The Ecological Implications of Body Size: Insights from an African Large Herbivore Assemblage

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Abstract

Variation in body size—6 orders of magnitude in mammals alone—is a defining feature of the animal kingdom and governs biological processes across levels of organization. Body size has pronounced effects on everything from rates of heat exchange to vulnerability to predation, and thus studies of scaling relationships have yielded many powerful insights into species' responses to environmental variation. Scaling relationships between body size and behavior, however, have received comparatively little attention, despite the fact that behavioral plasticity is typically the first line of defense in the face of environmental change. I first investigated how body size modulates behavioral and physiological strategies used by two closely related antelope (bushbuck, *Tragelaphus sylvaticus*, and greater kudu, *Tragelaphus strepsiceros*) to cope with diminishing water availability during the austral dry season. Surface water dependence is generally considered to scale positively with body size, but using a diverse suite of data streams, I found the opposite in my study species: bushbuck, the smaller species, consumed more preformed water, spent more time drinking, and strengthened their selection for surface water later in the dry season more than kudu. These results support the contention that body size alone is insufficient for predicting surface water dependence, and that interspecific differences in surface water dependence could have important effects on the distribution and composition of large-herbivore communities as temperatures warm and droughts become more frequent and severe. Next, I evaluated how key components of individuals' niche space (e.g., diet composition, habitat use and movement patterns) differ between male and female bushbuck, kudu, and a third congener, nyala (*Tragelaphus angasii*), that all exhibit pronounced sexual size dimorphism. I found considerable variation among species in the axes along which males and females partitioned their foraging behaviors, and this variation appeared to be driven by factors other than size dimorphism. Bushbuck, the smallest species, showed the greatest sex-dependent difference in diet quality. Nyala showed the greatest difference in habitat use, and kudu, the largest species (~4× larger than bushbuck), showed the greatest difference in movement behavior. This variation suggests that sex-dependent niche partitioning, and ultimately sexual segregation, is influenced by a suite of ecological factors that do not all covary with sexual size dimorphism in ungulates. Finally, I investigated whether foraging behavior differs between tusked and tuskless female African elephants (*Loxodonta africana*) in a population where tusklessness evolved rapidly in response to intensive poaching pressure. Elephants are a keystone species, and they are responsible for landscape-scale changes in vegetation structure and composition via their destructive foraging behaviors, at least some of which involve the use of tusks (e.g., stripping bark, toppling trees). I found that tuskless females showed stronger selection for grassland habitat than tusked females; however, both tusk morphs selected woodland habitats more strongly than grassland regardless of season. These results suggest that intensive poaching for ivory could have consequences for savanna ecosystems that are mediated by elephant behavior.

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Dedication

To my wife who selflessly followed me to a small town in northern Idaho in the middle of a pandemic, who held down the fort while I was in Africa, and who helped raise two beautiful and amazing young kids. Thank you for your grace, courage, love, and support.

To my beautiful daughters, may you always be in awe of the natural word around you.

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Disclaimer

The views expressed in this dissertation are those of the author and do not reflect the official policy or position of the United States Air Force, Department of Defense, or the U.S. Government.

Chapter 1: Decomposing water requirements of savanna ungulates: seasonal and interspecific differences in plant- vs. surface-water intake by browsing antelopes

ABSTRACT

Many tropical savannas are characterized by marked seasonal variation in surface water availability, and large herbivores in these systems often exhibit behavioral and/or physiological adjustments to cope with this variation. Natural selection should favor strategies that reduce dependence on surface water, yet whether and to what degree such strategies are modulated by body size is poorly understood. Indeed, much research on water dependence in savanna herbivores has focused on comparing the requirements of grazing versus browsing ungulates rather than on understanding the mechanisms that generate variation within these guilds. Moreover, although surface water dependence is often assumed to scale positively with body mass, recent evidence suggests that this relationship is driven by a few species at the extreme ends of the size continuum. We used a diverse suite of data streams, including GPS-tracking of individual movements, analyses of dietary water and nutrient contents, and both camera- and bio-logger-based metrics of time spent drinking, to test the prediction that surface water dependence scales positively with body mass in two closely related species of browsing antelope, bushbuck (*Tragelaphus sylvaticus*) and greater kudu (*Tragelaphus strepsiceros*). We found no support for this prediction: kudu (~4× larger than bushbuck) consumed less preformed water, spent less time drinking, and relaxed their selection for surface water as the dry season progressed. In contrast, bushbuck consumed more preformed water, spent more time drinking, and strengthened their selection for surface water later in the dry season. Neither fecal water content nor mean proximity to surface water differed between species. Much of Africa is projected to become hotter and drier in the coming century, and a thorough understanding of how large herbivores are likely to cope with such changes is imperative for predicting population- and community-level responses. Our results suggest that body size alone is insufficient for predicting surface water dependence in diverse large-herbivore communities and highlight the importance of using

mechanistic tools and techniques to quantify nuanced responses of animals to environmental variation.

