individuals is 81%, a level of dental impairment which has been reported for more than one lemur over the 10 years of dental ecology research conducted at this site [Orange 170, (Cuozzo and Sauther, 2004, 2006a); Black 226, this study]. Figure 1.1 shows an example of a living BMSR ring-tailed lemur with extensive tooth loss and wear relative to an individual with a low amount of dental impairment.

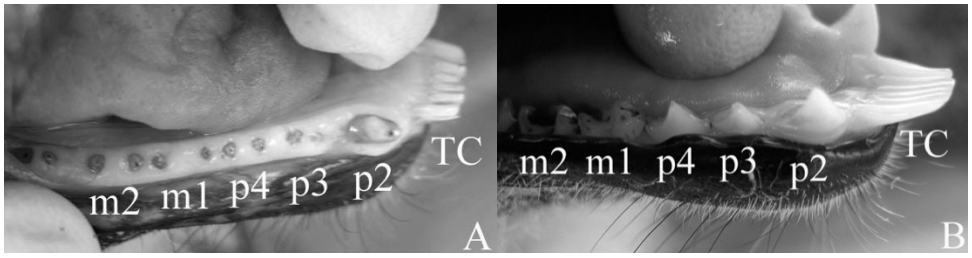


Figure 1.1. Tooth loss in a BMSR ring-tailed lemur. (A) Severely worn *Lemur catta* mandibular teeth, with only worn roots remaining (Yellow 195), also note the worn toothcomb (=TC). (B) Lightly worn mandibular teeth in *L. catta* (Teal 205). Note the unworn toothcomb (=TC). Images from Cuozzo and Sauther, 2006a.

Patterns of tooth loss at BMSR are not related strictly to the process of aging (e.g., wear increasing as a function of chronological age), but rather appear to be a function of diet (see below). Highlighting this pattern, in a comparative study contrasting lemurs over the age of 10 years at BMSR with those >10 years housed at the Indianapolis Zoo, Cuozzo et al. (2010) found that relatively few captive individuals were missing teeth in comparison to wild BMSR individuals. Of those captive individuals missing teeth (N=2/7), one was missing only a single tooth, while the other individual demonstrated 25% loss. This individual was, however, a decade older than any individual ever reported at BMSR. All individuals known to be >10 years of age at BMSR demonstrated tooth loss ranging from 6% to 50%. Interestingly, the two individuals in this sample demonstrating the least amount of loss at 6% and 14% were the youngest and oldest individuals respectively. This suggests that tooth wear and loss in this population is not an

accurate indicator of age, but also highlights the potential of resource use and ranging patterns as a source of tooth wear and loss among this population. Cuozzo et al. (2010) note that individuals with the highest frequencies of tooth loss are most commonly living in groups found along the border of the Sakamena River (see range map in Chapter 2). These areas are heavily forested with *Tamarindus indica* trees, the fruit of which may result in heavy dental wear (Sussman and Rakotozafy, 1994; Sussman and Ratsirarson, 2006; see below), but are also areas where ring-tailed lemurs have been observed to use human cultivated crops (Sauther and Cuozzo, 2009; Millette, personal observations).

Such variances in habitat-related dental wear patterns and onset of early dental wear have also been confirmed using GIS-based dental topographic methods. Young adult individuals inhabiting disturbed areas demonstrate early-aged onset of reduced topographic scores for "occlusal lift" (a combination of slope and relief) in comparison to those inhabiting protected areas or with access to human-sourced food items (Cuozzo et al., 2014, see also Head, 2011). Likewise, Yamashita et al. (2015a) report reduced topographic scores among individuals living in areas with more mechanically-challenging diets (e.g., BMSR Parcel 1 and 2 where tamarind is a common food source vs. Tsimanampetsotsa National Park where tamarind is rarely consumed), or where exogenous grit is likely to be included in the diet (e.g., *L. catta* of BMSR Parcel 2, where dietary grit is likely common, demonstrate higher rates of wear than found in Parcel 1). These data indicate that diet, rather than age alone, is related to patterns of dental wear observed at BMSR (see below; see also Chapter 4 for a full discussion of ring-tailed lemur dental topography).

Mammalian dental wear reflects a complex interaction among potential variables, including: behavior, food item mechanical properties, food availability and quality, food

processing and mastication, dental morphology, and enamel microstructure (Molnar, 1971, 1972; Rensberger, 1973; Graham and Burkart, 1976; Smith et al., 1977; Smith, 1984; Lanyon and Sanson, 1986; Janis and Fortelius, 1988; Teaford and Oyen, 1989; Hillson, 1996; Gandara and Truelove, 1999; Maas and Dumont, 1999; Verrett, 2001; Kaifu et al., 2003; Lucas, 2004; Lussi et al., 2004; Nussey et al., 2007)**.** Extensive tooth loss among the BMSR ring-tailed lemurs appears to result primarily from severe and extensive wear rather than dental damage or disease, as is more commonly observed for wild nonhuman primates with dental impairment and/or tooth loss (Sauther et al., 2002; Cuozzo and Sauther, 2004, 2006a,b). For BMSR *L. catta*, multiple studies have implicated consumption of ripe tamarind fruit (*Tamarindus indica*) as the primary source of severe tooth wear and loss found in this population (Sauther et al., 2002; Cuozzo and Sauther, 2004, 2006a,b; Cuozzo et al., 2008; Sauther and Cuozzo, 2009; Yamashita et al., 2012, 2015a). Ripe tamarind fruit is the hardest and toughest of all foods regularly consumed by ring-tailed lemurs at BMSR in terms of its mechanical properties (Yamashita, 1996, 2000, 2008b, Yamashita et al., 2012), and is moreover a key fallback food during the resource-depleted dry season for ring-tailed lemurs inhabiting the riverine gallery forests at BMSR (Sauther, 1992, 1998; Cuozzo and Sauther, 2006a; Simmen et al., 2006; Sauther and Cuozzo, 2009). In addition to its challenging mechanical properties, tamarind fruit is also physically large in size, and is the largest fruit regularly consumed by ring-tailed lemurs at this site, although individuals also occasionally consume other fruits of similar or larger size (e.g., *Ficus coccifolia* or *Crateva excelsa*). As a result of its large size, the processing and consumption of this food requires the use of a large portion of the dentition, and which may promote wear across multiple postcanine positions (Cuozzo and Sauther, 2006a,b; also see below).

As noted above, mechanical properties play a central role in how food items fragment during processing and subsequent mastication, and are also linked to those dental forms used to process and break down food items. As tamarind fruit demonstrates both hard and tough properties (Yamashita, 2000, 2008b; Yamashita et al., 2012), and is physically large, it is an exceptionally challenging resource for ring-tailed lemurs to process, breakdown and consume. Tannins found in tamarind fruit, which may reduce lubrication by saliva (Prinz and Lucas, 2000; but see de Wijk and Prinz, 2005), likely also contribute to the pattern of wear in the BMSR lemurs (Cuozzo et al., 2008). Consumption of mechanically "hard" foods, such as tamarind, is frequently associated with thick dental enamel and blunt cusp morphology. Ring-tailed lemurs, however, do not demonstrate thick molar enamel as is commonly found among hard-object feeding primates; rather they possess exceptionally thinly-enameled teeth in comparison to other primate taxa. In addition, *Lemur catta* also demonstrate comparatively elongated shearing crests relative to other lemurids, and which are similar to those of folivorous lemurs [e.g., *Propithecus* (Kay et al., 1978; Dumont, 1995; Shellis et al., 1998; Yamashita, 1998a,b, 2008b; Martin et al., 2003; Cuozzo and Sauther, 2004, 2006a,b; Lambert et al., 2004; Lucas, 2004; Godfrey et al., 2005)]. Consequently, ring-tailed lemur dental morphology appears ill-suited to processing large hard and tough tamarind fruit, resulting in the high rates of tooth wear and loss found within this population. In contrast, sympatric *Propithecus,* despite also frequently consuming tamarind fruit, rarely display severe molar wear and exhibit little tooth loss (Cuozzo and Sauther, 2006a, 2013). Such patterns among *Propithecus* are primarily related to their focus on unripe tamarind fruit, which is neither as hard nor as tough as the ripe tamarind fruit typically consumed by ring-tailed lemurs (Yamashita, 2003, 2008b; Cuozzo and Sauther, 2006a). Patterns of tooth wear and loss observed among ring-tailed lemurs at BMSR may therefore reflect a potential "ecological

mismatch" or evolutionary disequilibrium between this species' dental morphology and the physical properties of food items consumed. As ring-tailed lemurs demonstrate some apparent adaptation to folivory (Yamashita, 1998a,b, 2008b), in consuming mechanically-challenging tamarind they are utilizing a fallback food which does not appear to match their dental morphology. This "disequilibrium" thus appears to be the result of an over-reliance on this fruit in the tamarind dominated gallery forests of southern Madagascar. Such disequilibrium may result from the recent extinction of other lemur species which thereby allowed ring-tailed lemurs to consume previously unavailable foods, and/or changes to the forest at BMSR which resulted increasing availability of tamarind fruit to ring-tailed lemurs. (Cuozzo and Sauther, 2006a, 2010, 2013; Cuozzo et al., 2008 Sauther and Cuozzo, 2009; Cuozzo et al., 2010). Likewise, it is uncertain whether tamarind fruit is endemic to Madagascar or was introduced there by humans, as is the duration for which it has served as a key food species for ring-tailed lemurs. Based upon its common use by both human and animal populations Du Puy et al. (2002) suggest that the species is endemic to the island, and Goodman and Jungers (2014) have argued for species endemism based on published genetic data. However, Diallo et al. (2007) suggest that Malagasy tamarind is not genetically divergent from that found in mainland Africa or India, and argue that tamarind may be of mainland African origin based on paleontological and cultural evidence (see also discussion in Cuozzo and Sauther 2015). While the length of time that tamarind has been available to ring-tailed lemurs at BMSR is currently uncertain, it is clear that this is a source of tooth wear and loss for this population. For example, in the spiny forests of southern Madagascar, where tamarind is rare (e.g., Tsimanampetsotsa National Park), ring-tailed lemurs display reduced tooth wear and rare tooth loss (Cuozzo and Sauther, 2006a, 2013; Sauther and Cuozzo, 2009; Yamashita et al., 2015a), further suggesting disequilibrium at BMSR.

As noted above, tamarind pods are a large and challenging food to process. They have a hard outer shell, and portions must be bitten off before ingestion; this is normally done using the postcanine teeth (Sauther, 1992; Yamashita, 2003). As a result, severe wear and tooth loss occurs most frequently in the postcanine positions, and is most common for positions involved directly in tamarind fruit processing (Cuozzo and Sauther, 2006a). M1 is the most frequently lost tooth position, followed (in descending order) by P3, P4 and M2. Because M1 is the first permanent position to erupt, the high rate of M1 loss likely reflects an interaction between eruption schedule and tamarind consumption, which coincides with and/or begins soon after weaning (Sauther et al., 2002; Cuozzo and Sauther, 2004; 2006a). The association between loss and eruption schedule is, however, weakly linked for other tooth positions. For example, P3 and P4 positions are more frequently absent than earlier-erupting positions (e.g. I1, I2, and M2). High rates of dental wear are also noted upon the deciduous dentition of subadult individuals at BMSR (Cuozzo and Sauther, 2006b). Ring-tailed lemurs at BMSR are weaned before the dry season and erupt their deciduous dentition during the period which *T. indica* serves as a key fallback resource. These individuals have been observed to follow the adult pattern of ripe tamarind fruit processing and consumption during this period. Sauther and Cuozzo (2009) note that wear is clearly visible in the deciduous dentition of individuals approximately 10 months of age, while little wear is noted for those with newly erupted teeth, suggesting rapid and early wearing of recently exposed teeth. These data suggest that the frequency of absence for a given position is related primarily to tooth function, and highlights the key role of tamarinds in the generation of tooth wear and antemortem loss in ring-tailed lemurs (Cuozzo and Sauther, 2006a). Similarly, Cuozzo et al. (2014) note that variation in tooth wear related to tamarind fruit consumption and forest use is visible among young adult individuals (e.g., 2-3 years of age) using topographic

methods, particularly for those individuals who inhabit disturbed areas (see below; see also Chapter 3) Additionally, severe tooth wear and tooth loss is more frequent for females living in altered habitats where the availability of non-tamarind-based resources is limited in comparison to protected areas of the reserve (Sussman and Rakotozafy, 1994; Sauther and Cuozzo, 2009). These data further highlight the importance of tamarind in the generation of tooth loss for individuals at BMSR, but also demonstrate that anthropogenic alterations to the environment may impact the relative availability, subsequent use, and resulting impacts upon the dentition, of tamarind fruit (see also Head, 2011; Cuozzo et al., 2014).

Severe wear and tooth loss, such as that observed among ring-tailed lemurs at BMSR, is often assumed to prohibit individual access to key food items, ultimately resulting in death (Lucas, 2004). While tooth wear and loss likely impedes the processing, breakdown and consumption of tamarinds and other key food items, at least three individuals have been observed to live in a nearly-edentulous state for up to 3 years (Sauther et al., 2002; Cuozzo and Sauther, 2004, 2006a). These data are further augmented by observations for two females at least 10 years old (they were first captured and tagged in 2003). Individual Blue 139 was last captured in 2004 with 39% tooth loss and was reported in the population as of 2008. In turn, Blue 138 who demonstrated 56% tooth loss in 2007 was still alive the following year (Cuozzo et al., 2010). Individuals with tooth loss have also been observed to survive into old age while remaining reproductively active. Preliminary data on reproduction and infant survival for these individuals suggests that significant tooth loss does not predict a lack of infant survival through weaning (Cuozzo et al., 2010). For example, individuals Orange 156 and Green 459 (14% and 19% loss respectively) have been observed to produce infants who survive through weaning. This pattern is in contrast to that described for *Propithecus* at Ranomafana National Park, where

degree of tooth loss was associated with infant survival during periods of reduced rainfall (King et al., 2005).

Tooth loss does appear to be associated with observed patterns of overall health to some extent, although the association of health status with dental impairment remains somewhat unclear. For example, tooth loss of greater than 8% has been associated with various measures of sub-optimal health (e.g., hair loss, dental abscesses, anemia, high white blood counts), with individuals with extensive tooth loss (more than 30%) exhibiting serious health issues, including potential hepatic inflammation (Loudon et al., 2006; Miller et al., 2007). However, in a survey of general health in 69 individuals, only three of nine animals rated in "fair" or "poor" health showed >10% tooth loss, and two out of four individuals with >50% tooth loss were observed in "good" health (Cuozzo and Sauther, 2004). Thus, while there appear to be health effects relatable to tooth wear and loss, the relationship is not direct, suggesting that animals may be adjusting their behaviors in compensation for dental impairment.

Behavioral Responses to Dental Impairment among Mammals and BMSR *L. catta.*

One way ring-tailed lemurs at BMSR may compensate for dental impairment is through behavioral mechanisms. Primates have been shown to modify their behaviors to deal with a variety of energetic challenges analogous to those potentially resulting from tooth loss. For a number of primates, lactating females must alter their activity budgets to accommodate the energetic costs of nursing (see review in Dufour and Sauther, 2002). For example, lactating gelada baboons increase energy intake by beginning to feed earlier in the day and withdrawing from less important social relationships (Dunbar, 1983). Likewise, lactating yellow baboon females increase time spent feeding in exchange for time normally spent resting or socializing

(Altmann, 1980). However, to my knowledge no other researchers have investigated individual behavioral responses to dental impairment among wild non-human primates for which individual dental data are available, although several such studies have been conducted on non-primate mammals (Gipps and Sanson, 1984; Perez-Barberia and Gordon, 1998; Logan and Sanson, 2002a,b,c). Among male koalas (*Phascolarctos cinereus*), Logan and Sanson (2002a,b,c) found that individuals compensated for extensive tooth wear by increasing time engaged in feeding behaviors while reducing behaviors not directly related to somatic maintenance. In comparison to individuals with low to medium wear, those with severe dental wear increased time spent feeding by 62%, while simultaneously increasing the volume of food consumed by 41% and chews per leaf consumed by 116% (Logan and Sanson, 2002c). In turn, time spent walking (e.g., traveling) or engaged in movement was significantly reduced in individuals with advanced tooth wear, as was home range size (Logan and Sanson, 2002b). Individuals with heavy wear also engaged in fewer social behaviors, suggesting a general reduction of non-maintenance expenses in favor of compensatory feeding (Logan and Sanson, 2002a). Increases in food volume intake similar to those observed in *P. cinereus* have also been reported for ring-tailed possums (*Pseudocheirus*) with experimentally ablated dentition (Gipps and Sanson, 1984). In addition, among captive red deer (*Cervus elaphus*), individuals with reduced masticatory efficiency (as measured by molar occlusal surface area) spend more time chewing food items than those with relatively higher masticatory efficiency (Perez-Barberia and Gordon, 1998).

Preliminary observations conducted at BMSR indicate ring-tailed lemurs may compensate for dental impairment through adjustments to their behavior (Cuozzo and Sauther, 2006a, Millette, 2007; Millette et al., 2009). As part of my Master's work, I conducted a two month study examining behavioral responses to tooth loss in BMSR ring-tailed lemurs (Millette,

2007; Millette et al., 2009). In this work I demonstrated that ring-tailed lemurs with tooth loss show a suite of behaviors divergent from those without tooth loss in terms of both activity budget and food processing. In particular, individuals with tooth loss exhibited reduced resting behavior and a trend towards increased foraging during the Early Afternoon, a period of the day when ring-tailed lemurs typically sleep and/or rest (Sauther, 1992). When limited to those with >10% tooth loss, however, such individuals also engaged in significantly higher frequencies of feeding, foraging and grooming and lower frequencies of resting during the Early Afternoon, indicating that the magnitude of tooth loss is related to the expression of compensatory behavior. Individuals with tooth comb wear also demonstrated greater mutual allogrooming and trends towards higher rates of autogrooming and total grooming, indicating that lemurs also compensate behaviorally for toothcomb damage-related reductions in grooming efficiency.

While I found no significant differences for feeding bout lengths between individuals with and without tooth loss, significant differences were present between those with >10% tooth loss and those with <10% tooth loss for fruit feeding ($p = 0.033$) and which were related to tamarind fruit consumption. Individuals with tooth loss also demonstrated significantly higher frequencies of licking food items, both total feeding bouts and for tamarind fruit feeding, indicating that licking may be a food processing behavior used in compensation for dental impairment.

While this initial study demonstrated that ring-tailed lemurs alter their behaviors in response to reduced dental functionality, it did not examine a number of other alternative means by which individuals may compensate behaviorally against the impacts of dental impairment. For example, individuals with dental impairment may also utilize food items which have been previously opened and then discarded by other individuals (see Cuozzo et al., 2006a), or utilize

foods which have been opened prior to initial dental processing. Likewise, during this initial study, at least one individual with extensive tooth loss (Blue 138) repeatedly opened tamarind fruit utilizing the hands, a behavior which is rarely observed among ring-tailed lemurs. The occurrence of such behaviors, and their relation to dental impairment, is however not yet well understood.

The Bezà Mahafaly Special Reserve: Characteristics and Resources Available to *L. catta.*

The Bezà Mahafaly Special Reserve (BMSR) (23˚30'S, 44˚40'E) is located in southwest Madagascar approximately 35km northeast of the town of Betioky Sud. The reserve consists of two officially protected forest parcels (Parcel 1 and Parcel 2), an annex connecting the two parcels, and a research camp. At 4,200 ha it is the largest continuing tract of riverine forest in southwestern Madagascar and is classified as an IUCN Category V Reserve. As with most research on BMSR ring-tailed lemurs, this project was conducted in or adjacent to Parcel 1, an 80ha western Malagasy dry deciduous gallery forest located along the western bank of the Sakamena River (Sauther et al., 1999; Ratsirarson, 2003). Parcel 1 has been surrounded by a barbed-wire fence since 1979 and has thus been relatively protected from anthropogenic impacts (e.g. logging or livestock grazing) by both the fence and through an agreement by the local Mahafaly population to not graze their animals within the reserve. The eastern portion of Parcel 1 is a mature riparian deciduous and semi-deciduous forest that becomes more xerophytic as one moves west away from the river [(Sauther, 1998; Ratsirarson, 2003) Figure 1.2]. This gallery forest is primarily dominated by *Tamarindus indica*, particularly on the parcel's wetter eastern portion. Eastern portions of the reserve are characterized by a closed canopy of roughly 15-20m in height and consisting primarily of tamarind, *Acacia rouvmae*, *Euphorbia tirucalli*, and

Salvadorea angustifolia. Below this canopy layer, and in western portions of Parcel 1, most trees are relatively short in height ranging from 2 to 15m. Dominant tree species in a given area of the Parcel 1 tends to follow the decreasing east-west gradient of water availability, although tamarind and *Azima tetracantha* are found throughout Parcel 1 (Sussman and Ratsirarson, 2006).

Unprotected gallery forest that covers approximately 200ha is located contiguously to both the north and south borders of Parcel 1 (Sussman and Ratsirarson, 2006). These unprotected areas immediately outside Parcel 1 demonstrate anthropogenically-reduced productivity and degradation of the forest understory resulting from the grazing and farming practices of local Mahafaly agro-pastoralists. Within these areas the availability of understory lianas and herbs is reduced in comparison to Parcel 1 (Sussman and Rakotozafy, 1994; Sauther, 1998; Sauther and Cuozzo, 2009). Also located immediately adjacent to Parcel 1, the camp area consists of several small buildings used for administrative and research purposes and camping facilities for researchers and visitors. The camp also features an outdoor kitchen with associated open trash pits that are sometimes raided by several lemur groups (Fish et al., 2007; Sauther et al., 2007; Millette, personal observations). Although some lemurs utilize these human-derived resources, there is no intentional provisioning of this population. Ring-tailed lemurs use all of these areas irrespective of anthropogenic alterations, with some groups using both reserve and anthropogenically-disturbed areas (Sauther et al., 2006, Millette, personal observations). Images of Parcel 1, the camp and surrounding areas are available in Figure 1.2.

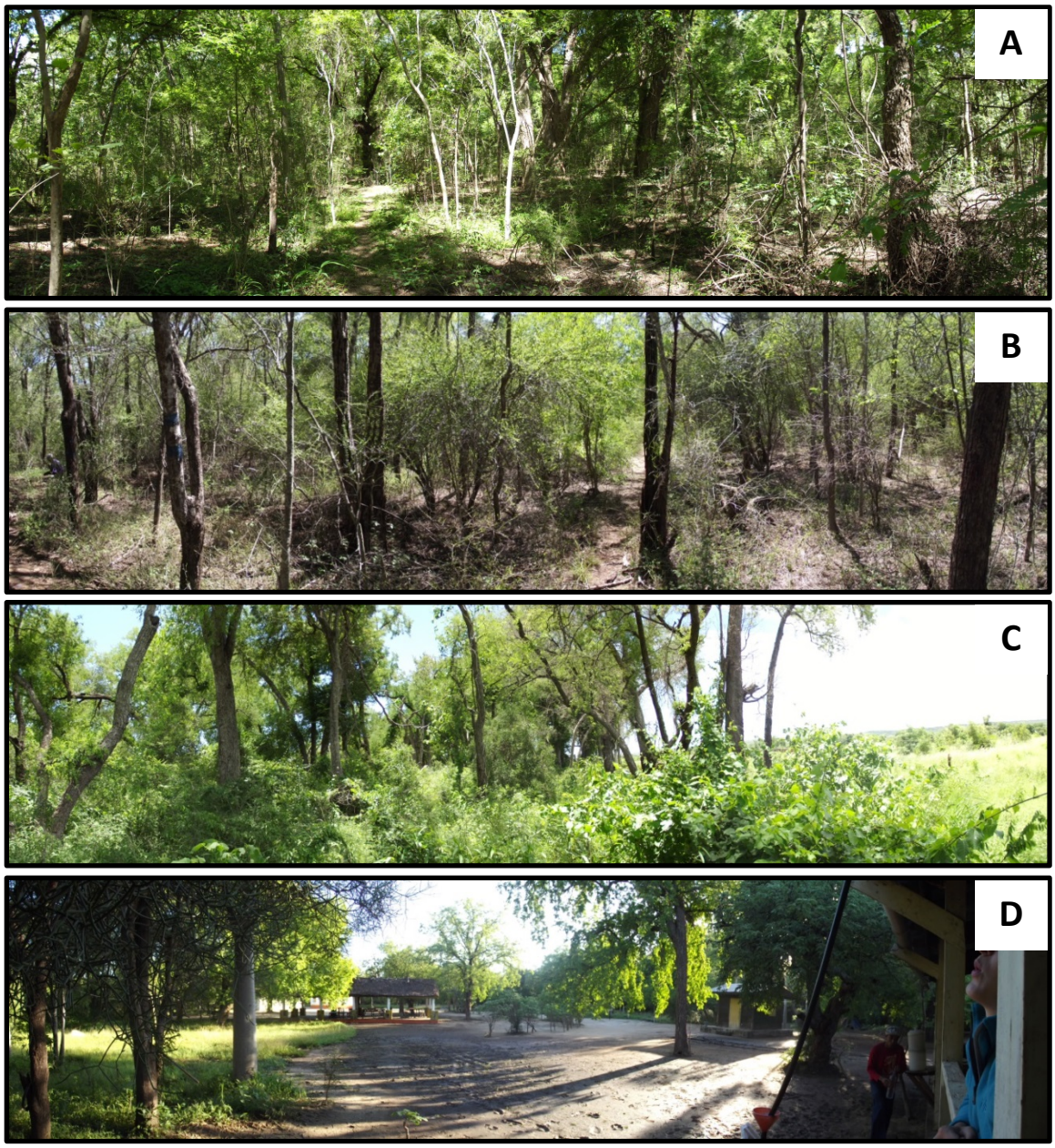


Figure 1.2. Images of BMSR. A shows the eastern portion of Parcel 1**. B** shows the western portion of Parcel 1. **C** shows disturbed forest to the south of the reserve along the Sakamena River. Note the reduced amount of canopy coverage. **D** shows the camp area.

Immediately west of Parcel 1 is a newly acquired protected annex of approximately 3400ha. Although the annex was only officially incorporated into the reserve in 2015, it was managed and patrolled by Madagascar National Parks during this study in 2012-2013 (see http://campuspress.yale.edu/bezamahafaly/decree-gives-legal-recognition-to-extension-of-the-beza-mahafaly-special-reserve/; Millette, personal observations). The portion of this annex immediately west of Parcel 1 (e.g., up to 200m west) was sometimes used by study animals of both Blue and Pink groups, which had core ranges found within western areas of Parcel 1 (see Chapter 2). The area of the annex used by lemurs during this study demonstrates a mosaic structure consisting primarily of shrubs and smaller trees, found in patches of dry deciduous forest, as well as patches of spiny forest (e.g., *Alluaudia* forest). In addition, there are a number of stands of tamarind and *Salvadora angustifolia* trees that are visited and fed upon by the lemurs of Blue and Pink groups (Millette, personal observations.). Similar to the areas north and south of Parcel 1, this area is relatively degraded by land use practices of the local Mahafaly people (Sussman and Ratsirarson, 2006; Millette, personal observations). As with Parcel 1, both ring-tailed lemurs and Verreaux's sifaka inhabit and utilize the annex area, although densities of sifaka are higher than those of ring-tailed lemurs in this region (Axel and Maurer, 2011; Millette, personal observations, Sauther, personal communication).

BMSR is characterized by a highly seasonal pattern of rainfall with distinct dry (April-October/November) and wet seasons (November/December-March). Rainfall averages 550mm per year with greater than 50% of precipitation (>100mm per month) falling from December to February. In contrast, rainfall from June to October averages less than 10mm per month (Sauther, 1998; Ratsirarson et al., 2001; Sussman et al., 2012). For this study, dry season observations were defined as occurring from July through October, while wet season observations were defined as occurring from November through March. The dry season was

determined as ending in late October as significant, daily rains began at this time (similar to that reported by Sauther, 1998). Likewise, although rainfall was not recorded directly (due to issues with consistent data collection between researchers and the BMSR staff), rainfall was relatively high during this study as two cyclones passed through the area during the wet season, resulting in flooding of the Sakamena River, which had not flooded since 2005.

Within and around Parcel 1, available resources tend to correspond to patterns of rainfall. Peak food availability occurs during the month of February and lowest food availability during July (Yamashita, 1996; Sauther, 1998; Gemmill and Gould, 2008; Sauther and Cuozzo, 2009; Millette, personal observations). During the height of the dry season from June to September, leaf, fruit and flower availability is at its nadir (Sussman and Ratsirarson, 2006). Ratsirarson et al. (2001) note that most species shed their leaves between the months of April and November. Flowering typically matches the rainy season, although many species flower during the end of the dry season, and tamarind trees critical to ring-tailed lemurs flower asynchronously during a period from November to June at this site (Sussman and Ratsirarson, 2006; Gemmill and Gould, 2008; Sauther and Cuozzo, 2009). *Tarenna pruinosum* also flowers and fruits asynchronously throughout the year, and serves as a dry-season resource for ring-tailed lemurs in Parcel 1, although it's fruit is not considered a fallback food as it's consumption does not appear to be related to phenological availability and/or availability of other preferred foods (Sauther and Cuozzo, 2009). Likewise, *Salvadora angustifolia* fruit serves as a key food item for ring-tailed lemurs during the transition between the dry and wet seasons, representing a key (e.g., approx. 70% of the diet) during this period, while flowers and leaves of this species serve as a key food immediately prior to this period (Millette, personal observations, see Chapter 3). Figure 1.3 highlights the variation in foliage between dry and wet seasons at this site.

Figure 1.3. Seasonal variation in forest foliage. Both images were taken at the same trail intersection in the western part of Parcel 1but at different points of the year. Image A shows the forest's condition during the dry season (September). Image B shows the forest during the wet season (January).

This Study: Its Goals, Aims and Structure.

In this dissertation I examine the impacts of dental impairment in the form of tooth wear and loss within a population or ring-tailed lemurs living at the Bezà Mahafaly Special Reserve, Madagascar (BMSR). As such, I use a broad dental ecology framework to link aspects of *Lemur catta* biology, health and behavior to dental anatomy in order to better understand the ontogeny and impacts of dental senescence among wild nonhuman primates (see Cuozzo and Sauther, 2012). As noted above, this population exhibits exceptionally high rates of tooth wear, and subsequent tooth loss due to the consumption of tamarind fruit (see Sauther et al., 2002; Cuozzo and Sauther, 2004, 2006a,b; Sauther and Cuozzo, 2009), and demonstrates individual rates of tooth loss that are among the highest recorded for any population of wild primates (Cuozzo and

Sauther, 2004, 2006a). Levels of individual tooth wear and loss observed in this population commonly reach those which are assumed to result in the functional loss of dental capacity (e.g., dental senescence; see Ungar et al., 2005, King et al., 2005), and which should theoretically result in negative impacts upon an individual's ability to maintain the capacity to adequately process, masticate and digest food items. Such impairment is also assumed to result in a reduced capacity for individuals to obtain sufficient nutrients necessary to maintain health, reproduction and survival (e.g., Gipps and Sanson, 1984; Lanyon and Sanson, 1986; Logan and Sanson, 2003a,b,c; Lucas, 2004; King et al., 2005; etc.).

While individuals within this population demonstrate frequent severe tooth wear and extensive loss (up to 81%), they have been observed to survive for a period of years in relatively good health (e.g., Sauther et al., 2004; Cuozzo and Sauther, 2006), suggesting that individuals may compensate behaviorally against the impacts of dental impairment either through alterations to activity budget or food processing behaviors (see Millette et al., 2009). However, few other field-based data are available regarding how dental impairment may impact aspects of behavior, health and nutrition directly among this population, or for wild primates generally. Although research conducted at this site suggests that tooth loss may result in a divergent capacity to process food items (Millette et al., 2012 / Chapter 6 this dissertation), little is known about how this may actually impact the animal's ability to process, digest and absorb nutrients from its diet and there has never been a direct test of the assumption that dental impairment leads to reduced nutritional intake among ring-tailed lemurs or, to my knowledge, for any other wild nonhuman primate. Likewise, while available data indicates that tooth loss may impact behavior, data on this aspect is of limited scope and is available only with respect to tooth loss rather than tooth wear alone. Moreover, few data focusing on tooth wear are available for wild nonhuman primate

populations generally (but see King et al, 2005; Venkataraman et al., 2014). Furthermore, teeth are expected to maintain their functionality prior to the total loss of function (Ungar and M'Kirera, 2003; M'Kirera and Ungar, 2003; King et al., 2005; Ungar, 2005) and may furthermore maintain their function through the course of wear until a fully senescent state is reached (King et al., 2005). How dental senescence develops and impacts aspects of behavior, health and nutrition is, however, not yet fully understood among nonhuman primates. Dental impairment has furthermore been cited as a driving force behind a variety of ecological and behavioral attributes, ranging from reduced infant survival in nonhuman primates (e.g., King et al., 2005) to the presumption of care-giving in the human fossil record (e.g., Lebel and Trinkaus, 2002; Tappen, 2009). Yet, no one has directly tested the assumption that dental impairment leads to reduced nutritional intake among nonhuman primates, and likewise few data are available to assess how tooth wear may lead to behavioral alterations among nonhuman primates.

In this dissertation I address these areas of interest though an analysis of ring-tailed lemur behavioral-ecological, nutritional and health data collected at BMSR from June 2012 to March 2013 in concert with fecal-nutritional and dental-topographic data collected at laboratories at both the University of Colorado (Dr. Matt Sponheimer's Nutritional and Isotopic Ecology Laboratory) and the University of Arkansas, Fayetteville (Dr. Peter Ungar's Paleoanthropology Laboratory) conducted from fall 2013 through summer 2015. This dissertation also includes work that I conducted concerning the relationship between fecal particle size and dental impairment status collected in the field during the 2006-2008 field seasons and assessed at the University of Colorado in 2010 prior to the commencement of this dissertation's primary field element.

During field elements of this dissertation, I collected a variety of data including approximately 920 hours of focal animal sampling to assess general dietary patterns across the study period, as well as general activity budget information. In addition, I also recorded all occurrences of feeding along with associated data including feeding bout duration, toothrow use, and use of food processing techniques with the potential to offset dental impairment (e.g., use of the hands, licking, use of open fruit, etc.). Although not utilized during this study, I also collected additional behavioral data with respect to group activity patterns (e.g., through group scan sampling) as well as data on dominance and agonistic interactions. In addition to behavioral data collection, I also recorded information related to individual health status in the form of coat and body condition scores as well as general indicators of health for each individual on at least a twice monthly basis. Finally, in order to assess nutritional status relative to dental impairment, I collected fecal samples for each focal animal across the course of the study. A full description of all field methods is available in Chapter 2.

In addition to field elements, I conducted a variety of lab analyses to understand the effects dental impairment on food processing behavior and individual nutritional status. First, I quantified postcanine tooth form for the lower dental arcade using dental topographic analysis (see Chapters 2 and 4). Data collection for topographic analysis was conducted at the Paleoanthropology Lab of the University of Arkansas, Fayetteville from October-November 2013, with subsequent data analysis conducted at the University of Colorado Boulder during spring 2014. This involved the development of and laser scanning of epoxy casts derived from impressions (collected for each study individual during health examinations at BMSR) and Geographic Information Systems-based analysis of resulting point clouds through use of generation surface models for the topographic measures of slope, relief and angularity (see

Chapters 2 and 4). Data generated from these analyses were then used to assess aspects of feeding behavior related to divergent patterns of food processing associated with dental impairment. In addition, I also conducted fecal-nutritional analyses in Dr. Matt Sponheimer's Nutritional and Isotopic Ecology Laboratory (NIEL) at the University of Colorado for fecal samples collected during field observations for a subset of individuals. Residual values for fecal crude protein, fiber (e.g., NDF and ADF), lignin and ash were assessed relative to individual tooth loss and tooth wear status as determined through an ordinal tooth wear scoring system (see Chapter 5).

Overall, in this dissertation I seek to provide a broad-based assessment on the impacts of dental wear on ring-tailed lemur food processing capacity (though behavioral and fecal food particle size measures), as well as on the ability of individuals to digest and absorb ingested food items (though assessment of fecal nutritional status). Additionally, I provide information on measures of ring-tailed lemur health status by examining seasonal variation in coat and body condition. This document is thus organized in the following format across a number of data-driven chapters:

Chapter 2 - Study Field Methods and Dental Topographic Methods: In the second chapter of this dissertation I describe all study methods utilized during field and dental topographic data collection. This includes a description of the study subjects used during this specific project, but also discusses the behavioral, health and dental methods carried out during the completion of this project.

Chapter 3 - General Patterns of BMSR *Lemur catta* Feeding Behavior: Chapter 3 details aspects of ring-tailed lemur feeding ecology during this field study. In this chapter I examine food item species and types consumed across the completion of this project, and provide a general context for understanding how feeding behavior and nutritional status are related to dental impairment in this population.

Chapter 4 - A Topographic Analysis of Ring-tailed Lemur Food Processing Behavior: Chapter 4 examines how the dental topographic measures of slope, relief, angularity across the dental arcade are related to patterns of food processing behavior. This chapter also introduces topographically-calculated 2D and 3D area measurements representative of the total "working area" available across the dental arcade as a means of quantifying dental impairment status. While this chapter focuses on a variety of feeding behaviors for the key fallback food of tamarind fruit, it also examines how feeding bout duration is impacted for a number of common BMSR *L. catta* food items. For data examined in this chapter, I hypothesize that feeding durations are negatively associated with dental impairment as measured using topographic methods, both for tamarind fruit and across food items generally. In addition, I hypothesize that increasing dental impairment is associated with the occurrence of a variety of food processing behaviors specific to tamarind fruit feeding (e.g., use of the hands, consumption of open fruit, consumption of fruit from the ground, etc.).

Chapter 5 - Fecal Nutritional Analysis in Relation to Dental Wear and Tooth Loss Status: In Chapter 5, I examine the relationship between postcanine dental wear and tooth loss relative to fecal nutritional status. Although it was not possible to examine the amount of nutrients

consumed directly (as all feeding could not be observed for each individual due to study design constraints), it was possible to assess relative amounts of nutrients remaining in feces following consumption and subsequent digestion. In this chapter I detail the relationship of dental wear and tooth loss to measures of dietary fiber (ADF and NDF), protein (e.g., fecal nitrogen), lignin content, and ash content. For these dietary components, I hypothesize that greater residual content will be present for individuals with tooth loss or increasing tooth wear for acid detergent fiber (ADF) and neutral detergent fiber (NDF) as well as for ratios of ADF, NDF, hemicellulose and cellulose to acid detergent lignin (ADL). I also hypothesize that fecal protein content will be greater for dentally-impaired individuals, while ADL content will decrease relative to other materials for individuals with dental impairment. Fecal ash values were not expected to vary by dental impairment status.

Chapter 6 - The Impact of Dental Impairment on Ring-tailed Lemur Food Processing Performance: This chapter, previously published in Millette et al. (2012) in the American Journal of Physical Anthropology, examines the relationship of fecal food particle size to tooth wear and tooth loss status. It therefore reports on results for fecal samples collected in the field from 2006-2008, and which were fractionated into different sizes and subsequently analyzed by fraction mass at the University of Colorado Boulder. Here, I hypothesize that individuals with dental impairment in the form of tooth wear and tooth loss demonstrate reduced proportions of small-sized food particles in their fecal material relative to those which do not demonstrate dental impairment.

Chapter 7 - Examining Visual Measures of Coat and Body Condition in Wild Ring-Tailed Lemurs at the Bezà Mahafaly Special Reserve, Madagascar: Although this chapter does not directly examine the relationship of dental wear to coat condition and body size, these are measures which may be used to assess health and nutritional status in ring-tailed lemurs. This chapter thus reports variation in coat condition and body mass for ring-tailed lemurs as related to reproductive status and seasonality based on data collected during dissertation field data collection from July 2012 to March 2013. As with Chapter 6, this chapter has previously been published previously, although this time in in the journal *Folia Primatologica* (Millette et al., 2015).

Chapter 8: Conclusion - Significant Findings and Directions for Future Research: In this chapter I summarize data collected during this project and its implications for understanding the relationships between dental impairment and ring-tailed lemurs' capacity to process and digest foods. I also discuss the impacts of dental impairment on ring-tailed lemur behavior and nutritional status. I finally discuss the broader impacts and implications of this study as well as potential areas for future research.

CHAPTER II:

STUDY FIELD METHODS AND DENTAL TOPOGRAPHIC METHODS.

In this dissertation I integrate field behavioral, health and fecal nutritional data collected from June-July 2012 to March 2013 at the Bezà Mahafaly Special Reserve, Madagascar. During this period, I collected approximately 920 hours of quantitative behavioral data from 36 (23 female, 13 male) ring-tailed lemurs (*Lemur catta*), using a combination of instantaneous focal animal sampling (to record activity budget data) and all occurrences sampling (to record feeding data) protocols. I also recorded health data using ordinal scoring systems for both coat and body condition (see Berman and Schwartz, 1998; Berg et al., 2009; Jolly, 2009a,b; Millette et al., 2015 / Chapter 7). Additionally, feces were sampled for each focal animal on a monthly basis, to be used in fecal nutritional assessments. Data presented in this dissertation reflect information collected from the height of the dry season (July to late October), through the transition between the dry and wet seasons (November) and onward until the end of the wet season [March (see Sauther, 1998; Rasamimanana, 2012; Millette, personal observations)]. Likewise, data for female lemurs also reflects the reproductive states of pregnancy (July to Sept/October, the birth season (September / October), as well as early to late lactation [September / October to the study's end (Millette, personal observations.)]. In this chapter, I detail study subjects and methods used to collect all behavioral, health and nutritional data resulting from and reported upon in this study.

Study Subjects and Sampling Methods.

Study Animals. Data for this study were collected from 36 (23 female, 13 male) adult ring-tailed lemurs from the Bezà Mahafaly Special Reserve, Madagascar and which were drawn initially from seven social groups (see below). All individuals were selected based upon their dental impairment status as determined by Dr. Frank P. Cuozzo during annual health inspections conducted in the dry seasons (June - July) of 2011 and 2012 (see below for capture protocols). Tooth loss status for study subjects ranged from 0% to at least 81% (Black 226, 2011 observations). Dental wear was also recorded for each subject using a 0-5 ordinal wear system, with average wear score across the postcanine dental arcade ranging from 1.55 to 4.96 (see below for a full description of ordinal scoring). Although all subjects were adults, individuals represented a variety of ages, from young adult (4 years) to old adult (13+ years). All study subjects were animals previously captured by the Bezà Mahafaly Lemur Biology Project (BMLBP), conducted since 2003 by Drs. Frank Cuozzo and Michelle Sauther. As a result of such captures, all study animals were identifiable using a numerical and color-based collar system. Data are also readily available for many individuals in terms of their life history, as well as their social and ecological histories. General health information and history is also available for most study animals. Please see Table 2.1 for a complete index of each study animal's identification, sex, social group, age and dental status.

It is notable that females outnumber males in this sample buy a sizeable margin (1.77:1). This imbalance is related to the effects of male migration on available study animals. While females most frequently reside in their natal group across their lifespan, males typically migrate from their natal group after adolescence, and often migrate repeatedly to other groups following

Table 2.1. Focal Animal Study Group and Individual Characteristics.

Group	Habitat	Subject ID	Year Obs	Sex	Age	Infant Status	Tooth Loss*	Postcanine Wear
Yellow	Reserve / Camp	155	11	F	11+	Survived	Low	3.909
Yellow	Reserve / Camp	172	11	F	11+	None	High	4.273
Yellow (Rain 1)	Reserve / Camp	208	11	M	11+	N/A	High	4.455
Yellow (Rain 1)	Reserve / Camp	223	12	M	9	N/A	No	3.45
Yellow (Rain 1)	Reserve / Camp	230	11	M	11+	N/A	Low	3.227
Yellow	Reserve / Camp	319	12	F	11	None	High	4.235
Yellow	Reserve / Camp	346	12	M	4	N/A	No	1.364
Red	Reserve	44	11	F	12+	Died	Low	3.727
Red	Reserve	231	12	F	9	Survived	No	3.45
Red	Reserve	347	12	F	4	Survived	No	1.55
Red (Green)	Reserve	23	11	F	12+	Died	High	3.818
Red (Lavender)	Reserve	38	12	F	13+	None	Low	3.82
Pink	Reserve	176	11	F	11+	Survived	No	3.318
Pink	Reserve	181	11	F	13+	Survived	No	3.727
Pink	Reserve	185	11	F	9+	Survived	No	3.818
Teal	Reserve	339	12	M	5	N/A	No	2.23
Teal	Reserve	340	11	F	10	None	No	3.412
Orange	Reserve / Camp / Marginal	154	11	F	11+	Survived	Low	3.81
Orange	Reserve / Camp / Marginal	268	12	F	8	Survived	No	2.55
Orange	Reserve / Camp / Marginal	273	12	M	?	N/A	No	3.5
Orange	Reserve / Camp / Marginal	307	12	M	6	N/A	No	2.55
Orange	Reserve / Camp / Marginal	308	12	M	6	N/A	No	3.14
Orange	Reserve / Camp / Marginal	316	12	F	6	Survived	No	2.32
Orange	Reserve / Camp / Marginal	341	12	F	4	Survived	No	1.68
Orange	Reserve / Camp / Marginal	343	12	F	5	Survived	No	1.682
Orange	Reserve / Camp / Marginal	368	12	F	11	Survived	No	3.91
Blue	Reserve / Camp / Marginal	217	12	F	8	Survived	No	2.91
Blue	Reserve / Camp / Marginal	218	11	M	8+	N/A	Low	3.118
Blue	Reserve / Camp / Marginal	246	12	F	10	Survived	Low	3.77
Blue	Reserve / Camp / Marginal	332	12	F	6	Died	No	1.82
Blue	Reserve / Camp / Marginal	348	11	F	11	None	High	4.647
Black	Camp / Marginal	226	11	M	13+	N/A	High	4.955
Black	Camp / Marginal	291	11	M	9+	N/A	No	3.273
Black (Rain 2)	Camp / Marginal	318	12	M	6	N/A	No	2.23
Black	Camp / Marginal	331	12	M	5	N/A	No	1.88
Black	Camp / Marginal	345	11	F	4	Survived	No	1.727

* No = No tooth loss, Low < 10% loss, High >10% loss (see Millette et al., 2009).

this initial movement. Males have been observed to migrate to groups outside of the Parcel 1 and/or the study area immediately surrounding the Parcel 1 (Cuozzo and Sauther, personal communication; Millette, personal observations). It is not feasible to follow these animals to their new groups. Likewise, it is not possible to immediately capture and collar all new adult male arrivals to the study area, and these males often go for some time in their new social groups before collaring. Such patterns of migration thus result in a comparatively lower number of collared male individuals in the study groups followed by the BMLBP and available for research by this project.

Study Groups and Their Locations. Subjects were drawn from and observed within a total of seven initial groups, with two additional social groups being observed after the migration of four adult males. All social groups were drawn from the area in and around of BMSR Parcel 1. Four of the seven study groups (Pink, Red, Yellow and Teal) ranged primarily within the reserve. Of these groups, Red, Yellow and Teal were found to move within the wetter, tamarind-dominated eastern portion of Parcel 1 to the somewhat more xerophytic and lower-canopied central portion of the forest. Although both Red and Yellow group utilized areas around the camp, this was only commonly observed for Yellow group. Teal group ranged from the tamarind gallery forest to the central areas of Parcel 1, and was never observed outside the reserve. Pink group utilized the scrub and small-tree dominated regions in the west and center of Parcel 1. This group also used the annex west of Parcel 1 and was seen in the camp area on a few occasions.

Two study groups, Orange and Blue, frequently utilized areas both within and outside of the Parcel 1. Orange group ranged from the eastern gallery forest along the western bank of the Sakamena River to the area approximately 100 - 200m west of the camp. Orange group also

extensively utilized disturbed areas immediately east of the camp area and was observed to frequently use the camp area to access resources there (e.g., open trash pits, water resources, mid-day sleeping sites). Blue group ranged primarily west of camp, both within and outside of the reserve. Disturbed areas frequently utilized included those south and west of the camp, as well as areas in xerophytic “spiny” forest immediately west of Parcel 1.

One initial group (Black) inhabited disturbed areas east and south of the camp area and south of Parcel 1. Although full-growth tamarind trees are common here, this region has been degraded significantly by anthropogenic impacts (see Whitelaw, 2010). Black group was observed to utilize areas from the farmlands west of the Sakamena and to frequently use camp resources.

Two other groups, in which study subjects were observed following male migrations, (Rainbow 1 and Rainbow 2) also inhabited disturbed areas south of camp. Rainbow 1 utilized areas both immediately south of the camp and occasionally utilized resources in the camp area. Rainbow 2 ranged primarily southwest of the camp in highly disturbed gallery forests along the banks of the Sakamena.

For groups ranging primarily within the reserve, I followed 15 study animals and collected data for seven initial individuals (4 male: 223, 230, 208, 346 and 3 female: 319, 155, 172) in Yellow group, three females in Pink group (176, 181, 185), and two individuals (1 male: 339, 1 female: 340) in Teal group. Red group included 3 females (44, 231, 347), although during observations two additional female individuals initially from other groups (Green 23, Lavender 38) were included with this group as they were attempting migrate into Red group throughout the study and associated frequently with Red group members. For groups using both areas inside and outside of the reserve, I observed14 individuals. Nine focal animals resided within Orange group

(3 males: 273, 307, 308, 6 females: 154, 268, 316, 341, 343, 368), while 5 focal animals were observed in Blue group (1 male: 218; 4 females: 217, 246, 332, 348). Five focal animals were observed within Black group (4 males: 226, 291, 318, 331; 1 female: 345). A map of all groups and their ranges is available below in Figure 2.1.

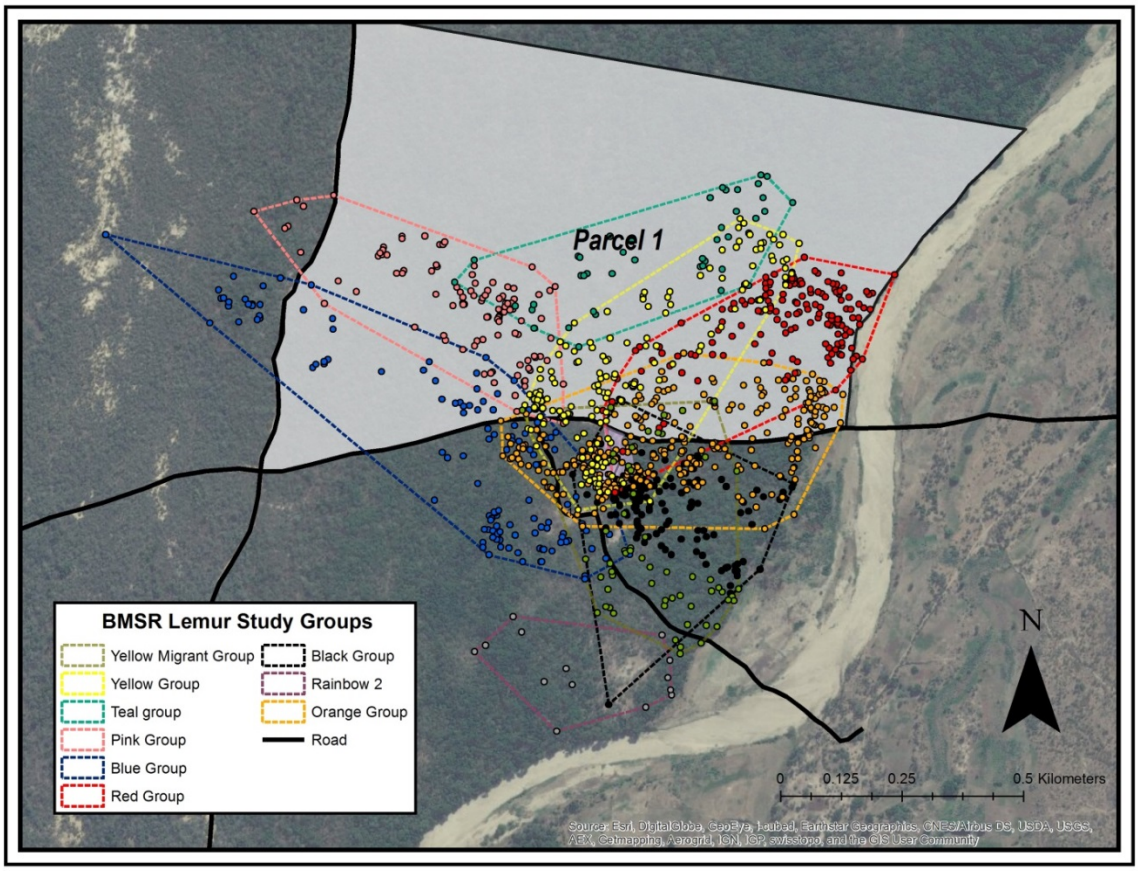


Figure 2.1. Ranges for each study group across the entire study period (July-Nov). Please note that the individual from Rainbow 2 (318) and the Yellow Migrants (208, 230, 346) separated from their initial groups (Black and Yellow) during the course of this study, and data presented here only represent that collected from late October-November onward.

Although not initially studied as a group, Rainbow 1 became a study group when three adult males from Yellow (208, 230, 346) migrated there during the onset of the wet season (November). Likewise, one male from Black group (318) was followed in Rainbow 2 after his migration to that group, also during the onset of the wet season. Such groups inhabited disturbed areas with few visits to the camp area.

Two study animals (Blue 218 and Teal 339) went missing during the completion of this study. It is likely that these individuals migrated from their groups as in both cases they disappeared along with another male, and it is common for males to migrate between groups in pairs (Sauther, personal communication).

Group size for all study groups ranged from 3 (Teal; July census) to 12 (Orange; all months) adult individuals. Including subadults and infants, groups ranged in size from 7 (Pink before infant births, and Teal in Dec-Jan) individuals to Orange group with 27 (potentially 28-29) individuals. Although only 4 individuals were noted in Teal group for July, this probably reflects a poor census count as no subadult individuals were reported, although these were observed in subsequent months. A monthly census for all groups is available in Table 2.2.

Sampling Strategy for Behavioral Follows. All behavioral data for each study subject were collected using 90-minute focal follows. I conducted behavioral follows for each individual at least twice monthly, with at least one follow conducted during the morning and at least one follow conducted during the afternoon. For follows that extended across noon (12:00), observations were counted as a morning follow if over 45 minutes of the follow was conducted prior to noon. If more than 45 minutes were recorded after noon, the follow was counted as an afternoon follow.

Table 2.2. Monthly Group Census Data by Age/Sex Classification.

Month	Sex/Age	Yellow	Red	Pink	Teal	Orange	Blue	Black	Rain 1	Rain 2
Jul.	**Male**	5	2(3)*	2	2	6	4	5	ND	ND
	Female	4	6	3	1	7	5	4	ND	ND
	Subadult / Juv	6	3	2	1	8	3	2	ND	ND
	Infant	0	0	0	0	0	0	0	ND	ND
	TOTAL	**15**	**11(12)**	**7**	**4**	**21**	**12**	**11**	**ND**	**ND**
Aug.	**Male**	5	3	2	4	5(6)	4	6	ND	ND
	Female	4	6	3	1(2)	7	5	4	ND	ND
	Subadult / Juv	6	3	2	3	7(8)	3	2(3)	ND	ND
	Infant	0	0	0	0	0	0	0	ND	ND
	TOTAL	**15**	**12**	**7**	**8(9)**	**19(20-21)**	**12**	**12(13)**	**ND**	**ND**
Sept.	**Male**	5	3	2	4	6	4	6	ND	ND
	Female	4	6	3	2	7	5	4	ND	ND
	Subadult / Juv	6	2	2	3	8	3	3	ND	ND
	Infant	2	2	0	0	7	3	3	ND	ND
	TOTAL	**17**	**13**	**7**	**9**	**28**	**15**	**16**	**ND**	**ND**
Oct.	**Male**	3	3	2	4	6	2	4	ND	ND
	Female	4	6	3	2	7	5	4	ND	ND
	Subadult / Juv	6	2	2	3	7 (8)	3	3	ND	ND
	Infant	2	5	3	0	7	3	3	ND	ND
	TOTAL	**15**	**16**	**10**	**9**	**27(28)**	**13**	**14**	**ND**	**ND**
Nov.	**Male**	3	3	1	4	6 (7)	4	5	ND	ND
	Female	4	6	3	2	7	5	4	ND	ND
	Subadult / Juv	6	2	2	3	7 (8)	3	3	ND	ND
	Infant	2	4	3	0	7	3	3	ND	ND
	TOTAL	**15**	**15**	**9**	**9**	**27(28-29)**	**15**	**15**	**ND**	**ND**
Dec.	**Male**	3	3	1	2	6	5	4	5	5
	Female	4	6	3	2	7	5	4	3	3
	Subadult / Juv	5(6)	2	2	3	7	3	3	3	3
	Infant	2	4	3	0	7	3	3	3	2
	TOTAL	**14(15)**	**15**	**9**	**7**	**27**	**16**	**14**	**14**	**13**
Jan.	**Male**	3	5	1	2	6	3	4	5	6(7)
	Female	4	6	3	2	7	5	4	3	4
	Subadult / Juv	6	1(2)	2	3	7	3	3	2	5
	Infant	3	3	3	0	7	3	3	3	2
	TOTAL	**16**	**15(16)**	**9**	**7**	**27**	**14**	**14**	**13**	**17(18)**
Feb.	**Male**	3	2(3)	3	4	6	3	5	4	5
	Female	4	6	3	2	7	5	4	3	3
	Subadult / Juv	5(6)	1(2)	2	3	1(7)	3	3	2(3)	5
	Infant	2	3	3	0	7	3	3	3	2
	TOTAL	**14(15)**	**12(13-14)**	**11**	**9**	**26**(27)**	**14**	**15**	**12(13)**	**15*** **
Mar.	**Male**	3	4	ND	ND	6	3	5	4	ND
	Female	4	6	ND	ND	7	5	4	3	ND
	Subadult / Juv	6	1(2)	ND	ND	6(7)	3	3	3	ND
	Infant	2	3	ND	ND	7	3	3	3	ND
	TOTAL	**15**	**14(15)**	**ND**	**ND**	**26(27)**	**14**	**15**	**13**	**ND**

*** Parenthesis indicates uncertain numbers of individuals in group / age-sex class.**

**** Number of individuals counted on March 1 rather than added numbers.**

***** Values for Rainbow 2 are approximate due to poor observational conditions.**

The sampling strategy that I used to determine animal follow times was nonrandom, with animals selected for observation based on the time of day. As noted above, animals were followed at least once in the morning and once in the afternoon per month. However, at 90 minutes, individual follows were shorter than the total amount of time available for observations in both the morning and afternoon periods. As such, follows were logged as occurring in the following general time slots: Early Morning (Dawn to 8:45), Late morning (start time 8:46-11:15), Early Afternoon (11:16-14:30) and Late Afternoon (14:31-Dusk). To ensure that all portions of the day were covered for each individual, observations conducted into four separate two-month periods from July to February (1: July-August, 2: September-October, 3: November-December, 4: January-February). If an animal was followed during the early morning for one month, it was followed for the late morning the next month. Likewise, if an animal was followed early in the afternoon for the first month, he or she was observed during the late afternoon in the second month. I utilized this sampling strategy in order to account for behavioral differences based on time of day, as animals tended to be more active during the early morning and late afternoon than they were during the late morning and early afternoon periods. As animals with dental impairment are hypothesized to be more active during periods of group inactivity, to assess this aspect of the study, it is important to cover all periods of the day for approximately the same amount of time for each animal (Millette et al., 2009; personal observations). A single set of observations was also conducted in March for a subsample of groups and animals. A full sample of observations was not possible for this month as the study ended on March 15th, approximately one half through a full observational cycle (see below).

Focal animals were observed during extended social group follows, where ideally each individual within a social group was followed before moving onto another social group. Social

groups were visited for focal animals ideally on a twice-monthly basis, except for in March I observed a subset of groups once. During each of these twice-monthly series of follows, I followed each focal animal one time. Study groups were followed using a rotating schedule, during which each animal was followed at least one time before moving onto the next group. Using this system, each group was observed at approximately 2 week intervals across the duration of the study period.

Focal Animal Behavioral Sampling Methods.

Activity Budget Data. I collected activity budget data for each individual using instantaneous focal animal sampling (Altmann, 1974). All data were collected using 90 minute focal follows, with each instantaneous observation occurring on a three-minute interval. The 90 minute length was selected due to difficulty identifying animals while in the trees, as extended amounts of time were required to identify individuals, precluding short follow lengths. Likewise, a longer follow length was not utilized as 90 minutes was the longest period I could observe animals before exhaustion affected the recording of data collected using all occurrences sampling (see below). A 90 minute follow with a three-minute interval was also effectively used during my Master's level work with BMSR lemurs, and adequately elucidated behavioral differences between individuals with and without tooth loss (Millette, 2007; Millette et al., 2009). Focal follows were dropped if the animal was out of view for more than three observation intervals, either for consecutive or non-consecutive intervals.

For each follow the following general data were recorded: individual ID, social group ID, individual sex, date, follow start time. On each three-minute interval, the following data were recorded: interval ID (e.g., 0:00, 0:03, 0:06 minutes into the follow), interval time (the time the

interval was recorded), behavior, food item type (e.g., fruit, leaves, young leaves, etc.), food item species, direction of behavior (for social behaviors), behavioral partner ID (for social behaviors), nearest neighbor ID, nearest neighbor distance (in meters), substrate usage, animal height above ground (in meters), nursing (for lactating females), infant contact and position (for females with infants). All behaviors were collected using an ethogram derived from that of Sauther (1992). A full ethogram of behaviors recorded is available in Appendix A. The behavior recorded was that displayed at the start of an interval timer beep, as the computer-based timers used (Time Left Free Edition, NesterSoft Inc., Etobicoke, Ontario, Canada and/or Free Countdown Timer, Comfort Software Group, Vancouver, British Columbia, Canada) sounded for a period of several seconds. Nearest neighbor distances were recorded for the closest weaned animal to the focal subject until the month of January, when infants had become comparatively independent. This was done so as to avoid female's infants being the most common nearest neighbor during the height of lactation, as this would preclude assessments of social relationships between adult individuals. From January onwards, infants were recorded as nearest neighbors, along with the nearest non-infant neighbor. Substrate was recorded using a quadrat system based on that of Sauther (1992). Height in the canopy was recorded using the nearest estimated meter below the individuals lowest point (e.g., 0 = 0-1 meters, 1 = 1-2 meters, etc.).

Feeding Data. I recorded all feeding bouts concurrently to instantaneous observations using all occurrences sampling. For each feeding bout, I recorded the following general information: focal animal ID, social group ID, focal animal sex, follow start time, bout start and end time, food type, food species. Feeding bout start time was recorded as soon as an animal began to feed on a given food item. Feeding bouts were ended after an animal stopped chewing a food item. While

this could be difficult to determine for food items eaten in quick succession (e.g., *Tarenna pruinosum* berries, *Salvadora angustifolia* fruit, small leaves, etc.), in such cases feeding bouts were typically ended if an animal was not observed to chew for a period of 5-10 seconds. For large, discrete food items (e.g., tamarind pods, Akaly fruit), feeding bouts typically reflected one food item.

In addition, for each feeding bout, I collected a variety of food processing data. These included the following: food processing position, use of licking behaviors, side of food processing, use of manual processing, use of previously opened foods, and use of pre-processed foods. Position on the tooth row used to process food items was recorded using an anterior-posterior system (mesial to C1/p2 = "Anterior;" distal to C1/p2 = "Posterior;" both anterior and posterior = "Both"). Licking was recorded if the animal was observed to repeatedly lick a food item with the tongue, while single licks were recorded if an animal did not repeatedly lick an item but used the tongue at least once to process a food item. The side of the mouth used to process a food item was also recorded (Right = only right side used, Left = only left side used or Both = both side used). Manual processing was recorded (Yes, No, or Possible) if the hands were used to processes and prepare a food item prior to ingestion. Manual processing does not include use of the hands to remove a food item from its source, but only the preparation of food for consumption (e.g., removing shells from a food item). Use of open food items was recorded (Yes, No, or Possible) if a food was clearly open prior to being processed and consumed by an animal. Use of pre-processed foods was recorded if a food eaten was clearly processed by another animal prior to ingestion by the focal animal.

Focal Animal UTM Locations.

I recorded each focal animal's approximate location using a Garmin eTrex VistaC GPS (Garmin International Inc., Olathe, Kansas, USA) unit immediately prior to and following the end of each follow. GPS data were recorded in the behavioral data recording sheet along with a waypoint name to index each point. Data points for dropped follows were not deleted as they are useful for identifying the range of each animal and social group. GPS points provided by this study represent approximate locations as signal quality was often reduced in forested areas, leading to reduced geospatial accuracy.

A map of group locations was produced by me from these data using ArcGIS 10.2 (ESRI Corp., Redlands, CA, USA). For each group, home ranges were plotted using the data points generated prior to and following each follow. These data points were then used to generate home range areas using the minimum convex hull function of the Minimum Bounding Geometry (Data Management) tool in ArcGIS. The resulting map is found above in Figure 2.1

Coat and Body Condition Data.

To assess the impact of dental impairment upon overall lemur health status, I collected visual measures of coat and body condition. For visual indicators of coat and body condition, I utilized methods modified from those of Berg et al. (2009; coat status, see also Jolly, 2009a,b), as well as Berman and Schwartz (1988; body size condition). For all health and coat indicators, data were collected for each individual on an ideally twice-monthly basis from July through February, although some individuals were infrequently assessed more than once per month, or only once monthly (see Millette et al., 2012 / Chapter 7).

Coat Condition Methods. I recorded coat and tail condition for all study animals using non-invasive, visual methods derived from those of Berg et al. (2009). Coat condition was determined using an ordinally-scaled system based on the quality and condition of the hair upon the back and limbs. Because the animal's ventral surface was often difficult to observe, and this area was not examined in previous studies of *L. catta* coat condition, I did not integrate ventral hair quality into the scoring system utilized in this study. Coat quality scores ranged from a state of excellent coat quality with no or very few imperfections (0-Good) to complete loss of the pelage (5-Bald). Scores 1 through 4 indicated intermediate coat quality. A score of 1 (Rough) indicated generally good fur condition with some unevenness to the coat. A score of 2 (Holes) indicated that more than three coin-sized holes were present in the coat, or that one or more larger sized holes (e.g., 4-5cm) were present. A coat condition of 3 (Rough) indicated that more than 25 percent, but less than 50 percent, of the coat was missing to half the fur's normal depth, while 4 (Sheared) indicated more than 50 percent of the fur was missing to half normal depth. For scores 1 and 2, one or two small holes (less than coin-sized) were permissible, as these are often occur due to interindividual agonism or sitting postures, and do not appear to be indicative of overall health status (Berg et al., 2009). For this study scores of 4-Sheared and 5-Bald were not observed, although all other coat conditions were recorded. A full listing of coat condition criteria and photos of each coat state are available in Table 2.3 and Figure 2.2.

In addition to ordinal coat scores, I also recorded alopecia status for all study animals. For this study, alopecia does not indicate the complete loss of body hair (e.g., Jolly, 2009a,b), rather it refers to any loss of hair from the animal's pelage. Alopecia was recorded using a Yes/No system where "Yes" was denoted if any hair was observed to be absent. If hair growth

Table 2.3. Coat Condition Scoring System Definitions (Based on Berg et al., 2009).

Coat Score	Defining Characteristics of Coat Condition Score
0 (Good)	**1.** Animal has excellent coat with no or few imperfections. **2.** Little to no hair creasing present. **3.** Hair is unidirectional in cranial-caudal direction. **4.** Little to no visible coat shagginess; coat is uniform. **5.** One or two small, coin-sized holes permissible.
1 (Rough)	**1.** Coat has slight to significant shagginess and/or unevenness. **2.** Creasing of the hair frequently present. **3.** Hair may have one or two small holes up to coin size. **4.** Score may be assigned when one larger hole present, but small sized (e.g. two or three coins).
2 (Holes)	**1.** Animal demonstrates > 2 coin-sized holes in hair. **2.** Holes are typically surrounded by rough hair. **3.** < 25% of hair missing in total. **4.** Hair may be partially grown back in the hole, but not predominantly filled w/ presence of hole still clear.
3 (Ragged)	**1.** Hair shows holes over >25% but <50% of body or limbs. **2.** Hair is < 1/2 normal length in affected areas.
4 (Sheared)	**1.** Hair less than half depth on >50% of body. **2.** **Not Observed at BMSR.**
5 (Bald)	**3.** More than 50% of hair on body or limbs absent. **4.** **Not Observed at BMSR.**

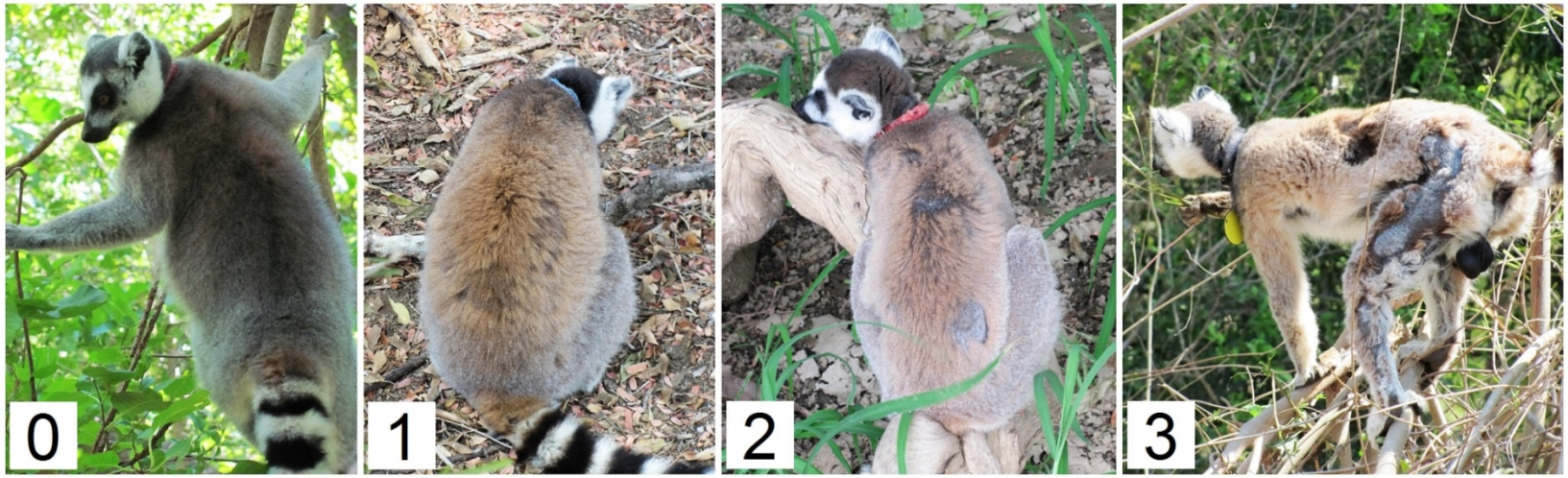


Figure 2.2. Coat condition scores for BMSR *L. catta*.. 0 = 0-Good, 1 = 1-Rough, 2 = 2-Holes, 3 = 3-Ragged. Individuals in images 2 and 3 demonstrate alopecia.

had occurred from where previously missing, alopecia was recorded if the missing patch was not yet predominantly filled in by new growth.

Body Condition Methods. I determined study animal body condition using visual methods derived from those of Berman and Schwartz (1988) which were developed for use in rhesus macaques (*Macaca mulatta*). While rhesus macaques were not observed during the construction of the system used by this study, observations by Pond and Pereira (1989) indicate that patterns of fat deposition are similar between *L. catta* and *M. mulatta.* As such, Berman and Schwartz's system is applicable to studies of *L. catta* such as the one presented here. All scores were also further refined for use in *L. catta* through field observations of body condition of animals of differing body mass during the completion of this study.

As with coat and tail condition scores, body condition was recorded using an ordinal system that rated animals on a scale from extremely thin (1) to obese (4). Although I did not measure body mass directly during this study, the scoring system used here represents a general indicator of body weight and/or body fat status. A score of 1 indicated that an animal had exposed skeletal features and extremely little body fat, while a score of 1.5 indicated that while an animal was visibly thin (e.g., exposed flanks and eye orbits) it did not have exposed skeletal features. Scores of 2 indicated that an animal was of average size and was lean with little excess fat, while a score of 2.5 indicated that an animal had excess amounts of fat with a "rounded" body. A score of 3 indicated that an animal demonstrated a large amount of fat, with bulging fat in the hips and/or stomach area. Scores of 3.5 and 4 were not observed, but were indicative of extremely high levels of fat and extremely high body mass. A full listing of body condition criteria and photos for each body state are available in Table 2.4 and Figure 2.3.

Table 2.4. Body Condition Scoring System Definitions.

Score	Defining Characteristics of Body Score
1	**1.** Animal is extremely thin. **2.** Underlying skeletal structure clearly visible. **3.** Score 1 was not observed during this study.
1.5	**1.** Animal shows low body mass and appears visibly thin. **2.** Body rail-like; hips are exposed with flanks depressed. **3.** Animal has taut skin with no excess fat. **4.** Eye orbits exaggerated and face appears thin. **5.** No bones or ribs visible through skin.
2	**1.** Animal is of average size, neither thin nor fat; animal is lean. **2.** No excess fat apparent with; animal is "sleek" in appearance. **3.** Face appears full; eye orbits do not protrude. **4.** Hips and flanks are not concave or only slightly so.
2.5	**1.** Animal of slightly high body mass than in score 2. **2.** Hips often rounded; fat often on hips and lower back. **3.** Body rounded; slight to moderate belly fat may be present. **4.** Face full; head may appear small in relation to body.
3	**1.** Animal is of high body mass. **2.** Girth > 2.5 w/ belly fat present; wide at hips and midsection. **3.** Sides bulge when sitting, fat may be present over legs. **4.** Head small in relation to body; animal is "light bulb" shape. **5.** Animal appears very full or "overstuffed."
3.5 - 4	**1.** Animal is of extremely high body mass. **2.** Extreme amounts of fat present. **3.** Scores 3.5 and 4 were not observed during this study.

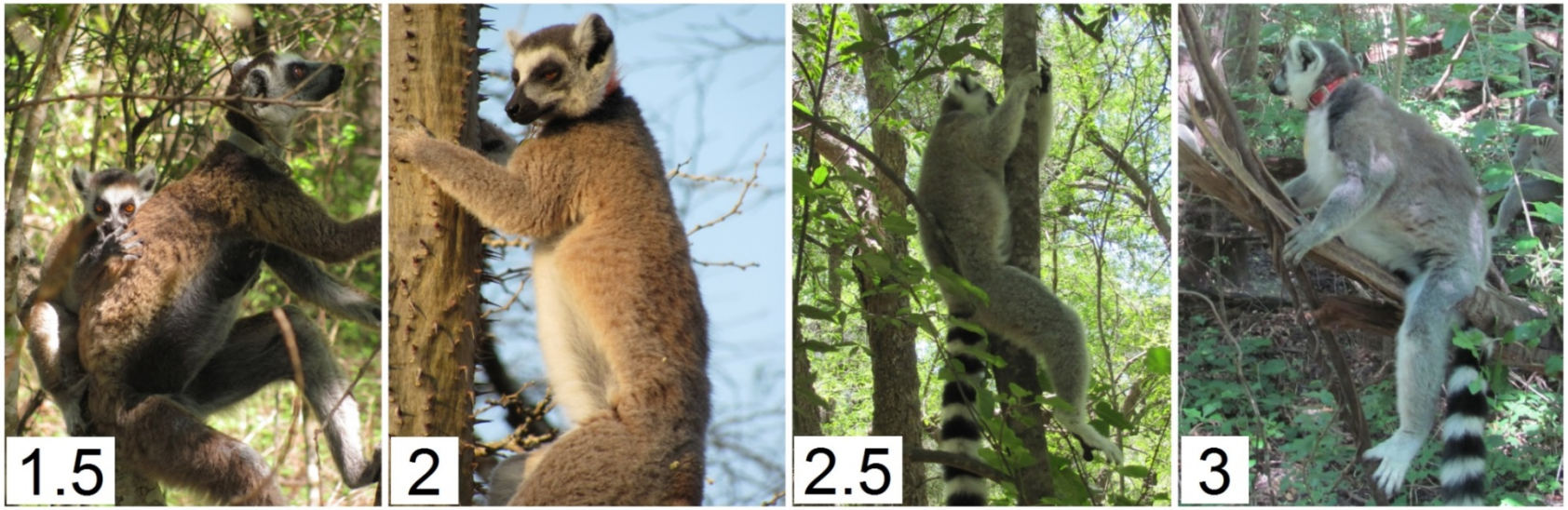


Figure 2.3. Body condition scores for BMSR *L. catta* 1.5 = Reduced body condition; 2 = Moderate body condition; 2.5 = Slightly high body condition; 3 = High body condition.

Photos of Coat and Body Condition. For each animal, I collected photographs of body and coat condition on a monthly basis. Tail condition was also recorded photographically as well, although this was recorded less frequently than coat and body condition. If indicators of poor health were observed, I also photographed these if possible. Photos were taken at the closest possible distance to the animal, preferably while the subject was on the ground. The majority of photos were taken at a range of less than 3 meters to the animal. Due to the coat condition method's focus upon the back and limbs, most photos were taken from the animal's rear and side aspects. Typically photos were taken on the same day as an animal was followed, and/or on which coat, body and health condition data were collected. If this was not possible (e.g., the animal was too far away to photograph adequately, it was too dark for photos, etc.), photos were taken as soon as possible, or at least before the end of the month during which coat and body data were taken.

Fecal Collection and Processing Methods.

Fecal samples were collected from June 2012 to March 2013 for use in fecal nutritional assessments (e.g., fat, protein and carbohydrate nutrient content, fiber content) conducted at the Dr. Matt Sponheimer's Nutritional and Isotopic Ecology Laboratory at the University of Colorado. Fecal samples were collected for each animal on a monthly basis. Although a minimum of one sample was collected for each individual per month, multiple samples were typically available for each individual during each monthly period. Multiple monthly samples are particularly available for individuals from larger study groups as the period of time spent with these groups during each month was longer than with smaller groups (behavioral observations

would take approximately 2-3 days for large groups such as Orange or Yellow, while smaller groups with one or two individuals would only be visited for a day or less at a time).

Fecal samples were collected primarily by Malagasy field assistants; although I collected fecal samples when possible. Most samples were collected by field assistants as it was extremely difficult to completely collect each fecal while conducting behavioral observations. From June to August, samples were collected independently by a student field assistant (Percy Yvon Rakoto of the University of Antananarivo). Later in the study, field assistants (either Naina Nicholas Rasolonjatovo of the University of Toliara, or members of the BMSR ecological monitoring team) typically collected samples while assisting with group follows. This change in procedure was done as the presence of a field assistant made finding and following groups and individuals much more efficient, and allowed for additional behavioral data to be collected.

All fecal samples were collected immediately after excretion from the animal and placed directly into an aluminum foil wrapper labeled with the animal's ID number. Fecal samples were then placed into a candle-powered camping oven where they were heated until dry the same day of collection. Although temperature was not recorded due to an unreliable oven thermometer, temperatures were typically low as small tea candles were used for heating. Samples were heated until thoroughly dried, typically requiring at least several hours of heating, and often extended periods of drying (e.g., overnight) during periods of high humidity and/or low ambient temperature.

Samples were removed from the oven once dried, and then placed into Nasco Whirl-Pak sample bags. Samples were also accompanied by a silica desiccant packet to ensure that no moisture was left within the sample, but also to absorb any atmospheric moisture during storage. Following preparation, samples were stored in buckets under ambient temperature in the

BMSR's on-site laboratory. Following return to the United States, all samples were stored in the Primate Biology Laboratory (Sauther Laboratory) of the University of Colorado at room-temperature. Photos of sample preparation are available in Figure 2.4.

Figure 2.4. Fecal sample preparation. Clockwise from top left: Samples were first collected in foil following excretion from the animal. The samples were then placed into a Coleman camp oven and then heated until dry using tea candles. Once fully dried (after heating for a period lasting several hours or to the next morning), all samples were placed into Whirl-Pak packets with silica desiccant.

Dental Examination Methods and Animal Captures.

Each study animal was captured in order to provide basic dental-morphological data and impressions necessary to produce high-resolution epoxy casts used for dental topographic

analysis. All animals were captured as a part of annual dental and general health examinations conducted during the field seasons (June-August) of 2011 and 2012 by Dr. Michelle Sauther and Dr. Frank Cuozzo. Individuals were captured using a Dan-Inject blow dart system (Dan-Inject, North America, Fort Collins, CO) and anesthetized using the drug Telazol (Fort Dodge Laboratories, Fort Dodge, IA). Doses were determined based on protocols developed over 20 years and over 400 safe captures of ring-tailed lemurs at BMSR (e.g., Sussman, 1991; Sauther et al., 2002, 2006; Cuozzo and Sauther, 2006a,b; Miller et al., 2007). Darting was conducted by a Malagasy field assistant with over 25 years of experience (Enafa, BMSR Ecological Monitoring Team), and all captures were conducted as early in the morning as possible to allow recovery for release the following morning. A trained veterinarian was on site to monitor the health of each captured lemur. All team members utilized protective masks and gloves to prevent disease transmission while handling lemurs, as is directed by the standards of the US CITES Management Authority. All capture protocols were approved by the Institutional Animal Care and Use Committees (IACUC) of the University of North Dakota and the University of Colorado, Boulder. All captures were also conducted with the approval of Madagascar National Parks, the institution governing research in Madagascar's protected areas.

During each capture, in addition to general health data (e.g., weight, body proportions, parasite load, etc.), a variety of dental-morphological data were collected. For each individual, the presence or absence of each tooth was recorded, while dental wear for each tooth position was assessed using a 0-5 ordinal scale (see Table 2.5). Likewise, the presence and position of any broken teeth was recorded, as were any other dental pathologies (e.g., caries, cracked teeth, excessive staining, etc.). All such assessments were conducted by Dr. Frank Cuozzo, who has

been conducting dental assessments for BMSR lemurs since the onset of the Bezà Mahafaly Lemur Biology Project in 2003.

Table 2.5. Gross Tooth Wear Ordinal Scale (from Millette, 2007; Derived From Cuozzo and Sauther, 2004 and 2006).

0 -- unworn occlusal surface.

1 -- small wear facets and no dentine or pulp exposure.

2 -- large wear facets and no dentine or pulp exposure.

3 -- some dentine and pulp exposure, few cusps still present; for canine and tooth comb, 1/2 remaining.

4 -- pulp exposure, with cusps gone, dentine or pulp exposed across most of the surface, or partial crown remaining; for canine and toothcomb, less than 1/4 remaining.

5 -- tooth worn to or below gum line with only roots/partial roots remaining (i.e., functional loss [Cuozzo and Sauther, 2004, 2006a]); OR no presence of the tooth remains (i.e., healed gingiva only, or in skeletal specimens remodeled alveoli).

In addition to dental-morphological data, for each study animal, dental impressions necessary to produce epoxy dental casts were collected. Dental impressions were produced for each of the dental arcade's four quadrants, as well as for the tooth comb. All impressions were made using President's Jet Plus Regular Body polyvinylsiloxane dental impression material (Coltène/Whaledent AG, Altstätten, Switzerland) and a custom dental impression tray. As with all field-based dental aspects of this project, all dental impressions were produced by Dr. Frank Cuozzo.

Dental Topographic Analysis Methods.

Dental Cast Production Methods. I produced graphite-teflon coated epoxy dental casts at the University of Arkansas Paleoanthropology Laboratory for each study animal. All casts were

manufactured from dental impressions collected from each individual during dental exams conducted at BMSR during 2011 and 2012 by Dr. Frank Cuozzo (see above). Casts were made for each quadrant of the dentition. For the upper dentition, all teeth were typically casted (I1-M3). Due to difficulties with producing accurate casts of the toothcomb, for the lower dentition, positions including and distal to the second premolar (p2-m3) were reproduced. The production of casts followed methods which have been utilized and validated by the Paleoanthropology Laboratory for laser-scanning-based GIS dental topographic analysis.

All impressions were first trimmed to remove excess material from their initial manufacture, taking care not to damage areas representing the occlusal surface of the teeth. Impressions were then sealed and externally coated using polyvinyl siloxane dental putty (President Soft Coltène/Whaledent AG, Altstätten, Switzerland) to prevent the escape of epoxy during cast production and to allow for centrifuging. Each impression was then partially filled (e.g., 50% filled) with EPO-TEK 301 epoxy (Epoxy Technology, Inc., Billerica MA, USA) that was dyed using a pigment-based colorant (to help prevent light penetration during subsequent laser scanning). Samples were then centrifuged for approximately two minutes to remove bubbles that commonly occur along the apex of cusps and crests. After centrifuging, epoxy was added until it filled the entire impression. The sample was then left to dry until fully hardened (typically two days).

Once casts were fully cured, they were removed from the impression material and then inspected for defects. If defects (e.g., bubbles, hairs, etc.) were found on the crown of any tooth, the cast was rejected and the casting procedure repeated for that sample. Samples with no apparent defects were then coated with a combination of graphite and Teflon to reduce penetration of the laser scanner into the epoxy material during laser scanning. Samples were first

coated in successive coats of graphite (Blaster Graphite Dry Lubricant, Blaster Group Pty LTD, Castle Hill, New South Wales, Australia) until fully black in color. These were then left to dry for at least one overnight period. Samples were then coated with a layer of Teflon (CRC Dry PTFE Lubricant, 16oz, CRC Industries America Group, Warminster PA, USA) until light grey in color. Samples were again allowed to cure until fully dry, at which they were ready for laser-scanning.

Laser-Scanning and GIS Dental Topographic Analysis Methods. GIS topographic analysis requires the generation of three-dimensional point clouds which are then converted into a 2.5D surface from which dental topographic measures (e.g., surface relief, slope and angularity) may be calculated. While a variety of methods may be used to collect such data (e.g., confocal microscopy, piezoelectric contact sensors, micro-cT scanning), in this study I utilized laser scanning to calculate *x-y-z* coordinates across the dental arcade of each individual (see Ungar and M'Kirera, 2003; M'Kirera and Ungar, 2003; Dennis et al., 2004; Bunn and Unger, 2009; Cuozzo et al., 2014).