INTRODUCTION

Water distribution and accessibility govern pattern and process across levels of organization, from the behavior and physiology of individual animals (Cain et al. 2006) to the distribution and composition of communities (Veldhuis et al. 2019). At the largest scales, water availability is a primary driver of spatiotemporal variation in biodiversity and is the strongest predictor of species richness in the tropics, subtropics, and warm temperate zones (Hawkins et al. 2003). Many of these systems (e.g., arid and semi-arid tropical environments) are strongly seasonal, and thus considerable variation in water availability and plant productivity is the rule rather than the exception (Valeix 2011, Bennitt et al. 2014). Such variation can enhance the fitness benefits of behavioral plasticity, and many animals, including large, herbivorous mammals, exhibit a wide array of behavioral adjustments to cope with water and other resource limitations (Hughes et al. 2011, Bennitt et al. 2014, Gedir et al. 2016, Veldhuis et al. 2019). For example, variable patterns of precipitation and forage productivity drive long-distance migrations of wildebeest (*Connochaetes taurinus*) in the Serengeti and saiga antelope (*Saiga tatarica*) in the semi-arid rangelands of central Asia (Pennycuick 1975, Frank et al. 1998, Holdo et al. 2009, Singh et al. 2010). Similarly, patterns of snowmelt often govern the progression of spring green up across elevational gradients, which affects the movement behavior of bison *(Bison bison*), elk (*Cervus canadensis)* and caribou (*Rangifer tarandus*) in North America (Fancy et al. 1989, Rickbeil et al. 2019, Abrahms et al. 2021, Laforge et al. 2021, Sheppard et al. 2021).

Animal responses to environmental variation are modulated by a diversity of extrinsic and intrinsic factors. One of the most fundamental of these is body size, which both facilitates and constrains herbivore behavior (Bell 1971, Geist 1974, Jarman 1974, Daskin et al. 2023). Dietary and water requirements, movement patterns, anti-predator behavior, and costs of thermoregulation all scale with body size (Dial et al. 2008, Owen-Smith and Mills 2008, Daskin et al. 2023), and the study of scaling relationships can provide mechanistic insights into how body size constrains the range of behaviors available to large herbivores as they negotiate complex tradeoffs (Veldhuis et al. 2019, Esmaeili et al. 2021). For example, optimal foraging theory predicts that species inhabiting heterogeneous landscapes should move more in search of resources than those that occupy more homogenous environments (Charnov 1976, Belovsky 1997, Abrahms et al. 2021). The ability to move more, however, is constrained by body size (du Toit 1990, Ofstad et al. 2016, Noonan et al. 2020).

Surface water dependence, and corresponding rates of water consumption, are also predicted to scale with body size. Numerous factors, however, add considerable noise to this scaling relationship. For example, gut morphology, feeding type (e.g., position on the browser–grazer continuum), and moisture content of forage (i.e., preformed water) interactively influence reliance on surface water (Woodall and Skinner 1993, Redfern et al. 2003, Cain et al. 2006, Valeix 2011, Kihwele et al. 2020). In theory, reduced dependence on surface water should confer a fitness advantage in systems with marked seasonal variation in water availability because it can decrease spatial overlap (and thus competition) between water dependent and independent species, and can reduce vulnerability to predation by making movements less predictable when surface water becomes scarce (Mosser et al. 2009, Veldhuis et al. 2019). Accordingly, natural selection should favor behavioral and physiological strategies that decouple herbivores from surface water. Whether and to what degree such strategies are constrained by body size, however, has been the subject of surprisingly few studies (but see Hayward and Hayward 2012, Veldhuis et al. 2019, Esmaeili et al. 2021).

Previous research on herbivore-water relationships in savanna systems has often centered on comparison of grazing versus browsing ungulates rather than on understanding the mechanisms that underpin variation in water dependence within these guilds. Browsers generally are regarded as being less dependent on surface water than grazers (Redfern et al. 2003, Valeix et al. 2009, Smit 2011, Valeix 2011, Hempson et al. 2015, Kihwele et al. 2020, Esmaeili et al. 2021), and some authors have suggested that browsers have lower water requirements (Hayward and Hayward 2012). Recently, however, Veldhuis et al. (2019) summarized evidence that grazers and browsers have equivalent water requirements but meet them in different ways (i.e., dietary water intake versus consumption of surface water). Those authors predicted that browsers are less dependent on surface water not because they have lower absolute water requirements, but because their diet is more succulent and contains more preformed water. This prediction has not been tested, however, and a nuanced understanding of how browsers meet their hydration needs is lacking. In tropical savannas, declining water content in forage might increase dependence of browsers on surface water as the austral dry season progresses. In addition, the role of body size in modulating seasonal changes in surface water dependence has also received little attention.