Casts for all study animals were laser scanned at the Paleoanthropology Laboratory of the University of Arkansas during October-November of 2013. Samples were digitized into three-dimensional *x-y-z* point clouds using a XSM multi-sensor scanning system (Xystum Corp., Turino, Italy) with an integrated OTM3 laser head (Dr. Wolf and Beck GmbH, Wangen, German). As GIS dental topographic analysis only permits one z value for each *x-y* coordinate, samples were oriented so that the greatest occlusal area was presented to the laser scanner in order to maximize the amount of surface available for subsequent topographic assessment (see M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004; Bunn and Unger,

2009; Cuozzo et al., 2014). Point clouds were generated by collecting three-dimensional data where z elevations were recorded at an interval of 25µm along the x and y axes. Resulting point clouds were then processed as ASCII files in Digiline™ software (Xystum Corp.) and were then imported into ArcGIS 10.2 Geographic Information Systems (GIS) software with Spatial Analysis and 3D Analysis Extensions (ESRI Corp., Redlands, CA, USA). For the upper dentition all positions distal to the canine were scanned. The canine was not scanned due to its recurved nature, which precluded scanning as the laser (and also ArcGIS) may only model one *z* elevation for each x-y coordinate. Maxillary incisors were not scanned as they are of exceptionally small size and would be difficult model in GIS. All positions distal to the p2 of the maxilla were scanned, with the toothrow being omitted due to difficulties in the production of these teeth in casts.

Following scanning, I conducted dental topographic analysis for mandibular positions from the p4 to m3. The caniform second lower premolar (lp2) position was not assessed due to its recurved lingual-buccal morphology, as this typically masked the buccal aspect of the tooth along the gumline resulting in a vertical surface when modeled in three-dimensions, while p3 was not assessed due to difficulties in determining a consistent point for cropping. Methods used for topographic analysis were taken from those of Klukkert et al. (2012b) and developed from those used by the University of Arkansas Paleoanthropology Laboratory. Teeth were modeled by interpolating a Digital Elevation Model (DEM) from the point-cloud data produced during scanning and subsequent processing in Digiline software. This was done by using ArcMap's Inverse Distance Weighted surface tool (IDW) to produce a 2.5D surface (e.g., 1 *z*-value for each *x-y* coordinate). The toothrow was then also modeled using at triangulated area network (TIN), as well as using topographic lines (topographic line tool). Each tooth's occlusal surface was

cropped using methods outlined by Klukkert et al. (2012b). This was done in order to allow the assessment of each tooth's functional surface in isolation from the rest of the toothrow and from its non-functional marginal regions (e.g., along the sides of the tooth and along the gumline). The position at which each tooth's occlusal surface was cropped was defined by first locating the lowest point in the tooth's distal basin (e.g., the talonid). If wear had removed the talonid, the lowest point on the tooth on the occlusal surface was selected. Typically this was located along the distal edge of the remnants of the talonid basin, although occasionally the low point was located anteriorly along the mesial edge of the remnants of the trigonid. After the low point on the occlusal surface was located, all points below this were removed from the surface to be assessed. To ensure that only functional surfaces were included for each cropped tooth, I cropped all teeth manually using both TIN and topographic lines to delineate the tooth's form. As such, each tooth was easily visible using these methods and was readily separated from adjacent teeth, particularly when using topographic lines as a guide. Each cropped tooth was then visually assessed to ensure that the entire occlusal surface was included, and also that non-occlusal surfaces (e.g., areas representing the gums) and/or adjacent teeth were excluded from analysis. Teeth for which little or no occlusal surface was present (e.g., total tooth loss), were excluded from topographic analysis.

For each tooth, slope, relief and angularity were assessed using tools available in ArcMap 10.2. Here, "slope" refers to the average slope (in degrees from horizontal) across the occlusal aspect of the cropped tooth. Slope was assessed using the Slope tool found in the 3D Analyst extensions of ArcMap 10.2. In turn, "relief" refers to the relationship of the 3D area of the tooth divided by the 2D area of the tooth. This measure was determined by dividing the 3D area of the cropped tooth as calculated by ArcMap in relation to the in relation to the 2D area of the cropped

occlusal table (e.g., the *x-y* profile of the tooth at the point where cropped). Relief provides a measure of tooth volume above the occlusal plane and is generally regarded as an analog of the traditional morphometric measure of shearing quotient (SQ). Finally, topographic angularity was assessed for each tooth. Topographic angularity refers to the average change in slope across the tooth's occlusal surface (e.g., the derivative of the tooth's average slope). This was determined by taking the layer generated when slope was initially calculated and then applying the slope tool a second time to this surface. Angularity represents the overall "jaggedness" of the tooth, and is thought to induce multi-directional forces within food items, thus facilitating their breakdown. As angularity is thought to only decrease with extensive wear, it is also thought to represent a measure of dental senescence (M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; but see Bunn and Ungar, 2009; Klukkert et al., 2012a).

CHAPTER III:

GENERAL PATTERNS OF BMSR *LEMUR CATTA* FEEDING BEHAVIOR

In this chapter I provide background data related to general patterns of ring-tailed lemur feeding behavior across the completion of this study from July 2012 through March of 2013. These data are intended to serve as a baseline from which to comprehend the differential effects of tooth loss and increasing dental wear on food item choice and/or behaviors utilized in compensation for dental impairment. In this chapter, I first examine food choice among BMSR *Lemur catta* across the course of the study in terms of both species and food item type (e.g., leaves, insects, fruit, etc.). Food choice is also examined in terms of the prevalence of consumption for each food species, as well as the types of food (e.g., leaves, fruit, etc.) consumed overall. In addition, I also present food species and food type data in relation to season (wet vs. dry) as well as on monthly basis from July through March. For this chapter, the proportion of time spent feeding on a given food item, as well as food item type and species data, are drawn from approximately 920 hours of instantaneous focal animal scan samples (see Altmann, 1974) collected on 3-minute interval, 90-minute follows conducted least twice monthly for each study animal. Although I also collected feeding data using all occurrences sampling, these are not presented here as they were collected in a manner intended to provide information specifically on feeding bout length and are not well suited to examining total frequencies of feeding (e.g., the lengths of feeding bouts where the animal was ingesting or processing food immediately prior or subsequent to the follow were recorded, thus making calculation of total

feeding time relative to total time observed difficult). Such duration based data are, however, presented relative to topographic status in Chapter 4. In contrast, instantaneous measures provide a means for examining patterns of food type and species consumption where feeding time is standardized relative to total observed behavior. Similarly, with instantaneous measures the total proportion of time a single species and type of food was consumed is standardized relative to all other foods. A summary of all food species (in common and scientific nomenclature) and food types consumed by BMSR lemurs during this study is available in Table 3.1.

Consumption of Food Items by Species Across the Entire Study Period.

Previously characterized as opportunistic omnivores, ring-tailed lemurs have been observed to consume a wide variety of foods in terms of both species as well as food item type (e.g., fruit, leaves, insects etc.). Likewise, *L. catta* frequently consume multiple food item types from a single food species depending on availability and/or forest phenology (Sauther, 1992; 1998; Sauther et al., 1999; Simmen et al., 2006; Yamashita et al, 2015b; LaFleur and Sauther, 2015; Millette, personal observations). During this project I observed similar patterns of food consumption, with study animal diets consisting of foods drawn from a multitude taxa. From July 2012 through March of 2013, study subjects consumed 74 individually identifiable food taxa and/or distinct foods that could not be identified taxonomically (e.g., foods which not have a clear taxonomic affinity, such as soil). All foods were identified using the assistance of local botanists at BMSR, through use of a herbarium and associated botanical samples located in BMSR's on-site museum, or through identification using a herbarium collected by Sauther during her observations in 1987-88 (See Sauther, 1992). Please see Table 3.1 for complete listing of all foods consumed.

Table 3.1. Foods and Food Types Consumed by BMSR *Lemur catta* Across the Study.

Scientific Name	**Common Name**	**Family**	**Food Type***
Acacia bellula	Tratriotse	Mimosaceae	YL, LB, FB, FL
Acalyphya decaryana	Tainajajamena	Euphorbiaceae	YL, FB, FR
Achyranthus apera	Tsipotaky/ Tsipoteke	Amaranthaceae	ML
***Achyranthus* sp.**	Tsipotekala	Amaranthaceae	ML
Allaphyllus decaryi	Sarivomanga	Sapivdaceae	FL
Alluaudia procera	Fantsiolotse	Didiereaceae	FB, ML
Aloe vahontsoy	Vahontsoy	Liliaceae	YL, FB, FR
Antidesma petiolare	Voafogme / Voafogne	Euphorbiaceae	FR
Argemone mexicana	Fatiboy	Papaveraceae	ML, ST
Aristolochia bernieri	Totonga	Aristolochiaceae	ML
Azima tetracantha	Filofilo	Salvadoraceae	FB, FL, FR
***Bridelia* sp.**	Tsikidrakatse	Euphorbiaceae	FB, FR
Byttneria voulili	Sarihasy	Sterculiaceae	ML
Cadaba virgata	Ndriamainty	Capparidaceae	FR
***Calopixis* sp.**	Sakaoanakoho	Combretaceae	ST
***Capparis chrysameia / Acacia* sp.**	Roihavitse	?	ML
Carica papaya	Papaya	Caricaceae	OT-FR
Cedrelopsis grevei	Katrafay	Ptaerxylaceae	FB, YL, ML
Cissus bosseri	Lelatrandrake	Vitaceae	FR
Citrullus lanatus	Voamanga (Melon crop)	Cucurbitaceae	YL, ML, FR
***Citrus* sp.**	Orange (crop fruit)	Rutaceae	OT-FR
Clerodendrum* sp. *cf. emirnense	Forimbitike	Verbenaceae	FB, FL, ML
***Combretum* sp.**	Tamenake / Tamenaka	Combretaceae	ML
***Commelina* sp.**	Andranahake / Andranahaka	Commelinaceae	ML
Commicarpus commersonii	Beamena	Nyctaginaceae	FB, FL, YL
***Commicarpus* sp.**	Bea	Nyctaginaceae	FB, FL
Commicarpus sp.	Beandahy / Beandahiny	Nyctaginaceae	ML
Corallocarpus greveii?	Kisenendolo / Voamangandolo	?	ML
Coridia ainensis	Varo	Boraginaceae	ML
Crateva excelsa	Akaly	Capparidaceae	FB, FL, YL, FR
***Cucurbita* sp. (?)**	Kisene (Squash Crop)	Cucurbitaceae (?)	YL, ML
Dialium madagascariensis	Karembolamitsy / Karembulamitsy	Cesalpinaceae	ML, FR
Dichrostachys humbertii	Avoha	Mimosaceae	YL, LB
Diospyros sakalavarum	Kibaintsihotse	Ebenaceae	ML
Discorea fandra	Kanjike	Discoreaceae	ML
Discorea nako	Nako	Discoreaceae	ML
***Discorea* sp.**	Ovy	Discoreaceae	ML
Dombeya analavelonae	Satro	Sterculiaceae	ML
Ficus cocculifolia	Adabo	Moraceae	FR
Gonocrypta grevei	Kompitse	Asclepiadaceae	ML

Table 3.1(cont). Foods and Food Types Consumed by BMSR *Lemur catta* Across the Study

***Gouania* sp.**	Masokara	Rhamnaceae	ML
Grewia fransiscana	Tainkafotse	Tiliaceae	FR
Grewia grevei	Kotipoke	Tiliaceae	YL, FB, FR
Grewia leucophylla	Tratramborondreo	Tiliaceae	FB, FR
***Grewia* sp.**	Malimatse	Tiliaceae	FR
Gymnosporia linearis	Tsingilofilo	Celastraceae	ML
Gyrocarpus americanus	Kapaypoty / Kapaipoty	Hernandiaceae	FB, FL, YL, ML
***Hibiscus* sp.**	Tabuaralolo / Tabuarandolo	Malvaceae	ML, ST
***Ipomoea carica* (?)**	Teloravina	Convolvulaceae	ML
Ipomoea majungensis	Velae	Convolvulaceae	FB, FL, FR
***Ipomoea batatas* (?)**	Bageda	Convolvulaceae	ML, OT
Ipomoea carica	Valae?	Convolvulaceae	FL, ML
Kochneria madagascariensis	Pisopiso	Lythraceae	FR
***Landolphia* sp.**	Piravola	Apocynaceae	ML, ST
Maerua filiformis	Suomongy / Somangy	Capparidaceae	FB, FL, ML, FR
Maerua sp.	Sarysuomongy	Capparidaceae	LB, ST
***Mangifera* sp.**	Mango	Anacardiaceae	OT-FR
Metaporana parvifolia	Kililo	Convolvulaceae	YL, ML, ST
***Musa* sp.**	Akondro (Banana)	Musaceae	FR
***Olax* sp.**	Tanjake	Olacaceae	FR
***Pentopetio* sp.**	Tsompia	Apocynaceae	YL, ML, ST
Physena sessiliflora	Fandriandambo / Fandreandambo	Flacourtiaceae	FR
Quisivianthe papionae	Valiandro	Meliaceae	FB, FL, FR
Salvadora angustifolia	Sasavy	Salvadoraceae	FB, FL, YL, ML, FR, ST, SO
Scutia murtina	Roiombilahy	Rhamnaceae	FR
***Secamone* sp.**	Angalora	Asclepiadaceae	YL, ML, ST
Seyrigia gracilis	Tsiridambo	Cucurbitaceae	FB, ST
Strychnos madagascariensis	Bakoa	Loganiaceae	FR
Talinella dauphinensis	Dango	Portulacaceae	LB, YL, ML
Tamarindus indica	Kily (Tamarind)	Cesalpinaceae	FB, FL, LB, YL, LB, FR
Tamelapsis linearis	Tamburue / Tamboro	Asclepiadaceae	YL, ML, FR
Tarenna) pruinosum	Mantsake	Rubiaceae	YL, FR
Tragia tiverneana	Sanatry	Euphorbiaceae	YL, ML, ST
Undetermined	Atratra	?	FL
Undetermined	Fale	?	FR
Undetermined	Fatikakoho	?	ML
Undetermined	Kidresy	?	ML
Undetermined	Lobakahjirike	?	FL
Undetermined	Mamyaho (Liana)	?	FB, FL
Undetermined	Sakarvironala / Sakaviro n'ala	?	ML
Undetermined	Saritabuara / Sarytabuara	?	YL, ML

* LB = Leaf Buds, YL = Young Leaves, ML = Mature Leaves, FB = Flower Buds, FL = Flowers, FR = Fruit, OT = Other, SO = Soil, ST = Stems

Of foods consumed by BMSR *L. catta*, 47 were identifiable to the species level, while an additional 13 foods were identifiable to the genus level. It is also of note that in at least one case several species were condensed into a single generic identifier (*Commicarpus* sp.; Malagasy names: bea, beandhiny, and beamena) as these were difficult to distinguish during field observations, and incorrect species-level identification was likely. A further 14 foods were singularly identified by their local Malagasy or English common names, but could not be identified to their generic or species names. Of these, several foods were agricultural products utilized by the lemurs and were thus identified by their common names (e.g., mango, voamanga melon, kisene squash, bageda sweet potato). Likewise, a number of singularly identifiable foods could not readily be classified by genus, species or Malagasy name (e.g., wood, feces, songbird eggs, insect species, soil, etc.), and are referred to by their common names in English. Foods which could not be identified either during or after consumption were classified as "unknown." Typically foods characterized as "unknown" were classified as such because the food item was either consumed quickly under poor observational conditions and/or no sample could be secured for subsequent identification (e.g., the animal consumed all leaves or fruit necessary for identification). The majority of food items classified as "unknown" are likely included within those species which could be identified, and only infrequently represent food species which could not be identified to at least some extent across the course of observations.

Three species accounted for the majority (58.76%) of foods consumed across the study (Figure 3.1, Table 3.2). These include *Tamarindus indica* ("tamarind", 28.87%), *Salvadora angustifolia* ("sasavy", 18.64%) and *Metaporana parvifolia* ("kililo", 11.25%). A total of 19 foods accounted for greater or equal to 1% of total feeding observed across the study. These include the following, identifiable to either the generic or species level: *Quisivianthe papionae*

("valiandro", 3.15%), *Acacia bellula* ("tratriotse", 3.05%), *Pentopetio* sp. ("tsompia", 2.41%), *Ipomoea majungensis* ("valae", 1.99%), *Tarenna pruinosum* ("mantsake", 1.93%), *Cedrelopsis grevei* ("katrafay", 1.41%), *Gyrocarpus americanus* ("kapaypoty", 1.41%), *Grewia leucophylla* ("tratramborondreo", 1.22%), S*ecamone* sp. ("angalora", 1.16%), *Talinella dauphinensis* ("dango", 1.09%)), *Bridelia* sp. ("tisikidrakatse", 1.06%), and *Crateva excelsa* ("akaly", 1.03%). Other foods accounting for ≥1% of time spent feeding, but which could not be identified taxonomically, included: wood (2.57%), caterpillars (1.22%) and soil (1.00%). Across the entire study, foods characterized as "unknown" accounted for 6.46% of total feeding observations.

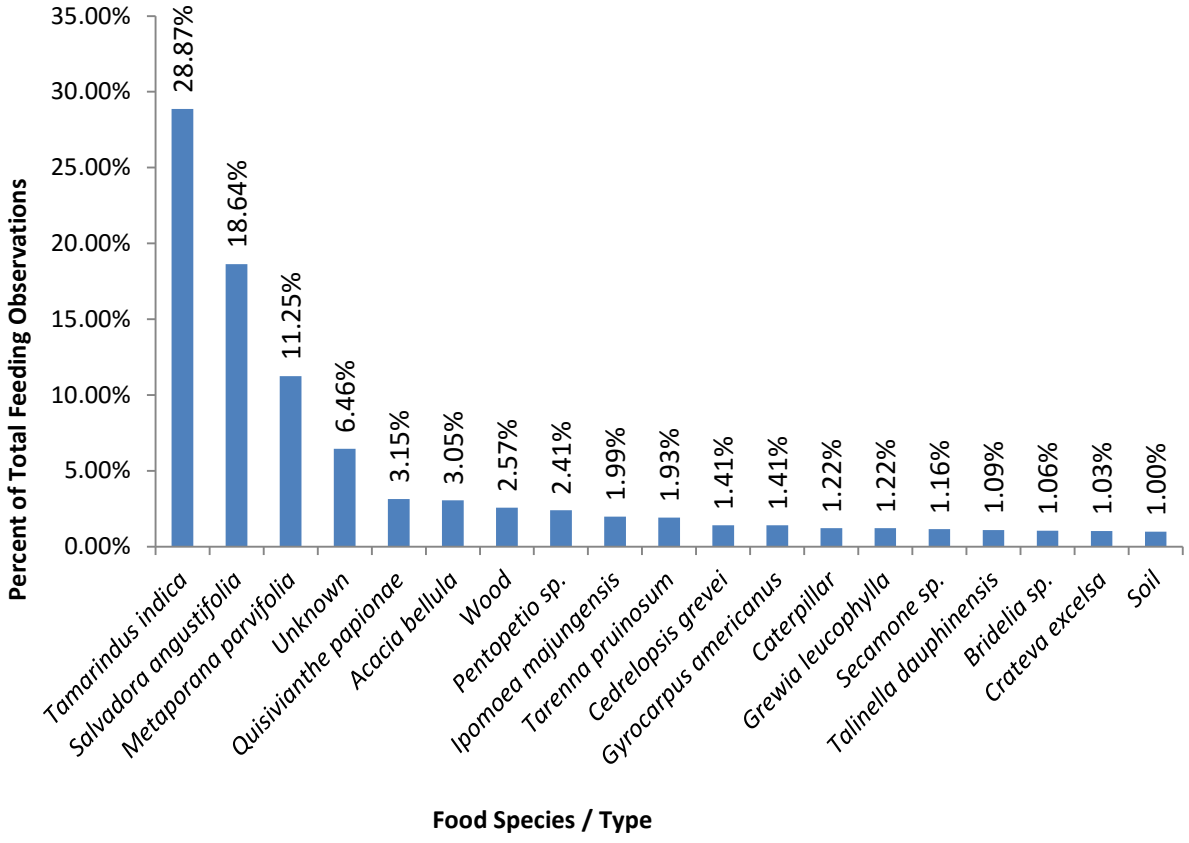


Figure 3.1. Percent of total feeding observed for all food species and types representing greater than or equal to one percent of total feeding across the study period (July 2012 – March 2013).

Table 3.2. Food Species and Food Items by Consumption Rank.

#	Species / Food item	# of Obs	% Feeding	#	Species / Food item	# of Obs	% Feeding
1	*Tamarindus indica*	898	28.87%	**39**	*Hibiscus* sp.	4	0.13%
2	*Salvadora angustifolia*	580	18.64%	**40**	Papaya	4	0.13%
3	*Metaporana parvifolia*	350	11.25%	**41**	Stone	4	0.13%
4	Unknown	201	6.46%	**42**	*Achyranthus apera*	3	0.10%
5	*Quisivianthe papionae*	98	3.15%	**43**	*Ficus cocculifolia*	3	0.10%
6	*Acacia bellula*	95	3.05%	**44**	Tomato	3	0.10%
7	Wood	80	2.57%	**45**	*Acalyphya decaryana*	2	0.06%
8	*Pentopetio* sp.	75	2.41%	**46**	*Achyranthus* sp.	2	0.06%
9	*Ipomoea majungensis*	62	1.99%	**47**	*Ipomoea carica*	2	0.06%
10	*Tarenna pruinosum*	60	1.93%	**48**	Kisene	2	0.06%
11	*Cedrelopsis grevei*	44	1.41%	**49**	Melon	2	0.06%
12	*Gyrocarpus americanus*	44	1.41%	**50**	Sakavironala	2	0.06%
13	Caterpillar	38	1.22%	**51**	Sarytabuara	2	0.06%
14	*Grewia leucophylla*	38	1.22%	**52**	Soil/Wood	2	0.06%
15	*Secamone* sp.	36	1.16%	**53**	*Alluaudia procera*	1	0.03%
16	*Talinella dauphinensis*	34	1.09%	**54**	*Aloe vahontsoy*	1	0.03%
17	*Bridelia* sp.	33	1.06%	**55**	*Antidesma petiolare*	1	0.03%
18	*Crateva excelsa*	32	1.03%	**56**	*Byttneria voulili*	1	0.03%
19	Soil	31	1.00%	**57**	*Cissus bosseri*	1	0.03%
20	Voamanga	26	0.84%	**58**	*Combretum* sp.	1	0.03%
21	Mamyaho	25	0.80%	**59**	*Diospyros sakalavarum*	1	0.03%
22	Mango	21	0.68%	**60**	*Discorea fandra*	1	0.03%
23	*Coridia ainensis*	19	0.61%	**61**	*Dombeya analavelonae*	1	0.03%
24	Feces	18	0.58%	**62**	Egg / Bird Nest	1	0.03%
25	Concrete	14	0.45%	**63**	Fale	1	0.03%
26	*Azima tetracantha*	9	0.29%	**64**	*Gouania* sp.	1	0.03%
27	*Capparis chrysameia / Acacia*	9	0.29%	**65**	*Grewia* sp.	1	0.03%
28	*Commicarpus* sp.	9	0.29%	**66**	*Gymnosporia linearis*	1	0.03%
29	*Maerua filiformis*	9	0.29%	**67**	Kisene and/or voamanga	1	0.03%
30	*Tamelapsis linearis*	9	0.29%	**68**	*Landolphia* sp.	1	0.03%
31	*Argemone mexicana*	8	0.26%	**69**	*Lobakahjirike*	1	0.03%
32	*Dialium madagascariensis*	8	0.26%	**70**	*Maerua* sp.	1	0.03%
33	*Grewia grevei*	8	0.26%	**71**	*Olax* sp.	1	0.03%
34	Cicada	7	0.23%	**72**	Orange	1	0.03%
35	*Discorea* sp.	7	0.23%	**73**	*Physena sessiliflora*	1	0.03%
36	*Seyrigia gracilis*	7	0.23%	**74**	Spider Nest	1	0.03%
37	Bageda	5	0.16%	**75**	*Strychnos madagascariensis*	1	0.03%
38	*Clerodendrum* sp. cf. *emirnense*	4	0.13%		**Grand Total**	**3111**	**100.00%**

Those 19 foods individually accounting for 1% or more of total feeding represent 90.94% of all feeding observations recorded. As a result, the remaining 56 recorded food species / items account for only 9.06% of intervals for where feeding was observed. These data indicate that ring-tailed lemurs frequently consumed variety of food items which do not contribute significantly to the overall diet across the course of this study. This finding is consistent with previous observations that BMSR ring-tailed lemurs often consume a wide variety of foods across the year, but often focus on specific foods depending on availability as related to seasonality and local phenology (see Sauther, 1998). While a majority of food items only account for a small proportion of total feeding observations, for a number taxa, the prevalence of consumption increases dramatically for specific points during the year and may account for a significant proportion of the diet at a given time (see below).

Consumption of Food Items by Species in Relation to Season.

For this study I separated the dry and wet seasons between the months of October and November, as significant rains commenced towards the end of October and occurred frequently (e.g., daily, or near-daily) following the start of November. The dry season is thus defined as occurring from the start of observations in July through the end of October. The wet season is defined as occurring from the start of November through the end of the study in March, when rains were beginning to subside. Similar dates for these seasonal patterns of rainfall at BMSR conform to those previously by other researchers (see Sauther, 1998; Ratsirarson et al., 2003). While I did not record direct phenological data during this study, in general the dry season was defined by reduced food availability, while overall food availability appeared to increase following the onset of rains. This is particularly true for the availability of leaf-bearing food taxa,

which became more prevalent during the wet season in contrast to the dry season during which the deciduous forest demonstrates reduced amounts of foliage.

For the dry season, BMSR *L. catta* consumed 43 species or otherwise singularly identifiable food items (Table 3.3). Of these, 12 species and items accounted for more than one percent of all feeding observations. As with total observations, *Tamarindus indica* and *Salvadora angustifolia* represented the top two major food items consumed, accounting for 35.91% and 16.55% of total feeding observations respectively. Unknown foods accounted for the third largest food group, representing 6.91% of feeding bouts during this period. During the dry season, many of these foods were truly unidentifiable as the animals frequently consumed detritus on the ground, and which appeared to consist of dried leaves, unknown dried fruits and/or potential feces. Other key species consumed during this period included: *Quisivianthe papionae* ("valiandro", 6.10%), *Acacia bellula* ("tratriotse", 5.91%), *Metaporana parvifolia* ("kililo", 4.48%), *Ipomoea majungensis* ("valae", 3.86%) *Tarenna pruinosum* ("mantsake", 3.67%) *Cedrelopsis grevei* ("katrafay", 1.93%), and *Cordia ainensis* ("varo", 1.18%). Wood and soil accounted for 2.55% and 1.18% of feeding bouts respectively. Overall, species accounting for one or more percent of feeding represented 92.97% of dry season feeding observations, while the remaining 31 species consumed accounted for 7.03% of feeding.

During the wet season, study subjects consumed 54 species or singularly identifiable food items (Table 3.4). As with the dry season, and for total observations, *Tamarindus indica* ("kily") and *Salvadora angustifolia* ("sasavy") accounted for the two top food species, representing 21.34% and 20.88% of foods consumed respectively. As with the total feeding

Table 3.3. Food Species / Singularly Identifiable Items Consumed During the Dry Season.

#	Species / Food Item	# Dry Obs	% Dry Obs	#	Species / Food Item	# Dry Obs	% Dry Obs
1	*Tamarindus indica*	577	35.91%	**23**	Mango	3	0.19%
2	*Salvadora angustifolia*	266	16.55%	**24**	*Azima tetracantha*	3	0.19%
3	Unknown	111	6.91%	**25**	*Grewia grevei*	3	0.19%
4	*Quisivianthe papionae*	98	6.10%	**26**	*Seyrigia gracilis*	3	0.19%
5	*Acacia bellula*	95	5.91%	**27**	Papaya	3	0.19%
6	*Metaporana parvifolia*	72	4.48%	**28**	Stone	3	0.19%
7	*Ipomoea majungensis*	62	3.86%	**29**	*Ficus cocculifolia*	3	0.19%
8	*Tarenna pruinosum*	59	3.67%	**30**	Tomato	3	0.19%
9	*Gyrocarpus americanus*	44	2.74%	**31**	*Crateva excelsa*	2	0.12%
10	Wood	41	2.55%	**32**	*Achyranthus* sp.	2	0.12%
11	*Cedrelopsis grevei*	31	1.93%	**33**	*Ipomoea carica*	2	0.12%
12	Soil	19	1.18%	**34**	*Talinella dauphinensis*	1	0.06%
13	*Coridia ainensis*	19	1.18%	**35**	*Commicarpus* sp.	1	0.06%
14	Feces	15	0.93%	**36**	*Tamelapsis linearis*	1	0.06%
15	*Secamone sp.*	10	0.62%	**37**	*Alluaudia procera*	1	0.06%
16	Concrete	9	0.56%	**38**	*Byttneria voulili*	1	0.06%
17	*Maerua filiformis*	9	0.56%	**39**	*Combretum* sp.	1	0.06%
18	*Argemone mexicana*	8	0.50%	**40**	*Diospyros sakalavarum*	1	0.06%
19	*Capparis chrysameia / Acacia* sp.	6	0.37%	**41**	*Gymnosporia linearis*	1	0.06%
20	*Pentopetio* sp.	5	0.31%	**42**	*Maerua* sp.	1	0.06%
21	Voamanga	5	0.31%	**43**	Orange	1	0.06%
22	Bageda	5	0.31%	**44**	Spider Nest	1	0.06%
					Grand Total	**1607**	**100%**

Table 3.4. Food Species / Singularly Identifiable Items Consumed During the Wet Season.

#	Species / Food Item	# Wet Obs	% Wet Obs	#	Species / Food Item	# Wet Obs	% Wet Obs
1	*Tamarindus indica*	321	21.34%	**29**	Feces	3	0.20%
2	*Salvadora angustifolia*	314	20.88%	**30**	*Capparis chrysameia / Acacia* sp.	3	0.20%
3	*Metaporana parvifolia*	278	18.48%	**31**	*Achyranthus apera*	3	0.20%
4	Unknown	90	5.98%	**32**	*Acalyphya decaryana*	2	0.13%
5	*Pentopetio* sp.	70	4.65%	**33**	Kisene	2	0.13%
6	Wood	39	2.59%	**34**	Melon	2	0.13%
7	Caterpillar	38	2.53%	**35**	Sakavironala	2	0.13%
8	*Grewia leucophylla*	38	2.53%	**36**	Sarytabuara	2	0.13%
9	*Talinella dauphinensis*	33	2.19%	**37**	Soil/Wood	2	0.13%
10	*Bridelia* sp.	33	2.19%	**38**	*Tarenna pruinosum*	1	0.07%
11	*Crateva excelsa*	30	1.99%	**39**	Papaya	1	0.07%
12	*Secamone sp.*	26	1.73%	**40**	Stone	1	0.07%
13	Mamyaho	25	1.66%	**41**	*Aloe vahontsoy*	1	0.07%
14	Voamanga	21	1.40%	**42**	*Antidesma petiolare*	1	0.07%
15	Mango	18	1.20%	**43**	*Cissus bosseri*	1	0.07%
16	*Cedrelopsis grevei*	13	0.86%	**44**	*Discorea fandra*	1	0.07%
17	Soil	12	0.80%	**45**	*Dombeya analavelonae*	1	0.07%
18	*Commicarpus* sp.	8	0.53%	**46**	Egg / Bird Nest	1	0.07%
19	*Tamelapsis linearis*	8	0.53%	**47**	Fale	1	0.07%
20	*Dialium madagascariensis*	8	0.53%	**48**	*Gouania* sp.	1	0.07%
21	Cicada	7	0.47%	**49**	*Grewia* sp.	1	0.07%
22	*Discorea* sp.	7	0.47%	**50**	Kisene and/or voamanga	1	0.07%
23	*Azima tetracantha*	6	0.40%	**51**	*Landolphia* sp.	1	0.07%
24	Concrete	5	0.33%	**52**	Lobakahjirike	1	0.07%
25	*Grewia grevei*	5	0.33%	**53**	*Olax* sp.	1	0.07%
26	*Seyrigia gracilis*	4	0.27%	**54**	*Physena sessiliflora*	1	0.07%
27	*Clerodendrum* sp. cf. *emirnense*	4	0.27%	**55**	*Strychnos madagascariensis*	1	0.07%
28	*Hibiscus* sp.	4	0.27%		**Grand Total**	**1504**	**100%**

across the study, *Metaporana parvifolia* (“kililo”) was the third most common food item, accounting for 18.48% of wet-season feeding. Fifteen foods accounted for more than one percent of total feeding observations. These include: *Pentopetio* sp. (“tsompia”, 5.98%), *Grewia leucophylla* (“tratramborondreo”, 2.53%), *Talinella dauphinensis* (“dango”, 2.19%), *Bridelia* sp. (“tsikidrakatse”, 2.19%), *Crateva excelsa* (“akaly”, 1.99%), *Secamone* sp. (“angalora”, 1.73%), Mamyaho (a taxonomically unidentified liana, 1.66%), voamanga melon (1.40%), and mango (1.20%). Unknown food species accounted for 5.98% of the diet, while wood and caterpillars accounted for 2.59% and 2.53% of foods consumed respectively.

As noted above, the proportion of each food item consumed during the dry and wet season varies by food species and type. This includes food items which were consumed in significant quantities during both wet and dry seasons. For example, while tamarind represented 35.91% of food items consumed during the dry season, tamarind-based foods accounted for only 21.34% of the diet during the wet season. Likewise, while accounting for only about 4% of the total diet during the dry season, *Metaporana parvifolia* accounted for over 18% of the diet during the wet season. Some major food items were only consumed during the wet or dry seasons. For example, *Quisivianthe papionae* and *Acacia bellula* were only consumed during the dry season, while the vast majority of *Pentopetio* sp. feeding observations were recorded during the wet season. Seasonal differences in the frequency of consumption for the top 10 most-consumed food items across the study period are presented in Figure 3.2. In addition, the types of food consumed from each species (e.g., leaves, fruit, etc.) vary by study month and season. For example, while tamarind fruit was commonly consumed at the study’s outset, tamarind leaves were commonly consumed during immediately prior to the onset of the dry season. Such variability in the type of food derived from each species is described below.

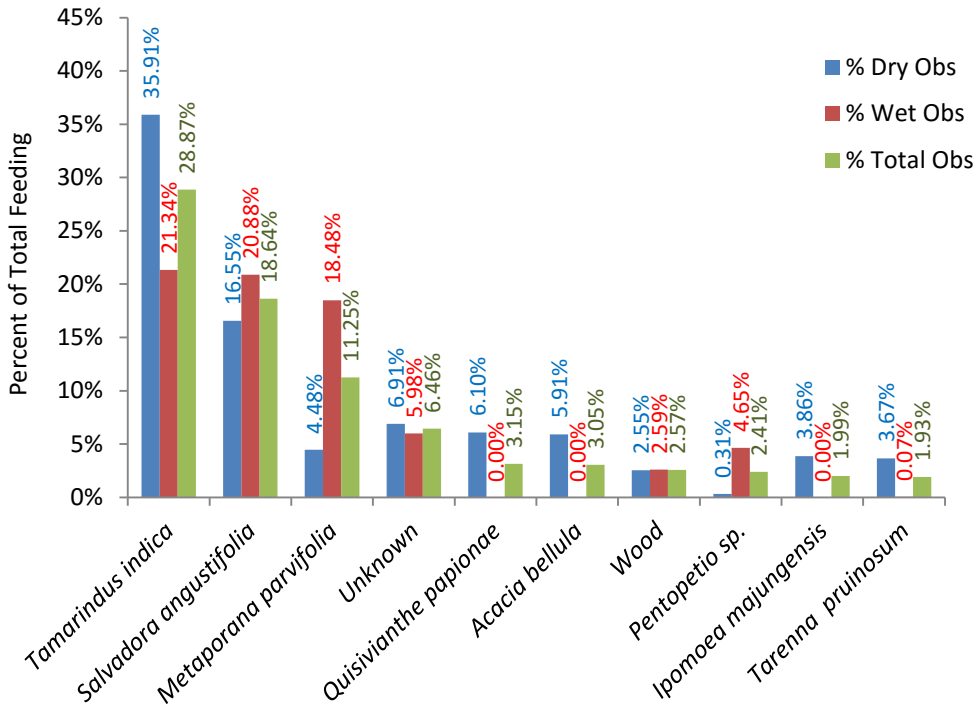


Figure 3.2. The percentage of feeding behavior recorded for the top 10 food species / item as consumed across the study period in relation to the dry season (July-October), wet season (November-March) and total feeding across the study.

Major Food Species by Month.

Although the main food items consumed by BMSR *Lemur catta* were represented by a few species (particularly *Tamarindus indica*, *Salvadora angustifolia* and *Metaporana parvifolia*), many of the species representing a small portion of the total diet were consumed at a higher rates during specific windows throughout the year. These spikes in consumption typically followed higher availability related to these species phenology. Such patterns of food item consumption have been previously described for BMSR ring-tailed lemurs and are not unique to this study (see Sauther 1992, 1998; Simmen et al., 2006).

Due to the large number of species consumed across the study, a full description of monthly variation in foods cannot be reported in this chapter. A full listing of species consumed

for each month is, however, available in Tables 3.5 and 3.6, where the monthly number of feeding observations and proportion of each food consumed is listed. However, it is possible to distinguish a number of patterns of feeding for these minor, but regularly consumed (e.g., >5% of observations), food items across the months of this study.

During July, *Tarenna pruinosum* ("mantsake") berries formed a major food item at 9.54% of feeding, but dropped to 2.35% of feeding the following month. In contrast, during August leaves of *Acacia bellula* accounted for 11.47% of feeding, while *Quisivianthe papionae* ("valiandro") flowers accounted for 8.53%. These species remained a major food during September with *Q. papionae* being the second most commonly consumed food at 14.57% of feeding observations and *A. bellula* accounting for 11.30%. In turn, *Cedrelopsis grevei* (10.2%) and *Gyrocarpus americanus* (7.24%) replaced these foods during October (Table 3.5).

Following the onset of the wet season, *Salvadora angustifolia* represented the dominant food species consumed at 68.78% of the diet with only three other species representing more than 5% of feeding observations (Tamarind: 7.69%; *Metaporana parvifolia*: 6.09%; *Talinella dauphinensis*: 5.58%). During December insects in the form of caterpillars accounted for the second most common food item (11.37%; Rank 1 = tamarind: 29.43%), although a number of minor plant species were also consumed regularly (*Pentopetio* sp.: 5.69%; *Crateva excelsa*: 5.02%). During January *Metaporana parvifolia* leaves (35.75%) and tamarind (27.42% formed the majority of the diet, while all other species accounted for less than 5% of feeding observations. During February genus *Bridelia* was consumed regularly (7.99%). In March this species accounted for 5.26% of feeding, while *Pentopetio* represented 6.58% of feeding (Table 3.6).

Table 3.5. Foods Consumed During the Dry Season (July-October) by Month.

Food Item	July		August		September		October	
	n =	% Total	n =	% Total	n =	% Total	n =	% Total
Acacia bellula	4	0.80%	39	11.47%	52	11.30%	0	0.00%
***Achyranthus* sp.**	0	0.00%	2	0.59%	0	0.00%	0	0.00%
Alluaudia procera	0	0.00%	0	0.00%	1	0.22%	0	0.00%
Argemone mexicana	6	1.19%	0	0.00%	2	0.43%	0	0.00%
Azima tetracantha	0	0.00%	0	0.00%	1	0.22%	2	0.66%
Bageda	1	0.20%	4	1.18%	0	0.00%	0	0.00%
Byttneria voulili	1	0.20%	0	0.00%	0	0.00%	0	0.00%
***Capparis chrysameia / Acacia* sp.**	4	0.80%	2	0.59%	0	0.00%	0	0.00%
Cedrelopsis grevei	0	0.00%	0	0.00%	0	0.00%	31	10.20%
***Combretum* sp.**	0	0.00%	1	0.29%	0	0.00%	0	0.00%
***Commicarpus* sp.**	0	0.00%	0	0.00%	1	0.22%	0	0.00%
Concrete	7	1.39%	1	0.29%	0	0.00%	1	0.33%
Coridia ainensis	17	3.38%	0	0.00%	2	0.43%	0	0.00%
Crateva excelsa	0	0.00%	0	0.00%	1	0.22%	1	0.33%
Diospyros sakalavarum	0	0.00%	0	0.00%	1	0.22%	0	0.00%
Feces	6	1.19%	1	0.29%	8	1.74%	0	0.00%
Ficus cocculifolia	3	0.60%	0	0.00%	0	0.00%	0	0.00%
Grewia grevei	3	0.60%	0	0.00%	0	0.00%	0	0.00%
Gymnosporia linearis	0	0.00%	0	0.00%	0	0.00%	1	0.33%
Gyrocarpus americanus	0	0.00%	2	0.59%	20	4.35%	22	7.24%
Ipomoea majungensis	2	0.40%	17	5.00%	31	6.74%	12	3.95%
Ipomoea carica	1	0.20%	1	0.29%	0	0.00%	0	0.00%
Maerua filiformis	3	0.60%	1	0.29%	4	0.87%	1	0.33%
***Maerua* sp.**	0	0.00%	0	0.00%	1	0.22%	0	0.00%
Mango	0	0.00%	0	0.00%	0	0.00%	3	0.99%
Metaporana parvifolia	54	10.74%	9	2.65%	6	1.30%	3	0.99%
Orange	0	0.00%	1	0.29%	0	0.00%	0	0.00%
Papaya	1	0.20%	2	0.59%	0	0.00%	0	0.00%
***Pentopetio* sp.**	0	0.00%	0	0.00%	0	0.00%	5	1.64%
Quisivianthe papionae	0	0.00%	29	8.53%	67	14.57%	2	0.66%
Salvadora angustifolia	118	23.46%	37	10.88%	52	11.30%	59	19.41%
***Secamone* sp.**	2	0.40%	7	2.06%	1	0.22%	0	0.00%
Seyrigia gracilis	0	0.00%	0	0.00%	3	0.65%	0	0.00%
Soil	11	2.19%	3	0.88%	5	1.09%	0	0.00%
Spider Nest	0	0.00%	1	0.29%	0	0.00%	0	0.00%
Stone	0	0.00%	0	0.00%	2	0.43%	1	0.33%
Talinella dauphinensis	0	0.00%	0	0.00%	0	0.00%	1	0.33%
Tamarindus indica	161	32.01%	128	37.65%	159	34.57%	129	42.43%
Tamelapsis linearis	1	0.20%	0	0.00%	0	0.00%	0	0.00%
Tarenna pruinosum	48	9.54%	8	2.35%	3	0.65%	0	0.00%
Tomato	0	0.00%	2	0.59%	1	0.22%	0	0.00%
Unknown	28	5.57%	37	10.88%	32	6.96%	14	4.61%
Voamanga	0	0.00%	5	1.47%	0	0.00%	0	0.00%
Wood	21	4.17%	0	0.00%	4	0.87%	16	5.26%
Grand Total	**503**	**100.00%**	**340**	**100.00%**	**460**	**100.00%**	**304**	**100.00%**

Table 3.6. Foods Consumed during the Wet Season (November-March) by Month.

	November		December		January		February		March	
Food Type	**n =**	**% Total**	**n =**	**% Total**	**n =**	**% Total**	**n =**	**% Total**	**n =**	**% Total**
Acalyphya decaryana	2	0.51%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Achyranthus apera	0	0.00%	1	0.33%	1	0.27%	1	0.28%	0	0.00%
Aloe vahontsoy	0	0.00%	0	0.00%	0	0.00%	1	0.28%	0	0.00%
Antidesma petiolare	0	0.00%	0	0.00%	0	0.00%	1	0.28%	0	0.00%
Azima tetracantha	2	0.51%	4	1.34%	0	0.00%	0	0.00%	0	0.00%
***Bridelia* sp.**	0	0.00%	0	0.00%	0	0.00%	29	7.99%	4	5.26%
***Capparis chrysameia / Acacia* sp.**	3	0.76%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Caterpillar	1	0.25%	34	11.37%	0	0.00%	3	0.83%	0	0.00%
Cedrelopsis grevei	7	1.78%	2	0.67%	0	0.00%	4	1.10%	0	0.00%
Cicada	7	1.78%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Cissus bosseri	0	0.00%	1	0.33%	0	0.00%	0	0.00%	0	0.00%
Clerodendrum* sp. cf. *emirnense	0	0.00%	0	0.00%	2	0.54%	2	0.55%	0	0.00%
***Commicarpus* sp.**	1	0.25%	3	1.00%	2	0.54%	2	0.55%	0	0.00%
Concrete	0	0.00%	2	0.67%	1	0.27%	0	0.00%	2	2.63%
Crateva excelsa	0	0.00%	15	5.02%	14	3.76%	1	0.28%	0	0.00%
Dialium madagascariensis	0	0.00%	8	2.68%	0	0.00%	0	0.00%	0	0.00%
Discorea fandra	0	0.00%	0	0.00%	1	0.27%	0	0.00%	0	0.00%
***Discorea* sp.**	0	0.00%	0	0.00%	2	0.54%	5	1.38%	0	0.00%
Dombeya analavelonae	0	0.00%	0	0.00%	1	0.27%	0	0.00%	0	0.00%
Egg / Bird Nest	1	0.25%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Fale	0	0.00%	0	0.00%	0	0.00%	1	0.28%	0	0.00%
Feces	0	0.00%	0	0.00%	3	0.81%	0	0.00%	0	0.00%
***Gouania* sp.**	0	0.00%	0	0.00%	1	0.27%	0	0.00%	0	0.00%
Grewia grevei	0	0.00%	1	0.33%	0	0.00%	4	1.10%	0	0.00%
Grewia leucophylla	0	0.00%	0	0.00%	0	0.00%	28	7.71%	10	13.16%
***Grewia* sp.**	0	0.00%	0	0.00%	0	0.00%	1	0.28%	0	0.00%
***Hibiscus* sp.**	0	0.00%	0	0.00%	1	0.27%	3	0.83%	0	0.00%
Kisene	0	0.00%	0	0.00%	2	0.54%	0	0.00%	0	0.00%
Kisene and/or voamanga	0	0.00%	0	0.00%	1	0.27%	0	0.00%	0	0.00%
***Landolphia* sp.**	0	0.00%	1	0.33%	0	0.00%	0	0.00%	0	0.00%
Lobakahjirike	0	0.00%	0	0.00%	1	0.27%	0	0.00%	0	0.00%
Mamyaho	0	0.00%	0	0.00%	0	0.00%	25	6.89%	0	0.00%
Mango	0	0.00%	18	6.02%	0	0.00%	0	0.00%	0	0.00%
Melon	0	0.00%	0	0.00%	2	0.54%	0	0.00%	0	0.00%
Metaporana parvifolia	24	6.09%	23	7.69%	133	35.75%	80	22.04%	18	23.68%
***Olax* sp.**	0	0.00%	1	0.33%	0	0.00%	0	0.00%	0	0.00%
Papaya	0	0.00%	0	0.00%	1	0.27%	0	0.00%	0	0.00%
***Pentopetio* sp.**	9	2.28%	17	5.69%	18	4.84%	21	5.79%	5	6.58%
Physena sessiliflora	0	0.00%	1	0.33%	0	0.00%	0	0.00%	0	0.00%
Sakavironala	0	0.00%	1	0.33%	1	0.27%	0	0.00%	0	0.00%
Salvadora angustifolia	271	68.78%	32	10.70%	5	1.34%	6	1.65%	0	0.00%
Sarytabuara	0	0.00%	1	0.33%	1	0.27%	0	0.00%	0	0.00%
***Secamone* sp.**	1	0.25%	5	1.67%	11	2.96%	6	1.65%	3	3.95%
Seyrigia gracilis	3	0.76%	1	0.33%	0	0.00%	0	0.00%	0	0.00%
Soil	1	0.25%	6	2.01%	0	0.00%	4	1.10%	1	1.32%
Soil/Wood	0	0.00%	0	0.00%	0	0.00%	2	0.55%	0	0.00%
Stone	0	0.00%	1	0.33%	0	0.00%	0	0.00%	0	0.00%
Strychnos madagascariensis	0	0.00%	1	0.33%	0	0.00%	0	0.00%	0	0.00%
Talinella dauphinensis	22	5.58%	3	1.00%	4	1.08%	2	0.55%	2	2.63%
Tamarindus indica	30	7.61%	88	29.43%	102	27.42%	82	22.59%	19	25.00%
Tamelapsis linearis	0	0.00%	0	0.00%	4	1.08%	3	0.83%	1	1.32%
Tarenna pruinosum	0	0.00%	1	0.33%	0	0.00%	0	0.00%	0	0.00%
Unknown	9	2.28%	18	6.02%	35	9.41%	24	6.61%	4	5.26%
Voamanga	0	0.00%	0	0.00%	13	3.49%	8	2.20%	0	0.00%
Wood	0	0.00%	9	3.01%	9	2.42%	14	3.86%	7	9.21%
Grand Total	**394**	**100.00%**	**299**	**100.00%**	**372**	**100.00%**	**363**	**100.00%**	**76**	**100.00%**

While a variety of species quickly rose and fell in consumption on a monthly basis, the major food species of *Tamarindus indica*, *Metaporana parvifolia* and *Salvadora angustifolia* were consumed frequently, although variably, across most months. *Tamarindus indica* accounted for the greatest number of feeding observations during the dry season months from July through October (32.01% - 42.43%), as well as during the wet season months of December (29.43%), February (22.59%) and March (25.00%). Tamarind fell to the second most commonly consumed species for November (7.61%) and January (27.42%), during which *Salvadora angustifolia* and *Metaporana parvifolia* were the dominant food items respectively. *Salvadora angustifolia* was typically the second most consumed food species for the duration of the dry season, accounting from 10.88% to 23.46% of feeding observations. Peak *Salvadora* consumption occurred during November when its fruit became exceptionally available, and during which this species accounted for a majority of feeding observations (68.78%). *Salvadora* consumption then fell to second place (10.70%) in December, before being consumed at a low rate during the months of January, February and March (1.34%, 1.65% and 0.00% respectively). *Metaporana parvifolia* was the third most commonly consumed item across the study, but demonstrated the most variability in consumption of these major taxa. This likely reflects the fact that this species was only consumed in the form of leaves (and occasionally young stems), and was only widely available during early dry season observations and during the wet season. Although accounting for 10.74% of feeding during July, *Metaporana* accounted for less than 3% of observations from August through October. After this point, consumption of this species increased monthly until it accounted for 35.75% of the diet in January. Consumption then declined to 22.04% and 23.68% of the diet in February and March respectively.

Consumption of Foods by Type Across the Study Period.

As indicated above, BMSR ring-tailed lemurs consume a wide variety of food species. From these taxa, a mix of different food item types are consumed. The types of foods eaten by lemurs include, but are not limited to, young leaves, mature leaves, leaf buds, stems, fruit, flowers, flower buds insects and stems. Likewise, it was also common for multiple food types to be consumed from single food species. For example, *Tamarindus indica* fruit, flowers, leaf buds, young leaves and mature leaves were eaten, while *Salvadora angustifolia* flower buds, flowers, fruit, young leaves and mature leaves were consumed. For species where multiple food types were consumed, different plant parts were typically eaten with regards to their availability as related to seasonality and/or food species phenology. However, in some cases, multiple food types were consumed from the same species at single point in time (e.g., leaves and fruit for tamarind, fruit and leaves/flowers for sasavy). In this section I detail patterns of feeding consumption as related to the food type across the course of the study period. Food item type data are then broken into seasonal and monthly patterns of feeding.

Food types are defined using a simplified system that combines flowers and vegetative portions of plants into a single category of "Flowers / Vegetation" (sometimes shortened to "vegetation" below). This category thus includes leaves, leaf buds, flowers, flower buds and stems. This system is utilized as for many taxa it was often difficult to determine the type of food consume as multiple food types were present simultaneously. This was commonly observed for the major food species consumed across the study. For example, when consuming Sasavy (*Salvadora angustifolia*), lemurs were observed to eat both flowers and young leaves during the same feeding bout. Furthermore, for tamarind, individuals were observed to consume both young leaves and mature leaves during a single feeding bout, while tratriotse (*Acacia bellula*) leaf buds

and young leaves were often consumed simultaneously. Likewise, multiple food types were often available across a food species due to asynchrony in phenology between individual plants (e.g., fruit was present in one plant while only leaves were available in another plant). Such a pattern of food availability is particularly common for tamarind, as this species demonstrates asynchronous fruiting, and fruit from this species was thus available across the study period (see Sauther 1998, Sauther and Cuozzo, 2009). Fruit consumption only refers to the consumption of fruits that were sourced either from the plant itself or were consumed from the ground after falling from a food species plant. Fruits sourced from the camp's trash or were raided from the camp's dining area were considered to be foods listed as "Trash / Other", as these were frequently processed prior to consumption, and thus their relationship to dental senescence likely differs from those obtained in a nonanthropogenic context. The "Other / Trash" classification also includes all other foods which could be identified but which could not be accounted for by alternative categories (e.g., bird's eggs, spider webs, etc.). Outside of the fruit, vegetation/flowers, and trash/other categorizations, foods were characterized in categories including: 1) Feces, 2) Insects (for caterpillars and cicadas), 3) soil (for geophagy, concrete and stone consumption, including by licking), 4) Wood (including termite excreta). Foods types which could not be characterized were classified as "unknown." As with food species characterized as "unknown", this was typically the result of poor observational conditions where a specific food item's type could not be determined, and many "unknown" foods likely fell into the above categories.

Across the year, vegetative material (e.g., flowers, leaves and stems) and fruit accounted for the vast majority of foods consumed (89.75%) of the diet. Of these, vegetation and flowers accounted for the 49.34% of feeding observations (n = 1535/3111 observations), while fruit

represented 40.41% of feeding observations (n = 1257/3111). Woody foods including termite excreta (which was often present upon rotting wood piles) accounted for the second most commonly consumed food type at 2.57% of feeding observations. Soil and insects accounted for 1.58% and 1.45% of observations respectively. Feces accounted for 0.58% of the diet, while flower buds / fruit of tsikidrakatse (*Bridelia* sp.) accounted for 0.51% of the diet. Foods in the "Other / Trash" category accounted for 1.38% of feeding observations. Unknown food types represented 2.19% of all feeding observations (Figure 3.3, Table 3.7).

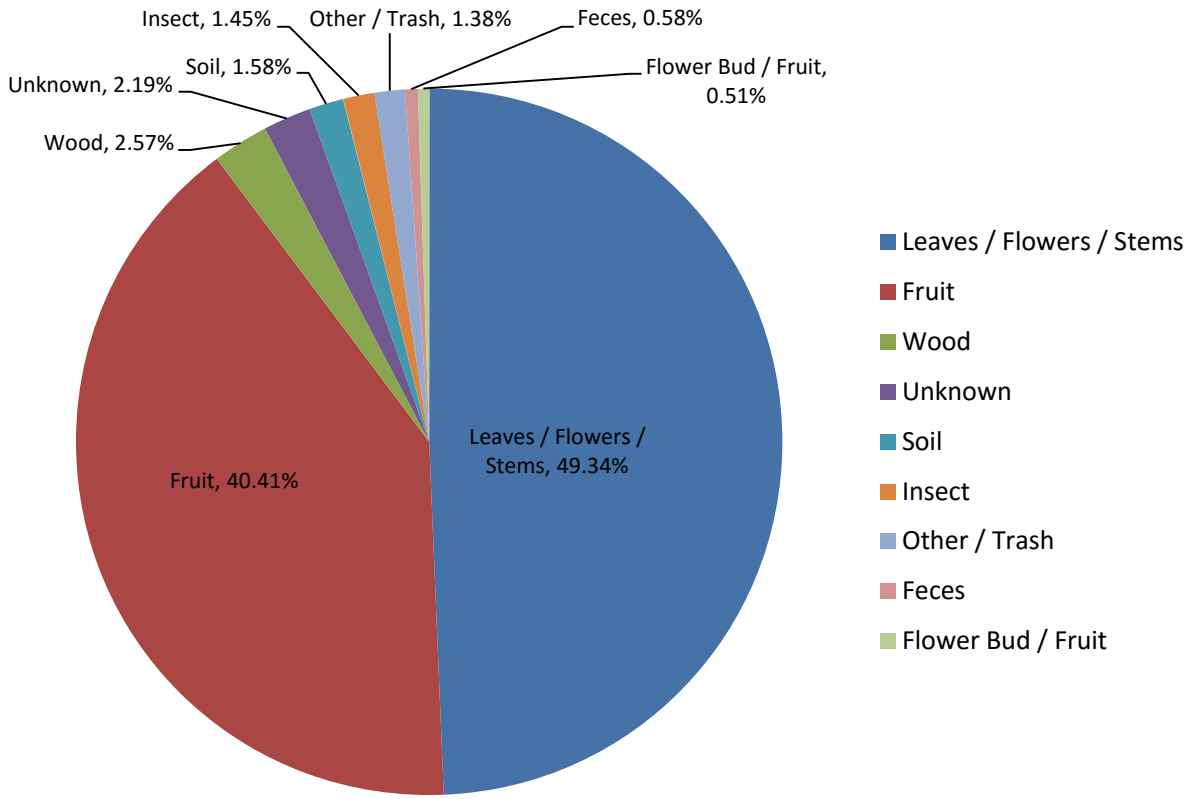


Figure 3.3. Total food type consumption across the entire study period. Fruit and vegetative material dominated the diet overall, with small amounts of alternative foods accounting for approximately 10% of the diet

Table 3.7. Food Types by Season and Across the Total Study.

	Dry Season		Wet Season		Total Study	
Food Type	**n =**	**% Total Dry**	**n =**	**% Total Wet**	**n =**	**% Total**
Leaves / Flowers / Stems	886	55.13%	649	43.15%	1535	49.34%
Fruit	567	35.28%	690	45.88%	1257	40.41%
Wood	41	2.55%	39	2.59%	80	2.57%
Unknown	44	2.74%	24	1.60%	68	2.19%
Soil	31	1.93%	18	1.20%	49	1.58%
Insect	0	0.00%	45	2.99%	45	1.45%
Other / Trash	22	1.37%	21	1.40%	43	1.38%
Feces	15	0.93%	3	0.20%	18	0.58%
Flower Bud / Fruit	1	0.06%	15	1.00%	16	0.51%
Grand Total	**1607**	**100.00%**	**1504**	**100.00%**	**3111**	**100.00%**

As related to vegetative material consumed across the course of the study, 11 species accounted for >1% of total feeding behavior. The most commonly consumed leaf species was represented by *Metaporana parvifolia* at 11.25% of total feeding observations. *Salvadora angustifolia* leaves and flowers likewise accounted for 7.33% of the diet, while tamarind leaves and flowers accounted for 7.07% of the diet. With regards to fruit, only 4 species accounted for more than 1% of the total diet. Tamarind was the most commonly consumed fruit, accounting for 21.79% of the total diet. *Salvadora angustifolia* berries accounted for a total of 11.06% of feeding observations. All other fruits were consumed at a comparatively low rate overall, with only *Tarenna pruinosum* and *Grewia leucophylla* accounting for more than 1% of the diet at 1.93% and 1.22% of feeding observations respectively.

As with food species, the proportion of each food type consumed varied between the wet and dry seasons. For the dry season vegetative material accounted for the largest proportion of the diet (55.13%), while fruit accounted for the second largest food type (35.28%). During the wet season this pattern reversed with fruit representing the greatest single food type (45.88%)

and vegetative material accounting for 43.15% of feeding observations. These two food item types thus accounted for 90.42% and 89.03% of the diet during the dry and wet seasons respectively. Other food types therefore only accounted for a comparatively small (<10%) of feeding observations seasonally. With relation to the consumption of such foods, it is notable that insects were only eaten during the wet season, while feces were more commonly consumed during the dry season. Percentages of observations consuming these foods during the dry season include the following, 1) Wood: 2.55%, 2) Soil: 1.93%, 3) Feces: 0.93, 4) Other / Trash: 1.37% 5) Flower Buds / Fruit: 0.06%. Unknown foods accounted for 2.74% of dry season feeding. Minor wet season foods include the following: 1) Insects: 2.99%, 2) Wood: 2.59%, 3) Other / Trash: 1.40%, 4) Soil: 1.20% 5) Flower buds and Fruit: 1.00%, 6) Feces: 0.20%. Unknown food types accounted for 1.60% of feeding during the wet season (Figure 3.4, Table 3.7).

With regards to species, during the dry season vegetation from *Salvadora angustifolia* represented 13.25% of feeding, while *Tamarindus indica* vegetation accounted for 10.33% of feeding. Flowers and flower buds from *Quisivianthe papionae* represented 6.1% of the diet while *Metaporana parvifolia* accounted for 4.48% of feeding observations. During the wet season, the most commonly consumed fruits were from *Salvadora angustifolia* (19.41% of total) and *Tamarindus indica* (17.75%). In terms of vegetative feeding, leaves and stems from *Metaporana parvifolia* represented the dominant food species during the wet season, accounting for 18.4% of feeding. *Pentopetio* sp. leaves and stems accounted for 4.59% of total feeding.

Monthly Variation in Food Item Type.

When examined at a monthly level, there is variation evident in the types of food consumed across the course of the study (Table 3.8; Figures 3.5 and 3.6). During the months of

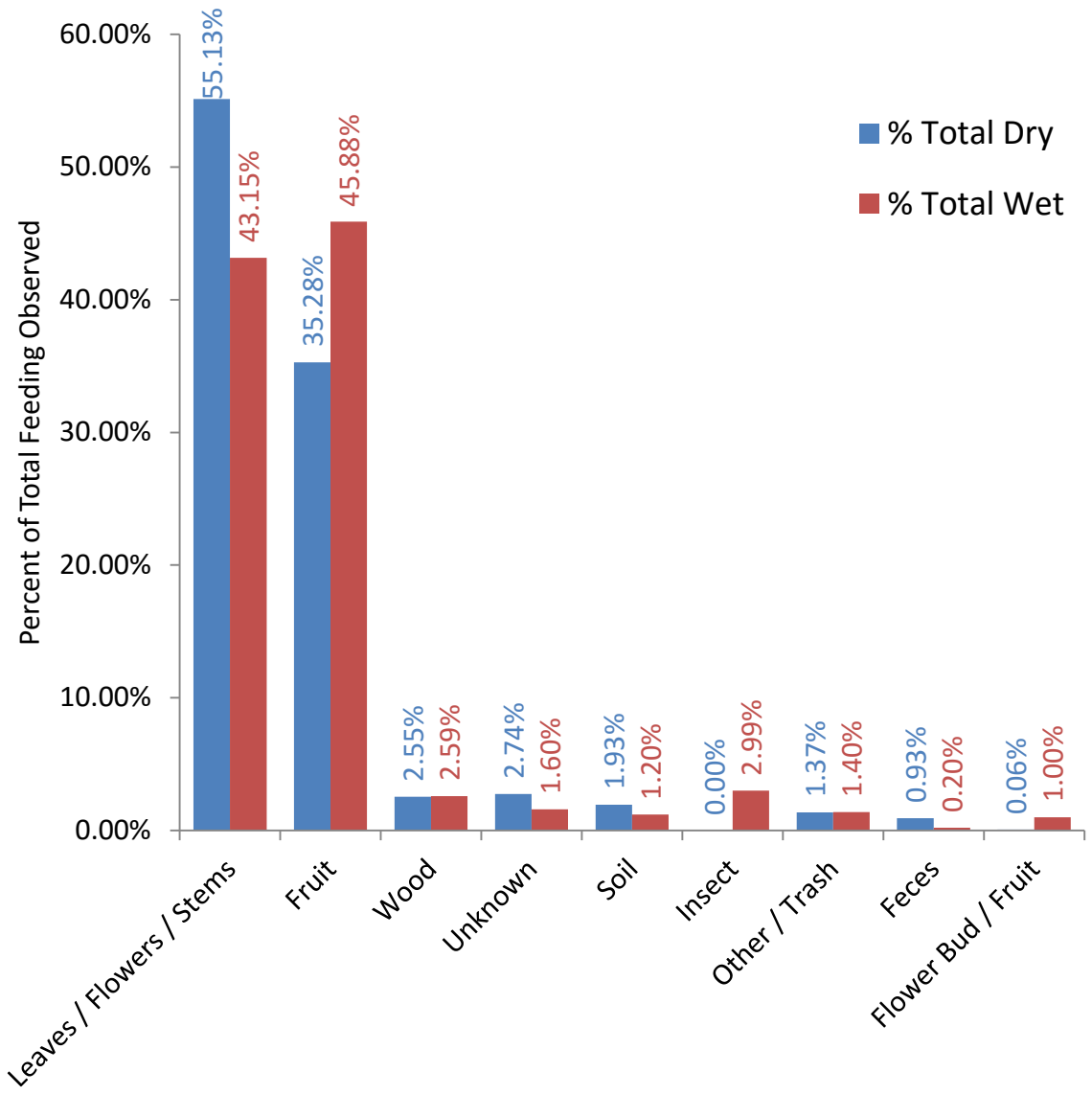


Figure 3.4. Differences in food type consumption by study season. During the Dry season vegetative material accounted for the greatest single food type (55.13%) while fruit accounted for the greatest single food type consumed during the wet season 45.88%. All other food types accounted for less than 3% of total for either season.

the dry season, vegetative material represented the primary food item consumed for each month (July: 45.33%, August: 49.41%, September: 68.70%, October: 57.24%). During the wet season, vegetation became the second most commonly consumed food items for November (25.89%) and December (26.42%), and became the most commonly consumed foods for January (64.52%), February (52.89%) and March (47.37%). Please note that results for March represent data for a reduced number of follows, and for a subsample of individuals and groups as the study ended during the middle of this month. Monthly data concerning variation in vegetative food species is presented in Table 3.9; these data are also visualized in Figures 3.7 and 3.8.

Table 3.8. Feeding by Food Item Type by Month.

		Jul.	Aug.	Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Total
Feces	**n =**	6	1	8	0	0	0	3	0	0	**18**
	%	1.19%	0.29%	1.74%	0.00%	0.00%	0.00%	0.81%	0.00%	0.00%	**0.58%**
Flower Bud / Fruit	**n =**	0	0	1	0	0	1	0	14	0	**16**
	%	0.00%	0.00%	0.22%	0.00%	0.00%	0.33%	0.00%	3.86%	0.00%	**0.51%**
Fruit	**n =**	219	142	100	106	280	137	115	129	29	**1257**
	%	43.54%	41.76%	21.74%	34.87%	71.07%	45.82%	30.91%	35.54%	38.16%	**40.41%**
Insect	**n =**	0	0	0	0	8	34	0	3	0	**45**
	% =	0.00%	0.00%	0.00%	0.00%	2.03%	11.37%	0.00%	0.83%	0.00%	**1.45%**
Leaves, Flowers, Stems	**n =**	228	168	316	174	102	79	240	192	36	**1535**
	% =	45.33%	49.41%	68.70%	57.24%	25.89%	26.42%	64.52%	52.89%	47.37%	**49.34%**
Other / Trash	**n =**	2	14	2	4	1	17	3	0	0	**43**
	% =	0.40%	4.12%	0.43%	1.32%	0.25%	5.69%	0.81%	0.00%	0.00%	**1.38%**
Soil	**n =**	18	4	7	2	1	9	1	4	3	**49**
	% =	3.58%	1.18%	1.52%	0.66%	0.25%	3.01%	0.27%	1.10%	3.95%	**1.58%**
Unk.	**n =**	9	11	22	2	2	13	1	7	1	**68**
	% =	1.79%	3.24%	4.78%	0.66%	0.51%	4.35%	0.27%	1.93%	1.32%	**2.19%**
Wood	**n =**	21	0	4	16	0	9	9	14	7	**80**
	% =	4.17%	0.00%	0.87%	5.26%	0.00%	3.01%	2.42%	3.86%	9.21%	**2.57%**
Total	**n =**	503	340	460	304	394	299	372	363	76	**3111**
	% =	100%	100%	100%	100%	100.%	100%	100%	100%	100%	**100%**

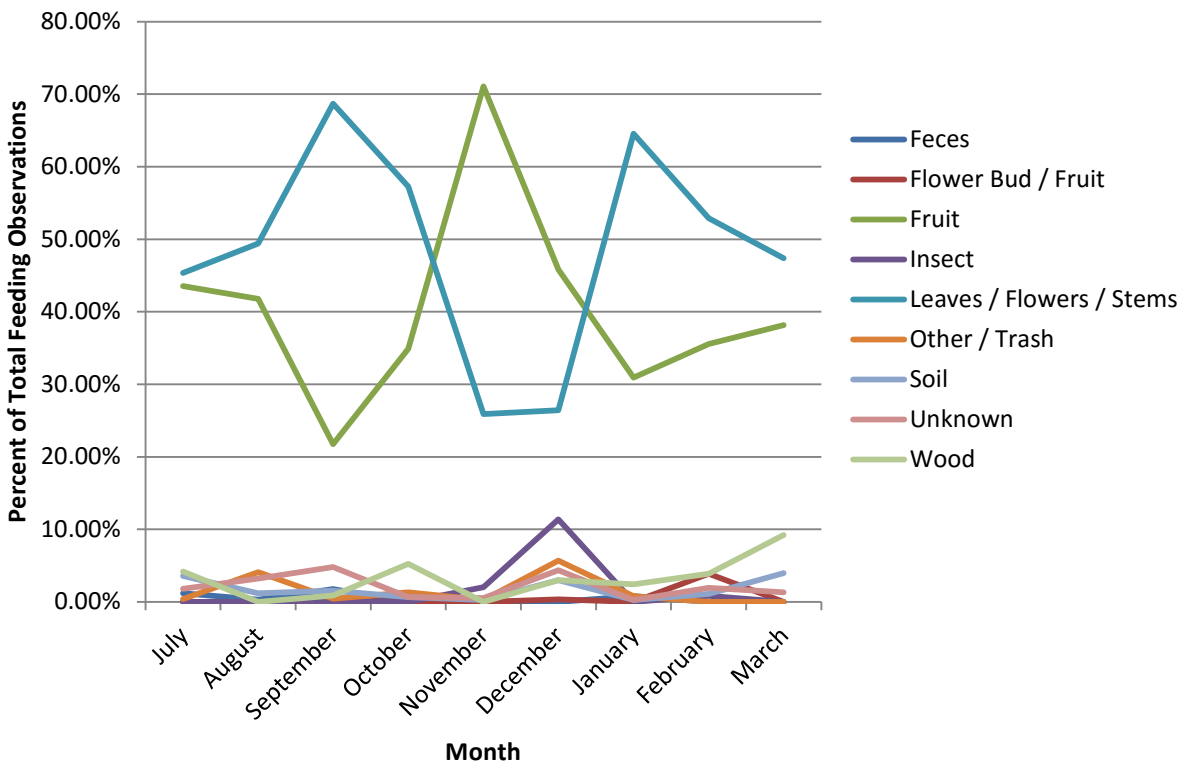


Figure 3.5. Percentage of all feeding observations for each type of food consumed across the study period on a month-to-month basis.

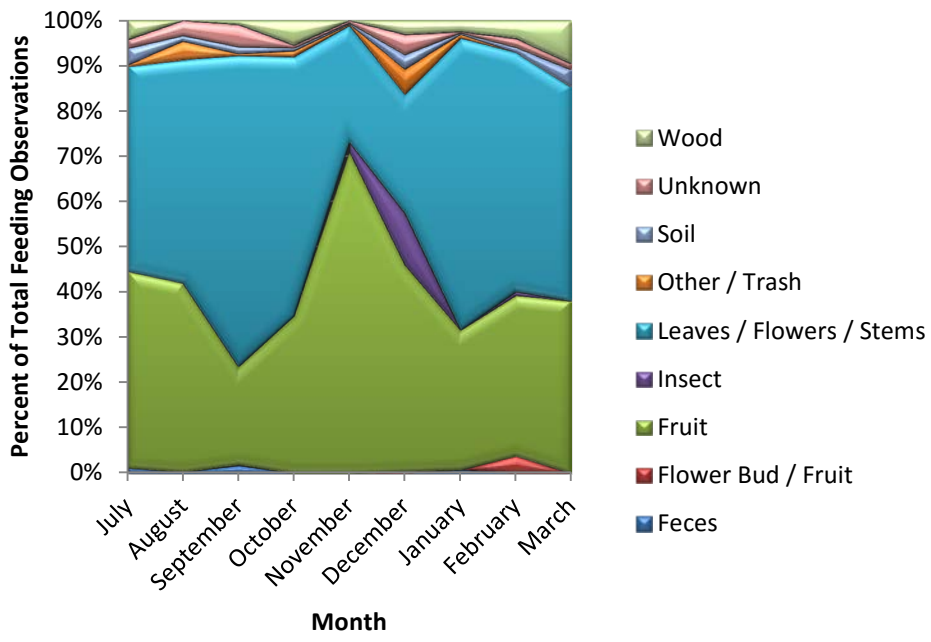


Figure 3.6. Percentage of each food item type as a percent of total feeding observations. Data is presented on a month-to-month basis.

Table 3.9. Vegetation Species Consumed from July to November as a Percent of Total Feeding.

	July		August		September		October		November	
Vegetation Food Species	**n**	**% =**	**n**	**% =**	**n**	**% =**	**n**	**% =**	**n**	**% =**
Metaporana parvifolia	54	10.74%	9	2.65%	6	1.30%	3	0.99%	24	6.09%
Salvadora angustifolia	118	23.46%	37	10.88%	52	11.30%	6	1.97%	0	0.00%
Tamarindus indica	1	0.20%	1	0.29%	75	16.30%	89	29.28%	27	6.85%
Unknown	15	2.98%	20	5.88%	11	2.39%	12	3.95%	8	2.03%
Quisivianthe papionae	0	0.00%	29	8.53%	67	14.57%	2	0.66%	0	0.00%
Acacia bellula	3	0.60%	39	11.47%	52	11.30%	0	0.00%	0	0.00%
***Pentopetio* sp.**	0	0.00%	0	0.00%	0	0.00%	5	1.64%	9	2.28%
Cedrelopsis grevei	0	0.00%	0	0.00%	0	0.00%	31	10.20%	7	1.78%
Gyrocarpus americanus	0	0.00%	2	0.59%	20	4.35%	22	7.24%	0	0.00%
Ipomoea majungensis	2	0.40%	17	5.00%	18	3.91%	1	0.33%	0	0.00%
***Secamone* sp.**	2	0.40%	7	2.06%	1	0.22%	0	0.00%	1	0.25%
Talinella dauphinensis	0	0.00%	0	0.00%	0	0.00%	1	0.33%	21	5.33%
Mamyaho	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Coridia ainensis	17	3.38%	0	0.00%	2	0.43%	0	0.00%	0	0.00%
***Commicarpus* sp.**	0	0.00%	0	0.00%	1	0.22%	0	0.00%	1	0.25%
Tamelapsis linearis	1	0.20%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Argemone mexicana	6	1.19%	0	0.00%	2	0.43%	0	0.00%	0	0.00%
***Discorea* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Maerua filiformis	3	0.60%	1	0.29%	3	0.65%	0	0.00%	0	0.00%
Seyrigia gracilis	0	0.00%	0	0.00%	3	0.65%	0	0.00%	3	0.76%
Capparis chrysameia	4	0.80%	2	0.59%	0	0.00%	0	0.00%	0	0.00%
Voamanga	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Clerodendrum* sp. cf. *emirnense	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Grewia grevei	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
***Hibiscus* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Achyranthus apera	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
***Achyranthus* sp.**	0	0.00%	2	0.59%	0	0.00%	0	0.00%	0	0.00%
Ipomoea carica	1	0.20%	1	0.29%	0	0.00%	0	0.00%	0	0.00%
Kisene	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Sakavironala	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Sarytabuara	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Acalyphya decaryana	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.25%
Alluaudia procera	0	0.00%	0	0.00%	1	0.22%	0	0.00%	0	0.00%
Aloe vahontsoy	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
***Bridelia* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Byttneria voulili	1	0.20%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
***Combretum* sp.**	0	0.00%	1	0.29%	0	0.00%	0	0.00%	0	0.00%
Crateva excelsa	0	0.00%	0	0.00%	0	0.00%	1	0.33%	0	0.00%
Diospyros sakalavarum	0	0.00%	0	0.00%	1	0.22%	0	0.00%	0	0.00%
Discorea fandra	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Dombeya analavelonae	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
***Gouania* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Gymnosporia linearis	0	0.00%	0	0.00%	0	0.00%	1	0.33%	0	0.00%
Kisene and/or voamanga	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
***Landolphia* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Lobakahjirike	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
***Maerua* sp.**	0	0.00%	0	0.00%	1	0.22%	0	0.00%	0	0.00%
Leaves / Flowers / Stems Total	**228**	**45.33%**	**168**	**49.41%**	**316**	**68.70%**	**174**	**57.24%**	**102**	**25.89%**

Table 3.10. Vegetation Species Consumed from December- March as a % of Total Feeding.

	December		January		February		March		Total	
Vegetation Food Species	**n**	**% =**	**n**	**% =**	**n**	**% =**	**n**	**% =**	**n =**	**% =**
Metaporana parvifolia	23	7.69%	133	35.75%	80	22.04%	18	23.68%	350	11.25%
Salvadora angustifolia	4	1.34%	5	1.34%	6	1.65%	0	0.00%	228	7.33%
Tamarindus indica	6	2.01%	9	2.42%	8	2.20%	4	5.26%	220	7.07%
Unknown	11	3.68%	34	9.14%	20	5.51%	3	3.95%	134	4.31%
Quisivianthe papionae	0	0.00%	0	0.00%	0	0.00%	0	0.00%	98	3.15%
Acacia bellula	0	0.00%	0	0.00%	0	0.00%	0	0.00%	94	3.02%
***Pentopetio* sp.**	17	5.69%	18	4.84%	20	5.51%	5	6.58%	74	2.38%
Cedrelopsis grevei	2	0.67%	0	0.00%	4	1.10%	0	0.00%	44	1.41%
Gyrocarpus americanus	0	0.00%	0	0.00%	0	0.00%	0	0.00%	44	1.41%
Ipomoea majungensis	0	0.00%	0	0.00%	0	0.00%	0	0.00%	38	1.22%
***Secamone* sp.**	5	1.67%	11	2.96%	5	1.38%	3	3.95%	35	1.13%
Talinella dauphinensis	3	1.00%	4	1.08%	2	0.55%	2	2.63%	33	1.06%
Mamyaho	0	0.00%	0	0.00%	25	6.89%	0	0.00%	25	0.80%
Coridia ainensis	0	0.00%	0	0.00%	0	0.00%	0	0.00%	19	0.61%
***Commicarpus* sp.**	3	1.00%	2	0.54%	2	0.55%	0	0.00%	9	0.29%
Tamelapsis linearis	0	0.00%	4	1.08%	3	0.83%	1	1.32%	9	0.29%
Argemone mexicana	0	0.00%	0	0.00%	0	0.00%	0	0.00%	8	0.26%
***Discorea* sp.**	0	0.00%	2	0.54%	5	1.38%	0	0.00%	7	0.23%
Maerua filiformis	0	0.00%	0	0.00%	0	0.00%	0	0.00%	7	0.23%
Seyrigia gracilis	1	0.33%	0	0.00%	0	0.00%	0	0.00%	7	0.23%
Capparis chrysameia	0	0.00%	0	0.00%	0	0.00%	0	0.00%	6	0.19%
Voamanga	0	0.00%	5	1.34%	0	0.00%	0	0.00%	5	0.16%
Clerodendrum* sp. cf. *emirnense	0	0.00%	2	0.54%	2	0.55%	0	0.00%	4	0.13%
Grewia grevei	0	0.00%	0	0.00%	4	1.10%	0	0.00%	4	0.13%
***Hibiscus* sp.**	0	0.00%	1	0.27%	3	0.83%	0	0.00%	4	0.13%
Achyranthus apera	1	0.33%	1	0.27%	1	0.28%	0	0.00%	3	0.10%
***Achyranthus* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	2	0.06%
Ipomoea carica	0	0.00%	0	0.00%	0	0.00%	0	0.00%	2	0.06%
Kisene	0	0.00%	2	0.54%	0	0.00%	0	0.00%	2	0.06%
Sakavironala	1	0.33%	1	0.27%	0	0.00%	0	0.00%	2	0.06%
Sarytabuara	1	0.33%	1	0.27%	0	0.00%	0	0.00%	2	0.06%
Acalyphya decaryana	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Alluaudia procera	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Aloe vahontsoy	0	0.00%	0	0.00%	1	0.28%	0	0.00%	1	0.03%
***Bridelia* sp.**	0	0.00%	0	0.00%	1	0.28%	0	0.00%	1	0.03%
Byttneria voulili	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
***Combretum* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Crateva excelsa	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Diospyros sakalavarum	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Discorea fandra	0	0.00%	1	0.27%	0	0.00%	0	0.00%	1	0.03%
Dombeya analavelonae	0	0.00%	1	0.27%	0	0.00%	0	0.00%	1	0.03%
***Gouania* sp.**	0	0.00%	1	0.27%	0	0.00%	0	0.00%	1	0.03%
Gymnosporia linearis	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Kisene and/or voamanga	0	0.00%	1	0.27%	0	0.00%	0	0.00%	1	0.03%
***Landolphia* sp.**	1	0.33%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Lobakahjirike	0	0.00%	1	0.27%	0	0.00%	0	0.00%	1	0.03%
***Maerua* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Leaves / Flowers / Stems Total	**79**	**26.42%**	**240**	**64.52%**	**192**	**52.89%**	**36**	**47.37%**	**1535**	**49.34%**

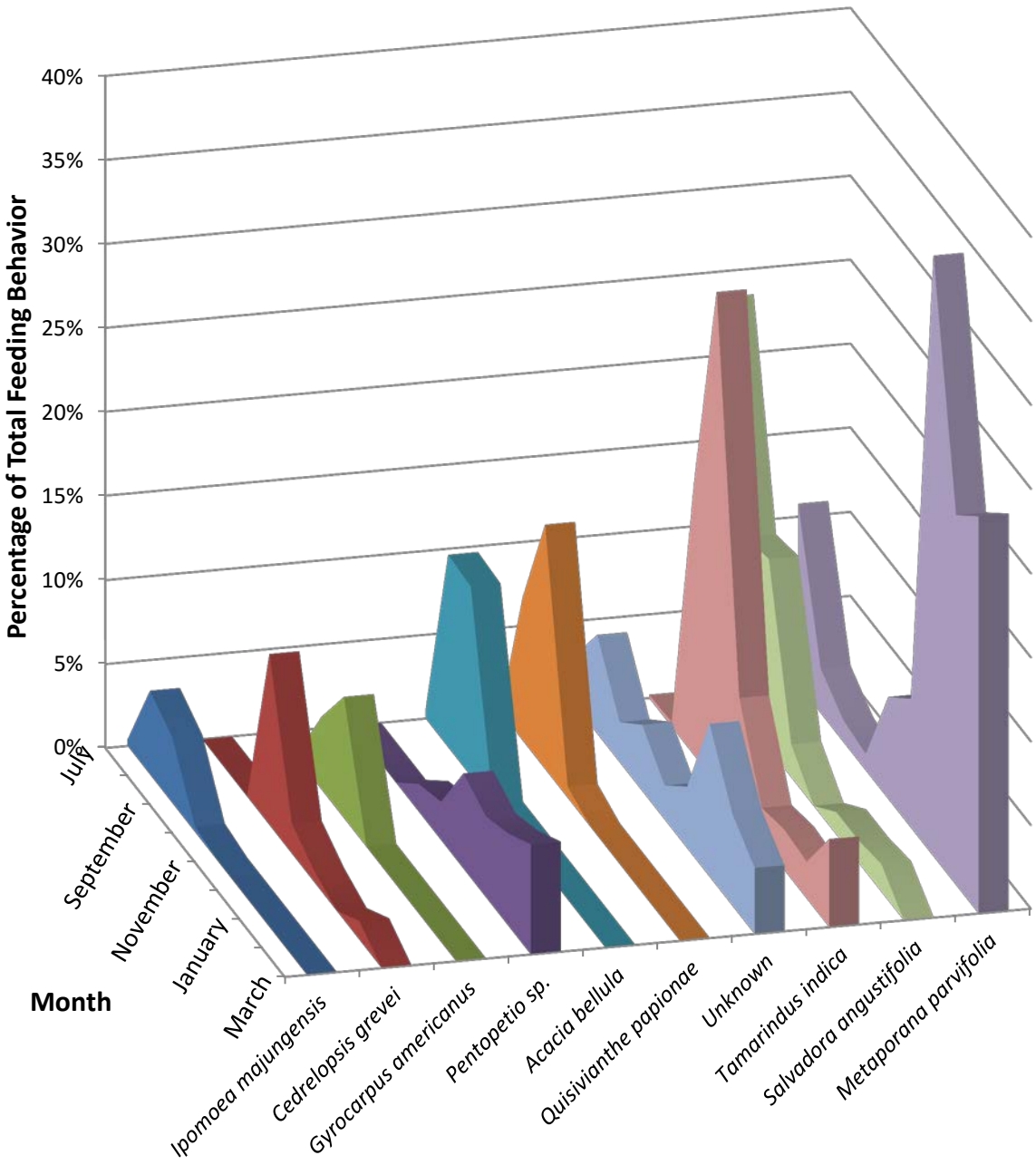


Figure 3.7. Percent of total feeding behavior for vegetation by species on a month-to-month basis.