Situated at the southern end of the Great Rift Valley, Mozambique's Gorongosa National Park is characterized by substantial seasonal variation in water availability and distribution (Tinley 1977). The Park receives an average of 840 mm of rain annually, most of which falls during the wet season (November to April), and the subsequent inundation of several rivers flowing into Lake Urema at the center of the park floods up to 40% of Gorongosa each year (Daskin et al. 2016). Conversely, <50 mm of rain typically falls each month during the dry season (May to October), leading to reduced availability of surface water and diminished forage abundance and quality. Of the many species of large herbivores occupying Gorongosa, two congeneric antelope—greater kudu (*Tragelaphus strepsiceros*) and bushbuck (*Tragelaphus sylvaticus*)—are ideal for studying size-dependent responses of browsing herbivores to seasonal changes in water availability. These two spiral-horned antelopes have similar life histories, are sympatric within woodland habitats, and are primarily browsers (Tinley 1977, Hempson et al. 2015, Daskin et al. 2023). Despite these similarities, however, the two species differ four-fold (based on average mass of adult females; Kingdon 2015) in body mass. These traits provide a unique opportunity to assess how body size modulates the behavioral and physiological strategies used by browsing herbivores to cope with changes in availability of surface and preformed water as both become more limited during the dry season.

We hypothesized that surface-water dependence (i.e., proximity to surface water and amount of time spent drinking) of browsing antelopes is modulated by body size and declining intake of preformed water as plants senesce during the dry season. To test this hypothesis we tracked individual movements using GPS collars, collected fecal samples for analysis of diet composition and water content, measured water and nutrient concentrations in key forage plants throughout the dry season, and recorded the frequency and duration of drinking bouts by bushbuck and kudu. We predicted that because surface-water dependence ostensibly scales positively with body size across ungulates (Hempson et al. 2015, but see Veldhuis et al. 2019), kudu would spend more time drinking during the dry season than bushbuck. We also predicted that both antelope species would increasingly consume plants that exhibited a lower rate of water loss (i.e., plants that retained a greater proportion of their preformed water content) as the dry season progressed (Warrick and Krausman 1989, Cain et al. 2006, Harkleroad and Krausman 2014, Kihwele et al. 2020), but that the consequences of this behavioral adjustment would differ between species. For bushbuck (the smaller species), we expected that this dietary shift, in combination with reduced fecal water loss (Ghobrial and Cloudsley-Thompson 1966, Maloiy and Hopcraft 1971, Cain et al. 2006), would preclude the need to increase consumption of surface water as the dry season progressed. Accordingly, we predicted that neither total time spent drinking nor strength of selection for surface water sources by bushbuck would change significantly during the dry season. In contrast, we predicted that kudu, owing to their larger body size and greater absolute water requirements, would increase time spent drinking and strength of selection for surface water as the dry season progressed despite their ostensibly greater ability to reduce fecal water content (Woodall and Skinner 1993).

MATERIALS AND METHODS

Study area.–Gorongosa National Park is situated at the southern end of the Great Rift Valley (18.96°S, 34.36°E) and supports a diverse community of large mammals. Our study area in the core of the park included Lake Urema and its expansive floodplain grassland, as well as *Acacia*, palm, and broadleaf savanna woodland habitats (Pansu et al. 2019, Stalmans et al. 2019). There are two primary seasons in Gorongosa: the wet season, which runs from November to April, and the dry season, which runs from May to October (Fig. 1.1). Annually the Park receives 700–900 mm of rainfall, most of which occurs from December to February (Tinley 1977). Monthly precipitation during the dry season averages <50 mm, and both water and high-quality forage become increasingly scarce as the dry season progresses.

Animal capture and handling.–In July 2021 and June 2022, we captured adult bushbuck (n = 26) and kudu (n = 23) as part of a long-term study of spiral-horned antelope ecology (Daskin et al. 2023). We chemically immobilized individuals of each species via remote injection of a combination of thiafentanil, ketamine, and azaperone. Each individual was fitted with an iridium satellite GPS collar (VERTEX Lite, Vectronic Aerospace) programmed to record locations every 3–30 min for kudu and every 60 min for bushbuck (13-m location error; Atkins et al. 2019). GPS collars were remotely triggered to drop off 1 year after deployment.