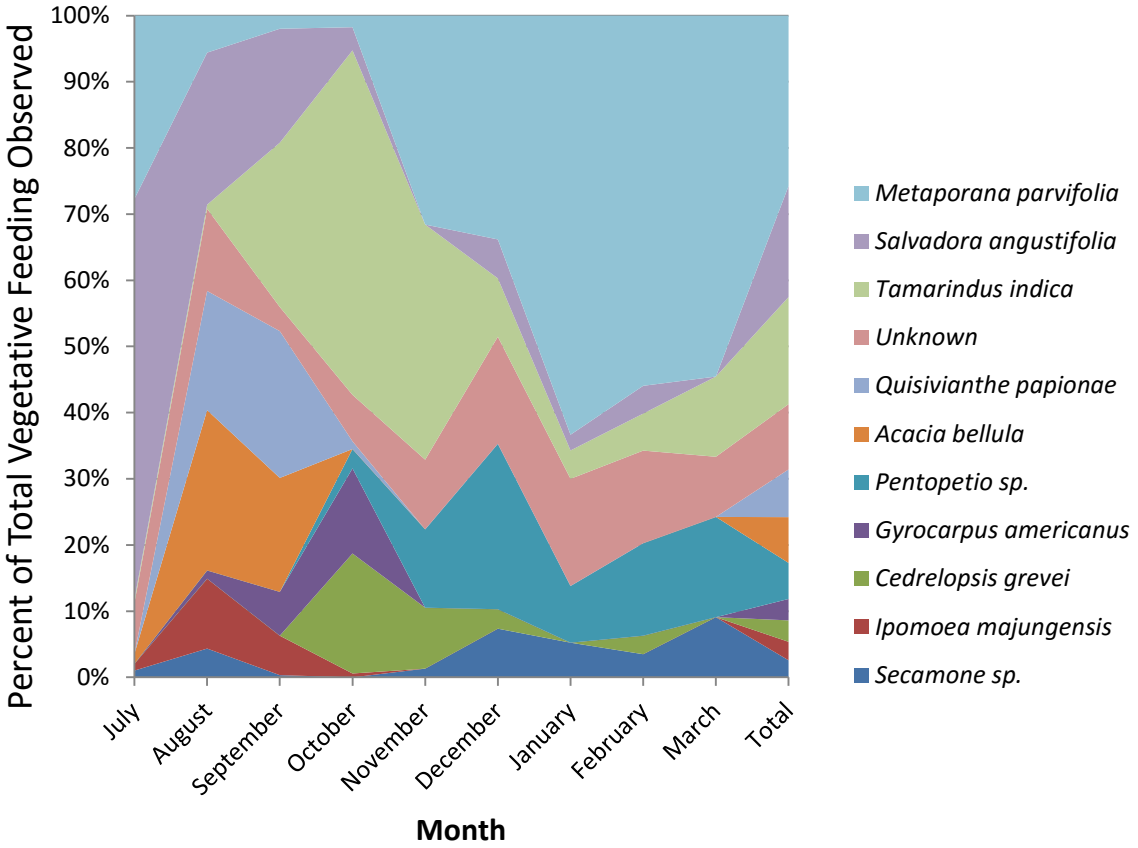


Figure 3.8. Percent of total vegetative feeding for the top 10 vegetative food species by month.

Fruit was the second most commonly consumed type of food across the study at 40.41% of total feeding observations. For all dry season months, fruit (primarily tamarind for most months), was the second most commonly consumed food type (July 43.54%, August 41.76%, September: 21.74%, October: 34.87%). During the wet season study months (November to March), fruit was the dominant food type for November and December at 71.01% and 45.82% of total feeding observations respectively. For the month of November, *Salvadora angustifolia* fruit accounted for 68.53% of total feeding observations, the only time during the study when one singular food species or type accounted for a majority of foods consumed. Monthly data for fruit species are available in Tables 3.11 and 3.12, while monthly data for the top 10 fruit species are visualized in Figures 3.9 and 3.10.

Table 3.11. Fruit Species Consumption as Percent of Total Feeding From July to November (Ordered by Total Rank).

	July		August		September		October		November	
Fruit Food Species	**n**	**% =**	**n**	**% =**	**n**	**% =**	**n**	**% =**	**n**	**% =**
Tamarindus indica	160	31.81%	127	37.35%	84	18.26%	40	13.16%	3	0.76%
Salvadora angustifolia	0	0.00%	0	0.00%	0	0.00%	52	17.11%	270	68.53%
Tarenna pruinosum	48	9.54%	8	2.35%	3	0.65%	0	0.00%	0	0.00%
Grewia leucophylla	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Crateva excelsa	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Ipomoea majungensis	0	0.00%	0	0.00%	12	2.61%	11	3.62%	0	0.00%
Voamanga	0	0.00%	5	1.47%	0	0.00%	0	0.00%	0	0.00%
***Bridelia* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Azima tetracantha	0	0.00%	0	0.00%	1	0.22%	2	0.66%	2	0.51%
Dialium madagascariensis	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Unknown	5	0.99%	1	0.29%	0	0.00%	0	0.00%	0	0.00%
Capparis chrysameia	0	0.00%	0	0.00%	0	0.00%	0	0.00%	3	0.76%
Ficus cocculifolia	3	0.60%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Grewia grevei	3	0.60%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Acalyphya decaryana	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.25%
Antidesma petiolare	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Cissus bosseri	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Fale	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
***Grewia* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Maerua filiformis	0	0.00%	0	0.00%	0	0.00%	1	0.33%	0	0.00%
Mango	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
***Olax* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Physena sessiliflora	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
***Secamone* sp.**	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Strychnos madagascariensis	0	0.00%	0	0.00%	0	0.00%	0	0.00%	0	0.00%
Talinella dauphinensis	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.25%
Tomato	0	0.00%	1	0.29%	0	0.00%	0	0.00%	0	0.00%
Fruit Totals	**219**	**43.54%**	**142**	**41.76%**	**100**	**21.74%**	**106**	**34.87%**	**280**	**71.07%**

Table 3.12. Fruit Species Consumption as Percent of Total From December to March (Ordered by Total Rank).

	December		January		February		March		Total	
Fruit Food Species	**n =**	**% =**	**n =**	**% =**	**n =**	**% =**	**n =**	**% =**	**n =**	**% =**
Tamarindus indica	82	27.42%	93	25.00%	74	20.39%	15	19.74%	678	21.79%
Salvadora angustifolia	22	7.36%	0	0.00%	0	0.00%	0	0.00%	344	11.06%
Tarenna pruinosum	1	0.33%	0	0.00%	0	0.00%	0	0.00%	60	1.93%
Grewia leucophylla	0	0.00%	0	0.00%	28	7.71%	10	13.16%	38	1.22%
Crateva excelsa	15	5.02%	14	3.76%	1	0.28%	0	0.00%	30	0.96%
Ipomoea majungensis	0	0.00%	0	0.00%	0	0.00%	0	0.00%	23	0.74%
Voamanga	0	0.00%	8	2.15%	8	2.20%	0	0.00%	21	0.68%
***Bridelia* sp.**	0	0.00%	0	0.00%	14	3.86%	4	5.26%	18	0.58%
Azima tetracantha	4	1.34%	0	0.00%	0	0.00%	0	0.00%	9	0.29%
Dialium madagascariensis	8	2.68%	0	0.00%	0	0.00%	0	0.00%	8	0.26%
Unknown	0	0.00%	0	0.00%	0	0.00%	0	0.00%	6	0.19%
Capparis chrysameia	0	0.00%	0	0.00%	0	0.00%	0	0.00%	3	0.10%
Ficus cocculifolia	0	0.00%	0	0.00%	0	0.00%	0	0.00%	3	0.10%
Grewia grevei	0	0.00%	0	0.00%	0	0.00%	0	0.00%	3	0.10%
Acalyphya decaryana	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Antidesma petiolare	0	0.00%	0	0.00%	1	0.28%	0	0.00%	1	0.03%
Cissus bosseri	1	0.33%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Fale	0	0.00%	0	0.00%	1	0.28%	0	0.00%	1	0.03%
***Grewia* sp.**	0	0.00%	0	0.00%	1	0.28%	0	0.00%	1	0.03%
Maerua filiformis	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Mango	1	0.33%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
***Olax* sp.**	1	0.33%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Physena sessiliflora	1	0.33%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
***Secamone* sp.**	0	0.00%	0	0.00%	1	0.28%	0	0.00%	1	0.03%
Strychnos madagascariensis	1	0.33%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Talinella dauphinensis	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Tomato	0	0.00%	0	0.00%	0	0.00%	0	0.00%	1	0.03%
Fruit Totals	**137**	**45.82%**	**115**	**30.91%**	**129**	**35.54%**	**29**	**38.16%**	**1257**	**40.41%**

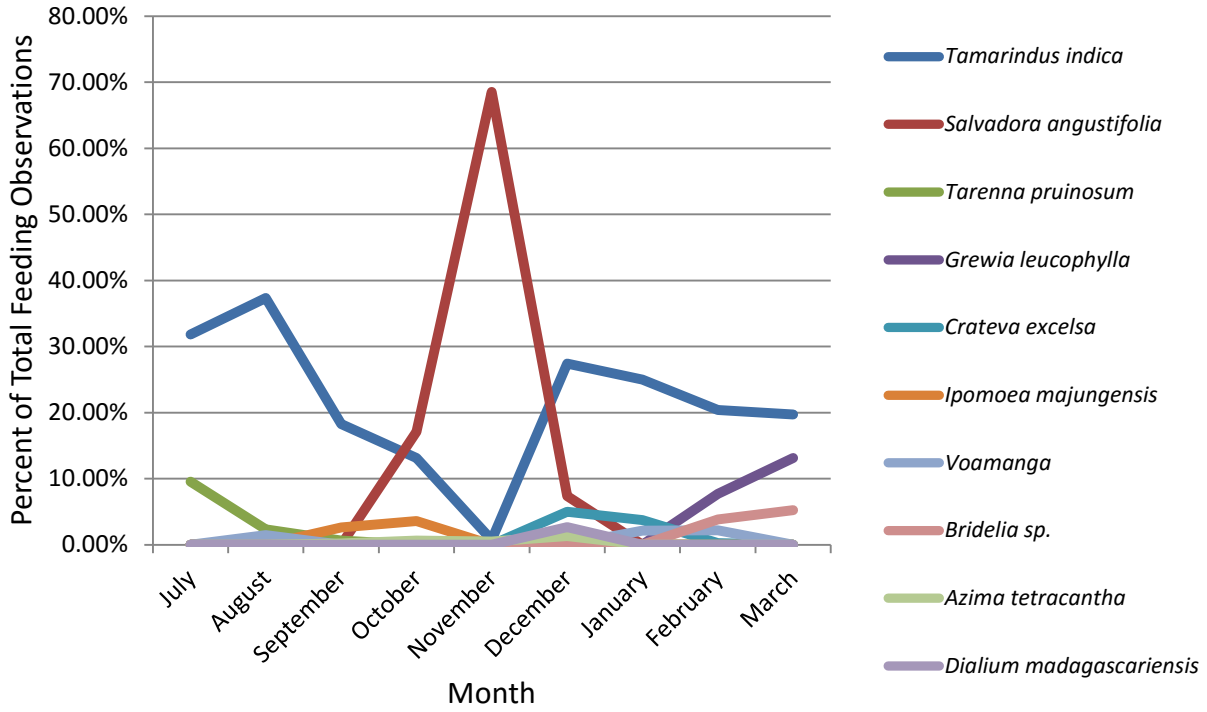


Figure 3.9. Percent of total feeding for the top 10 fruit species by month.

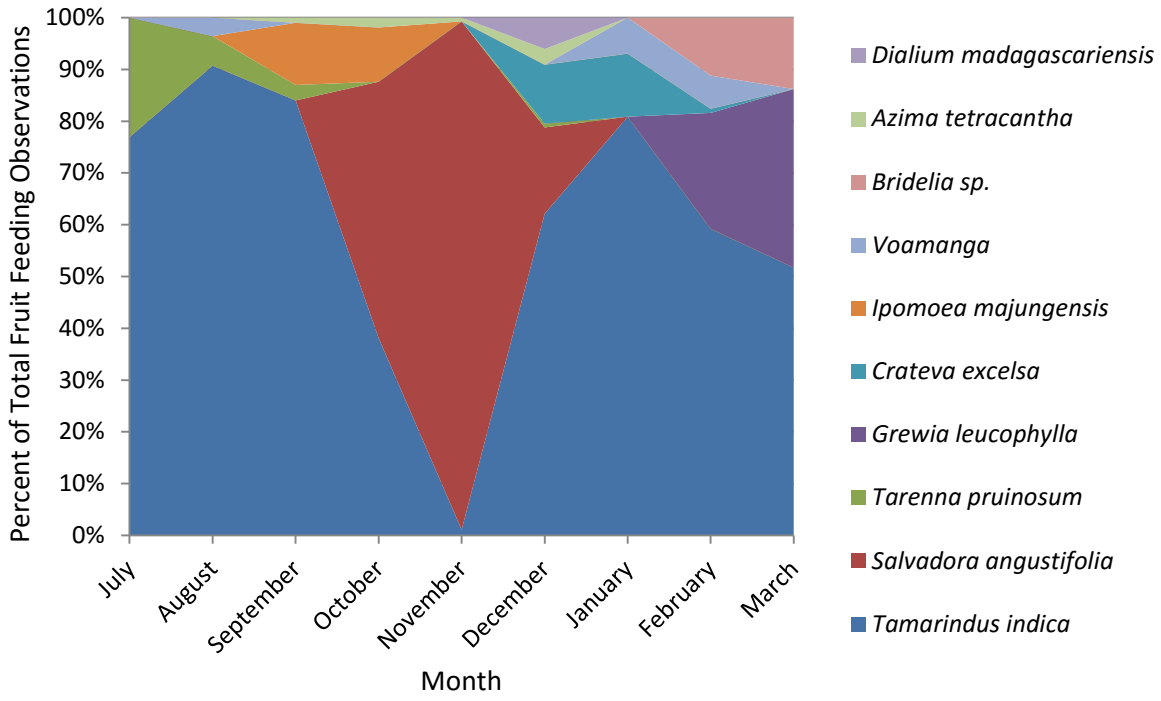


Figure 3.10. Percentage of the top ten fruit species as a percentage of total fruit feeding by month.

Across all study months, food types other than fruit and vegetative material typically only accounted for a comparatively small proportion of the diet (e.g., <10% of total feeding observations). A number of food types did demonstrate several minor spikes in feeding for specific months during the study (Table 3.8). For example, insects were only consumed during three months of the year, with the majority of insects consumed in the form of cicadas and caterpillars during the month of December where they accounted for 11.37% of the total diet. Insects were also consumed during November (2.03%) and February (0.83%). Insect consumption appeared to track their availability to the lemurs. As such, during November and December, large amounts of caterpillars were present in scrub and grassy areas of the forest. Likewise, cicadas were readily available during this time which represented their breeding period, and were frequently present in large mating groups and/or were loudly sounding their location. Additionally, lemurs consumed feces during the dry season months of July (1.19%), August (0.29%), and September (1.74%) as well as the wet season month of January (0.81%). The prevalence of fecal feeding during the dry season likely reflects reduced food availability during these months, and generally fits pattern of fecal feeding by *L. catta* at BMSR as reported by Fish et al. (2007). Additionally, soil appeared to be consumed frequently during the study, and was observed to be eaten during each month (Min: 0.25%, November; Max 3.95%, March). Wood was also consumed frequently across the study period, particularly during the months of July (4.17%), October (5.26%), December (3.01%), January (2.42%), February (3.86%) and March (9.21%). Determining the source of variation between months in wood consumption is difficult as this food type was continuously available across the study period. Additionally, the lemurs may have also been consuming termite excreta upon the wood, although this was difficult to assess during observations. Finally, Other / Trash food items typically drawn from camp

sources were commonly consumed during August (4.12%) and December (5.69%). For all other months such foods accounted for less than 2% of the diet. It is likely that the use of these foods during August reflects reduced food availability. During the height of the dry season, frequent use of camp trash pits as a food source was noted for a number of groups (e.g., Orange, Black, Yellow, and Blue). Use of such foods during December reflects consumption of mangoes in the camp area, as these were frequently consumed by camp residents before remaining scraps were taken by lemurs from the camp's trash pit.

Chapter Summary: BMSR *Lemur catta* Diet.

While BMSR *Lemur catta* consumed a wide variety of singly identifiable foods (n = 74), over the course of this study, over 90% of the diet was accounted for by 19 food species. Of these, only three taxa, *Tamarindus indica, Salvadora angustifolia* and *Metaporana parvifolia,* accounted for the majority (58.76%) of all feeding observations. Variation in food species consumed was present on a seasonal basis, although tamarind and sasavy remained the two most commonly eaten food species during both the wet and dry seasons. Bezà Mahafaly demonstrates highly seasonal patterns of rainfall, with a corresponding increase in food availability and higher diversity of food species accessible following the onset of rains in late October. As a result, lemurs consumed a wider variety of foods during the wet season (54 foods) than during the dry season (44 foods). Likewise, extensive monthly variation was present in the species of food consumed, with some species being consumed only for short periods (e.g., several weeks) across the study period. For example, *Acacia bellula* accounted for over 11% of feeding during the months of August and September, but was not consumed across the rest of the study. Similarly, mantsake (*Tarenna pruinosum*) represented 9.54% of the diet in July, but quickly fell to <3% for

August, and did not account for more than 1% of feeding for the rest of the study. Similar spikes in feeding were also noted for major food items. For instance, while *Salvadora angustifolia* accounted for almost 70% of feeding during November it accounted for less than 1% of feeding from January through March. As such, foods which appear to be exceptionally common food sources on a seasonal basis often varied greatly when assessed on a more fine-grained monthly basis. Again, although I collected no phenological data during this study, such brief increases in species prevalence in the diet appeared to reflect food species phenology and the resulting availability of a given food species.

Across the study period, the parts of plants consumed and types of foods consumed varied across the study period, while overall fruit (40.41%) and vegetative material (49.34%) represented the major types of foods consumed. During the dry season leaves accounted for the majority of feeding (55.13%) while fruit accounted for 35.28% of feeding, reflecting reduced fruit availability during this period (with the exception of kily fruit). In contrast, during the wet season, fruit accounted for the largest proportion of food consumed (45.88%) while vegetative material accounted for 43.15% of feeding. All other food types accounted for less than 3% of total feeding on an individual basis, overall and by season. Monthly variation was, however, present for food types in a manner similar to that seen for food species. For example, fruit ranged from a low of 30.91% of feeding observations in January to a maximum of 71.07% of observations in November, while vegetative materials accounted for only 25.89% in November, and a maximum of 68.7% of the diet in September. Minor food types also varied considerably on a month-to-month basis. This is particularly true for insects which accounted for 11.37% of the diet during December, but were only consumed in small amounts during two other months. Likewise, foods such as wood, soil and feces varied by quite a large amount depending on

month, often in relation to the overall availability of these resources or in the case of foods such as feces lack of alternative foods.

Overall, patterns of food consumption during this study generally followed those reported by previous observers at the Bezà Mahafaly Special Reserve. As such, a wide variety of species and plant parts were consumed by the *L. catta* inhabiting the areas in and around the reserve. Those species and types of eaten, likewise, generally correspond to those reported by previous researchers (see Sauther, 1992, 1998; Simmen et al., 2006; LaFleur and Sauther, 2015, Yamashita, 2015b). As also reported previously, dietary content varied both seasonally and monthly. Such variation appeared to be primarily related to food availability associated with changing species phenology (see Sauther, 1998).

CHAPTER IV:

A TOPOGRAPHIC ANALYSIS OF RING-TAILED LEMUR FOOD PROCESSING BEHAVIOR.

Part 1 Introduction and Methods.

In this chapter I examine the relationship between postcanine dental topography for the lower dental arcade from p4-m3 to feeding behaviors among BMSR *L. catta*. This chapter is divided into two main parts. In Part I, I examine the impact of increasing dental wear and tooth loss on tamarind fruit processing times as well as the expression of compensatory processing behaviors for this food in relation to measures of dental wear determined through GIS-based dental topographic analysis. In Part II, I examine the relationship of dental topography to food processing times, both for food item types (e.g., fruit, insects, leaves) and for food item types for individual food species.

While traditional methods for assessing dental functional morphology (e.g., shearing quotient) have proven effective for the determination of broad dietary categories (e.g., high SQ = leaves or other "tough" foods, low SQ = consumption of fruits or "hard" foods), such methods are difficult to use when assessing the functional capacities of worn teeth. Such difficulties arise as these measures typically require the presence of distinct morphological landmarks (e.g., shearing crests and cusps), that are often modified or removed through the course of tooth wear.

As a result the calculation of traditional measures of functionality becomes difficult for individuals with significantly worn teeth, thus limiting the utility of such measures for addressing relationships between dental wear and aspects of primate ecology, nutrition and behavior (Kay, 1975, 1981; Kay and Covert, 1984; M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004).

Over approximately the last 15 years, computer-intensive, three-dimensional methods for modeling dental morphology and function have been developed by a number of researchers (Zuccotti et al., 1998; Jernvall and Selanne, 1999; Ungar and Williamson, 2000; Ungar and M'Kirera, 2003; M'Kirera and Ungar, 2003; Ungar, 2004; Dennis et al., 2004, King et al., 2005; Evans et al., 2007; Boyer, 2009; Bunn et al., 2011; Godfrey et al., 2012; Klukkert et al., 2012a; Winchester et al., 2014; Cuozzo et al., 2014; Yamashita et al., 2015a). Although studies using 3D methods vary in their exact methods of data capture (e.g., laser scanning, confocal microscopy, or micro-cT) and modes of analysis [GIS Dental topographic analysis for slope, relief and angularity; Relief Index (RFI); Orientation Patch Count (OPC); Dirichlet Normal Energy (DNE), 3D calculation of compensatory shearing crests, etc.)], 3D methods have proven effective for interrogating a number of areas in dental-morphological and dental-ecological research. These include examinations of dental-diet relationships (Evans et al., 2007; Boyer, 2008; Bunn et al., 2011; Winchester et al., 2014), species-level patterns of dental wear and their ontogeny (Ungar and M'Kirera, 2003; Dennis et al., 2004; Klukkert et al., 2012a; Cuozzo et al., 2014; Yamashita et al., 2015a), the relationship of dental form to ecology (Dennis et al., 2004; Head, 2011; Godfrey et al., 2012; Cuozzo et al., 2014; Yamashita et al., 2015a), the impact of dental wear on reproductive ecology (King et al., 2005), and have also been successfully used for interpretation of the paleontological record (Ungar, 2004, 2007; Merceron et al., 2006; Boyer et al., 2010;

Godfrey et al., 2012). In addition, such 3D methods, particularly those which use topographic methods which do not require morphological landmarks, have proven effective for assessing the functionality of worn dental form (M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004; King et al., 2005; see below for details on worn tooth analysis).

In this study I use Geographical Information Systems (GIS)-based 3D dental topographic analysis to quantify worn tooth form and function. This method has been effectively utilized by a variety of researchers across a wide variety of primate taxa including great apes (M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Merceron et al., 2006; Klukkert et al., 2012a), African Cercopithecidae (Bunn and Ungar, 2009), howling monkeys (Dennis et al., 2004), and extinct hominins such as *Australopithecus* (Ungar, 2004). This methodology has also been successfully used for assessing dental-ecological relationships for BMSR ring-tailed lemurs (Head, 2011; Cuozzo et al., 2014; Yamashita et al., 2015a).

GIS-based topographic analysis models dental morphology as a landscape within Geographic Information Systems software packages (in the case of this study ArcMap 10.2, although other packages such as GRASS GIS and ESRI ArcView have also been effectively utilized to complete GIS analysis). Although initially designed to model the Earth's surface, GIS software can accept data from a number of sources, including point clouds and/or digital elevation models (DEMs) derived from laser scanning of high-resolution epoxy dental casts (see Chapter 2 for details). From these data, a tooth's entire occlusal surface can be modeled topographically using tools for assessing landscapes incorporated into GIS. By modeling the morphology of the entire tooth's surface simultaneously to produce mean topographic scores, this method does not require the use of specific morphological landmarks. GIS-based dental topographic analysis is thus well suited to analyzing the form of worn teeth, and has proven

effective for assessing the functionality of worn teeth and the progression of wear on a species-specific level (M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Merceron et al., 2006; Head, 2011; Klukkert et al., 2012a; Cuozzo et al., 2014; Yamashita et al., 2015a).

To date, a number of topographic measures have been utilized to assess dental morphology using GIS methods. The key measures produced during topographic analysis are: 1) *Slope*, 2) *Relief* and 3) *Angularity*. These measures may be used to assess wear state, as well as the functionality of worn teeth (see M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004). Slope refers to the average change in elevation across the tooth's surface, and generally decreases with escalating wear. Relief is defined as the ratio of occlusal surface area to planometric area, and functionally serves as an analog to shearing quotient. Finally, angularity refers to the rate of change in slope across the occlusal surface, and reflects the tooth's overall "jaggedness." A full description of each of these measures is available below.

With respect to dental senescence, angularity may be a particularly important measure. Tooth angularity is thought to produce multi-directional forces in food items facilitating their breakdown (Ungar and M'Kirera, 2003). It is also of note that unlike slope and relief which generally decline with wear state, angularity is typically maintained until severe wear occurs among primates taxa for which data are available (but see Klukkert et al., 2012; Glowacka et al., 2016). Angularity may serve as a measure of reduced tooth functionality (Ungar and Williamson, 2000; M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004), as declines in this measure appear to occur only with high wear states that are associated dental senescence, a marked drop in functional efficiency of the tooth for chewing (see Ungar, 2005; King et al., 2005). The hypothesis that a decline in angularity indicates reduced dental function has not been fully explored across primate taxa nor has it yet been linked to factors such as individual health

and nutrition. Moreover, occlusal angularity has not been assessed with regards to individual patterns of feeding behavior. Likewise, the impacts of angularity on the maintenance of dental functional morphology remains somewhat murky as several researchers have reported that angularity does decrease with wear state and dental functionally may be maintained through alternative morphological features, such as compensatory shearing crests (Klukkert et al., 2012a; Glowacka et al., 2016). However, it is notable that the greatest reductions in angularity in these studies were found for individuals with higher wear states generally, or for individuals of advanced age, suggesting that these individuals may have been approaching dental senescence (Klukkert et al., 2012a; Glowacka et al., 2016).

Linkages between angularity and feeding behaviors reported in this chapter may serve as a means for assessing if angularity does serve as an indicator of worn tooth functionality among nonhuman primates. For example, if angularity is negatively correlated with factors such as feeding bout duration, or food processing behaviors associated with tooth wear and loss (e.g., use of licking or open food items), this study can provide support for the hypothesis that this measure does, in fact, indicate a loss of dental functionality.

In addition to the measures of slope, relief and angularity, it is also possible to calculate the total 2D and 3D areas of a given tooth using GIS topographic methods. As topographic relief is calculated using both 2D and 3D area measures, these data are easily collected during analysis. 2D area refers to the planometric area of the tooth at the level of the occlusal plane where cropping occurs during analysis (see Chapter 2), while 3D area reflects the area of the tooth's occlusal surface above the occlusal plane. While I know of no studies which have used these measures to date, they likely provide a general measure of tooth wear and function as dental wear typically erodes both the 2D and, often more severely, the 3D area of teeth (Millette,

personal observations collected during topographic analysis). Likewise, the general ease of collecting these data suggest that, if effective, they may be a faster means of conducting topographic analysis than traditional slope, relief and angularity calculations.

It is also of note that this current study differs from most others in its examination of an extended portion of the postcanine dental arcade to better understand the effects of tooth wear. I also integrate data from both sides of the dental arcade, rather than focusing on just one side. All GIS-based topographic studies to date of which I am aware, only examine one or two lower molar positions (typically m1 or m2) in a unilateral fashion. Dental wear among nonhuman primates does not, however, necessarily occur only upon these teeth, but may occur for multiple positions and at different rates for each position dependent upon its use and specific location within the dental arcade. For example, dental wear and tooth loss among BMSR ring-tailed lemurs is a progressive phenomenon which starts at an early age immediately after teeth erupt, and begins typically around p4 or m1 before spreading to the rest of the dentition, starting with posterior positions and followed by anterior positions (see Sauther et al., 2002; Cuozzo and Sauther, 2006a; Cuozzo et al., 2010; Head, 2011; Cuozzo et al., 2014). Such patterns of tooth wear and subsequent loss at BMSR are associated with the use of tamarind fruit, and are also related to ring-tailed lemurs' dental morphology which is ill-suited to consuming this food item, which is both mechanically hard and tough and which is also large in size (Yamashita, 1998a, 2008b; Cuozzo and Sauther, 2006a; Cuozzo et al., 2010; Yamashita et al., 2012, 2015a; see below for more concerning this tamarind fruit-dental mismatch). While ring tailed lemurs typically utilize the posterior dentition to process tamarind fruit, patterns of dental wear and food processing behaviors observed at BMSR indicate that more than one tooth may be used to do so (Yamashita, 2003; Sauther et al., 2002; Cuozzo and Sauther, 2006a,b; Millette, personal

observations). Ring-tailed lemurs at BMSR with extensive tooth loss have also been observed to "target" teeth which remain in the dental arcade in order to process food items (Millette, personal observations). For example one individual (Black 226) was repeatedly observed to utilize his remaining third molar to process tamarind fruit despite the loss of all other postcanine positions. Thus, when attempting to understand the potential impacts of dental impairment on food processing and consumption, it is critical that a multiple tooth analysis is utilized as a single or two-tooth analysis cannot account for variability in tooth wear throughout the dental arcade, and /or the effects of variable wear on behavior. This study therefore utilizes mean values for all teeth across both sides of the dental arcade from to p4-m3 to assess the topographic measures slope, relief, and angularity, as well as for 2D and 3D area measurements.

In addition, as topographic measures are difficult to apply to missing teeth (as there is no surface on which to conduct analysis), this study also examines the sum of 2D and 3D areas for all positions present within the dental arcade. As these data do not include areas for missing positions, those individuals with extensive tooth wear and loss demonstrate lower area sum scores than those without, thus providing a measure of the available dental "working area" within the toothrow. As such, a 2D and 3D sum data may provide a topographic means for assessing dental function among individuals who are missing teeth, despite methodological limitations imposed by missing teeth for the traditional measures of slope, relief and angularity. A full description of each measure used in this study is presented below.

Dental Topography in Ring-tailed Lemurs. GIS-based dental topographic analyses have been conducted for BMSR ring-tailed lemurs by a number of researchers (e.g., Head, 2011; Cuozzo et al., 2014; Yamashita et al., 2015a), as well as for Tsimanampetsotsa *L. catta* by Yamashita et al.

(2015a). Data reported by Head (2011) and published in Cuozzo et al. (2014) focus on the effects of microhabitat variation on tooth wear patterns on m1 and m2 for young-aged individuals (2-4 years). This study examined wear for individuals from groups living in disturbed and non-disturbed areas of BMSR using the measure of angularity as well as by combining slope and relief into a single variable called "occlusal lift." These researchers observed that occlusal lift varied relative to microhabitat use (e.g., disturbed vs. protected), by troop (which use different habitats) and also by sex. In this case, troops which utilized marginal areas demonstrated higher wear than did those from within the protected reserve, suggesting that differences in mechanical properties of foods available and consumed by lemurs (as well as potential differences in the amount of grit on these resources) may result in divergent patterns of tooth wear. Likewise, use of the anthropogenically-altered camp area (where lemurs often eat foods discarded by humans) was associated with higher occlusal lift, suggesting consumption of easily-processed and less-abrasive human-sourced foods (see Sauther et al., 2006) by these individuals may result in reduced dental wear. Sex-based differences reported by this study (specifically for m2, but not m1), were also believed to be related to priority of access for females, who may be less likely to consume mechanically-challenging tamarind fruit than are males (see Gemmill and Gould, 2008). Overall, these data suggest that dental wear at BMSR can be detected at an early age (e.g., young adult) using topographic measures, and that the patterns of wear observed are linked to foods consumed and sex-based access to resources. Interestingly, this study also noted that angularity did not vary among individuals within this population. These data may suggest that this measure, in fact, does not decline with tooth wear and may be linked to the maintenance of dental function as reported by researchers working with other taxa (M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004; Bunn and Ungar, 2009; but see chimpanzee

results for Klukkert et al., 2012a). This study does not, however, indicate if angularity is related to differences in food processing or behavioral patterns associated with dental impairment.

In their study, Yamashita et al. (2015a) reported on ring-tailed lemur dental topography for m1 and m2 in relation to food item mechanical properties for individuals from areas in and around BMSR Parcels 1 and 2 as well as for individuals from Tsimanampetsotsa National Park (TNP). Data from this study indicated that individuals from BMSR Parcel 2 showed the greatest reductions of topographic measures followed, in order, by Parcel 1 and TNP. Angularity was furthermore most pronounced among individuals from TNP, in comparison to the BMSR parcels. Topographic measures indicating increased wear (e.g., slope and relief) were correlated with dietary mechanical variables of elastic modulus and toughness (but not hardness), with the overall diet being most mechanically-challenging at BMSR Parcel 2, and followed (in order) by BMSR Parcel 1 and TNP. These authors implicate dry-season consumption of tamarind fruit (in concert with this species' generally thin dental enamel) in both BMSR Parcels as the source of the greater dental wear found here in comparison to TNP where tamarind is limited, seasonal and thus rarely consumed (see LaFleur, 2012). Likewise, while these authors implicate tamarind as a source of wear at BMSR possibly due to dental fractures radiating from the Enamel-Dentine Junction similar to that reported by Lucas et al., 2008 and Constantino et al., 2009 and which have also been reported for ring-tailed lemurs during enamel microindentation mechanical testing by Campbell et al. (2012), they also suggest that dental wear for Parcel 2 may be higher than that of Parcel 1 due to the heightened inclusion of exogenous grit at this site (see Chapter on fecal nutrition for a further discussion of exogenous grit and tooth wear at BMSR).

While these studies provide important contextual information on sources of tooth wear for ring-tailed lemurs (e.g., both between and within populations of this species) in relation to

diet, microhabitat, sex status and food item mechanical properties, they do not investigate the potential impacts of such wear on individual behaviors (including those in compensation for dental impairment), nor the effect of dental wear on individual's capacity to process critical food items. These studies, furthermore, only focus on two teeth within the dental arcade, which limits their ability to generalize how wear may impact the use of the entire toothrow, or how the development of wear throughout the toothrow may be related to food processing. This study differs from these in that it seeks to assess how dental wear relates to how foods are consumed, rather than simply how those foods consumed impact the dentition. I build on these previous works by addressing how dental wear impacts behavior directly, and examine if dental wear results in a reduced capacity to process food items (e.g., dental senescence) rather than simply attempting to identify sources of tooth wear.

It is interesting to note, however, that both of these studies indicate that angularity may be a key factor in tooth morphology which maintains dental function through wear. Cuozzo et al. (2014) find that younger individuals do not demonstrate reductions in angularity despite reductions in slope and relief status. In those populations where significant dental impairment has been reported (BMSR Parcels 1 and 2), angularity was lower than at TNP where significant dental impairment has not been observed to occur. These sources, thus, provide tantalizing hints that angularity may serve as a measure of dental function in ring-tailed lemurs, although no direct evidence of this hypothesis is presented by their authors. This study's focus on food processing seeks to confirm the efficacy of this measure with respect to direct measures of feeding behavior (see below).

Tamarind Fruit and Dental Wear at BMSR. As I noted in the introductory chapter, BMSR *L. catta* demonstrate exceptionally high rates of dental wear and subsequent wear related tooth loss as a function of high rates of tamarind fruit feeding, particularly during the resource depleted dry season (Sauther et al., 2002; Cuozzo and Sauther, 2006a,b; Sauther and Cuozzo 2009; Yamashita et al., 2012; Yamashita et al., 2015a,b). Dental wear and tooth loss observed at this site is associated particularly with postcanine positions associated with the dental processing of this food item, which is the most mechanically-challenging food regularly consumed by BMSR *L. catta* (Yamashita, 2003, 2008b; Cuozzo and Sauther, 2004, 2006a,b; Cuozzo et al., 2010; Yamashita et al., 2012). Such wear in turn appears to be the result of a dental-dietary mismatch between this fruit (which is both hard and tough) and ring-tailed lemur's relatively thin dental enameled and somewhat folivorously-adapted postcanine dentition (Kay et al., 1978; Lucas, 2004; Lambert et al., 2004; Godfrey et al., 2005; Cuozzo and Sauther, 2004, 2006a,b, 2008, 2013; Sauther and Cuozzo, 2009).

Extensive tooth wear such as that seen at BMSR is often presumed to result in a reduced capacity to process, masticate and digest food items, particularly challenging foods such as tamarind fruit. Tamarind fruit is an important fallback resource during the dry season (Sauther, 1992, 1998; Simmen et al., 2006; Sauther and Cuozzo, 2009). Although this food is consumed during both the wet and dry seasons, it appears to fit Marshall and Wrangham's (2007) definition of a staple fall back food as it may serve as the only fruit food source when other preferred foods are unavailable (Sauther and Cuozzo, 2009). Thus, failure to access this food item may have significant consequences for the animal in terms of meeting its nutritional needs, potentially impacting health, survival and reproduction. Such difficulties consuming this food have been reported in previous work that I have conducted at the site focusing on dental impairment and the

consumption of tamarind fruit. First, individuals with tooth loss spend more time processing this food item than those without tooth loss, suggesting that these individuals are less capable of accessing this food resource (Millette, 2007; Millette et al., 2009). During this study, individuals with tooth loss were also observed to engage in more frequent licking of tamarind fruit than those without, suggesting that they utilize this method to process in lieu of dental processing. Secondly, in addition to demonstrating larger fecal particle sizes, during the dry season, individuals with tooth loss have also been observed to show fewer fruit particles in their fecal material, suggesting that they do not as effectively consume this food item in comparison to those who remain unimpaired (Millette et al., 2012 / Chapter 6). These previous studies indicate that tamarind fruit is more difficult for individuals with tooth loss to process than for those without, but also indicate that individuals may utilize compensatory behaviors to access this fruit through alternative means. Such behavioral mechanisms, if present, may allow individuals to survive and reproduce in relatively good health despite tooth wear and loss, as has been reported for BMSR *Lemur catta* (see Sauther et al., 2002; Cuozzo and Sauther, 2004).

A primary goal of this chapter is to examine the relationship between dental topography and tamarind use by examining patterns of dental topography for both the left and right dental arcade from p4-m3 for all study individuals in relation to a variety of behavioral measures related to tamarind fruit processing. These behavioral measures include feeding bout duration data, as well as food processing behaviors including: 1) use of manual food processing, 2) consumption of open tamarind fruits, 3) use of tamarind fruits which have been previously opened by conspecific individuals, 4) divergent use of the dental arcade (e.g., posterior vs. anterior), 5) use of licking-based processing behavior and 6) consumption of tamarind fruit from the ground. When combined with dental morphological data, these measures provide a broad-

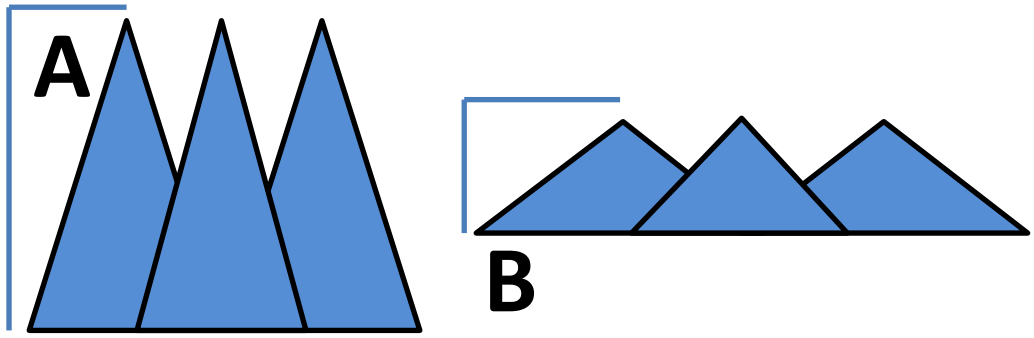
based framework for assessing how tamarind fruit processing and consumption is impacted by dental impairment, as well as a means for examining how individuals may or may not engage in behavioral compensation for such impairment. A full description of all behavioral types is provided below.

Topographic Data Analysis Measures.

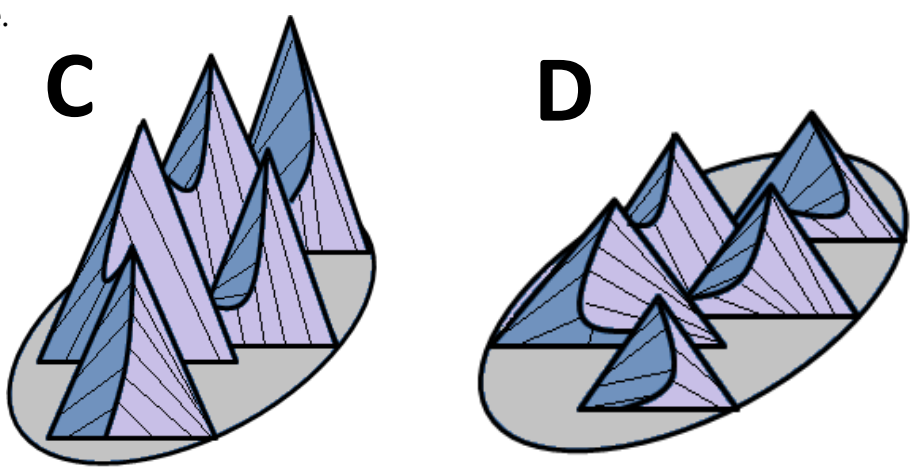
I collected dental topographic data for 31 individuals (19 Female, 12 Male) based on scans conducted at the University of Arkansas Paleoanthropology Laboratory and which were made upon casts produced from dental impressions collected by Dr. Frank Cuozzo during annual health inspections conducted during 2011 and 2012. GIS dental topographic analyses were conducted using methods developed by members of the Paleoanthropology Lab and presented in Klukkert et al. (2012b). A full description of topographic data collection techniques utilized by this study is available in the chapter focusing on methods (Chapter 2). A variety of topographic measures were collected for the dentition from p4 to m3 on both sides of the tooth row. Each topographic measure reflects a different aspect of tooth (or toothrow) morphology, and provides a theoretically different indicator when concerning the functional implications of tooth wear (see introductory section above). A description of the topographic measures used here are as follows: Visualizations for each measure and for topographic outputs are available in Figures 4.1-4.3.

Mean Slope. Topographic slope represents the average slope of the tooth's occlusal surface (e.g., the average rise / run from one digital elevation model point to the next across the tooth's surface). Teeth with high slope demonstrate more vertical surfaces than those with low slope. Worn teeth generally demonstrate reduced slope in comparison to those which are less worn

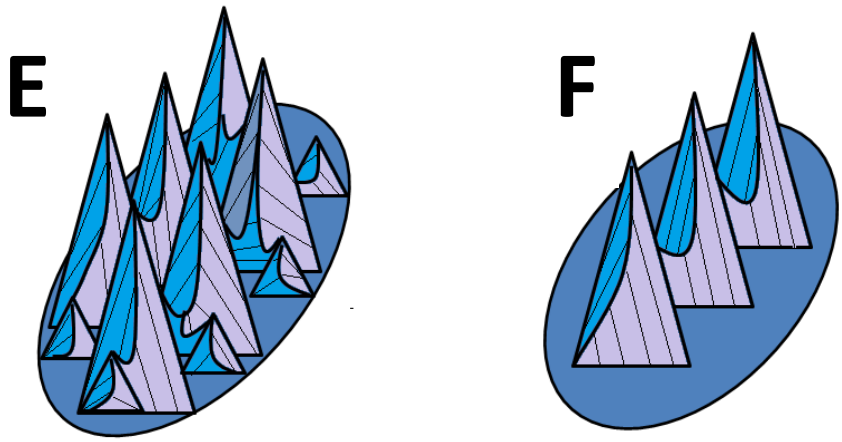
Figure 4.1. Graphical depiction of slope, relief and angularity.



Slope represents the average change in elevation (e.g., rise / run) across the tooth's surface. **A** represents a tooth of higher slope, while **B** represents a tooth with lower slope.



Relief represents the 3D surface area of the tooth to its 2D planometric area at the elevation of cropping. **C** shows an idealized tooth of higher relief than for tooth **D.**



Angularity is the average change in slope over the tooth's surface. **E** shows an idealized tooth of high angularity, while **F** shows a tooth with reduced angularity. All major cusps demonstrate the same slope, although angularity is higher for **E.**

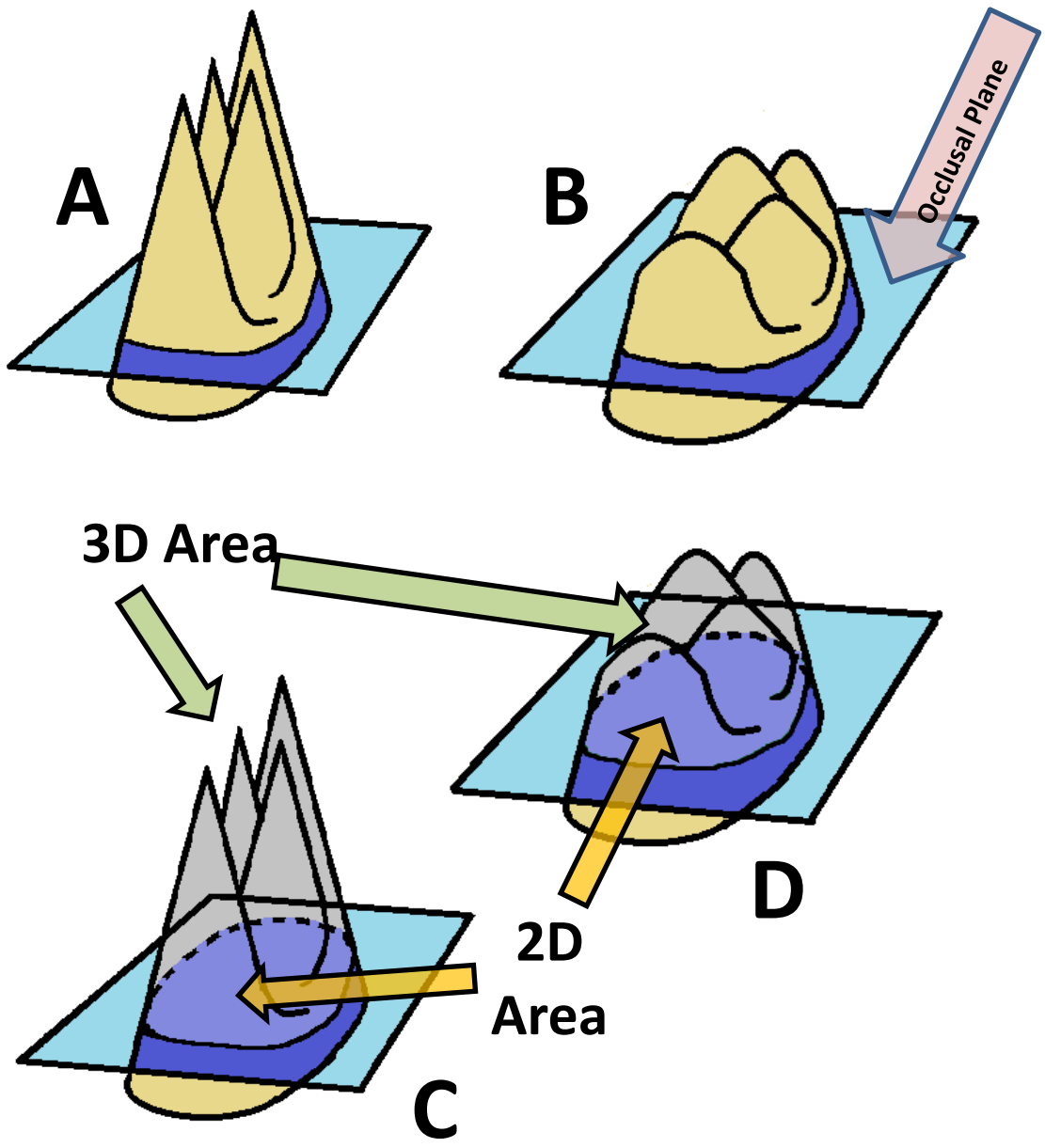


Figure 4.2 A depiction of 2D and 3D area measures. A and **B** show idealized teeth with high (**A**) and low (**B**) relief. The "occlusal plane" is the plane of cropping at the lowest z-point of the talonid basin. **C** and **D** show 2D and 3D area measures. **2D area** is that circumscribed by the occlusal plane. **3D area** reflects all area above the occlusal plane. **Relief** reflects 3D / 2D area. For both teeth, **2D area** is similar, although **C** is of higher 3D area than **D**. **C** thus has a higher relief index than does **D.**

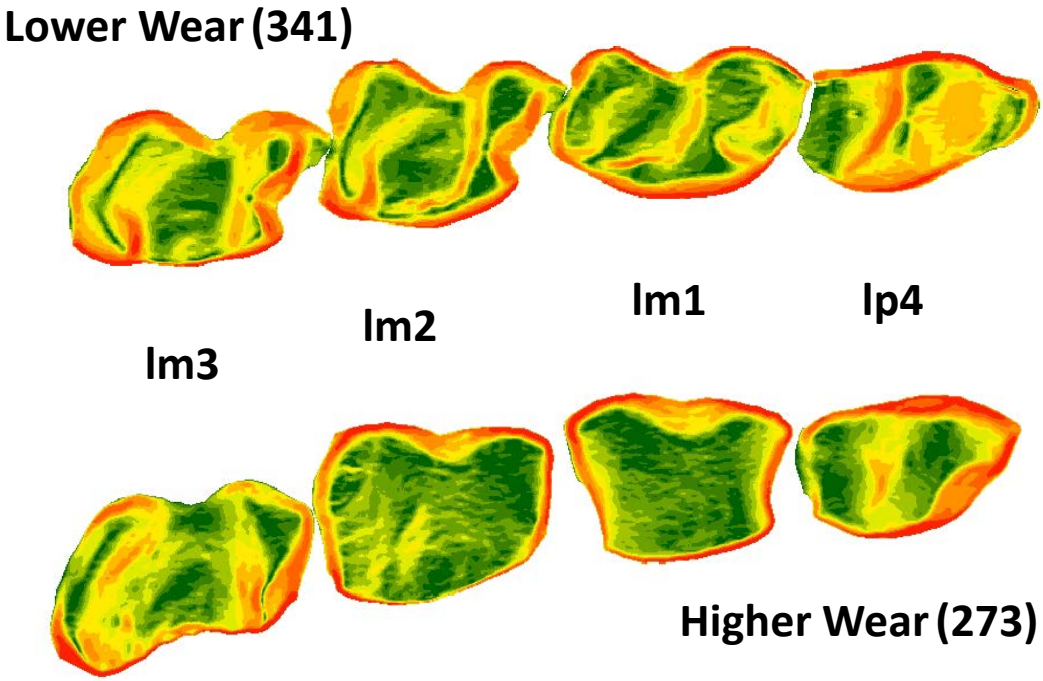


Figure 4.3. A visualization of dental topographic data generated during GIS analysis. Toothrows from two individuals are shown, one with a low amount of wear (341) and one with a higher amount of wear (273) for the measure of slope where green represents areas of low slope and red indicates areas of higher slope). Note the general reduction of slope (and relief) found between these two individuals, although all teeth are present within the dental arcade.

(M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Ungar and Bunn, 2008; Bunn and Ungar, 2009). For this study, the slope value for each individual tooth present from p4-m3 was utilized to calculate a mean slope value used during analyses for food processing. When means were determined, missing teeth were excluded from this calculation as slope cannot be determined for such positions. Thus, the mean slope values utilized reflect only the average slope of those teeth present and do not integrate data from missing positions.

Mean Relief. Relief is defined as the relationship of the tooth's occlusal surface 3D area to the 2D planometric area of the occlusal surface at the z-axis level at which the tooth was cropped (see Chapter 2). Relief is analogous to shearing quotient in terms of functional morphology [e.g., tough diets are associated with high relief, while fruit based/or hard diets are associated with lower relief) see M'Kirera and Ungar, 2003; Boyer, 2008; Winchester et al., 2014)], and generally decreases as wear increases (see Ungar and M'Kirera, 2003; M'Kirera and Ungar, 2003; Ungar and Bunn, 2008; Bunn and Ungar, 2009; Klukkert et al., 2012a, Yamashita et al., 2015a.). As with slope, mean relief was calculated for those teeth present on both sides of the dental arcade, and thus does not directly integrate the impact of tooth loss on dental processing capacity.

Mean Angularity. Angularity reflects the average change in slope (e.g., the derivative of the slope) across the surface of the tooth. Angularity reflects the overall "jaggedness" of the tooth, and is thought to be a key feature in maintaining tooth function through the course of wear (see Ungar and M'Kirera, 2003; Dennis et al., 2004) as angularity typically only changes with extreme tooth wear. Mean angularity was calculated from the scores of those teeth present in the

tooth row from p4-m3, and as with slope and relief does not integrate values for those positions for which teeth were missing from the toothrow.

2DAverage. This measure refers to the average size of the 2D area for each tooth present in the dental row. This data was generated using 2D area calculated to produce relief values. As with the aforementioned measures, data for missing teeth were not included in the calculation of this value. 2DAverage is believed to be reduced among those individuals with extensive tooth wear as this process often leads to the reduction of the tooth's area along the occlusal plane (e.g., the point at which the tooth was cropped, see Chapter 2). To my knowledge, the overall 2D average area of positions within the toothrow has not been used in any primate topographic study to date.

2DSum. This measure refers to the summed 2D areas of each tooth across the dental arcade from All teeth present were utilized in the calculation of this value (from p4-m3), while missing teeth were not included in this value as 2D area measures were possible for these positions. As with 2DAverage, to my knowledge, no topographic study has yet utilized this measure across the toothrow. As 2DSum does not consider areas from missing teeth, but sums only those positions present, this measure may serve as a means for assessing the impact of tooth loss on food processing behavior as individuals with tooth loss will have fewer positions summed than those without tooth loss for the final value used when examining behavioral measures.

3DAverage. Similar to 2DAverage, 3DAverage reflects the average 3D surface area of the teeth present for both sides of the dental row from p4-m3. This data was derived from that used to calculate relief scores for each individual teeth, and as with 2DAverage, was not determined

using values for missing positions. As with 2DAverage, 3DAverage values are expected to decline with increasing wear as this worn teeth generally demonstrate a reduced occlusal surface. To my knowledge this measure has not been used in any topographic study to date.

3DSum. 3DSum measures the total sum of 3D area present on the tooth row from p4-m3 on both sides of the dental arcade. As this measure does not include 3D areas of teeth which are missing, it likely allows for analysis of the impacts of tooth loss on food processing capacity as individuals with tooth loss will have a reduced number of positions summed in comparison to those without tooth loss. As with other area measurements, this value is believed to decrease with increasing wear and tooth loss as these processes remove enamel from the teeth. This measure has not been utilized in any topographic study of which I am aware.

Tamarind Feeding Behavioral Measures.

Tamarind feeding was recorded as a subset of data collected for all feeding behaviors during 90-minute focal follows conducted for each study animal on a minimum of a twice-monthly basis from July 2012 - March 2013. All feeding bouts, including those for tamarind feeding, were recorded using an all-occurrences sampling strategy, where each feeding bout was recorded in concert with all other forms of behavioral data (e.g., instantaneous data, agonism data, etc.). Full descriptions of behavior data collection methods are available in Chapter 2.

A number of variables were recorded with respect to tamarind feeding during observations. In addition to tamarind fruit feeding bout durations, a range of food processing behaviors associated with this fruit were recorded during observations. A description of each processing behavior associated with tamarind feeding is as follows:

Feeding Bout Duration. Tamarind feeding bout durations have been positively associated with tooth loss status among ring-tailed lemurs at BMSR, where individuals with tooth loss demonstrated significantly longer feeding bout lengths than those without tooth loss (Millette et al., 2009). In contrast, in this study I seek to assess the impact of topographic measures (allowing assessment of tooth wear in addition to tooth loss) on feeding behavior. Tamarind fruit feeding durations were recorded during each feeding bout. As detailed in Chapter 2, durations were calculated automatically using a recursive "=Now(" function in Microsoft Excel. The duration of each feeding bout was recorded in real time during behavioral focal follows of each animal conducted from July 2012 – March 2013. Feeding bouts commenced when the animal placed the fruit in the mouth and began to orally process and/or masticate the food item. Bouts were ended once the individual finished consuming the food item completely, or discarded the food item. Likewise, bouts were ended if the animal stopped consuming the fruit for a period of 5 seconds or more, as the fruit was typically discarded after this amount of time of non-feeding. If feeding was resumed after this 5 second period, the bout was then restarted and the total time of consumption used in analysis. Additionally, tamarind fruit is frequently licked during consumption. As they were typically interspersed with dental processing, and could not easily be subtracted from total dental processing times, licking behaviors were included in the total tamarind feeding times recorded. Those bouts with suspect durations (e.g., those where either a start or end time was not properly recorded, or which were clearly wrong due to miss-entry) were removed from analysis during the data cleaning phase of this project.

The calculation of bout lengths for tamarind was designed to reflect only the use of one fruit at a time, although occasionally two fruits may have been consumed during a bout due to poor observation conditions or if the animal switched to another fruit so quickly that two bouts

could not accurately be recorded. However, the vast majority of bouts reflect only single tamarind fruit. To ameliorate this potential issue, both mean and median durations were utilized during analysis as median durations should be less susceptible to the inclusion of a small number of multiple-fruit feeding bouts than are mean durations.

Manual Tamarind Processing. Tamarind fruit possesses a mechanically-challenging outer shell which requires removal prior to consumption of the fruit's softer inner portion. Ring-tailed lemurs typically remove this shell using the posterior dental apparatus to break through and subsequently fragment the shell. These fragments are then either discarded or simply consumed by the animal (Millette, personal observations).

In contrast, a number of individuals were observed to manually remove the shells of ripe tamarind fruit using the hands. This behavior has been previously observed among BMSR *L. catta*, but to date no data are available to assess the direct relationship between manual processing and dental impairment (see Millette et al., 2009). Manual processing was recorded if the shell was primarily removed using the hands rather than using the dentition, although occasionally this behavior was recorded if the shell was partially processed dentally but primarily removed using the hands. Such use of the hands in concert with the dentition was rare, and typically no dental action accompanied manual processing. In most instances manual processing was used to fully remove the fruit's shell in one motion. This was usually done with the fruit still attached to the tree by its stem to provide stability necessary to remove the shell, and where the animal would first grasp the area around the stem with one hand before using the other to pull the shell from the fruit. Once removed, the shell was discarded by the animal, and the edible portion of the fruit was consumed. Occasionally, manual processing was observed for

tamarinds which had fallen from the tree, but in such instances, removal of the majority of the shell using the hands was required for this behavior to be recorded.

Use of Open Tamarind Fruit. The consumption of tamarind fruit with shells opened prior to initial processing by the focal animal was frequently observed during this study. Opened fruit is likely easier to process into a form suitable for consumption than are fruits where the shell maintains its structural integrity. I recorded the use of open fruit if the shell was clearly fractured prior to initial processing by the animal. A conservative scoring system was used when scoring open tamarinds, and typically internal portions of the fruit were visible when this behavior was recorded. "Possibly open" was recorded if the fruit appeared to be open, but could not be fully confirmed either due to observation conditions or due to rapid consumption by the animal that prevented a clear view prior to processing and ingestion.

All fruits with clearly fractured shells prior to processing were recorded as "open," and no specific source of fracture was required as was the case for "pre-processed" tamarind fruits (see below). Tamarind fruit appeared to be opened by a number of potential sources. First, extremely ripe and/or extremely old fruits often appeared to be open simply due to their age, with the older pods appearing to be opened simply as a result of weathering. In addition, many fruits on the ground appeared to be opened due to their fall to the forest floor, or potentially by being crushed by humans or their livestock transiting through the forest. Open fruit may have also been the result of lemurs (either *L. catta* or sympatric *Propithecus verreauxi*) opening and rejecting a specific fruit or due to birds such as the sickle billed vanga which commonly consume this food (Millette, personal observations). Human-opened tamarind fruit from the camp area

was not included with open fruits, although only one bout of tamarind opened by humans was observed to have taken place.

Use of Conspecific-Processed Fruit. Use of pre-processed food items has been observed previously at BMSR by Cuozzo and Sauther (2006a), although no quantitative data are yet available with which to assess the relationship of this behavior to dental impairment status. As with pods opened prior to processing by the focal animal generally, use of conspecific-processed tamarinds should provide individuals with dental impairment with an increased ability to use this food source. In contrast to data collected for "open" tamarinds, use of pre-processed tamarinds was only recorded if a conspecific individual was observed to have opened a fruit prior to consumption by the focal animal. For use of pre-processed tamarind to be recorded, the tamarind had to be directly opened and subsequently discarded by another *L. catta* individual. In this case, the animal which processed the fruit initially could open the fruit in any manner, either manually or through use of the dentition. As when recording "open" fruit, a conservative approach was used when recording use of this behavior. As it was often difficult to observe the animal that processed the fruit, if a fruit had been potentially processed prior to consumption by the focal animal, this was recorded rather than a confirmed use of pre-processed fruit.

Location on the Toothrow of Tamarind Processing. As with most large and/or mechanically-challenging food items, tamarinds are generally processed on the posterior dentition (e.g., the postcanine positions distal to C1/p2). Such posterior processing of tamarind fruit also frequently results in the wear and loss of teeth for these positions (Yamashita, 2003; Cuozzo and Sauther, 2004; Cuozzo and Sauther, 2006a). The ablation of these positions is likely to result in a reduced

capacity to dentally process tamarind fruit using the standard posterior fashion. As a result, individuals may utilize remaining anterior positions to process tamarind fruit in compensation for the loss of postcanine functionality.

For each feeding bout, I recorded the location on the toothrow of tamarind processing. If the animal processed the fruit using positions distal to the upper canine (e.g., on the premolars and molars), this was recorded as being a "posterior" bout. If the fruit was only processed on the canine or mesially (including use of licking if no posterior use was observed), the feeding bout was recorded as being an "anterior" bout. If both anterior and posterior positions were utilized, this was recorded as a feeding event where "both" positions were utilized. As it was expected that most individuals would utilize the posterior dentition to process tamarind, and that individuals with dental impairment would use the posterior dentition to process this food less frequently, analysis was limited to examination of the dental topography to the frequency of posterior toothrow use. That said, as it was often difficult to determine all times the posterior dentition was used clearly, in a manner where similar to where "possible" observations were examined for other behaviors, topography was also assessed in relation to the use of both posterior and anterior dentition as well.

Use of Licking Behavior to Process Tamarind Fruit. Previous research at BMSR indicates that tooth loss is associated with higher frequencies of licking behavior to process and consume tamarind fruit (Millette, 2007; Millette et al., 2009). This research, however, did not examine if such behavior was related to wear state, either prior to, or following, tooth loss. Likewise, no study has examined this behavior with respect to dental topography.

For this study, I recorded licking behavior if the focal animal was observed to use the tongue to process tamarind fruit at any point during a single feeding bout. Similar use of licking behavior has been reported by other researchers at this site as a means for processing tamarind fruit (Sauther, 1992; see also Millette et al., 2009). Licking was not recorded if the individual utilized only a single lick (e.g., as if to primarily taste the fruit), but was recorded only if the animal repeatedly used the tongue to process and break down the fruit. Possible licking was also recorded if the animal appeared to be licking a fruit, but visibility was too poor to confirm the use of the tongue (e.g., the animal's head was moving as if licking, but the tongue itself could not be seen).

Use of Tamarind Fruit from the Ground. Tamarind fruit is used by individuals both from arboreal sources (e.g., directly from the source tree) and on the ground (e.g., fruit which has fallen or has otherwise been removed from its source). Based on personal observations, fruits found terrestrially were frequently of lower quality, but also of reduced mechanical integrity because they had remained on the forest floor for some time. Such fruits often appeared to be less challenging mechanically than those found within the trees and were frequently very easy to open upon examination and/or manipulation. Many fruits on the ground also appeared to be open, either due to their fall to the forest floor or trampling by humans and their animals which often utilize areas in and around the reserve (Millette, personal observations). It was therefore anticipated that individuals with higher amounts of dental impairment would preferentially use tamarind fruit found on the ground. Use of tamarind fruit from the ground was recorded if the fruit was clearly found upon the ground prior to processing and consumption. If the tamarind

fruit was found in any other state, (e.g., in the trees, on human structures, etc.) it was not recorded as being a "ground tamarind."

Data Analysis Methods.

The relationships between topographic measures and feeding behaviors were assessed utilizing non-parametric methods (Spearman's correlations). Non-parametric tests of significance were utilized as measures of dental topography are typically not interval/ratio-scaled data, but rather reflect an ordinal measure of dental form. Previous research utilizing topographic measures of primate dental morphology have typically utilized rank-ordered data when assessing statistical significance (see M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004, Klukkert et al., 2012a, etc.), thereby converting the data into ordinal form. Additionally, for some of the behavioral measures, particularly manual processing, not all individuals engaged in the behavior being recorded, or did so at very low rates in comparison to others, thus the datasets tended to not be normally distributed. In this case, use of ordinal methods is likely more appropriate than parametric methodologies. That said, parametric analyses were utilized for examining tamarind feeding durations in relation to dental area measurements. This was done as in both cases, the data were scaled properly for analysis (e.g., both were interval-ratio scaled) using parametric correlations (e.g., Pearson's r), likewise all individuals engaged in this behavior allowing for a proper distribution of the dataset.

For feeding durations, and parametric correlations for area measures, Pearson's correlations were used to assess the relationship between each topographic measure and feeding bout mean and median durations (see below). For food processing behaviors, frequencies of the occurrences of each were calculated from total counts of each behavior in comparison to the total

number of feeding bouts recorded. In addition, as the total number of feeding bouts included those where “unknown” was recorded, or the specific behavior was not recorded (i.e., it was still unknown), for all food processing behaviors except ground tamarind feeding (where no unknowns were recorded), the frequency of each behavior was recorded relative to the total number of observations where the behavior was clearly recorded to those where it clearly did not occur (e.g., Yes / Yes + No). All behaviors were also examined for frequencies including possible occurrences of the behavior (Yes + Possible / Yes + No + Possible). For tamarind processing location, the same scheme was followed, but instead of possible observations, this case examined simultaneous use of the anterior and posterior dentition (e.g., both areas of the toothrow) in addition to the use of the posterior dentition in comparison to all feeding bouts and those where only use of the anterior or posterior dentition were recorded.

In addition to being examined for all individuals overall (n = 31), each behavioral measure was examined by individual sex class. For females, 19 individuals were available for analysis, while 12 males were assessed. Statistical power is thus highly reduced for male-only analyses, and the interpretation of the results should reflect the reduced sample size among males for which topographic data are available. It is also of note that two males went missing during this study (Blue 218, and Teal 339), during September and November respectively. As individual frequencies, rather than individual counts, were utilized for the analysis of feeding behaviors, these individuals were recorded in the analyzed dataset. However, it should be noted that wet season observations are not generally available for these individuals, although seasonality was not directly examined during this chapter. For all tests, significance was set at the $\alpha \leq 0.05$ level with all results rounded to two decimal places (see Weiss, 2011).

Tamarind Fruit Processing Hypotheses.

A number of hypotheses were examined with respect to tamarind fruit processing. These were based on the assumption that reduced scores for each topographic measure are indicative increasing tooth wear or loss (see Ungar and M'Kirera, 2003; M'Kirera and Ungar, 2003; Dennis et al., 2004, Bunn and Ungar, 2009; Klukkert et al., 2012a; Yamashita et al., 2015a). Previous studies using dental topographic analysis generally report that increasing wear is associated with reduced slope, relief and angularity, although reductions in angularity may only result from exceptionally high wear states (e.g., Ungar and M'Kirera, 2003; M'Kirera and Ungar, 2003; Dennis et al., 2004; Cuozzo et al., 2014; Yamashita et al., 2015a). Although not used by previous studies the measures of 2DAverage, 3DAverage, 2DSum and 3DSum are also believed to be indicative of dental impairment, as wear should remove surface area from the dentition over time. I visually observed such a reduction in surface area during cast preparation and analysis, although reductions in surface area have not been quantified for BMSR *L. catta* prior to this study. 2DSum and 3DSum scores, in particular, should reflect dental impairment related to tooth loss as these represent the sum of all positions (p4-m3) where missing positions are entered as having no surface area, which should result in a lower total surface area than if even the base of the tooth remains within the dental arcade.

Ha1: Tamarind feeding durations: All topographic measures will be negatively correlated with the duration of each tamarind fruit feeding bout, quantified either using individual median or mean durations as a measure of central tendency.

Ha2: Manual processing of tamarind fruit: All topographic measures will be negatively associated with the manual processing of tamarind fruit, as topographic measures decline the frequency of manual tamarind fruit processing will increase.

Ha3: Use of open tamarind fruit: Use of open tamarind fruit was expected to be greater among those subjects with dental impairment. All topographic scores were hypothesized to be negatively correlated with frequencies of open tamarind fruit use.

Ha4: Use of conspecific-processed tamarind fruit: Use of conspecific processed tamarind was expected to be greater among subjects with dental impairment. All topographic scores were hypothesized to be negatively correlated with frequencies for use of pre-processed tamarind fruit.

Ha5: Toothrow processing location: Tamarind fruit is typically processed upon the posterior dentition, which is also where tooth wear is most common within this population. It was hypothesized that individuals with reduced topographic scores would demonstrate reduced amounts of processing along on the posterior dentition, or would demonstrate reduced amounts of processing on both the anterior and posterior dentition (e.g., “both” where recorded during observation).

Ha6: Use of licking behavior to process tamarind fruit: Licking behavior was hypothesized to be associated with dental impairment. All topographic measures were hypothesized to be negatively correlated with individual frequencies of licking behavior during tamarind fruit feeding.

Ha7: Use of tamarind from the ground: Tamarind fruit from the ground was frequently used by BMSR *L. catta*. Such fruit was apparently less mechanically challenging than fresh fruit found in trees. I thus hypothesized that use of tamarind fruit from the ground would be associated with lower topographic scores for all measures.

Tamarind Fruit Processing Results.

Feeding Bout Duration and Dental Topography. The relationship of tamarind fruit feeding bout length to dental form was calculated for all topographic measures using non-parametric correlations (Spearman's rho). While nonparametric tests of significance have generally been used in previous studies using GIS-based dental topography (see Ungar and M'Kirera, 2003; Dennis et al., 2004, Klukkert et al., 2012a, etc.), it is likely that topographic measures do not provide data scaled in a manner required necessary for parametric tests (i.e., the topographic scores of slope, relief and angularity may not scale linearly). In addition, tamarind feeding durations were assessed using both mean and median durations for each individual. Tamarind feeding bout lengths varied widely, with several instances of extremely long bouts being present (e.g., bouts of 9 minutes in length). Such long bouts may reflect the consumption of more than one tamarind, and may result in higher-than representative mean values. Median values are thus provided as the vast majority of tamarind feeding bouts only included one tamarind. In addition, the relationship of dental topography to feeding bout duration was assessed for both sexes combined as well as individually.

When all individuals were included (N = 31), the only topographic variable which demonstrated a significant positive correlation was for angularity in relation to mean tamarind

bout duration (rho = 0.4097; p = 0.0221). No other topographic measures (e.g., Slope, Relief, 2DAverage, 2DSum, 3DAverage, or 3DSum) were significantly correlated with feeding bout length; either when using the mean or median duration values (see Table 4.1 and Figure 4.2).

For females, the only significant correlation between feeding bout duration and topography was a negative correlation between 3DAverage and median bout length (rho = -0.0507, p = 0.0507). In contrast, for males angularity was significantly and positively associated with mean feeding bout length (rho = 0.6354, p = 0.0261). These data suggest that males likely drive the overall correlation between angularity and feeding bout length in this sample (Tables 4.1 and 4.2).

Table 4.1. Tamarind Feeding Bout Duration by Topographic Measure (Spearman's rho).

	Total mean		**Total median**		**Female mean**		**Female median**		**Male mean**		**Male median**	
Measure	**rho =**	**p =**	**rho =**	**p =**	**rho =**	**p =**	**rho =**	**p =**	**rho =**	**p =**	**rho =**	p =
Slope	-0.077	0.6805	-0.0706	0.706	-0.2947	0.2206	-0.3912	0.0977	0.2028	0.5273	0.1958	0.5419
Angularity	0.4097	***0.0221***	0.2871	0.1174	0.3772	0.1114	0.207	0.3951	0.6364	***0.0261***	0.4056	0.1908
Relief	-0.0004	0.9983	-0.0391	0.8345	-0.1456	0.552	-0.2561	0.2898	0.2098	0.5128	0.0769	0.8122
2DAverage	0.0504	0.7877	-0.1165	0.5324	-0.207	0.3951	-0.3842	0.1044	0.1049	0.7456	-0.0979	0.7621
2DSum	-0.0855	0.6475	-0.1077	0.5643	-0.3825	0.1061	-0.4088	0.0823	0.1399	0.6646	-0.007	0.9828
3DAverage	-0.0105	0.9554	-0.1	0.5925	-0.2825	0.2413	-0.4544	***0.0507***	0.3217	0.3079	0.1259	0.6967
3DSum	-0.0629	0.7367	-0.0694	0.7108	-0.3684	0.1206	-0.4035	0.0867	0.3497	0.2652	0.1818	0.5717

Table 4.2. Tamarind Feeding Bout Duration by Topographic Measure Summary (Spearman's Correlations).

	Total		Female		Male	
	mean	**median**	**mean**	**median**	**mean**	**median**
Slope	↔	↔	↔	↔	↔	↔
Angularity	↗	↔	↔	↔	↗	↔
Relief	↔	↔	↔	↔	↔	↔
2DAverage	↔	↔	↔	↔	↔	↔
2DSum	↔	↔	↔	↔	↔	↔
3DAverage	↔	↔	↔	↘	↔	↔
3DSum	↔	↔	↔	↔	↔	↔

While likely inappropriate for the topographic measures of slope, relief and angularity, parametric measures may, however, be appropriate for area measurements as these demonstrate interval-ratio scaling (e.g., the difference between data points is linearly-scaled). For parametric pairwise correlations of area, 2DAverage, was negatively correlated with both mean and median bout length for all individuals (mean: r = -0.03568, p = 0.0488; median: r = -0.4179, p = 0.0193). A negative trend was also observed for both mean and median durations with respect to 3DAverage (mean: r = -0.3357, p = 0.0648; median: r = -0.3357, p = 0.0648; median: r = -0.03409, p = 0.0606). When examined by sex, females demonstrated significant negative correlations for 2DAverage for both mean and median bout lengths, as well as for 2DSum, 3DAverage and 3DSum (2DAverage mean: r = -0.5916, p = 0.0076; 2DAverage median: r = -0.6599; p = 0.0021; 2DSum mean: r = --0.5403; p = 0.0169; 2DSum median: r = -0.5068; p = 0.0268; 3DAverage mean: -0.05457; p = 0.0157; 3DAverage median: r = -0.5833; p = 0.0088; 3DSum mean: r = -0.4915, p = 0.0326; 3DSum median: -0.4587; p = 0.0482). No similar

patterns were noted among males, suggesting that females are driving the overall patterns observed. Interestingly, when examined parametrically, angularity values are also positively associated with longer feeding bouts, overall and mean values females (All individuals mean: r = 0.4927; p = 0.0049; All individuals median: r = 0.4105; p = 0.0218; Female mean: 0.4902; p = 0.0315; Female median trend: r = 0.4165, p = 0.0761). These data may indicate that topography in terms of angularity is associated positively with tamarind feeding bout length (Table 4.3 and 4.4).

Table 4.3. Tamarind Bout Durations by Topographic Measure (Parametric Correlations).

	Total mean		**Total median**		**Female mean**		**Female median**		**Male mean**		**Male median**	
Measure	**r =**	**p =**	**r =**	**p =**	**r =**	**p =**	**r =**	**p =**	**r =**	**p =**	**r =**	**p =**
Slope	-0.1836	0.3228	-0.0709	0.7045	-0.3044	0.2051	-0.2471	0.3077	0.0002	0.9995	0.245	0.4428
Angularity	0.4927	***0.0049***	0.4105	***0.0218***	0.4942	***0.0315***	0.4165	*0.0761*	0.5025	0.096	0.4119	0.1833
Relief	-0.1075	0.5649	-0.0373	0.8423	-0.1729	0.4791	-0.1617	0.5085	-0.0451	0.8893	0.144	0.6553
2DAverage	-0.3568	***0.0488***	-0.4179	***0.0193***	-0.5916	***0.0076***	-0.6599	***0.0021***	-0.0526	0.8711	-0.1114	0.7303
2DSum	-0.2806	0.1262	-0.2886	0.1153	-0.5403	***0.0169***	-0.5068	***0.0268***	-0.0424	0.896	-0.1077	0.7391
3DAverage	-0.3357	*0.0648*	-0.3409	*0.0606*	-0.5457	***0.0157***	-0.5833	***0.0088***	-0.0627	0.8465	-0.0141	0.9653
3DSum	-0.2755	0.1336	-0.2588	0.1597	-0.4915	***0.0326***	-0.4587	***0.0482***	-0.049	0.8798	-0.0586	0.8564

Table 4.4. Tamarind Feeding Bout Duration by Topographic Measure Summary (Parametric Correlations).

	Total		**Female**		**Male**	
	mean	**median**	**mean**	**median**	**mean**	**median**
Slope	↔	↔	↔	↔	↔	↔
Angularity	↗	↗	↗	↗	↔	↔
Relief	↔	↔	↔	↔	↔	↔
2DAverage	↘	↘	↘	↘	↔	↔
2DSum	↔	↔	↘	↘	↔	↔
3DAverage	↘	↘	↘	↘	↔	↔
3DSum	↔	↔	↘	↘	↔	↔

Manual Tamarind Fruit Processing. Ring-tailed lemurs typically do not process food items using the hands, although individuals with tooth loss have previously been reported to use their hands to completely remove the shell of this fruit (see Millette et al., 2009). While these earlier data were presented on an anecdotal basis, data presented below indicate that manual processing of tamarind fruit is associated with topographic measures. All results are available in Table 4.5.

Table 4.5. Manual Processing of Tamarind Fruit (Spearman's Correlation).

		Yes			**Yes / Yes+No**			**Y+P / Y+P+N**		
All Individuals	**n=**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	31	-0.2431	0.1875	↔	-0.2477	0.1792	↔	-0.2936	0.1089	↔
Angularity	31	-0.433	***0.015***	↘	-0.433	***0.015***	↘	-0.3786	***0.0357***	↘
Relief	31	-0.2512	0.1729	↔	-0.2542	0.1676	↔	-0.3262	0.0733	↘
2DSum	31	-0.4362	***0.0142***	↘	-0.4242	***0.0174***	↘	-0.47	***0.0076***	↘
2DAverage	31	-0.2542	0.1676	↔	-0.2567	0.1633	↔	-0.278	0.13	↔
3DSum	31	-0.4491	***0.0113***	↘	-0.4415	***0.0129***	↘	-0.4765	***0.0067***	↘
3DAverage	31	-0.4184	***0.0192***	↘	-0.4194	***0.0188***	↘	-0.4356	***0.0143***	↘
Females Only	**n=**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	19	-0.3949	0.0942	↔	-0.3904	0.0984	↔	-0.5524	***0.0142***	↘
Angularity	19	-0.3361	0.1595	↔	-0.3406	0.1536	↔	-0.2615	0.2795	↔
Relief	19	-0.3021	0.2087	↔	-0.3067	0.2016	↔	-0.4774	***0.0388***	↘
2D Sum	19	-0.2342	0.3344	↔	-0.2161	0.3741	↔	-0.3547	0.1362	↔
2DAverage	19	-0.0113	0.9633	↔	-0.0294	0.9048	↔	-0.1318	0.5908	↔
3DSum	19	-0.3768	0.1118	↔	-0.3655	0.1238	↔	-0.4966	***0.0306***	↘
3DAverage	19	-0.3474	0.145	↔	-0.3565	0.1341	↔	-0.4855	***0.0351***	↘
Males Only	**n=**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	12	-0.1123	0.7282	↔	-0.1123	0.7282	↔	-0.0546	0.8662	↔
Angularity	12	-0.5034	0.0952	↔	-0.5034	0.0952	↔	-0.4679	0.1251	↔
Relief	12	-0.2996	0.3442	↔	-0.2996	0.3442	↔	-0.2729	0.3907	↔
2DSum	12	-0.7946	***0.002***	↘	-0.7946	***0.002***	↘	-0.7018	***0.011***	↘
2DAverage	12	-0.7031	***0.0107***	↘	-0.7031	***0.0107***	↘	-0.5693	***0.0534***	↘
3DSum	12	-0.6657	***0.0181***	↘	-0.6657	***0.0181***	↘	-0.5771	***0.0495***	↘
3DAverage	12	-0.6282	***0.0287***	↘	-0.6282	***0.0287***	↘	-0.5147	0.0869	↔

Yes vs. All Feeding Bouts. When I assessed the frequency of manual processing in relation to total tamarind feeding observations for all individuals, reduced angularity was associated with higher rates of manual processing (rho = -0.433, p = 0.015), as well as lower 2DSum (rho = -0.4362, p= 0.0142), 3DSum (rho = -0.4491, p = 0.0142), 3DSum (rho = -0.4491, p = 0.0113) and 3DAverage area (rho = -0.4181, p = 0.0192). As no significant patterns were observed for females, these patterns appear to be driven by males. Males demonstrated significant correlations for 2DSum (rho = -0.7946; p = 0.002), 2DAverage (rho = -0.7031, p = 0.0107), 3DSum (rho = -0.6657, p = 0.0181), and 3DAverage [(rho = -0.6282, p = 0.0287) Table 4.5]. Thus, for all individuals angularity was associated with higher rates of manual processing for individuals overall. Likewise, reduced tooth area measurements were also associated with increased use of the hands generally and for males.

Yes / Yes + No. Similar patterns were also noted when the data were examined with regard to the relative percentage of "Yes" scores to the total number of observations where Yes or No was recorded (e.g., no "unknown" or "possible" manual use observations were observed). As before, for all individuals, significant negative correlations were found for the measures of angularity (rho = -0.433, p = 0.015), 2DSum (rho = -0.4242, p = 0.0174), 3Dsum (rho = -0.4415, p = 0.0129) and 3DAverage (rho = -0.4194, p = 0.0188). As before, these patterns appear to be driven by males, with significant negative correlations occurring for 2DSum (rho = -0.7946, p = 0.002), 2DAverage (rho = -0.7031, p = 0.0107), 3DSum (rho = -0.6657, p = 0.0181) and 3DAverage (rho = -0.6282, p = 0.0287). No significant correlations were noted for females (Table 4.5). As when all feeding observations were included, these data indicate that angularity reductions and reduced area measures are associated with increased use of the hands for the

study sample overall, but when examined by sex only for males demonstrated significant results, suggesting that they are driving patterns of manual food processing for the study sample overall.

Possible Observations Included. Finally, the relationship of manual processing to topographic scores was determined when also including possible manual feeding bouts. In most cases these possible bouts were likely to be positive cases of this behavior, and I recorded "possible" rather than "yes" primarily due to observational conditions (e.g., low light, presence of foliage, etc.) and the conservative observational strategy that I used during this study. For all individuals, the patterns observed with possible manual processing included are similar to those reported above. Significant negative correlations were found for angularity (rho = -0.3786, p = 0.0357), 2DSum (rho = -0.4700, p = 0.0076), 3DSum (rho = -0.4765, p = 0.0067), and 3DAverage (rho = -0.4356, p = 0.0143). In addition, a trend towards a negative correlation was also noted for the measure of relief [(rho = -0.3262, p = 0.0733) Table 4.5]. As before, these data further indicate that angularity and area measures are associated with use of manual fruit processing, where rates of hand use increase with reduced topographic scores.

In contrast to prior observations, when possible manual processing was included, females also demonstrated significant correlations for slope (rho = -0.5524, p = 0.0142), relief (rho = -0.4774, p = 0.0388), 3DSum (rho = -0.4966, p = 0.0306) and 3DAverage (rho = -0.4855, p = 0.351). Male patterns of manual processing were less significant when possible observations were included, with only 2DSum (rho = -0.7018, p = 0.011), 2DAverage (rho = -0.5693, p = 0.0534) and 3DSum (rho = -0.5771, p = 0.0495) being negatively correlated with manual processing. (Table 4.5). These data indicate that when possible occurrences are included, correlations are present in females for relief and slope (which are common measures of wear)

and area measures. For males, only area measurements were negatively correlated use of the hands, although this pattern is similar to those observed for when examining all observations or when limiting analysis to “Yes” or “No” observations.

Use of Open Tamarind Fruit. Ring-tailed lemurs often consume tamarind fruit with shells which have been previously been opened prior to initial processing by the animal. Fruits were only recorded as “open” if the shell was clearly fractured, revealing the internal structure of the fruit. Possibly opened shells were recorded as well if it appeared that the shell was open, but this could not be confirmed. Here the source of the opened shell is not taken into account. Results for use of open tamarind fruit are available in Table 4.6.

Open / Total Observations. When assessed in terms of opened tamarinds compared to all feeding observations (e.g., including unknown samples), several patterns were noted for the study sample overall and for males specifically. For all individuals, no significant patterns of open tamarind use were present with respect to any non-area topographic measure. However, a trend towards a negative correlation was present for the measure of 3DSum (rho = -0.3380, p = 0.0629). While no significant correlations were noted for females for any measure, several significant correlations were present for males. For males, slope and relief values were negatively correlated with open tamarind use (slope: rho = -0.5874, p = 0.0446; relief: r = -0.5874, p = 0.0446). In addition, negative correlations were also present for 3DSum (rho = -0.05804, p = 0.0479) and 3DAverage measures [(rho = -0.5874, p = 0.0446) Table 4.6]. These data indicate that for males use of open tamarind fruit was associated with measures of increased wear state, as well as reduced occlusal surface area.

Table 4.6. Open Tamarind Fruit Use by Topographic Measure (Spearman's Correlations).

		Yes			Yes / Yes+No			Y+P / Y+P+N		
All Individuals	**n =**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	31	-0.0752	0.6875	↔	-0.2339	0.2053	↔	-0.2929	0.1099	↔
Angularity	31	0.0097	0.9588	↔	-0.2266	0.2202	↔	-0.2949	0.1073	↔
Relief	31	-0.0853	0.6482	↔	-0.285	0.1202	↔	-0.3639	***0.0442***	↘
2DSum	31	-0.3019	0.0988	↔	-0.3251	*0.0743*	↘	-0.4066	***0.0232***	↘
2DAverage	31	-0.2309	0.2114	↔	-0.1812	0.3292	↔	-0.2737	0.1363	↔
3DSum	31	-0.338	*0.0629*	↘	-0.4184	***0.0192***	↘	-0.498	***0.0044***	↘
3DAverage	31	-0.2981	0.1034	↔	-0.3457	*0.0568*	↘	-0.4403	***0.0132***	↘
Females Only	**n =**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	19	0.1423	0.5612	↔	0.0272	0.9119	↔	-0.0509	0.836	↔
Angularity	19	-0.0044	0.9858	↔	-0.4534	***0.0512***	↘	-0.5171	***0.0234***	↘
Relief	19	0.1581	0.518	↔	-0.0923	0.7072	↔	-0.1589	0.5158	↔
2DSum	19	-0.3399	0.1545	↔	-0.3295	0.1683	↔	-0.3582	0.1321	↔
2DAverage	19	-0.2486	0.3048	↔	-0.0668	0.7859	↔	-0.0896	0.7154	↔
3DSum	19	-0.2574	0.2875	↔	-0.3146	0.1896	↔	-0.3494	0.1425	↔
3DAverage	19	-0.1686	0.4901	↔	-0.1397	0.5683	↔	-0.187	0.4433	↔
Males Only	**n =**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	12	-0.5874	***0.0446***	↘	-0.7203	***0.0082***	↘	-0.7215	***0.0081***	↘
Angularity	12	0.014	0.9656	↔	0.0699	0.829	↔	-0.0981	0.7617	↔
Relief	12	-0.5874	***0.0446***	↘	-0.6503	***0.022***	↘	-0.7846	***0.0025***	↘
2DSum	12	-0.3077	0.3306	↔	-0.0629	0.8459	↔	-0.1541	0.6325	↔
2DAverage	12	-0.3217	0.3079	↔	-0.014	0.9656	↔	-0.1331	0.6801	↔
3DSum	12	-0.5804	***0.0479***	↘	-0.4895	0.1063	↔	-0.662	***0.019***	↘
3DAverage	12	-0.5874	***0.0446***	↘	-0.4895	0.1063	↔	-0.683	***0.0144***	↘

Open / Open + Not Open. When examined in terms where use was limited strictly to open and non-open fruit, use of open tamarind fruit was negatively associated with 3DSum data for all individuals (rho = -0.4184, p = 0.192), while trends were also noted for 2DSum (rho = -0.3251, p = 0.0743) and 3DAverage (rho = -0.3457, r = 0.0568). Again, for males a number of significant findings were recorded. Significant negative correlations were found for slope (rho = -0.7203, p = 0.0082) and relief (rho = -0.6503, p = 0.022), although 3DSum and 3DAverage values were no

longer correlated with open tamarind use. For females a negative correlation between angularity and open tamarind use was observed (rho = -0.4532, p = 0.0512). When only open or non-open fruit were considered, use of open fruit was associated with reduced tooth area measures. When assessed by sex, males demonstrated an association with slope and relief, while females with reduced angularity used open tamarind fruit at increased frequencies (Table 4.6).

Open + Possibly Open Included. When possible open tamarinds are included, a number of patterns emerge from the dataset. For all individuals, significant negative correlations are present for relief (rho = -0.3639, p = 0.0442) as well as for 2DSum, (rho = -0.4066, p = 0.0232), 3DSum (rho = -0.498, p = 0.0044) and 3DAverage (rho = -0.4403, p = 0.0132). Few significant patterns were noted among females, although angularity scores were significantly correlated negatively with open fruit use (rho = -0.5171, p = 0.0234). In contrast, I found a number of correlations for male lemurs, with significant negative correlations observed for slope (rho = -0.7215, p = 0.0081), relief (rho = -0.7846, p = 0.0025), 3DSum (rho = -0.662, p = 0.0190) and 3DAverage (rho = -0.683, p = 0.0144). For all individuals, open fruit use was associated with decreased relief and the area measures of 2D and 3D sums, as well as 3DAverage. When examined by sex, reduced angularity was associated with increased open fruit use for, while for males there were negative correlations for slope, relief, 3DSum and 3DAverage (Table 4.6).

Conspecific-Processed Tamarind Fruit. In addition to merely pre-opened tamarinds, data were also recorded on the use of tamarinds which were opened previously by other conspecifics. For "processed" to be recorded, another individual had to be directly observed to open and discard a tamarind prior to being utilized by the focal individual.

For both the fraction of processed fruits to total observations and for the fraction of processed to processed and non-processed total (e.g., processed / processed + non-processed), no significant correlations were found for any measure. This was the case for all individuals combined, or when examined by sex status. When possibly processed tamarinds were also included, the only significant result was a negative correlation for slope among males (rho = -0.5926, p = 0.0423). No other significant findings or trends were observed (Table 4.7).

Table 4.7. Use of Conspecific Processed Tamarind Fruit by Topographic Measure.

		%Yes			Yes / Yes+No			Y+P/Y+P+N		
All Individuals	**n =**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	31	0.1059	0.5707	↔	0.1001	0.5921	↔	-0.1571	0.3986	↔
Angularity	31	0.2283	0.2167	↔	0.2322	0.2088	↔	-0.0428	0.8191	↔
Relief	31	0.141	0.4492	↔	0.1342	0.4718	↔	-0.146	0.4333	↔
2DSum	31	0.1321	0.4786	↔	0.1441	0.4392	↔	-0.0086	0.9635	↔
2DAverage	31	0.2011	0.2781	↔	0.2159	0.2433	↔	0.0505	0.7872	↔
3DSum	31	0.1206	0.5182	↔	0.1232	0.5092	↔	-0.1486	0.4251	↔
3DAverage	31	0.1683	0.3655	↔	0.1714	0.3566	↔	-0.1443	0.4387	↔
Females Only	**n =**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	19	0.1871	0.443	↔	0.1871	0.443	↔	0.0766	0.7554	↔
Angularity	19	0.2807	0.2444	↔	0.2807	0.2444	↔	-0.1713	0.4831	↔
Relief	19	0.2828	0.2407	↔	0.2828	0.2407	↔	0.0565	0.8183	↔
2DSum	19	0.0617	0.802	↔	0.0617	0.802	↔	-0.0501	0.8385	↔
2DAverage	19	0.1467	0.5489	↔	0.1467	0.5489	↔	0.0647	0.7924	↔
3DSum	19	0.1063	0.6649	↔	0.1063	0.6649	↔	-0.1148	0.6397	↔
3DAverage	19	0.1744	0.4753	↔	0.1744	0.4753	↔	-0.0592	0.8096	↔
Males Only	**n =**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	12	-0.0269	0.9339	↔	-0.0269	0.9339	↔	-0.5926	***0.0423***	↘
Angularity	12	0.1828	0.5697	↔	0.1828	0.5697	↔	0.2261	0.4797	↔
Relief	12	-0.0161	0.9603	↔	-0.0161	0.9603	↔	-0.4367	0.1558	↔
2DSum	12	0.5214	0.0821	↔	0.5214	0.0821	↔	0.1872	0.5603	↔
2DAverage	12	0.5214	0.0821	↔	0.5214	0.0821	↔	0.0936	0.7724	↔
3DSum	12	0.172	0.5929	↔	0.172	0.5929	↔	-0.2573	0.4194	↔
3DAverage	12	0.172	0.5929	↔	0.172	0.5929	↔	-0.3509	0.2634	↔

.

Toothrow Processing Location. Ring-tailed lemurs predominantly process tamarind fruit upon the posterior dentition [(e.g., the distal premolars and on the molars) see Sauther, 1992;

Yamashita, 2003]. Individuals with significant dental impairment, however, may have postcanine dentitions which no longer effectively break down the shell of tamarind fruit. These individuals may selectively utilize the anterior dentition as this tends to wear at a reduced rate within this population (Cuozzo, pers. comm.; Millette, personal observations). Toothrow processing location results are available in Table 4.8.

Table 4.8. Toothrow Processing Location for Tamarind Fruit by Topographic Measure.

		% Post			Post / Post+Ant			Post+Both / P+B+A		
All Individuals	**n =**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	31	0.3112	0.0883	↔	0.2962	0.1057	↔	0.2962	0.1057	↔
Angularity	31	0.3312	*0.0687*	↗	0.3104	0.0893	↔	0.3104	0.0893	↔
Relief	31	0.3453	*0.0571*	↗	0.3365	*0.0642*	↗	0.3365	*0.0642*	↗
2DSum	31	0.4357	***0.0143***	↗	0.3113	0.0883	↔	0.3113	0.0883	↔
2DAverage	31	0.2092	0.2588	↔	0.1665	0.3707	↔	0.1665	0.3707	↔
3DSum	31	0.4795	***0.0063***	↗	0.3894	***0.0304***	↗	0.3894	***0.0304***	↗
3DAverage	31	0.4069	***0.0231***	↗	0.3635	***0.0444***	↗	0.3635	***0.0444***	↗
Females	**n=**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	19	0.2554	0.2913	↔	0.2803	0.2451	↔	0.2803	0.2451	↔
Angularity	19	0.5801	***0.0092***	↗	0.475	***0.0398***	↗	0.475	***0.0398***	↗
Relief	19	0.3352	0.1606	↔	0.2707	0.2624	↔	0.2707	0.2624	↔
2DSum	19	0.3695	0.1195	↔	0.2071	0.3948	↔	0.2071	0.3948	↔
2DAverage	19	0	1	↔	-0.069	0.7788	↔	-0.069	0.7788	↔
3DSum	19	0.4677	***0.0434***	↗	0.2859	0.2355	↔	0.2859	0.2355	↔
3DAverage	19	0.3256	0.1738	↔	0.2002	0.4111	↔	0.2002	0.4111	↔
Males	**n=**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	12	0.2657	0.4038	↔	0.4032	0.1938	↔	0.4032	0.1938	↔
Angularity	12	-0.1329	0.6806	↔	-0.0484	0.8813	↔	-0.0484	0.8813	↔
Relief	12	0.3776	0.2262	↔	0.5698	***0.0531***	↗	0.5698	***0.0531***	↗
2DSum	12	0.4545	0.1377	↔	0.4999	0.0979	↔	0.4999	0.0979	↔
2DAverage	12	0.3706	0.2356	↔	0.5698	***0.0531***	↗	0.5698	***0.0531***	↗
3DSum	12	0.3916	0.2081	↔	0.5698	***0.0531***	↗	0.5698	***0.0531***	↗
3DAverage	12	0.3636	0.2453	↔	0.6397	***0.0251***	↗	0.6397	***0.0251***	↗

Postcanine / Total. When I examined the number of postcanine processing bouts out of all observations (e.g., unknowns included) for all individuals, significant positive correlations were found for 2DSum (rho = 0.4357, p = 0.0143), 3DSum (rho = 0.4795, p = 0.0063) and 3DAverage

(rho = 0.4069, p = 0.0231) measures. In addition, trends were noted for angularity scores (rho = 0.3312, p = 0.0687), and relief (rho = 0.3453, p = 0.0571). When limited to females, significant correlations were present for angularity (rho = 0.5801, p = 0.0092) and 3DSum (rho = 0.4677, p = 0.0434). No significant correlations were present for males. For the study sample overall, increased use of the posterior dentition was associated with higher 2DSum, 3DSum and 3DAverage, with a trend towards increased postcanine angularity. When assessed by sex, significant results were present only for females, where the measures of angularity and 3DSum were associated with increased postcanine use (Table 4.8).

Postcanine / Postcanine + Anterior. When examining the dataset with regard to the percent of posterior processing when limited strictly to anterior and posterior processing observations, both 3DSum and 3DAverage were positively correlated with posterior toothrow use (3DSum: rho = 0.3894, p = 0.0304; 3DAverage : rho = 0.3635, p = 0.0444). For females, only angularity was significantly correlated with food processing position (rho = 0.475, p = 0.0398), while for males 3DAverage was positively correlated with postcanine usage (rho = 0.6397, p = 0.0251), as was 2DAverage (rho = 0.5698, p = 0.0531), 3DSum (rho = 0.5698, p = 0.0531) and relief (rho = 0.5698, p = 0.0531). For all individuals, the area measures of 3DSum and 3DAverage were associated with increasing topographic scores. For females, only angularity was associated with topography, although for males the measures of relief, 2D and 3D average, and 3DSum were positively associated with postcanine use (Table 4.8)

Postcanine + Both Included. Occasionally individuals utilized both the anterior and posterior positions of the toothrow to process tamarind fruit. When these observations were included in the

analysis, a number of patterns emerged. First, when all individuals were included both 3DSum and 3DAverage generated significant positive correlations (3DSum: rho = 0.3894, p = 0.0304, 3DAverage 0.3635, p = 0.0444). A positive trend was also observed for relief (rho = 0.3365, p = 0.0642). When examined by sex, a positive correlation was present for relief (rho = 0.475, p = 0.0398) among females. In turn, males demonstrated a positive correlations for relief (rho = 0.5691, p = 0.0531), 3DAverage (rho = 0.6397, p = 0.0251), 3DSum (rho = 0.5698, p = 0.0531). and 2DAverage (rho = 0.5698, p = 0.0531). When bouts where anterior and posterior positions were used, positive correlations were noted overall for 3DSum and 3DAverage measures. By sex, relief was correlated for females, while relief, 3DAverage, 3DSum and 2DAverage were associated with use of both anterior and posterior positions for males (Table 4.8).

Tamarind Licking Behavior. Previous research at BMSR has demonstrated that ring-tailed lemurs often lick tamarind fruit as a means of extracting softer inner portions of this fruit (Sauther, 1992). Licking has also been suggested as a non-dental means for processing food items as related to tooth loss (Millette et al., 2009). Results for licking behavior are available in Table 4.9.

Yes / Total Observations. As a percentage of all observations (including unknown) the only significant correlation was a positive correlation for angularity (rho = 0.4355, p = 0.0143) when all individuals were included. No other correlations were noted either for all individuals or by sex (Table 4.9).

Table 4.9. Use of Licking to Process Tamarind Fruit by Topographic Measure (Spearman's Correlations).

		Yes			Yes / Yes+No			Y+P / Y+P+N		
All Individuals	**n =**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	31	0.1976	0.2867	↔	0.1425	0.4444	↔	0.15	0.4206	↔
Angularity	31	0.4355	***0.0143***	↗	0.3395	*0.0617*	↗	0.3428	*0.0591*	↗
Relief	31	0.2573	0.1624	↔	0.156	0.4019	↔	0.1623	0.3831	↔
2DSum	31	0.0375	0.8413	↔	0.0981	0.5996	↔	0.0997	0.5935	↔
2DAverage	31	0.1044	0.5761	↔	0.1841	0.3215	↔	0.1857	0.3172	↔
3DSum	31	0.1081	0.5628	↔	0.1702	0.3601	↔	0.1732	0.3515	↔
3DAverage	31	0.1649	0.3753	↔	0.2202	0.2339	↔	0.2247	0.2243	↔
Females Only	**n =**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	19	0.0386	0.8753	↔	-0.2491	0.3037	↔	-0.2352	0.3324	↔
Angularity	19	0.4035	0.0867	↔	0.0263	0.9148	↔	0.0298	0.9035	↔
Relief	19	0.1596	0.5138	↔	-0.1737	0.477	↔	-0.1597	0.5137	↔
2DSum	19	-0.4088	0.0823	↔	-0.6316	***0.0037***	↘	-0.6283	***0.004***	↘
2DAverage	19	-0.1737	0.477	↔	-0.2684	0.2665	↔	-0.2642	0.2745	↔
3DSum	19	-0.2965	0.2177	↔	-0.5368	***0.0178***	↘	-0.5309	***0.0193***	↘
3DAverage	19	-0.1105	0.6524	↔	-0.3509	0.1408	↔	-0.3414	0.1526	↔
Males Only	**n =**	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	12	0.2587	0.4168	↔	0.5446	*0.0671*	↗	0.5446	*0.0671*	↗
Angularity	12	0.4545	0.1377	↔	0.6763	***0.0158***	↗	0.6763	***0.0158***	↗
Relief	12	0.2867	0.3663	↔	0.5161	0.0859	↔	0.5161	0.0859	↔
2DSum	12	0.1678	0.6021	↔	0.3595	0.2511	↔	0.3595	0.2511	↔
2DAverage	12	-0.021	0.9484	↔	0.1744	0.5877	↔	0.1744	0.5877	↔
3DSum	12	0.3077	0.3306	↔	0.6086	***0.0357***	↗	0.6086	***0.0357***	↗
3DAverage	12	0.2098	0.5128	↔	0.5197	0.0833	↔	0.5197	0.0833	↔

Yes / Yes + No. When licking was assessed in relation to observations where it's presence or absence could be confirmed (Yes / Yes + No), a trend towards a positive correlation was noted for angularity (rho = 0.3395, p = 0.0617) for all individuals. When examined by females alone, a negative correlation was present for both 2DSum (rho = -0.6316, p = 0.0037), and 3DSum (rho = -0.5368, p = 0.0178). Correlations were present for males, but in the opposite direction from that of the females. Here angularity (rho = 0.6763, p = 0.0158) and 3DSum (rho = 0.6086, p = 0.0357). In addition a trend towards a positive correlation with slope was observed [(rho = 0.5446, p = 0.0671) Table 4.9].

Yes + Possible Licking Included. When possible licking observations were included in data analysis, all patterns remained the same. Overall, only angularity was associated with licking (rho = 0.3428, p = 0.0591). When assessed by sex, females again had negative correlations for 2DSum (rho = -0.6283, p =0.004), and 3DSum (rho = -0.5309, p = 0.0193). In contrast, males had positive correlations for angularity and 3DSum (angularity: rho = 0.6763, p =0.0158; 3DSum (rho = 0.6086, p = 0.0357), as well as a positive trend for slope (rho = 0.5446, p = 0.0671). These data indicate that when possible cases are included, increased angularity is associated with increased licking for individuals overall. Females lick tamarind more frequently with reduced topographic scores for 2D and 3D sums, although males demonstrate increased licking in association with higher topographic scores for angularity and 3DSum (Table 4.9).

Use of Tamarind from the Ground. During each feeding bout it was recorded whether the animal took a tamarind fruit from the ground. Often tamarind fruit from the ground are open and/or extremely ripe. It was thus posed that these may be easier for individuals to consume and

would thus be preferentially utilized by those with worn teeth. When both males and females were combined, no significant correlations or trends were found. For females, a positive correlation between ground tamarind use and slope was noted (rho = 0.5579, p = 0.013), although no other correlations were noted. No significant correlations were present for males, although a very weak trend towards a negative correlation with 2DAverage was noted [(rho = -0.5254, p = 0.0794) Table 4.10].

Table 4.10. Use of Tamarinds From the Ground (Spearman's Correlations).

	Total			**Females**			**Males**		
	rho =	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Slope	0.2426	0.1886	↔	0.5579	***0.0131***	↗	-0.1261	0.6962	↔
Angularity	-0.0337	0.8573	↔	-0.2211	0.3631	↔	0.1821	0.571	↔
Relief	0.2089	0.2594	↔	0.3544	0.1366	↔	0.021	0.9483	↔
2DSum	-0.2186	0.2375	↔	-0.1123	0.6472	↔	-0.4203	0.1737	↔
2DAverage	-0.2934	0.1092	↔	-0.1456	0.552	↔	-0.5254	*0.0794*	↘
3DSum	-0.0875	0.6397	↔	0.0158	0.9488	↔	-0.2277	0.4767	↔
3DAverage	-0.0718	0.7012	↔	0.0982	0.6891	↔	-0.2697	0.3966	↔

Discussion.

Tamarind fruit consumption represents a critical aspect of BMSR ring-tailed lemur feeding ecology. While tamarind is available to, and is consumed by, lemurs throughout the year, this food also serves as a key fallback food during the resource-depleted dry season (Sauther, 1992; Simmen et al., 2006; Sauther and Cuozzo, 2009; Millette, this study). Tamarind is also exceptionally challenging in terms of its mechanical properties, being both the hardest and toughest food regularly consumed by BMSR *L. catta* (Yamashita, 2008b; Cuozzo and Sauther, 2006a; Cuozzo et al., 2010; Yamashita et al., 2012). However, ring-tailed lemur dental morphology, which is both high crested and thin-enameled, appears to be ill-suited to the

consumption of this food. This fruit is a major source of dental wear and tooth loss among this population of ring-tailed lemurs, as populations where tamarind fruit is rarely consumed (e.g., ring-tailed lemurs at Tsimanampetsotsa National Park) demonstrate lower rates of tooth wear and loss (Sauther et al., 2002; Godfrey et al., 2005; Cuozzo and Sauther, 2006a,b; LaFleur, 2012; Yamashita et al., 2015a). Given its challenging mechanical properties, significant tooth wear and loss are presumed to inhibit an individual's capacity to process and subsequently consume this food item. For example, pervious research on tamarind fruit processing indicates that individuals with tooth loss divergently process tamarind fruit, engaging in longer feeding bouts and more frequently licking this food item than those without tooth loss (Millette, 2007; Millette et al., 2009). Individuals with tooth loss also demonstrated higher amounts of fruit material in their feces relative to leafy materials, suggesting that they are impaired in the processing of this food item (Millette et al., 2012 / Chapter 6; see also Chapter 5).

Tamarind Feeding Durations. Previous research at BMSR suggests that tooth loss is associated with increased tamarind fruit feeding durations (Millette, 2007; Millette et al., 2009). This earlier study, however, did not examine if tooth wear alone was sufficient to result in divergent patterns of tamarind fruit feeding in terms of feeding bout duration. The results of this study indicate that tamarind fruit feeding duration is related to dental wear status when measured using topographic methods. When limited to nonparametric assessments, significant associations between tooth form and feeding bout length were only noted for angularity measurements. In this case, tamarind fruit feeding duration increased with higher average angularity. While this was true for the dataset overall, this pattern appears to be driven primarily by males as significant correlations were noted only for male individuals and no significant pattern was noted for females. It is

notable that these tamarind bout length durations were in an unexpected direction, as it was hypothesized that reduced angularity would be associated with reduced food processing capacity due to the presumed association of this measure with the generation of multidirectional forces within challenging food items (see Ungar and M'Kirera, 2003; M'Kirera and Ungar, 2003).

One explanation for these unexpected findings is that individuals with higher angularity values may be spending more time to open tamarind fruit because they maintain a greater ability to process more challenging tamarind fruits than do individuals with lower angularity scores. Higher dental angularity values may permit access to fruits which take a longer period to open, but may be worth the additional time spent processing if they possess contents of higher nutritional value. In contrast, those with lower angularity may focus on fruits which only take a minimal effort to process, and do not attempt to process and/or simply reject fruits which are particularly challenging to process. Data presented here on the consumption of open fruits also suggest that individuals with dental impairment may be preferentially selecting less challenging fruits (see below). This explanation, however, must be assessed in the future using nutritional and mechanical-properties testing methods to determine if tamarinds which take longer to process do, in fact, possess greater nutritional content and are more mechanically challenging.

When tamarind fruit were assessed using parametric methods, which are likely appropriate for examining area-based topographic measures (e.g., unlike topographic scores they demonstrate interval-ratio scaling) a number of additional patterns emerged. For the total study sample, a significant negative correlation was present for 2DAverage for both mean and median feeding times, while a trend is present towards a negative correlation for 3DAverage and mean duration ($p = 0.0648$) and median duration ($p = 0.0606$). Only females demonstrated significant correlations between tooth form and duration, with significant negative correlations being

present for 2DAverage, 2DSum, 3DAverage and 3DSum measures for both median and mean bout durations. No such patterns were present for male individuals, suggesting that the overall patterns observed are driven by females, although the larger sample size available for females (19 vs. 12) may be the root of these sex-based differences.

In contrast to angularity, the directionality of the significant sum and average area results were as hypothesized. These data indicate that reduced topographic measures of tooth area were associated with a lower capacity to fragment tamarind fruit. Only 2DAverage and 3DAverage, but not 2DSum or 3DSum, were of significant or trending towards significant, for the total study sample. While these measures only reflect the average size of teeth present, they do not account for missing teeth as do the measures of 2DSum or 3DSum. They do, however, provide a general measure of dental wear as individuals with extensively worn teeth visibly demonstrated teeth which were of reduced size as a function of wear (Millette, personal observations during cast scanning and topographic analysis). When limited to females, all area measurements were negatively associated with increased tamarind fruit processing durations, suggesting that 3DSum and 2DSum area measurements are of utility for inferring the impacts of tooth loss on behavior. In all cases, as area decreases the amount of time necessary to process tamarind fruit increases, suggesting that dental impairment impacts the animal's ability to process this food item.

No significant patterns were found for male feeding durations with regard to any topographic area measure. This result is potentially an effect of small sample size, although significant results for angularity among males (and significant results other food processing behaviors), suggest that sample size was sufficiently large enough to discern food processing behavior differences among male lemurs relative to topography. One potential explanation for these findings is that males may simply not differ in their tamarind feeding bout lengths in

relation to dental topography, but rather their feeding bout lengths are determined by other factors. As this species displays strict female dominance, it is possible that males do not have access to prime tamarind fruits due to female priority of access. If males are differentially forced to consume tamarinds which are more difficult to open, or are forced to utilize overripe or unripe fruits that result in widely divergent feeding times, distinct patterns related to feeding bout durations may fail to emerge. Alternatively, females were frequently observed to displace and/or interrupt males feeding on tamarind fruit (Millette, personal observations). Such intersexual dominance-related interactions could result in irregular feeding bout lengths among males, thus eliminating the occurrence of clear patterns of feeding bout length.

Although the data are not well suited to parametric analysis, it is notable that angularity was associated with increased feeding bout lengths when examined using parametric correlations for all animals (both mean and median times) and for female mean durations. These data may confirm that angularity is indeed associated with increased feeding bout lengths, although the patterning of sex differences here is different than when nonparametric methods are utilized. Again, such patterns likely indicate that individuals with high average angularity may be selecting more challenging fruits than those with lower angularity as they may be better capable of processing these foods.

Manual Processing of Tamarind Fruit. Lemurs in general have reduced hand dexterity (Bishop, 1962). Ring-tailed lemurs generally do not utilize their hands to process food items into a state suitable for consumption. While the hands are frequently used to manipulate items to the mouth for dental processing, they are typically not used to remove portions of food items prior to placement in the mouth or to remove mechanically-challenging portions of food items [(e.g.,

shells, seeds, etc.) Sauther, 1992; Millette et al., 2009; Millette, personal observations]. While this is generally the case, previous research at BMSR has found that some individuals occasionally utilize the hands to completely (or less frequently, partially) remove tamarind fruit shells (Millette, 2007; Millette et al., 2009). Although no quantitative data were collected for manual food processing in relation to dental impairment status during this previous study, it was observed that only those individuals with extensive tooth wear engaged in this behavior, suggesting a link between tooth loss and manual food processing. Such a similar pattern was observed during this study, with the individuals with the highest amounts of tooth loss (Black 226 and Blue 348) engaging in this behavior most frequently of all study individuals.

When examined by all individuals, significant correlations were present for angularity, 2DSum, 3DSum and 3DAverage. In all cases, these correlations were in the negative direction, suggesting that increasing tooth wear and/or tooth loss (in the case of 2D and 3D sums) results in higher frequencies of manual tamarind fruit processing. These patterns hold true when analyzed for the frequencies of manual processing to all feeding observations, when assessed only for those observations where “yes” or “no” were recorded, or when possible manual processing was recorded. These correlations were generally strongest when possible manual processing events were included (and a trend towards a negative correlation with relief was found), suggesting that these possible events likely recorded true manual processing events.

It is notable that significant correlations were limited to angularity and measures of area. It has been posited previously that angularity is a key aspect which allows the maintenance of dental function through tooth wear (see Ungar and M’Kirera, 2003; Dennis et al., 2004, but see Klukkert et al., 2012a) as this value generally changes only with extensive tooth wear (in comparison to relief and slope which generally decline continuously with wear). That this

measure is linked with a compensatory behavior in the form of increased use of the hands to process foods provides direct evidence for the hypothesis that angularity is critically associated with the maintenance of dental function, and which has to date not been tested using behavioral data.

Topographic area measurements provide a measure of the size of teeth and the amount functional occlusal surface available for food processing. As dental wear is associated with the physical ablation of the tooth, reduced occlusal area is thus associated with increasing dental impairment status (particularly for 2D and 3DSum measures which integrate tooth loss). The association of increased manual use for individuals with diminished 2D and 3D surface areas further suggests that wear and/or tooth loss result in the use of this behavior in compensation for dental impairment.

When examined by sex, females only demonstrated significant patterns of manual tamarind processing when assessed with the inclusion of possible events. Here, slope, relief, 3DSum and 3DAverage were negatively correlated with the frequency of manual (or possible) food processing. As when all individuals were included, these occur in a negative direction, coherent with the concept that increasing dental wear and/or tooth loss is associated with higher rates of behavioral compensation in the form of manual processing among females. These data may also provide support for the hypothesis that the measures of slope and relief are also correlated with the reduction of dental function, although it is surprising that angularity is not associated with manual processing among females. As noted below, however, the relatively restricted range of variation for angularity scores as well as the relatively small sample size of females may impact the statistical power with which this measure is correlated to behavior. It is critical to note that, as the relationship of this behavior to topographic measures only become

significant when including possible events, such relationships appear to be somewhat limited among females in comparison to that observed for males (see below).

In contrast to females, when limited to males the measures of relief, slope or angularity were not associated with manual processing for any means of examining the frequency of manual processing (e.g., yes / all observations, yes / yes + no, yes with possible included). However, strong patterns are present for area measurements for the frequency of manual processing when all observations utilized in analysis, as well as when limited to clear observations where either "yes or no" were recorded. When possible manual processing was included, 2DSum, 2DAvearage and 3DSum were significantly correlated with this behavior. Among males, it therefore appears that non-area based topographic scores do not explain the presence of this behavior, suggesting that tooth form alone (e.g., overall slope, relief or angularity) does not necessarily result in the occurrence of manual processing. Among male study subjects, it therefore appears that the total loss of dental function through reduced surface area leads to the use of manual processing.

Overall, there do appear to be sex differences in the occurrence of manual processing, with area measures being more closely associated with these behaviors among males (at least for area measurements). However, it is intriguing that the associations of these behaviors are only associated with the measure of angularity when both males and females are included in the analysis. This may reflect reduced sample size available when the dataset is split by sex, but may also indicate that these scores are more sensitive to sample size than are topographic area. As such, angularity values did appear to have a relatively restricted range in comparison to area scores, slope or relief, and may thus require larger sample sizes to reach significance.

Use of Open Tamarind Fruit. Ring-tailed lemurs have often been observed to consume tamarind fruit which has been previously opened prior to initial processing by the animal. While individuals may use fruits which have been opened and then discarded by other individuals (see Cuozzo and Sauther, 2006a), open fruits are also available from other sources. These may include shells which have simply been broken open (e.g., by a fall to the forest floor from their initial point of origin within the trees), previous opening by other primate taxa at BMSR (e.g., Sifaka), or opening by non-primate taxa [(e.g., birds such as the sickle bill vanga, which is commonly observed to consume these fruits) Millette, personal observations]. Irrespective of source, opened ripe tamarind appears to be significantly less challenging to process as once breached, tamarind shells tend to rapidly break apart (Yamashita, 2002, 2008b; Yamashita et al., 2012; Millette, personal observations). Thus, I hypothesized that individuals with dental impairment would be more likely to utilize previously opened tamarind fruit than do those of lower tooth wear or loss status.

When examined across the total study sample, when the use of open fruits was compared to all observations of tamarind feeding, only a trend towards a negative correlation for 3DSum was noted. When only “Yes” or “No” observations were included, 3DSum became a significant correlation while trends were present for 2DSum and 3DAverage. When possibly open fruits were included, each of these measures demonstrated significant negative correlations while relief was also negatively associated with use of open fruit. These overall data indicate that reduced area and relief are associated with use of open fruits, suggesting that this behavior is utilized in compensation for dental impairment and that such open fruit are indeed more accessible to individuals with impaired dentitions.

In this case, it is also notable that these patterns likely only became significant with the inclusion of possibly opened fruit as it was often difficult to determine with absolute certainty if fruits were open, although it was usually easy to determine if the fruit was unopen. However, I believe that in the majority of cases these fruits were in fact open as the observational strategy utilized was extremely conservative (e.g., the shell had to be clearly breached or internal structures were visible). Thus, for distant observations in the trees, where focal animals could be up to 20 meters or more away, it is likely that the frequency of opened fruit use was underestimated when possibly open fruits were excluded from analysis.

Sex based differences were apparent in the patterning of open fruit use with respect to topographic measures. Females demonstrated negative correlations for angularity when assessed by confirmed open or unopened fruits, as well as when possibly opened fruits were included. In contrast, for all groupings of open fruit use, males demonstrated significant negative correlations for at least some measures. When all feeding bouts were included, this held for slope, relief, 3DSum and 3DAverage. In contrast, when limited to “yes and no” observations, only slope and relief were negatively correlated. However, when possibly opened fruits were included, stronger negative correlations were present when assessed for slope, relief, 3DSum and 3DAverage. Likewise, all data are in the expected direction, suggesting that dental wear status is associated with the use of opened fruits. These data suggest that males are driving overall patterns of open tamarind fruit use with respect to dental impairment status. Moreover, these data also suggest that males may be benefiting more from use of open tamarinds than do females, which may be related to priority of access in this species (see below). In contrast to manual use, among males these data are also well associated with measures of slope and relief in addition to area measurements, although the source of this difference between these behaviors is unclear.

Nevertheless, they do suggest that increased dental wear status is associated with the use of open tamarind fruit for males.

Why these measures should be more highly correlated among males than females is uncertain as it would appear that the use of opened fruits would render this food more accessible to individuals with dental impairment irrespective of sex. It is possible that males with dental impairment are more likely to use open fruits as they will have less access to easily accessible or preferred unopened fruits. Essentially, it is possible that male individuals are unable to utilize easy-to-process but unopened fruits as females have priority of access to this resource. Alternatively, females with dental impairment may not as frequently use opened fruits as they may be of lower quality. For example, under inspection open tamarind fruit found on the ground I found they were frequently contaminated with mold-like material, or were partially rotten and/or apparently fermented, particularly during the wet season (Millette, personal observations). If females preferentially avoid such open fruits due to increased access to preferred resources, this could explain the differences in the patterning of results reported here.

Use of Conspecific-Processed Tamarind Fruit. In addition to generally opened fruit (e.g., fruit that was opened prior to processing by the focal animal irrespective of source), ring-tailed lemurs have also been observed to utilized food items which have been previously opened and subsequently discarded by other conspecific individuals. Such use of conspecific-processed foods has been posited as a behavior used in compensation for dental impairment by other researchers (see Cuozzo and Sauther, 2006a), although this has not been examined for BMSR lemurs specifically using quantitative methods. For the purposes of this study, use of pre-processed tamarind was recorded if a fruit was clearly opened by one individual and

subsequently utilized by the focal animal. As before, possible use of pre-processed foods was recorded as well, as observational conditions frequently made confirming this behavior difficult.

Overall, only one significant correlation was noted for this behavioral measure, with a negative correlation for males being present for slope when possible observations were included. While this correlation is significant, no other data is present to support a link between dental impairment as measured through topographic analysis. Dental topography is not closely linked with the use of pre-processed food items, for all individuals overall or for either sex. Thus, it appears that the use of conspecific-opened tamarind fruit is not a significant compensatory strategy among BMSR ring-tailed lemurs with dental impairment.

Despite previous qualitative observations, this behavior appears to be not related to dental impairment or tooth wear among BMSR *Lemur catta.* It is possible that individuals do not engage in this behavior simply due to the fact that tamarinds rejected by another individual were done so reasons that would keep another individual from consuming the fruit irrespective of tooth wear or loss status. For example, rejected tamarinds could possibly be not ripe enough for consumption (ring-tailed lemurs typically prefer ripe tamarind) or were spoiled beyond the point preferred for consumption (as is common for tamarinds found on the ground). This explanation does not, however, explain why those individuals with higher tooth wear status would preferentially utilize tamarinds which were opened from all sources. Data for all opened fruits may be significant because there were simply a greater number of opportunities to consume such fruits, while the consumption of pre-processed tamarinds was a relatively rare occurrence. Likewise, such tamarinds could be opened for reasons that would not their involve rejection following initial processing (e.g., they were opened by birds, dropping to the forest floor, etc.), which may result in such fruits being generally more palatable to the lemurs overall.

Toothrow Processing Location. Ring-tailed lemurs primarily utilize the posterior toothrow to process large and challenging food items (e.g., tamarind, *Crateva excelsa* fruit, *Ficus coccifolia* fruit, discarded mangoes, etc.) while the anterior toothrow is primarily used to consume smaller, less challenging items [(e.g., small fruits and leaves) Sauther, 1992; Yamashita, 2003; Cuozzo and Sauther, 2006a; Millette, personal observations)]. At BMSR the tooth wear and resulting loss most commonly occurs for postcanine positions directly associated with tamarind fruit processing. (Sauther et al., 2002; Cuozzo and Sauther, 2004, 2006a). With the ablation of these positions, it is likely difficult for individuals to breakdown and consume this fruit using the posterior dentition. As a result, I posited that individuals could utilize alternative processing locations, particularly the anterior dentition (e.g., the incisors and canines). While this is a non-ideal location biomechanically for processing this food (e.g., most tamarind is processed posteriorly), use of the anterior dentition may nevertheless provide a means for individuals to access this resource following ablation of the posterior dentition.

The overall dataset suggests that individuals demonstrated positive associations with 3DAverage and 3DSum for each means of examining posterior processing (e.g., Posterior / Total; Posterior / Posterior + Anterior, and Both + Posterior /Posterior + Anterior + Both). As measures of overall toothrow size and average tooth size, these data indicate that individuals with reduced amounts of tooth wear are more likely to utilize the posterior dentition, suggesting that those with significant postcanine impairment utilize the anterior dentition (or both the anterior and posterior dentition) more frequently to process this food. Similarly, trends are present across all means of assessing processing location towards positive correlations for relief, and angularity when limited to Posterior / Total Observations, further suggesting a relationship between reduced tooth wear and use of the posterior dentition. These data suggest that

individuals with reduced amounts of dental impairment upon the postcanine dentition are more likely to use this portion of the toothrow. Thus, it is plausible that this is a compensatory behavior which allows individuals to continue processing this food item despite the loss of function for the postcanine dentition (see below).

When examined by sex, for all means of examining the data, females demonstrated significant positive correlations for the measure of angularity, while 3DSum was also associated with increasing use of the posterior dentition when rates of posterior use were calculated for all observations. In contrast, males demonstrated no significant correlations when the posterior measures were compared to all tamarind feeding observations. However, significant positive correlations were present for relief, 2DAverage, 3DSum and 3DAverage when limited to anterior and posterior observations, as well as when use of both anterior and posterior positions were included.

It is difficult to assess why patterns of posterior toothrow morphology and use differs between males and females. Again, reductions in sample size when splitting analyses by sex likely account for such issues. However, it is clear that all significant correlations are in a positive direction, as was also noted for the study sample overall. However, as angularity is believed to be associated with dental senescence, and reduced 2D/3D area averages and sums are associated with increasing wear and/or tooth loss, it appears that use of the posterior dentition most commonly occurs for those individuals with lower dental impairment status. These results are consistent with our previous observations that patterns of dental impairment at BMSR may cause impairment which interferes with the typical use of the posterior toothrow of ring-tailed lemurs to process larger food items. These data also indicate that individuals may behaviorally compensate against this effect by using alternative regions of the toothrow (e.g., the anterior

dentition) to breakdown this food item. Such divergent use of the toothrow to process food items may furthermore help explain the continued survival of individuals with significant damage to areas of the tooth row typically used to process this food key fallback food. While anterior positions likely receive damage due to consumption of this food, their use may provide the animal with an additional period of survival (and reproduction) following loss of function for the posterior dentition.

Use of Licking Behavior. Ring-tailed lemurs often utilize licking behaviors to process tamarind fruits following initial processing. Typically, this behavior occurs following opening of the shell using the dental apparatus, and the tongue is used to consume the food item through repeated strokes. This behavior likely removes softer, pulpy interior portions of the fruit, but may also soften the fruit through application of saliva (Sauther, 1992; Millette et al., 2009).

My previous master's level research at BMSR (Millette, 2007) indicates that licking occurs more commonly for individuals with tooth loss than for individuals without tooth loss. These previously collected data indicate that significant dental impairment may result in the use of this behavior in compensation for tooth loss, however, the impact of tooth form or wear status was not examined at that time.

For the total dataset, the only significant result was a positive correlation for angularity scores when the fraction of licking observances was compared to total tamarind feeding observations. Trends in the same direction were also present for licking when limited to confirmed yes or no observations, and when possible observations were included. These data provide evidence again that licking behavior is linked to a measure of dental impairment, although the directionality of this behavior is in the opposite of that expected. Angularity was

hypothesized to be negatively correlated with licking if this behavior is related to increasing dental wear. It may, however, be the case that increased angularity does allow individuals to better access fruits, thus allowing the licking behavior which occurs afterwards.

When assessed by sex, females demonstrated negative correlations for 2DSum and 3DSum with respect to when limited to “yes and no” observations and when possible observations were recorded. No significant observations were noted for any direct topographic measure. As these measures are linked directly with tooth loss status, these are convergent with those reported by Millette et al., 2009. Licking behavior thus appears to be linked with compensation for dental impairment among female individuals.

In contrast, data derived from males demonstrated positive correlations for angularity and 3DSum for analyses limited to confirmed “yes or no” observations and for those with possible observations included. These data suggest that males with reduced dental impairment engaged in higher frequencies of licking behavior. Likewise, for these groupings of the data, positive trends were also observed for slope. As with the overall dataset, results for males run contrary to the expected direction, and from the direction observed for females for 3DSum. These data may indicate that males may not use licking to process tamarind fruit in the same manner as females, and no data presented here suggest that males use this behavior in compensation for dental impairment. As noted before, individuals who are more able to access foods may simply be more likely to lick them because they have the opportunity to do so. While no differences were noted among females for angularity, the significant correlations for males suggest that these individuals may be also driving general positive trends and correlations observed. Although no significant patterns of angularity were observed for females, their correlations were in a positive direction, thus explaining the overall patterns of increased licking in association with higher angularity.

In all, it appears that data presented here provide limited support for the use of licking as a compensatory behavior among females, but overall patterns and those for males do not provide support for this hypothesis. This is different than observed during my earlier research at BMSR which suggested a general link between tooth loss and licking behavior. One explanation for the divergence in behaviors observed during this study is that the previous study only examined the impact of tooth loss, while this study measured dental wear in addition to tooth loss. It is of note that the 3DSum and 2DSum measures for females were as expected, and these measures should more accurately reflect the results of tooth loss than the other measures presented here. This, however, does not explain divergent patterns between males and females for 3DSum measures, although these differences may be related to the small sample size available for males or due to differences in access to preferred (or tamarinds which may be effectively broken down by licking). Additionally, it is also important to note that data collected during this study were recorded across both the wet and dry seasons while my previous research only focused on the dry season. Differences in the patterning of licking behavior relative to dental impairment may therefore reflect seasonal variations in tamarind phenological state (e.g., average ripeness of fruit by season). Tamarind fruit may be more suitable for licking during this time than during the wet season, thus impacting the patterns of results seen here in comparison to work conducted previously at this site.

Use of Tamarinds from the Ground. Tamarinds sourced from the ground were frequently used by ring-tailed lemurs during this study. It was hypothesized that these tamarinds would likely be easier to open than those sourced from trees as I had often observed that fruits found on the forest floor were often overripe and/or generally easy to break apart. Data presented in the

chapter do not provide any significant results other than a single positive correlation between use of tamarinds from the ground and slope for females. A trend towards a negative correlation was also noted for 2DAverage for males.

The data provide little evidence that dental impairment is associated with use of tamarind fruit from the ground. The only significant correlation is for slope for females and is in the opposite direction from that hypothesized. Overall, there is not a strong association between dental form and use of tamarind from the ground, suggesting that these fruit do not differ in their ease of access or processing from those found within the trees. While many tamarinds found on the ground do appear to be weaker and easier to access, it is likely that such fruits are less preferable to those found within the trees and are thus not consumed at a higher rate among those with dental impairment. It may also be likely that individuals only use those which have recently fallen and are thus of similar mechanical properties to those sourced arboreally, which could limit the use of these fruits by those with dental impairment.

Tamarind Processing and Dental Impairment.

The Impact of Dental Impairment on Tamarind Processing Capacity. Tamarind fruit provides a major food source for ring-tailed lemurs at BMSR, particularly during the dry season (Sauther, 1998; Sauther and Cuozzo, 2009; Yamashita et al., 2012, 2015b). Given this food item's challenging mechanical properties, I anticipated that feeding behaviors would reflect the impact of dental impairment. Likewise, I anticipated that behaviors related to processing this fruit would reflect those associated with an impaired ability to consume this food (e.g., increased feeding durations), as well as the occurrence of those which are used in compensation for dental impairment (use of the hands, licking, use of open pods, etc.).

With respect to negative impacts on capacity to feed on tamarinds, I hypothesized that individuals would take longer to process each pod, resulting in an overall reduced capacity to consume nutrients from this source. Overall, when limited to non-parametric tests, there is little evidence that increasing dental impairment is associated with longer feeding bout lengths. However, a number of negative correlations are present for 2DAverage for individuals overall and for females with respect to area measures when examined parametrically. These data, suggest that individuals with reduced postcanine area may indeed be less capable at processing this food item, at least for female individuals. No such patterns were noted for males, possibly due to sample size. However, angularity was positively associated with increased feeding times for all individuals and for males when assessed nonparametrically, and for all individuals and females when assessed parametrically. These data suggest that individuals with less impairment take longer to process this fruit, counter to expectations. While this may indicate that those with greater angularity spend more time on each tamarind, it does not support the concept that individuals with dental impairment take longer to process this food. Overall, these results are equivocal with regards to individual capacity to process tamarind fruit and it is difficult to make any firm statements concerning the impact of dental impairment on tamarind processing times or ability to access this resource from feeding times alone. Data from females concerning area do suggest that large changes in tooth area available reduce capacity to process this food, suggesting that high-level damage to the teeth does impact ability to access this resource (as reported previously when examining tooth loss status). Nevertheless, the overall lack of significant results may indicate that individuals may compensate behaviorally for dental impairment using alternative food processing techniques (see below).

Behaviors Used in Compensation for Dental Impairment. A number of the behaviors that I examined appear to be associated with behavioral compensation for dental impairment. In particular, manual processing and use of opened tamarind fruit appear to be most strongly associated with compensation for postcanine dental impairment. While significant negative correlations are present for a variety of measures across the dataset overall, use of open fruits tend to be most common among males, and females do not appear to engage in this behavior with respect to dental wear at the same frequencies (except for when considering angularity). Thus, while these data do indicate that this behavior is likely used in compensation for behavior, it appears that it is primarily a behavior utilized by males. As noted above, this may be related to differential access to resources between the sexes, and males with dental impairment may be more likely to eat opened pods as they do not have equal access to relatively easy-to-process fruit. Likewise, use of manual processing appears to be associated with compensation for dental impairment for this population. While these data are significant for a variety of measures overall and for males, as values for females only become significant when possible observations were recorded, use of the hands also appears to be primarily used by males. Again, patterns observed here are likely related to priority of access, but do suggest that this behavior is related to compensation for dental impairment within this population. Finally, data presented for food processing location indicate that animals adjust their use of the dental arcade to account for reduced postcanine function. For the population overall use of the posterior dentition is particularly associated with area measurements, which suggests that tooth loss or extensive tooth wear of this area that removes significant portions of the tooth are associated with reduced use of the posterior dentition. These data indicate that behavioral compensation for tooth wear and or

loss may not only include use of alternative foods or use of divergent processing techniques, but also may simply be achieved by shifting the positioning of the fruit during processing.

The other behaviors examined appear to be less likely to be used by individuals as a means of compensation for dental impairment. No significant negative associations were observed for use of tamarind from the ground, suggesting that individuals use these fruits irrespective of dental impairment status. As these fruits tend to be older and are often rotted or fermented (Millette, personal observations), it is likely that animals only eat these when newly fallen irrespective of tooth loss status. Likewise, only one significant negative correlation was found for use of pre-processed food items relative to impairment status (slope for males, when possibly processed fruits were included), suggesting that consumption of conspecific-processed foods is not used as a strategy against the impacts of dental impairment in this population. As noted above, as these conspecific foods have been rejected by others rather than eaten, these may be generally unpalatable tamarinds and are thus not consumed irrespective of dental status. Finally, equivocal data are present for use of licking behavior with respect to topographic measures. While, 3D and 2D sum measures suggest that females may use this behavior in compensation for dental impairment in the form of tooth loss, correlations in the opposite direction for males for angularity and 3DSum indicate that increasing impairment is likely not associated with this behavior among males. While licking may be a compensatory behavior observed among females, there is little to suggest that it is used by male individuals. The source of this difference, however, remains unknown at this time. The use of licking behavior, while a potentially compensatory behavior, therefore, remains unconfirmed for individuals overall at this time and must be subject to future inquires.

Part I Conclusion.

Overall, data presented in this chapter indicate that postcanine dental wear and loss are associated with divergent patterns of tamarind fruit consumption for BMSR ring-tailed lemurs. The impact of reduced postcanine dental topography appears to be relatively limited in terms of its negative effects upon time spent processing tamarind fruit when limited to strict topographic measures (slope and relief), although parametric measures do suggest that reduced surface area is associated with reduced capacity to effectively process this food among females (and overall for 2DAverage). This finding is coherent with previous work demonstrating that individuals with tooth loss take longer to process tamarinds than those who do not. The relative lack of significant findings for topographic measures in the expected direction, however, suggest that alternative food processing techniques may counter the impact of dental impairment on individual's ability to process foods.

The use of manual processing, consumption of previously opened fruit, and reduced use of the posterior dentition, appears to be the major means by which individuals may compensate for postcanine impairment when eating tamarind fruit. Likewise, licking behavior may be utilized as a means for accessing fruits despite dental impairment among females. These behavioral mechanisms may be why dental impairment is only weakly linked to feeding duration for measures that do not account for tooth loss. If individuals can compensate through behavioral mechanisms, than accessing tamarind fruit should in theory take little to no more time than if processed using only the dentition.

Interestingly, there do appear to be differences between the sexes in their expression of compensatory behaviors. For both manual processing and use of open tamarinds, a greater number of topographic measures are associated with the occurrence of these behaviors for males

than for females. In turn, compensatory licking behaviors appear to be more closely associated with females (as males demonstrate a positive correlation with reduced dental wear and this behavior). These findings may be related to differences in sample sizes between the sexes available for this study, as the number of males analyzed was lower than that for females. Alternatively, it is likely that males and females may utilize divergent patterns of behavior to compensate for dental wear and/or tooth loss. Such divergent patterns may be related to patterns of intersexual dominance in this species, as females should have preferential access to foods which are easier to process dentally. This may explain why clearly compensatory behaviors (such as the use of the hands or open fruit) appear to be more closely associated with dental impairment among males, as they have reduced access preferred fruits. Likewise, if males are more likely to utilize alternative food processing behaviors than are females, this may also provide some explanation for why female durations are longer for those increasing impairment when area measurements are considered parametrically. If female individuals do not learn to use such behaviors prior to the onset of significant tooth wear, it is possible that they simply spend more time processing tamarinds to access this food once a significant wear state has been achieved.

Finally, it is of note that this study has linked dental topography to interindividual differences in the expression of feeding behavior. I know of no study which has, to date, linked dental topography to behavioral patterns on an individual basis (although dental impairment has been suggested to reduce ability to access resources for individuals based on general age-based tooth wear patterns in Geladas; see Venkataraman et al., 2014). The results of this study suggest that dental topographic analysis provides data which are of sufficient resolution to distinguish divergent patterns of behavior in relation to dental impairment. It is also of note that simple area

measurements were generally more effective than were the measurements of slope, relief and angularity. This suggests that dental impairment may be quantified by simply examining the amount of dental area present for processing and mastication of food resources. It is also critical to emphasize that use of the entire dental row appears to be valid way of completing dental topographic analysis and may be successfully linked to behavior. This methodology differs from standard topographic studies which examine only one or two teeth. Use of the entire toothrow allows the calculation of 3D and 2D area sums which provide a means for assessing the impact of tooth loss within a topographic framework. This has been a major issue for topographic studies which cannot easily quantify the impacts of tooth loss, as this is difficult to model in GIS studies which focus on one or two teeth. While non-area measurements tended to be less commonly associated with dental impairment, angularity did appear to be relatively commonly associated with compensatory behaviors and durations. This is of note as this measure has been proposed as a key measure of dental functionality (Ungar and M'Kirera, 2003; M'Kirera and Ungar, 2003; see also Dennis et al., 2004). Data presented in this chapter may provide additional support for this hypothesis, as this measure tended to be more closely related with behavior than did relief or slope. Thus, changes in angularity may be an accurate indicator of dental senescence among nonhuman primates.

Part II Intro and Methods - Feeding Durations by Food Item Type, Species and Topography.

In Part II of this chapter I examine food processing behaviors associated with non-tamarind fruit food items. As with tamarind fruit, I collected data on the consumption of all other foods consumed by subject animals during focal follows. For each feeding bout, I recorded all

the forms of food processing data reported for tamarind above. However, the majority of food processing data that I recorded were specific to those behaviors associated with tamarind fruit feeding (e.g., use of open foods, manual processing, etc.), and were not very well suited to the assessment of dental topography for other food species, as these behaviors were rarely recorded during non-tamarind fruit feeding bouts. In contrast to tamarind fruit, the consumption most food items (e.g., fruits such as tsikidrakatse or mantsake) did not involve alternative food processing techniques such as use of the hands or use of open or preprocessed food items, and appeared to be associated with relatively stereotypical patterns of food processing. Feeding durations are thus one of the few means for examining food processing behaviors for non-tamarind fruit food items. It must be noted, however, that most feeding durations for non-tamarind fruit food items were not recorded for each individual item consumed (e.g., one fruit or leaf), as most foods eaten at BMSR are small in size and are consumed at a rapid rate that makes recording the consumption of individual food items difficult while recording multiple forms of food processing data simultaneously. With the exception of large food items (e.g., *Crateva excelsa*, Mango fruit, etc.) bout durations recorded reflect the consumption of multiple fruits, leaves, flowers, etc. As a result, the data I present in Part II of this chapter cannot provide direct insight into the relationships between the total amount of food and/or energy consumed by individuals during the course of feeding and dental topographic status. The production of such information will require the integration of feeding durations with total time spent feeding, and will be completed in future projects (see Conclusion / Chapter 8). However, divergences in feeding bout length may provide some information relation to the relationship between food processing ability and dental topographic measures as individuals with dental impairment are expected to be less efficient at

processing each individual item, and are therefore expected to take longer during each feeding bout.

As noted above, the same methods as those used for tamarind fruit feeding were employed when recording data for non-tamarind fruit food items. Likewise, durations for each food item type and species were examined using both mean and median duration lengths. As before, median durations were examined due to high variability in durations for the consumption of some foods. This was particularly the case for foods where it was difficult to determine bout end points due to rapid switching between individual foods, and for foods where extremely long bouts occurred on an occasional basis (e.g., wood / termite exudates). For example, small leaves (e.g., *Metaporana parvifolia*) or small fruits (e.g., *Salvadora angustifolia* or *Tarenna pruinosum*) were consumed extremely rapidly, which precluded recording bout lengths for each item individually, but also made determining bout end points difficult because the animals switched between plants so quickly that only one bout could be recorded. As such, while most feeding bouts were short, occasionally individuals would engage in feeding without a break needed to start a new bout for extended periods (e.g., up to approximately 15 minutes). Thus, use of median durations may provide a more accurate measure of central tendency for examining the relationship of feeding bout length to dental topography.

Additionally, I examined feeding bout durations relative to dental impairment using multiple classification systems for food item types. For example, it was often difficult to distinguish flowers from leaves for a number of food species (e.g., *S. angustifolia* flowers and leaves were difficult to distinguish during consumption), and determining the exact type of food consumed was often difficult due to observational conditions (see Chapter 3). As a result, leaves and flowers were combined into one category for assessment (e.g., Vegetative Material). That

said, as it was sometimes possible to distinguish the different food item types, data are also presented for leaves and leaf buds alone, as well as for flowers and flower buds alone for those instances where food type was clearly visible. Nevertheless, it must again be acknowledged that it was often difficult to distinguish the consumption of some foods by type as multiple food types were often consumed together at the same time and at a rapid rate. On a species level, however, this was usually less of a problem for major food species, and thus results are reported using both formats.

As with tamarind fruit, I assessed all data using non-parametric methodologies (Spearman's correlations) due to the potentially non-linear nature of the topographic measures employed. In addition, as not all food item types or species were eaten by each study individual, and some food species were only eaten by a very few individuals, only those foods which were consumed by more than 10 individuals were analyzed in this section. Due to generally small sample sizes for most food species, duration data were only examined for the study cohort overall, as separating the dataset into male and female categories typically resulted in sub-samples which were too small for statistical analysis.

Results.

Food Type vs. Topographic Measures. When leaves and flowers were combined into a single category, the topographic measure of relief was negatively associated median duration times (rho = -0.4008, p = 0.0254) and with mean durations for 3DSum (rho = -0.3528, p = 0.0516). Negative trends were observed form median slope and mean relief durations (Median slope: rho = -0.3406, p = 0.0608; Mean relief; rho = -0.03363, p = -0.0644). When assessed more specifically when leaves and flowers were separated, significant results were only noted for relief

for median durations for leaves and leaf buds in respect for relief (rho = -0.04444, p = 0.0123) (Table 4.12). Likewise, for mean durations, flower and flower buds were positively associated with angularity (rho = 0.3461, p = 0.044) and negatively associated with 2DAverage (rho = -0.3835, p = 0.0332) mean durations. A negative trend was noted for median leaves and leaf buds for slope (rho = -0.3383, p = 0.0627), while a negative trend was also noted for mean durations for 3DAverage and flowers and flower buds [(rho = -0.3411, p = 0.0604) Tables 4.11-4.14]

Table 4.11. Mean Durations for Foods with Vegetative Materials Included.

	Slope			Relief			Angularity		
	rho =	p =		rho =	p =		rho =	p =	
Wood	0.4053	0.0852	↔	0.507	***0.0267***	↗	0.3877	0.101	↔
Vegetative	-0.2782	0.1296	↔	-0.3363	*0.0644*	↘	0.1133	0.5439	↔
Soil	-0.1724	0.3623	↔	-0.2352	0.211	↔	-0.2102	0.2648	↔
Insect	0.1545	0.5036	↔	0.0558	0.81	↔	-0.3182	0.1598	↔
Fruit	-0.1472	0.4295	↔	-0.0931	0.6182	↔	0.2726	0.1379	↔
Feces	-0.1091	0.7495	↔	-0.0818	0.811	↔	0.0091	0.9788	↔

	3DSum			3DAverage			2DSum			2DAverage		
	rho =	p =		rho =	p =		rho =	p =		rho =	p =	
Wood	0.2895	0.2293	↔	0.2614	0.2797	↔	0.0807	0.7426	↔	-0.107	0.6628	↔
Vegetative	-0.3528	***0.0516***	↘	-0.3298	*0.07*	↘	-0.3012	0.0996	↔	-0.244	0.186	↔
Soil	-0.1773	0.3486	↔	-0.1528	0.4201	↔	-0.2018	0.2849	↔	-0.0736	0.699	↔
Insect	-0.1623	0.482	↔	-0.1013	0.6622	↔	-0.361	0.1078	↔	-0.2649	0.2458	↔
Fruit	-0.1544	0.4068	↔	-0.1573	0.3982	↔	-0.1669	0.3694	↔	-0.0984	0.5985	↔
Feces	0.0727	0.8317	↔	-0.3273	0.3259	↔	0.1	0.7699	↔	-0.4818	0.1334	↔

Table 4.12. Median Durations for Foods with Vegetative Materials Included.

	Slope			Relief			Angularity		
	rho =	p =		rho =	p =		rho =	p =	
Wood	0.714	***0.0006***	↗	0.8035	***<.0001***	↗	0.6333	***0.0036***	↗
Vegetative	-0.3406	*0.0608*	↘	-0.4008	***0.0254***	↘	-0.0752	0.6876	↔
Soil	-0.0923	0.6275	↔	-0.1172	0.5372	↔	-0.164	0.3866	↔
Insect	-0.1987	0.3879	↔	-0.2896	0.2029	↔	-0.526	***0.0143***	↘
Fruit	0.0089	0.9622	↔	0.0601	0.7481	↔	0.367	***0.0423***	↗
Feces	0.2545	0.45	↔	0.3	0.3701	↔	0.0636	0.8525	↔

	3DSum			3DAverage			2DSum			2DAverage		
	rho =	p =		rho =	p =		rho =	p =		rho =	p =	
Wood	0.5877	***0.0081***	↗	0.5807	***0.0091***	↗	0.3737	0.115	↔	0.1947	0.4243	↔
Vegetative	-0.2428	0.1882	↔	-0.2307	0.2119	↔	-0.1159	0.5345	↔	-0.0054	0.9768	↔
Soil	-0.0759	0.6903	↔	-0.0674	0.7234	↔	-0.087	0.6476	↔	-0.0069	0.9711	↔
Insect	-0.4662	***0.0331***	↘	-0.3961	*0.0755*	↘	-0.5481	***0.0101***	↘	-0.4117	*0.0637*	↘
Fruit	-0.0232	0.9015	↔	-0.0173	0.9262	↔	-0.0702	0.7076	↔	-0.0302	0.8717	↔
Feces	0.3273	0.3259	↔	0.0364	0.9155	↔	0.3455	0.2981	↔	-0.1455	0.6696	↔

Table 4.13. Mean Bout Duration by Food Type by Topographic Measure.

	Slope			Relief			Angularity		
By Mean Duration	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Flower Bud / Flower	-0.1617	0.3848	↔	-0.1536	0.4093	↔	0.3641	***0.044***	↗
Flower Bud / Fruit	-0.3	0.6238	↔	-0.3	0.6238	↔	-0.6	0.2848	↔
Flowers and Leaves	0.2025	0.3014	↔	0.127	0.5196	↔	-0.0115	0.9537	↔
Fruit	-0.1472	0.4295	↔	-0.0931	0.6182	↔	0.2726	0.1379	↔
Insect	0.1545	0.5036	↔	0.0558	0.81	↔	-0.3182	0.1598	↔
Leaves / Leaf Buds	-0.2149	0.2456	↔	-0.2976	0.104	↔	-0.0552	0.7679	↔
Other / Trash	-0.0466	0.8591	↔	-0.0833	0.7505	↔	0.0441	0.8665	↔
Soil Mean	-0.1724	0.3623	↔	-0.2352	0.211	↔	-0.2102	0.2648	↔
Wood	0.4053	0.0852	↔	0.507	***0.0267***	↗	0.3877	0.101	↔

	3DSum			3DAverage			2DSum			2DAverage		
	rho =	**p =**		**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Flower Bud / Flower	-0.3153	0.084	↔	-0.3411	*0.0604*	↘	-0.3008	0.1001	↔	-0.3835	***0.0332***	↘
Flower Bud / Fruit	0.3	0.6238	↔	0.3	0.6238	↔	0.5	0.391	↔	0.5	0.391	↔
Flowers and Leaves	0.0099	0.9603	↔	0.1544	0.4329	↔	-0.0772	0.6963	↔	0.1845	0.3474	↔
Fruit	-0.1544	0.4068	↔	-0.1573	0.3982	↔	-0.1669	0.3694	↔	-0.0984	0.5985	↔
Insect	-0.1623	0.482	↔	-0.1013	0.6622	↔	-0.361	0.1078	↔	-0.2649	0.2458	↔
Leaves / Leaf Buds	-0.2645	0.1504	↔	-0.2488	0.1771	↔	-0.2681	0.1447	↔	-0.2194	0.2358	↔
Other / Trash	-0.0466	0.8591	↔	-0.0294	0.9108	↔	-0.1103	0.6735	↔	-0.0025	0.9926	↔
Soil Mean	-0.1773	0.3486	↔	-0.1528	0.4201	↔	-0.2018	0.2849	↔	-0.0736	0.699	↔
Wood	0.2895	0.2293	↔	0.2614	0.2797	↔	0.0807	0.7426	↔	-0.107	0.6628	↔

Table 4.14. Median Bout Duration of Food Type Bouts by Topographic Measure.

	Slope			Relief			Angularity		
By Median Duration	**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Flower Bud / Flower	-0.1399	0.4528	↔	-0.1601	0.3897	↔	0.2415	0.1905	↔
Flower Bud / Fruit	-0.1	0.8729	↔	-0.1	0.8729	↔	-0.2	0.7471	↔
Flowers and Leaves	0.2458	0.2075	↔	0.243	0.2127	↔	-0.0022	0.9912	↔
Fruit	0.0089	0.9622	↔	0.0601	0.7481	↔	0.367	***0.0423***	↗
Insect	-0.1987	0.3879	↔	-0.2896	0.2029	↔	-0.526	***0.0143***	↘
Leaves / Leaf Buds	-0.3383	*0.0627*	↘	-0.4444	***0.0123***	↘	-0.2407	0.1921	↔
Other / Trash	-0.0931	0.7222	↔	-0.0662	0.8008	↔	0.1103	0.6735	↔
Soil Mean	-0.0923	0.6275	↔	-0.1172	0.5372	↔	-0.164	0.3866	↔
Wood	0.714	***0.0006***	↗	0.8035	***<.0001***	↗	0.6333	***0.0036***	↗

	3DSum			3DAverage			2DSum			2DAverage		
	rho =	**p =**		**rho =**	**p =**		**rho =**	**p =**		**rho =**	**p =**	
Flower Bud / Flower	-0.244	0.186	↔	-0.2452	0.1837	↔	-0.1883	0.3104	↔	-0.2206	0.2331	↔
Flower Bud / Fruit	0.6	0.2848	↔	0.6	0.2848	↔	0.7	0.1881	↔	0.7	0.1881	↔
Flowers and Leaves	0.0974	0.6219	↔	0.2162	0.2692	↔	-0.0744	0.7066	↔	0.1275	0.5178	↔
Fruit	-0.0232	0.9015	↔	-0.0173	0.9262	↔	-0.0702	0.7076	↔	-0.0302	0.8717	↔
Insect	-0.4662	***0.0331***	↘	-0.3961	*0.0755*	↘	-0.5481	***0.0101***	↘	-0.4117	*0.0637*	↘
Leaves / Leaf Buds	-0.2254	0.2228	↔	-0.2133	0.2493	↔	-0.0694	0.7108	↔	0.0669	0.7205	↔
Other / Trash	-0.0588	0.8226	↔	-0.0686	0.7935	↔	-0.1127	0.6666	↔	-0.0637	0.808	↔
Soil Mean	-0.0759	0.6903	↔	-0.0674	0.7234	↔	-0.087	0.6476	↔	-0.0069	0.9711	↔
Wood	0.5877	***0.0081***	↗	0.5807	***0.0091***	↗	0.3737	0.115	↔	0.1947	0.4243	↔

With respect to fruit, higher angularity was associated with increased median durations (rho = -0.367, p = -0.0423). In contrast, insects were associated with negative median feeding bout lengths for angularity (rho = -0.526, p = 0.0143) and 3DSum (rho = -0.4662, p = 0.0331) and 2DSum (-0.5481, p = 0.0101). Likewise, for insects, median durations were trending towards negative correlations for 3DAverage (rho = 0.3961, p = 0.0755), and 2DAverage (rho = -0.4117, p = 0.0637). No significant correlations, however, were noted for insects for mean measures. Of all food types, wood feeding was most closely associated with median feeding durations, with significant positive correlations being present for slope (rho = 0.714, p = 0.0006), relief (rho = 0.8035, p < 0.0001), angularity (rho = 0.6333, p = 0.0036), 3DSum (rho = 0.5877, p = 0.0081), 3DAverage (rho = 0.0587, p = 0.0091). Likewise a positive correlation was present for mean wood feeding times for relief [(rho = 0.507, p = 0.0267) Tables 4.11-4.12].

Food Species and Type by Topographic Measures.

Leaves and Flowers Combined into Vegetative Materials. When leaves and flowers were combined, no significant correlations were noted for any measure for the species of *Acacia bellula, Cedrelopsis grevei, Commicarpus* sp., *Salvadora angustifolia or Secamone* sp. (Tables 4.15-4.16). Significant negative correlations were however noted for a variety of topographic measures across a number of other species (see below), with all significant correlations in a negative direction except for one positive correlation for angularity for *Gyrocarpus americanus* (rho = 0.5417, p = 0.0247). As such, for feeding bout durations a number of significant negative correlations were present for tamarind and *Tamelapsis linearis.* For tamarind this includes relief (rho = -0.4038, p = 0.0269), 2DSum (rho = -0.378, p = 0.0395), 2DAverage (rho = 0.4567, p = 0.0112), 3DSum (rho = -0.4268, p = 0.0186) and 3DAverage (rho = -0.4968, p = 0.0052) for

Table 4.15. Vegetative Portions of Plants by Mean Duration and Species (when n > 10).

	A. bellula				*C. grevei*				*Commicarpus* sp.			
	n =	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	15	0.2429	0.3831	↔	19	0.2281	0.3477	↔	11	0.2545	0.45	↔
Angularity	15	-0.0214	0.9396	↔	19	-0.1526	0.5328	↔	11	0.0455	0.8944	↔
Relief	15	0.1036	0.7134	↔	19	0.1772	0.468	↔	11	0.0545	0.8734	↔
2DSum	15	0.0893	0.7517	↔	19	-0.1105	0.6524	↔	11	-0.1364	0.6893	↔
2DAverage	15	-0.1107	0.6945	↔	19	-0.0526	0.8306	↔	11	-0.4455	0.1697	↔
3DSum	15	-0.0071	0.9798	↔	19	0.0825	0.7372	↔	11	-0.1	0.7699	↔
3DAverage	15	-0.0179	0.9496	↔	19	0.1105	0.6524	↔	11	-0.2	0.5554	↔

	M. parvifolia				*Pentopetio* sp.				*Q. papionae*			
	n =	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	31	0.0851	0.6491	↔	27	-0.3205	0.1031	↔	18	-0.1889	0.4529	↔
Angularity	31	0.3367	*0.064*	↘	27	-0.105	0.6022	↔	18	-0.0237	0.9255	↔
Relief	31	0.1359	0.4661	↔	27	-0.3321	0.0906	↔	18	-0.1868	0.458	↔
2DSum	31	0.1548	0.4056	↔	27	-0.3016	0.1263	↔	18	-0.3437	0.1626	↔
2DAverage	31	0.0944	0.6136	↔	27	-0.1984	0.3211	↔	18	-0.3127	0.2065	↔
3DSum	31	0.2024	0.2748	↔	27	-0.3523	*0.0715*	↘	18	-0.4551	*0.0577*	↘
3DAverage	31	0.1948	0.2938	↔	27	-0.3284	0.0944	↔	18	-0.4241	*0.0794*	↘

	T. dauphinensis				*T. indica*				*T. linearis*			
	n =	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	22	-0.1169	0.6045	↔	30	-0.3157	0.0892	↔	13	-0.511	*0.0743*	↘
Angularity	22	0.013	0.9543	↔	30	-0.1648	0.384	↔	13	0.1648	0.5905	↔
Relief	22	-0.0582	0.7971	↔	30	-0.4038	***0.0269***	↘	13	-0.5604	***0.0463***	↘
2DSum	22	-0.4523	***0.0346***	↘	30	-0.378	***0.0395***	↘	13	-0.3846	0.1944	↔
2DAverage	22	-0.4207	***0.0512***	↘	30	-0.4567	***0.0112***	↘	13	-0.2582	0.3943	↔
3DSum	22	-0.3586	0.1013	↔	30	-0.4269	***0.0186***	↘	13	-0.7033	***0.0073***	↘
3DAverage	22	-0.3337	0.1291	↔	30	-0.4968	***0.0052***	↘	13	-0.6813	***0.0103***	↘

	Secamone sp.				*G. americanus*				*S. angustifolia*			
	n =	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	20	0.1383	0.5608	↔	17	0.0368	0.8886	↔	30	0.0154	0.9358	↔
Angularity	20	-0.2827	0.2272	↔	17	0.5417	***0.0247***	↗	30	0.0323	0.8656	↔
Relief	20	-0.0301	0.8998	↔	17	0.2377	0.3582	↔	30	-0.0483	0.8	↔
2DSum	20	-0.2812	0.2297	↔	17	0.1912	0.4623	↔	30	-0.0416	0.8272	↔
2DAverage	20	-0.0226	0.9248	↔	17	0.2451	0.343	↔	30	0.1996	0.2904	↔
3DSum	20	-0.0932	0.6958	↔	17	0.1373	0.5994	↔	30	-0.0905	0.6342	↔
3DAverage	20	0.0451	0.8502	↔	17	0.1863	0.4741	↔	30	0.0318	0.8675	↔

	I. majungensis			
	n =	**rho =**	**p =**	
Slope	23	-0.3785	*0.075*	↘
Angularity	23	-0.2787	0.1979	↔
Relief	23	-0.4368	0.0372	↔
2DSum	23	-0.3399	0.1125	↔
2DAverage	23	-0.2332	0.2842	↔
3DSum	23	-0.4002	*0.0585*	↘
3DAverage	23	-0.3874	*0.0678*	↘

Table 4.16. Vegetative Portions of Plants by Median Duration and Species (when n > 10).

	A. bellula				*C. grevei*				*Commicarpus* sp.			
	n =	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	15	-0.0893	0.7517	↔	19	0.1333	0.5863	↔	11	0.0727	0.8317	↔
Angularity	15	0.1321	0.6387	↔	19	-0.1754	0.4725	↔	11	-0.1818	0.5926	↔
Relief	15	-0.0536	0.8496	↔	19	0.0912	0.7103	↔	11	-0.1091	0.7495	↔
2DSum	15	0.0571	0.8397	↔	19	-0.086	0.7264	↔	11	0.2091	0.5372	↔
2DAverage	15	0.0214	0.9396	↔	19	0.0018	0.9943	↔	11	0.0545	0.8734	↔
3DSum	15	0.0429	0.8795	↔	19	0.0439	0.8585	↔	11	0.0818	0.811	↔
3DAverage	15	0.0429	0.8795	↔	19	0.0877	0.721	↔	11	0.0182	0.9577	↔

	M. parvifolia				*Pentopetio* sp.				*Q. papionae*			
	n =	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	31	-0.0673	0.7189	↔	27	-0.3654	*0.0609*	↘	18	0.0299	0.9062	↔
Angularity	31	-0.0056	0.976	↔	27	0.0626	0.7565	↔	18	-0.0444	0.8612	↔
Relief	31	-0.031	0.8683	↔	27	-0.3819	***0.0493***	↘	18	-0.0753	0.7664	↔
2DSum	31	0.0089	0.9622	↔	27	-0.1563	0.4362	↔	18	-0.1744	0.4888	↔
2DAverage	31	0.0952	0.6106	↔	27	-0.2641	0.1832	↔	18	-0.1496	0.5534	↔
3DSum	31	0.0431	0.8177	↔	27	-0.2409	0.2261	↔	18	-0.3498	0.1547	↔
3DAverage	31	0.0698	0.7092	↔	27	-0.345	*0.078*	↘	18	-0.3251	0.1881	↔

	T. dauphinensis				*T. indica*				*T. linearis*			
	n =	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	22	0.0457	0.8398	↔	30	-0.3535	*0.0553*	↘	13	-0.4505	0.1223	↔
Angularity	22	0.1067	0.6364	↔	30	-0.1933	0.306	↔	13	0.0934	0.7615	↔
Relief	22	0.135	0.5493	↔	30	-0.386	***0.0351***	↘	13	-0.511	*0.0743*	↘
2DSum	22	-0.2208	0.3235	↔	30	-0.2334	0.2146	↔	13	-0.4451	0.1275	↔
2DAverage	22	-0.1801	0.4225	↔	30	-0.2356	0.2101	↔	13	-0.2747	0.3637	↔
3DSum	22	-0.1191	0.5974	↔	30	-0.3241	0.0806	↔	13	-0.7143	***0.0061***	↘
3DAverage	22	-0.0807	0.7209	↔	30	-0.3722	***0.0428***	↘	13	-0.6648	***0.0132***	↘

	Secamone sp.				*S. angustifolia*				*G. americanus*			
	n =	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	20	-0.0241	0.9198	↔	30	0.0625	0.7428	↔	17	-0.2426	0.348	↔
Angularity	20	-0.3218	0.1665	↔	30	0.0794	0.6765	↔	17	0.4534	*0.0675*	↘
Relief	20	-0.188	0.4274	↔	30	0.0274	0.8859	↔	17	-0.0662	0.8008	↔
2DSum	20	-0.3053	0.1906	↔	30	-0.1408	0.4579	↔	17	0.1176	0.6529	↔
2DAverage	20	-0.009	0.9699	↔	30	-0.0033	0.986	↔	17	0.1422	0.5863	↔
3DSum	20	-0.1835	0.4388	↔	30	-0.1043	0.5832	↔	17	0.0368	0.8886	↔
3DAverage	20	-0.0346	0.8849	↔	30	-0.022	0.908	↔	17	0.049	0.8518	↔

	I. majungensis			
	n =	**rho =**	**p =**	
Slope	23	-0.1354	0.5378	↔
Angularity	23	-0.1285	0.5589	↔
Relief	23	-0.2264	0.2989	↔
2DSum	23	-0.0974	0.6585	↔
2DAverage	23	-0.1626	0.4584	↔
3DSum	23	-0.1211	0.582	↔
3DAverage	23	-0.1567	0.4752	↔

mean durations, as well as relief (rho = -0.386, p = 0.0351) and 3DAverage (rho = -0.3722, p = 0.0428) for median durations. For *Tamelapsis linearis*, negative correlations were present for mean durations with respect to relief (rho = -0.5604, p = 0.0463), 3DSum (rho = -0.7033, p = 0.0073) and 3DAverage (rho = -0.6813, p = 0.0103), as well as for median durations for 3DSum (rho = -0.7143, p = 0.0061) and 3DAverage (rho = -0.6648, p = 0.0132). Finally, significant negative correlations were noted for *Talinella dauphinensis* for mean feeding durations 2DSum (rho = -0.4523, p = 0.0346) and 2DAverage (rho = -0.4207, p = 0.0512) and *Pentopetio* sp. for median durations and relief (rho = -0.3819, p = 0.0493). Additionally, a number of trends (0.05 < p < 0.08) were noted in a negative direction for a number of measures and species. While not discussed directly here, all of these were in the same direction as the majority of significant values (e.g., they were negative associations between topographic measures and durations). All trends are visible in Tables 4.15 and 4.16.

Flowers and Flower Buds. Lemurs were only observed to consume four flower species at rates high enough to examine statistical patterns (e.g., consumption by 10 or more individuals), these included *Gyrocarpus americanus*, *Ipomoea majungensis*, *Quisivianthe papionae*, and *Salvadora angustifolia*. For these species, significant negative correlations were noted for median durations for slope for *G. americanus* (rho = -0.5179, p = 0.048) and relief for mean durations for *I. majungensis* (rho = -0.4368, p = 0.0372). Negative trends, were also observed for mean flower feeding durations for a number of measures for *I. majungensis* for slope (rho = -0.3785, p = 0.075), 3DSum (rho = -0.4002, p = 0.0585), and 3DAverage (rho = -0.3874, p = 0.0678), as well as for *Q. papionae* for 3DSum (rho = -0.4551, p = 0.0577) and 3DAverage [(rho = -0.4241, p = 0.0794) Table 4.17].

Table 4.17. Feeding Bout Duration for Flowers and Flower Buds (where n > 10).

	G. americanus				*I. majungensis*			
Mean Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	15	-0.225	0.4201	↔	23	*-0.3785*	*0.075*	↘
Angularity	15	0.4214	0.1177	↔	23	-0.2787	0.1979	↔
Relief	15	0.0071	0.9798	↔	23	-0.4368	***0.0372***	↘
2DSum	15	0.1786	0.5243	↔	23	-0.3399	0.1125	↔
2DAverage	15	0.175	0.5327	↔	23	-0.2332	0.2842	↔
3DSum	15	0.0643	0.8199	↔	23	*-0.4002*	*0.0585*	↘
3DAverage	15	0.05	0.8595	↔	23	*-0.3874*	*0.0678*	↘
Median Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	15	-0.5179	***0.048***	↘	23	-0.1354	0.5378	↔
Angularity	15	0.3286	0.2318	↔	23	-0.1285	0.5589	↔
Relief	15	-0.3143	0.2539	↔	23	-0.2264	0.2989	↔
2DSum	15	0.1357	0.6296	↔	23	-0.0974	0.6585	↔
2DAverage	15	0.1	0.7229	↔	23	-0.1626	0.4584	↔
3DSum	15	-0.0214	0.9396	↔	23	-0.1211	0.582	↔
3DAverage	15	-0.0893	0.7517	↔	23	-0.1567	0.4752	↔
	Q. papionae				***S. angustifolia***			
Mean Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	18	-0.1889	0.4529	↔	12	-0.074	0.7436	↔
Angularity	18	-0.0237	0.9255	↔	12	-0.1158	0.608	↔
Relief	18	-0.1868	0.458	↔	12	-0.092	0.6837	↔
2DSum	18	-0.3437	0.1626	↔	12	-0.1869	0.4049	↔
2DAverage	18	-0.3127	0.2065	↔	12	-0.1316	0.5595	↔
3DSum	18	-0.4551	*0.0577*	↘	12	-0.0559	0.8048	↔
3DAverage	18	-0.4241	*0.0794*	↘	12	-0.0695	0.7588	↔
Median Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	18	0.0299	0.9062	↔	12	-0.0322	0.8869	↔
Angularity	18	-0.0444	0.8612	↔	12	-0.092	0.6837	↔
Relief	18	-0.0753	0.7664	↔	12	-0.0446	0.8437	↔
2DSum	18	-0.1744	0.4888	↔	12	-0.2309	0.3011	↔
2DAverage	18	-0.1496	0.5534	↔	12	-0.1801	0.4225	↔
3DSum	18	-0.3498	0.1547	↔	12	-0.0582	0.7971	↔
3DAverage	18	-0.3251	0.1881	↔	12	-0.0774	0.7322	↔

Leaves and Leaf Buds. With respect to leaves and leaf buds, 10 species were available for analysis (i.e., more than 10 individuals were observed to consume these, see Table 4.18). As with flowers and flower buds, the vast majority of correlations present were in a negative direction between durations and topography. In this case, significant negative correlations for mean durations were noted for a number of measures for *Tamarindus indica*, including relief (rho = -

0.4109, p = 0.0241), 2DAverage (rho = -0.4403, p = 0.0149), 3DSum (rho = -0.4247, p = 0.0193) and 3DAverage (rho = -0.4981, p = 0.0051). For this species, median durations were associated with negative correlations for slope (rho = -0.418, p = 0.0215) and relief (rho = -0.4002, p = 0.0284). Similarly, a variety of negative correlations were present for *Tamelapsis linearis* mean feeding durations (relief: rho = -0.5604, p = 0.0463; 3DSum: rho = -0.7033, p = 0.0073; 3DAverage: rho = -0.6813, p = 0.0103), and for median feeding durations for the measures of 3DSum (rho = -0.7143, p = 0.0061) and 3DAverage (rho = -0.6648, p = 0.0132). Negative correlations were also noted for mean feeding durations and 2DSum and 2DAverage for *T. dauphinensis* (2DSum: rho = -0.4523, p = 0.035; 2DAverage: rho = -0.4207, p = 0.0512), and for median feeding durations for *Pentopetio* sp. for relief (rho = -0.3819, p = 0.0493). A number of positive correlations were found for *Salvadora angustifolia* leaves for mean feeding durations for angularity (rho = 0.6348, p = 0.0062), 2DSum (rho = 0.6275, p = 0.007), 2DAverage (rho = 0.4926, p = 0.0445), 3DSum (rho = 0.5368, p = 0.0263), and 3DAverage (0.5221, p = 0.0316). In turn, for *S. angustifolia* median durations, positive correlations were noted for angularity (rho = 0.6373, p = 0.0059) and 2DSum (rho = 0 .4828, p = 0.0496) and 3DSum (rho = 0.4755, p = 0.0537). Additionally a positive correlation was noted for Commicarpus slope for mean durations (rho = 0.6000, p = 0.0510). As with “vegetative” feeding, a number of trends were also noted, and were generally in the negative direction for most measures (but not for *Metaporana parvifolia* and *S. angustifolia*). All significant correlations and trends are reported in Tables 4.18 and 4.19

Table 4.18. Mean Leaf and Leaf Bud Durations by Species (where n > 10).

	A. bellula				*C. grevei*				*Commicarpus* sp.			
By Mean Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	**11**	0.0455	0.8944	↔	19	0.2281	0.3477	↔	11	0.6	***0.051***	↗
Angularity	**11**	-0.1364	0.6893	↔	19	-0.1526	0.5328	↔	11	0.1273	0.7092	↔
Relief	**11**	-0.2091	0.5372	↔	19	0.1772	0.468	↔	11	0.3636	0.2716	↔
2DSum	**11**	0.2455	0.4669	↔	19	-0.1105	0.6524	↔	11	0.0273	0.9366	↔
2DAverage	**11**	0.1636	0.6307	↔	19	-0.0526	0.8306	↔	11	-0.1909	0.5739	↔
3DSum	**11**	0.0091	0.9788	↔	19	0.0825	0.7372	↔	11	0.1273	0.7092	↔
3DAverage	**11**	0.0091	0.9788	↔	19	0.1105	0.6524	↔	11	0.1182	0.7293	↔

	S. angustifolia				*Secamone* sp.				*T. dauphinensis*			
By Mean Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	17	0.3064	0.2317	↔	26	0.1383	0.5608	↔	22	-0.1169	0.6045	↔
Angularity	17	0.6348	***0.0062***	↗	26	-0.2827	0.2272	↔	22	0.013	0.9543	↔
Relief	17	0.3676	0.1466	↔	26	-0.0301	0.8998	↔	22	-0.0582	0.7971	↔
2DSum	17	0.6275	***0.007***	↗	26	-0.2812	0.2297	↔	22	-0.4523	***0.0346***	↘
2DAverage	17	0.4926	***0.0445***	↗	26	-0.0226	0.9248	↔	22	-0.4207	***0.0512***	↘
3DSum	17	0.5368	***0.0263***	↗	26	-0.0932	0.6958	↔	22	-0.3586	0.1013	↔
3DAverage	17	0.5221	***0.0316***	↗	26	0.0451	0.8502	↔	22	-0.3337	0.1291	↔

	M. parvifolia				*Pentopetio* sp.			
By Mean Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	31	0.0851	0.6491	↔	27	-0.3205	0.1031	↔
Angularity	31	0.3367	*0.064*	↗	27	-0.105	0.6022	↔
Relief	31	0.1359	0.4661	↔	27	-0.3321	0.0906	↔
2DSum	31	0.1548	0.4056	↔	27	-0.3016	0.1263	↔
2DAverage	31	0.0944	0.6136	↔	27	-0.1984	0.3211	↔
3DSum	31	0.2024	0.2748	↔	27	-0.3523	*0.0715*	↘
3DAverage	31	0.1948	0.2938	↔	27	-0.3284	0.0944	↔

	T. indica				*T. linearis*			
By Mean Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	30	-0.3353	*0.0701*	↘	13	-0.511	*0.0743*	↘
Angularity	30	-0.1355	0.4753	↔	13	0.1648	0.5905	↔
Relief	30	-0.4109	***0.0241***	↘	13	-0.5604	***0.0463***	↘
2DSum	30	-0.3531	*0.0557*	↘	13	-0.3846	0.1944	↔
2DAverage	30	-0.4403	***0.0149***	↘	13	-0.2582	0.3943	↔
3DSum	30	-0.4247	***0.0193***	↘	13	-0.7033	***0.0073***	↘
3DAverage	30	-0.4981	***0.0051***	↘	13	-0.6813	***0.0103***	↘

.