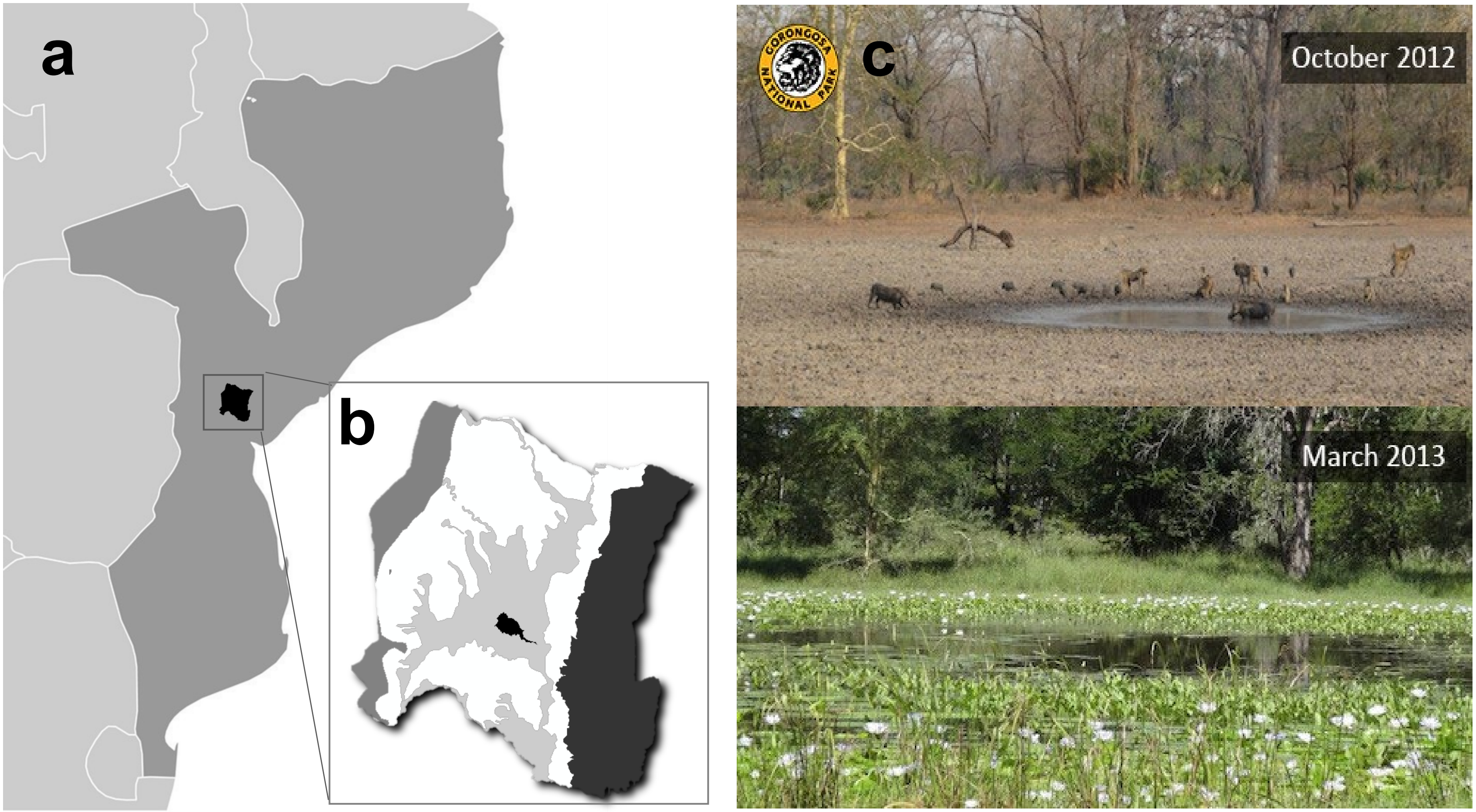

Figure 1.1. – (a) Gorongosa National Park is located in central Mozambique and (b) consists of four major habitat zones (from left: western escarpment [medium gray], woodland [white], floodplain [light gray], and eastern escarpment [dark gray], as well as Lake Urema [black]). (c) A perennial pan during the dry (October) and wet (March) seasons in Gorongosa. Photo used with permission from Gorongosa National Park.

At the time of collaring, bio-loggers capable of recording up to 900 h of 8-kHz audio, 50-Hz accelerometer and 10-Hz magnetometer data (Miquel et al. 2022) were affixed to 8 female kudu collars (Fig. A.1) to record continuous sound data for each individual in 2021. Audio loggers have been used successfully to investigate aspects of physiology, behavior, and ecology of free-ranging wildlife through acoustic signal processing (Lynch et al. 2013, Wijers et al. 2018, Greif and Yovel 2019, Studd et al. 2021). For example, acoustic data have been used to identify behaviors (e.g., eating, drinking, walking, ruminating) of Indian crested porcupines (*Hystrix indica*; Alkon et al. 1989), mule deer (*Odocoileus hemionus*; Lynch et al. 2013), and lions (*Panthera leo*; Wijers et al. 2018). We used data from the audio loggers in conjunction with GPS-collar data to identify and quantify the duration of drinking bouts by kudu (see *Surface water consumption*). Loggers were too heavy to place on bushbuck collars, so we used a camera-trap-based approach to quantify drinking by bushbuck (details below). All animal-handling procedures were approved by the Animal Care and Use Committees of the University of Idaho (No. IACUC-2019-32) and Princeton University (No. 2075F-16) and were in accordance with guidelines established by the American Society of Mammologists (Sikes & The Animal Care and Use Committee of the American Society of Mammologists 2016).