Table 4.19. Median Leaf and Leaf Bud Durations by Species (where n > 10).

	A. bellula				*C. grevei*				*Commicarpus* sp.			
By Median Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	11	0.0818	0.811	↔	19	0.0263	0.9148	↔	11	0.2273	0.5015	↔
Angularity	11	0.0636	0.8525	↔	19	-0.2632	0.2764	↔	11	-0.1182	0.7293	↔
Relief	11	-0.0273	0.9366	↔	19	-0.0035	0.9886	↔	11	0.0364	0.9155	↔
2DSum	11	0.4273	0.1899	↔	19	-0.0263	0.9148	↔	11	0.2727	0.4171	↔
2DAverage	11	0.3727	0.2589	↔	19	0.0982	0.6891	↔	11	0.2091	0.5372	↔
3DSum	11	0.3455	0.2981	↔	19	0.0333	0.8922	↔	11	0.2091	0.5372	↔
3DAverage	11	0.3455	0.2981	↔	19	0.0754	0.7589	↔	11	0.2	0.5554	↔
	S. angustifolia				*Secamone* sp.				*T. dauphinensis*			
By Median Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	17	0.3186	0.2126	↔	26	-0.0241	0.9198	↔	22	0.0457	0.8398	↔
Angularity	17	0.6373	***0.0059***	↗	26	-0.3218	0.1665	↔	22	0.1067	0.6364	↔
Relief	17	0.3922	0.1195	↔	26	-0.188	0.4274	↔	22	0.135	0.5493	↔
2DSum	17	0.4828	***0.0496***	↗	26	-0.3053	0.1906	↔	22	-0.2208	0.3235	↔
2DAverage	17	0.3946	0.117	↔	26	-0.009	0.9699	↔	22	-0.1801	0.4225	↔
3DSum	17	0.4755	***0.0537***	↗	26	-0.1835	0.4388	↔	22	-0.1191	0.5974	↔
3DAverage	17	0.473	*0.0551*	↗	26	-0.0346	0.8849	↔	22	-0.0807	0.7209	↔
	M. parvifolia				*Pentopetio* sp.							
By Median Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**					
Slope	31	-0.0673	0.7189	↔	27	-0.3654	*0.0609*	↘				
Angularity	31	-0.0056	0.976	↔	27	0.0626	0.7565	↔				
Relief	31	-0.031	0.8683	↔	27	-0.3819	***0.0493***	↘				
2DSum	31	0.0089	0.9622	↔	27	-0.1563	0.4362	↔				
2DAverage	31	0.0952	0.6106	↔	27	-0.2641	0.1832	↔				
3DSum	31	0.0431	0.8177	↔	27	-0.2409	0.2261	↔				
3DAverage	31	0.0698	0.7092	↔	27	-0.345	*0.078*	↘				
	T. indica				*T. linearis*							
By Median Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**					
Slope	30	-0.418	***0.0215***	↘	13	-0.4505	0.1223	↔				
Angularity	30	-0.0679	0.7216	↔	13	0.0934	0.7615	↔				
Relief	30	-0.4002	***0.0284***	↘	13	-0.511	*0.0743*	↘				
2DSum	30	-0.1359	0.4739	↔	13	-0.4451	0.1275	↔				
2DAverage	30	-0.1462	0.4409	↔	13	-0.2747	0.3637	↔				
3DSum	30	-0.305	0.1012	↔	13	-0.7143	***0.0061***	↘				
3DAverage	30	-0.3531	*0.0557*	↘	13	-0.6648	***0.0132***	↘				

Fruit. Five species (other than tamarind) were consumed by more than 10 individuals (*Bridelia* sp., *Crateva excelsa*, *Grewia leucophylla*, *S. angustifolia*, and *Tarenna pruinosum*. Of these, significant correlations for durations were only noted for *S. angustifolia.* Here positive correlations were noted for median durations for both slope (rho = 0.5211, p = 0.0045), and relief [(rho = 0.5261, p = 0.0040) Table 4.20].

Table 4.20. Feeding Durations by Fruit Species (where n > 10, Tamarind Excluded).

	***Bridelia* sp.**				***C. excelsa***				***G. leucophylla***			
By Mean Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	13	0.0549	0.8585	↔	12	-0.2168	0.4986	↔	13	-0.4396	0.1329	↔
Angularity	13	0.4835	0.0941	↔	12	-0.1958	0.5419	↔	13	0.2747	0.3637	↔
Relief	13	0.2033	0.5053	↔	12	-0.1469	0.6488	↔	13	-0.4176	0.1557	↔
2DSum	13	0.1154	0.7074	↔	12	0.3217	0.3079	↔	13	-0.1154	0.7074	↔
2DAverage	13	0.0989	0.7479	↔	12	0.2517	0.4299	↔	13	-0.1374	0.6545	↔
3DSum	13	0.1978	0.5171	↔	12	0.042	0.897	↔	13	-0.2967	0.3249	↔
3DAverage	13	0.1538	0.6158	↔	12	-0.0769	0.8122	↔	13	-0.3132	0.2974	↔
By Median Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	13	0.2308	0.4481	↔	12	-0.2657	0.4038	↔	13	-0.4066	0.168	↔
Angularity	13	0.3462	0.2466	↔	12	-0.1958	0.5419	↔	13	0.1593	0.6031	↔
Relief	13	0.3077	0.3064	↔	12	-0.2098	0.5128	↔	13	-0.3901	0.1876	↔
2DSum	13	0.1978	0.5171	↔	12	0.3497	0.2652	↔	13	-0.1593	0.6031	↔
2DAverage	13	0.1813	0.5533	↔	12	0.2727	0.3911	↔	13	-0.2198	0.4706	↔
3DSum	13	0.3407	0.2547	↔	12	0.021	0.9484	↔	13	-0.3022	0.3156	↔
3DAverage	13	0.3462	0.2466	↔	12	-0.1049	0.7456	↔	13	-0.3516	0.2387	↔

	S. angustifolia				***T. pruinosum***			
By Mean Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	28	0.306	0.1133	↔	21	0.0766	0.7413	↔
Angularity	28	0.1314	0.5052	↔	21	0.1429	0.5367	↔
Relief	28	0.3186	0.0985	↔	21	0.1519	0.5109	↔
2DSum	28	-0.198	0.3135	↔	21	-0.061	0.7927	↔
2DAverage	28	-0.281	0.1478	↔	21	0.0403	0.8624	↔
3DSum	28	0.0411	0.8357	↔	21	0.0104	0.9643	↔
3DAverage	28	0.0454	0.8184	↔	21	0.1143	0.6218	↔
By Median Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	28	0.5211	***0.0045***	↗	21	-0.1156	0.6178	↔
Angularity	28	0.3003	0.1206	↔	21	0.1792	0.437	↔
Relief	28	0.5261	***0.004***	↗	21	0.0013	0.9955	↔
2DSum	28	0.0161	0.935	↔	21	-0.0792	0.7328	↔
2DAverage	28	-0.055	0.7831	↔	21	0.0455	0.8449	↔
3DSum	28	0.2565	0.1877	↔	21	-0.061	0.7927	↔
3DAverage	28	0.2929	0.1304	↔	21	0.0455	0.8449	↔

Other Foods. For insects, caterpillars were negatively associated with median feeding durations for angularity (rho = -0.4902, p = 0.0282), 2DSum (rho = -0.5805, p = 0.0073) and 3DSum (rho = -0.5098), and negative trends were present for 2DAverage (rho = -0.5098, p = 0.0217) and 3DAverage (rho = -0.415, p = 0.0688). Interestingly, no mean durations were significantly associated for any dental measure for this food, likely due to high variability in caterpillar feeding lengths. For wood feeding bouts, duration was positively associated with median durations for slope (rho = 0.714, p = 0.0006), angularity (rho = 0.6333, p = 0.0036), Relief (rho = 0.8035, p < 0.0001), 3DSum (rho = 0.5877, p = 0.0081) and 3DAverage (rho = -0.5807, p = 0.0091). No correlations were present for mean durations, likely due to high variability in individual feeding bout lengths for this food item. A trend towards a negative correlation was also present for mean soil feeding length and angularity [(rho = -0.3601, p = 0.055) Table 4.21].

Table 4.21. Other Food Items by Duration and Topographic Status.

	Concrete				Soil				Stem, *S. gracilis*			
By Mean Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	14	-0.1297	0.6586	↔	29	-0.2468	0.1968	↔	9	-0.3833	0.3085	↔
Angularity	14	0.2	0.493	↔	29	-0.3601	*0.055*	↘	9	-0.2	0.6059	↔
Relief	14	0.0857	0.7708	↔	29	-0.2946	0.1208	↔	9	-0.4333	0.244	↔
2DSum	14	-0.0374	0.8991	↔	29	-0.0961	0.6201	↔	9	0.3167	0.4064	↔
2DAverage	14	0.0418	0.8873	↔	29	0.0369	0.8491	↔	9	0.35	0.3558	↔
3DSum	14	-0.1165	0.6917	↔	29	-0.0995	0.6076	↔	9	-0.25	0.5165	↔
3DAverage	14	-0.0901	0.7593	↔	29	-0.0833	0.6677	↔	9	-0.25	0.5165	↔
By Median Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	14	-0.0462	0.8755	↔	29	-0.1429	0.4597	↔	9	-0.1667	0.6682	↔
Angularity	14	0.3011	0.2955	↔	29	-0.2067	0.2821	↔	9	-0.1333	0.7324	↔
Relief	14	0.156	0.5942	↔	29	-0.1379	0.4755	↔	9	-0.2667	0.4879	↔
2DSum	14	0.0462	0.8755	↔	29	0.0525	0.7869	↔	9	0.2167	0.5755	↔
2DAverage	14	0.1077	0.714	↔	29	0.1412	0.4652	↔	9	0.2667	0.4879	↔
3DSum	14	-0.0593	0.8403	↔	29	0.0537	0.782	↔	9	-0.1667	0.6682	↔
3DAverage	14	-0.0198	0.9465	↔	29	0.0468	0.8095	↔	9	-0.1667	0.6682	↔

	Caterpillars				Wood			
By Mean Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	20	0.0887	0.7099	↔	19	0.4053	0.0852	↔
Angularity	20	-0.3173	0.1728	↔	19	0.3877	0.101	↔
Relief	20	-0.0466	0.8453	↔	19	0.507	0.0267	↔
2DSum	20	-0.3699	0.1084	↔	19	0.0807	0.7426	↔
2DAverage	20	-0.2256	0.339	↔	19	-0.107	0.6628	↔
3DSum	20	-0.2331	0.3227	↔	19	0.2895	0.2293	↔
3DAverage	20	-0.1414	0.5522	↔	19	0.2614	0.2797	↔
By Median Duration	**n =**	**rho =**	**p =**		**n =**	**rho =**	**p =**	
Slope	20	-0.197	0.4052	↔	19	0.714	***0.0006***	↗
Angularity	20	-0.4902	***0.0282***	↘	19	0.6333	***0.0036***	↗
Relief	20	-0.3293	0.1562	↔	19	0.8035	***<0.0001***	↗
2DSum	20	-0.5805	***0.0073***	↘	19	0.3737	0.115	↔
2DAverage	20	-0.4105	*0.0722*	↘	19	0.1947	0.4243	↔
3DSum	20	-0.5098	***0.0217***	↘	19	0.5877	***0.0081***	↗
3DAverage	20	-0.415	*0.0688*	↘	19	0.5807	***0.0091***	↗

Part II Discussion.

When examined by food type alone, several significant correlations are present for feeding durations with respect to dental topography. Overall, median durations demonstrate a greater efficacy for assessing connections between behavior and food item type. For example, ten out of twelve significant correlations were for median durations when leaves and flowers were combined into the vegetative category, while ten out of thirteen were present for median durations when flowers and leaves were examined independently. All significant correlations were in the same direction for each food item type where more than one trend or correlation was present, except for mean flower bud and flowers durations (Angularity positive and 2DAverage negative). As such, feeding durations and topographic status are likely linked to the type of food being consumed and their general mechanical and morphological properties (e.g., leaves demonstrating 2D morphologies and displacement limited failure, fruits generally demonstrating 3D morphologies and stress-limited failure; see Lucas, 2004). Such findings are consistent with general observations that specific dental morphologies are more or less appropriate to the consumption of broad dietary categories (e.g., Kay et al., 1979; Kay and Covert, 1981; Evans et al., 2007; Boyer, 2008). For vegetative foods, significant negative correlations were present for mean 3DSum and median relief, while negative trends were present for median slope and mean relief. Thus, individuals with tooth wear and loss generally take longer for each feeding bout for flowers and leaves, and may thus be less capable of processing these food items generally. With respect to the constituent categories making up the vegetative category, significant negative correlations were noted for leaves and leaf buds for median durations and relief only, although a negative trend was also noted for slope, suggesting that these dental features are critical for processing these food types. This is consistent with the concept that these measures, and relief

particularly, are generally indicative of shearing capacity (see M'Kirera and Ungar, 2003; Boyer, 2008; Ungar and Bunn, 2008) and thus may be a good measure of the ability to break down leafy materials. For flower and flower buds, angularity was positively associated with longer mean feeding bout durations although 2DAverage was negatively associated with mean durations. While the 2DAverage data are in the expected direction angularity is not. However, as noted for tamarind fruit, individuals may be more likely to consume these foods for a longer period if they have a better chance of processing them, although this goes against expectations that reduced angularity would be associated with longer food processing times. Similarly, for fruit, the only significant correlation (in a positive direction) was for angularity when median durations were used. Such data are consistent with tamarind fruit observations when measured alone, and may indicate that individuals with low dental wear, particularly those who do not have senescent teeth, are more likely to take time necessary to process fruit-based foods. As tamarind was included in this analysis for fruits overall, it is possible that these patterns are driven by the aforementioned associations between tamarind feeding and this measure.

With respect to minor food types (e.g., wood, soil and insects), there were surprisingly numerous significant correlations between feeding duration and dental topographic status. For wood consumption (which is likely the consumption of termite excreta, although this could not be confirmed as termites themselves or their products were rarely, if ever, observed), only relief was significantly associated with mean feeding durations. However, for median durations slope, relief, angularity, 3DSum and 3DAverage were positively associated with feeding duration times. First, these data suggest that feeding times for this food type are highly variable (as was observed during data recording, with some wood feeding bouts continuing for 10-20 minutes), and are thus more suitable to the use of median as a measure of central tendency when

examining interindividual patterns of consumption. It is also of note that all correlations here were in a positive direction, suggesting that individuals with reduced tooth wear status take longer to complete each feeding bout of this food type. While topographic data presented here reflect only that for the posterior dentition, consumption of wood was typically completed using licking and/or the anterior dentition to gouge soft pieces of wood, and which were usually too large for posterior dental processing. As such, the animals most frequently consumed wood/termite excreta from large semi-rotten stumps and/or branches located on the ground. It is unclear why individuals with greater topographic scores (indicating less tooth wear and/or loss), would spend more time consuming this food for each individual bout. It may be the case that individuals with reduced dental wear are more likely to spend time feeding on this food item for extended periods than are those with higher dental impairment statuses as they are more likely to be able to access other food items, and thus can spend longer periods on this specific food item. Additionally, these individuals may spend more time processing this food as wood is likely to be quite fibrous and extended processing may allow for more effective digestion following ingestion, although this explanation was not tested directly. Nutritional data for wood food items are not available for this study, and the species of wood for each feeding bout was not recorded as decomposition rendered this typically impossible to determine. Thus, it is difficult to state exactly why this pattern is observed within the dataset, although it is likely that the nutritional content of this food item is limited, at least in terms of macronutrient availability. It is possible that individuals with reduced dental impairment may be using wood as a fallback food (in addition to tamarind fruit) in a manner similar to that of bark feeding reported for chimpanzees (Nishida, 1976; Yamakoshi, 1998) or Japanese macaques (Agetsuma and Nakagawa, 1998) during periods of reduced resource (and especially fruit) availability. However, this explanation

must be tempered by the observation that wood was consumed throughout the study, although it was consumed most frequently during the resource-depleted months of July and October. Alternatively, it may be the case that individuals with increased dental impairment are simply choosing to consume other, higher-value food items rather than spending their time consuming wood/termite excreta, which may be difficult to process due to its fibrous nature.

In addition, median insect feeding durations were significantly and negatively correlated with a number of topographic measures (angularity, 3DSum and 2DSum) while negative trends were noted for 3DAverage and 2DAverage. No significant correlations were noted for mean durations. Again, this difference is probably attributable to the highly variable nature of feeding bout lengths for insects (where were represented primarily by caterpillars), although in this case most feeding bouts were of very short length, on the order of a few seconds, with few longer outliers. These data do, however, suggest that dental senescence (as indicated by angularity) and tooth loss (3DSum and 2DSum) are associated with longer feeding bouts for this food type. As with tamarind fruit, insect consumption typically reflected the consumption of only one individual caterpillar, or occasionally one cicada. Therefore, these data indicate that those individuals with dental impairment may lack the same capacity to process and consume this food item type as those without impairment. This is in line with the fact that significant dental wear at BMSR destroys those features which are associated with the breakdown of tough food items, including insect exoskeletons (see Lucas et al., 2004). Most of the insects consumed at BMSR by study subjects were caterpillars which lack a thick chitinous exoskeleton. However, inspection of caterpillars within the feces did indicate that these remained relatively difficult to break apart during fecal sample processing even following digestion, suggesting that these may be a comparatively tough food item in terms of their mechanical properties. Live caterpillars,

however, were not examined as many of these at BMSR cause skin reactions (e.g., inflammation and swelling) when touched (Millette, personal observations).

Food Type by Species with Respect to Dental Topographic Measures.

Vegetative Portions. When data were examined combining all leaf and flower materials, mean values were significant for only four species of the 13 examined. Of these, for *T. indica* and *T. linearis* and *T. dauphinensis* were multiple correlations were present. For tamarind, relief, 2D and 3D sums, and 2D and 3D averages were negatively associated with feeding duration. Likewise, for *T. linearis* ("tamboro"), relief and 3DSum and 3DAverage were negatively associated with mean durations. For *T. dauphinensis* 2DSum and 2DAverage were negatively associated with mean feeding duration. Somewhat similar patterns were also noted for median durations for *T. indica* and *T. linearis,* although significant correlations were only present for relief and 3DAverage, and for 3DSum and 3DAverage, respectively. Likewise, for median durations *Pentopetio* vegetation were negatively associated with for relief. For both mean and median values, a number of trends were present for a variety of measures across a number of other species, all in the negative direction. The only significant correlation in the positive direction was for angularity for *G. americanus,* likely for flowers, which were the primary food consumed from this species.

Overall, these data indicate that vegetative materials from only a few species are associated significantly with feeding bout lengths. These were most frequently associated for only two species, *T. indica* and *T. linearis,* suggesting that species differences in mechanical properties for both flower and leaves may be critical when discussing the impacts of dental impairment. While, *T. linearis was* consumed by a minority of individuals, *T. indica* vegetative

material was consumed by 30 of 31 individuals. Thus, it is apparent that along with tamarind fruit, other materials from this species are also commonly consumed by lemurs at BMSR. Likewise, as durations are longer for individuals with dental impairment, impairment may be negatively impacting their capacity to access this food species, the leaves of which are commonly used immediately prior to, or during the transition to, the wet season (e.g., during October, see Chapter 3). It is also of note that all trends and correlations present, except for one, are in the negative direction, suggesting that even for less common food items, dental impairment may result in a reduced capacity to consume vegetative food items overall, although additional data is necessary to confirm these findings as the number of individuals who consumed each species tended to be restricted during this study.

Flowers and Flower Buds. Flower buds and flowers were only consumed by greater than 10 individuals for four species. Of these, significant correlations were only present for two species, *I. majungensis* (mean durations and relief) and *G. americanus* (median durations for slope). Negative trends were also present for slope, 3DSum and 3DAverage for mean durations for *I. majungensis*, and for 3DAverage and 3DSum for *Quisivianthe papionae*. Although few significant correlations or trends are present, *all* are in the same direction, indicating that flowers of these species are harder to consume by those with dental impairment. However, given the limited data available from this study, it is difficult to assess if dental impairment impacts the capacity of individuals to process this food type on a species basis or overall. Additionally, for the species of *I. majungensis*, both flowers and new fruit were often simultaneously present, which may complicate the interpretation of these data. Thus the data presented here should be

examined with caution, although they do indicate that flower feeding for this species may be impacted by dental impairment status.

Leaves and Leaf Buds. With respect to leaves, negative correlations were present for tamarind and *T. linearis* for multiple measures for mean durations (relief, 3DSum, 3DAverage for both, and 2DAverage for *T. indica*). For median durations, negative correlations were likewise present for slope and relief and a negative trend for 3DAveragefor *T. indica*. For *T. linearis* negative correlations were present for 3DSum and 3DAverage, with a negative trend for relief. Significant negative durations were also present for median durations for relief for *Pentopetio* and mean durations for *T. dauphinensis* with respect to 2DSum and 2DAverage. These data are consistent with those observed when leaves and flowers were combined into one "vegetative" category, suggesting that those values observed reflect differences primarily in leaf and leaf bud feeding. Likewise, all trends present for these species are in a negative direction. For these species, it is thus likely that leaf feeding is impacted by dental impairment status. As noted before, this is consistent with patterns of tooth wear at this site which generally remove shearing features associated with the processing of leaf materials and/or remove positions from the postcanine dentition with the mastication and breakdown of leaves, particularly larger leaves (see Yamashita, 2003; Sauther et al., 2004; Cuozzo and Sauther, 2006a,b; Millette, personal observations).

However, positive correlations are present for *S. angustifolia* mean durations for angularity, 2DSum, 2DAverage, 3DSum and 3DAverage, and for median durations for angularity, 2DSum and 3DSum. Similarly a significant positive correlation was present for mean feeding durations and slope for *Commicarpus* sp., while a trend towards longer mean durations

was present for *M. parvifolia* with respect to angularity. These data are in the opposite direction than expected. For *S. angustifolia* this may relate to this plant's unique leaf, which is shaped similarly to that of a "green bean" rather than like a standard flat leaf. Thus, differences here may reflect this difference in morphology. Also, it was often difficult to distinguish when animals were consuming *S. angustifolia* leaves or when they were consuming flowers, as both were often consumed rapidly during the same bout. These data may therefore reflect the consumption of flowers for this species rather than leaves alone. This, however, would not explain why those individuals with tooth wear or impairment would spend less time consuming this food species on a per bout basis. As such, these patterns go against those found for all other species where a significant correlation was noted, and are therefore difficult to explain. These data do suggest that patterns of dental impairment interact with feeding bout lengths differently on a species-level basis. It is possible that the physical properties of these plants may be better suited to the tooth form of those individuals with higher topographic scores, and these individuals may spend more time processing this food as those with increased impairment simply cannot adequately process these species into a form suitable for consumption, but this remains open to future investigation (see Chapter 8).

Fruit Feeding. For fruit feeding, five species other than tamarind were consumed by >10 individuals (*Bridelia* sp., *C. excelsa*, *G. leucophylla*, *S. angustifolia* and *T. pruinosum*). In each case no significant correlations were present except for positive correlations for slope and relief for mean durations for *S. angustifolia.* These data indicate that dental impairment status is not associated with feeding durations for most fruits, or at least it does not lead to increased feeding durations. These data are also roughly consistent with those for tamarind fruit feeding when

examined using nonparametric methods, and when fruit was examined overall as a food type class. It appears that fruit feeding duration is not impacted by dental impairment in the manner expected. That said, all fruits here except for *Crateva excelsa* are of small size and are consumed relatively quickly, and multiple fruits are often consumed in a single bout. Although it is not possible to count each individual fruit consumed, when combined with shorter feeding bout lengths overall, it is possible that individuals with dental impairment are consuming fewer individual fruits during each feeding bout. Data necessary to confirm this hypothesis (e.g., bit count and rate data) remain to be collected, and this question must be addressed in future studies. That individuals with dental impairment are likely consuming fewer food items, however, does not explain why significantly longer bouts are found for *S. angustifolia* for those with reduced dental impairment. Such patterns, however, may reflect this fruit's morphology. *Salvadora* fruit demonstrates a thin fleshy covering over a relatively large seed. If individuals are attempting to gain energy from this fruit by breaking into and consuming the seed, it may stand that those with less impairment are spending more time during each feeding bout attempting to do so in a manner similar to that posed above for tamarind feeding. However, informal examinations of fecal material indicate that lemurs do not typically breakdown these seeds, but rather they pass through the digestive tract unaltered. Thus, this explanation may not fit the data very well, and the source of the longer time of *Salvadora* consumption for those with higher slope and relief remains somewhat unclear.

Part II Conclusions.

Although patterns of food processing duration are highly variable across food types and food species in terms of their association with each specific measure and form of bout

measurement (e.g., mean vs. median durations), it is evident that a number of significant correlations do emerge from the data available. As such, for major food item types (e.g., vegetative materials, flowers, leaves, insects) all correlations between duration and topography are in the negative direction (except for angularity for mean flower feeding durations), suggesting that feeding bout length generally increases as dental impairment also increases. Similarly, the vast majority of non-significant trends are also in this direction, indicating that dental impairment is associated with increased feeding bout lengths. While most foods were consumed in a manner where multiple items were consumed in a single bout, these data do provide support that individuals with higher amounts of dental impairment are less effective at processing food items than those with reduced amounts of dental impairment. The exception to this pattern appears to be for fruit when examining scores for angularity. As noted before, this result likely reflects primarily the inclusion of tamarind in this analysis, as angularity was associated with longer feeding times for this food, which was the most commonly consumed fruit during this study Nevertheless, these data do suggest that increased angularity may be associated with longer attempts by individuals to access fruit-based food items similar to what was found for flowers and angularity. Wood/termite excreta consumption was also associated with increased bout lengths for individuals with reduced measures of dental impairment. However, here, the source of this difference in the expected direction of the data remains unclear, although it does appear that those with reduced impairment are likely to spend more time processing and consuming this food at a given time. Given that such variances in the direction of food processing times emerge by food item type, it is apparent that dental impairment does not simply result in longer feeding bout lengths overall, but rather that the

length of feeding bouts among food types may vary differently with respect to dental impairment status.

It is also notable that when examined on a species specific level, only a few taxa demonstrate multiple associations with measures of dental topography. For example, for leaves and leaf buds (or vegetative materials), durations were primarily associated with topography for the species of *T. indica* and *T. linearis* in the negative direction and *S. angustifolia* in a positive direction, but were of limited association with dental topography for other food item taxa. While these findings may reflect reduced sample size for some food species, they may also indicate that there are differences between species with regard to how dental wear and/or tooth loss interact with feeding duration. It may be more difficult for individuals with increasing dental wear to process some species, while others remain easier to consume despite dental impairment. It is likely that the morphology and physical properties of these taxa result in such divergences, particularly for the longer feeding bout durations observed for those with higher dental topographic measures for *S. angustifolia*, which demonstrates atypical leaves and fruit with challenging seeds. Thus, it is apparent that dental impairment may interact with feeding and food processing ability differently between species, and that overall patterns observed by food item type reflect primarily the impacts of those key species where impairment results in a clear difference in their patterns of consumption. Nevertheless, as most correlations between duration and topography are in the negative direction, or trend in that direction, dental impairment is generally associated with increased feeding times for most species (particularly with regards to vegetative and leaf feeding), indicating that dental impairment is likely associated with a reduced capacity to process these foods.

When examining combined food item categories, median durations appear to have more efficacy for discerning patterns of feeding bout length to dental topography than do mean durations. The measure of central tendency used, however, appears to be less critical when examining the data on a species-specific level, although there are some instances where either the mean or median values appear to be better associated with topography for some foods (e.g., caterpillars). Variability in mean feeding bout lengths between general food types may muddle the data when examined using mean data, as certain species may take much longer or shorter to consume per bout than others on average. For examinations of general food item types, from the data presented here it appears that use of the median as a measure of central tendency is more appropriate for studies examining the relationship of dental topography to feeding bout length than is the mean.

Finally, a range of variability in which topographic measures were significantly associated with feeding durations was apparent across species, depending on if mean or median values were utilized (e.g., if one measure was significant when mean was used, but not when median was used, or vice versa). Different measures may be more responsive to use of mean or median measures of feeding bout length when examining the data on a species-specific level. Although the source of such variation is difficult to determine, certain measures of topography are likely more responsive to within-species variability in mechanical properties, which could lead to divergent patterns of feeding bout length for the same food items a given species. Such variations could cause outliers in feeding bout length, which could lead to divergent patterns between mean and median durations seen here. Such data, however, remains to be collected and tested using a dental topographic analysis framework.

CHAPTER V:

FECAL NUTRITIONAL ANALYSIS IN RELATION TO DENTAL WEAR AND TOOTH LOSS STATUS.

Introduction.

In this chapter I will examine the nutritional content of feces collected from a sample of ring-tailed lemurs observed during this study period (June 2012 – March 2014). In particular I focus on how dental impairment, in the form of tooth loss and tooth wear, impacts the individual's capacity to digest and subsequently uptake food items relative to those without dental impairment. Data presented here are based upon nutritional analyses conducted for 154 fecal samples collected at BMSR during the dry season (June-July) 2012 and wet season (January) 2013 for 14 adult individuals of three social groups. Analyses were conducted for the following elements: dietary fiber (ADF and NDF), lignin content (ADL), protein (measured by fecal nitrogen content), and fecal ash. Ratios of fecal ADF and NDF to lignin (ADL), as well as ratios of hemicellulose and cellulose to ADL, were also calculated from the collected data. Each variable examined provides a measure of digestive and fermentative capacity as related to tooth loss and dental wear status (see below).

The study discussed in this chapter is based on the concept that individuals with tooth wear and/or tooth loss will be less able to reduce foods to small particles than are individuals without dental impairment. Dentally-impaired individuals will, on average, ingest comparatively larger food particles following food processing and mastication (see Gipps and Sanson, 1986;

Lentle et al., 2003; Millette et al., 2012; Ellis et al., 2015). The consumption of such larger food particles reduces the surface to volume ratio on which gut microbes and enzymes may act in comparison to smaller-sized particles. As a result, if passage rates and fermentation times remain unaltered, larger particle sizes will subsequently result in a reduced capacity to digest and absorb ingested foods (see Bjorndal et al., 1990; Gipps and Sanson, 1986; Lentle et al., 2003, etc.). This study examines the outcome of such a dental impairment-related failure to breakdown food items by analyzing nutrient content in fecal material. It is posited, and has been observed (see below; Chapter 6), that individuals with dental impairment demonstrate a reduced ability to extract nutrients from ingested food items, resulting in greater amounts of nutrients remaining within their feces. In this chapter, I examine fecal nutritional measures as a potential means for assessing how tooth loss and wear alters the ability of animals to process, ferment and extract nutrients from dietary fiber (measured in the form of ADF and NDF), as well as their capacity to absorb protein. I also examine the acid detergent lignin (ADL) and ash (e.g., insoluble inorganic material, such as silicates) content remaining within the sample. I furthermore discuss ratios of NDF, ADF, hemicellulose and cellulose to ADL, as these measures may serve as a means for assessing the effects of tooth loss and wear on the individual's ability to ferment the hemicellulose and cellulose fractions of dietary fiber (see Fahey and Jung, 1983; Van Soest, 1984; Rothman et al., 2012).

Digestion and Fiber Fermentation in Ring-tailed Lemurs. Ring-tailed lemurs are characterized as "opportunistic omnivores," regularly consuming a wide variety of food items, both in terms of food item type (e.g., leaves, fruit, flowers, and feces) and food item species (Sauther et al., 1999). Specific foods consumed by BMSR ring-tailed lemurs varies not only seasonally, but also

on a month-to-month basis related to food item availability. Likewise, foods eaten vary based upon their location, as a gradient from xeric (e.g., dry, spiny forest) to deciduous gallery forest is present within the reserve. Food selection may also be influenced by individual group home ranges and whether they include protected areas or surrounding non-protected areas that are more affected by human disturbance (Sauther, 1998; Sauther et al., 1999; Ratsirarson et al., 2001; Yamashita, 2002; Simmen et al., 2006; Fish et al., 2007; Sauther and Cuozzo, 2009; Whitelaw, 2010; Cuozzo et al., 2014; Yamashita et al., 2015a,b).

During the resource-depleted dry season, BMSR *Lemur catta* from areas around Parcel 1 frequently fall back on the mechanically-challenging tamarind (*Tamarindus indica*) fruit.(Sauther, 1998; Cuozzo and Sauther 2006; Simmen et al., 2006; Sauther and Cuozzo, 2009; Yamashita et al., 2012, 2015). This does not, however, mean that other foods are not consumed during the dry season (see Sauther, 1998; Simmen et al., 2006; Yamashita et al., 2015b). During this study, in the months of the dry season (June and July) for which fecal matter was examined (feeding data are only available for July, although feeding patterns during late June should be similar to that of July; Millette personal observations) a number of foods were consumed. For example, in July, while tamarind fruit represented the single largest proportion of foods consumed (31.81% total feeding), foods including *Tarenna pruinosum* fruit (9.54% feeding) and vegetation and flowers from species including *Salvadora angustifolia* (Primarily flowers with some young leaves eaten simultaneously: 23.46%) and *Metaporana parvifolia* (Leaves: 10.74%) were also consumed. While leaves and flowers are less available during the dry season period examined during this study (Sauther, 1998), these can and do account for a major portion of time spent feeding during the period examined (45.33%). In turn, fruit, primarily of *T. indica* and *T. pruinosum*, accounted for 43.54% of July feeding.

During the wet season, the availability of leaves, and particularly those preferred by *L. catta* (e.g., young leaves and those of lianas), increases at BMSR (Sauther, 1998; Rasamimanana et al., 2012; Millette, personal observations). For the wet season month examined in this chapter (January), this is clearly reflected in feeding patterns demonstrated by the animals, with vegetative material (e.g., leaves, stems and flowers) accounting for 64.52% of feeding, the highest of any month during this study. *M. parvifolia* leaves (with a few stems) accounted for 35.75% of all observed feeding during January, with vegetation of other many other species (e.g., *Pentopetio* sp.: 4.84%; *Secamone* sp.: 2.96%; unknown: 9.14%) accounting for remainder of vegetative feeding. In contrast, during January all fruit consumed accounted for 30.91% of total feeding, which is less feeding time than that observed for *M. parvifolia* leaves alone. Tamarind made up a high proportion of fruits consumed, accounting for 25% of total feeding, with voamanga melon and *Crateva excelsa* accounting for all other fruits consumed during this month. Chapter 3 provides a full description of all feeding behaviors and food items consumed.

Although often considered an opportunistic omnivore, leaf and/or flower-based material frequently account for a significant proportion of ring-tailed lemurs' diet on a seasonal basis (Sauther, 1998; Sauther et al., 1999; Millette, this study). Being able to access resources from leaves is likely to be an important aspect of the ring-tailed lemur's adaptive profile, and these animals should demonstrate morphological, physiological and/or behavioral adaptations towards the consumption of such materials. Dentally, ring-tailed lemurs exhibit higher shearing capacity than that found among primarily frugivorous lemur species. Although they do not demonstrate shearing capacity akin to folivorous taxa (e.g., *Propithecus* sp.), ring-tailed lemurs do possess molars with relatively elongated shearing crests suitable to breaking down tough food items such as leaves, as well as thin enamel which is likely suited the production of auxiliary shearing crests

appropriate for the consumption of leaves and patterns of wear resulting from a folivorous diet (Rensberger, 1973; Kay et al., 1978; Janis and Fortelius, 1988; Yamashita, 1998a,b, 2008b; King et al., 2005). The finding ring-tailed lemurs at BMSR demonstrate high amounts of tooth wear and/or loss likely reflects an interaction of a semi-folivorous tooth form and the diet available during the dry season, which emphasizes morphologically (e.g., large size) and mechanically-challenging tamarind fruit, and which also appears to be ill-suited to being broken down with a tooth that is both cusped with thin enamel and demonstrates elongated shearing crests (Kay et al., 1978; Dumont, 1995; Yamashita, 1998b, 2008b; Cuozzo and Sauther, 2004, 2006a,b; Lambert et al., 2004; Lucas, 2004; Godfrey et al., 2005; Campbell et al., 2012). Such observations may also provide evidence that *L. catta* demonstrate dental morphology suited to vegetative material rather than hard food items such as *T. indica* fruit. Significant tooth wear and loss resulting in dental impairment has also been observed to result in an inability to comminute leaves into small fragments, and individuals with significant tooth wear and/or loss have been observed to ingest and pass leaves which are of several centimeters square in area (Millette et al., 2012 / Chapter 6; Millette, personal observations), indicating that dental impairment may significantly impact the ability to consume foliage-based food sources to which *L. catta* is otherwise adapted.

Dietary fiber consists of nonstartch polysaccharides of both soluble and insoluble forms (i.e., soluble and insoluble fiber). Soluble fiber includes elements such as pectins, which are readily fermentable. Insoluble fiber is represented by the structural polysaccharides hemicellulose, cellulose and lignin (Lambert, 2007a,b). Although no mammal is known to possess enzymes capable of breaking down cellulose, this dietary element represents a large proportion of energy potentially available within plant foods (Blaxter, 1962; Alexander, 1993; Lambert, 2007b). Energy found within cellulose may however be harnessed through

fermentation by protozoa, fungi or bacterial gut microbiota. Similarly, hemicellulose also requires bacterial fermentation in order to be utilized by the animal, although it appears that this structural polysaccharide may be partially broken down within a low pH stomach (Milton and Demment 1987, National Research Council 2003 in Lambert, 2007b; Lambert, 2007b). In contrast to cellulose or hemicellulose, lignin cannot be broken down by bacterial fermentation and no mammal maintains enzymes which break down lignin. Lignin is thus totally unavailable to consumers. Insoluble fiber is commonly measured using the measures of NDF and ADF, with NDF representing cellulose, hemicellulose and lignin content, while ADF represents cellulose and lignin content (Lambert, 2007b; Saha et al., 2013).

Studies of ring-tailed lemur gut morphology and cellulose digestion indicate this species possesses an ability to effectively digest, ferment and extract energy and nutrients from leaf-based or foods otherwise heavy in cell wall components (e.g., flower petals, stems, etc.). Although mammalian taxa do not possess enzymes with which to breakdown insoluble fiber found in the cell walls of plants, animals may derive significant energy from such foods through utilizing byproducts produced by the fermentation of gut microbes. Microbial action can hydrolyze the structural carbohydrates (e.g., cellulose and hemicellulose) from which dietary fiber is constructed into short-chained fatty acids that are subsequently absorbed and utilized by the host animal (Van Soest, 1994; Lambert, 1998, 2007; Edwards and Ullrey, 1999a,b; Campbell et al., 2004; Lambert and Fellner, 2012). Among primates (and non-primate mammals) the consumption of diets high in foliage is frequently associated with specialized gut morphologies adapted to maintaining gut microbes necessary to break down structural carbohydrates found in the cell walls of leaf material. This is most dramatically seen among members of the Colobinae, which demonstrate foregut adaptations consisting of a modified “sacculated stomach” similar to

that seen among ungulates, and which permits the consumption of diets high in structural carbohydrates, but is also susceptible to changes in pH that may result from consumption of acidic foods (Stevens and Hume, 1995; Lambert, 1998; Edwards and Ullrey, 1999b). Among non-colobine species, diets high in structural carbohydrates are frequently associated with adaptations towards post-stomach fermentation of ingested foods. Such "caeco-colic" (e.g., hindgut) fermenters often utilize an enlarged caecum and/or colon to maintain bacteria necessary for fermentation (Lambert, 1998; Edwards and Ullrey, 1999b; Campbell et al., 2000, 2004; Campbell, 2003). These patterns of gut morphology have been reported for a number of folivorous strepsirrhine taxa. For example, during necropsies conducted on *Propithecus tattersalli* and *Propithecus coquereli*, Campbell (2000) found these sifaka species demonstrated elongated intestines with large surface areas, as well as a lengthy, spiraled colon and an extensively sacculated caecum, both of which were highly vascularized. This was in contrast to the fruit feeding *Varecia variegata* which showed limited sacculations or teniae for either the cecum or colon, and which has also been shown to demonstrate a comparatively reduced ability to ferment fiber (Edwards and Ullrey, 1999a; Campbell et al., 2004). Campbell found that the digestive tract of *L. catta* showed a morphology intermediate between *Propithecus* and *Varecia* but consistent with the breakdown of structural carbohydrates. As Campbell states: "The caecum of *L. catta* was large and haustrated and therefore suitable for microbial breakdown of plant cell wall, allowing for seasonal dependence on a predominantly leaf diet" (Campbell et al, 2000: 29). Additionally, experimental studies conducted by Sheine (1979) indicate that ring-tailed lemurs maintain a capacity to ferment and utilize significant amounts of dietary cellulose when presented in the form of processed particles. Such data provide support that ring-tailed lemur gut physiology can maintain sufficient bacteria for the fermentation of structural carbohydrates such

as those found in leaves. These data are also consistent with Campbell's observations, as well as from this and other (e.g., Sauther, 1992, 1998) field studies that ring-tailed lemurs can process, utilize and survive on a diet seasonally high in leaves and their cell wall contents.

It is likely that significant dental impairment such as that observed at BMSR can reduce the individual's ability to breakdown folivorous material into a state where effective bacterial fermentation is possible, resulting in the reduced production and uptake of short-chained fatty acids (SFCA). While *L. catta* demonstrate a dental morphology capable of effectively milling leaves into small particles suitable for fermentation in an unworn or low-wear state, significant dental wear and/or resulting tooth loss can result in the loss of shearing capability, reducing the individual's ability to reduce leaf material into small particles. Such a capacity is critical for increasing the relative surface to volume area of ingested food items, and provides gut bacteria a viable substrate on which to act (Van Soest and McQueen, 1973; Stevens, 1988; Bjorndal et al., 1990; Mackie, 2002; Lentle et al., 2003; Ellis et al., 2015). Although I know of no studies directly linking food particle size to short chained fatty acid production among nonhuman primates, smaller-sized bran particles have been observed to enhance SFCA production relative to larger particles when fermented using human-derived innocula *in vitro* (see Stewart and Slavin, 2009). Data previously collected from BMSR *L. catta* indicate that individuals with tooth wear and/or significant tooth loss do not process food items as effectively as do those without dental impairment, leading to the consumption of fewer particles of small size. It is also of note that such individuals also appear to have greater amounts of leaf material within their feces in comparison to those without dental impairment (Millette et al., 2012 / Chapter 6). Such individuals may thus be particularly impacted in their ability to utilize ingested foods. Likewise, experimental studies by Sheine (1979) found that when fed soy-hull sourced cellulose particles

of larger size, ring-tailed lemurs demonstrated a reduced ability to ferment and uptake their experimental diet in comparison to when fed particles of smaller size.

With regard to fecal fiber content, it is probable that individuals with tooth loss or extensive dental wear will pass greater amounts of fiber, measured in the form of ADF or NDF, through their gut than those without dental impairment. I posit that this will hold true across seasons, although it is likely that such an impact will be greatest during the wet season when leaves form a higher proportion of the diet. Yet, as vegetation forms a large portion of the diet during the dry season in the form of *M. parvifolia* herbs and *S. angustifolia* flowers (which resemble leaves), such a pattern may also be apparent during the months of June and July.

Fecal Lignin Content. Animals may consume divergent diets based upon their tooth wear status, resulting in differences in the total amount of fiber consumption by an individual, and which may impact the proportion of fiber present within the feces relative to other dietary elements. For example, if animals with tooth loss consume greater amounts of leaves, as is indicated by previous examinations of fecal material generated at BMSR, such animals may demonstrate greater fecal ADF and NDF content overall. Examination of lignin content relative to measures of total fiber content (e.g., ADF and NDF) as well as the ratio of lignin to the fermentable structural carbohydrates of cellulose and hemicellulose may provide a better means to account for such differences in fiber intake.

In contrast to the hemicellulose or cellulose fraction of ADF or NDF, lignin resists fermentation by gut bacteria, and no mammalian species maintains gut enzymes capable of breaking down this compound (Van Soest, 1994). Given its non-digestible nature in comparison to other cell wall components, lignin may serve as an internal measure with which to gauge the

digestion of other components of dietary fiber if present in sufficient quantities (e.g., >5%) While hemicellulose and cellulose fractions are fermented by gut bacteria and subsequently utilized by the animal, lignin should pass through relatively undigested (Fahey and Jung, 1983; Van Soest, 1994; Rothman et al., 2012). As a result, the ratio of total fiber content (either ADF or NDF), or the ratio of fermentable structural carbohydrates (e.g., cellulose and hemicellulose) to lignin can provide information on the animal's ability to break down these food items. Likewise, as lignin will not be digested by the animal, particle size should not impact the passage of lignin through the digestive tract. Thus, an inability to fragment foods should not result in a change to the amount of lignin entering and subsequently exiting the digestive system.

It should be noted; however, that lignin does represent an "anti-quality" element within food items, and is associated with reduced overall forage digestibility (Moore and Jung, 2001). By physically limiting the effects of enzymatic and bacterial action within the gut, foods with higher amounts of lignin will limit the ability of an animal (regardless of tooth loss status) to ferment the structural carbohydrates within ingested food items, thus reducing the total amount of dietary energy available (Moore and Hatfield, 1994; Jung and Allen, 1995; Moore and Jung, 2001). Lignin also reduces the overall amount of digestible dry matter that may be consumed by an animal, limiting the animal's ability to gain nutrition from forage (Moore et al., 1993 in Moore and Jung, 2001; Mertens, 1994). Animals may thus select foods which are low in lignin content to avoid such issues, although this may complicate the use of lignin as an internal standard if animals divergently select foods with less lignin based on tooth loss or wear status. Several studies have indicated that some mammalian taxa (e.g., pandas) maintain bacteria capable of oxidizing lignin and/or have demonstrated the loss of lignin during transit through the digestive tract (as has been observed for several Colobine primate species), suggesting that lignin

can be degraded to some extent (Dierenfeld et al., 1992; Nijboer, 2006; Feng et al., 2012). The amount of such loss during transit, however, appears to be relatively low in comparison to cell wall structural carbohydrates (i.e., hemicellulose and cellulose). While these limitations must be noted for the use of lignin as an internal marker, for this study, it was not possible to assess overall digestibility of food items as these data are extremely difficult to collect given the extent of this study (e.g., food samples must be collected and analyzed, and fecal contents from specific individuals must be matched with those derived from observed feeding, etc. (see conclusion). Despite these limitations, for this study, lignin offers one of the few means available for assessing the impact of dental impairment on digestive capacity, and is one of the few means for controlling for potentially divergent patterns of fiber excretion resulting from food selection.

Fecal Ash Content. In addition to measures of fecal fiber content, the variables of fecal protein (e.g., fecal nitrogen) content and fecal ash were also examined. Ash represents the non-organic fraction of a food item, or in the case of this study, of fecal matter resulting from the consumption of food items. Fecal ash content is determined by heating the sample to high temperatures (e.g., @500^{0}C) for a period of several hours in order to burn away all organic material, thus leaving the sample's mineral fraction (Saha et al., 2013). As they are not burned away during heating, dietary and fecal ash content can provide a measure of dietary silicates. The ingestion of dietary silicates has been implicated in the generation of tooth wear and/or evolution of morphological adaptations (e.g., hypsodonty, thick enamel, etc.) believed to counter the effects of tooth wear upon masticatory capacity. Fecal ash content can provide a general measure of endogenous silica (e.g., phytoliths), exogenous grit (e.g., sand, dust, etc.) adhering to foods consumed by the animal, and/or other silicates consumed by the animal [e.g., due to geophagy

(Baker et al., 1959, 1961; Healy and Ludwig, 1965; Ludwig et al., 1966; Beyer et al., 1994; Hummel et al., 2010, Rabenold and Pearson, 2011; Madden, 2014)].

Dietary silicates have been implicated in the generation of tooth wear across a wide range of mammalian taxa through both studies carried out *in vitro* and *in vivo*, although the source of such wear (e.g., phytoliths vs. grit) remains under debate (Baker et al., 1959; Ludwig et al., 1966; Walker et al., 1978; Covert and Kay, 1981; Kay and Covert, 1983; Danielson and Reinhart, 1998; Ciochon et al., 1990; Ungar et al., 1995; Clauss et al., 2007; Kaiser et al., 2008; Rabenold and Pearson, 2011, 2014; Sanson et al., 2007; Lucas et al., 2013, 2014; Ungar, 2015). Work by Sanson et al., (2007) and Lucas et al. (2013, 2014) suggests that phytoliths may not be mechanically hard enough to remove dental enamel; however, the generation of tooth wear and/or adaptations against tooth wear have been linked to phytoliths in a number of mammalian taxa (for example see: Baker et al., 1959: domestic sheep; Walker et al., 1978: hyraxes; Ciochon et al., 1990: *Gigantopithecus blacki*; Danielson and Reinhart, 1998, 2005: modern humans; Rabenold and Pearson, 2011: multiple primate taxa). Although debate exists about the wear generating qualities of phytoliths, exogenous silica is known to be harder than enamel (Lucas et al., 2013, 2014) and has been implicated as a of cause dental wear among mammalian species (e.g., Ludwig et al., 1966; Mainland, 2003: sheep; Covert and Kay, 1981; Kay and Covert, 1983: *Didelphis* opossums fed pumice; Jardine et al., 2012: ungulates and Glires; Lucas et al., 2013). Discerning the source of wear between phytoliths and grit is likely difficult as both may result in similar patterns of wear, although silica-based wear sources may be distinguished using detailed microscopy (Sanson et al., 2007; Lucas et al., 2013, 2014). For the purposes of this study, the source of wear is of less importance as both grit and phytoliths would be included in any measure of fecal ash content. Given that either endogenous or exogenous silicates likely

contribute to the wear of mammalian teeth, for this study total fecal ash content is examined in relation to tooth wear and/tooth loss status.

A number of researchers have demonstrated the utility of fecal measures of silica and/or fecal ash when assessing the inclusion of inorganic matter (e.g.., phytoliths or soil) within the diet (e.g., Beyer et al., 1994). Similarly, patterns of tooth wear (and adaptations against tooth wear) for a number of mammalian taxa have been examined using fecal methods. For example, using fecal acid detergent insoluble ash (ADIA) as an indicator of dietary silicates, Hummel et al. (2011) found that among large African herbivores, hypsodonty was correlated with higher fecal ash content. These authors suggested that increased abrasiveness and resulting tooth wear due to dietary silicates is likely indicative of a selective pressure (either from phytoliths or grit) in the evolution of wear-resistant dentitions. Similarly, fecal silica content has been related directly to tooth wear among sheep populations. Ludwig et al. (1966) found that incisal wear was closely associated with the amount of soil ingested by grazing New Zealand sheep, and reported that rates of wear were highest during portions of the year when fecal soil content [as measured by Acid Insoluble Residue (AIR)] was greatest. In turn, wear rates were lower when reduced amounts of soil were ingested, suggesting that fecal measures can detect seasonal variations in silica ingestion and/or tooth wear. Among wild Canadian bighorn sheep, Skipworth (1974) found seasonally variable levels of fecal AIR, with silica content reaching similar levels as those among New Zealand captive animals. Although Skipworth did not measure tooth wear in this study, he did indicate that the consumption of grit (likely due to use of soils for mineral content) in the levels observed could lead to tooth wear with physiologically-significant ramifications. Mainland (2003) found patterns of increased dental wear striations were related to higher amounts of grit-related dietary silica in the feces of grazing sheep than in those which consumed

comparatively browse-based diet. Fecal contents in this case indicated that grit, rather than phytoliths, were the primary source of such wear in this population. In contrast, among humans, Danielson and Reinhard (1998, see also Reinhard and Danielson, 2005) found high calcium oxalate phytolith content in coprolites was associated with extensive tooth wear (both in terms of microwear and pathological gross tooth wear) among a skeletal archaeological population of hunter-gatherers from the Lower Pecos region of Texas. In this case, silica from other sources (e.g., grinding stones) did not appear to be a major source of tooth wear, leading the authors to conclude that phytoliths (possibly from chewing agave quids) were the primary source of dental wear observed in this population. Unfortunately, fecal ash or silica data appear to be exceptionally limited for primates with respect to tooth wear. I have not found any researchers or documentation regarding the relationship of dental wear with fecally-measured dietary silica for any primate species. As such, results from this study maintain a potential to enhance knowledge of how exogenous grit may impact tooth wear in nonhuman primates, as well as improve means for measuring the impact of dietary silicates on tooth wear in nonhuman primates.

Fecal Protein Content. Fecal nitrogen was used as a measure for the excretion of protein within the feces of study animals. Dietary protein is a key macronutrient required by primates, and is utilized as a substrate for energy as well as for growth and somatic maintenance. Dietary protein also provides animals with amino acids necessary for the production of endogenous proteins, most critically essential amino acids, which cannot be manufactured by the animal itself (Oftedal, 1991; Leonard, 2000; Lambert, 2007b). Protein may be sourced from a variety food types, but is particularly concentrated within leaves, insects and animal matter. Fruit material, in contrast, does not typically provide high amounts of protein, and consumption of fruit alone is

likely insufficient to account for all of the animal's protein requirements (Whitten et al., 1991; Leighton, 1993 both in Lambert et al., 2007b). Animal or insect material represented an exceptionally small portion of the diet during the months examined by this chapter, and insects or animal-based foods were not consumed during any interval-based feeding observation during the study months of July or January. It is unlikely that these sources provided a major source of protein for BMSR lemurs during this study, although insects were consumed during other months, as were eggs from a bird's nest. Leaves did, however, form a considerable portion of the diet during the dry season, and were consumed at even higher rates during the wet season. While no nutritional data are available from food items directly, it is thus likely that leaves formed the backbone of protein intake during this study.

Individuals with dental impairment are predicted to be particularly challenged by leaves, as dental wear observed at BMSR frequently removes features of the tooth (e.g., shearing crests) associated with folivorous diets and the comminution of leaves (see above). Likewise, individuals from this population with extensive tooth loss and tooth wear have been observed to be less efficient at breaking down leaves and other vegetative materials. Such leaves often pass through the digestive tract in large, semi-complete particles (e.g., several cm^2) and which are clearly identifiable as leaves following excretion (Millette et al., 2012 / Chapter 6; Millette, personal observations). Therefore, it is probable that individuals with tooth wear and loss are unable to obtain protein resources from leaf materials as efficiently as do those without dental impairment, as leaf cell walls must be broken down through mastication to allow access to proteins held within (Lambert, 1998).

Fecal nitrogen is frequently used as a measure of dietary (e.g., forage) quality among ungulate taxa, particularly for grazing forms. Fecal nitrogen is believed to reflect the amount of

crude protein available within foods consumed by the animal (e.g., Holechek et al., 1982; Leslie and Starkey, 1985; Codron et al., 2006; Leslie et al., 2007). This relationship between fecal nitrogen content and dietary protein is, nonetheless, complicated by a number of factors. Fecal nitrogen may be impacted by the incorporation of secondary compounds such as dietary tannins which can bind to ingested proteins and/or the ingestion of lignins that may also interfere with digestion. Such dietary factors may reduce protein uptake by the animal, and result in higher amounts of fecal nitrogen than expected based upon diet alone (Robbins et al., 1987; Meissner et al., 1999 in Codron et al., 2006; Verheyden et al., 2011). Additionally, while fecal nitrogen reflects protein that passes through the digestive tract along with undigested portions of the diet, fecal nitrogen is also indicative of metabolic nitrogen, which in large herbivores, may represent the majority of nitrogen content within the feces. Metabolic nitrogen may result from the physiological processes of the animal itself or from the action (or excretion) of gut bacteria. Gut microbiota are often cited as the primary source of fecal nitrogen, and differences in forage quality detected by fecal nitrogen are often thought to reflect subsequent increases in fermentation and gut microbial turnover due to increased digestibility associated with high dietary protein content (Holechek et al., 1982; Robbins 1983; Putman 1984; Wehausen, 1995; Chapman et al., 2005; Schwarm et al., 2009). Such variability in gut microbial action may complicate the analysis of fecal nitrogen as a measure of dietary quality, as metabolic nitrogen may overwhelm direct dietary signals (e.g., Milton et al, 1980; Chapman et al., 1995; Leslie et al., 2007). As a result of such issues, Leslie and Starkey (1987) and Leslie et al. (2007), pose that number of factors should be considered when using fecal nitrogen as a measure of dietary quality. These authors indicate that dietary nitrogen should be limited to examinations of 1) interseasonal dietary changes for a single population, 2) single season examinations of diet for a

single population between years when assessing variation in forage availability, 3) within-season comparisons of different populations that occupy similar habitats and consume similar diets. These authors also indicate that cross-species analysis should be avoided, due to differences in digestive physiology between taxa. This study generally fits these guidelines, although study animals with and without dental impairment were drawn from the same population during the same year to examine within-season variation in fecal nitrogen (e.g., guideline #3). However, it may be argued that animals with and without dental impairment represent divergent populations physiologically due to differences in their ability to comminute food items.

Although most research concerning fecal nitrogen content as a measure of protein intake has been limited to ungulate taxa, a number of researchers have successfully utilized fecal protein measures to discern dietary intake and/or differences in digestive physiology among nonhuman primates (e.g., Dunbar and Bose, 1991; Chapman et al., 2005; Codron et al., 2006). For example, Dunbar and Bose (1991) found that gelada baboons and "*Papio*" baboons, demonstrated a reduced capacity to digest protein within feces than either cattle or zebra. Interestingly, although geladas demonstrated an increased ability to fragment food items in comparison to *Papio* baboons, they did not demonstrate reduced protein within their feces in comparison to this species, although remaining fecal energy content was comparatively less than within the baboons. These authors indicated similarities in fecal protein content between species may have resulted from divergences in diet between the two taxa as baboons consumed fruits at a higher rate than did the more gramnivorous geladas. Such a lack of differences may also have resulted due to dissimilarities between species in digestive physiology, as per cautions of Leslie et al. (2007). Similarly, Chapman et al. (2005) found that fecal protein content for *Colobus guereza* and *Pilocolobus tephrosceles* was reduced within areas with reduced resource

availability in poor quality habitats in comparison to those drawn from areas with greater resource availability, indicating that differences in habitat and nutritional stress resulted in divergent patterns of fecal nitrogen excretion related to diet. These researchers also found that captive *Colobus guereza* showed higher fecal protein content when fed a high protein diet than when fed diets with medium or low protein content. Interestingly, when fed a low protein diet, these animals demonstrated higher fecal protein content than those fed the medium protein diet, suggesting that fecal protein does not necessarily follow a direct linear relationship with dietary protein for this species. These authors suggest that such fecal protein increases may reflect primarily microbial protein, as animals may secrete urea within the gut in response to the low protein treatment in order to promote bacterial colonies necessary for fermentation during dietary restriction (see Kay and Davies, 1994). Likewise, Codron et al. (2006) found that fecal nitrogen content for chacma baboons (*Papio ursinus*) varied between study sites (e.g., Waterberg and Kruger Park), likely reflecting site-specific differences in food item % nitrogen, although within-site microhabitat-related differences in nitrogen were limited. Seasonal variation of fecal nitrogen content was also found to follow changes in % nitrogen of available foods, generally increasing from the dry to wet seasons. These authors also noted that fecal nitrogen was higher for baboons than for sympatric ungulates These authors therefore suggested that baboons select and consume foods of higher crude protein content than do these ungulates, although it is also likely that taxonomic differences in dental morphology and/or gut physiology between baboons and ungulates could result in such divergences in fecal nitrogen (as per the critique of Codron et al.'s (2006) study posed by Leslie et al., 2007). While primate-related fecal nitrogen data studies are relatively uncommon, those studies which do exist indicate that fecal nitrogen provides a coherent means with which to better understand diet, physiology and ecology among nonhuman

primates. Nevertheless, is important to note that work by Milton et al (1980), indicating that fecal nitrogen resulted primarily from excretion of metabolic nitrogen for *Allouatta palliata* does temper these studies' results. Thus, while fecal measures of protein maintain a potential for understanding aspects of primate diet, dietary factors cannot be considered alone and physiological factors must also be examined when assessing primate fecal nitrogen.

This study differs from most studies of fecal protein content in that the area of research does not focus primarily on determining the source of dietary protein inputs or the examination of forage quality. For this study, fecal protein content in BMSR *L. catta* is assessed as a potential measure of the animal's ability to breakdown food items in relation to dental wear and tooth loss. As noted, fecal nitrogen may result from either dietary sources of nitrogen and/or metabolic processes. In the case of dental wear, I posit that an inability to process food items (particularly leaves) will result in a pattern where the animal cannot access protein contained within. Given that individuals with severe dental wear and/or tooth loss demonstrate exceptionally large portions of leaves, it is likely that fecal protein measures will reflect primarily the fraction of protein remaining in foods consumed by these individuals. However, it must be noted that individuals may also demonstrate fecal nitrogen related to physiological processes and/or resulting from gut microbes. Individuals without extensive tooth wear or loss may be more capable of comminuting challenging food items than do those with such impairment, resulting in greater protein availability to gut microbes. This may result in a situation where, even if identical diets are consumed, food items processed and presented to gut bacteria by those without dental impairment more closely resemble that found in a high-protein, higher-quality diet. Given that fecal protein content in many herbivores is directly related to the amount of protein digested by gut microbes and reflects resulting increases in microbial protein, it is possible that individuals

with reduced amounts of dental wear or loss will have higher fecal protein content than those with dental impairment. Additionally, seasonal variations may impact the relative availability of those foods available to individuals with and without dental impairment. For example, if dental impairment results in a failure to efficiently process *T. indica* fruit, this may reduce the overall capacity of individuals to ingest this food. This may subsequently result in divergent patterns of protein excretion during periods when this food is commonly consumed (e.g., during the dry season).

Study Hypotheses. I posed hypotheses relevant to each fecal measure with regard to both tooth loss and tooth wear status. All study hypotheses were based on the concept that individuals with tooth loss and/or high amounts of tooth wear would be less capable at fragmenting food items and subsequently uptake fewer nutrients from foods ingested. All hypotheses were constructed under the assumption that individuals would consume similar diets, as differential feeding between study samples could impact fecal nutritional content. This assumption was made as ring-tailed lemurs typically forage and feed as a group, with individuals typically consuming the same food item simultaneously (Simmen et al., 2003; Millette, personal observations). I also hypothesized that the direction of results were to be similar between seasons, as it was not anticipated that seasonal effects would change the overall impact of tooth wear or loss on each nutritional variable. Each study hypothesis thus encompasses data for 1) the dry season and 2) the wet season.

Fecal Fiber Content. Fecal fiber content for acid detergent fiber (ADF) and for neutral detergent fiber (NDF) was expected to be dependent on the individual's ability to effectively fragment

food items, thus permitting higher amounts of bacterial fermentation of cellulose and hemicellulose fiber fractions. For fecal fiber content the following were hypothesized:

Ha1: Fecal ADF and NDF content will be higher among those with tooth loss than for those without tooth loss, while fecal ADF and NDF content will be positively correlated with tooth wear status.

Fecal Lignin Content. Lignin (measured here as acid detergent lignin, ADL) is an indigestible dietary element and the total amount of ADL within the feces is not expected to vary from that consumed. However, given that other measured dietary elements do undergo digestion, fermentation, and absorption by the animal, it is expected that proportion of lignin present in the feces will be greater for those individuals with enhanced digestibility. Thus, it is expected that individuals with tooth loss and wear will be less capable of digesting non-lignin dietary elements, resulting in lower proportions of lignin within their feces. With regard to ADL, my hypothesis was as follows:

Ha2: Fecal ADL measures will be lower for individuals with tooth loss, and will be negatively correlated with tooth loss status.

Ratios of NDF, ADF, Hemicellulose and Cellulose to Lignin. It is expected that the amount of each measured nutritional variable will decrease relative to lignin content during digestion, fermentation and subsequent uptake by the animal as lignin is not fermentable by gut microbiota. As individuals with dental impairment are predicted to demonstrate a reduced ability to ferment structural carbohydrates, it is posited that these values will decrease less than for those without

dental impairment. Such a pattern of fermentation / digestion will result in a greater ratio of each measure relative to lignin among dentally impaired individuals.

Ha3: Ratios of all measured fiber (NDF / ADF) or structural carbohydrate (hemicellulose / cellulose) values to ADL will be higher for individuals with tooth loss than those without, while ADL ratios will also be positively correlated with tooth wear status.

Fecal Ash Content. It was predicted that the individual's ability to masticate and fragment food items would not impact fecal ash content. I therefore posed the following hypothesis for fecal ash content:

Ha4: Fecal ash content does not differ between individuals with relation to tooth status, nor is fecal ash content significantly correlated with tooth wear status.

Fecal Protein Content. As with fiber content, it is presumed that inability to fragment food items will impede the animal's access to protein held within food items. Individuals with dental impairment are expected to demonstrate higher amounts of protein in their feces than are those without.

Ha5: Fecal protein content will be higher in the fecal matter of those with tooth loss than for those without tooth loss, and will be correlated positively with tooth wear status.

Methods.

Fecal nutritional assays were conducted for a subset of study individuals for months representative of the dry (June and July) and wet (January) seasons. As sample preparation and assessment required extensive laboratory time, it was not possible to assay samples for all

individuals or months. In all, 154 samples were assessed for fiber (NDF and ADF), acid detergent lignin (ADL), crude protein and ash content. From these data, ratios of NDF and ADF to ADL were determined, as were the proportions of hemicellulose and cellulose to ADL.

Study Subjects. Three study groups were chosen based on their location within and around the reserve as related to home range microhabitat, as well as the presence of suitable individuals (e.g., animals with variety of tooth wear / loss statuses were present). Samples were thus processed for Red, Black and Blue groups. Red group (including Green 23 and Lavender 38, who were both attempting to migrate into Red Group) inhabited primarily eastern areas of the reserve along the western bank of the Sakamena River. Black group utilized marginal areas immediately south of Parcel 1 and also frequently visited the camp area. Blue group ranged in both marginal areas west of the camp as well as in areas within and around the western Parcel 1 For the month of January, data were also collected for one individual (318) who migrated from Black group to Rainbow II group, which ranged in marginal areas to the south and west of the camp along the bank of the Sakamena River (see map of study area in Chapter 2 for group locations).

Fecal samples for 14 study individuals in the aforementioned groups were assessed for nutritional content. The individuals from which data were collected are as follows: Red: 23, 38, 44, 231, 347 (5 females); Black: 226, 291, 318, 331, 345 (4 males 1 female); Blue: 217, 246, 332, 348 (4 females). Males are outnumbered by females in this sample primarily due to male migration, which limited the number of collared males in each group. Likewise, inter-group sex ratios are skewed somewhat, as for each group there were a limited number of collared individuals for which up to date tooth wear and loss data (e.g., 2011 or 2012) were available. As

a result, the number of suitable males and/or females differed between groups. Study individuals drawn from these groups maintained a variety of tooth wear and tooth loss conditions, ranging from little wear and no tooth loss (e.g., Black: 291, 318, 331, 345; Red: 231, 347; Blue: 217, 332) to those with extensive tooth wear and/or tooth loss (e.g., Red: 23, 38, 44; Black: 226; Blue: 246, 348). All measures are based on dental data collected by Dr. Frank Cuozzo during 2011-2012 field seasons (see Chapter 2). See Table 5.1 for tooth wear and tooth loss data for individuals used in this chapter.

Table 5.1. Study Subject General and Dental Characteristics.

Group	Habitat	Subject ID	Year Obs	Sex	Age	Tooth Loss	Postcanine Wear
Red	Reserve	44	11	F	12+	Yes	3.727
Red	Reserve	231	12	F	9	No	3.45
Red	Reserve	347	12	F	4	No	1.55
Red (Green)	Reserve	23	11	F	12+	Yes	3.818
Red (Lavender)	Reserve	38	12	F	13+	Yes	3.82
Blue	Reserve / Camp / Marginal	217	12	F	8	No	2.91
Blue	Reserve / Camp / Marginal	246	12	F	10	Yes	3.77
Blue	Reserve / Camp / Marginal	332	12	F	6	No	1.82
Blue	Reserve / Camp / Marginal	348	11	F	11	Yes	4.647
Black	Camp / Marginal	226	11	M	13+	Yes	4.955
Black	Camp / Marginal	291	11	M	9+	No	3.273
Black (Rain 2)	Camp / Marginal	318	12	M	6	No	2.23
Black	Camp / Marginal	331	12	M	5	No	1.88
Black	Camp / Marginal	345	11	F	4	No	1.727

Fecal Sample Collection and Preparation. Fecal samples were collected on an *ad libitum* basis following excretion by the study animal. Samples were collected from the time of contact with the group (typically prior to 07:00), until the mid-to-late afternoon (e.g., @15:00-16:00). Fecals were rarely collected after 15-16:00, as the time necessary to process and dry samples before the next day would not be sufficient after this point. For this study, field assistants collected the

majority of samples. During the dry season, fecals were collected primarily by Percy Yvon Rakoto of the University of Antananarivo, while wet season fecal samples were collected primarily by members of the BMSR ecological monitoring team (Enafa, Elahavelo, Efiteria, and Edouard) and field assistant Naina Nicolas Rasolonjatovo of the University of Toliara. When possible, I also collected fecal samples, although this proved difficult while conducting observations during periods of group activity. Most samples that I collected personally were obtained between observational periods or when study subjects were resting.

During the dry season, fecal samples were collected independently from behavioral observations, while wet season samples were obtained alongside behavioral observations. This change was made primarily because having assistants collect fecal samples alongside observational follows contributed to my ability to maintain contact with both the group and focal individuals, as dense wet-season foliage made detailed observations difficult without assistance. Due of this change in collection methods, dry season samples were typically obtained for each individual on a number of days across the month, while during the wet season fecals for each individual were typically collected on the two to six days during which their study group was observed monthly. As a result, for the dry season, samples collected for a given individual generally reflect a wider period of time than those collected during the wet season.

As refrigeration is not available on-site at BMSR, I preserved the fecal samples through drying (Rothman et al., 2012), and using a method similar to those which have been previously used at BMSR (see previous work by Brockman, Whitten and O'Mara). All fecals were initially collected using aluminum foil to encase the sample. The foil was then reopened upon return to camp, and the sample was then dried upon the foil in a Coleman (Coleman Company Inc., Golden, CO, USA) camp oven heated using tea candles. Although temperature within the oven

was not recorded due to a faulty thermometer, temperatures were generally low (e.g., approx. <50^{0}C). Samples were heated until fully dried, typically over a period of several hours to overnight in duration. During the wet season when humidity was comparatively high, drying times were sometimes longer for large samples, with desiccation taking place overnight and continuing for several hours during the next day. Following drying, all samples were placed within a Whirl-Pak (Nasco, Fort Atkinson, WI, USA) plastic bag along with a silica desiccant packet and then stored in dry, dark area prior to transport to the United States for processing and analysis.

Post-Field Sample Processing. Following return to the US, samples were initially processed by removing any seeds present prior to nutrient analyses. Seeds were removed as these were not typically digested by animals and appeared to pass through the digestive tract unaltered. If left within the sample, seeds would inflate fecal nutrient values, as these represented a portion of the diet that should not contribute to the animal's nutrient intake (see Urquiza-Hass et al., 2008). For tamarind fruit, if seeds retained their seed coat, this was removed and returned to the sample as this portion of the fruit demonstrates a sticky, pulpy layer which can provide nutritive content and often contains other fecal contents (e.g., leaf fragments) which adhere to its surface following excretion. Sample weight was recorded both prior to and following seed removal, and from these data, total seed mass was also determined. The number and type of seeds removed were also recorded. As the majority of seeds were represented by *T. indica* and *T. pruinosum*, the number of seeds from these taxa were recorded. Seeds from other taxa, which were typically unidentifiable, were recorded as "unknown."

Prior to assessment for nutritional content, all samples were ground into a fine powder using a centrifugal mill with a 2 mm screen. Milling was necessary to reduce samples into particles suitable for analysis. Milling also homogenized samples, a critical step as only a small proportion of each was utilized for each assay. Prior to milling, all samples were examined once more for the presence of seeds, which if found, were removed and the sample was then reweighed. Once milled, fecal materials were then placed into covered, ventilated plastic soufflé cups and allowed to dry in a food dehydrator (at 40^0C) for a period of at least 12 hours (e.g., overnight). Due to the low temperature of this dehydrator, samples were also stored in this condition prior to analysis to maintain low moisture content.

Fecal Nutritional Assays. I personally conducted all fecal nutritional assays for fiber (ADF and NDF), lignin (ADL), ash and crude protein content in Dr. Matt Sponheimer's Nutritional and Isotopic Ecology Laboratory at the University of Colorado Boulder. For fiber contents, assays were completed for neutral detergent fiber (NDF: cellulose, hemicellulose and lignin), acid detergent fiber (ADF: cellulose and lignin), and for lignin [(ADL organic matter (om)]. Fiber analyses were run using an ANKOM 2000 (ANKOM Technologies, Macedon, NY, USA) fiber analyzer using a subtractive method, where NDF was first determined, followed in series by ADF and ADL. Crude protein analysis was carried out using a LECO FP-528 (LECO Corporation, St. Joseph, MI, USA) nitrogen analyzer. For all measures, results are presented as the percent of total sample mass.

Neutral Detergent Fiber Analysis. NDF was assessed by first placing approximately 0.4500g to 0.5000g of ground sample within an ANKOM F57 fiber filter sample bag, which was then closed

using an impulse bag heat sealer (ANKOM Model 915 Heat Sealer). All sample bags were weighed prior to filling in order to allow determination of fiber content independent of bag weight. Twenty-three samples were then placed within supplied bag suspender trays before insertion into the fiber analyzer, along with two blank bags (for use in correction factors related to fiber leakage from the bags) and a control sample consisting of cut and milled grass.

Once placed within the ANKOM 2000 fiber analyzer, samples were processed using the machine's NDF protocol. This procedure exposes the sample to reagents, temperatures and pressures necessary to remove soluble fiber from each sample. During this cycle, the sample was exposed automatically to approximately one liter of pre-mixed NDF solution available from ANKOM and consisting of the following diluted in distilled water: Sodium dodecyl sulfate (USP; 30g/liter), ethylenediaminetetraacetic disodium salt (dehydrate; 18.61g/liter), sodium borate (6.81g/liter), sodium phosphate dibasic (anhydrous; 4.56g/liter), and triethylene glycol (10g/liter). In addition, 4ml of alpha-amylase and 20g of sodium sulfite (Na_2SO_3) were added to the NDF solution upon filling the fiber analyzer at the start of the procedure, but prior to closing the machine's pressure vessel. An additional 8ml of alpha-amylase diluted with deionized water was placed into an amylase dispenser attached to the side of the machine (port B), to be injected automatically during rinse cycles conducted during processing. Once all reagents were inserted, the machine was left to cycle until the NDF extraction process was complete (approximately 1 hour).

After extraction, all samples were removed from the fiber analyzer, placed within a 250ml beaker and pressed to remove excess water. The beaker was then filled with acetone (to assist with drying), and the samples were allowed to soak for approximately 5 minutes. The acetone was then drained and samples again pressed to remove excess liquid. All filter bags were

then air dried for approximately 20-25 minutes to allow excess acetone to evaporate (to avoid fire danger during desiccation). Samples were then placed within a food dehydrator at 40^0C, where the samples were allowed to dry completely (typically overnight). Following drying, all samples, blanks and controls were then removed from the food dehydrator, placed into a plastic bag with silica desiccant, and allowed to return to room temperature. All sample filter bags were then weighed individually to determine weight lost during the procedure. These data were then entered into a worksheet available from ANKOM to calculate the percentage of NDF within each sample.

Acid Detergent Fiber Procedure. Acid detergent fiber (cellulose and lignin) was determined for each sample following weighing during the NDF procedure. The method utilized is broadly similar to that used for NDF, with 23 samples, two blanks and one standard again being processed in an ANKOM 2000 fiber analyzer under heat, pressure and with reagents necessary to remove the hemicellulose fraction from each sample. In contrast to the NDF procedure, once the samples were placed within the analyzer, no additional reagents were added prior to closing of the fiber analyzer's pressure vessel. In addition, prior to commencing the ADF protocol, the temperature within the fiber analyzer was cooled to below 20^0C using crushed ice. ADF analyses, like NDF, utilized a commercially available (from ANKOM) acid detergent solution to process all samples. This premixed solution consisted of cetyl trimethylammonium bromide (CTAB: 20g/Liter) diluted in standardized 1.00M H_2SO_4. Samples were then left to run through a standard ADF extraction cycle within the fiber analyzer. Following removal from the fiber analyzer, the same post-extraction procedures as used for NDF were completed. Samples were first pressed to remove water, and then allowed to soak in acetone for 5 minutes. These were

then dried overnight, and then weighed to the nearest 0.0001 gram. Sample weights were then entered into the ADF template, from which the percent ADF in each sample was calculated.

Lignin Analysis and Ash. Acid detergent lignin and ash analyses were completed for each sample bag previously used to determine ADF/NDF fecal content. Lignin content was determined by removing the cellulose fraction from that remaining after the ADF analysis and then subtracting the ash content from the mass of the sample following removal of the cellulose fraction.

Prior to measuring ADL content, samples were dried overnight using a food dehydrator at approx. 40 degrees C, after storage following the completion of ADF analyses. Likewise, prior to the day of analysis, a mixture of 72% H_2SO_4 was made by diluting 1200g of concentrated sulfuric acid with 350ml of deionized water. Due to the heat generated by this step, the diluted mixture was allowed to cool overnight in an open container under a fume hood. Following these initial steps, samples were loaded into partitioned glass vessels (24 samples/vessel), and covered in approximately 500ml of the diluted sulfuric acid. The samples were then placed within an ANKOM Daisy II model incubator where they were rotated at room temperature for a period of 3 hours.

Following incubation, samples were removed and placed into a custom wash container and then flushed with tap water for a period of approximately 40 minutes. Flushing continued until the samples reached a relatively neutral pH (e.g., pH = 6), equal to that of the water used neutralize the samples. Commercially-available pH paper was utilized to determine the acidity of the sample following removal from the incubation vessels. Once samples were neutralized, they were pressed to remove excess water, and placed into acetone (to aid drying) for 5 minutes. Following removal from the acetone, all samples were pressed to remove excess fluid, and then

dried overnight at 40^0C. Samples were then weighed to determine mass loss resulting from exposure to H_2S0_4.

To determine ash content, samples were combusted for a period of approximately six hours. For each run, 24 samples were first placed upon pre-weighed combustion tins, and then placed within a high-temperature muffler. The muffler was then heated to 600 degrees Celsius over a period of 3 hours, after which samples were allowed to cool to room temperature. Ash remaining in each combustion tin was then weighed the following day after all samples had returned to room temperature. Percent ash content was then determined using a worksheet provided by ANKOM.

Crude Protein Analysis. Fecal protein content was determined using a LECO FP-528 Nitrogen/Protein Analyzer (LECO Corporation, St. Joseph, MI, USA). This device is commonly used to assess protein content in forage samples, and has been used by the Nutritional and Isotopic Ecology Laboratory (University of Colorado Boulder, Department of Anthropology) to assess a variety of food items drawn from African hominin sites. To maintain as similar a profile to samples run previously, all study samples were assessed using the "Cereal" setting (e.g., a standard 6.25 multiplication factor, commonly used in ecological studies), which has proven effective for examining forage collected by members of this laboratory.

Prior to assessing samples, the machine was first pressure checked for both oxygen and helium gasses, and then calibrated to account for atmospheric nitrogen by running a number of blank samples. Samples were then run by placing approximately 0.1500g of ground and dried fecal material into foil capsules, although in a number of cases a smaller amount of sample was utilized due to insufficient sample quantity. For smaller samples, no analysis issues were

apparent due to reduced sample mass. In order to homogenize the samples, each one was stirred prior to placing it within the capsule. Samples were also assessed visually to ensure homogeneity. This step was critical as samples were often heterogeneous in nature (e.g., woody material was often present in larger particles after grinding, while smaller powder-like particles were present for leafy material). Capsules were then combusted completely within the nitrogen analyzer, which then ascertained nitrogen content and determined crude protein content automatically. Additionally, prior to running the samples, the machine was conditioned by running yellow soy (approx. 0.1500g). Samples were also run with standards consisting of approx. 0.1500g of EDTA and approx. 0.1500g of brown rice flour.

Statistical Analyses. Following collection, data produced from each sample were collated into a Microsoft Excel spreadsheet before being entered into JMP Pro 11 (SAS Institute INC, Cary NC, USA) for analysis. To reduce the data into a form suitable for assessment (e.g. to eliminate issues associated with non-independent repeated measures), the mean value for each nutritional variable (e.g., NDF, ADF, etc.) was calculated for each individual. Such values were calculated for the dry and wet seasons individually. As each fecal sample was processed and assayed using a ground subsample, all resulting data were reported in percentages of total sample mass. From the initial dataset, ratios of ADF and NDF relative to ADL were also determined for all individuals. Likewise, the ratios of % hemicellulose to % lignin (%HC/%L) as well as % cellulose to % lignin (%C/%L) were calculated.

The impact of dental impairment upon these variables was examined using nonparametric methods. To assess the relationship of tooth loss to each variable, a Wilcoxon (i.e., Mann-Whitney U) test was performed. In this case, animals were grouped into “Loss” or “No Loss”

categories based on the presence or absence of missing teeth within the dental arcade. Each nutritional variable was assessed in relation to postcanine dental wear status. Postcanine wear status was determined during dental assessments conducted by Dr. Frank Cuozzo during the field seasons of 2011 and 2012. During these assessments the wear of each tooth was assigned a score of 0-5 (no wear to wear resulting in tooth loss). The wear index used here thus reflects the mean value of the scores generated for each study animal's postcanine teeth. As this is an ordinal scoring system, Spearman's correlations were used to assess the relationship between tooth wear and each fecal nutritional variable. For all tests, significance was set at the $p = 0.05$ level using a two-tailed distribution.

Results.

Significant differences between Loss and No Loss groups were noted for a number of fecal nutritional variables, with between-samples being present seasonally. In addition, there were a number of significant correlations between dental wear and fecal nutritional status when data were examined on a seasonal basis.

Fecal Nutritional Status by Tooth Loss and Season.

Dry Season Results. Significant differences were noted between Loss and No Loss groups for samples collected during the dry season (June-July) for all fecal nutritional measures (Table 5.2, Figure 5.1). NDF and ADF measures were higher among individuals without tooth loss than among those with tooth loss (NDF: No Loss = 59.34%, Loss = 53.99%, $p = 0.017$, $S = 26$, $Z = -2.39$; ADF: No Loss = 44.41%, Loss = 40.41%, $p = 0.024$, $S = 27$, $Z = -2.26$). Likewise, ADL was higher among individuals without tooth loss (ADL: No Loss = 14.73%, Loss = 10.45%, p =

0.002, S = 21, Z = -3.03). In contrast to fiber values, ash and protein results were higher among those with tooth loss than those without (Ash: No Loss = 2.80%, Loss = 6.35%, p = 0.002, S = 69, Z = 3.03; protein: No Loss = 10.86%, Loss = 13.99%, p = 0.012, S = 65, Z = 2.52). NDF/ADL and ADF/ADL ratios were higher among those with tooth loss than without (NDF/ADL: No Loss = 4.15, Loss = 5.57, p = 0.002, S = 69, Z = 3.03; ADF/ADL: No Loss = 3.09, Loss = 4.15, p = 0.002, S = 69, Z = 3.03). Similar patterns were also present for hemicellulose / lignin and cellulose / lignin ratios [(%HC/%L: No Loss = 1.06, Loss = 1.42, p = 0.008, S = 66, Z = 2.65; %C/%L: No Loss = 2.02, Loss = 3.15, p = 0.002, S = 69, Z = 3.03) Table 5.2, Figure 5.1].

Table 5.2. Dietary Variables by Tooth Loss for the Dry Season.