Diet composition.–We quantified composition of bushbuck and kudu diets using fecal DNA metabarcoding following previously established protocols (Atkins et al. 2019, Branco et al. 2019, Guyton et al. 2020, Potter et al. 2022, Daskin et al. 2023, Walker et al. 2023). At the time of collaring, we collected a fecal sample (>5 pellets) directly from the rectum of immobilized individuals for subsequent analysis of diet composition and fecal water content. To capture changes in diet across the dry season, we collected additional fresh fecal samples from both species in the early (May/June 2022; *n* = 18 bushbuck, 28 kudu), mid (July/August 2021; *n* = 25 bushbuck, 32 kudu), and late (September/October 2021; *n* = 15 bushbuck, 16 kudu) dry season. We used GPS data and radiotelemetry to locate collared individuals, and we observed them and any conspecifics using high-powered optics from a distance of 10-100 m until defecation occurred. At the time of defecation, we noted the individual's distance from the observer and the nearest landmarks to the site of defecation. We then searched the area for the fecal sample and collected a minimum of 5 fecal pellets that were clear of debris using nitrile gloves and an unused, plastic zip top bag. Samples were stored on ice in a portable cooler for ≤ 5 hrs until they were processed and preserved at the Gorongosa laboratory. Each sample was homogenized, and a pea-sized sub-sample was transferred to a labeled tube containing silica beads and DNA preservation buffer (Xpedition Stabilization/Lysis Solution, Zymo Research Corporation). To lyse cells, tubed samples were vortexed for 1 minute and were then frozen at -80°C until transport to the U.S. Prior to transport, each sample was subjected to an anti-viral heat treatment (72°C for 30 minutes) and re-frozen in accordance with the requirements of the United States Department of Agriculture (Permit 130123 to Robert M. Pringle).

Analysis of bushbuck and kudu diets was conducted at Princeton University following established protocols (Kartzinel et al. 2015, 2019, Atkins et al. 2019, Branco et al. 2019, Pansu et al. 2019, Guyton et al. 2020, Kartzinel and Pringle 2020, Becker et al. 2021). Briefly, we amplified the P6 loop of the chloroplast *trn*L(UAA) intron, a region commonly used to metabarcode an array of plant taxa, using primers that contain a unique 8-nt tag at the 5' end, enabling pooling of uniquely identifiable PCR products for sequencing in a single high-throughput run (Taberlet et al. 2007). Extraction and sequencing were performed on an Illumina HiSeq 2500 and processed via the OBITools pipeline (Boyer et al. 2016). We discarded low-quality sequences, and the remaining sequences were considered molecular operational taxonomic units (mOTUs). After rarefying to the minimum number of sequence reads per sample, the dataset included 272 dietary mOTUs (136 in the early dry season, 136 in the mid and late dry seasons). Post bioinformatic filtering, we identified plant sequences by matching them to an extensive reference library of DNA from plant specimens collected in Gorongosa (Pansu et al. 2019), and secondarily to a global reference library derived from the European Molecular Biology Laboratory database. Guyton et al. (2020) provide a detailed description of these protocols. From these data, we determined presence/absence and relative read abundance (RRA, the proportional representation of each mOTU per sample; Deagle et al. 2019) of each plant species contained in each sample. We only included samples in subsequent analyses when mOTUs for which we had both diet quality and/or preformed water data accounted for ≥50% of the RRA in the sample.

Dietary and fecal water content.–Previous research in Gorongosa has shown that bushbuck and kudu consume many of the same forage plants, albeit in different proportions (Pansu et al. 2019, Daskin et al. 2023). We used those data to generate a list of 33 plant species commonly consumed by bushbuck and/or kudu during the dry season. We then collected green leaves and stems (petioles) from 3 different individuals of each plant species in July (mid-dry) and October (late-dry) 2021 and May (early-dry) 2022 to quantify changes in preformed water content of those species during the dry season. Samples were weighed immediately after collection using a Pesola Balance Light Line spring scale (precision ± 0.3%), dried to a constant mass (i.e., no additional mass loss after 24 hrs) at 100°C, and then reweighed to calculate water content as a proportion of wet mass. We calculated preformed water content using the following equation (1.1), where W_{wet} and W_{dry} are the weights of the plant material before and after drying:

$$Preformed\ Water\ (\%) = \frac{W_{wet} - W_{dry}}{W_{wet}}\ x\ 100$$

We combined data on diet composition with estimates of plant water content to calculate weighted averages of preformed water in each sampled antelope diet using the RRA of each plant species in the diet as the weighting factor. We then estimated mean preformed water content in the diets of bushbuck and kudu during the early, mid, and late-dry season as the weighted average of water content across individuals using the total proportion of the diet accounted for (based on RRA, minimum of 50% required for inclusion) as the weighting factor.

Throughout the study, we set aside a subset of the fecal pellets collected for diet analysis to estimate fecal water content (*n* = 62 bushbuck, 65 kudu). Fecal samples were weighed using the same Pesola Balance Light Line spring scale and dried in a drying oven to a constant weight at 100°C. We calculated the water content of fecal samples (%) using the same equation used to calculate water content of plants (Lee et al. 2009).

*Diet quality.–*We were interested in quantifying potential nutritional consequences of dietary shifts made by antelope to increase preformed water consumption late in the dry season. Accordingly, we combined data on diet composition with detailed, plant species-specific data on nutritional quality obtained during previous studies (Atkins et al. 2019, Potter et al. 2022, Daskin et al. 2023) to estimate digestible energy (DE) and protein (DP) content of sampled bushbuck and kudu diets during the dry season. We calculated weighted averages of DE and DP for each sampled diet by using the RRA of each plant species in the diet (determined from the metabarcoding analysis) as the weighting factor (Atkins et al.

2019, Branco et al. 2019, Pansu et al. 2022, Walker et al. 2023). We then estimated mean DE and DP of each species' dry-season diet as the weighted average of DE and DP estimates across individuals using the total proportion of the diet accounted for (based on RRA, minimum of 50% required for inclusion) in each sample as the weighting factor. Sample sizes differed between the two nutritional metrics because we had estimates of DP for a much larger number of plant species than DE (*n* = 66 for DE and *n* = 99 for DP) and were therefore able to include more sampled diets in the DP analysis based on the RRA threshold. Mean RRA of samples analyzed for DE and DP was 66% and 75%, respectively, for bushbuck, and 83% and 87%, respectively, for kudu.

Selection for surface water.–We quantified selection for surface water by bushbuck and kudu using resource selection functions (RSFs) and a use-availability design (Boyce et al. 2002, Johnson et al. 2006) at the home range scale (i.e., 3rd-order selection; Johnson 1980). We used 100% minimum convex polygons (MCP) estimated from each collared individual's GPS locations (*n* =12 bushbuck, 12 kudu) to delineate home-range boundaries (for purposes of defining available habitat) during the 2021 dry season (5 July– 31 October 2021). We generated a set of random points equal to the number of used points within each home range during each week (*n* = 17 weeks) of the dry season to assess temporal changes in selection of surface water as the dry season progressed. We used high-resolution photographic imagery (10-cm accuracy) and a digital terrain model (DTM) of Gorongosa derived from airborne light detection and ranging (LiDAR) data collected in August 2019 (mid-dry season; Daskin et al. 2023, Walker et al. 2023) to hand-digitize rivers within our study area. Perennial pans were digitized from satellite imagery (Stalmans 2019). We used the *near* function in ArcGIS 10.8.1 to calculate the distance between each used and random location and the nearest surface water source, whether river or perennial pan. We standardized distances by subtracting the mean and dividing by the standard deviation (Kutner et al. 2004, Cade 2015). We then fit generalized linear mixed models (Gillies et al. 2006, Bolker et al. 2009, Zuur et al. 2009) with a binomial error distribution and a logit link function to the used (antelope GPS locations, coded 1) and random (available locations within each individual's home range, coded 0) locations in the *lme4* package in R (Bates et al. 2022). We fit separate models for each species both on a seasonal (i.e., all dry-season data included) and a weekly (i.e., separate models fit for each week) basis. All models included a random intercept and random slope (Zuur et al. 2009), grouped by individual animal, so that the effect of distance to surface water (the sole predictor variable) on selection was allowed to vary among individuals.

We used simple linear regression to quantify changes in the strength of selection for surface water (quantified using standardized coefficients from weekly RSFs) across the dry season. We also fit weighted versions of these models in which the inverse of the *SE* associated with each GLMM coefficient was used as the weighting factor to account for uncertainty in estimated coefficients; results did not differ qualitatively between weighted and unweighted models. Finally, we used a two-sample *t*-test to compare mean distance to water between bushbuck and kudu independent of the RSFs.

Surface water consumption.–On average, bushbuck home ranges in Gorongosa are small (<0.5 km^2; Atkins et al. 2019), which facilitated the identification of key surface water sources used by GPS-collared individuals, as well as any conspecifics with overlapping home ranges. We used the *adehabitatHR* package (Calenge 2019) in R to estimate 95% fixed-kernel home ranges for GPS-collared bushbuck in 2021 using location data from the first 4 weeks after collar deployment. We overlaid home ranges on the LiDAR-derived DTM and then used the DTM and corresponding high-resolution imagery to identify potential surface-water sources within each bushbuck home range. We then visually confirmed the status of each water source (e.g., dry or containing water) by traversing home ranges on foot. To estimate the frequency and duration of drinking bouts by bushbuck we placed 38 camera traps (Bushnell Trophy Cam E3) near water sources (mostly small, perennial pans), aiming to maximize coverage of the water's edge and key access points. We placed cameras approximately 0.5 m above the ground and programmed each camera to record 15-s videos with an interval of 3 s between recordings. Six weeks after initial deployment, we revisited cameras and removed them from dried pans or repositioned them as necessary to follow receding water levels. We removed all cameras from surface water sources 3 weeks later (October 20, 2021). In total, we recorded 62 bushbuck drinking bouts. We began timing a drinking bout when an individual's muzzle contacted the water and it began rhythmically swallowing, and the bout ended when the muzzle left the water. We considered drinking bout duration to be additive if we observed the same individual drink multiple times during the same visit.