	%NDF		**%ADF**		**%ADL**		**%Ash**		**%Protein**	
Loss status	No	Yes	No	Yes	No	Yes	No	Yes	No	Yes
n =	8	6	8	6	8	6	8	6	8	6
Mean	**59.34**	53.99	**44.41**	40.41	**14.73**	10.45	2.80	**6.35**	10.86	**13.99**
Std Dev	3.27	1.96	2.57	2.05	0.96	1.26	1.10	1.44	1.65	1.58
Std Err Mean	1.16	0.80	0.91	0.84	0.34	0.51	0.39	0.59	0.58	0.64
Lower 95%	56.61	51.93	42.26	38.26	13.93	9.13	1.88	4.84	9.48	12.33
Upper 95%	62.08	56.04	46.55	42.57	15.53	11.77	3.72	7.87	12.24	15.64
Score Sum	79	26	78	27	84	21	36	69	40	65
Expected Score	60	45	60	45	60	45	60	45	60	45
Score Mean	9.88	4.33	9.75	4.50	10.50	3.50	4.50	11.50	5.00	10.83
(Mean-Mean0)/Std0	2.39	-2.39	2.26	-2.26	3.03	-3.03	-3.03	3.03	-2.52	2.52
S-score	26		27		21		69		65	
Z-score	-2.39		-2.26		-3.03		3.03		2.52	
p =	***0.017***		***0.024***		***0.002***		***0.002***		***0.012***	

	NDF/ADL		**ADF/ADL**		**%HC/%L**		**%C/%L**	
Loss Status	No	Yes	No	Yes	No	Yes	No	Yes
n =	8	6	8	6	8	6	8	6
Mean	4.15	**5.57**	3.09	**4.15**	1.06	**1.42**	2.09	**3.15**
Std Dev	0.24	0.85	0.16	0.60	0.09	0.29	0.16	0.60
Std Err Mean	0.09	0.35	0.06	0.25	0.03	0.12	0.06	0.25
Lower 95%	3.95	4.67	2.96	3.52	0.98	1.12	1.96	2.52
Upper 95%	4.35	6.46	3.22	4.78	1.14	1.72	2.22	3.78
Score Sum	36	69	36	69	39	66	36	69
Expected Score	60	45	60	45	60	45	60	45
Score Mean	4.50	11.50	4.50	11.50	4.88	11.00	4.50	11.50
(Mean-Mean0)/Std0	-3.03	3.03	-3.03	3.03	-2.65	2.65	-3.03	3.03
S-score	69		69		66		69	
Z-score	3.03		3.03		2.65		3.03	
p =	***0.002***		***0.002***		***0.008***		***0.002***	

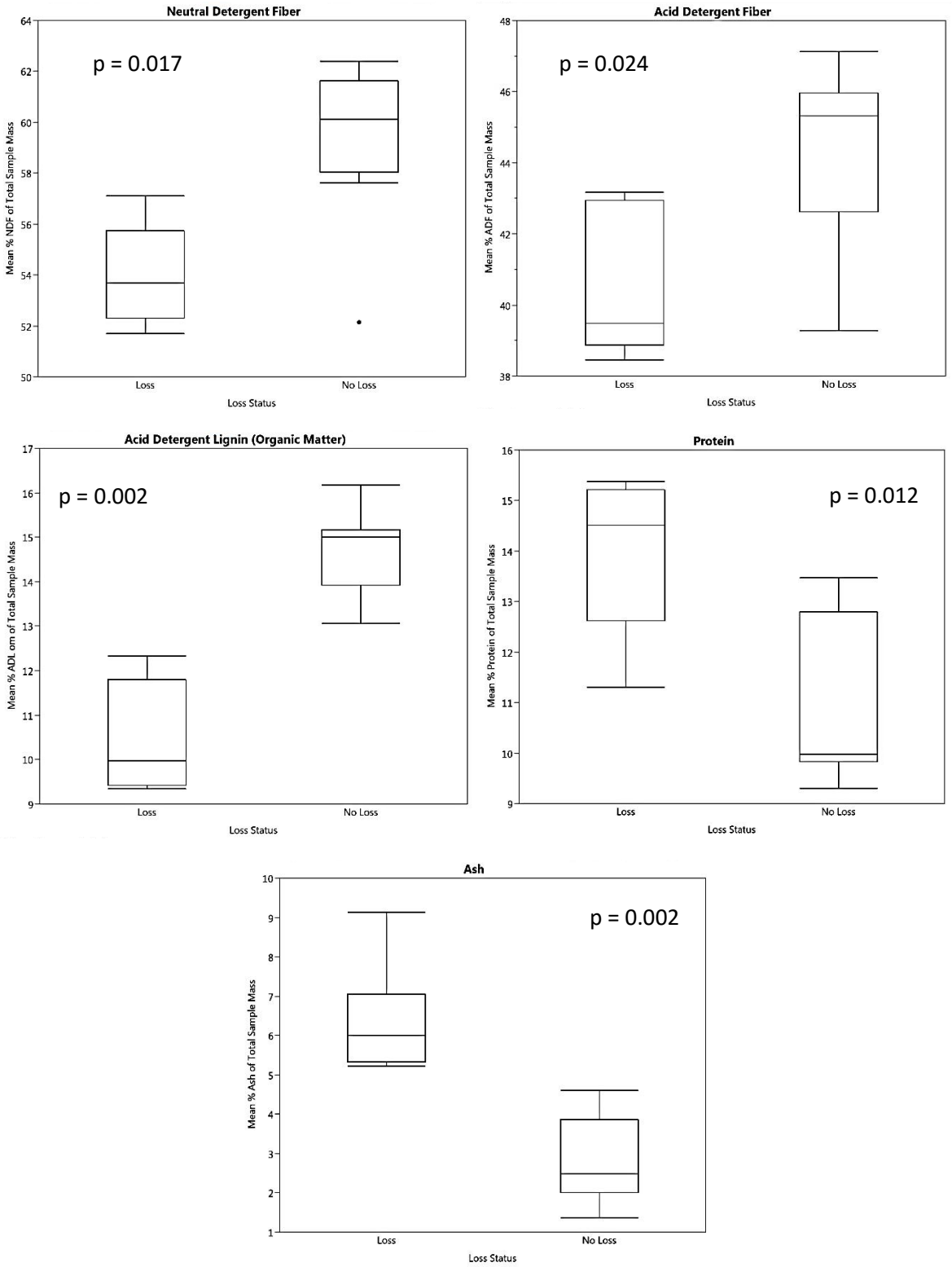


Figure 5.1. Dietary variables by tooth loss status for individuals with and without tooth loss for the dry season.

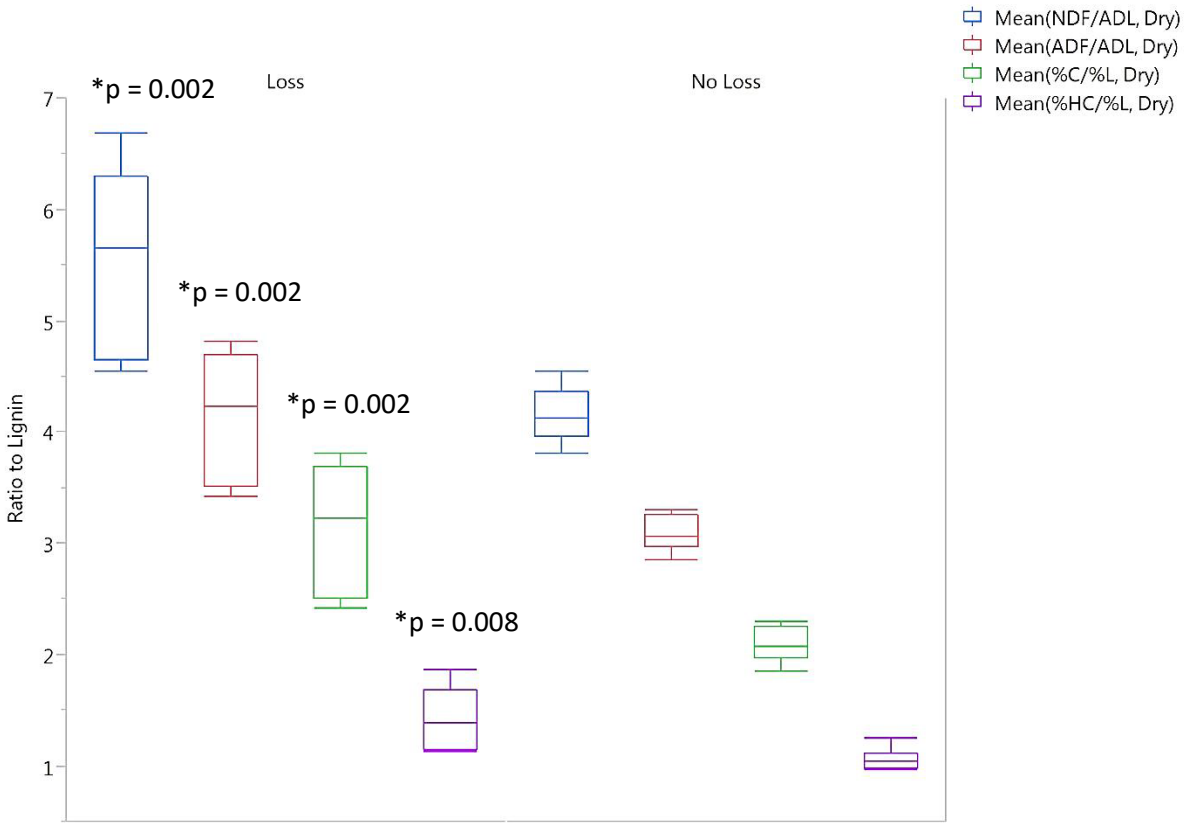


Figure 5.2. Ratios of fecal fiber and structural carbohydrate content to lignin by tooth loss status for the dry season. All differences between tooth loss groups are significant at the $p < 0.05$ level.

Wet Season Results. As with the dry season and for the dataset overall, significant differences were present for a number of fecal nutritional variables (Table 5.3, Figures 5.3-5.4). In contrast to the dry season dataset, NDF and ADF were higher among those with tooth loss than those without tooth loss (NDF: No Loss = 46.32%, Loss = 51.66%, p = 0.033, S = 62, Z = 2.13; ADF: No Loss = 34.51%, Loss = 38.90%, p = 0.033, S = 62, Z = 2.13. Ash values were also significantly higher among those with tooth loss than for those without (Ash: No Loss = 4.08%, Loss = 8.96%, p = 0.045, S = 61, S = 2.00, Z = 2.52). With regard to NDF/ADL and ADF/ADL, only NDF/ADL differed by tooth loss status (NDF/ADL: No Loss = 5.40, Loss = 6.65, p = 0.033, S = 62, Z = 2.13). Although not statistically significant, trends towards greater ratios were

present for individuals with tooth loss for ADF/ADL and %C/%L (ADF/ADL: No Loss = 4.02, Loss = 5.01, p = 0.061, S = 60, Z = 1.87; %C/%L: No Loss = 3.02, Loss = 4.01, p = 0.061, S = 60, Z = 1.87). No significant differences were noted between groups for ADL, protein or %HC/%L during the wet season (ADL: No Loss = 9.15%, Loss = 8.37%, p = 0.220, S = 35, Z = -1.23; Protein: No Loss = 14.52%, Loss = 16.42%, p = 0.401, S = 52, Z = 0.84; %HC/%L: No Loss = 1.38, Loss = 1.64, p = 0.106, S = 58, Z = 1.62).

Table 5.3. Dietary Variables by Tooth Loss for the Wet Season.

	%NDF		**%ADF**		**%ADL**		**%Ash**		**%Protein**	
Loss Status	**No**	**Yes**	**No**	**Yes**	**No**	**Yes**	**No**	**Yes**	**No**	**Yes**
n =	8	6	8	6	8	6	8	6	8	6
Mean	46.32	**51.66**	34.51	**38.90**	9.15	8.37	4.08	**8.96**	14.58	16.42
Std Dev	4.83	4.11	3.93	4.14	1.55	2.10	2.20	5.05	3.14	2.73
Std Err Mean	1.71	1.68	1.39	1.69	0.55	0.86	0.78	2.06	1.11	1.11
Lower 95%	42.28	47.35	31.22	34.56	7.85	6.17	2.24	3.66	11.95	13.56
Upper 95%	50.36	55.97	37.79	43.24	10.44	10.57	5.92	14.26	17.20	19.29
Score Sum	43	62	43	62	70	35	44	61	53	52
Expected Score	60	45	60	45	60	45	60	45	60	45
Score Mean	5.38	10.33	5.38	10.33	8.75	5.83	5.50	10.17	6.63	8.67
(Mean-Mean0)/Std0	-2.13	2.13	-2.13	2.13	1.23	-1.23	-2.00	2.00	-0.84	0.84
S-score	62		62		35		61		52	
Z-score	2.13		2.13		-1.23		2.00		0.84	
p =	***0.033***		***0.033***		0.220		***0.045***		0.401	

	NDF/ADL		**ADF/ADL**		**%HC/%L**		**%C/%L**	
Loss Status	**No**	**Yes**	**No**	**Yes**	**No**	**Yes**	**No**	**Yes**
n =	8	6	8	6	8	6	8	6
Mean	5.40	**6.65**	4.02	5.01	1.38	1.64	3.02	4.01
Std Dev	0.78	1.21	0.52	1.03	0.29	0.23	0.52	1.03
Std Err Mean	0.28	0.49	0.18	0.42	0.10	0.09	0.18	0.42
Lower 95%	4.75	5.38	3.59	3.93	1.14	1.40	2.59	2.93
Upper 95%	6.06	7.92	4.45	6.09	1.63	1.88	3.45	5.09
Score Sum	43	62	45	60	47	58	45	60
Expected Score	60	45	60	45	60	45	60	45
Score Mean	5.38	10.33	5.63	10.00	5.88	9.67	5.63	10.00
(Mean-Mean0)/Std0	-2.13	2.13	-1.87	1.87	-1.62	1.62	-1.87	1.87
S-score	62		60		58		60	
Z-score	2.13		1.87		1.62		1.87	
p =	***0.033***		*0.061*		0.106		*0.061*	

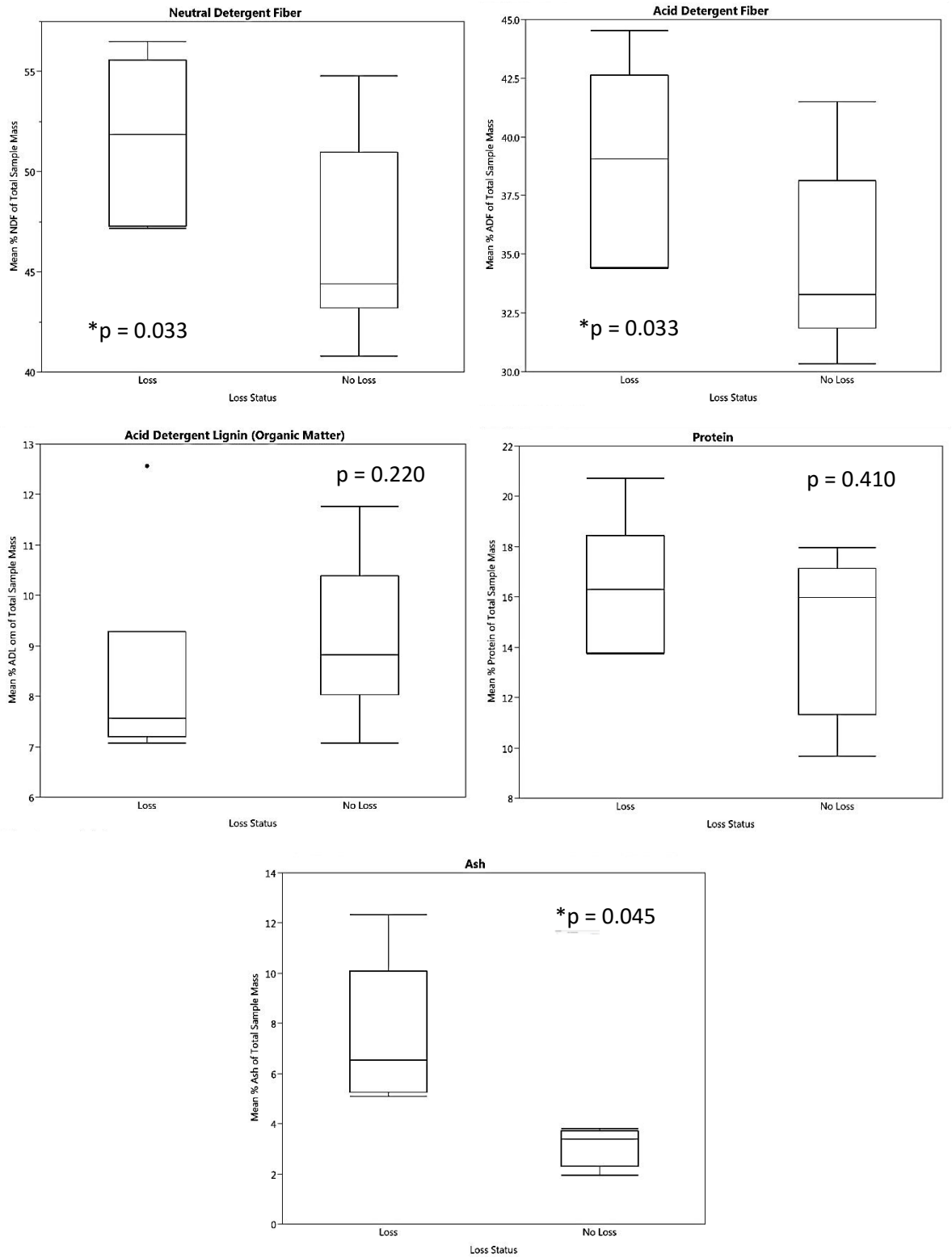


Figure 5.3. Dietary variables by tooth loss status for individuals with and without tooth loss for the wet season.

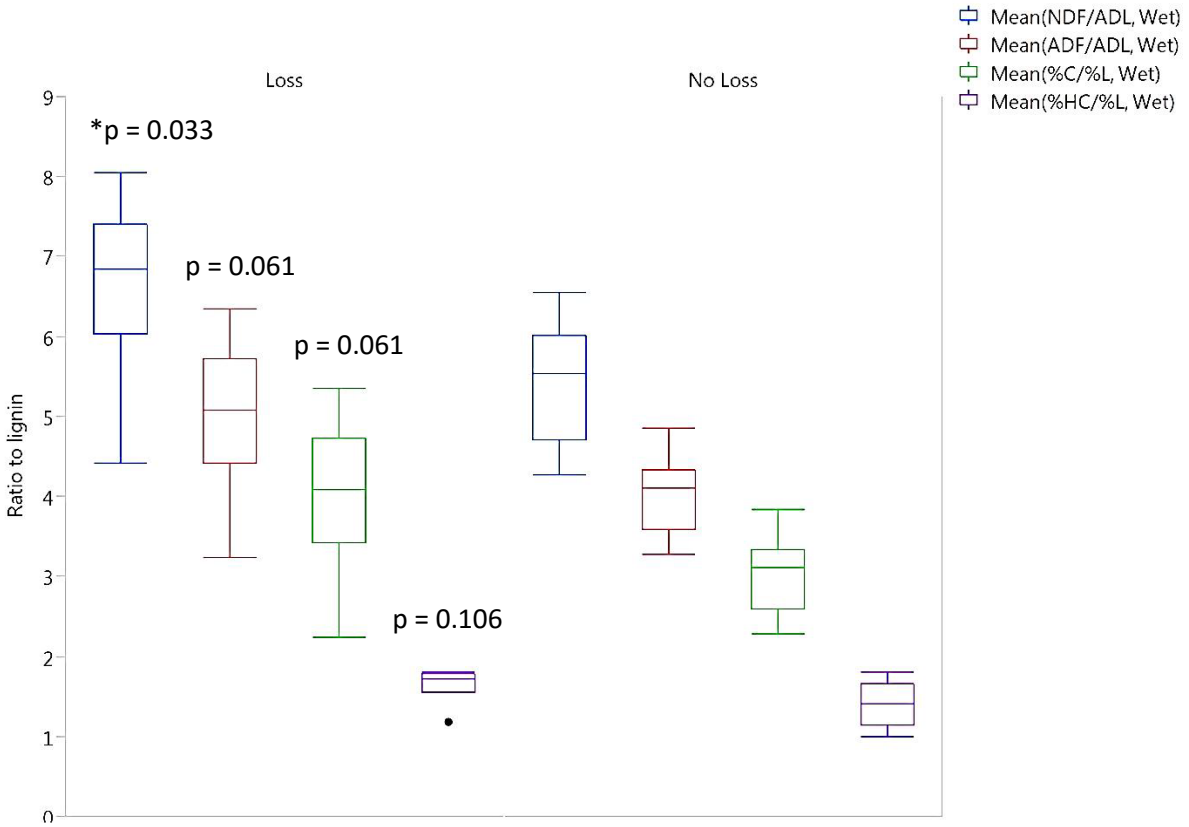


Figure 5.4. Ratios of fecal fiber and structural carbohydrate content to lignin by tooth loss status for the wet season. *Significant differences ($p < 0.05$) between tooth loss and no loss groups are present only for NDF/ADL, while trends are present between ADF/ADL and %HC/%

Fecal Nutritional Status by Tooth Wear and Season. To examine the impact of tooth wear upon fecal nutritional status, ordinal tooth wear scores for the postcanine dentition of each focal animal was examined in relation to each fecal nutritional variable. All ordinal wear scores were determined using data produced by Dr. Frank Cuozzo during the 2011 and 2012 field seasons. Due to the ordinal nature of the postcanine wear scores, nonparametric correlations (Spearman's rho) were utilized to assess the relationship between tooth wear and fecal nutritional status for both the wet and dry seasons.

Dry Season Results. During the dry season, NDF was significantly and negatively associated with tooth wear (rho = -0.538, p = 0.047), while ADF was not significantly correlated with tooth wear (rho = -0.459, p = 0.098). As with the two-season dataset, ADL remained negatively correlated with tooth wear (ADL: rho = -0.692, p = 0.006). Likewise, for the dry season both protein and ash values correlated positively with tooth wear [(Protein: rho = 0.653, p = 0.011; Ash: rho = 0.754, p = 0.002). Similar positive correlations for NDF/ADL, ADF/ADL, %HC/%L, and %C/%L were also present [(NDF/ADL: rho = 0.714, p = 0.004; ADF/ADL: rho = 0.727, p = 0.003; %HC/%L: rho = 0.622, p = 0.018; %C/%L: rho = 0.727, p = 0.003) Table 5.4, Figures 5.5-5.10].

Table 5.4. Tooth Wear Status in Relation to Fecal Nutritional Values (Spearman's Correlations).

	Dry Season		**Wet Season**	
Nutritional Value	**rho =**	**p =**	**rho =**	**p =**
%NDF	-0.538	***0.047***	0.459	0.098
%ADF	-0.459	0.098	0.486	*0.078*
%ADL	-0.692	***0.006***	-0.332	0.246
%Ash	0.754	***0.002***	0.512	*0.061*
%Protein	0.653	***0.011***	0.310	0.281
NDF/ADL	0.714	***0.004***	0.596	***0.025***
ADF/ADL	0.727	***0.003***	0.547	***0.043***
%HC/%L	0.622	***0.018***	0.460	0.098
%C/%L	0.727	***0.003***	0.547	***0.043***

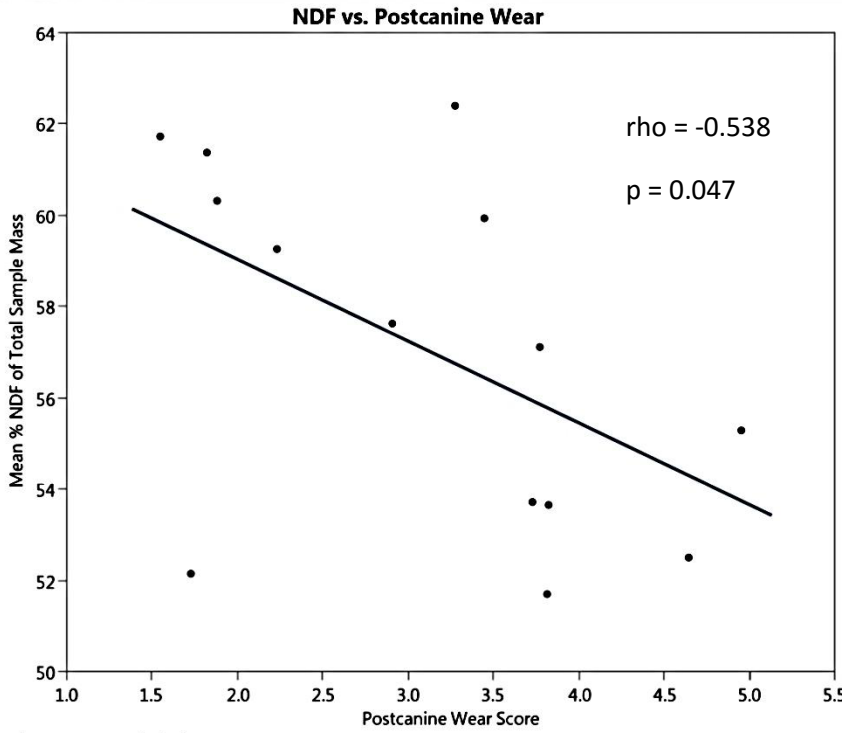


Figure 5.5. Dry season correlation between %NDF and postcanine wear score.

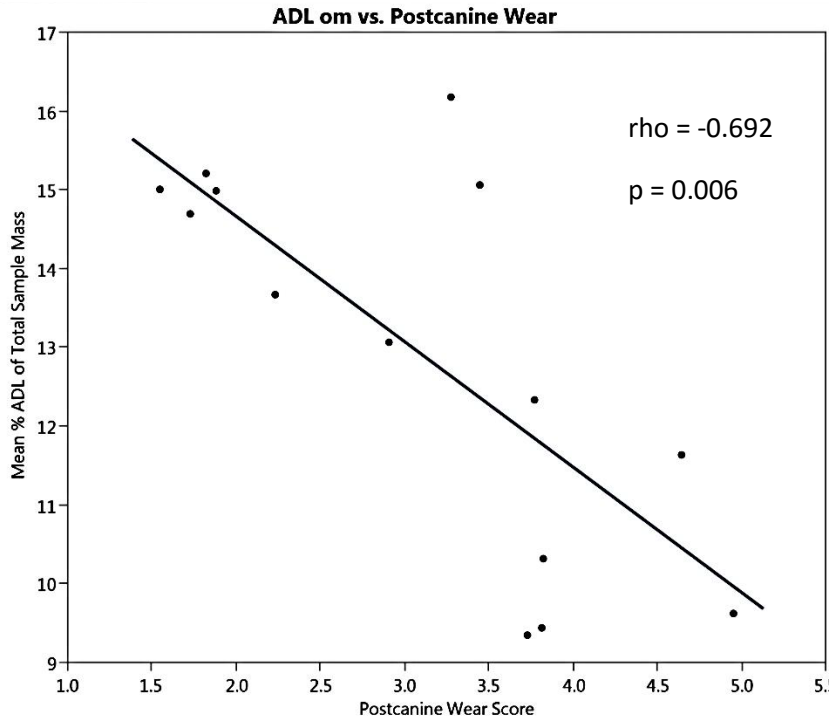


Figure 5.6. Dry season ADL content by postcanine wear score.

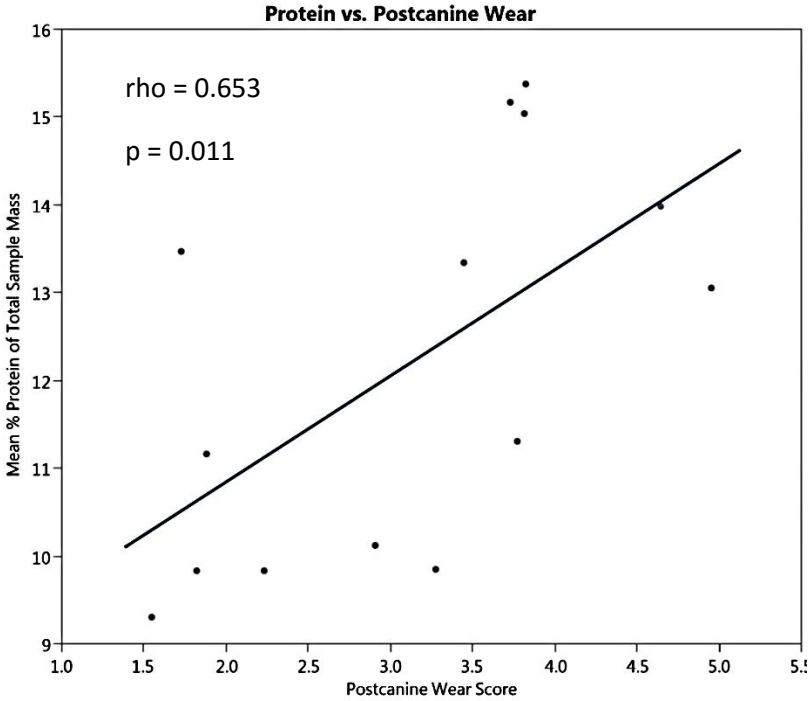


Figure 5.7. Dry season fecal protein by postcanine wear score.

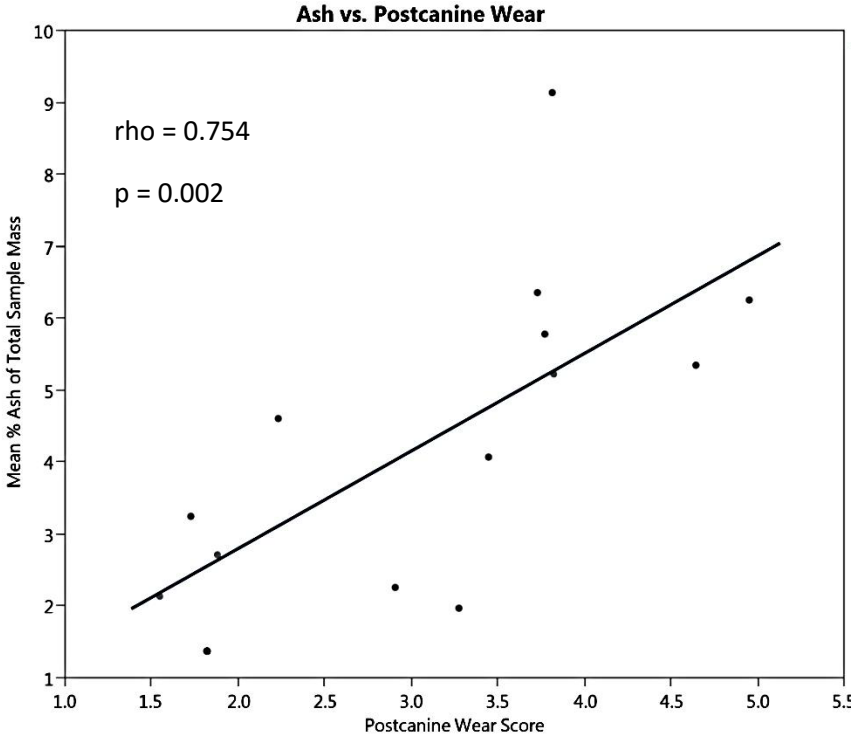


Figure 5.8. Dry season fecal ash content by postcanine wear score.

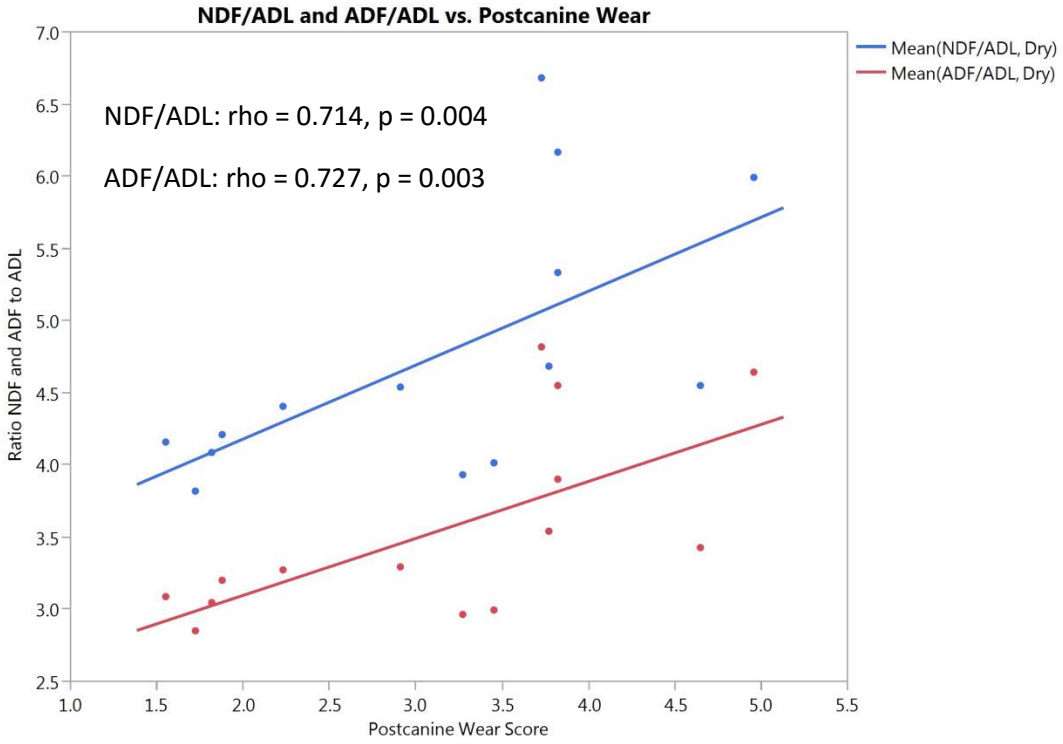


Figure 5.9. Dry season ADF/ADL and NDF/ADL by postcanine wear score.

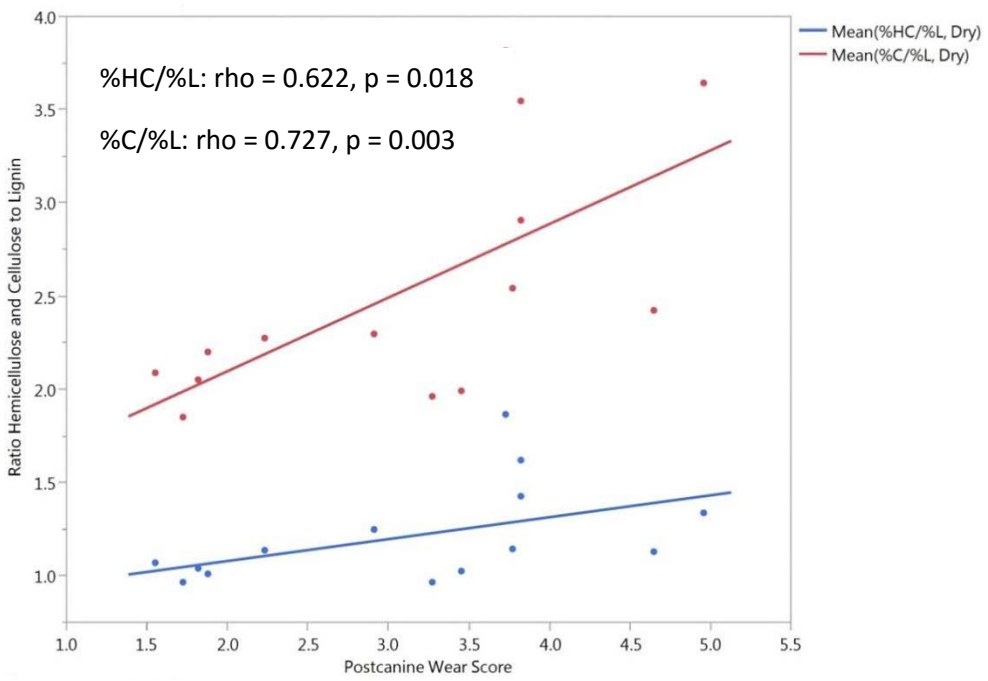


Figure 5.10. Dry season hemicellulose and cellulose to lignin ratios by tooth wear status.

Wet Season Results. No significant correlations were found between ADF, NDF or ADL and tooth wear status during the wet season, although a positive trend was present towards for ADF [(NDF: rho = 0.459, p = 0.098; ADF: rho = 0.486, p = 0.078; ADL: rho = -0.332, p = 0.246) Table 5.4]. In contrast to data collected during the dry season, protein was not correlated with tooth wear status [(rho = 0.310, p = 0.281) Table 5.4]. However, a trend towards increasing amounts of ash relative to dental wear was present (rho = 0.510, p = 0.061). Ratios of NDF to ADL, and ADF to ADL demonstrated a positive correlation with tooth wear status (NDF/ADL: rho = 0.596, p = 0.025; ADF/ADL: rho = 0.547, p = 0.043). No correlation was found between % hemicellulose and % lignin (rho = 0.460, p = 0.098), although a correlation between % cellulose and % lignin (rho = 0.547, p = 0.043) and dental wear status was found (Table 5.4, Figures 5.11-5.12).

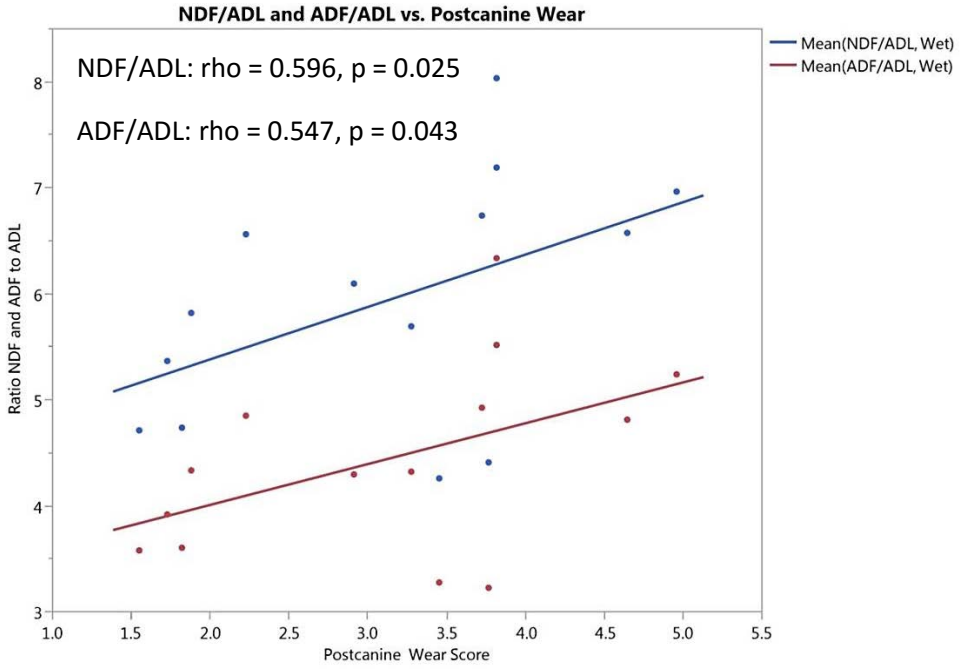


Figure 5.11. NDF/ADL and ADF/ADL vs. postcanine wear during the wet season.

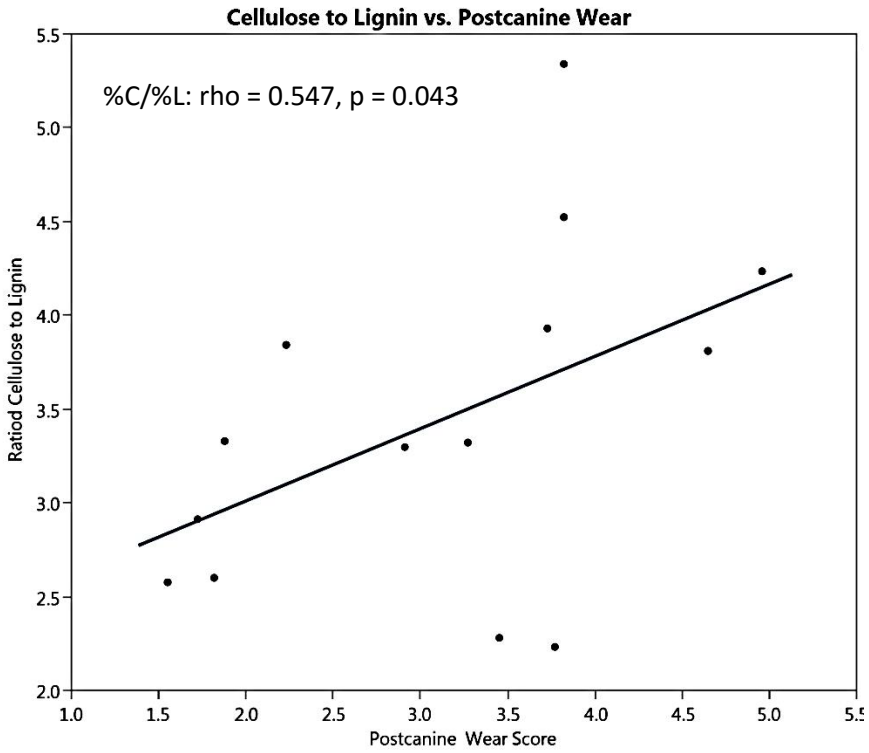


Figure 5.12. Wet Season cellulose to lignin ratio by postcanine wear.

Overall, a number of nutritional variables were either significantly associated with tooth loss status and/or with tooth wear status. Seasonally variable differences noted for a number of dietary measures related to cell wall contents and protein, with wet season associations with dental impairment typically being less significant than found during the dry season. In addition, increased ratios of dietary fiber to lignin content, as well as ratios of structural carbohydrates to lignin content were generally associated with measures of dental impairment. Ash content was furthermore generally associated with tooth wear and loss during both seasons. Tables 5.5 and 5.6 summarize general patterns of fecal nutrient content observed during this study with regards to both tooth loss and wear status. The first table (Table 5.5) visualizes differences in fecal nutritional content for those with tooth loss in comparison to those without, while the second (Table 5.6) visualizes correlations of fecal content with increasing dental wear.

Table 5.5. Do Individuals with Tooth Loss Demonstrate Divergent Patterns of Fecal Nutrient Excretion?

Measured Variables:	Seasons Combined	Dry Season	Wet Season
NDF	↔	↓	↑
ADF	↔	↓	↑
ADL	↓	↓	↔
Protein	↑	↑	↔
Ash	↑	↑	↑
Lignin Ratios:			
NDF / ADL	↑	↑	↑
ADF / ADL	↑	↑	↗
%HC / %L	↑	↑	↔
%C / %L	↑	↑	↗

↔ = No Difference ↑ = Significantly Higher
↓ = Significantly Lower ↗ = Trend Higher

Table 5.6. Does Tooth Wear Status Correlate With Fecal Nutrient Excretion?

Measured Variables:	Seasons Combined	Dry Season	Wet Season
NDF	↔	↘	↔
ADF	↔	↔	↗
ADL	↘	↘	↔
Protein	↗	↗	↔
Ash	↗	↗	↗
Lignin Ratios:			
NDF / ADL	↗	↗	↗
ADF / ADL	↗	↗	↗
%HC / %L	↗	↗	↔
%C / %L	↗	↗	↗

↔ = No Correlation ↗ = Positive Correlation
↘ = Negative Correlation ↗ = Trend Positive

Discussion.

Mammalian dental morphology demonstrates forms that facilitate the processing and subsequent mechanical breakdown of food items (Kay, 1975; Seligsohn, 1977; Kay et al., 1978; Kay and Covert, 1984; Lanyon and Sanson, 1986; Yamashita 1998a; Logan and Sanson, 2002; Lucas, 2004; Evans et al., 2007). Although low to moderate tooth wear may not impact the individual's ability to fragment food items, and may even facilitate tooth function (see Rensberger, 1973; Janis and Fortelius 1988; M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004; King et al., 2005), significant dental wear and or tooth loss may alter the morphology of the dental apparatus in a manner that impedes the breakdown of food items (Gipps and Sanson, 1984; Lanyon and Sanson, 1986; Logan and Sanson 2002; Millette et al., 2012 / Chapter 6). Tooth loss and/or tooth wear are expected to impact the individual's ability to effectively fragment food items, resulting in the ingestion of food particles of larger sizes. As larger particle sizes demonstrate a higher surface to volume ratio than do smaller particles, tooth wear may impact subsequent food item fermentation, digestion and uptake as both digestive enzymes and gut microbes have a reduced surface to act upon relative to food item volume (Sheine, 1979; Lanyon and Sanson, 1986; Bjorndal et al., 1990; Lentle et al., 2003; Millette et al., 2012 / Chapter 6; Ellis et al., 2015). Consumption of food particles of larger size may, in particular, negatively impact the individual's ability to harness energy available from insoluble fiber due to reduced fermentation capacity as mammals lack enzymes capable of hydrolyzing either hemicellulose or cellulose without microbial action (Sheine and Kay, 1979; Sheine, 1979; Bjorndal et al., 1990; Lambert, 1998; Gipps and Sanson, 2004; Lentle et al., 2003). Impediment of food item breakdown related to tooth loss and/or wear has been previously reported for BMSR ring-tailed lemurs, where animals with significant tooth loss demonstrated reduced amounts of

small-sized particles in their feces than did those without tooth loss, while increasing tooth wear was also correlated with lower amounts of small-sized particles in the feces (Millette et al., 2012 / Chapter 6). Likewise, captive digestive studies by Sheine (1979) demonstrate that ring-tailed lemurs more effectively ferment and utilize cellulose presented in smaller particles than they do cellulose of larger particle sizes. Thus, it is not unreasonable to hypothesize that BMSR *L. catta* with tooth loss and/or significant dental wear demonstrate reduced capacity to ferment and digest ingested food items, which is predicted to result in divergent patterns of fecal nutrient excretion. Results presented in this chapter suggest that this is likely the case, although the patterning of fiber excretion is directionally inconsistent across seasons (see below).

Fecal Fiber and Cell Wall Contents.

Neutral Detergent Fiber and Acid Detergent Fiber. For this study, I hypothesized that fiber content would form a higher percentage of fecal mass for individuals with tooth loss than for those without tooth loss during the dry season and for the wet season. This hypothesis was based on the assumption that individuals with tooth loss would demonstrate a reduced ability to fragment food items into fragments suitable for fermentation by gut microbes (see above). I also expected that individuals without tooth loss and/or wear would pass less fiber through their digestive tract in comparison to those with tooth loss and/or wear, as this material would be converted into short chain fatty acids (e.g., volatile fatty acids) which are subsequently absorbed and utilized by the animal (e.g., Van Soest, 1994; Lambert, 1998, 2007b).

The data presented here do not support the hypotheses that dietary fiber measured in the form of ADF or NDF is excreted at higher rates for individuals with tooth loss or wear during dry season. During this period, individuals without tooth loss demonstrated higher fecal NDF

and ADF values than did individuals with loss. This failure of the data to provide support for the active hypothesis may result from differential access to food resources during this period between those with and without dental impairment (see below). In contrast, fecal samples from the wet season do provide support for the hypothesis that individuals with dental impairment excrete more fiber within their feces than do those without dental impairment, as NDF and ADF values were higher for those with tooth loss during this period.

During the dry season, individuals with tooth loss demonstrated lower levels of NDF and ADF in their feces than did those without tooth loss. Similarly, increasing tooth wear status was associated with reduced fecal NDF, although fecal ADF was not associated with tooth wear (but was in the same direction as for NDF). These dry season results run counter to initial hypotheses that dental impairment would be associated with higher fecal NDF and ADF content. One explanation for these data is that individuals without tooth loss or significant wear are consuming foods with higher overall fiber content than those without such impairment during the dry season. Much of the undigested portion of the diet processed during the dry season was represented by *T. indica* shells remaining in the fecal material and which were not removed from the samples during processing. Shells of this fruit appear to be very difficult to digest as they pass through the digestive tract in a clearly identifiable form and demonstrates a woody, fibrous consistency. Although no data is available for the nutritional content of tamarind shell, based on its comparatively indigestible nature and woody nature, it is likely that tamarind shell contains significant amounts of dietary fiber. Additionally, my previous work at BMSR (Millette et al., 2009; Millette, personal observations) demonstrates that individuals at BMSR with tooth loss demonstrate difficulties processing tamarind fruit, and spend more time feeding on tamarind per feeding bout. Such patterns of impairment when processing this food were also found during this

study, as individuals with reduced tooth area demonstrated longer feeding bout lengths for *T. indica* fruit (see Chapter 4). Dentally impaired individuals may consume fewer tamarind fruit than individuals without tooth loss, resulting in lower overall fiber content in their feces. This finding is somewhat perplexing, however, given previous observations (Millette et al., 2012 / Chapter 6) that during the dry season individuals with tooth loss demonstrate greater proportions of leaf material remaining in their feces than do those without tooth loss. While these findings suggest an inability to access tamarind fruit, it is also probable that such remaining leaf material would be relatively high in insoluble fiber. The pattern observed during this study, however, could be achieved if the amount of fiber present in tamarind shell in the feces of those with lower amounts of dental impairment is greater than that found remaining in the leaves present in the feces of those with increased amounts dental impairment.

During the wet season, both fecal ADF and NDF content were higher for those with tooth loss than for those without. This pattern of ADF and NDF excretion is coherent with my initially proposed hypothesis, and provides evidence that dentally impaired individuals may less efficiently ferment insoluble fiber. Although *T. indica* remained a common food item during this time, and was present in the feces of the animals, the wet season at BMSR is characterized by a fluorescence of vegetative (e.g., leaves and flowers) materials (Sauther, 1998; Rasamimanana et al,, 2012; Millette, personal observations). In contrast to the dry season when much of the forest is without vegetation (excluding *M. parvifolia* and *T. indica* mature leaves), during the wet season a variety of leaves and flowers become available to, and are frequently consumed by, ring-tailed lemurs (see chapter on lemur feeding ecology). This may explain the differences between individuals with and without tooth loss, as it is likely that individuals with tooth loss are less able to breakdown leaf or flower based materials than are those without tooth loss, resulting

in higher fecal ADF and NDF values. It is notable that fecal NDF and ADF are not significantly correlated with tooth wear during the wet season. This may suggest that tooth wear alone is not enough to significantly impact the individual's ability to fragment leaves into small fragments suitable for the fermentation of dietary fiber. Thus, it is likely that significant wear causing dental impairment in the form of tooth loss is necessary to result in divergent patterns of fecal fiber excretion.

Acid Detergent Lignin. Lignin is considered an "anti-quality" element within feeds that reduces the amount of energy available from structural carbohydrates hemicellulose and cellulose. This phenolic polymer is a key structural component of cell walls and adds strength and rigidity to plant tissues, but also impedes the fermentation of structural carbohydrates by physically blocking access to these by gut bacteria. Total lignin content is positively associated with indigestible dry matter content in plant forage, and may also limit the amount of forage an animal can consume and subsequently utilize (Jung and Deetz, 1993; Moore and Hatfield 1994; Mertens, 1994; Van Soest, 1994; Jung and Allen, 1995; Buxton et al., 1996; Hatfield et al. 1999; Moore and Jung, 2001; Jung, 2012).

The ADL data presented here are consistent with the hypothesis that the fraction of ADL present within the feces is lower for those with tooth loss for the dry season. The proportion of ADL within the feces of those individuals with tooth loss was reduced in comparison to those individuals without tooth loss during this time. Similarly, there was a significant negative correlation between tooth wear status and fecal lignin content during the dry season. Lignin content, however, did not differ between loss categories or by wear status during the wet season, although the general direction of the data remained consistent with that from the dry season.

Lignin remains undigested when travelling through the digestive tract relative to other dietary contents as mammals generally lack gut microbes with a capacity to break down this compound (Van Soest, 1994). As all data are reported in terms of percent of total sample mass, the lower lignin content in the feces of those with dental impairment indicate that these animals are failing to digest and absorb other nutrients. As the relative proportion of other nutrients increases within the feces, the relative amount ADL present will correspondingly decrease. Significantly higher values of protein, ash and fiber are variably present for individuals with tooth loss depending on season, and significant correlations for these variables in relation to tooth wear status are also present. As such, these data indicate that ADL decreases with tooth wear, and is also associated with tooth loss, during the dry season is likely indicative of a reduced digestive capacity for dentally-impaired individuals during these periods.

This explanation does have difficulties accounting for the lack of significant effects during the wet season. While NDF and ADF are higher for individuals with tooth loss during this period, ADL does not differ significantly between samples. However, the magnitude of differences for NDF and ADF are lower between tooth loss status categories during the wet season, and there is also no difference for protein during this period. Similarly, no significant correlations are present for any of these measures during the wet season. Such smaller dental impairment-related effects for these dietary components may thus permit relative percentages of ADL to remain similar between tooth loss categories and/or limit variability related to tooth wear status.

While the dry season lignin data do appear to indicate differences in digestibility related to tooth loss, they may also indicate that dental impairment status is associated with selection of food items based upon their lignin content. Non-dentally impaired individuals potentially

maintain a comparatively higher capacity to breakdown more mechanically-challenging food items, and may thus consume these at a higher rate than those with impairment. In contrast, those with dental impairment may be selecting food items with reduced lignin content if these foods are easier to process. This is plausible as lignin is a key structural element in plant cell walls, and adds rigidity to plant tissues. Likewise, as lignin can limit energy available from cell wall components, low-lignin foods should also be more digestible overall than those with higher amounts of lignin. By consuming food items with lower lignin content, these individuals may be better able to access energy stored in hemicellulose and cellulose despite having a reduced capacity to fragment foods during mastication.

Ratios of Fiber and Structural Carbohydrates to Acid Detergent Lignin. Perhaps the greatest indicator of dental impairment impacting the individual's ability to utilize food resources comes from data assessing ratios of NDF, ADF, hemicellulose and cellulose individually to ADL content. Lignin is indigestible relative to the hemicellulose and cellulose fractions of dietary fiber (Van Soest, 1994). As a result, dental impairment is hypothesized to result in higher ratios of NDF and ADF to ADL, as well as higher ratios of hemicellulose and cellulose to ADL. This hypothesis is based on the reasoning that larger ingested particle sizes will result in a comparatively reduced ability to ferment the hemicellulose and cellulose fraction of the diet, while particle size will not impact lignin content in the sample due to its indigestible nature. In this manner, lignin may serve as an internal marker of digestive capacity when examined in conjunction with other more digestible fractions of the diet. It is, however, important to note that these data are subject to the limitation that lignin consumption could not be measured for prior to ingestion, and total lignin consumption may have varied based on dental impairment status.

Lignin may also demonstrate limited fermentability and some lignin may be lost during digestion, thus impacting observed ratios with other cell wall components (see above).

NDF and ADF to ADL Ratios. During the dry season ADF/ADL and NDF/ADL ratios are significantly higher for those with tooth loss. In contrast, during the wet season while only the ratio of NDF to ADL was significantly higher for those with tooth loss, a trend in the same direction was present for ADF to ADL. These data indicate that individuals with tooth loss may have a more difficult time fermenting the structural carbohydrate fractions of NDF (cellulose, hemicellulose) and ADF (cellulose). With respect to tooth wear status, a similar pattern holds true for both the dry and wet season, where the ratio of ADF/ADL and NDF/ADL are positively correlated with tooth loss status. Similar to patterns with tooth loss status, these data are particularly strong during the dry season, with correlations of lower significance being noted during the wet season. These data are consistent with the hypothesis that tooth loss and tooth wear significantly impact the individual's ability to process and subsequently harness energy from the fiber fraction of their diet.

As noted above, during the dry season ADL was higher for those individuals without tooth loss than for those with tooth loss. These data are consistent with the observed lower ratios of ADF and NDF to ADL for those without tooth loss. While NDF/ADL and ADF/ADL ratios are interesting, both the NDF and ADF fiber fractions demonstrate the presence of lignin. Higher lignin content within a given sample will also inflate overall ADF and NDF values, potentially impacting the resulting ratio. Thus, the ratio of hemicellulose to lignin and cellulose to lignin may provide a better measure of fermentative capacity (see below).

Hemicellulose and Cellulose to ADL Ratios. Overall, the ratios of hemicellulose to lignin (%HC/%L) and cellulose to lignin (%C/%L) demonstrate patterns in relation to tooth loss similar to those seen with ADF/ADL and NDF/ADL ratios. Seasonally, higher ratios were also present for those with tooth loss during the dry season, but not during the wet season, although a trend towards higher ratios of %C/%L was found for individuals with tooth loss. All ratios were significantly and positively correlated with tooth wear seasonally, except for %HC/%L for the wet season.

These ratio data generally fit the posed study hypotheses (see above), and suggest that individuals with dental impairment demonstrate an inability to ferment structural carbohydrates with the same capacity as those without dental impairment. All significant differences are in the same direction, with higher ratios always occurring among individuals with tooth loss, or with increasing tooth wear status. These data are consistent for the dry season, although values are not significant for the wet season for tooth loss or for tooth wear for %HC/%L. These dry season data, and wet season data for %C/%L with respect to tooth wear and tooth loss, are consistent with expectations that individuals who possess dental impairment will also demonstrate a relatively impaired ability to break down food items during mastication, ingest particles of comparatively larger size and subsequently fail to ferment structural carbohydrates found within cell walls. As noted previously, ingestion of foods which are of greater particle size will reduce the amount of surface area on which digestive enzymes and gut bacteria may act upon relative to particle volume. Research with BMSR *L. catta* has demonstrated that individuals with dental impairment show reduced amounts of small sized food particles in their feces, which provides a mechanism for dental impairment to impact food absorption and thus these lignin ratios (Millette et al., 2012 / Chapter 6).

One key finding from these data is that there are seasonal differences in the relationship of tooth loss status to the ratio of either ADF or NDF to ADL, and also for hemicellulose or cellulose to ADL. That differences are clearly present for the dry season, but are only limited to ADF/ADL for the wet season is likely related to food availability. For ADF, NDF and ADL, dry season fecal values are higher overall than for the wet season, suggesting that the availability of low fiber / low lignin resources is reduced during the dry season. Significant differences between loss and no loss samples during the dry season may thus indicate that individuals with tooth loss are comparatively less able to digest the diet available during this time. Tamarind forms a key dry season fallback food, and is both difficult for individuals to process and breakdown due to its physically large size and mechanically challenging mechanical properties (Cuozzo and Sauther, 2006a,b; Sauther and Cuozzo, 2009; Millette et al., 2009; Yamashita et al., 2012). Likewise, during this period the availability of less challenging foods such as young leaves and alternative fruit species is limited (see Sauther 1998; Millette, personal observations). The lack of significant differences between tooth wear loss categories during the wet season, for all but NDF/ADL ratios, may thus reflect an increase in preferred forage during this time, and an increased ability of individuals with dental impairment to comminute and subsequently digest such foods. However, significant correlations for tooth wear status relative to lignin ratios are present during the wet season except for %HC/%L, as are trends towards differences between tooth loss status categories. These findings indicate, that while the impact of dental impairment is somewhat attenuated during this period, animals with tooth loss or extensive wear are still likely lacking in their ability to ferment structural carbohydrates during the wet season. As such, the general patterning of these “lignin ratios,” both overall and seasonally, indicate that individuals

with dental impairment are less capable in their ability to ferment dietary fiber and its component structural carbohydrates.

Fecal Nitrogen / Protein Content. I hypothesized that animals with dental impairment would demonstrate higher fecal protein than those without dental impairment. As with fecal fiber content, this hypothesis was based on the assumption that individuals with dental impairment would be less likely to fragment food items into small sized particles than those without dental impairment. When consuming vegetative materials, such as leaves, it is necessary for an animal to break open cell walls through mastication to access proteins contained within (Hladik, 1978; Lambert, 1998, 2007b). I anticipated that individuals with dental impairment would be unable to do so due to their inability to finely reduce food particles relative to individuals without dental impairment, thus leaving greater proportions of residual protein within their feces. I also, however, posited that individuals with tooth wear and/or loss could show lower amounts of protein within their feces due to reduced protein available to gut bacteria, which could reduce the amount of metabolic nitrogen found within the feces.

For tooth loss status, protein content in the feces was higher for those with tooth loss for the dry season. During the wet season, protein content did not differ significantly between loss and no loss samples. When examined by tooth wear status, a similar pattern was also noted, with fecal protein content being positively correlated with tooth wear status for the dry season, and with no significant correlations present for the wet season.

These data provide additional evidence that those individuals with dental impairment also show impairment when digesting and utilizing food items. As with fecal fiber and structural carbohydrate to lignin ratios, such impairment appears to occur primarily during the dry season,

with little evidence for impairment of protein uptake during the wet season. Such patterns may stem from overall differences in protein intake between seasons. Fecal protein content was higher during the wet season than during the dry season. These data are consistent with the general increase in vegetative material available to animals at BMSR during the wet season, as leaves tend to be relatively high in potentially available protein (Lambert, 1998, 2007b). Similar patterns of fecal nitrogen increase related to increasing nitrogen content in forage have also been noted between the dry and wet seasons for South African baboons (Codron et al., 2006). The data presented in this study, however, contrasts with that of Yamashita (2008a), who found that protein intake did not vary significantly between seasons for BMSR *L. catta.* For this study, I only examined data drawn from two months over the course of the year, in contrast to 9 months for Yamashita's study. LaFleur and Sauther (2015) have, however, reported that dietary protein for BMSR *lemur catta* varies on a month to month basis. For example, LaFleur and Sauther found that the highest protein content in top food items for BMSR *L. catta* (from September through March) occurred during the wet season month of February (41% crude protein), with highly variable amounts of protein available for other months (21-32%). While January crude protein was not particularly high for LaFleur and Sauther's study (23%), differences in rainfall between Sauther's 1987-88 and my research in 2012-2013 (when there was enough rainfall to cause flooding of the Sakamena river on two separate occasions -- the first flood since the cyclone of 2005) may have resulted in divergent patterns of forest phenology and food availability between these studies.

Higher availability of protein during the wet season could also potentially lead to a ceiling effect where individuals cannot digest all protein available irrespective of tooth loss status, leading to the lack of differences between tooth loss categories and/or wear state.

Alternatively, it may simply be more difficult for individuals to access protein-dense resources during the dry season, as food availability is limited (and is dominated by mechanically-challenging *T. indica* fruit) during this period. Dental impairment may exacerbate the individual's ability to access protein-rich resources, leading to the divergent patterns of fecal protein content observed. Divergences in tannin consumption between groups as related to dental impairment may also account for dry season differences. If individuals without tooth loss or low wear differentially access foods containing tannins this may increase the amount of protein within the feces, as these have been positively associated with increased fecal nitrogen content (Robbins et al., 1987; Verheyden et al., 2011). Alternatively, dry season differences between loss-no loss samples and correlations with tooth wear status related to fecal nitrogen content may reflect divergences in the ability to maintain gut bacteria by those with dental impairment. Diet quality in terms of available protein is associated with increased fecal nitrogen / protein content related to the inclusion of gut microbes and their byproducts within the feces for a number of species (e.g., Schwarm et al., 2009). If animals with dental impairment cannot comminute their foods into particles suitable for fermentation, then lower gut bacterial content could result in reduced amounts of fecal nitrogen. This is inconsistent with values observed, suggesting that such an explanation does not account for patterns of fecal protein observed during this study. However, it must be noted that animals may be following patterns of fecal nitrogen excretion similar to that of Chapman et al.(2005), where animals fed a low protein diet demonstrated higher fecal nitrogen levels than did those fed a diet with moderate protein content, possibly due to endogenous urea excretion into the gut. Irrespective of source, these data suggest that individuals with tooth loss and or increasing tooth wear may have a more difficult time accessing protein from food resources during the dry season.

Fecal Ash Content. Fecal ash content varied between tooth loss status categories across both seasons. These patterns of ash excretion were not consistent with that expected (e.g., that ash would not vary by tooth loss or wear status). Individuals with tooth loss demonstrated higher ash content than did those without across both seasons. This pattern also held with respect to wear, with positive correlations between wear and ash content being present for the dry season. A trend ($p = 0.061$) was present for the wet season, although a significant correlation was not found.

Assessing where such patterns of fecal ash content originate with respect to dental impairment is somewhat problematic, although differences in dietary content are likely the source of such variation. Individuals with tooth loss may simply be consuming food items with higher endogenous silica content (e.g., from phytoliths), which pass through the digestive tract unaltered. However, little data to support this assumption are available as no ash or endogenous silica values for food items consumed during this study are currently available. Perhaps it is more likely that individuals with dental impairment are consuming greater amounts of food items exogenous silica adhering to their surface. BMSR *L. catta* frequently consume food items which have fallen to the forest floor. This is particularly true for *T. indica* fruit, although other large fruits (e.g., those of *Crateva excelsa*, "Akaly") are also consumed in such a manner. Individuals with tooth loss may be consuming these fruits at a differentially high rate, as these have frequently been opened and discarded by other individuals (or have been otherwise opened before or while on the forest floor) and thus do not require extensive dental processing to consume (see Cuozzo and Sauther, 2006a). Such foods may contain higher amounts of grit, sand or "dirt" than do those that have been sourced from trees directly (although see Ungar et al., 1995). Data on food processing behaviors (Chapter 5), however, suggest that individuals with higher amounts of dental wear are not consuming tamarind from the ground at increased rates,

which does temper this hypothesis. Nevertheless, several individuals with tooth loss have been observed to consume small chunks of tamarind from ground on a regular basis ("tambits", particularly Black 226 and Red 44) and to which grit likely adheres due to exposure of the tamarind's sticky, pulpy interior. Although use of such foods was not recorded directly during this study, they may serve as a means for increasing the amount of grit consumed by individuals with tooth loss. Lemurs at BMSR also consume a number of foods with a high likelihood of containing exogenous grit which are not sourced from the ground directly. Such foods include leaves and fruits from the farmed areas adjacent to the reserve (particularly those of kisene squash and voamanga melon) which are grown in sandy soils, and the introduced plant *Argemone mexicana* that also grows in sandy soils. Fecal matter sourced from the forest floor is also a common food item consumed during the dry season for many individuals (Fish et al., 2007), and could also include high amounts of exogenous silica. If animals with tooth loss are consuming these food items at a higher rate than those without, then fecal ash content is likely to be higher for these individuals.

Fecal ash content may also result from soil adhering to samples following collection. While some sand was noted in a few samples, when these were assayed clearly visible grit was removed from the portion of the sample utilized during analysis. As such, this explanation is probably unlikely to represent a major source of ash within the feces of the study animals. Likewise, this explanation would not explain why patterns of ash excretion differ between tooth loss categories or wear state.

Higher ash content in the fecal material of individuals with tooth loss and/or higher wear scores is also interesting with regard to the source of dental wear in this population. Although tooth wear and loss here has been associated with tamarind fruit feeding (see Sauther et al., 2002;

Cuozzo and Sauther, 2004, 2006; Cuozzo et al., 2008), such patterns may also be related to the inclusion of exogenous grit within the diet (Cuozzo and Sauther, personal communication) as this is common source of tooth wear among mammalian species (e.g., Ludwig et al., 1966; Hummel et al., 2011; Lucas et al., 2013, 2014). If exogenous grit is the source of divergent patterns of ash in the feces of BMSR lemurs, it is likely that individuals with tooth wear and/or tooth loss are consuming more silicates in their diet than those without impaired dentitions. Such fecal ash thus may not only result from consuming foods on the ground or otherwise with higher amounts of grit adhering to their surfaces, but may also represent a source of tooth wear in this population. Individuals with tooth loss may not be only consuming foods which contain higher grit than those without tooth loss due to an inability to access unopened resources, but rather these individuals may also have higher measures of dental impairment due to their consumption of such resources. In selecting such foods due to their ease of processing, individuals may enter a vicious cycle where grit causes further wear and additional dental impairment. If this is the case, examination of fecal ash content may thus provide key insights into the development of dental wear in this and other primate populations.

Conclusion - Fecal Nutritional Measures and Dental Impairment: What Does it All Mean?

Data presented here provide a relevant, yet somewhat complicated, picture of the relationship between tooth wear and tooth loss to fecal nutritional content. The relationship between fecal ADF and NDF and dental impairment is particularly difficult to disentangle. While significant differences are present between tooth loss categories for both the dry and wet season, the direction of such differences is inconsistent between seasons. Unexpectedly high values for those without tooth loss during the dry season may indicate that individuals are divergently

selecting different foods than those with tooth loss. It is likely that individuals with tooth loss are choosing items of comparatively reduced fiber content, despite a lack of food availability during this time. Alternatively, they may simply not be able to access key dry season food items such as tamarind, which has a fibrous shell and tough internal fibers. In contrast, during the wet season, individuals with tooth loss demonstrate higher amounts of fecal fiber, perchance suggesting that they maintain a reduced ability to process and ferment fibrous foods during this period.

Significant differences by tooth loss status and correlations for tooth wear for lignin ratios do suggest that individuals with dental impairment maintain a reduced ability to ferment structural carbohydrates. These patterns are particularly strong during the dry season, while less significant results were found for the wet season. Similar patterns were also noted for protein content in fecal samples, with individuals with tooth loss demonstrating higher amounts of protein for the dry season. These data likely indicate that dental impairment constrains the ability of individuals to process, ferment/digest and subsequently absorb ingested food items. The source of this variation, whether related to nitrogen within food items or nitrogen resulting from metabolic sources, however, remains murky. Nevertheless, such impairment appears to impact individuals the most during the dry season, a finding which is coherent with general patterns of food availability during this period.

Finally, fecal ash content is also of interest, varying significantly by tooth loss status for both seasons, with individuals with tooth loss having higher amounts of ash. Similar patterns are also present with respect to tooth wear status, although only a near-significant trend was observed during the wet season. These data likely indicate that individuals with dental impairment are consuming greater amounts of exogenous grit than those without. Such grit may

be both a result of tooth loss if sourced from pre-processed food items from the forest floor, but also may be a source of tooth loss in this population if consumed in sufficient quantities.

While the data presented here are complex, it is clear that patterns of fecal nutrient content vary significantly based upon tooth loss and dental wear status. While the source of such variation is currently difficult to determine, it is evident that tooth wear and/or loss is connected to the patterns observed here on some level. These data provide preliminary evidence that fecal nutritional analysis can provide a valid means for assessing the impact of dental impairment on animal digestive physiology, and potentially for better understanding aspects of behavior and ecology among wild primate populations.

Future research is however needed in order to disentangle if these patterns result from divergent patterns of feeding (e.g., for fiber or protein intake during the dry season), or from less efficient digestion and subsequent maintenance of gut microbiota, as related to tooth loss. Such research could feasibly be completed by recording individual feeding over an extended period of time, determining the nutrient content of foods consumed and then assaying resulting fecal material using methods presented here. Unfortunately, the data collected during this study with regard to individual feeding patterns was of limited scope (typically only 3-4.5 hours per individual per month) and are not well suited to assessing food intake on an individual basis. Digestive research similar to that proposed has however been completed *in vivo* among captive lemurs as well as for other captive primate and non-primate mammal taxa (Kay and Sheine, 1979; Sheine, 1979; Bjorndal et al., 1990; Edwards and Ullrey, 1999a,b; Campbell et al., 2004; Schmidt et al., 2005a,b). Likewise, dietary nutrient content has previously been examined at BMSR by a number of researchers (e.g., Sauther, 1992; Yamashita, 2008a; LaFleur and Sauther, 2015). While difficult, such work could likely be completed in a well-designed study.

Additionally, data from this research is limited to a small number of individuals (n = 14) drawn from a small number of social groups. Continued research must also examine a wider number of individuals of a variety of ages and drawn from a variety of habitat areas (e.g., marginal areas, BMSR Parcel 2 as well as Parcel 1). The limited sample size currently presented precludes examination of factors such as age and habitat use in relation to tooth wear and loss status, although future inclusion of all individuals and samples could likely provide insight into these areas of interest (see below). In particular, age-related variation in fecal ash content could provide data vital towards understanding the development of dental impairment within this population, particularly if combined with a longitudinal framework. Finally, future research must examine a greater number of months during both the wet and dry seasons with regard to each fecal variable. The period of time during which the data presented here were collected reflects only a limited portion of the year. Work by LaFleur and Sauther (2015) demonstrates that fiber and crude protein content (and carbohydrate content) vary by month at BMSR, suggesting that data from a wider portion of the year than available here could be useful for pinpointing sources of variation in fecal nutrient content related to seasonal changes in phenology.

I only assayed a limited number of samples during this study; this was primarily due to limitations in time available for laboratory analyses. Currently, this project includes only a fraction of all individuals for which both dental and fecal data are available (e.g., 14 of 36 potential individuals), and only examined a fraction of samples generated by those animals reported here. Although approximately 1050 additional fecal samples were collected during this study for all 36 individuals across a period of 10 months (June-March), these remain to be assayed for nutritional content. Future analysis of these samples is currently planned for the near

future, and I anticipate that these will provide additional insight into fecal nutritional content, and its relationship to diet and dental impairment status among BMSR *L. catta*.

CHAPTER VI:

THE IMPACT OF DENTAL IMPAIRMENT ON RING-TAILED LEMUR FOOD PROCESSING PERFORMANCE.

Published previously in the *American Journal Physical Anthropology* as:

Millette JB, Sauther ML, Cuozzo FP. 2012. The impact of dental impairment on ring-tailed lemur food processing performance. American Journal of Physical Anthropology 148:238-248.

Introduction.

Teeth are the primary structures by which foods are initially processed and reduced before subsequent swallowing and digestion. For particular diets, mammalian dental morphology therefore demonstrates forms that facilitate the generation of forces necessary to cause runaway crack propagation within foods characterized by specific physical and structural properties (Lucas, 2004). For example, researchers have long noted that primarily folivorous and insectivorous species demonstrate high-cusped and crested teeth with shearing capabilities necessary to initiate and continue cracks within foods characterized by physically tough structural carbohydrates such as found within cell walls (e.g., cellulose) and chitinous insect exoskeletons. In turn, primarily frugivorous and hard-object consuming taxa demonstrate morphologies of lower relief suited to breaking open the cell walls of fruits through grinding and crushing, or opening hard foods (e.g., seeds) without incurring damage to the tooth (Kay, 1975; Seligsohn, 1977; Kay et al., 1978; Yamashita, 1998a; Lucas, 2004; Evans et al., 2007). Dental morphology, however, may be altered significantly through an animal's lifespan due to a variety of processes including dental disease, wear from physical interactions with food items, food

processing and masticatory behaviors (including culture among humans), dental morphology and microstructure, chemical erosion and/or physical damage resulting from exposure to high forces (Molnar, 1971, 1972; Rensberger, 1973; Gahram and Burkart, 1976; Smith et al., 1977; Smith, 1984; Lanyon and Sanson, 1986; Janis and Fortelius, 1988; Teaford and Oyen, 1989; Hillson, 1996; Gandara and Truelove, 1999; Maas and Dumont, 1999; Verrett, 2001; Kaifu et al., 2003; Lucas, 2004; Lussi et al., 2004; Nussey et al., 2007). Severe wear that ablates critical functional features of the tooth, and/or tooth loss which results in the failure of occlusion between opposing positions, is presumed to hinder the breakdown and subsequent utilization of food items thus negatively impacting aspects of individual health (Gipps and Sanson, 1984), life history, reproduction (e.g., Logan and Sanson, 2002; King et al., 2005), and survival (Lucas, 2004).

A key aspect of dental function is the reduction of foods from their initial form into particles of decreased size. Breaking down food into particles permits individuals to consume food items which are too large to swallow in their original state and/or may permit access to resources that would otherwise pass through the digestive tract unutilized [(e.g., seeds) Lucas, 2004]. Particle size may also play a role in the efficient digestion and utilization of food items as particle size corresponds to the surface area to volume ratio of ingested foods. Particles of relatively smaller size demonstrate an increased surface area to volume ratio in comparison to particles of relatively larger size, and therefore present a larger surface on which digestive enzymes and gut bacteria may act. Individuals who reduce foods into relatively smaller-sized particles should thus digest and utilize foods more effectively than those who reduce foods into larger particles (McLeod and Minson, 1969; Bjorndal et al., 1990; Lentle et al., 2003; Lucas, 2004).

The impact of dental morphology on masticated food particle size distributions, and the impact of food particle size upon subsequent digestion and utilization of food items, has been demonstrated for a number mammalian species (Sheine and Kay, 1977; Kay and Sheine, 1979; Sheine, 1979; Gipps and Sanson, 1984; Lanyon and Sanson, 1986; Bjorndal et al., 1990; Lentle et al., 2003). Experiments by Bjorndal et al. (1990) demonstrate, that when digested *in vitro* in inoculant drawn from cattle rumen, grass particles of smaller size digest more completely than do particles of comparatively greater size, and also that grass particles 3mm in size digest at a greater rate (1.7x) than 30mm-sized particles when placed in the rumen of cattle. Likewise, in their study of four wallaby species, Lentle et al. (2003) noted that reduced food particle size was associated with enhanced organic matter, nutrient detergent fiber and acid detergent fiber digestibility. Additionally, among browser and browser/grazer wallaby species, ingested food particle sizes were of larger size than those found among grazer and grazer/browser species, suggesting predominantly herbivorous species must more finely reduce their diets to ensure effective utilization of resources than those which persist on a less herbaceous diet (see below). Similarly, Gipps and Sanson (1984) noted that, when fed a controlled diet, ring-tailed possums (*Pseudocheirus* sp.) with significant dental wear showed reduced neural-detergent fiber and dry matter digestion, as well as weight loss. These authors also reported that individuals with experimentally ablated dentitions had greater amounts of large food particles and vascular bundles within their stomachs, but did not demonstrate weight loss, reduced digestibility, or differences in ingested food particle size, suggesting increased intake of digestible materials. Likewise, in koalas, Lanyon and Sanson (1986) noted significant tooth wear was associated with larger particle sizes in the stomach and caecum in comparison to individuals with lower wear.

While similar datasets are limited among nonhuman primates, Sheine and Kay (1977) noted that insectivorous *Galago senegalensis* demonstrated smaller food particles in their feces than did more frugivorous *Cheirogaleus medius.* These authors suggested that the reduction of foods into finer particles by *G. senegalensis* reflects the higher proportion of insects consumed by this species and the need to break down chitinous exoskeletons into small fragments towards their efficient enzymatic digestion. This presumption is supported by later experimental work (Kay and Sheine, 1979) demonstrating that galagos more efficiently digested chitin of smaller particle sizes than they did chitin particles of larger particles. Likewise, Sheine (1979) also reported that *Lemur catta* and *Eulemur fulvus* sbsp. digested greater amounts of dietary cellulose when presented in experimental diets in small particles (0.425-0.710mm) than when provided in larger sized particles (0.850-1.70mm), suggesting these species demonstrate adaptations suited towards breaking down and utilizing fibrous material, but also highlighting that particle size may play a role in the assimilation of such resources.

The extent to which particle size impacts digestion is also contingent, in part, upon the nature of the diet consumed. For instance, food particle size should be of great importance for diets high in structural carbohydrates, particularly for herbaceous materials of which cellulose-based cell walls cannot be digested by mammals without microbial action within the gut (e.g., Van Soest and McQueen, 1973; Stevens, 1988; Mackie, 2002). Reducing foods into particles of small size may be of less importance than previously assumed in the digestion of herbaceous materials, provided that consumers can cause numerous cracks and fissures through the indigestible cuticle of larger sized particles. Such cracks may effectively increase the relative surface area of otherwise large-sized particles (Bjorndal et al., 1990). In contrast, fruit materials may require less breakdown than do herbaceous materials as these are comparatively easy to

digest. In such cases thoroughly breaking down fruits may, however, be beneficial as this permits access to resources stored within cell walls and which would otherwise be more difficult to utilize. Despite these caveats, all things being equal, individuals who maintain an ability to reduce foods into particles of smaller size will demonstrate enhanced digestive efficiency and utilization of ingested resources than those individuals who do not (e.g., Lucas, 2004).

Dental Impairment in Bezà Mahafaly Lemur catta. *Lemur catta* living in and around Parcel 1 of the Bezà Mahafaly Special Reserve, Madagascar, show exceptionally high rates of tooth wear and frequencies of wear-related tooth loss, with 26.5% of individuals exhibiting the total loss (e.g., complete ablation of the crown) of at least one tooth and 6.4% of all assessed positions being absent. This is the highest reported frequency of tooth loss for any sample of extant nonhuman primates (Cuozzo and Sauther, 2006a). The loss of multiple teeth for individuals is common, with 10.8% of lemurs showing >30% loss, and 4.6% with >50% tooth loss. The highest loss observed in a living individual is 81% (Cuozzo and Sauther, 2004, 2006a).

The high frequencies of wear-related antemortem tooth loss observed among BMSR *L. catta* results primarily from the consumption of tamarind fruit (*Tamarindus indica*). Ripe tamarind fruit is a dominant dietary component during the dry season for ring-tailed lemurs living in semi-deciduous riverine gallery forest at BMSR and serves as an important fallback food resource during this period as the availability of alternative food items is limited due to seasonal effects (Sauther, 1992, 1998; Ratsirarson et al., 2001; Cuozzo and Sauther, 2004, 2006a,b; Simmen et al., 2006; Cuozzo et al., 2008; Sauther and Cuozzo, 2009). This fruit is exceptionally difficult for *L. catta* to consume as ripe tamarind is physically the largest food item eaten at BMSR, has a casing (exocarp) which is harder and tougher than any other food eaten by *L. catta* at BMSR and

also possesses tough fibers surrounding edible portions of the fruit (Cuozzo and Sauther, 2006a,b; Yamashita, 2000, 2008b; Yamashita et al., 2012). Ring-tailed lemur dental morphology appears to be poorly-adapted to breaking down diets in which large, hard and tough foods compose a major proportion of items consumed. In contrast to hard-object feeding adapted species, which frequently demonstrate blunt cusp morphology and comparatively thick enamel, ring-tailed lemurs possess dental morphology characterized by elongated shearing crests and enamel which is among the thinnest observed for any extant primate taxa (e.g., Shellis et al., 1998; Godfrey et al., 2005). Ring-tailed lemur dental morphology thus appears to be adapted towards a more folivorous diet than is consumed at BMSR (Kay et al., 1978; Dumont, 1995; Yamashita, 1998a; Cuozzo and Sauther, 2004, 2006a,b; Lambert et al., 2004; Godfrey et al, 2005; see also review in Cuozzo and Yamashita, 2006). Tooth wear and loss thus appears to result from using a comparatively thin-enameled and crested tooth to process the hard and tough casing of the tamarind fruit and scraping the fruit's tough internal fibers across the tooth during consumption (Cuozzo and Sauther, 2004, 2006a,b; Cuozzo et al., 2008; Sauther and Cuozzo, 2009; Yamashita et al., 2012).

Severe tooth wear and loss is particularly common for postcanine tooth positions directly involved in tamarind fruit processing, with the first molars being the most frequently lost position, followed by (in descending order) P3, P4 and M2. Because M1 is the first permanent position to erupt, the high rate of M1 loss likely reflects an interaction between eruption schedule and tamarind consumption, which corresponds with and/or begins soon after weaning (Sauther et al., 2002; Cuozzo and Sauther, 2004, 2006a). The association between loss and eruption schedule is, however, weakly linked for other tooth positions. For example, the P3 and P4 positions commonly used in tamarind fruit processing are more frequently absent than are earlier-erupting

positions which are used less during tamarind fruit processing (e.g., I1, I2, and M2). High rates of dental wear are also noted in the deciduous dentition of subadults (which demonstrate adult patterns of tamarind processing) further highlighting the role of tamarind in the generation of tooth wear and loss observed for BMSR *L. catta* and indicating that wear commences and proceeds rapidly from an early age (Cuozzo and Sauther, 2006b; Sauther and Cuozzo, 2009).

Behavior, Tooth Loss and Food Particle Size. Although dental impairment is presumed to reduce the efficiency with which foods are processed and utilized, BMSR ring tailed lemurs have been observed to adjust their behaviors to compensate for tooth loss (Millette et al., 2009). Individuals with tooth loss show divergent food processing behaviors in comparison to those without tooth loss. For example, these individuals demonstrate longer tamarind fruit feeding bout lengths than do individuals without tooth loss, but do not show longer feeding bouts when consuming less-challenging foods which do not require extensive initial processing (e.g., small *Tarenna pruinosum* berries which are swallowed whole without processing). These data suggest that lemurs with tooth loss may be less capable at consuming tamarind fruit than individuals without tooth loss (Millette et al., 2009). As tamarind fruit is a top fruit resource during the dry season at BMSR, the relative proportion of tamarind in the feces of individuals without tooth loss may be greater than those without loss as these lemurs may more easily access and consume greater quantities of this fruit per feeding bout.

Ring-tailed lemurs, however, may alter their behaviors to compensate for reduced tamarind fruit processing abilities. For example, individuals with tooth loss have been observed to engage in higher frequencies of licking behavior when consuming tamarind than individuals without tooth loss. Individuals have also been observed to manually process tamarind fruit and

consume tamarind pods previously opened and discarded by other group members. Examination of excreted food particles may help understand the relationship of these behaviors towards the compensation for tooth loss, particularly that directed towards consumption of tamarind fruit. If these behaviors are fully compensatory, the relative proportion of fruit material within the feces of these individuals may not differ from those without tooth loss. As such, examining the relative proportion of fruit material within the feces of individuals with dental impairment in comparison to those without dental impairment may test the efficacy of these behavioral adjustments.

Study Goals and Hypotheses. In this paper we examine the relationship between dental impairment and 1) fecal particle size as an indicator of ingested food particle size, and 2) the relative proportion of leaf matter to fruit matter with respect to fecal particle size in ring-tailed lemurs of the Bezà Mahafaly Special Reserve. We hypothesize that individuals with dental impairment will demonstrate increased fecal food particle size in comparison to those without dental impairment. We also hypothesize that individuals will demonstrate divergent patterns of processing for different types of food (e.g., fruits vs. leaves). We suggest that dentally impaired individuals will demonstrate reduced proportions of fruit material within their feces as these individuals are expected to demonstrate reduced efficiency when processing tamarind fruit.

Methods.

Study Site. The Bezà Mahafaly Special Reserve (BMSR) (23˚30’S, 44˚40’E) is located in southwest Madagascar approximately 35km northeast of the town of Betioky Sud. The reserve consists of two protected parcels, an annex connecting the two parcels, and a research camp. This study was conducted using samples procured from *L. catta* living in or adjacent to Parcel 1,

an 80ha western Malagasy dry semi-deciduous gallery forest located on the western bank of the Sakamena River (Sauther et al., 1999; Ratsirarson, 2003). Since 1979 Parcel 1 has been relatively free from anthropogenic impacts (e.g., logging or livestock grazing) by both a barbed-wire fence and an agreement by the local Mahafaly population to not graze their animals within the reserve. The eastern portion of Parcel 1 is a mature riparian deciduous and semi-deciduous forest which progressively becomes more xerophytic in western areas away from the river (Sauther, 1998; Ratsirarson, 2003). This gallery forest is dominated by tamarind (*Tamarindus indica*), particularly on the parcel's wetter eastern portion. Eastern portions of the reserve are characterized by a closed canopy of roughly 15-20m in height and consisting primarily of tamarind, *Acacia rouvmae, Euphorbia tirucalli,* and *Salvadora angustifolia*. Below this canopy layer, and in western portions of Parcel 1, most trees are relatively short in height ranging from 2 to 15m. Dominant tree species in a given area of the Parcel 1 tends to follow the decreasing east-west gradient of water availability, although tamarind and *Azima tetracantha* are found throughout Parcel 1 (Sussman and Ratsirarson, 2006).

Unprotected gallery forest covering approximately 200ha is located contiguously to the north and south borders of Parcel 1 (Sussman and Ratsirarson, 2006). These areas demonstrate reduced productivity and degradation of the forest understory due to grazing and farming (Sussman and Rakotozafy, 1994; Sauther, 1998; Sauther and Cuozzo, 2009). The quantity and quality of understory lianas and herbs available for use by ring-tailed lemurs is reduced within these areas in comparison to Parcel 1 (Sussman and Rakotozafy, 1994; Sauther, 1998; Sauther and Cuozzo, 2009). Located immediately south of Parcel 1, the camp area consists of several small administrative and research buildings, a small museum and camping facilities for researchers and visitors. These facilities include a kitchen, a closed water well, and open trash

pits that are sometimes raided by several lemur groups (Sauther et al., 2006; Fish et al., 2007; Millette, personal observations). While lemurs have been observed to procure both food and water from these sources, BMSR ring-tailed lemurs are not provisioned. Ring-tailed lemurs range through and utilize all aforementioned areas irrespective of anthropogenic alterations, with several social groups using both protected and disturbed areas (Sauther et al., 2006; Fish et al., 2007).

Dental Assessment and Capture Protocol. Individual tooth wear and loss data were determined by F.P.C. for 33 BMSR ring-tailed lemurs during annual health assessments (see capture and assessment protocol below) conducted during the 2006, 2007 and 2008 field seasons. Tooth loss was recorded only for positions where the crown was worn to the gumline and/or was totally missing, leaving at most only worn roots (e.g., Cuozzo and Sauther, 2006a; Cuozzo et al., 2010). Such positions are not in occlusion and represent a total loss of tooth function. Of the 33 study subjects, seven individuals were found to be missing at least one tooth, with total tooth loss ranging from 8% to 56%. Of these, all but one individual demonstrated >10% tooth loss (e.g., heavy loss). When limited to the postcanine dentition, tooth loss ranged from 8% to 79%. For this study, postcanine dentition is defined as maxillary tooth positions distal to the canine, and mandibular positions distal to the lower second premolar, as this tooth canine-like in form and function (e.g., Cuozzo and Yamashita, 2006). To provide an indicator of individual tooth wear status, a wear index was calculated. For each individual, all tooth positions were scored using a 0 to 5 ordinal wear scale (0 representing a low-wear state, 5 representing the total ablation of the tooth to the gumline, see Table 6.1 and Figure 6.1). Wear indices were determined by dividing the sum of wear scores by the number of teeth present for 1) the total dental arcade and 2) for the

postcanine dentition. Total wear indices ranged from 0.500 to 4.750, while postcanine wear ranged from 0.500 to 4.733. A complete listing of tooth wear and loss statistics for individuals used in this study is available in Table 6.2.

Table 6.1. Tooth Wear Ordinal Scale.

0 -- unworn occlusal surface
1 -- small wear facets and no dentine or pulp exposure
2 -- large wear facets and no dentine or pulp exposure
3 -- some dentine and pulp exposure, few cusps still present; for canine and tooth comb, 1/2 remaining
4 -- pulp exposure, with cusps gone, dentine or pulp exposed across most of the surface, or partial crown remaining; for canine and toothcomb, less than 1/4 remaining
5 -- tooth worn to or below gum line with only roots/partial roots remaining (i.e., functional loss [see Cuozzo and Sauther, 2004, 2006a]); OR no presence of the tooth remains (i.e., healed gingiva only, or in skeletal specimens remodeled alveoli)

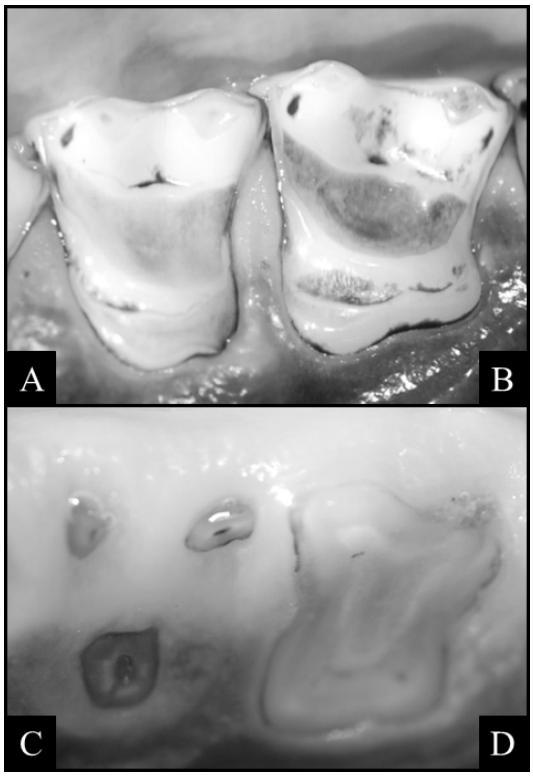


Figure 6.1. Tooth wear scores ranging from 2 to 5 upon the first and second maxillary molars for two living BMSR *L. catta.* The top photo shows wear scores of 2 (B) and 3 (A). The bottom photo shows wear scores of 4 (D) and 5 (C).

Table 6.2. Individual Tooth Wear and Loss Status by Year.

Animal ID	Capture Year	Age at Capture*	Tooth Loss Status	% Total Missing	% Postcanine Missing	Total Wear Index	Postcanine Wear Index
104	2006	JUV	NO	0.00	0.00	0.50	0.05
104	2007	SA	NO	0.00	0.00	0.97	1.14
105	2006	YA	NO	0.00	0.00	1.11	1.18
106	2006	AD	NO	0.00	0.00	2.28	2.91
168	2006	AD	NO	0.00	0.00	1.36	1.59
168	2007	AD	NO	0.00	0.00	2.00	2.18
211	2007	SA	NO	0.00	0.00	0.94	1.00
212	2007	AD	NO	0.00	0.00	2.20	2.46
227	2007	AD	NO	0.00	0.00	1.25	1.36
253	2006	YA	NO	0.00	0.00	0.75	0.73
253	2006	YA	NO	0.00	0.00	0.75	0.73
253	2007	AD	NO	0.00	0.00	1.44	1.46
259	2007	AD	NO	0.00	0.00	1.64	2.05
263	2006	AD	NO	0.00	0.00	1.64	2.05
264	2007	YA	NO	0.00	0.00	1.06	1.09
271	2006	SA	NO	0.00	0.00	0.53	0.86
271	2007	YA	NO	0.00	0.00	1.19	1.32
275	2006	SA	NO	0.00	0.00	0.76	1.09
276	2007	SA	NO	0.00	0.00	0.94	1.00
280	2007	AD	NO	0.00	0.00	1.64	2.05
282	2007	SA	NO	0.00	0.00	0.78	1.00
284	2007	YA	NO	0.00	0.00	1.19	1.32
286	2007	.	NO	0.00	0.00	0.50	0.50
295	2007	AD	NO	0.00	0.00	3.00	3.64
297	2008	YA	NO	0.00	0.00	1.11	1.18
298	2007	AD	NO	0.00	0.00	1.53	1.86
299	2007	.	NO	0.00	0.00	0.50	0.50
300	2006	AD	NO	0.00	0.00	1.22	1.36
300	2007	AD	NO	0.00	0.00	1.53	1.86
300	2008	AD	NO	0.00	0.00	1.72	2.18
308	2008	SA	NO	0.00	0.00	0.88	1.00
316	2008	SA	NO	0.00	0.00	0.88	0.91
130 (330)	2007	AD	NO	0.00	0.00	1.67	1.96
6	2007	OA	YES	28.00	45.83	4.00	4.46
138	2006	OA	YES	44.00	75.00	4.53	4.68
138	2007	OA	YES	56.00	79.17	4.75	4.77
156	2006	OA	YES	11.00	8.33	3.44	3.64
156	2007	OA	YES	11.00	8.33	3.76	3.91
226	2006	OA	YES	22.00	29.17	3.84	4.09
226	2007	OA	YES	33.00	41.67	4.29	4.41
293	2007	AD	YES	8.00	12.50	3.28	3.91
459	2007	OA	YES	19.00	25.00	3.94	4.18
488	2007	OA	YES	22.00	33.33	4.14	4.36

All captures were conducted as part of a long-term study (since 2003) of ring-tailed lemur health, behavior and life history conducted by M.L.S. and F.P.C. (see Cuozzo and Sauther, 2004; 2006a,b; Sauther and Cuozzo, 2009). All lemurs were captured using a Dan-Inject blow dart system (Dan-Inject, North America, Fort Collins, Colorado, USA) in conjunction with the drug Telazol® (Fort Dodge Laboratories, Fort Dodge, Iowa, USA). Darting, capture and examination protocols followed methods developed over 20 years of research and over 400 safe captures at BMSR (e.g., Sussman, 1991; Sauther et al., 2002, 2006; Cuozzo and Sauther, 2006a,b; Miller et al., 2007; Larsen et al., 2011) During each examination, a veterinarian and veterinary students were present to monitor the health of the captured lemur. All methods and materials received approval by and followed standard animal handling guidelines and protocols of the Institutional Animal Care and Use Committees (IACUC) of 1) the University of North Dakota, and 2) the University of Colorado. All data collection and captures were approved by MNP (Madagascar National Parks, formerly ANGAP), the body governing research in Madagascar's protected areas.

Fecal Methods. Forty-three fecal samples were collected during the dry seasons (June-August) of 2006, 2007 and 2008 from 33 study subjects. Ten fecal samples were collected from individuals demonstrating tooth loss, while 33 samples were collected from individuals without tooth loss. Fecal samples were collected from excreta produced during annual health inspections, either while lemurs were being examined and assessed, or from the portable kennels in which the lemurs recover, following their release. Following collection, samples were washed with water through three sieves of decreasing size (11.2, 4.75 and 1mm), producing three sub-samples of the following sizes: 1.0-4.75mm, 4.75-11.2mm and >11.2mm. Seeds were not removed from the

sample, with most of these deposited in the 4.75mm and 11.2mm sieves (Figure 6.2). Samples were then dried and transported to the University of Colorado where the mass of each fraction was weighed. As samples collected during field observations were often impossible to collect in their entirety, and some of each sample <1mm in size was lost during sieving, each sieved proportion was assessed in relation to the total dry mass recovered following sieving. Each resulting mass proportion was then assessed in relation to individual wear and loss status using non-parametric methods (e.g., Mann-Whitney U and Spearman rho). Non-parametric methods were utilized as masticated food particle sizes are typically not normally distributed (see Lucas, 2004). For each test, statistical significance was set at the $\alpha = 0.05$ level.

Figure 6.2. Sieving methods. Food particle size fractions were produced by sieving each fecal sample through sieves of decreasing size (A). Each fraction was then dried individually before being weighed and assessed as a proportion of the total mass of recovered material (B). Image C demonstrates fecal materials trapped within the 11.2mm sieve, including tamarind seeds and leaf material.

To assess the potential relationship between tooth loss and the consumption of divergent food item types, for a 20-fecal sub-sample (10 from individuals with tooth loss, 10 from those without loss) the relative proportion of fruit to leaf mass was determined within each size fraction. For each sieve, fruit material and leaf material were manually separated. These proportions were then weighed and assessed in relation to their total mass. The relative proportions of these materials within each sieve size were then assessed in relation to tooth loss status using non-parametric methods (e.g., Mann-Whitney U). Statistical significance was set at $\alpha = 0.05$ level. While leaf material (e.g., undigested leaf portions, leaf vasculature, stems) was easily distinguished from fruit material (e.g., seeds, kily shell, remnants of fruit "meat") it was not possible to reliably distinguish the species of each leaf, although several fruit species were readily identifiable (e.g., *E. pruinosum* and *T. indicia*). As species could not be reliably determined for leaves, a simple leaf/fruit distinction was utilized to assess the relationship between dental impairment and the relative proportion of dietary components within each sieved portion. Insect material was not observed, and thus not considered, when assessing fecal contents.

Results.

Dental Impairment and Particle Size. Tooth loss status was associated with fecal particle size. Individuals with tooth loss demonstrated relatively smaller proportions of material 1.0-4.75mm in size than did individuals without tooth loss. This pattern also holds true for individuals with greater than 10% tooth loss, and also when assessed with respect to the postcanine dentition. Unexpectedly, no differences were found between individuals with tooth loss and without tooth loss for material recovered in the 4.75-11.2mm or >l1.2mm fractions. This pattern remains when

examined in terms of the postcanine dentition and/or limited to individuals with greater than 10% tooth loss (Table 6.3, Figure 6.3).

Table 6.3. Mean Percentage of Total Fecal Mass for Each Sieved Fraction in Relation to Tooth Loss Status.

Tooth Loss (N = 10)	22.4	7.5	47.9	12.3	29.7	12.6
No Tooth Loss (N = 33)	36.6	16.3	42	11.7	21.3	15.2
Mann-Whitney U-score	75		114		110	
P-value =	***0.0097***		0.1426		0.1127	
>10% Tooth Loss (N = 9)	21.5	7.3	48.6	12.2	29.9	13.3
<10% Tooth Loss (N = 34)	36.5	16.1	42	12.1	21.5	15
Mann-Whitney U-score	62		111		104	
P-value =	***0.0066***		0.6217		0.1422	
Postcanine Loss (N = 10)	22.4	7.5	47.9	12.3	29.7	12.6
No Postcanine Loss (N = 33)	36.6	16.3	42	11.7	21.3	15.2
Mann-Whitney U-score	75		114		110	
P-value =	***0.0097***		0.1426		0.1127	

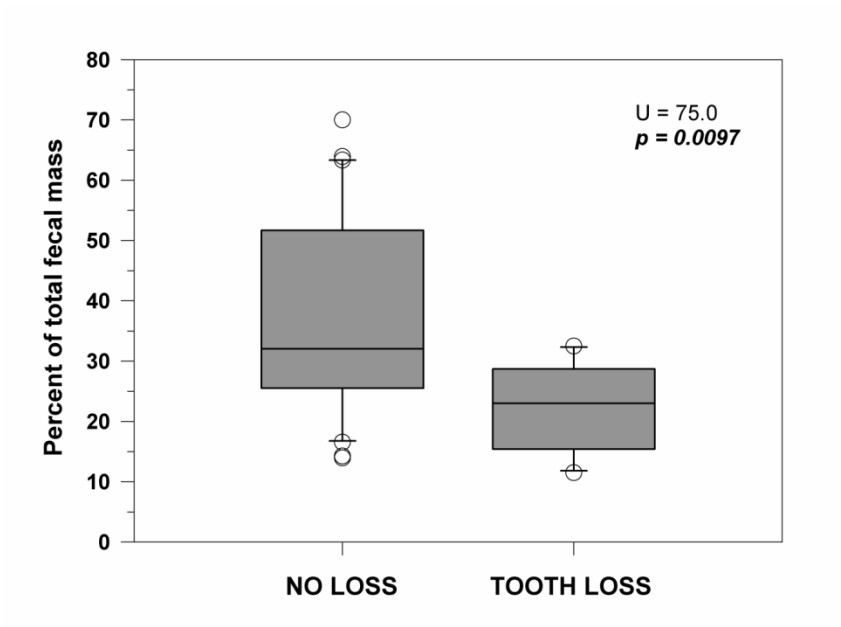


Figure 6.3. Percentage of total fecal mass accounted for by the 1.0-4.75mm sieve fraction based upon tooth loss status. Individuals without tooth loss demonstrate significantly greater proportion of their fecal materials within this sample than do individuals without tooth loss. No differences were noted for either the 4.75-11.2mm or >11.2mm fractions between individuals with and without tooth loss.

When particle sizes were examined with respect to tooth wear indices, increasing wear was associated with a smaller proportion of particles in the small 1.0-4.75mm proportion. When limited to postcanine wear, significantly more particles were also present in the large >11.2mm sample with increasing wear scores. When examined with respect to tooth wear indices, patterns observed with respect to tooth loss status hold remarkably well. This is not unexpected as individuals with extensive tooth loss demonstrate significantly higher rates of tooth wear, as wear is the primary modality of tooth loss in this population (e.g., Cuozzo and Sauther, 2006a). However, when assessed by excluding individuals without tooth loss, but demonstrating tooth wear, no significant patterns for any of the three size categories were present (Table 6.4 and Figure 6.4).

Table 6.4. Relationship Between Tooth Wear Status and Sieved Fecal Food Particle Size.

		Sieved Fraction Size					
		1.0 - 4.75mm		**4.75 - 11.2mm**		**>11.2mm**	
Tooth Wear Status	**n =**	***rho* =**	***p* =**	***rho* =**	***p* =**	***rho* =**	***p* =**
Total Wear	43	-0.379	***0.014***	0.257	0.095	0.282	*0.067*
Postcanine Wear	43	-0.387	***0.012***	0.244	0.114	0.307	***0.047***
Total Wear (No Loss)	33	-0.145	0.411	0.182	0.303	0.151	0.393
Postcanine Wear (No Loss)	33	-0.174	0.326	0.169	0.339	0.187	0.289

Tooth loss is associated with greater proportions of leaves and reduced proportions of fruit within the 1.0-4.75mm sample and the >11.2mm sample. A similar but non-significant pattern is also present in the 4.75-11.2mm sample. These data indicate that leaves are present in greater quantities in the feces of individuals with tooth loss, and suggest these individuals ingest fewer fruits than do individuals without tooth loss during BMSR's dry season (Figure 6.5 and Table 6.5).

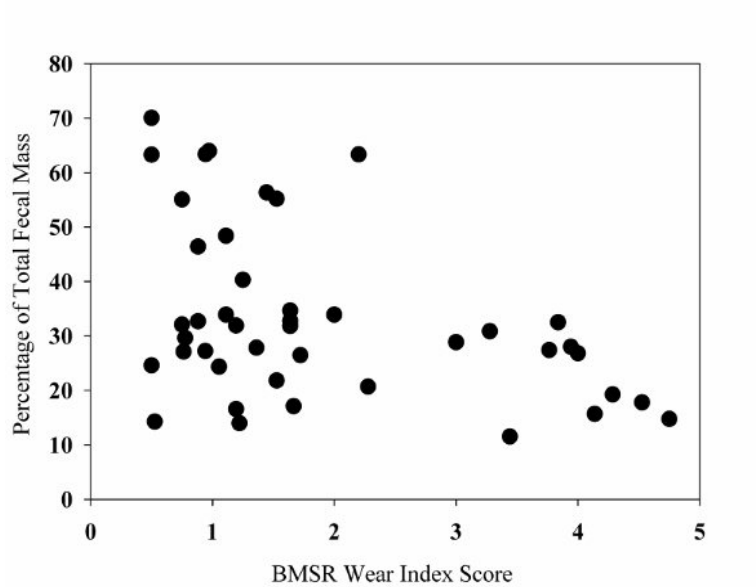


Figure 6.4. The relationship between tooth wear status and fecal food particle size for the 1.0-4.75mm sized sample. Increasing tooth wear status is associated with a reduced proportion of their fecal material within this fraction, while lower tooth wear is associated with a greater proportion of material sized 1.0-4.75mm.

Table 6.5. Type of Food Within Each Sieved Fraction by Tooth Loss Status.

Sieve Fraction Size	Loss Status	n =	% Fruit	*sd*	% Leaf	*sd*
1.0 - 4.75mm	**Tooth Loss**	10	23.9	24.6	76.1	24.6
	No Tooth Loss	10	49.7	29.1	50.3	29.1
	U-score = 22	**p = *0.034***				
4.75 - 11.2mm	**Tooth Loss**	10	49.3	28.8	50.7	28.8
	No Tooth Loss	10	75.7	28.4	24.3	28.4
	U-score = 25	p = *0.063*				
>11.2mm	**Tooth Loss**	10	62.8	39	37.2	39
	No Tooth Loss	8*	99.6	0.7	0.4	0.7
	U-score = 15	**p = *0.018***				

* No >11.2mm sized particles were present for two individuals without tooth loss.

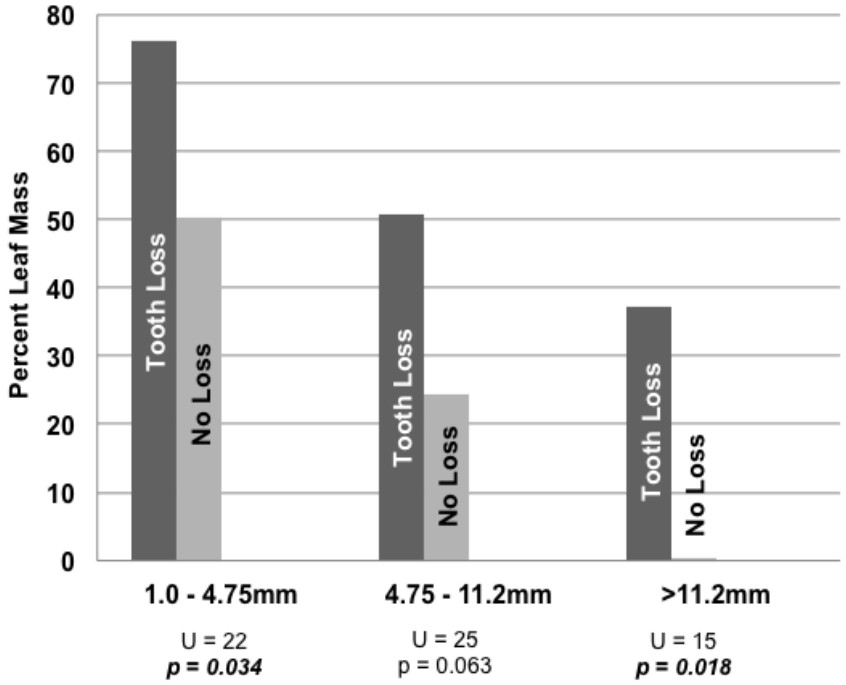


Figure 6.5. Percentage of leaf material within each fecal sieve size for individuals with and without tooth loss. Individuals with tooth loss demonstrated increased proportions of leaf material within their fecal material than did individuals without tooth loss for both the 1.0-4.75mm and >11.2mm fractions than did individuals without tooth loss. These data suggest that individuals with tooth loss ingest relatively lower proportions of fruit material than do individuals with without tooth loss.

Discussion.

Dental Impairment and Food Processing. Our data indicate a link between dental impairment and fecal food particle size. Individuals with tooth loss demonstrate significantly higher proportions of material in the 1.0-4.75mm sample. Wear index was also associated with fecal particle size for the 1.0-4.75mm sample, and also the >11.2mm sample when examined in terms of postcanine wear. Therefore, fecal particle size provides a means for assessing the impact of dental impairment on an individual's ability to process and utilize food items among ring-tailed lemurs. Individuals with significant dental impairment, such as severe tooth wear and/or tooth loss, demonstrate a reduced ability to break down foods into particles of small size in comparison

with those with lower amounts of dental impairment. Given the link between food particle size and the effective utilization of food items, these lemurs may demonstrate a comparatively lower capacity to digest and harness nutrients of foods consumed. This may be especially true in terms of capacity to utilize leaves, which appear to be a major dietary component for lemurs with tooth loss in this study and given that food particle size-related loss of cellulose digestion has been reported for *L. catta* by Sheine (1979).

No differences were found between individuals with and without tooth loss for the 4.75-11.2mm or >11.2mm categories. One explanation for these data is that the generation of particles in these size ranges is attainable regardless of tooth loss status, although further breakdown is not readily possible for dentally-impaired individuals. A second explanation is that dentally-impaired individuals may be selecting items which more easily fragment than do those without impairment. We suggest, however, that individuals with tooth loss or extensive wear do not divergently select food items as *L. catta* feed predominantly in a synchronous manner where all members of a social group consume a single species or type of food simultaneously (Sauther et al., 1999) and tend to focus on a narrow variety and limited number of food species as they become seasonally available (Sauther, 1998). Patterns of food consumption should thus be similar between individuals irrespective of tooth loss status. Additionally, if impaired individuals are selecting foods based on ease of breakdown, their observed particle size distributions should be similar to those without impairment. As this is not the case, it appears that dentally-impaired individuals comparatively are less capable of reducing food items into particles of small size.

Individuals with tooth loss demonstrate relatively higher proportions of leaves and reduced proportions of fruit in their fecal matter. One explanation for these findings is that individuals may be selectively consuming leaves in lieu of fruit. Alternatively, these data may

reflect an inability to access fruit resources, particularly tamarind fruit, which is a primary dry-season fall back food that is exceptionally challenging to process (e.g., Sauther and Cuozzo, 2009). Dentally-impaired individuals may not be able access tamarind as efficiently as those without dental impairment, thus reducing the relative percentage of fruit within each fecal sample. This interpretation is supported by observations that individuals with tooth loss demonstrate difficulty opening tamarind pods, and exhibit longer feeding bout lengths for tamarind fruit (Millette et al., 2009, personal observations). Given that herbaceous material is of comparatively lower quality than the ripe tamarind fruit consumed during the dry season, which is a high quality food rich in sugar (see Yamashita, 2008a), and observations that dentally-impaired individuals do not as finely comminute leaf material (see Figure 6.5), it is likely that they show reduced energetic intake in comparison to individuals without dental impairment.

When limited to individuals without tooth loss, tooth wear indices are not related to food particle size. These data suggest that tooth wear alone does not result in reduced masticatory abilities for ring-tailed lemurs at BMSR and may provide support for the hypothesis that teeth have evolved in a manner where functionality is maintained despite the presence of wear. As primate teeth do not regenerate or remodel their unworn morphology, they should evolve in a manner which permits the efficient breakdown of diets consisting of food items of specific physical and structural properties through wear (Rensberger, 1973; Kay, 1981; Teaford, 1983; Lanyon and Sanson, 1986; Ungar and M'Kirera, 2003; Dennis et al, 2004). For example, among folivorous species, differential wear between enamel and dentin may result in the maintenance of dental function through the exposure of shearing crests where dentin ablates at a higher rate than does enamel (Rensberger, 1973; Lanyon and Sanson, 1986; King et al., 2005).

Several researchers have demonstrated that functional aspects of primate dental morphology are maintained through the course of wear. For example, using three-dimensional GIS-based topographic methods on worn teeth of chimpanzees and gorillas, Ungar and M'Kirera (2003; see also M'Kirera and Ungar, 2003) found that despite reductions in overall slope and relief with wear, these species remain constant in their topographic angularity, a measure of the average change in slope of the tooth associated with the breakdown of tough material. In this case, gorillas maintained comparatively higher angularity through the course of wear than did chimpanzees, consistent with their (in comparison to chimpanzees) tougher and more folivorous diet. As such, these authors suggest that angularity represents a means through which dental function is maintained despite the onset of wear. Similar maintenance of angularity until extreme wear has been reported for howling monkeys by Dennis et al. (2004). Likewise, King et al. (2005) found that extreme dental wear was associated with increased maternal infant mortality among *Propithecus edwardsi* during periods of low rainfall, although individuals with lower wear did not demonstrate rainfall-linked patterns of reduced infant survival.

Ring-tailed lemurs at BMSR may demonstrate a similar maintenance of dental function through the course of wear as, when limited to individuals without tooth loss, increasing wear status is not associated with the reduction of foods into larger sized particles. As a result, wear alone appears insufficient to limit an individual's ability to efficiently masticate foods into particles. The finding that wear status is associated with increasing proportions of larger-sized food particles when individuals with tooth loss are included is not unexpected, as loss represents the total elimination of function for a given tooth position. Nevertheless, the hypothesis that dental function is maintained despite wear for BMSR *L. catta* remains speculatory. Individuals without tooth loss may simply not possess wear of sufficient severity to impact particle size.

Individuals with extensive wear also likely show comparatively higher levels of wear than do individuals without tooth loss. These data do, however, provide initial evidence that dental function is maintained despite wear among BMSR lemurs prior to the onset of tooth loss.

Behavior and Dental Impairment. Although food particle size appears related to dental impairment in BMSR ring-tailed lemurs, the impact of dental impairment upon these individuals is not yet fully understood. As we have discussed elsewhere (Millette et al., 2009), ring-tailed lemurs compensate for tooth loss through behavioral mechanisms related to food processing. Individuals with tooth loss may gain access to foods by simply spending more time processing each individual food item. Also, individuals have been observed processing foods using alternative means, including non-dental food processing such as licking food items and/or use of the hands. Alternatively, individuals with dental impairment utilize foods that have been previously processed and discarded by others (Cuozzo and Sauther, 2006a). Individuals with tooth loss, particularly extensive tooth loss, have also been observed to remain active during periods of group inactivity and resting during the early afternoon, using this period instead to feed, forage and groom (Millette et al., 2009).

Individuals may also compensate for dental impairment using strategies that have not yet been investigated. For example, dentally-impaired individuals may preferentially select food items which are less challenging to process and consume. Such alterations to the diet may explain differences in the contents of fecal samples assessed during this study. If this is the case, fecal particle size may also be impacted by food choice. While the presence and direction of any such change remains speculatory, dental impairment is still expected to result in the consumption of larger sized food particles for a given diet.

Our data indicate that dental impairment is associated with increased food particle sizes and reduced fruit intake. Behaviors associated with compensation for tooth loss do not appear to enhance the efficiency with which food items are broken down into particles, or to confer compensatory food processing abilities. Behavioral responses to tooth loss thus appear to reflect compensation through increasing overall food intake and/or access to foods, but likely do not impact the utilization of food resources once acquired, processed and ingested.

Conclusion.

Our data indicate that individuals with dental impairment demonstrate divergent patterns of fecal food particle size than do individuals without dental impairment. Tooth loss and wear appear to negatively impact an individual's ability to breakdown of food items and indicate that dental impairment leads to reduced masticatory performance. The presence of larger food particle sizes within the feces of individuals with dental impairment indicates that these individuals likely utilize ingested food items less effectively. The extent to which the increased particle size impacts ring-tailed lemur health, survival and reproduction is not yet known or well understood. Ring-tailed lemurs with extensive tooth loss living at BMSR, however, have been observed to survive for extended periods (e.g., survival for > 5 years in a near-edentulous state) while in good health, and do not demonstrate reduced reproductive success in comparison to those without tooth loss (Cuozzo and Sauther, 2004, 2006a,b; Cuozzo et al., 2010). The impact of tooth loss alone, and thus increased particle size, is not inextricably linked to individual health and survival in this population. We suggest that individuals with tooth loss attempt to compensate for this reduction of masticatory abilities through behavioral mechanisms, including alterations to activity budget and food processing behaviors (see Millette et al., 2009). These

alterations, however, do not appear to result in similar particle size distributions between individuals with and without dental impairment. These data indicate that behaviors used in compensation for dental impairment do not act through reducing food particle size. Rather, such behaviors likely increase overall access to foods and/or permit the initial breakdown foods, which could otherwise not be consumed

Data presented here also suggest that individuals with tooth loss demonstrate divergent patterns of food consumption, ingesting more leaves than they do fruit material. We suggest that this is potentially a function of reduced tamarind fruit processing capabilities among individuals with tooth loss, as this fruit is exceptionally difficult for *L. catta* to process and consume. Alternatively, these individuals may be emphasizing leaves over fruit, regardless of a reduced ability to comminute leaves into finely-sized particles and a presumably impaired capacity to extract nutrients from this resource. Although we as of yet have no feeding behavior data regarding patterns of leaf consumption among individuals with dental impairment, these individuals may be attempting to offset reductions in their ability to utilize this resource by increasing the total volume of leaves consumed. Such alterations to feeding behavior may be particularly prevalent if access to important alternative foods is restricted by dental impairment, as is likely the case with ripe tamarind fruit. Our ongoing work at BMSR will further clarify such aspects of food item choice and consumption in relation to dental impairment.

Tooth wear was not associated with increased particle sizes when individuals with tooth loss were excluded. These data indicate that tooth loss is a key element in the reduction of masticatory efficiency for individuals living at BMSR, and that dental function may be maintained through the course of wear prior to tooth loss. This interpretation supports the

hypothesis that masticatory function is maintained among individuals with tooth wear, and is only reduced when extensive wear and/or tooth loss is present.

In conclusion, our data suggest the following is true for ring-tailed lemurs at BMSR:

1) Tooth loss is associated with reduced proportions of small sized particles <4.75 mm in size, but is not associated with divergent patterns for particles 4.75-11.2mm or >11.2mm in size. As a result, individuals with tooth loss may less effectively process food items and may utilize foods less efficiently than do those without tooth loss.

2) Increasing dental wear is associated with fewer proportions of 1.0-4.75mm sized particles and, when limited to post canine wear scores, also greater proportions of large >11.2mm sized particles. No such differences are present when individuals with tooth loss are removed from the sample. Tooth wear alone may thus not result in a loss of dental function among BMSR *L. catta.*

3) Individuals with tooth loss demonstrate greater proportions of leaf material in their fecal material than do individuals without tooth loss, suggesting that individuals with tooth loss consume less fruit during the dry season at BMSR.

CHAPTER VII:

EXAMINING VISUAL MEASURES OF COAT AND BODY CONDITION IN WILD RING-TAILED LEMURS AT THE BEZÀ MAHAFALY SPECIAL RESERVE, MADAGASCAR.

Published previously in *Folia Primatologica* as:

Millette JB, Sauther ML, Cuozzo FP. 2015. Examining Visual Measures of Coat and Body Condition in Wild Ring-Tailed Lemurs at the Bezà Mahafaly Special Reserve, Madagascar. Folia Primatologica 86:44-55.

Introduction.

Measures of coat condition and body mass present an avenue for assessing general health status among nonhuman primates, but also reflect a variety of factors including individual reproductive state, social ecology, resource use and availability, and seasonality. Poor coat condition and/or hair loss may result from a number of potential sources including seasonality and aging, reproductive condition, nutritional and hormonal imbalances, social stress, parasites and infections, and diet. Although a range of variables may impact coat quality and/or result in hair loss, coat condition indices can present a valuable non-invasive measure of general health status, but may also provide insights into aspects of social and feeding ecology as well as reproductive state (Isbell, 1995; Pride, 2003; Beisner and Isbell, 2009; Jolly, 2009a,b; Novak and Meyer, 2009; Zhang, 2011; Borg et al., 2014). Similarly, measures of body mass can provide information about resource availability, nutritional and health status, and insight into reproductive condition. For example, body mass has been positively associated with increased access to resources (Eley et al., 1989; Altmann et al., 1993; Olupot, 1999; Borg et al., 2014), and

is also positively associated with reproductive outcomes (Bercovitch, 1987; Richard et al., 2000). Likewise, body condition may also vary seasonally due to reproductive condition (e.g., pregnancy and lactational status, seasonal timing of reproduction) and/or seasonal variation in food availability (Sauther, 1998; Richard et al., 2000; Lewis and Kappeler, 2005).

Among ring-tailed lemurs (*Lemur catta*), coat condition has been associated with seasonality, resource use, reproductive state, physiological stress and health status (Pereira et al., 1999; Pride, 2003; Crawford et al., 2006a,b; Junge and Sauther, 2006; Miller et al., 2007; Jolly, 2009a,b; LaFleur, 2012; Ichino et al., 2013b). Variation in body mass that is related to resource availability, seasonality and lactational status has also been reported for *L. catta* in both wild and captive settings (Pereira, 1993; Sauther, 1998; Ichino et al., 2013a). While these measures may be evaluated through direct veterinary examinations, visual scoring systems for body mass and coat condition provide a cost effective and less time consuming alternative. In this paper we present data using remote visual measures of coat condition and body condition related to body mass in ring-tailed lemurs from the Bezá Mahafaly Special Reserve (BMSR). Although visual measures of coat condition have been presented for *L. catta* from Berenty and Tsimanampetsotsa (e.g., Pride, 2003; Jolly, 2009a,b; LaFleur, 2012), to date no such data have been presented for BMSR ring-tailed lemurs. Likewise, to our knowledge, no researchers have published quantitative visual indices of body mass condition for wild ring-tailed lemurs, or for any other wild strepsirrhine primate. As such, in this paper we examine month-to-month variation for body size, coat condition and alopecia (hair loss). We also provide information on sex differences for each of these measures on a within-month basis, as well as data examining variability in monthly body condition between individuals with and without infants.

Materials and Methods.

Coat and body condition evaluations were conducted for 36 (23 female, 13 male) adult ring-tailed lemurs (*Lemur catta*) at the Bezá Mahafaly Special Reserve, in southwestern Madagascar (23.6667° S, 44.6000° E) from July 2012 to March 2013 as part of an ongoing long-term study (since 2003) of *L. catta* dental and general health. Individuals were drawn from 7 initial study groups, with observations being recorded in 9 groups following migration of 4 male study subjects over the course of observations. All individuals were identified using a numerical collar system (Cuozzo and Sauther, 2006; Sauther and Cuozzo, 2008). There were more females in the study population than males due to male immigration. J.B.M assessed coat condition, body condition and the presence of alopecia for each study animal ideally on a twice-monthly basis from July 2012 to February 2013. Bezá Mahafaly demonstrates a highly seasonal pattern of rainfall with the majority of precipitation falling during the wet season from between October-November to the end of March (Sauther, 1998). Therefore data presented herein were collected from the height of the dry season, until the end of the wet season. At BMSR, food resources available to *Lemur catta* track patterns of rainfall with reduced food availability occur during the height of the dry season and increased resource availability occurring during the wet season (Sauther, 1998; Millette, personal observations). BMSR *L. catta* demonstrate a seasonal pattern of reproduction with the majority of births occurring during the months of September and October (Sauther 1998; Millette, personal observations), and infant weaning commencing by March before the subsequent mating season [May - June (Sauther, 1998)]. Observations for study animals were occasionally, but rarely, made more than twice monthly. Likewise, on several occasions animals were only observed once during a month (e.g., the animal was too far away to be observed adequately, or could not be located), although this occurred infrequently

from July to February. A smaller subset of individuals (n = 29) was scored by J.B.M. once during March 2013.

Coat condition was scored using a 0-5 ordinal system (Table 7.1, Figure 7.1) derived from that of Berg et al. (2009), and which has been used for assessing *L. catta* at Berenty Private Reserve and Tsimanampetsotsa National Park (see Berg et al., 2009; Jolly, 2009a,b; LaFleur, 2012). Alopecia was documented using a Yes/No system, and was recorded if holes with missing hair were present in the coat. If hair regrowth had started where hair had been previously missing, alopecia was recorded if these patches were not predominantly filled by new growth. Body condition for each subject was recorded using a 0-4 ordinal system based on that designed for quantifying relative body fat in *Macaca mulatta* by Berman and Schwartz (1988) and adapted for use in *L. catta* (Table 7.2, Figure 7.2). Our system represents a general measure of relative body mass (e.g., from extremely thin to obese), although we use the term "body condition" as study animals were not weighed directly. While we did not directly compare body condition scores between BMSR lemurs and rhesus macaques, Pereira and Pond (1995) found patterns of fat deposition to be similar between both taxa.

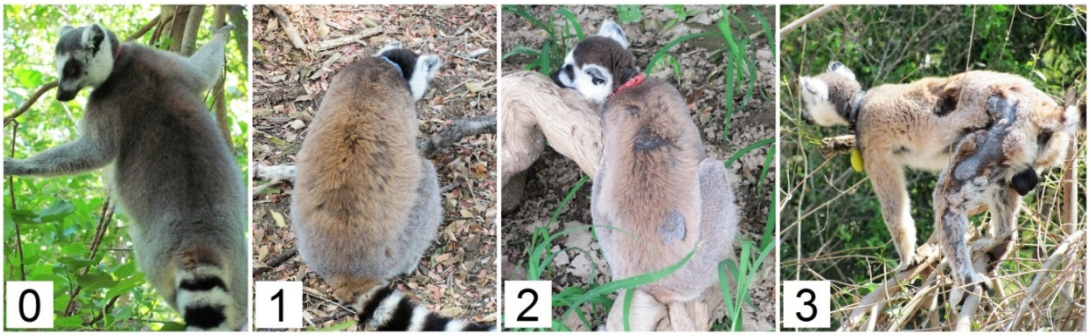


Figure 7.1. Coat condition scores for BMSR *L. catta*.. 0 = 0-Good, 1 = 1-Rough, 2 = 2-Holes, 3 = 3-Ragged. Individuals in images 2 and 3 demonstrate alopecia.

Table 7.1. Coat Condition Scoring System Definitions (Based on Berg et al., 2009).

Coat Score	Defining Characteristics of Coat Condition Score
0 (Good)	1. Animal has excellent coat with no or few imperfections. 2. Little to no hair creasing present. 3. Hair is unidirectional in cranial-caudal direction. 4. Little to no visible coat shagginess; coat is uniform. 5. One or two small, coin-sized holes permissible.
1 (Rough)	1. Coat has slight to significant shagginess and/or unevenness. 2. Creasing of the hair frequently present. 3. Hair may have one or two small holes up to coin size. 4. Score may be assigned when one larger hole present, but small sized (e.g. two or three coins).
2 (Holes)	1. Animal demonstrates > 2 coin-sized holes in hair. 2. Holes are typically surrounded by rough hair. 3. < 25% of hair missing in total. 4. Hair may be partially grown back in the hole, but not predominantly filled w/ presence of hole still clear.
3 (Ragged)	1. Hair shows holes over >25% but <50% of body or limbs. 2. Hair is < 1/2 normal length in affected areas.
4 (Sheared)	1. Hair less than half depth on >50% of body. NOT OBSERVED.
5 (Bald)	1. More than 50% of hair on body or limbs absent. NOT OBSERVED.

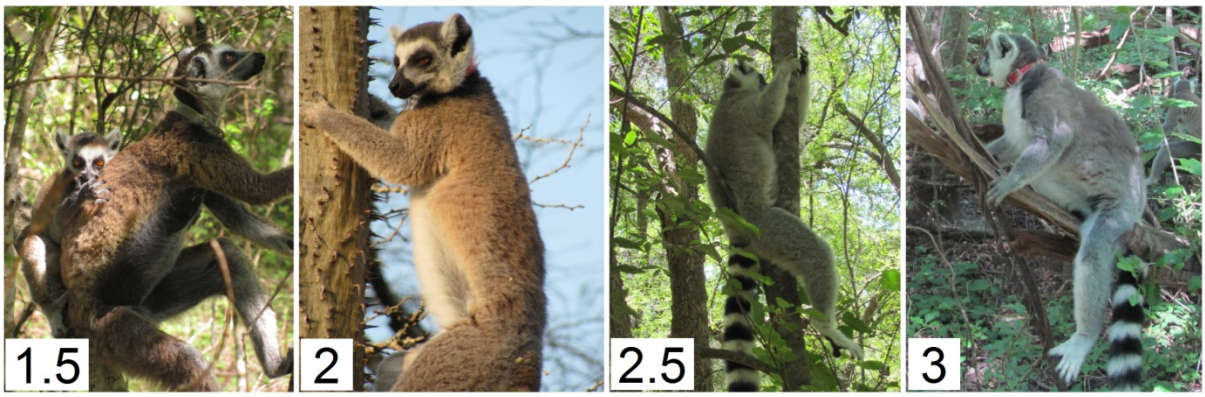


Figure 7.2. Body condition scores for BMSR *L. catta.* 1.5 = Reduced body condition; 2 = Moderate body condition; 2.5 = Slightly high body condition; 3 = High body condition.

Table 7.2. Body Condition Scoring System Definitions.

Score	Defining Characteristics of Body Score
1	1. Animal is extremely thin. 2. Underlying skeletal structure clearly visible. 3. Score 1 was not observed during this study.
1.5	1. Animal shows low body mass and appears visibly thin. 2. Body rail-like; hips are exposed with flanks depressed. 3. Animal has taut skin with no excess fat. 4. Eye orbits exaggerated and face appears thin. 5. No bones or ribs visible through skin.
2	1. Animal is of average size, neither thin nor fat; animal is lean. 2. No excess fat apparent with; animal is "sleek" in appearance. 3. Face appears full; eye orbits do not protrude. 4. Hips and flanks are not concave or only slightly so.
2.5	1. Animal of slightly high body mass than in score 2. 2. Hips often rounded; fat often on hips and lower back. 3. Body rounded; slight to moderate belly fat may be present. 4. Face full; head may appear small in relation to body.
3	1. Animal is of high body mass. 2. Girth > 2.5 w/ belly fat present; wide at hips and midsection. 3. Sides bulge when sitting, fat may be present over legs. 4. Head small in relation to body; animal is "light bulb" shape. 5. Animal appears very full or "overstuffed."
3.5 - 4	1. Animal is of extremely high body mass. 2. Extreme amounts of fat present. 3. Scores 3.5 and 4 were not observed during this study.

All scores were preferably collected when subjects were on the ground and at a close range to the observer. While observation distance was not recorded, observations were typically conducted less than 3 meters from the animal. To account for multiple monthly observations, each animal's status was assessed using the highest recorded monthly value for coat and body condition, while alopecia was reported if observed during at least one monthly assessment.

Coat, body condition and alopecia scores were evaluated for month-to-month differences and for within-month sex differences using nonparametric methods (Wilcoxon Mann-Whitney and Wilcoxon signed-rank tests). Between-month differences for alopecia were determined using

McNemar's exact test. Within-month sex differences for alopecia were determined using Fisher's exact test. To examine the effect of reproductive status on within-month variation in body condition, females who were never observed with a new infant and males were compared to females who had infants using a Wilcoxon Mann-Whitney test. For all tests, significance was set at the $\alpha \leq 0.05$ level using a two-tailed distribution with values approaching $p = 0.05$ (e.g., $p = 0.052$) being rounded down (see Weiss, 2011).

Results.

Coat Scores. Significant differences for coat scores were observed between the months of December and January ($p = 0.008$, $S = -40.05$, $N = 34$, Dec mean = 1.294, Jan mean = 0.912) as well as between February and March ($p = 0.002$, $S = -40.00$, $N = 29$, Feb mean = 0.794, March mean = 0.345). Coat scores improved several months following the transition from the dry season to the wet season (November), during which poor coat scores (e.g., 2-Holes and 3-Ragged) became less common and/or disappeared. While females generally had better coats than males during the late dry season, males generally demonstrated better coats when compared with females for the months following infant birth and subsequent lactation (e.g., September / October onward). However, significant within-month coat differences between sexes were not observed except when males had lower (better) coat scores in January (Wilcoxon Mann-Whitney test: $p = 0.052$, $S = 159.5$, $Z = -1.95$; ♂: $n = 11$, rank mean = 14.5; ♀: $n = 23$, rank mean = 18.9; Table 7.3). Although individual coat scores ranged from 0-Good to 3-Ragged, no animals were observed to have coats in the 4-Sheared or 5-Bald categories.

Table 7.3. Monthly Coat Scores by Sex (Wilcoxon Mann-Whitney Test).

		Jul.	Aug.	Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.
Males	**n =**	13	13	13	12	12	11	11	11	10
	Score Sum	267	256	235.5	222	200.5	152	159.5	163	129
	Expect Sum	240.5	240.5	240.5	216	216	192.5	192.5	192.5	150
	Score Mean	20.5	19.7	18.1	18.5	16.7	13.8	14.5	14.8	12.9
Females	**n =**	23	23	23	23	23	23	23	23	19
	Score Sum	399	410	430.5	408	429.5	443	435.5	432	306
	Expect Sum	425.5	425.5	425.5	414	414	402.5	402.5	402.5	285
	Score Mean	17.4	17.8	18.7	17.7	18.7	19.3	18.9	18.8	16.1
Stat Values	**S - Score**	267	256	235.5	222	200.5	152	159.5	163	129
	Z	1.05	0.58	-0.19	0.27	-0.66	-1.64	-1.95	-1.52	-1.14
	p =	0.292	0.563	0.851	0.784	0.512	0.102	***0.052***	0.128	0.254

Alopecia Scores. No significant within-month differences were observed between males and females. Exact McNemar's tests indicate that alopecia became more common during the height of the dry season from July to August (p = 0.031, Jul: No Alopecia n = 28, Alopecia n = 8; Aug: No Alopecia n = 22, Alopecia n = 14), along with a similar trend between November and December (p = 0.070, Nov: No Alopecia n = 14, Alopecia n = 21; Dec: No Alopecia n = 8, Alopecia n = 26). Individuals demonstrated alopecia with decreased frequency from December to January (p = 0.0002, Dec: No Alopecia n = 8, Alopecia n = 26; Jan: No Alopecia n = 21, Alopecia n = 13) approximately a month after the start of the wet season, with a trend towards reduced alopecia between January and February (p = 0.070, Jan: No Alopecia n = 21, Alopecia, n = 13; Feb: No Alopecia n = 27, Alopecia n = 7; Figure 7.3).

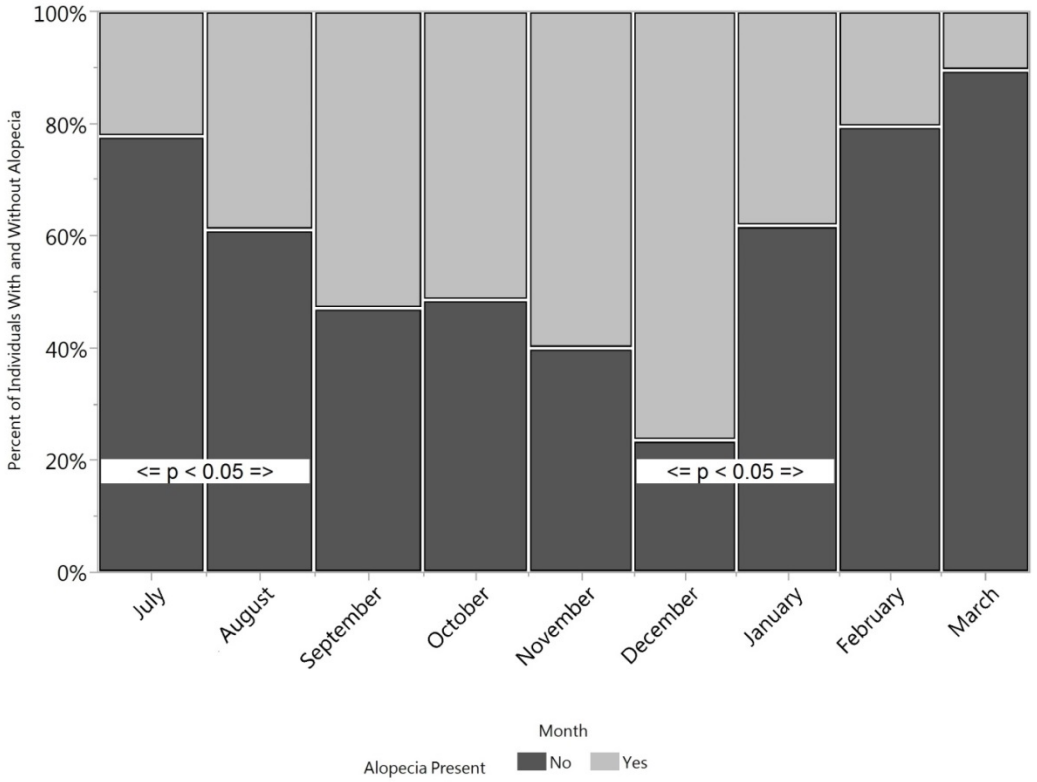


Figure 7.3. Presence of alopecia for BMSR *L. catta.* Alopecia increased in frequency from July to December, before becoming less common from January to March.

Body Condition Scores. Body condition scores did not differ significantly between months. Likewise, body condition did not differ by sex except for February when males had higher body condition scores than did females (Wilcoxon Mann-Whitney test: p = 0.049, S = 226, Z = 1.97; ♂: n = 11, rank mean = 20.6; ♀: n = 23, rank mean = 16.0). When males and non-infant bearing females were compared to females that gave birth, however, significantly higher body condition scores were recorded for males and non-infant bearing females for November, January, February and March [(Wilcoxon Mann-Whitney test: Nov: p = 0.052, S = 339, Z = 1.94; ♂ & ♀ w/o Infant: n = 17, rank mean = 19.9; ♀ w/ Infant: n = 18, rank mean = 16.2 | Jan: p = 0.027, S = 320, Z = 2.21; ♂ & ♀ w/o Infant: n = 16, rank mean = 20.0; ♀ w/ Infant: n = 18, rank mean = 15.3| Feb: p = 0.020, S = 322, Z = 2.33; ♂ & ♀ w/o Infant: n = 16, rank mean = 20.1; ♀ w/ Infant: n =

18, rank mean = 15.2 | Mar: p = 0.004, S = 263, Z = 2.92; ♂ & ♀ w/o Infant: n = 14, rank mean = 18.8; ♀ w/ Infant: n = 15, rank mean = 11.5) Figure 7.4]. Individuals with exceptionally low (e.g., Score 1) or high (Scores 3.5 and 4) body condition scores were not observed.

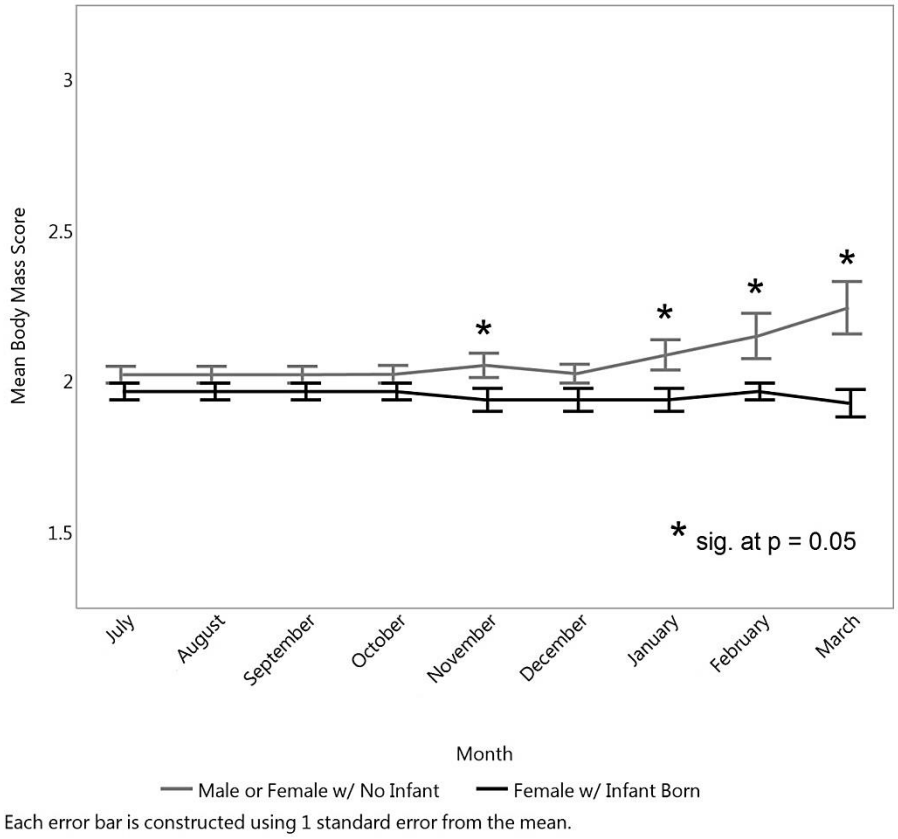


Figure 7.4. Body condition scores for BMSR *L. catta* by reproductive status. Males and females who were not observed to give birth demonstrated higher body condition scores than females who had infants for November, January, February and March.

Discussion.

Coat Condition. Our data show that coat condition varies seasonally among BMSR *Lemur catta*. Coat condition generally improved following the transition from the dry season to the wet season (late October to early November; see Sauther, 1998; Millette, personal observations) when resource availability increases (see Sauther, 1998). Coat scores do not improve immediately,

indicating a delay occurs before hair growth and/or replacement. While the observational method used in this study may not be of high enough resolution to determine fine differences in coat condition between the sexes, from October through March, when females give birth (September to October), nurse (Birth to March) and wean their infants (March: Sauther, 1998; Millette, personal observations), males generally had better average coat scores than did females, although this only reached significance during late lactation (January). Similar patterns of coat condition have also been documented among ring-tailed lemurs at Tsimanampetsotsa, where females demonstrated poorer-quality coats than males overall, but also showed a more rapid deterioration of coat condition during the dry season and slower recovery of coat condition during the wet season (LaFleur, 2012). Seasonal variation in coat condition has also been observed at Berenty, where Jolly (2009a) found coat status declined from September to November for males and for females with infants. Likewise, females with infants showed reduced coat condition in contrast to those without infants in October and November, although no overall male-female differences in coat condition were found.

When compared to data collected for Berenty *L. catta* from September to November (Jolly, 2009a,b), our data demonstrate a restricted range of scores as BMSR lemurs do not show 4-Sheared or 5-Bald categories. This reduced range of variation likely reflects the absence of *Leucaena leucocephala* at BMSR, an introduced food species which has been implicated in the occurrence of bald lemur syndrome among Berenty *L. catta* (Crawford et al., 2006a; Jolly, 2009b; Ichino et al., 2013b). The frequency of alopecia also varied seasonally, generally increasing from July to December, before declining until the end of the study. These alopecia data roughly correspond with our ordinal measures of coat condition across the study period, suggesting that alopecia frequency may provide a general proxy for overall coat condition. As

with coat condition, scores no significant differences were noted between males and females, although informal observations suggest that females and males demonstrate divergent patterns of alopecia. BMSR mothers often appear to lose hair in large (5-10cm) patches where their infants consistently grasp (e.g., the flanks and back) when being carried. Similar hair loss has been reported for Berenty mothers, and has also been reported among Japanese macaques (*Macaca fuscata*), suggesting such patterns are not unique to BMSR lemurs (Crawford et al., 2006a,b; Jolly, 2009a; Zhang, 2011). Additionally, observed patterns of alopecia may also reflect seasonal variation in the rate of hair growth. Captive ring-tailed lemurs have been observed to grow tail hair primarily between the spring and fall equinoxes (Pereira et al., 1999), and similar cessations in body hair growth could possibly impact the occurrence and duration of missing hair patches among wild *L. catta.* Patterns of alopecia at BMSR may also be influenced by the presence of notable tooth comb wear among some lemurs in this population, as individuals with toothcomb wear/damage have been observed to demonstrate increased rates of hair loss (Sauther and Cuozzo, 2013).

Body Condition. While a nonsignificant trend towards greater body condition scores was observed following the new year, no significant monthly differences for body condition were found. Likewise, no sex differences for body condition were noted except for February, when males demonstrated higher body condition scores than did females. This lack of significant differences between months and sexes may reflect the relatively small size of our study sample and/or the coarse nature of our scoring system. Alternatively, a lack of month-to-month and sex differences during the dry season may relate to the relatively longer coats that animals possess during this time (Millette, personal observations). Dry season coats may obscure individual

differences in body status as lengthier hair not only makes animals appear visually larger, but may also hide diagnostic traits characterizing low body condition scores (e.g., exposed skeletal features, depressed flanks, etc.). Future studies may benefit from the use of coat and body condition scoring systems which account for seasonal variability in coat length.

Our results do suggest that the body condition scoring system employed is useful and effective following the onset of the wet season when coats become shorter and females with infants are undergoing lactation. When contrasting males and females who did not have infants against females who did, significant differences for body condition were observed in November, January, February and March. These data suggest that the energetic demands of infant care in terms of lactation and infant carriage may limit weight gain following resource increases during the wet season, while females without infants and males unencumbered by these demands proceed to gain weight. Likewise, all scores of 1.5 (the lowest recorded body condition score) were recorded for females who bore infants, further suggesting that female lactational status and/or infant carrying is associated with reduced body condition. These data are analogous to those reported by Sauther (1998) who noted that females became observably thin during lactation. Similarly, Pereira (1993) reported weight loss among captive lactating *Lemur catta* females, and lactation-related weight loss has also been reported among olive baboons (Bercovitch, 1987). Our data showing that individuals without infants visually gain mass is also similar to Pereira's (1993) observations that captive males and females gain weight prior to the mating season, although reproductive status appears to have a mediating effect on weight gain. While we acknowledge that our study did not collect measured body weights with which to confirm our visual methods, the congruence of our findings with those of others studies suggests that our method represents a valid, useful measure of body condition in wild ring-tailed lemurs

Conclusion.

Our results provide support for the efficacy of visual methods for assessing coat and body condition among BMSR ring-tailed lemurs. Such methods provide a valuable tool permitting examination of individual condition and health status without requiring direct health assessments under veterinary care that are both time-consuming and expensive. Such examinations are also potentially disruptive to animals and also present small, but not insignificant, risk of injury during capture. Our results also correspond with those by other ring-tailed lemur studies, in which poor coat and/or body condition may reflect female reproductive costs (Sauther, 1998; Pride, 2003; Jolly, 2009a; LaFleur, 2012). Additionally, ring-tailed lemur health status may be impacted by factors such as seasonal variation in food availability or stochastic events resulting in reduced food availability, such as cyclones and/or droughts which occur commonly at BMSR (Sauther, 1998; Gould et al, 1999; LaFleur and Gould, 2009; Rasamimanana et al., 2012; Sauther et al., 2012). Health status may also be impacted by factors that influence individual access to nutritional resources including social group size, habitat quality and/or severe dental impairment [(a common occurrence among BMSR *Lemur catta*) Pride, 2005; Cuozzo and Sauther, 2006; Sauther et al., 2006]. The coat and body condition scoring methods presented here may be used to enhance studies investigating the impact of these factors on lemur health. While we do not directly address the impact of these factors here, we are currently assessing how the measures presented in this paper also interact with aspects of lemur behavior and health (e.g., tooth loss status, indicators of illness, etc.).

CHAPTER VIII:

CONCLUSION – SIGNIFICANT FINDINGS AND DIRECTIONS FOR FUTURE RESEARCH.

In this chapter I summarize the main findings of this dissertation concerning the impacts of dental impairment on food processing behavior, fecal-nutritional status, and food processing capacity. I furthermore examine those coat and body condition measures presented in this dissertation for their potential use for better understanding the impacts of dental impairment on ring-tailed lemur health and nutritional status. For each section of this report I detail the major findings and their implications, but also discuss potential areas for future research on the impacts of dental impairment on nutrition, ecology and behavior.

Dental Topographic Analysis of Food Processing Behavior.

Dental topographic analysis conducted indicates that reduced measures of occlusal topography (and thus increased dental impairment status) are associated with behaviors related to food processing, particularly for fruit of the key fallback species *Tamarindus indica*. For this food species, reduced dental area was associated with increased feeding bout lengths data were examined parametrically (for 2DAverage, with a trend towards 3DAverage scores) overall. These effects, however, were limited primarily to females, who demonstrated significant negative correlations between duration and all area measures. Higher angularity was associated with increased durations for time processing this food item both overall and for males.

These data support that dental wear and/or tooth loss status do impact the consumption of tamarind fruit in terms of duration, although the patterning of such behaviors is not consistent in terms of hypothesized directionality (i.e., that all measures would be negatively associated with feeding duration). That individuals with higher angularity spend more time consuming tamarind fruit suggests that they may do so because they maintain an increased ability to access this food source. In contrast, those with lower dental area appear to spend more time processing this food, suggesting that dental wear sufficient to remove surface area does impact food processing capacity. Yet, overall these data indicate that while angularity is associated with dental function (e.g., Ungar and M'Kirera, 2003; Dennis et al., 2004), simple measures of occlusal area are also effective for assessing dental function when data for the full dental arcade are available.

Topographic data also correspond to a number of specific tamarind fruit feeding behaviors including the use of open fruit, manual processing and food item processing location and support the hypothesis that these behaviors are executed in compensation for dental impairment. However, not all food processing behaviors examined were associated with topographic status. For example, the use of conspecific pre-processed foods was not associated with increasing wear as was hypothesized. Likewise, although licking was associated with tooth loss in previous studies (Millette, 2007; Millette et al., 2009), a compensatory association of this behavior was only present for females. The occurrence of compensatory behaviors may be related to other factors rather than simply tooth loss. Males and females may differentially use this behavior due to divergent access to resources, as females may have access to preferred tamarind fruit (e.g., Gemmill and Gould, 2008) that may be better suited to processing through use of the tongue.

Overall, however, the dental-behavioral data presented *does* indicate that individuals with high levels of impairment demonstrate a reduced capacity to process tamarind fruit, although there does appear to be some behavioral compensation present. Such behavioral compensation, particularly the use of open fruit, manual processing and alterations to tooth row use, may permit lemurs in this population to survive and reproduce in relatively good health (as has been reported in Cuozzo and Sauther 2004, 2006a, etc.) despite a reduced capacity to process this food.

Assessment of feeding durations by type and species indicates dental impairment is frequently associated with longer feeding bout periods, and that impairment may be associated with a reduced capacity to process food items generally (e.g., in addition to tamarind fruit). However, topographic scores appear to be most closely related to feeding durations for a limited number of species and food item types. Leaves and/or vegetative materials are most closely associated with increased feeding times when examined on a species level (particularly for *Tamelapsis linearis and Tamarindus indica*). Insects (primarily in the form of caterpillars) are also associated with increased feeding bout durations. In general, most topographic scores were negatively associated with feeding duration; as wear increases (lower scores = more wear) so does time spent feeding during each bout. It is also noteable that certain species and food types demonstrated durations positively associated with topographic scores (for example, *Salvadora angustifolia* leaves and fruit, as well as for wood/termite excreta). Differences between the physical and mechanical properties of these food items may lead to differences in the amount of time an animal chooses to spend on a specific food item species or type. These differences suggest that individuals who maintain a greater ability to process a certain type of food may spend *more* time doing so during each bout because they *can* effectively process this food. As such, simple use of time spent feeding may not be an indicator of reduced dental capacity for

certain food items, and future studies should be designed with this caveat in mind. The finding that certain species appear to be more challenging to those with reduced dental capacity is also important as it indicates that individuals with dental impairment may be more limited by these foods than they are for species which are not associated with dental topographic status. This has implications for the design of future studies examining the impacts of tooth wear as they may allow for finer analyses of how interactions between specific species and dental impairment negatively impact aspects of health and behavior for this and other primate populations. These data also have implications for the management of this population as they may allow for better assessment of where lemurs with dental impairment are likely to thrive based on resources available within a specific environment.

A wide range of variation for the association between specific topographic measures and each specific plant species and food item type was also present. Such variation may be related to small sample sizes available for some foods, but may also indicate that each measure is differentially associated with the breakdown of specific food items. This is not unexpected, as different topographic measures are associated with different aspects of dental function. For example, higher relief has been associated with increased shearing capacity, while slope and relief are associated with general wear state, and angularity is often associated with the maintenance of dental function (Ungar and M'Kirera, 2003; Dennis et al., 2004; Boyer, 2008; Yamashita et al., 2015a). Likewise, Yamashita et al., (2015a) suggest that more mechanically challenging diets and/or exogenous grit consumption are associated with reduced topographic measures for BMSR lemurs, but the relationship of such variables to food processing behavior is currently unknown. Therefore, in future studies examining the connection between dental

topography and behavior, it may be informative to examine the mechanical properties of each food item relative to each dental topographic measure and individual processing behaviors.

There are also a number of other means by which ring-tailed lemurs may compensate behaviorally for dental wear that have not been investigated using topographic methods, such as through alterations to their activity budget. It is also not yet clear if impaired individuals are less capable of accessing and gaining energy from foods overall despite demonstrating a reduced capacity to access foods on a per-bout basis (e.g., do individuals with dental impairment spend more time feeding overall and/or consume a similar amount of food in total when compared to those with lower impairment?). While activity budget has been assessed for dental impairment in the form of tooth loss, an analysis of this data with respect to dental wear and/or topography has not yet been completed. Such data may provide further insight into how dental wear impacts lemur behavior, but also may indicate that individuals compensate for wear-related reductions in dental function though increasing time spent feeding and foraging, or through other alterations to their activity patterns. Such data may clairify if individuals with dental impairment truly are consuming less food overall and thus capturing less energy in total or per time spent feeding as a function of reduced processing capacity. Alternatively, such future data collection may provide evidence that individuals are effectively compensating for dental impairment by increasing the total amount of food ingested despite requiring longer feeding times per item consumed.

While not presented here, during field work associated with this dissertation, I collected behavioral data sufficient to examine potential compensatory alterations to activity budget in the future. I hypothesize that these data will resemble those presented in Millette et al., 2009 (see Chapter 1), and that individuals with reduced topographic scores are likely to demonstrate increases in feeding, foraging and grooming with reduced resting. I furthermore expect that these

behaviors will be completed in a manner consistent with group living (e.g., they will be done during the early afternoon rest period, or during the early morning period when most animals are sunning or resting). Seasonal variation in compensatory behaviors may also be examined, as it is likely that individuals with dental impairment demonstrate higher rates of behavioral compensation during the dry season, as challenging tamarind fruit represents the predominant food source during this time (Sauther, 1992, 1998; see also Chapters 3 and 4). Alternatively, as dental wear appears to impact the processing of leaves (see Chapter 4 and 5), impaired individuals may also demonstrate higher rates of feeding during the wet season when the use and availability of such resources becomes more common (Sauther, 1992, 1998; Millette, personal observations). Dentally-impaired lemurs may also preferentially select food items with less challenging mechanical properties. Such an analysis will, however, require integration of individual feeding and dental data with information on food item mechanical properties. As such data are currently available for this site (see Yamashita, 2000, 2008b; Yamashita et al., 2015a), such an analysis is expected to be completed in the near future.

It is important to highlight that the topographic methods that I employed during this study examined the entirety of the dental arcade, from p4-m3 bilaterally. This is in comparison to previously conducted GIS-based dental topographic research which has focused on one or two teeth at most (e.g., M'Kirera and Ungar, 2003; Ungar and M'Kirera, 2003; Dennis et al., 2004; Ungar and Bunn, 2008; Bunn and Ungar, 2009; Cuozzo et al., 2014; Yamashita et al., 2015a). This study therefore demonstrates the efficacy of such a "whole mouth" approach to studying the impacts of dental wear with GIS methods. Dental wear may occur across a number of positions, and may variably affect the performance of some teeth relative to others which remain unworn (see Cuozzo and Sauther, 2004, 2006a). Likewise, animals may alter their placement of foods to

compensate against dental impairment for specific teeth (Chapter 4). Use of multiple teeth during topographic analysis therefore sheds additional light on the impacts of dental impairment beyond what may be assessed using a one or two tooth-based methodology. In addition, both 2D and 3D area measurements were associated with a variety of indicators of reduced dental function (e.g., feeding duration and use alternative food processing strategies). Prior to this project, GIS-based dental topographic analysis has suffered from a "missing tooth" problem, and until now it has not been possible to assess the potential impacts of missing dental positions using the topographic measures of slope, relief or angularity as these require the presence of a tooth to calculate (Ungar, personal communication). When combined with data drawn from the entire tooth row, the measures of 3DSum and 2DSum, however, provide a means for quantifying the total tooth area present within the dental arcade which may be used for food processing. As missing teeth do not contribute to these calculations, individuals with greater tooth loss demonstrate reduced area sum scores. Thus, these measures provide a means for quantifying the morphological consequences of tooth loss using topographic methods, and are readily calculated following the determination of individual tooth relief scores. Future use of 2D and 3D sum scores during topographic analysis may therefore contribute to our understanding of the impacts of tooth loss in addition to wear alone. In turn, in the future it may also be beneficial to integrate the measures of slope, relief and angularity with the use of area sums to simultaneously provide a measure of working area as well as dental topographic form. In doing so such analyses may better quantify the relative impacts of tooth loss and variably worn topography, which may enhance investigations related to the study of dental impairment's impacts on factors such as health, survival, nutrition and may help to further clarify the onset of dental senescence.

Fecal-nutritional Analysis. My analysis of fecal contents for a subsample (n = 14) of individuals indicates that dental wear and loss are associated with a reduced ability to digest and absorb nutrients from food items following their ingestion. During the dry season, tooth loss was associated with reduced amounts of NDF and ADF within the feces, although protein content was higher for such individuals. In contrast, during the wet season NDF and ADF were positively associated with tooth loss status. Similar patterns were present for tooth wear with NDF being negatively associated with tooth wear during the dry season, and ADF trending towards a positive association with wear during the wet season. These data were in contrast to the study hypothesis that fiber content would be higher among those with tooth loss across both seasons, and may suggest that differences in the intake of total amounts fiber may vary based on seasonal food availability or by food choice. Although this hypothesis remains to be investigated thoroughly, divergent patterns of fruit vs. leaves in fecal material relative to dental impairment status for the dry season (see Chapter 6) suggest that such differences in diet are likely. The finding that NDF and ADF levels were associated with tooth loss during the wet season, does however, suggest that individuals may be impaired in their ability to utilize fibrous resources and may also reflect the higher availability of leaves during this period, (see Sauther, 1998; see also Chapter 6).

When fiber and structural carbohydrate content were standardized as a ratio to acid detergent lignin (ADL), tooth loss was associated with higher ratios for NDF, ADF, hemicellulose and cellulose for the dry season. For the wet season, NDF was significantly associated with tooth loss status, with trends for ADF and cellulose also present. These ratio-based data suggest that individuals with tooth loss and/or tooth wear are not as capable at fermenting structural carbohydrates within the fiber fraction of their diet. This finding provides

direct evidence that the tooth loss and/or wear observed at BMSR may negatively impact the ability of these animals to absorb nutrients from structural carbohydrates found within their diets. This finding must, however, be tempered by the lack of dietary input data for each fecal sample. As noted above, individuals could be ingesting quantities of foods which compensate for a reduced capacity to ferment and absorb structural carbohydrates. However, these data do provide initial evidence that dental impairment does negatively impact individuals in terms of their capacity to effectively utilize ingested food items.

It is also notable that protein (e.g., fecal nitrogen) content was higher among individuals with tooth loss than those without for the dry season, suggesting that impairment reduces the capacity to absorb protein from their diets during this period. Concurrently, increasing tooth wear was also associated with greater proportions of protein within the feces during the dry season. No such patterns were noted for protein during the wet season, although generally fecal protein was higher during this season for animals overall. Such differences are likely related to dietary differences between the seasons, and may reflect a ceiling effect with regards to protein digestion during the wet season as potentially protein-rich leaves become more common during this period. Alternatively, patterns observed here could result from divergent gut microbiota or from excretion of endogenous urea into the gut for individuals with tooth loss, resulting in higher nitrogen in the feces during the dry season (see Chapman et al., 2005; Schwarm et al., 2009). While there does appear to be a relationship between tooth loss status and fecal nitrogen / protein content, at present the source of this variation remains somewhat unclear.

Although relationships between fecal nutrient content and dental impairment status are present, these must be subject to a number of future investigations to confirm their utility. Currently it is unclear if fecal content is primarily a function of divergent patterns of digestive

capacity and/or physiology or is related more closely to dietary patterns. Future research which carefully controls for individual dietary intake relative to residual nutrients in the feces is required. Although this could not be completed within the confines of this study, such studies have been completed *in vivo* for a number of taxa (see Kay and Sheine, 1979; Sheine, 1979; Bjorndal et al., 1990; Edwards and Ullrey, 1999a,b; Campbell et al., 2004; Schmidt et al., 2005a,b, etc.), and could feasibly be completed among wild BMSR *L. catta.* Ideally, such a study would also include more individuals than were present in this preliminary work which demonstrated a limited sample size that precludes assessment of fecal nutritional patterns by age, sex and habitat use. It may also be of use to assess the impact of dental impairment using topographic techniques. As demonstrated in Chapter 4, these methods are of sufficient resolution to distinguish behavioral variation related to dental impairment. They are thus likely to also distinguish differences in fecal nutritional content, which may provide greater insight into the relationship of topography to dental function. For example, inclusion of topographic methods could provide further evidence for the onset of dental senescence if reduced angularity is associated with increased fecal nutrient content (see Chapters 1 and 4).

Fecal Ash Content.

Fecal ash represents the mineral content within the diet consumed by an individual, and serves as a measure of dietary silicates, either in the form of phytoliths or exogenous grit adhering to foods. Such dietary silicates are likewise presumed to be a major source of dental wear among mammalian taxa (Baker et al., 1959; Sanson et al., 2007; Rabenold and Pearson, 2011; Lucas et al., 2013,2014). For BMSR *L. catta* tooth loss was associated with increased ash

content for both the dry and wet season, while tooth wear was associated with increasing ash content during the dry season (with a positive trend also present during the wet season).

The association of fecal ash with dental wear and tooth loss status indicates that individuals with higher amounts of dental impairment are consuming higher amounts of wear-inducing dietary silicates. This finding augments our understanding of the ontogeny of dental wear in this population. While tamarind is frequently associated with tooth wear and loss here (e.g., Cuozzo and Sauther, 2004; Cuozzo and Sauther, 2006a; Yamashita et al., 2012), these data suggest that tooth wear may also result from inclusion of siliceous particles in the diet. Such a source of wear has been proposed for Parcel 2 *L. catta* by Yamashita et al. (2015a), and this study provides evidence that grit and/or phytoliths may be causing wear for individuals in Parcel 1 as well. However, the exact source of such particles is not yet well understood and must be subject to further research. It is possible that the amount of silica consumed is related to habitat use, which is plausible given that tooth wear at BMSR has been observed to vary based on location (Cuozzo et al., 2014; Yamashita et al., 2015a). It remains to be tested, however, if ash content is higher for those individuals which use “dustier” areas of the reserve (e.g., close to the Sakamena river) in comparison to those with lower ash content, or if the amount of dust present on food items varies based on location. Similarly, it may be that individuals with greater amounts of tooth wear are selecting foods which are easier to consume, but higher in grit or phytolith content. This is possible if these individuals preferentially use opened or easy to process food items sourced from the ground. While this does not appear to be the case for tamarind consumption, this hypothesis has not yet been tested for other food items (e.g., akaly, human sourced mangoes, or crops grown in sandy soils such as melons and squash, etc.). Thus, while ash appears to be associated with wear in this population, the source of these silicates and their

interactions with lemur ecology remain unclear and must be subject to future inquiry to determine their origin.

Food Particle Size and Dental Impairment.

That effects of dental impairment were relatable to fecal-nutritional status is not surprising given that significant tooth wear and or loss are expected to reduce the animal's ability to reduce foods into a form that is suitable for subsequent digestion and/or fermentation. Failure to sufficiently masticate food items may result in the ingestion of food particles of larger size, thus reducing the surface area relative to volume on which digestive enzymes and/or gut bacteria can act upon in comparison smaller sized particles. In turn, the animal is likely to be less capable at harnessing nutrients available in larger-sized particles ((McLeod and Minson, 1969; Kay and Sheine, 1979; Sheine, 1979; Gipps and Sanson, 1984; Lanyon and Sanson, 1986; Bjorndal et al., 1990; Lentle et al., 2003; Lucas, 2004).

Data presented in Chapter 6 indicate that individuals with dental impairment demonstrate larger sized particles in their feces, which provides a mechanism for the divergent patterns of nutrient excretion reported in Chapter 5. In particular, individuals with tooth loss demonstrated reduced amounts of small sized particles (1.00-4.75mm) relative to those without tooth loss, but did not demonstrate differences in larger sized particles (4.75-11.2mm or >11.2mm). Similarly, increasing wear was associated with reduced numbers of particles within the 1.00 – 4.75mm sieve, and the >11.2mm portion when limited to postcanine wear score, although wear was not associated with particle sizes when limited to those individuals without tooth loss. Overall, these data indicate that individuals with dental impairment are less capable at processing food items than are those without lower levels of dental impairment, although it does appear that tooth loss

and/or significant dental impairment is necessary for these effects to occur. It is also of note that individuals with tooth loss demonstrate more leaves and less fruit in their fecal material for the 1.0 – 4.75mm and >11.2mm samples than do those without. This suggests that there may be dietary differences relatable to tooth loss within this population, and given that these data were collected during the dry season, may indicate that such individuals are less capable at consuming tamarind fruit as is similarly indicated by data presented in Chapter 4.

As noted above, data presented in Chapter 6 provide a mechanism with which to explain fecal nutritional differences noted in Chapter 5. Food particle size is likely impacting the capacity of individuals to adequately utilize ingested resources. However, data collected in this chapter could be expanded to provide a greater understanding of the role of food particle size. As this study utilized a coarse weighed sieve fraction methodology to understand particulate size, it could not account for particles smaller than <1mm or calculate the exact distribution of particles within the feces. One way to improve on this methodology would be to calculate the exact size and surface area of particles present, but also the distribution of food particle sizes within in the fecal material. Such data may be collected using either optical scanning, optical imaging or laser diffraction laser diffraction techniques, which provide enhanced information for the distribution of particles on the lower end of the size spectrum and which likely demonstrate the highest potential fermentability from a surface to volume standpoint (see Vanderbilt et al., 1993; Hoebler et al., 2009). As most differences related to dental impairment in this study were found within the smaller-sized fraction, such methods may provide enhanced information on the sources of divergent patterns of fecal-nutrient excretion related to dental impairment in this and other primate population.

In addition, it is notable that dental wear alone was not associated with food particle size within the study sample, suggesting that significant dental impairment is necessary for significant patterns of food particle size to emerge. As such, when combined with topographic analysis food particle size data may be useful for assessing the function of the worn dentition. In particular, angularity analyses may be well suited to this understanding this question given their potential relationship to dental senescence. If food particle sizes only differ based upon angularity measurements, this could be an excellent means of assessing if this measure is in fact a signal of the onset of dental senescence. Finally, fecal food particle size may be of interest for future studies utilizing fecal nutritional analysis. Given that smaller particles are likely to be digested more completely, it is likely that individuals with smaller particles demonstrate reduced fecal nutritional content. Such an association for ring-tailed lemurs, however, remains to be examined.

Coat and Body Condition.

Coat condition was observed to vary seasonally within this population of ring-tailed lemurs for ordinal measures of coat condition and for the presence of alopecia. In particular, coats were observed to improve from the dry season to the wet season, indicating that coat greater condition may be linked to increases in resource availability. Similarly, males generally demonstrated better coat condition than did females, particularly during the period of late lactation, also implying that energetic status impacts coat condition. No significant patterns of body condition were noted, although a trend towards larger body sizes was noted during the wet season when resources increase. However, body size differences were noted for individuals with

and without infants, supporting that energetic demands imposed by infant care may impact body condition in a manner similar to that reported for coat condition.

While the relationship of body size and coat condition were not assessed directly in relationship to dental impairment status, these measures may provide an additional means for assessing the impact of dental wear and/or tooth loss. It is notable that coat and body condition appeared to vary relative to seasonal differences in available resources and/or lactational status. If dental impairment impacts the individual's capacity to harness energy within their diet, it is likely that similar patterns of body size and or coat condition may occur. For example, Miller et al., report that one individual (lemur 188) with extensive tooth loss (e.g., 56%) was noted to be of low body mass and demonstrate a high ectoparasite load, which is associated with dermatitis-related poor coat condition in ring-tailed lemurs (see Junge and Sauther, 2006). Similarly, in this study, the individual with the highest amount of tooth loss (Black 226, 81% loss) consistently demonstrated the worst coat condition (and often appeared to be undersized) relative to other individuals. However, it is also notable that Blue 348, who demonstrated the second highest amount of tooth loss (>40%) in this sample did not appear to be of reduced body size (in fact she was often larger than other individuals) or to have a coat with poor condition, suggesting that tooth loss alone is not the sole source of low body size or poor coat condition. These variables, and in particular coat condition, may not be only related to gross tooth loss status, but may primarily reflect tooth comb status. Impairment of the tooth comb and a failure to groom efficiently may lead increased ectoparasite load, which has been observed to negatively impact coat condition and/or result in hair loss (Junge and Sauther, 2006; Sauther and Cuozzo, 2013). Future studies are required to more firmly establish relationships between dental impairment status, coat and body condition. By examining the relative effects of tooth wear/loss within the

dental arcade in contrast to that related to toothcomb condition we may be also be able to better determine if coat (and possibly body) condition is related to energetic costs associated with dental impairment or with ectoparasite loads resulting from reduced grooming capacity. As such, coat and body condition scores collected during the field element of this study will be analyzed in the near future using postcanine topographic and dental comb data collected by Dr. Frank Cuozzo at BMSR, to provide resolution to such questions.

Additional Broader Impacts and Study Implications.

Results presented in this dissertation also have implications for the broader understanding of dental wear, tooth loss and dental senescence among primates and other mammals. Significant dental wear and/or tooth loss has been linked to a variety of negative health, nutritional and life history outcomes among mammalian taxa, with effects ranging from a reduced capacity to utilize food resources (e.g., Gipps and Sanson, 1986; Lanyon and Sanson Logan and Sanson, 2002b,c; Venkataraman et al., 2014) to death due to starvation resulting from an inability to process food items (Buss, 1990; Lucas, 2004). However, to date, I know of no other studies which have actually quantified the impacts of dental impairment on nutritional status among wild nonhuman primates (or other animals) using dental data drawn directly from study individuals. For example, among sifaka King et al. (2005) report that dental senescence leads to increased infant death during periods of resource stress resulting from dehydration due to inadequate milk production. However, King et al. (2005) did not directly measure individual tooth wear or nutritional status. Conversely, data presented here do suggest that individuals with dental impairment not only demonstrate a reduced capacity to process and breakdown food items, but also a reduced capacity to absorb nutrients from their diet. My data may therefore provide some

support for King et al.'s interpretations. However, it is also notable that infant survival at BMSR appears to be weakly linked to tooth loss status (Cuozzo et al., 2010), suggesting that there are significant differences in the overall impacts of dental impairment between these two taxa on reproductive life history.

Additionally, data presented here indicate that individuals utilize behavioral compensation to survive and maintain their health status despite possessing extremely worn dentitions which are less efficient at processing food items. For example, individuals with reduced topographic scores utilized alternative food processing techniques for tamarind fruit (e.g., manual processing, licking behavior, alternative tooth row use and use of open tamarind pods). These observations highlight that measures of dental impairment alone may not be sufficiently indicative of reduced health or survival without considering the behavioral context associated with such dental impairment. Yet, as dental impairment was associated with fecal measures of reduced nutritional uptake during this study, I must acknowledge that there are likely limits to the extent that behavioral compensation can attenuate the impacts of dental impairment, although this remains an open area of inquiry.

Information presented in this dissertation may also help inform interpretations of the primate paleontological record as teeth (including worn teeth) are among the most common items found within fossil assemblages. For example, ring-tailed lemur dental morphology is analogous to that of Eocene Adapiformes (Gebo, 2012), and notable postcanine tooth wear has been observed for a number of Eocene Notharctine primates (Sauther and Cuozzo, 2012). Data provided by this study suggest that such wear may have had significant negative impacts on the capacity of these primates to process and absorb food items. Likewise, full dental arcade topographic methods employed by this project may provide for enhanced future assessment of

these primate's dental functional morphology despite tooth wear (see also Ungar and M'Kirera, 2003). Additionally, this study contributes to our understanding of indicators and behaviors associated with conspecific care among fossil hominin taxa. The occurrence of conspecific care has been invoked to explain the extended survival of hominins with significant tooth loss (Neanderthals: Lebel et al., 2001; Lebel and Trinkaus, 2002; Dmanisi *Homo*: Lordikipandize et al., 2005, 2006). However, BMSR ring-tailed lemurs survive significant tooth loss similar to that of these hominins without the expression of such care, suggesting that tooth loss alone cannot be used to implicate the expression of this behavior, nor can tooth loss be used to implicate the occurrence of "morality" (see Dettwyler, 1991; DeGusta, 2002, 2003; Cuozzo and Sauther, 2004; Millette et al., 2009). Data from this study (similar to that presented in Millette et al., 2009), highlight that the survival of such individuals may have been a function of alternative food processing strategies similar to those observed among BMSR ring-tailed lemurs rather than a function of conspecific care, although it must also be acknowledged that dentally impaired hominins would likely have also utilized cultural adaptations to engage in such food processing behaviors (e.g., use of tools and cooking).

Concluding Statement: How Does Dental Impairment Impact BMSR *Lemur catta?*

The overarching goal of this dissertation was to examine the possible impacts of dental wear and tooth loss among ring-tailed lemurs using a broad dental-ecological framework combining behavioral, morphological and nutritional information collected in both field and laboratory-based settings. A key aspect of this work was to determine in what manner dental impairment impacts these individuals in terms of their ability to not only process, but also to utilize ingested food items. Overall, data presented here indicate that tooth wear and loss do have

a generally negative effect on these facets of ring-tailed lemur behavior and biology. Individuals with increasing topographically-measured wear demonstrate patterns of food processing behavior indicative of a reduced capacity to process tamarind fruit dentally. Given the significance of this food as a dry season fall back resource, it is likely that dental wear and loss results in real and significant challenges for individuals with significant dental impairment. Feeding bout durations for other major food types and species generally follow in a similar direction, with longer bout lengths typically being associated with dental impairment. Although these associations may vary for some foods, it appears that there is a generally negative relationship between the ability to process foods and dental impairment status.

In addition to behavioral evidence for the impact of dental impairment, data presented in this study suggest that individuals with dental wear do not as effectively process food items into fragments suitable for fermentation and/or digestion (see Chapter 6). This appears to result in increased residual nutrient content within the fecal material, particularly for the fraction of structural carbohydrates (see Chapter 5), indicating that with increasing impairment individuals do not as effectively ferment and utilize this potential source of energy. As with the topographic-behavioral dataset, these nutritional data suggest that dental impairment has a real and significant negative impact on the ability of individuals to harness resources available at BMSR.