To quantify frequency and duration of kudu drinking bouts, we affixed low-power bio-loggers containing 3 MEMS sensors (Latorre and Chamaille-Jammes 2020, Latorre et al. 2021, Miquel et al. 2022) to eight female kudu collars at the time of capture in 2021. Audio data were recorded using an MP34DT05 MEMS microphone powered by a 1950mAH Li-Ion battery and were stored on a 32 gigabyte micro-SD card. Miquel et al. (2022) provide a detailed description of the components and construction of the bio-loggers.

Bio-loggers recorded continuously until the batteries failed 6–51 days after deployment (mean = 33.7 days; 1 logger did not record any usable data). We collected loggers when GPS collars dropped off 1 year after deployment. We used kudu movement data (3-minute fixes for the first 3-4 weeks, followed by 30 min fixes), in combination with high-resolution LiDAR imagery, to inform when to review audio data for drinking bouts (i.e., based on proximity of collared kudu to a water source; Fig. A.2). We reviewed audio data both aurally and visually (spectrographs; Fig. A.2) in Audacity 3.2.5 (Audacity Team 2023) for approximately 15 min before and after GPS data indicated an individual was near a water source to minimize the possibility of missing drinking bouts. We identified drinking bouts based on the sound of disturbed water and regular, rhythmic swallowing, in combination with the spectrograph (Wijers et al. 2018, 2021). Again, drinking bout duration was additive if the same individual drank multiple times during the same visit to a water source. In total we recorded 102 drinking bouts by kudu.

To validate the video (bushbuck) and audio (kudu) recordings as effective means of identifying drinking bouts, we used GPS locations from collared individuals in 2021 to estimate the number of visits to perennial pans and the corresponding number of drinking bouts (not all visits to pans lead to drinking) by both species between 13 July and 31 August 2021. We focused on these dates because the camera traps and bio-loggers were operating concurrently during this period. We calculated the mean radius of 2,486 surface water polygons in the Park (mean radius = 20 m) as a starting point for identifying pan visits by antelope. We then used GPS locations obtained during our 7-week focal period to determine when each collared individual was within 33 m (20 m average radius + 13 m GPS collar error; Atkins et al. 2019) of a perennial pan (*n* = 5,074 pans; Stalmans 2019). GPS locations that fell within 33 m of a pan were then sorted temporally to distinguish and enumerate separate pan visits for each collared individual. We then estimated the number of drinking bouts for each collared individual by scaling the number of pan visits by the proportion of visits that led to drinking bouts by each species, which we estimated from camera trap videos. In total, cameras recorded 83 instances of female bushbuck at perennial water sources, and of those 83 recordings, 41 of them included drinking behavior (49%). Likewise, of the 9 recordings of female kudu at perennial pans, 6 of them included drinking behavior (67%). We compared the weekly number of drinking bouts estimated from GPS-collar data to the weekly number of drinking bouts recorded aurally using bio-loggers using linear regression. Informal comparison of GPS-derived estimates of drinking bouts by bushbuck with camera-derived estimates indicated that the 60-min fix rate of bushbuck collars grossly underestimated visits to pans, and thus we did not formalize this comparison for bushbuck.

Testing predictions about surface-water dependence required generating estimates of total time spent drinking from the camera trap data for bushbuck and the audio logger data for kudu. Total time spent drinking is the product of drinking bout frequency and mean bout duration. For bushbuck, linear regression analysis indicated that the mean number of drinking bouts per week did not change as the dry season progressed ($\beta = -0.123$, $P = 0.887$). Accordingly, and because we lacked individual-level data for bushbuck, we estimated mean total time spent drinking by bushbuck in each week of camera-trap deployment (17 August–20 October 2021) by multiplying the mean number of drinking bouts per week (calculated across all weeks) by the mean bout length per week. For kudu, which were individually monitored, we estimated mean total time spent drinking by multiplying the mean number of drinking bouts per week (calculated across monitored individuals within each week) by the mean bout length per week during the monitoring period (13 July–28 August 2021).

Statistical analyses.–We used general linear models (GLMs) to evaluate the effects of species and season on dietary water and nutrient (DE, DP) content. We included species (bushbuck or kudu), Julian day, and the corresponding 2-way interaction as explanatory variables. We tested for seasonal (early versus late dry season) differences in preformed water content of each dietary plant species using ANOVA. We used results of that analysis to categorize plant species based on whether preformed water content differed significantly ($\alpha = 0.10$) between the early and late dry seasons. We then used 2-sample Z-tests to determine whether the proportion of bushbuck and kudu diets comprised of plant species in each category (i.e., plants that did or did not exhibit a significant decline in water content late in the dry season) changed between the early and late dry seasons (potentially indicative of a shift in diet toward or away from plants with relatively more water content). We also sorted plant species based on their DP and DE content and identified species that fell into the upper and lower quartiles of the distribution of each nutritional metric. We used 2-sample Z-tests to determine whether the proportion of bushbuck and kudu diets comprised of plant species in each category (i.e., high versus low quality) changed between the early and late dry seasons. We used GLMs to test our predictions that both antelope species would increase water retention as the dry season progressed, and that kudu would have lower rates of fecal water loss (i.e., lower fecal water content) than bushbuck. We included species, Julian day, and their interaction as covariates.