Yet, it is also apparent that individuals at BMSR with significant dental impairment (and even senescent dentitions) can survive for an extended period of time in relatively good health (Cuozzo and Sauther, 2004, 2006a). For example, the most dentally-impaired individual in this study sample (Black 226) was observed to survive across the study period despite being almost edentulous. This individual's health furthermore appeared to improve visually as the study progressed from the dry to wet seasons (Millette, personal observations). Food processing data

for tamarind presented here indicate that behavioral compensation for dental wear and/or loss in the form of food item selection (e.g., use of open pods) and processing techniques (e.g., divergent use of the dental arcade and hands) may permit such survival and maintenance of health status. It is also of note that some of these techniques, particularly the use of the hands, appear to be most common in those with exceptionally severe wear, suggesting that the form and rate behavioral compensation is linked to the extent of impairment observed. This is not to say that such behavioral compensation for dental impairment is complete, as health effects have been associated with significant tooth loss in this population (see Miller et al., 2007). The extent to which behavioral adjustments may compensate for the health effects of dental impairment however remains to be quantified. Given that multiple variables may contribute to health status among ring tailed lemurs (e.g., age, sex, resource and ranging patterns, social rank, reproductive status, etc.) in addition to dental impairment, this remains a challenging area of interest to examine. Likewise, alternative behavioral compensatory methods, such as the possible divergent use of food types and species, and/or adjustments to activity budget have not yet been assessed relative to wear or topographic status, nor have dental measures yet been compared to visual measures of health status. While these areas of inquiry remain open and will be addressed using data collected during this study which have not yet been examined, their resolution will also likely require the collection of additional dental-ecological data among BMSR *L. catta.*

In all, a number of salient points emerge from this dissertation project with respect to the effects of dental impairment on ring-tailed lemur biology and behavior:

1) Dental impairment impacts aspects of individual feeding behavior, particularly for tamarind fruit. This includes divergent patterns of time spent processing foods, as well as techniques used by animals to access tamarind fruit-based resources.

2) Dental topographic techniques possess resolution sufficient for assessing behavioral aspects of dental impairment on an individual level. In particular, the “whole arcade” approach utilized by this study provides a means for assessing the impacts of both wear as well as for tooth loss, which is difficult to achieve using a single or two-tooth methodology.

3) Fecal food particle size and fecal content analyses indicate that dental impairment in the form of tooth loss and wear reduces the individual’s capacity to utilize ingested food resources. Fecal data also indicate that dietary silicates may result in tooth wear among BMSR *L. catta* in addition to that which has been associated with the use of tamarind fruit.

4) Visual coat and body condition scores appear to vary with resource seasonality and with reproductive state, suggesting that these measures may be related to individual energetic status. Such measures may therefore be of use in future studies of dental impairment and its impact for BMSR ring-tailed lemurs as it also appears that these factors influence the ability of individuals to harness ingested resources.

5) Individuals at BMSR do appear to compensate behaviorally for the impacts of dental impairment in the form of food processing. The extent to which such behavioral modifications counter the impacts of dental impairment is not yet known, nor is how the occurrence of other potential behavioral mechanisms (e.g., activity budget alterations) may counter the impacts of dental impairment. Future research is necessary to quantify the role of behavioral mechanisms in compensation for dental impairment.

BIBLIOGRAPHY

Agetsuma N, Nakagawa N. 1998. Effects of habitat differences on feeding behaviors of Japanese monkeys: Comparison between Yakushima and Kinkazan. Primates 39:275-289.

Alexander RM. 1993. The relative merits of foregut and hindgut fermentation. Journal of the Zoological Society of London 231:391-401.

Altmann J. 1974. Observational study of behavior: Sampling methods. Behaviour 49:227-267.

Altmann J. 1980. Baboon Mothers and Infants. Cambridge: Harvard University Press.

Altmann J, Schoeller D, Altmann SA, Muruthi P, Sapolsky RM. 1993. Body size and fatness of free-living baboons reflect food availability and activity levels. American Journal of Primatology 30:149-161.

Anon. Decree gives legal recognition to extension of the Bezà Mahafaly Special Reserve Accessed online at: http://campuspress.yale.edu/bezamahafaly/decree-gives-legal-recognition-to-extension-of-the-beza-mahafaly-special-reserve/

Ashby MF. 1989. On the engineering properties of materials. Acta Metallurgica 37:1273-1293.

Ashby MF. 1999. Materials Selection in Mechanical Design, 2nd edition. Oxford: Butterworth Heinemann.

Axel AC, and Maurer BA. 2011. Lemurs in a complex landscape: Mapping species abundance in tropical dry forests of southwestern Madagascar. American Journal of Primatology 73:38-52

Baker G, Jones LHP, Wardrop ID. 1959. Cause of wear in sheeps' teeth. Nature 184:1583-1584.

Baker G, Jones LHP, Wardrop ID. 1961. Opal phytolith and mineral particles in the rumen of sheep. Australian Journal of Agricultural Research 12:462-471.

Barton R. 1985. Grooming site preferences in primates and their functional implications. International Journal of Primatology 6:519-532.

Beisner BA, Isbell LA. 2009. Factors influencing hair loss among female captive rhesus macaques (*Macaca mulatta*). Applied Animal Behavior Science 119: 91-100.

Bercovitch FB. 1987. Female weight and reproductive condition in a population of olive baboons (*Papio anubis*). American Journal of Primatology 12:189-195.

Berg W, Jolly A, Rambeloarivony H, Andrianome V, Rasamimanana H. 2009. A scoring system for coat and tail condition in ringtailed lemurs, *Lemur catta*. American Journal of Primatology 71:183-190.

Berman CM, Schwartz S. 1988. A nonintrusive method for determining relative body fat in free-ranging monkeys. American Journal of Primatology 14:53-64.

Beyer WN, Connor EE, Gerould S. 1994. Estimates of soil ingestion by wildlife. The Journal of Wildlife Management 58:375-382.

Bishop A. 1962. Control of the hand in lower primates. Annals of the New York Academy of Sciences 102:316-337.

Bjorndal KA, Bolton AB, Moore JE. 1990. Digestive fermentation in herbivores: Effect of food particle size. Physiological Zoology 63:710–721.

Blaxter KL. 1962. The Energy Metabolism of Ruminants. London: Hutchinson.

Borg C, Majolo B, Qarro M, Semple S. 2014. A comparison of body size, coat condition and endoparasite diversity of wild Barbary macaques exposed to different levels of tourism. Anthrozoös 27:49-63.

Boyer DM. 2008. Relief index of second mandibular molars is a correlate of diet among prosimian primates and other euarchontan mammals. Journal of Human Evolution 55:1118-37.

Boyer DM, Evans AR, Jernvall J. 2010. Evidence of dietary differentiation among late Paleocene-early Eocene Plesiadapids (Mammalia, Primates). American Journal of Physical Anthropology 142:194-210.

Buettner-Janusch J, Andrew RS. 1962. The use of the incisors by primates in grooming. American Journal of Physical Anthropology 20:127–129.

Bunn JM, Boyer DM, Lipman Y, St. Clair EM, Jernvall J, Daubechies I. 2011. Comparing Dirichlet normal surface energy of tooth crowns, a new technique of molar shape quantification for dietary inferences, with previous methods in isolation and in combination. American Journal of Physical Anthropology 145:247-261.

Bunn JM, Ungar PS. 2009. Dental topography and diets of four old world monkey species. American Journal of Primatology 71:466-477.

Buss IO. 1990. Elephant life. Fifteen years of high population density. Ames: Iowa State University Press.

Campbell JL. 2003. Malagasy primates: Nutrition and gastrointestinal physiology with an emphasis on fiber utilization. PhD Dissertation, North Carolina State University.

Campbell JL, Eisemann JH, Williams CV, Glenn KM. 2000. Description of the gastrointestinal tract of five lemur species: *Propithecus tattersalli*, *Propithecus verreauxi coquereli*, *Varecia variegata*, *Hapalemur griseus*, and *Lemur catta*. American Journal of Primatology 52:133–142.

Campbell JL, Williams CV, Eisemann JH. 2004. Use of total dietary fiber across four lemur species (*Propithecus verreauxi coquereli*, *Hapalemur griseus griseus*, *Varecia variegata*, and *Eulemur fulvus*): Does fiber type affect digestive efficiency? American Journal of Primatology 64:323-335.

Campbell SE, Cuozzo FP, Sauther ML, Sponheimer M, Ferguson ML. 2012. Nanoindentation of lemur enamel: An ecological investigation of mechanical property variations within and between sympatric species. American Journal of Physical Anthropology 148:178-190.

Chapman CA, Webb T, Fronstin R, Wasserman MD, Santamaria AM. 2005. Assessing dietary protein of colobus monkeys through faecal sample analysis: A tool to evaluate habitat quality. African Journal of Ecology 43:276-278.

Ciochon RL, Piperno DR, Thompson RG. 1990. Opal phytoliths found on the teeth of the extinct ape *Gigantopithecus blacki*: Implications for paleodietary studies. In: Delson E, Tattersall I, Van Couvering J, editors. Paleoanthropology Annuals New York: Garland Publishing. p 47-52.

Clauss M, Franz-Odendaal TA, Brash J, Castell JC, Kaiser T. 2007. Tooth wear in captive giraffes (*Giraffa cameloparadalis*): Mesowear analysis classifies free-ranging specimens as browsers but captive ones as grazers. Journal of Zoo and Wildlife Medicine 38:433-445.

Codron D, Lee-Thorp JA, Sponheimer M, de Ruiter D, Codron J. 2006. Inter- and intrahabitat dietary variability of chacma baboons (*Papio ursinus*) in South African savannas based on fecal δ13C, δ15N, and %N. American Journal of Physical Anthropology 129:204-214.

Constantino PJ, Lucas, PW, Lee JJ-W, Lawn BR, 2009. The influence of fallback foods on great ape tooth enamel. American Journal of Physical Anthropology 140:653-660.

Covert HH, Kay RF. 1981. Dental microwear and diet: Implications for determining the feeding behaviors of extinct primates, with a comment on the dietary pattern of *Sivapithecus.* American Journal of Physical Anthropology 55:331-336.

Crawford GC, Andriafaneva L, Blumenfeld-Jones K, Calaba G, Clarke L, Gray L, Ichino S, Jolly A, Koyama N, Mertl-Millhollen A, Ostpak S, Pride RE, Rasamimanana H, Simmen B, Soma T, Tarnaud L, Tew A, Williams G. 2006a. Bald lemur syndrome and the miracle tree: Alopecia associated with *Leucaena leucocephala* at Berenty Reserve, Madagascar. In: Jolly A, Sussman RW, Koyama N, Rasamimanana H, editors. Ringtailed Lemur Biology. New York, Springer. p 332-342.

Crawford GC, Jolly A, Ostapak SE. 2006b. Kwashiorkor (protein energy malnutrition syndrome) in ringtailed lemurs (*Lemur catta*) at Berenty Reserve, Madagascar. XXI International Primatological Congress, Entebbe, Uganda.

Cuozzo FP. Head BR, Sauther ML, Ungar PS, O'Mara MT. 2014. Sources of tooth wear variation early in life among known-aged wild ring tailed lemurs (*Lemur catta*) at the Bezà Mahafaly Special Reserve. American Journal of Primatology 76:1037-1048.

Cuozzo FP, Sauther ML. 2004. Tooth loss, survival and resource use in wild ring-tailed lemurs (*Lemur catta*): implications for inferring conspecific care in fossil hominids. Journal of Human Evolution 46:623-631.

Cuozzo FP, Sauther ML. 2006a. Severe wear and tooth loss in wild ring-tailed lemurs (*Lemur catta*): a function of feeding ecology, dental structure, and individual life history. Journal of Human Evolution 51:490-505.

Cuozzo FP, Sauther ML. 2006b. Temporal change in tooth size among ring-tailed lemurs (*Lemur catta*) at the Beza Mahafaly Special Reserve, Madagascar: Effects of an environmental fluctuation. In: Jolly A, Sussman RW, Koyama N, Rasamimanana H, editors. Ringtailed Lemur Biology. New York: Springer. p 343-366.

Cuozzo FP, Sauther ML. 2008. Habitat variation in patterns of ring-tailed lemur (*Lemur catta*) gross tooth wear and functional tooth loss indicates ''ecological disequilibrium'' in Madagascar. American Journal of Physical Anthropology 135(S46):85.

Cuozzo FP, Sauther ML. 2012. What is dental ecology? American Journal of Physical Anthropology 140:163-170.

Cuozzo FP, Sauther ML. 2013. Dental evidence suggests evolutionary disequilibrium among sympatric diurnal lemurs in south-western Madagascar. Folia Primatologica 84:262-263.

Cuozzo FP, Sauther ML. 2015. Patterns of dental macrowear in subfossil *Lemur catta* from Ankilitelo Cave, Madagascar: Indications of ecology and habitat use over time. Folia Primatologica 86:140-149.

Cuozzo F, Sauther M, Gould L, Sussman R, Villers L, Lent C. 2010. Dental wear and tooth loss in known-aged, older ring-tailed lemurs (*Lemur catta*): A comparison between wild and captive individuals. American Journal of Primatology 72:1026-1037.

Cuozzo FP, Sauther ML, Yamashita N, Lawler RR, Brockman DK, Godfrey LR, Gould L, Jacky Youssouf IA, Lent C, Ratsirarson J, Richard AF, Scott JR, Sussman RW, Villers LM, Weber MA, Willis G. 2008. A comparison of salivary pH in sympatric wild lemurs (*Lemur catta* and *Propithecus verreauxi*) at Beza Mahafaly Special Reserve, Madagascar. American Journal of Primatology 70:363-371.

Cuozzo FP, Yamashita N. 2006. Impact of ecology of the teeth of extant lemurs: a review of dental adaptations, function, and life history. In: Gould L, Sauther ML, editors. Lemurs: Ecology and Adaptation. New York: Springer. p 67-96.

Danielson DR, Reinhard KJ. 1998. Human dental microwear caused by calcium oxalate phytoliths in prehistoric diet of the lower Pecos region, Texas. American Journal of Physical Anthropology 107:297-304.

Dennis JC, Ungar PS, Teaford MF, Glander KE, 2004. Dental topography and molar wear in *Alouatta palliata* from Costa Rica. American Journal of Physical Anthropology 125:152–161.

DeGusta D. 2002. Comparative skeletal pathology and the case for conspecific care in Middle Pleistocene hominids. Journal of Archaeological Science 29:1435-1438.

DeGusta D. 2003. Aubesier 11 is not evidence for neanderthal conspecific care. Journal of Human Evolution 45:91-94.

Dettwyler KA. 1991. Can paleopathology provide evidence for "compassion?" American Journal of Physical Anthropology 84: 375-384.

Diallo BO, Joly HI, McKey D, Hossaret-McKey M, Chevallier MH. 2007. Genetic diversity of *Tamarindus indica* populations: Any clues on the origin from its current distribution? African Journal of Biotechnology 6:853-860.

Dierenfeld ES, Koontz FW, Goldstein RS. 1992. Feed intake, digestion and passage of the proboscis monkey (*Nasalis larvatus*) in captivity. Primates 33:399-405.

Dufour DL, Sauther M. 2002. Comparative and evolutionary dimensions of energetic of human pregnancy and lactation. American Journal of Human Biology 14:584-602.

Dumont ER. 1995. Enamel thickness and dietary adaptation among extant primates and chiropterans. Journal of Mammalogy 76:1127-1136.

Dunbar RIM. 1983. Structure of gelada baboon reproductive units. IV. Integration at group level. Zeitschrift fur Tierpsychologie 63:265-282.

Dunbar RIM. 1988. Primate Social Systems. Ithaca: Comstock Publishing Associates.

Du Puy DJ, Labat JN, Rabevohitra R, Villiers JF, Bosser J, Moat J. 2002. The Leguminosae of Madagascar. Kew: Royal Botanical Gardens.

Edwards MS, Ullrey DE. 1999a. Effect of dietary fiber concentration on apparent digestibility and digesta passage in non-human primates. I. Ruffed lemurs (*Varecia variegata variegata* and *V. v. rubra*). Zoo Biology 18:529-536.

Edwards MS, Ullrey DE. 1999b. Effect of dietary fiber concentration on apparent digestibility and digesta passage in non-human primates. II. Hindgut- and foregut-fermenting folivores. Zoo Biology 18:537-549.

Eley RM, Strum SC, Muchemi G, Reid GDF. 1989. Nutrition, body condition, activity patterns and parasitism of free-ranging troops of olive baboons (*Papio anubis*) in Kenya. American Journal of Primatology 18: 209-219.

Ellis W, Attard R, Johnston S, Theileman P, McKinnon A, Booth D. 2014. Faecal particle size and tooth wear of the koala (*Phascolarctos cinereus*). Australian Mammalogy 36:90-94.

Evans AR. 2005. Connecting morphology, function and tooth wear in microchiropterans. Biological Journal of the Linnaean Society 85:81-96.

Evans AR, Sanson GD. 1998. The effect of tooth shape on the breakdown of insects. Journal of Zoology 246:391-400

Evans AR, Wilson GP, Fortelius M, Jernvall J. 2007. High-level similarity of dentitions in carnivorans and rodents. Nature 445:78-81.

Feng W, Fang Z, Zhou P, Chang F, Hong Y, Zhang X, Peng H, Xiao Y. 2012. Evidence for lignin oxidation by the giant panda fecal microbiome. PLoS One 7:e50312.

Fish KD, Sauther ML, Loudon JE, Cuozzo FP. 2007. Coprophagy by wild ring-tailed lemurs (*Lemur catta*) in human-disturbed locations adjacent to the Beza Mahafaly Special Reserve, Madagascar. American Journal of Primatology 69:713-718.

Fleagle JG. 1999. Primate Adaptation and Evolution. New York: Academic Press.

Gandara BK, Truelove EL. 1999. Diagnosis and management of dental erosion. Journal of Contemporary Dental Practice 1:1-17.

Gebo DL. 2002. Adapiformes: phylogeny and adaptation. In: Hartwig WC, editor. The Primate Fossil Record. Cambridge: Cambridge University Press. p 21-43.

Gemmill A, Gould L. 2008. Microhabitat variation and its effects on dietary composition and intragroup feeding interactions between adult female *Lemur catta* during the dry season at Beza Mahafaly Special Reserve, southwestern Madagascar. International Journal of Primatology 29:1511–1533.

Gingerich PD, 1974a. Function of pointed premolars in *Phenacolemur* and other mammals. Journal of Dental Research 53:497.

Gingerich PD, 1974b. Dental function in the Paleocene primate *Plesiadapis*. In: Martin, RD, Doyle GA, Walker AC, editors. Prosimian Biology. London: Duckworth. p 531–542.

Gipps JM, Sanson GD. 1984. Mastication and digestion in *Pseudocheirus*. In: Smith AP, Hume ID, editors. Possums and Gliders. Sydney: Australian Mammal Society. p 237-246.

Glowacka H, McFarlin SC, Catlett KK, Mudakikwa A, Bromage TG, Cranfield MR, Stoinski TS, Schwartz GT. 2016. Age-related changes in molar topography and shearing crest length in a wild population of mountain gorillas from Volcanoes National Park, Rwanda. American Journal of Physical Anthropology. Accessed online at: http://onlinelibrary.wiley.com/doi/10.1002/ajpa.22943/epdf.

Godfrey LR, Semprebon GM, Schwartz GT, Burney DA, Jungers WL, Flanagan EK, Cuozzo FP, King SJ. 2005. New insights into old lemurs: The trophic adaptations of the Archaeolemuridae. International Journal of Primatology 26:825-854.

Godfrey LR, Winchester JM, King SJ, Boyer DM, Jernvall J. 2012. Dental topography indicates ecological contraction of lemur communities. American Journal of Physical Anthropology 148:215-227.

Godinot M, Mahbouti M. 1992. Earliest known simian primate found in Algeria. Nature 357:324-326.

Goodman SM, Jungers WL. 2014. Extinct Madagascar: Picturing the Island's Past. Chicago: University of Chicago Press.

Gould L, Sussman RW, Sauther ML. 1999. Natural disasters and primate populations: The effects of a 2-year drought on a naturally occurring population of ring-tailed lemurs (*Lemur catta*) in southwestern Madagascar. International Journal of Primatology 20: 69–84.

Graham EE, Burkart J. 1976. A preliminary analysis of antemortem tooth loss among the Fremont. American Antiquity 41:534-537.

Gregory WK. 1922. The Origin and Evolution of the Human Dentition. Baltimore: Williams and Wilkins.

Hatfield RD, Ralph J, Grabber JH. 1999. Cell wall structural foundations: Molecular basis for improving forage digestibilities. Crop Science 39:27-37.

Head BR. 2011. A longitudinal study of dental wear in ring-tailed lemurs. MA Thesis. University of Arkansas-Fayetteville.

Healy WB, Ludwig TG. 1965. Wear of sheep's teeth. I. The role of ingested soil. New Zealand Journal of Agricultural Research 8:753-62.

Hillson S. 1996. Dental Anthropology. Cambridge: Cambridge University Press.

Hladik CM. 1978. Adaptive strategies of primates in relation to leaf eating. In: GG Montgomery, editor. The Ecology of Arboreal Folivores. Washington: Smithsonian Institution Press. p373-395.

Hoebler C, Devaux MF, Karinthi A, Belleville C, Barry JL. Particle size of solid food after human mastication and *in vitro* simulation of oral breakdown. International Journal of Food Sciences and Nutrition 51:353-366.

Hogue AS. 2004. Molar shear crests as dietary indicators: evidence from primate ecological analogs. American Journal of Physical Anthropology 123(S38):114.

Holecheck JL, Vavre M, Pieper RD. 1982. Methods for determining the nutritive quality of range ruminant diets: A review. Journal of Animal Science 54:363–376.

Hummel J, Findeisen E, Südekum K, Ruf I, Kaiser TM, Bucher M, Clauss M, Codron D. 2010. Another one bites the dust: Faecal silica levels in large herbivores correlate with high-crowned teeth. Proceedings of the Royal Society B 278:1742-1747.

Ichino S, Chatani K, Kawamoto Y, Sato H, Schnöll A, Soma T, Koyama N, Aimi M, Takahata Y. 2013a. Decrease in the body mass of wild ringtailed lemurs at Berenty Reserve in Madagascar with environmental changes. African Study Monographs 34:109-118.

Ichino S, Soma T, Koyama N. 2013b. The impact of alopecia syndrome on female reproductive parameters in ring-tailed lemurs (*Lemur catta*) in Berenty Reserve, Madagascar. In: Masters J, Gamba M, Génin F, editors. Leaping Ahead: Advances in Prosimian Biology, Developments in Primatology: Progress and prospects. New York: Springer. p 377-386.

Isbell LA. 1995. Seasonal and social correlates of changes in hair, skin, and scrotal condition in vervet monkeys (*Cercopithecus aethiops*) of Amboseli National Park, Kenya. American Journal of Primatology 36: 61-70.

Janis C. 1984. Prediction of primate diets from molar wear patterns. In: Chivers DJ, Wood BA, Bilsborough A, editors. Food Acquisition and Processing in Primates. New York: Plenum Press.

Janis CM, Fortelius M. 1988. On the means where by mammals achieve increased functional durability of their dentitions with special reference to limiting factors. Biological Reviews 63:197-230.

Jardine PE, Janis CM, Sahney S, Benton MJ. 2012. Grit not grass: Concordant patterns of early origin hypsodonty in Great Plains ungulates and Glires. Paleogeography, Paleoclimatology, Paleoecology 366:1-10.

Jolly A. 2009a. Coat condition of ringtailed lemurs, Lemur catta at Berenty Reserve, Madagascar: I. Differences by age, sex, density and tourism, 1996-2006. American Journal of Primatology 71:191-198.

Jolly A. 2009b. Coat condition of ringtailed lemurs, *Lemur catta*, at Berenty Reserve, Madagascar: II. Coat and tail alopecia associated with *Leucaena leucocephala*, 2001-2006. American Journal of Primatology 71:199-205.

Jung HG. 2012. Forage Digestibility: The intersection of cell wall lignification and plant tissue anatomy. 23rd Florida Ruminant Nutrition Symposium, January 31-February 1, 2012, University of Florida, Gainesville, Florida.

Jung HG, Allen 1995. Characteristics of plant cell walls affecting intake and digestibility of forages by ruminants. Journal of Animal Science 73:2774-2790.

Jung HG, Deetz DA. 1993. Cell wall lignification and degradability. In: Jung HG, Buxton DR, Hatfield RD, Ralph J, editors. Forage Cell Wall Structure and Digestibility. Madison: ASA-CSSA-SSSA. p315-346.

Junge RE, Sauther ML. 2006. Overview on the health and disease ecology of wild lemurs: conservation implications. In: Gould L, Sauther ML, editors. Lemurs: Ecology and Adaptation. New York: Springer. p 423-440.

Kaifu Y, Kasai K, Townsend GC, Richards LC. 2003. Tooth wear and the “design” of the human dentition: A perspective from evolutionary medicine. Yearbook of Physical Anthropology 46:47-61.

Kay RF. 1975. The functional adaptations of primate molar teeth. American Journal of Physical Anthropology 43:195-216.

Kay RF. 1977. The evolution of molar occlusion in the Cercopithecidae and early catarrhines. American Journal of Physical Anthropology 46:327-352.

Kay RF. 1978. Molar structure and diet in extant Cercopithecidae. In: Butler P, editor. Development, Function, and Evolution of Teeth. London: Academic Press. p 309-339.

Kay RF. 1981. The nut-crackers - a new theory of the adaptations of the Ramapithecinae. American Journal of Physical Anthropology 55:141-151.

Kay RF, Covert HH. 1983. True grit: A microwear experiment. American Journal of Physical Anthropology 61:33-38.

Kay RF, Covert HH. 1984. Anatomy and behavior of extinct primates. In: Chivers DJ, Wood BA, Bilsborough A, editors. Food Acquisition and Processing in Primates. New York: Plenum. p 467-508.

Kay RF, Hiiemae KM. 1974. Jaw movement and tooth use in recent and fossil primates. American Journal of Physical Anthropology 40:227-256.

Kay RF, Hylander WL. 1978. The dental structure of mammalian folivores with special reference to primates and Phalangeroidea (Marsupialia) In: Montgomery GG, editor. The Ecology of Arboreal Folivores. Washington, DC: Smithsonian Institution Press. p 173–191.

Kay RF, Sheine WS. 1979. On the relationship between chitin particle size and digestibility in the primate *Galago senegalensis.* American Journal of Physical Anthropology 50:301-308.

Kay RF, Sussman RW, Tattersall I. 1978. Dietary and dental variations in the genus *Lemur*, with comments concerning dietary-dental correlations among Malagasy primates. American Journal of Physical Anthropology 49:119-128.

King SJ, Arrigo-Nelson SJ, Pochron ST, Semprebon GM, Godfrey LR, Wright PC, Jernvall J. 2005. Dental senescence in a long-lived primate links infant survival to rainfall. Proceedings of the National Academy of Sciences of the United States of America 102:16579-16583.

Kirk EC, Simons EL. 2001. Diets of fossil primates from the Fayum depression of Egypt: A quantitative analysis of molar shearing. Journal of Human Evolution 40:203–229.

Klukkert ZS, Teaford MF, Ungar PS. 2012a. A dental topographic analysis of chimpanzees. American Journal of Physical Anthropology 148:276-284.

Klukkert ZS, Dennis JC, M'Kirera F, and Ungar PS. 2012b. Dental topographic analysis of the molar teeth of primates. In: Bell LS, editor. Forensic Microscopy for Skeletal Tissues: Methods and Protocols. Humana Press: New York. p 145-152.

LaFleur M. 2012. Ecology of ring-tailed lemurs (*Lemur catta*) at the Tsimanampetsotsa National Park, Madagascar: Implications for female dominance and the evolution of lemur traits. PhD Dissertation. University of Colorado at Boulder.

LaFleur M, Sauther ML. 2015. Seasonal feeding ecology of ring-tailed lemurs: A comparison of spiny and gallery forest habitats. Folia Primatologica 86:25-34.

Lambert JE. 1998. Primate digestion: Interactions among anatomy, physiology and feeding ecology. Evolutionary Anthropology 7:8-20.

Lambert JE. 2007a. The biology and evolution of ape and monkey feeding. In: Henke W, Hardt T, Tattersall I, editors. Handbook of Paleoanthropology. New York: Springer-Verlag. p. 1207-1234.

Lambert JE. 2007b. Primate nutritional ecology: Feeding biology and diet at ecological and evolutionary scales. In: Campbell CJ, Fuentes A, Mackinnon KC, Panger M, Bearder SK, editors. Primates in Perspective. New York: Oxford University Press. p 482-495.

Lambert JE, Chapman CA, Wrangham RW, Conklin-Brittain NL. 2004. Hardness of cercopithecine foods: implications for the critical function of enamel thickness in exploiting fallback foods. American Journal of Physical Anthropology 125:363-368.

Lambert JE, Fellner V. 2012. *In vitro* fermentation of dietary carbohydrates consumed by African apes and monkeys: Preliminary results for interpreting microbial and digestive strategy. International Journal of Primatology 33:263-281.

Lanyon JM, Sanson GD. 1986. Koala (*Phascolarctos cinereus*) dentition and nutrition. II. Implications of tooth wear in nutrition. Journal of Zoology 209:169-181.

Larsen RS, Moresco A, Sauther ML, Cuozzo FP. 2011. Field anesthesia of wild ring-tailed lemurs (*Lemur catta*) using Tiletamine-Zolazepam, Medetomidine, and Butorphanol. Journal of Zoo and Wildlife Medicine 42:75-87.

Lebel S, Trinkaus E. 2002. Middle Pleistocene human remains from the Bau de l'Aubesier. Journal of Human Evolution 43:659–685.

Lebel S, Trinkaus E, Faure M, Fernandez P, Guérin C, Richter D, Mercier N, Valladas H, Wagner GA. 2001. Comparative morphology and paleobiology of Middle Pleistocene human remains from the Bau de l'Aubesier, Vaucluse, France. Proceedings of the National Academy of Science of the United States of America 98:11097–11102.

Leighton M. 1993. Modeling dietary selectivity by Bornean orangutans: Evidence for integration of multiple criteria in fruit selection. International Journal of Primatology 14:257-314.

Lentle RG, Hume ID, Stafford KJ, Kennedy M, Springett BP, Haslett SJ. 2003. Observations on fresh forage intake, ingesta particle size and nutrient digestibility in four species of macropod. Australian Journal of Zoology 51:627–636.

Leonard WR. 2000. Human nutritional evolution. In: Stinson S, Bogin B, Huss-Ashmore R, O'Rourke D, editors. Human Biology: An Evolutionary and Biocultural Approach. New York: Wiley-Liss. p 295-343.

Leslie DM, Bowyer RT, Jenks JA. 2007. Facts from feces: Nitrogen still measures up as a nutritional index for mammalian herbivores. The Journal of Wildlife Management 72:1420-1432.

Leslie DM Jr, Starkey EE. 1985. Fecal indices to dietary quality of cervids in old-growth forests. The Journal of Wildlife Management 49:142–146.

Lewis RJ, Kappeler PM. 2005. Seasonality, body condition and timing of reproduction in *Propithecus verreauxi verreauxi* in the Kirindy Forest. American Journal of Primatology 67:347-364.

Logan M, Sanson GD. 2002a. The association of tooth wear with sociality of free-ranging male koalas (*Phascolarctos cinereus* Goldfuss). Australian Journal of Zoology 50:621-626.

Logan M, Sanson GD. 2002b. The effects of tooth wear on the activity pattern of free-ranging koalas (*Phascolarctos cinereus* Goldfuss). Australian Journal of Zoology 50:281-292.

Logan M, Sanson GD. 2002c. The effect of tooth wear on the feeding behavior of free-ranging koalas (*Phascolarctos cinereus*, Goldfuss). Journal of Zoology 256:63-69.

Loudon J, Sauther M, Fish K, Hunter-Ishikawa M, Ibrahim J. 2006. One reserve, three primates: applying a holistic approach to understand the interconnections among ring-tailed lemurs (*Lemur catta*), verreaux's sifaka (*Propithecus verreauxi*), and humans (*Homo sapiens*) at Beza Mahafaly Special Reserve, Madagascar. Ecological and Environmental Anthropology 2:54-74.

Lordkipanidze D, Vekua A, Ferring R, Rightmire GP, Agusti J, Kiladze G, Mouskhelishvili A, Nioradze M., Ponce de Leon S, Tappen M, Zollikofer CPE. 2005. The earliest toothless hominin skull. Nature 434:717-718.

Lordkipanidze D, Vekua A, Ferring R, Rightmire GP, Zollikofer CPE, Ponce de Leon MS, Agusti J, Kiladze G, Mouskhelishvili A, Nioradze M, Tappen M. 2006. A fourth hominin skull from Dmanisi, Georgia. The Anatatomical Record Part A 288A:1146-1157.

Lucas PW. 1991. Fundamental physical properties of fruits and seeds in the diet of Southeast Asian Primates. In: Ehara A, Kimura T, Takenaka O, Iwamoto M, editors. Primatology Today. Amsterdam: Elsevier. p 128-152.

Lucas PW. 2004. Dental Functional Morphology: How Teeth Work. Cambridge: Cambridge University Press.

Lucas PW, van Casteren A, Al-Fadhalah K, Almusallam AS, Henry AG, Michael S, Watzke J, Reed DA, Diekwisch TGH, Strait DS, Atkins AG. 2014. The role of dust, grit and phytoliths in tooth wear. Annales Zoologici Fennici 51:143-152.

Lucas P, Contantino P, Wood B, Lawn B. 2008. Dental enamel as a dietary indicator in mammals. BioEssays 30:374-385.

Lucas PW, Omar R, Al-Fadhalah K, Almusallam AS, Henry AG, Michael S, Thai LA, Watzke J, Strait DS, Atkins AG. 2013. Mechanisms and causes of wear in tooth enamel: Implications for hominin diets. Journal of the Royal Society Interface 10:20120923.

Ludwig TG, Healy WB, Cutress TW. 1966. Wear of sheep's teeth. New Zealand Journal of Agricultural Research 9:157-164.

Lussi A, Jaeggi T, Zero D. 2004. The role of diet in the aetiology of dental erosion. Caries Research. 38S1:34-44.

Maas MC, Dumont ER. 1999. Built to last: the structure, function and evolution of primate dental enamel. Evolutionary Anthropology 8:133-152.

Mackie RI. 2002. Mutualistic fermentative digestion in the gastrointestinal tract: diversity and evolution. Integrative and Comparative Biology 42:319–326.

Madden RH. 2014. Hypsodonty in Mammals: Evolution Geomorphology, and the Role of Earth Surface Processes. Cambridge: Cambridge University Press.

Mainland IL. 2003. Dental microwear in grazing and browsing Gotland sheep (*Ovis aries*) and its implications for dietary reconstruction. Journal of Archaeological Science 30:1513-1527.

Marshall AJ. 2004. The population ecology of gibbons and leaf monkeys across a gradient of Bornean forest types. PhD thesis. Harvard University.

Marshall AJ, Boyko CM, Feilen KL, Boyko RH, Leighton M. 2009. Defining fallback foods and assessing their importance in primate ecology and evolution. American Journal of Physical Anthropology 140:603-614.

Marshall AJ, Wrangham RW. 2007. Evolutionary consequences of fallback foods. International Journal of Primatology 28:1219-1235.

Martin LB, Olejniczak AJ, Maas MC. 2003. Enamel thickness and microstructure in pitheciin primates, with comments on dietary adaptations of the middle Miocene hominoid *Kenyapithecus*. Journal of Human Evolution 45:351-367.

McLeod MN, Minson DJ. 1969. Sources of variation in the *in vitro* digestion of tropical grasses. Journal of the British Grassland Society 24:244-249.

Meissner HH, Zacharias PJK, O'Reagain PJ. 1999. Forage quality (feed value). In: Tainton NM, editor. Veld Management in South Africa. Pietermaritzburg, South Africa: University of Natal Press. p 139–168.

Merceron G, Taylor S, Scott R, Chaimanee Y, Jaeger J. 2006. Dietary characterization of the hominoid *Khoratpithecus* (Miocene of Thailand): Evidence from dental topographic and microwear texture analyses. Naturwissenschaften 93:329-333.

Mertens DR. 1994. Regulation of forage intake. In: Fahey Jr. GC, editor. Forage Quality Evaluation and Utilization. Madison: ASA, CSSA, SSA p450-493.

Miller DS, Sauther ML, Hunter-Ishikawa M, Fish KD, Culbertson H, Cuozzo FP, Campbell TW, Chavey S, Nachreiner R, Rumbeiha W, Stacewicz-Sapantzakis M, Lappin MR. 2007. Biomedical evaluation of free-ranging ring-tailed lemurs (*Lemur catta*) in three habitats at the Beza Mahafaly Special Reserve, Madagascar. Journal of Zoo and Wildlife Medicine 38:201-216.

Millette JB. 2007. Behavioral responses to tooth loss in ring-tailed lemurs (*Lemur catta*) at the Beza Mahafaly Special Reserve, Madagascar. MA thesis. University of Colorado Boulder.

Millette JB, Sauther ML, Cuozzo FP. 2009. Behavioral responses to tooth loss in wild ring-tailed lemurs (*Lemur catta*) at the Beza Mahafaly Special Reserve, Madagascar. American Journal of Physical Anthropology 140:120-134.

Millette JB, Sauther ML, Cuozzo FP, Ness JL. 2012. The impact of dental impairment on ring-tailed lemur food processing performance. American Journal of Physical Anthropology 148:238-248.

Milton K, Demment MW. 1987. Digestion and passage kinetics of chimpanzees fed high and low fiber diets and comparison with human data. Journal of Nutrition 118:1082-1088.

Milton K, Van Soest PJ, Robertson J. 1980. Digestive efficiencies of wild howler monkeys. Physiological Zoology 53:402-409.

Mitchell P. 1990. Social behaviour and communication of koalas. In: Lee AK, Handasyde KA, Sanson GD, editors. Biology of the Koala. Sydney: Surrey Beatty & Sons. p 151–171.

M'Kirera F, Ungar PS. 2003. Occlusal relief changes with molar wear in *Pan troglodytes troglodytes* and *Gorilla gorilla gorilla.* American Journal of Primatology 60:31-41.

Molnar S. 1971. Human tooth wear, tooth function, and cultural variability. American Journal of Physical Anthropology 46:447-454.

Molnar S. 1972. Tooth wear and culture: a survey of tooth functions among some prehistoric populations. Current Anthropology 13:511-526.

Moore KJ, Hatfield RD. 1994. Carbohydrates and forage quality. In: Fahey Jr. GC, editor. Forage Quality Evaluation and Utilization. Madison: ASA, CSSA, SSSA. p 229-280.

Moore KJ, Jung HG. 2001. Lignin and fiber digestion. Journal of Range Management 54:420-430.

Moore KJ, Vogel KP, Hopkins AA, Pedersen JF and Moser LE. 1993. Improving the digestibility of warm-season perennial grasses. Proceedings of the XVI International Grassland Congress. p 447–448.

Nijboer J. 2006. Effect of diet on the feces quality in Javan langur (*Trachypithecus auratus auratus*). Doctoral Thesis. University of Utrecht.

Nishida T. 1976. The bark-eating habits in primates, with special reference to their status in the diet of wild chimpanzees. Folia Primatologica 25:277–287.

National Research Council. 2003. Nutrient Requirements of Nonhuman Primates. Washington DC: National Academies Press.

Novak MA, Meyer JS. 2009. Alopecia: Possible causes and treatments, particularly in captive nonhuman primates. Comparative Medicine 59: 18-26.

Nussey DH, Metherell B, Moyes K, Donald A, Guinness FE, Clutton-Brock TH. 2007. The relationship between tooth wear, habitat quality and late-life reproduction in a wild red deer population. Journal of Animal Zoology 76:402-412.

Oftedal O. 1991. The nutritional consequences of foraging in primates: The relationship of nutrient intakes to nutrient requirements. Philosophical Transactions of the Royal Society of London B. 334:161-170.

Olupot W. 1999. Mass differences among male mangabey monkeys inhabiting logged and unlogged forest compartments. Conservation Biology 14:833-843.

Pereira ME. 1993. Seasonal adjustment of growth rate and adult body weight in ringtailed lemurs. In: Kappeler PM, Ganzhorn JU, editors. Lemur Social Systems and Their Ecological Basis. Plenum: New York. p 205-221.

Pereira ME, Pond CM. 1995. Organization of white adipose tissue in Lemuridae. American Journal of Primatology 35:1-13.

Pereira ME, Strohecker RA, Cavigelli SA, Hughes CL, Pearson DD. 1999. Metabolic strategy and social behavior in Lemuridae. In: Rakotosamimanana B, Rasamimanana H, Ganzhorn J, Goodman S, editors. New Directions in Lemur Studies. New York: Plenum Press. p 93-118.

Perez-Barberia FJ, Gordon IJ. 1998. The influence of molar occlusal surface area on the voluntary intake, digestion, chewing behavior and diet selection of red deer (*Cervus elaphus*). Journal of Zoology 245:307-316.

Pride RE. 2003. The socio-endocrinology of group size in *Lemur catta*. PhD Thesis. Princeton, Princeton University.

Pride RE. 2005. Optimal group size and seasonal stress in ring-tailed lemurs (*Lemur catta*). Behavioral Ecology 16:550-560.

Prinz JF, Lucas PW. 2000. Saliva tannin interactions. Journal of Oral Rehabilitation 27:991-994.

Rabenold D, Pearson OM. 2011. Abrasive, silica phytoliths and the evolution of thick molar enamel in primates with implications for the diet of *Paranthropus boisei.* PLoS ONE 6:e28379. Doi:10.1371/journalpone.0028379.

Rabenold D, Pearson OM. 2014. Scratching the surface: A critique of Lucas et al. (2013)'s conclusion that phytoliths do not abrade enamel. Journal of Human Evolution 74:130-133.

Rasamimanana N, Ratsirarson J, Richard AF. 2012. Influence de la variabilité climatique sur la phenologie de la forêt de la Reserve Speciale de Bezà Mahafaly. Malagasy Nature 6:67–82.

Ratsirarson J. 2003. Reserve Speciale de Beza Mahafaly. In: Goodman SM, Benstead JP, editors. The Natural History of Madagascar. Chicago: The University of Chicago Press. p 1520-1525.

Ratsirarson J, Randrianarisoa J, Ellis E, Emady J, Efitroarany, Ranaivonasy J, Razanajaonarivalona EG, Richard AF. 2001. Serie Sciences Biologiques: No 18 - Beza Mahafaly: Ecologie et Realites Socio-Economiques. Antananarivo: Recherches Pour Le Developpement.

Remis MJ. 1997. Feeding ecology and position behavior of western lowland gorillas (*Gorilla gorilla gorilla*) in Central African Republic. PhD dissertation, Yale University.

Rensberger JM. 1973. An occlusion model for mastication and dental wear in herbivorous mammals. Journal of Paleontology 47:515-528.

Reinhard KJ, Danielson DR. 2005. Pervasiveness of phytoliths in prehistoric southwestern diet and implications for regional and temporal trends for dental microwear. Journal of Archaeological Science 32:981-988.

Richard AF, Dewar RE, Schwartz M, Ratsirarson J. 2000. Mass change, environmental variability and female fertility in wild *Propithecus verreauxi*. Journal of Human Evolution 39:381-391.

Robbins CT, 1983. Wildlife Feeding and Nutrition. New York: Academic Press.

Robbins CT, Hanley TA, Hagerman AE, Hjeljord, Baker DL, Schwartz CC, Mautz WW. 1987. Role of tannins in defending plants against ruminants - reduction in protein availability. Ecology 68:98-107.

Rothman JM, Chapman CA, Van Soest PJ. 2012. Methods in primate nutritional ecology: A users guide. International Journal of Primatology 33:542-566.

Ryan AS. 1979. Tooth sharpening in primates. Current Anthropology 20:121-122.

Saha U, Sonon L, Hancock D, Hill N, Stewart L, Heusner G, Kissel DE. 2013. Common terms used in animal feeding and nutrition. University of Georgia Cooperative Extension. College of Agricultural and Environmental Sciences; College of Family and Consumer Sciences. Accessed online at: http://extension.uga.edu/publications/files/pdf/B%201367_2.PDF, August 20, 2015.

Sanson GD, Kerr SA, Gross KA. 2007. Do silica phytoliths really wear mammalian teeth? Journal of Archaeological Science 34:526-531.

Sauther ML. 1992. Effect of reproductive state, social rank and group size on resource use among free-ranging ring-tailed lemurs (*Lemur catta*) of Madagascar. Unpublished Ph.D. Dissertation. Department of Anthropology, Washington University.

Sauther ML. 1998. The interplay of phenology and reproduction in ring-tailed lemurs: implications for ring-tailed lemur conservation. Folia Primatologica Suppl 69:309-320.

Sauther ML, Cuozzo FP. 2008. Somatic and dental variation in living, wild ring-tailed lemurs. Folia Primatologica 79:58-78.

Sauther ML, Cuozzo FP. 2009. The impact of fallback foods on wild ring-tailed lemur biology: A comparison of intact and anthropogenically disturbed habitats. American Journal of Physical Anthropology 140:671-686.

Sauther ML, Cuozzo FP. 2012. Understanding Eocene primate paleobiology using a comprehensive analysis of living primate ecology, biology and behavior (a special issue: *Messel and the Eocene* – Proceedings of the 22nd annual Senkenberg Conference, Frankfurt, Germany). Paleobiodiversity and Paleoenvironments. 92:573-583.

Sauther ML, Cuozzo FP. 2013. Toothcomb function and use in wild ring-tailed lemurs: Implications for the evolution of the prosimian toothcomb. Bulletin of the International Association for Paleodontology 7:123.

Sauther ML, Cuozzo FP, Larsen R, Moresco A, Norris A, Simai A. Singleton C, Smith A, Stegmann GF, Weber M, Jacky Youssouf I. 2012. Climatic effects on wild ring-tailed lemur biomedical values at the Beza Mahafaly Special Reserve, Madagascar. American Journal of Physical Anthropology S54:259.

Sauther ML, Fish KD, Cuozzo FP, Miller DS, Hunter-Ishikawa M, Culbertson H. 2006. Patterns of health, disease and behavior among wild ringtailed lemur, *Lemur catta*: Effects of habitat and sex. In: Jolly A, Sussman RW, Koyama N, Rasamimanana H, editors. Ringtailed Lemur Biology. New York: Springer. p 313-331.

Sauther ML, Sussman RW, Cuozzo FP. 2002. Dental and general health in a population of wild ring-tailed lemurs: a life history approach. American Journal of Physical Anthropology 117:122-132.

Sauther ML, Sussman RW, Gould L. 1999. The socioecology of the ringtailed lemur: Thirty-five years of research. Evolutionary Anthropology 8:120-132.

Schmidt DA, Kerley MS, Dempsey JL, Porton IJ, Porter JH, Griffen ME, Ellersieck MR, Sadler WC. 2005a. Fiber digestibility by the orangutan (*Pongo abelii*): *In vitro* and in vivo. Journal of Zoo and Wildlife Medicine 36:71-580

Schmidt DA, Kerley MS, Porton IJ, Porter, JH, Dempsey JL, Griffin ME, Ellersieck MR, Sadler WC. 2005b. Fiber digestibility by black lemurs (*Eulemur macaco macaco*). Journal of Zoo and Wildlife Medicine 36:204-211.

Schofield RMS, Nesson MH, Richardson KA. 2002. Tooth-hardness increases with zinc content in mandibles of young leaf cutter ants. Naturwissenschaften 89:579-583.

Schwarm A, Schweigert M, Ortmann S, Hummel J, Janssens GPJ, Streich WJ, Clauss M. 2009. No easy solution for the fractionation of faecal nitrogen in captive wild herbivores: Results of a pilot study. Journal of Animal Physiology and Animal Nutrition 93:596-605.

Seligsohn D. 1977. Analysis of species-specific molar adaptations in strepsirrhine primates. Contributions to Primatology 11:1-116.

Sheine WS. 1979. The effect of variations in molar morphology on masticatory effectiveness and digestion of cellulose in prosimian primates. PhD Dissertation. Department of Anthropology, Duke University.

Sheine WS, Kay RF. 1977. An analysis of chewed food particle size and its relationship to molar structure in the primates *Cheirogaleus medius* and *Galago senagalensis* and the insectivoran *Tupaia glis*. American Journal of Physical Anthropology 47:15–20.

Shellis RP, Beynon AD, Reid DJ, Hiiemae KM. 1998. Variations in molar enamel thickness among primates. Journal of Human Evolution 35:597-522.

Simmen B, Hladik A, Ramasiarisoa P. 2003. Food intake and dietary overlap in native *Lemur catta* and *Propithecus verreauxi* and introduced *Eulemur fulvus* at Berenty, Southern Madagascar. International Journal of Primatology 24:949-968.

Simmen B, Sauther ML, Soma T, Rasamimanana HR, Sussman RW, Jolly A, Tarnaud L, Hladik A. 2006. Plant species fed on by *Lemur catta* in gallery forests of the southern domain of Madagascar. In: Jolly A, Sussman RW, Koyama N, Rasamimanana H, editors. Ringtailed Lemur Biology. New York: Springer. p 55-68.

Skipland JP. 1974. Ingestion of grit by bighorn sheep. The Journal of Wildlife Management 38:880-883.

Smith BH. 1984. Patterns of molar wear in hunter-gatherers and agriculturalists. American Journal of Physical Anthropology 63:39-56.

Smith JD, Genoways HH, Jones JK. 1977. Cranial and dental anomalies in three species of platyrrhine monkeys from Nicaragua. Folia Primatologica 28:1-42.

Stewart ML, Slavin J. Particle size and fraction of wheat bran influence short-chain fatty acid production *in vitro.* British Journal of Nutrition 102:1404-1407.

Strait SG. 1993a. Differences in occlusal morphology and molar size in frugivores and faunivores. Journal of Human Evolution 25:471–484.

Strait SG. 1993b. Molar morphology and food texture among small-bodied insectivorous mammals. Journal of Mammalogy 74:391–402.

Stevens CE. 1988. Comparative Physiology of the Vertebrate Digestive System. New York: Cambridge University Press.

Stevens CE, Hume ID. 1995. Comparative Physiology of the Vertebrate Digestive System. 2nd edition. New York: Cambridge University Press.

Sussman RW. 1991. Demography and social organization of free-ranging *Lemur catta* in the Beza Mahafaly Reserve, Madagascar. American Journal of Physical Anthropology 84:43–58.

Sussman RW, Rakotozafy A. 1994. Plant diversity and structural analysis of a tropical dry forest in southwestern Madagascar. Biotropica 26:241-254.

Sussman RW, Ratsirarson J. 2006. Beza Mahafaly Special Reserve: a research site in southwestern Madagascar. In: Jolly A, Sussman RW, Koyama N, Rasamimanana H, editors. Ringtailed Lemur Biology. New York: Springer. p 43-51.

Sussman RW, Richard AF, Ratsirarson J, Sauther ML, Brockman DK, Gould L, Lawler R, Cuozzo FP. 2012. Beza Mahafaly Special Reserve: Long-term research on lemurs in southwestern Madagascar. In: Kappeler PM, Watts DP, editors. Long-Term Field Studies of Primates. New York: Springer-Verlag Berlin Heidelberg. p 45-66.

Swindler DR. 2002. Primate Dentition: An Introduction to the Teeth of Non-human Primates. Cambridge: Cambridge University Press.

Szalay FS. 1968. The beginnings of primates. Evolution 22:19–36.

Tappen M. 2009. The wisdom of the aged and out of Africa I. In: Shea JJ, Lieberman DE, editors. Transitions in prehistory: essays in honor of Ofer Bar-Yosef. Oakville: Oxbow Books. p 33-53.

Tattersall I, Delson E, and Van Couvering J. 1988. Teeth. In: Tattersall I, Delson E, Van Couvering J, editors. Encyclopedia of Human Evolution and Prehistory. New York: Garland Publishing.

Teaford MF. 1983. Differences in molar wear gradient between adult macaques and langurs. International Journal of Primatology 4:427-444.

Teaford MF, Glander KE. 1996. Dental Microwear and diet in a wild population of mantled howling monkeys (*Allouatta palliata*). In: Norconk MA, Rosenberger AL, Garber PA, editors. Adaptive Radiations of Neotropical Primates, New York: Plenum Press.

Teaford M, Maas M, Simons EL. 1996. Dental Microwear and microstructure in early Oligocene primates from the Fayum, Egypt: implications for diet. American Journal of Physical Anthropology 101:527-544.

Teaford MF, Oyen OJ. 1989. Differences in the rate of molar wear between monkeys raised on different diets. Journal of Dental Research 68:1513-1518.

Tutin CEG, Fernandez M, Rogers ME, Williamson EA, McGrew WC. 1991. Foraging profiles of sympatric lowland gorillas and chimpanzees in the Lopé reserve, Gabon. Philosophical Transactions of the Royal Society B 334:179-186.

Tutin CEG, Fernandez M. 1993. Composition of the diet of chimpanzees and comparisons with that of sympatric lowland gorillas in the Lope Reserve, Gabon. American Journal of Primatology 30:195-122.

Ulhaas L, Kullmer O, Schrenk F, Henke W. 2004. A new 3-d approach to determine functional morphology of cercopithecoids molars. Annals of Anatomy 186:487-493.

Ungar PS. 1996. Relationship of incisor size to diet and anterior tooth use in sympatric Sumatran anthropoids. American Journal of Primatology 38:145-156.

Ungar P. 1998. Dental allometry, morphology and wear as evidence for diet in fossil primates. Evolutionary Anthropology 6:205-217.

Ungar PS. 2004. Dental topography and diets of *Australopithecus afarensis* and early *Homo*. Journal of Human Evolution 46:605-622.

Ungar PS. 2005. Reproductive fitness and tooth wear: Milking as much as possible out of dental topographic analysis. Proceedings of the National Academy of Sciences of the United States of America 102:16533-16534.

Ungar PS. 2007. Dental topography and human evolution with comments on the diets of *Australopithecus africanus* and *Paranthropus.* In: Bailey SE, Hublin JJ, editors. Dental Perspectives on Human Evolution: State-of-the-Art Research in Dental Paleoanthropology. Dordrecht: Springer. p 321-344.

Ungar PS. 2014. Teeth: A Very Short Introduction. New York: Oxford University Press.

Ungar PS. 2015. Mammalian dental function and wear: A review. Biosurface and Biotribology 1:25-41.

Ungar PS, Bunn JM. 2008. Primate dental topographic analysis and functional morphology. In: Irish JD, Nelson GC, editors. Technique and Application in Dental Anthropology. New York: Cambridge University Press. p 253-265.

Ungar PS, Kay RF. 1995. The dietary adaptations of European Miocene catarrhines. Proceedings of the National Academy of Sciences of the United States of America 92:5479–5481.

Ungar PS, M'Kirera F. 2003. A solution to the worn tooth conundrum in primate functional anatomy. Proceedings of the National Academy of Sciences of the United States of America.100:3874-3877.

Ungar PS, Teaford MF, Glander KE, Pastor RF. 1995. Dust accumulation in the canopy: A potential cause of dental microwear in primates. American Journal of Physical Anthropology 107:93-99.

Ungar P, Williamson M. 2000. Exploring the effects of tooth wear on functional morphology: A preliminary study using dental topographic analysis. Paleo-electronica: http://paleo-electronica.org/2000_1/gorilla/main.htm

Urquiza-Haas T, Serio-Silva JC, Hernàndez-Salazar LT. 2008. Traditional nutritional analyses of figs overestimates intake of most nutrient fractions: A study of *Ficus perforate* consumed by howler monkeys (*Alouatta palliata mexicana*). American Journal of Primatology 70:432-438.

Van Der Bilt A, Abbink JH, Mowlana F, Heath MR. 1993. A comparison between data analysis methods concerning particle size distributions obtained by mastication in man. Archives of Oral Biology 38:163-167.

Van Soest PJ. 1994. Nutritional Ecology of the Ruminant, 2nd edition. Ithaca: Cornell University Press.

Van Soest PJ, McQueen RW. 1973. The chemistry and estimation of fibre. Proceedings of the Nutrition Society 32:123–130.

Van Valkenburgh B, Teaford MF, Walker A. 1990. Molar microwear and diet in large carnivores Journal of Zoology 22:319-340.

Venkataraman VV, Glowacka H, Fritz J, Clauss M, Seyoum C, Nguyen N, Fashing PJ. 2014. Effects of dental fracture toughness and dental wear on chewing efficiency in geladas (*Theropithecus gelada*). American Journal of Physical Anthropology 155:17-32.

Verheyden H, Aubry L, Merlet J, Petibon P, Chauveau-Duriot B, Guillon N, Duncan P. 2011. Faecal nitrogen, an index of diet quality in roe deer *Capreolus capreolus*? Wildlife Biology 17:166-175.

Verrett RG. 2001. Analyzing the etiology of an extremely worn dentition. Journal of Prosthodontology 10:224-233.

Walker A, Hoeck HN, Perez L. 1978. Microwear of mammalian teeth and an indicator of diet. Science 201:908-910.

Wehausen JD. 1995. Fecal measures of diet quality in wild and domestic ruminants. Journal of Wildlife Management 59:816-823.

Whitelaw, D. 2010. Ecological impacts of forest disturbance on ring-tailed lemurs (*Lemur catta*) in the Beza Mahafaly Special Reserve Region: Implications for conservation in an altered landscape. PhD Dissertation. Department of Anthropology, University of Colorado Boulder.

Whitten A. Byrne RW, Barton RA, Waterman PG, Henzi SP. 1991. Dietary and foraging strategies of baboons. Philosophical Transactions of the Royal Society of London B. 344:187-197.

de Wijk RA, Prinz JF. 2005 The role of friction in perceived oral texture. Food Quality and Preference 16:121-129.

Williamson EA, Tutin CEG, Rogers ME, Fernandez M. 1990. Composition of the diet of lowland gorillas at Lope in Gabon. American Journal of Primatology 21:265-277.

Winchester JM, Boyer DM, St. Clair EM, Gosselin-Ildari AD, Ledogar JA. 2014. Dental topography of platyrrhines and prosimians: Convergence and contrasts. American Journal of Physical Anthropology 153:29-44.

Yamakoshi G. 1998. Dietary responses to fruit scarcity of Wild Chimpanzees at Bossou, Guinea: Possible implications for ecological importance of tool use. American Journal of Physical Anthropology 106:283-295.

Yamashita N. 1996. Seasonal and site specificity of mechanical dietary patterns in two Malagasy lemur families (Lemuridae and Indriidae). International Journal of Primatology 17:355-387.

Yamashita N. 1998a. Functional dental correlates of food properties in five Malagasy lemur species. American Journal of Physical Anthropology 106:169-188.

Yamashita N. 1998b. Molar morphology and variation in two Malagasy lemur families (Lemuridae and Indriidae) Journal of Human Evolution. 35:137-162.

Yamashita N. 2000. Mechanical thresholds as a criterion for food selection in two prosimian primate species. In: Spatz HC, Speck T, editors. Proceedings of the 3rd Plant Biomechanics Conference, Freiburg-Badenweiler August 27th to September 2nd. Stuttgart: Thieme Verlag. p 590-595.

Yamashita N. 2002. Diets of two lemur species in different microhabitats in Beza Mahafaly Special Reserve Madagascar. International Journal of Primatology 23:1025-1051.

Yamashita N. 2003. Food procurement and tooth use in two sympatric lemur species. American Journal of Physical Anthropology 121:125-133.

Yamashita N. 2008a. Chemical properties of the diets of two lemur species in southwestern Madagascar. International Journal of Primatology 29:339–364.

Yamashita N. 2008b. Food physical properties and their relationship to morphology: the curious case of kily. In: Vinyard C, Ravosa MJ, Wall CE, editors. Primate Craniofacial Function and Biology. New York: Kluwer Academic Press. p 387-406.

Yamashita N, Cuozzo FP, Sauther ML. 2012. Interpreting food processing through dietary mechanical properties: A *Lemur catta* case study. American Journal of Physical Anthropology 148:205-214.

Yamashita N, Cuozzo FP, Sauther ML, Fitzgerald E, Riemenschneider A, Ungar P. 2015a. Mechanical food properties and dental topography differentiate three populations of *Lemur catta* in southwest Madagascar. doi:10.1016/j.jhevol.2015.09.006

Yamashita N, Sauther ML, Cuozzo FP, Youssouf JIA. 2015b. Beyond the gallery forest: Contrasting habitat and diet in *Lemur catta* Troops at Bezà Mahafaly Special Reserve. Folia Primatologica 86:35-43.

Yeager CP. Kool K. 2000. The behavioral ecology of Asian colobines. In: Whitehead PF, Jolly CJ, editors. Old World Monkeys. Cambridge: Cambridge University Press. p 496-521.

Weiss KE. 2011. The 5% solution: How do we make decisions in science? Evolutionary Anthropology 20:81-84.

Zhang P. 2011. A non-invasive study of alopecia in Japanese macaques *Macaca fuscata*. Current Zoology 57:26-35.

APPENDIX

A: BEHAVIORAL ETHOGRAM

Definitions are based upon an ethogram for ring-tailed lemurs developed and utilized by Sauther (1992) and further refined by Millette (2007) with the addition of **Stink Fight (SF)** and **Displace (DP)** as a separate behaviors.

No distinction is made between State and Event behaviors.

Anogenital Scent Mark (AG) - Focal animal marks a target object (most frequently a small branch or tree) utilizing scent glands in the anal/genital region. Females mark by placing the vulva upon the target object while males press the testicles upon the target. Individuals may support much of their weight using their forelimbs while engaging in AG. AG is frequently preceded by inspecting and smelling the target object. Behavior has been combined into the category of "Other" for purpose of analysis.

Approach (AP) - Focal animal locomotes within one meter of a social partner, or a social partner locomotes within one meter of the focal animal. AP is a directional behavior. *DIRECTED* is scored if the focal animal locomotes to within one meter of a social partner. *RECEIVED* is scored if a social partner locomotes to within one meter of the focal animal." Behavior has been combined into the category of "Other" for purpose of analysis.

Bite (BI) - The mouth is used to forcefully and agonistically grasp a social partner with the dental apparatus. Biting behavior is directional, and may be *DIRECTED* by the animal against a social partner or may be *RECEIVED* from a social partner. Behavior has been combined into the category of "Agonism" for purpose of analysis.

Carpal Scent Mark (CA) - Focal animal marks a target object (most frequently a small branch or tree) by first charging the carpal spurs with the sternal glands and subsequently pulling the carpal spurs across the target, often with enough force to notch the bark of a small tree. Individuals frequently repeat pulling of the spurs across the target object. CA is most often performed from a vertical posture where the focal animal's weight is supported by the hindlimbs while the carpal spurs are utilized in marking. CA is frequently preceded by inspecting and smelling the target object. Behavior has been combined into the category of "Other" for purpose of analysis.

Cuff (CU) - Animal makes contact with the palm of the hand or wrist against the head or face of a social partner. Cuffing behavior is directional, and may be *DIRECTED* by the animal towards a social partner or may be *RECEIVED* from a social partner. Behavior has been combined into the category of "Agonism" for purpose of analysis.

Chase (CH) - Animal locomotes towards and pursues a social partner who withdraws and departs away from the animal directing the chase. The directing animal peruses the social partner following withdraw for at least one meter. Chasing is a directional behavior and may be *DIRECTED* by the focal animal towards a social partner or may be *RECEIVED* from a social partner. Behavior has been combined into the category of "Agonism" for purpose of analysis.

Defecate (DF) - Focal animal excretes feces through the anus. Behavior has been combined into the category of "Other" for purpose of analysis.

Displace (DP) - Animal moves towards another animal and causes that animal to vacate its location. This is a directional behavior, and may be *DIRECTED* by the focal animal towards a social partner or may be *RECEIVED* from a social partner. Behavior has been combined into the category of "Agonism" for purpose of analysis.

Drink (DK) - Focal animal ingests water through the mouth. Water may be ingested either by licking a water source or by using the hand to "cup" water from a source. DK may be scored for either ingestion of water from a concentrated source such as a bucket of water or a puddle, or by licking items as dew-wet leaves or damp concrete. Behavior has been combined into the category of "Other" for purpose of analysis.

Feed Mouth (FM) - Focal animal selects, manipulates, processes, and ingests food items. Animal utilizes the mouth to manipulate, process, and ingest foods. Hand use may accompany FM, but is limited to manipulating stems, vines, or other structures as to bring that item to the mouth -- the hand does not directly contact the food item. Behavior commences with processing a food item or mastication. FM behavior is broken once the focal animal has been observed to cease mastication. Behavior has been combined into the category of "Feed" for purpose of analysis.

Feed Hand (FH) - Focal animal selects, manipulates, processes, and ingests food items. Hands are utilized to manipulate foods, including: removal of food items from the plant, processing food items, and bringing food items to the mouth for processing, mastication and ingestion. Hands are in direct contact with the part of the food item consumed. FH is scored if the animal is grasping a food item by the stem, but the hand also contacts the item. Behavior commences with processing of food item or mastication. Behavior has been combined into the category of "Feed" for purpose of analysis.

Fight (FI) - Agonistic encounter between the focal animal and a social partner involving biting, grappling, chasing and physical contact. FI is a directional behavior. *DIRECTED* is scored if focal animal unidirectionally commences FI towards a social partner. *RECEIVED* is scored if a social partner unidirectionally commences FI towards the focal animal. *MUTUAL* is scored if

both animals initiate fighting simultaneously. Behavior has been combined into the category of "Agonism" for purpose of analysis.

Food Steal (FS) - Animal displaces an animal engaged in feeding and then consumes that animal's food item. FS may also involve taking a food item from an animal through use of agonistic behaviors such as fighting, cuffing or biting. FS is a directional behavior. *DIRECTED* is scored if focal animal takes a social partners food. *RECEIVED* is scored if a social partner takes the focal animal's food item. Behavior has been combined into the category of "Agonism" for purpose of analysis.

Forage (FG/FO) - Focal animal searches for and/or inspects food items. Individual actively searches for food items while being close proximity to a food source (e.g. a *Tamarindus* tree, kililo patch, trash pit, *Tarenna* bush, etc.). Animal may visually search for food, locomote between items or patches of food items, and inspect, sniff or taste food items. Animal does not ingest food during FO activities. FO becomes FM or FH following the first processing or ingestion of a food item.

Groom (GR) - Individual is observed pass the toothcomb or anterior portion of the muzzle (in cases of toothcomb loss) through the fur. Grooming is most frequently observed to feature a rhythmic anterior-posterior movement of the head during which the toothcomb is used to remove ectoparasites. Grooming behavior is frequently marked by licking in addition to passing the toothcomb through the fur. Grooming is a directional behavior. Social grooming may consist of *RECEIVED* grooming (R) in which the focal animal is subject to grooming by conspecific partner, *DIRECTED* grooming (D) where the focal animal grooms a conspecific partner, and *MUTUAL* grooming (M) in which the focal directs grooming towards a social partner while simultaneously receiving grooming.

Jump Fight (JF) - Agnostic encounter in which one individual attacks another through jumping. Behavior has been combined into the category of "Agonism" for purpose of analysis.

Mate (MA) - Focal animal engages in copulatory behavior with a conspecific social partner and most frequently includes intromission of the penis into the vulva. Intrasexual mounting behaviors are also included within MA. *THIS BEHAVIOR WAS NOT OBSERVED.*

Movement (MT) - Individual locomotes across a distance not less than one meter but no greater than 10 meters. Movement includes all forms of locomotion (e.g. walking, leaping, climbing, running and, galloping), but does not include minor repositioning or postural changes while engaged in other behaviors.

Nip (NP) - Animal uses the anterior dentition to quickly and lightly grasp a social partner with the anterior portion of the dental apparatus. Nipping behavior is directional, and may be *DIRECTED* by the animal towards a social partner or may be *RECEIVED* from a social partner. Behavior has been combined into the category of "Agonism" for purpose of analysis.

Nose Poke (NO) - Focal animal and a social partner briefly touch noses. Nose poking is most frequently seen as two animals approach and "greet." NO is a *MUTUAL* social behavior.

Play (PY) - Individual engages in behaviors which may include mock fighting, non-agonistic chasing, light wrestling, jumping, and high-velocity movement or travel. Play may be a social behavior or a solitary behavior. Social play is typically accompanied by a "play face," marked by a wide gape of the mouth without bearing of teeth. Behavior has been combined into the category of "Other" for purpose of analysis.

Rest (RE) - Focal animal is not engaged in any other defined behavior. Resting behavior features a lack of any locomotor activity. Focal animal may be in physical contact with social partner while engaging in RE. If an observed behavior cannot be categorized, RE is assigned.

Slap (SL) - Animal makes contact with the palm of the hand against a social partner. This behavior is directional, and may be *DIRECTED* by the animal towards a social partner or may be *RECEIVED* from a social partner. Behavior has been combined into the category of "Agonism" for purpose of analysis.

Sleep (SL) - Focal animal is motionless with eyes closed. SL may be scored with the eyes slightly open (e.g. the pupil is obscured) as individuals frequently only partially close their eyes. If eyes open while attempting to determine closure status or the eyes are obscured RE is scored. Animal may be in physical contact with a social partner while engaging in SL. Behavior has been combined into the category of "Rest" for purpose of analysis.

Stand (ST) - Individual maintains a vertical posture in which the posterior limbs are used to support the entire weight of the body and the anterior limbs are not in contact with the ground or other substrate. Standing may be accompanied by visual monitoring. Standing bouts are short and appear to be an *event behavior*. Behavior has been combined into the category of "Other" for purpose of analysis.

Stink Fight (SF) - An agonistic encounter in which an animal rubs the carpal spurs against the sternal glands, "charges" the tail with the carpal spurs, and subsequently directs the tail towards the face of a social partner. Direction of the tail towards a social partner is also accompanied by movement of ears to a posterior position in which they lie flat against the head. Directing SF is limited to male individuals, although males may attempt to SF with females. SF is a directional behavior. *DIRECTED* is scored if focal animal unidirectionally commences SF towards a social partner. *RECEIVED* is scored if a social partner unidirectionally commences SF against the focal animal. *MUTUAL* is scored if both animals initiate SF simultaneously. Behavior has been combined into the category of "Agonism" for purpose of analysis.

Sun (SU) - Focal animal exposes the ventral surface towards the sun in a stereotyped vertical sitting position where the arms are placed laterally to the torso and legs. Sunning animals often close the eyes or turn the head away from the sun. In such cases of closed eyes, SL is not scored. SU may be scored if one arm is not placed laterally to the torso or inside legs, if as to maintain balance or prevent a fall. SU occurs most frequently during cold periods. Behavior has been combined into the category of "Other" for purpose of analysis.

Travel (TL) - Individual locomotes for a distance greater than 10 meters. Movement includes all forms of locomotion (e.g. walking, leaping, climbing, running and, galloping). Individual may pause for a short period (no more than 2-3 seconds) during a travel bout, as to engage in activities such as locating the next point of movement or viable substrate, prepare to leap, or wait for another individual to clear from the line of movement. Behavior has been combined into the category of "Move" for purpose of analysis.

Urinate (UR) - Focal animal excretes urine through the urethral opening. Behavior has been combined into the category of "Other" for purpose of analysis.

Watch Observer (WO) - Individual visually monitors and/or visually tracks the observer. Visual monitoring must be clearly directed towards the observer. Short "glances" directed towards the observer or monitoring in which the animal monitors several visual targets in rapid succession are not included within this behavioral category. Behavior has been combined into the category of "Other" for purpose of analysis.

B. TOPOGRAPHIC SCORES

Table B.1. Topographic scores for all individuals.

		Topographic Measure						
ID	**Group**	**Slope**	**Angularity**	**Relief**	**2DAverage**	**2DSum**	**3DAverage**	**3DSum**
226	Black	33.962	88.643	1.440	8.275	8.275	11.913	11.913
291	Black	31.654	89.051	1.447	13.302	106.418	18.944	151.549
318	Black	39.125	89.163	1.646	14.217	113.733	23.125	185.004
331	Black	40.124	88.856	1.585	14.411	115.284	22.475	179.802
345	Black	40.272	88.840	1.599	15.682	125.458	24.836	198.684
217	Blue	31.677	89.180	1.505	14.442	115.539	21.299	170.396
218	Blue	32.520	88.795	1.468	14.620	116.956	20.823	166.586
332	Blue	37.558	89.163	1.627	14.049	112.393	22.425	179.403
348	Blue	26.771	88.696	1.416	14.528	72.641	19.597	97.985
154	Orange	32.078	88.661	1.415	12.666	63.329	17.750	88.749
268	Orange	40.419	88.880	1.629	13.275	106.198	21.400	171.202
273	Orange	31.115	88.724	1.440	14.152	113.214	20.238	161.902
307	Orange	40.289	88.981	1.703	13.363	106.908	22.344	178.751
308	Orange	34.540	88.779	1.549	12.555	100.443	18.990	151.916
316	Orange	40.933	88.880	1.652	11.995	95.956	19.641	157.132
341	Orange	40.829	88.875	1.633	12.663	101.308	20.425	163.399
343	Orange	38.616	88.718	1.584	13.584	108.671	21.322	170.579
368	Orange	31.033	88.760	1.447	12.574	100.594	17.870	142.963
181	Pink	30.368	88.756	1.423	12.035	72.208	16.726	100.354
38	Red	35.485	88.866	1.527	13.297	93.079	20.079	140.553
44	Red	36.195	88.711	1.593	12.751	102.010	20.005	160.043
231	Red	32.315	88.725	1.463	12.703	101.622	18.512	148.099
347	Red	42.097	88.784	1.722	12.836	102.691	21.696	173.564
339	Teal	41.356	88.841	1.679	13.303	106.420	21.959	175.673
340	Teal	30.567	88.662	1.396	12.750	101.998	17.600	140.802
155	Yellow	26.688	88.764	1.367	11.073	88.587	14.930	119.442
172	Yellow	32.205	88.997	1.477	8.285	49.712	12.041	72.244
223	Yellow	34.885	88.752	1.504	14.795	118.359	22.028	176.222
230	Yellow	32.401	88.584	1.464	13.458	94.209	19.333	135.334
319	Yellow	30.985	88.680	1.371	12.327	73.964	16.557	99.343
346	Yellow	42.566	88.774	1.686	13.794	110.349	22.889	183.108
	Average	**35.214**	**88.824**	**1.531**	**13.025**	**96.727**	**19.670**	**146.861**

C. INDIVIDUAL FECAL NUTRTITIONAL DATA.

Table C.1. Fecal Nutrition Data for All Individuals for Dry and Wet Seasons.

	% NDF			% ADF			% ADL			% Ash			% Protein		
ID	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**
217	12	53.21	8.44	12	38.42	7.49	12	11.04	3.67	12	3.26	1.61	12	13.38	4.29
218	6	62.81	9.83	6	47.58	9.42	6	13.09	3.61	6	7.20	5.24	6	12.56	4.56
226	10	53.05	7.16	10	40.76	6.35	10	8.73	2.42	10	6.59	2.53	10	14.83	3.16
23	10	54.09	10.44	10	41.48	8.77	10	8.35	2.79	10	12.33	6.49	9	14.48	4.19
231	15	52.47	9.89	15	39.57	7.34	14	13.11	3.08	14	3.50	2.55	14	14.38	4.14
246	12	56.50	8.34	12	42.28	7.57	12	12.42	2.00	12	5.33	5.11	11	11.97	2.89
291	7	59.51	6.55	7	44.99	5.05	7	14.41	4.14	7	3.77	3.47	7	9.80	0.67
318	15	51.65	10.05	15	38.22	7.93	15	11.01	4.36	15	3.81	3.43	12	12.34	4.40
331	12	47.31	10.61	12	35.56	8.40	12	9.71	4.44	12	2.02	1.51	10	11.83	2.50
332	9	55.53	13.12	9	41.76	10.31	9	13.23	4.18	9	1.95	1.33	9	11.95	3.98
345	11	49.52	8.95	11	36.94	8.01	11	12.37	4.64	11	3.15	1.78	11	14.54	5.43
347	9	59.40	5.34	9	44.44	3.93	9	13.93	2.34	9	3.53	3.26	9	9.87	1.78
348	11	50.61	4.59	11	37.65	4.07	11	10.22	2.60	11	5.08	2.72	9	15.47	3.90
38	12	53.80	7.06	12	40.52	6.33	12	9.44	2.39	12	9.34	7.32	11	15.28	3.88
44	9	52.25	4.96	9	37.98	4.01	9	8.84	3.34	9	6.50	3.86	8	15.80	3.19
Avg:	**10.67**	**54.11**	**8.35**	**10.67**	**40.54**	**7.00**	**10.60**	**11.33**	**3.33**	**10.60**	**5.16**	**3.48**	**9.87**	**13.23**	**3.53**

	NDF/ADL			ADF/ADL			%HC/%L			%C/%L		
ID	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**
217	12	5.19	1.46	12	3.71	0.99	12	1.48	0.50	12	2.71	0.99
218	6	4.92	0.67	6	3.71	0.55	6	1.22	0.27	6	2.71	0.55
226	10	6.38	1.36	10	4.88	1.01	10	1.49	0.40	10	3.88	1.01
23	10	7.10	2.36	10	5.45	1.86	10	1.66	0.67	10	4.45	1.86
231	14	4.12	0.34	14	3.11	0.30	14	1.01	0.12	14	2.11	0.30
246	12	4.59	0.67	12	3.44	0.63	12	1.16	0.11	12	2.44	0.63
291	7	4.43	1.27	7	3.35	0.96	7	1.08	0.32	7	2.35	0.96
318	15	5.41	2.28	15	4.01	1.76	15	1.41	0.57	15	3.01	1.76
331	12	5.28	1.13	12	3.96	0.82	12	1.33	0.32	12	2.96	0.82
332	9	4.30	0.51	9	3.23	0.42	9	1.07	0.12	9	2.23	0.42
345	11	4.38	1.31	11	3.24	0.96	11	1.15	0.40	11	2.24	0.96
347	9	4.34	0.59	9	3.25	0.46	9	1.09	0.19	9	2.25	0.46
348	11	5.29	1.52	11	3.93	1.13	11	1.36	0.41	11	2.93	1.13
38	12	6.11	1.96	12	4.58	1.42	12	1.53	0.62	12	3.58	1.42
44	9	6.69	2.58	9	4.84	1.82	9	1.85	0.79	9	3.84	1.82
Avg:	**10.60**	**5.24**	**1.33**	**10.60**	**3.91**	**1.01**	**10.60**	**1.33**	**0.39**	**10.60**	**2.91**	**1.01**

Appendix C.2. Individual Dietary Variables for the Dry Season.

	%NDF			%ADF			%ADL			%Ash			%Protein		
ID	**N**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**
217	7	57.63	7.36	7	42.15	6.99	7	13.06	3.08	7	2.26	0.53	7	10.11	1.88
218	6	62.81	9.83	6	47.58	9.42	6	13.09	3.61	6	7.20	5.24	6	12.56	4.56
226	6	55.28	5.28	6	42.86	4.82	6	9.63	2.33	6	6.25	1.25	6	13.05	1.57
23	5	51.70	12.50	5	38.46	9.50	5	9.44	3.33	5	9.14	2.83	5	15.04	4.68
231	8	59.92	7.29	8	44.73	6.25	8	15.06	2.50	8	4.07	3.25	8	13.34	5.19
246	8	57.12	9.47	8	43.17	8.56	8	12.34	2.29	8	5.78	6.16	8	11.31	2.48
291	5	62.40	3.61	5	47.14	3.21	5	16.17	2.53	5	1.98	0.80	5	9.85	0.76
318	8	59.26	7.40	8	44.09	6.02	8	13.66	2.49	8	4.60	3.71	8	9.83	1.93
331	4	60.31	4.60	4	45.97	3.93	4	14.98	3.85	4	2.71	2.21	4	11.16	3.43
332	6	61.36	12.14	6	45.98	10.20	6	15.20	3.67	6	1.38	0.63	6	9.84	2.38
345	7	52.16	8.79	7	39.29	7.73	7	14.69	4.29	7	3.25	1.83	7	13.46	5.21
347	6	61.71	4.70	6	45.92	3.99	6	15.01	1.80	6	2.13	1.14	6	9.30	1.86
348	7	52.49	4.27	7	39.51	3.61	7	11.63	1.21	7	5.35	3.32	7	13.98	2.90
38	7	53.64	6.91	7	39.48	5.44	7	10.32	1.90	7	5.23	5.23	7	15.37	3.83
44	7	53.70	4.17	7	39.00	3.28	7	9.34	3.67	7	6.36	4.40	6	15.16	3.11
Avg:	**6.47**	**57.43**	**7.22**	**6.47**	**43.02**	**6.20**	**6.47**	**12.91**	**2.84**	**6.47**	**4.51**	**2.84**	**6.40**	**12.23**	**3.05**

	NDF/ADL			ADF/ADL			%HC/%L			%C/%L		
ID	**N**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**
217	7	4.54	0.69	7	3.29	0.39	7	1.25	0.33	7	2.29	0.39
218	6	4.92	0.67	6	3.71	0.55	6	1.22	0.27	6	2.71	0.55
226	6	5.99	1.37	6	4.65	1.13	6	1.34	0.27	6	3.65	1.13
23	5	6.17	2.80	5	4.55	1.97	5	1.62	0.85	5	3.55	1.97
231	8	4.02	0.29	8	2.99	0.17	8	1.03	0.14	8	1.99	0.17
246	8	4.68	0.76	8	3.54	0.72	8	1.14	0.12	8	2.54	0.72
291	5	3.93	0.61	5	2.96	0.40	5	0.97	0.21	5	1.96	0.40
318	8	4.41	0.60	8	3.27	0.43	8	1.14	0.22	8	2.27	0.43
331	4	4.21	0.99	4	3.20	0.71	4	1.01	0.28	4	2.20	0.71
332	6	4.09	0.48	6	3.05	0.38	6	1.04	0.14	6	2.05	0.38
345	7	3.82	1.28	7	2.85	0.93	7	0.97	0.40	7	1.85	0.93
347	6	4.16	0.55	6	3.09	0.35	6	1.07	0.23	6	2.09	0.35
348	7	4.55	0.56	7	3.42	0.43	7	1.13	0.14	7	2.42	0.43
38	7	5.33	1.07	7	3.90	0.69	7	1.42	0.42	7	2.90	0.69
44	7	6.68	2.93	7	4.81	2.03	7	1.87	0.91	7	3.81	2.03
Avg:	**6.47**	**4.77**	**1.04**	**6.47**	**3.55**	**0.75**	**6.47**	**1.21**	**0.33**	**6.47**	**2.55**	**0.75**

Table C.3. Individual Dietary Variables for the Wet Season.

	%NDF			%ADF			%ADL			%Ash			%Protein		
ID	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**
217	5	47.02	5.73	5	33.21	4.76	5	8.21	2.40	5	4.66	1.60	5	17.96	0.58
226	4	49.69	9.06	4	37.61	7.77	4	7.38	2.10	4	7.12	4.00	4	17.50	3.18
23	5	56.48	8.64	5	44.51	7.74	5	7.25	1.84	5	15.52	7.84	4	13.77	4.05
231	7	43.94	2.69	7	33.67	2.03	6	10.51	1.29	6	2.75	0.91	6	15.76	1.66
246	4	55.25	6.53	4	40.50	5.73	4	12.57	1.54	4	4.42	2.32	3	13.72	3.75
291	2	52.31	7.73	2	39.64	5.63	2	10.01	4.79	2	8.24	3.72	2	9.65	0.62
318	7	42.96	2.54	7	31.52	2.53	7	7.98	4.13	7	2.91	3.10	4	17.37	3.42
331	8	40.80	4.77	8	30.35	3.37	8	7.07	0.84	8	1.67	1.03	6	12.27	1.89
332	3	43.88	3.76	3	33.34	2.33	3	9.30	1.15	3	3.09	1.77	3	16.17	3.03
345	4	44.89	8.25	4	32.82	7.68	4	8.32	0.80	4	2.99	1.93	4	16.41	6.03
347	3	54.79	3.32	3	41.49	1.55	3	11.77	1.81	3	6.33	4.65	3	11.00	1.05
348	4	47.33	3.36	4	34.39	2.63	4	7.75	2.61	4	4.60	1.49	2	20.72	0.74
38	5	54.03	8.08	5	41.99	7.82	5	8.20	2.66	5	15.10	5.94	4	15.12	4.56
44	2	47.18	5.12	2	34.43	5.65	2	7.07	0.68	2	7.00	1.47	2	17.71	3.63
Avg:	**4.50**	**48.61**	**5.68**	**4.50**	**36.39**	**4.80**	**4.43**	**8.81**	**2.05**	**4.43**	**6.17**	**2.98**	**3.71**	**15.37**	**2.73**

	NDF/ADL			ADF/ADL			%HC/%L			%C/%L		
ID	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**	**n**	**Mean**	**SD**
217	5	6.09	1.84	5	4.29	1.31	5	1.80	0.54	5	3.29	1.31
226	4	6.96	1.27	4	5.23	0.82	4	1.73	0.48	4	4.23	0.82
23	5	8.04	1.58	5	6.34	1.38	5	1.70	0.52	5	5.34	1.38
231	6	4.26	0.38	6	3.28	0.37	6	0.99	0.09	6	2.28	0.37
246	4	4.41	0.48	4	3.23	0.41	4	1.18	0.12	4	2.23	0.41
291	2	5.69	1.95	2	4.32	1.50	2	1.37	0.45	2	3.32	1.50
318	7	6.56	2.97	7	4.84	2.35	7	1.71	0.70	7	3.84	2.35
331	8	5.82	0.78	8	4.33	0.61	8	1.49	0.19	8	3.33	0.61
332	3	4.73	0.18	3	3.60	0.20	3	1.13	0.02	3	2.60	0.20
345	4	5.37	0.60	4	3.91	0.60	4	1.46	0.12	4	2.91	0.60
347	3	4.71	0.58	3	3.58	0.55	3	1.13	0.06	3	2.58	0.55
348	4	6.57	1.90	4	4.81	1.50	4	1.77	0.40	4	3.81	1.50
38	5	7.20	2.50	5	5.52	1.72	5	1.67	0.86	5	4.52	1.72
44	2	6.74	1.37	2	4.93	1.27	2	1.81	0.10	2	3.93	1.27
Avg:	**4.43**	**5.94**	**1.31**	**4.43**	**4.44**	**1.04**	**4.43**	**1.50**	**0.33**	**4.43**	**3.44**	**1.04**