We tested our prediction that kudu would spend more time drinking than bushbuck during the dry season using a GLM with species and time (Julian day) as covariates. To test the prediction that kudu, but not bushbuck, would increase consumption of surface water as the dry season progressed, we fit GLMs with time (Julian day), species, and their interaction as covariates. We used a weekly time step to maximize within-time sampling rate without washing out meaningful temporal variation in time spent drinking. As a result, however, the GLM was likely underpowered (n = 8 weeks for bushbuck and 7 weeks for kudu). Therefore, to reduce the likelihood of a type 2 error we also used species-specific linear regressions to test for temporal trends in time spent drinking by bushbuck and kudu.

To test for seasonal differences in dietary dissimilarity between species we used nonmetric multidimensional scaling (NMDS; Daskin et al. 2023; Walker et al. 2023) followed by permutational multivariate analysis of variance (perMANOVA). To better understand how plant traits influenced preformed water content across the dry season we used linear regression to investigate the relationship between seasonal percent water loss (early to late-dry season) and leaf mass per area (a metric of vulnerability to evaporative water loss) and plant height (related to ‘woodiness’) of each key forage species.

RESULTS

Dietary water.–Preformed water content declined significantly between the early and late dry season in roughly half (54%) of plant species consumed by bushbuck and/or kudu (Table A.1). Of the remaining plant species, only one (*Trichilia emetica*) contained more water late in the dry season than early (Table A.1). The composition of bushbuck diets shifted in tandem as the dry season progressed, reflecting a consistent effort by bushbuck to increase intake of preformed water. In the early dry season, a majority ($P = 0.002$) of bushbuck diets were comprised of plants that contained significantly ($P = 0.02$, 2-sample *t*-test) more preformed water during that period, but that subsequently declined in water content as the dry season progressed; Fig. 1.2; Table A.1). In the late-dry season, however, bushbuck shifted to consuming a significantly ($P = 0.01$) greater proportion of plants that maintained their preformed water content throughout the dry season (Fig. 1.2, Table A.1). Kudu exhibited no significant ($P = 0.346$) preference for plant species based on water content during the early dry season, but, similar to bushbuck, shifted to consuming a significantly ($P = 0.018$) greater proportion of plants that maintained their preformed water content late in the dry season (Fig. 1.2, Table A.1). Despite shifting diet composition toward plants that contained more water late in the dry season, preformed water content of both bushbuck and kudu diets still declined significantly as the dry season progressed ($\beta_{\text{Julian}} = -0.07$, $P = 0.001$, adj. $R^2 = 0.288$; Fig. 1.3). This decline, however, differed significantly between species, with kudu diets containing less preformed water ($\beta_{\text{Species}} = -9.49$, $P = 0.09$). Concomitantly, fecal water content declined significantly during the dry season for both species ($\beta_{\text{Julian}} = -0.12$, $P < 0.001$, adj. $R^2 = 0.388$; Fig. 1.4) and did not differ between bushbuck and kudu ($\beta_{\text{Species}} = -7.07$, $P = 0.14$).

Diet composition and quality.–We observed significant differences in taxonomic composition of bushbuck and kudu diets across the dry season, both between (perMANOVA: pseudo-$F_{1,146} = 11.194$, $P < 0.001$, $R^2 = 0.071$) and within species (bushbuck, early vs. mid: pseudo-$F_{1,41} = 3.21$, $P = 0.002$, $R^2 = 0.073$; mid vs. late: pseudo-$F_{1,38} = 3.11$, $P = 0.001$, $R^2 = 0.076$; early vs. late: pseudo-$F_{1,31} = 5.2$, $P = 0.001$, $R^2 = 0.144$; kudu, early vs. mid: pseudo-$F_{1,72} = 1.93$, $P = 0.062$, $R^2 = 0.026$; mid vs. late: pseudo-$F_{1,48} = 5.14$, $P = 0.002$, $R^2 = 0.097$; early vs. late: pseudo-$F_{1,54} = 5.35$, $P = 0.001$, $R^2 = 0.09$; Fig. A.3). During the early dry season, bushbuck and kudu consumed relatively equal proportions ($P = 0.373$ and $P = 0.679$, respectively) of low- and high-DP plant species (Fig. 1.2). In the late dry season, however, bushbuck consumed a significantly ($P = 0.092$) greater proportion of plants with high DP (Fig. 1.2), whereas kudu diet composition remained unchanged with respect to plant protein content ($P = 0.17$; Fig. 1.2).

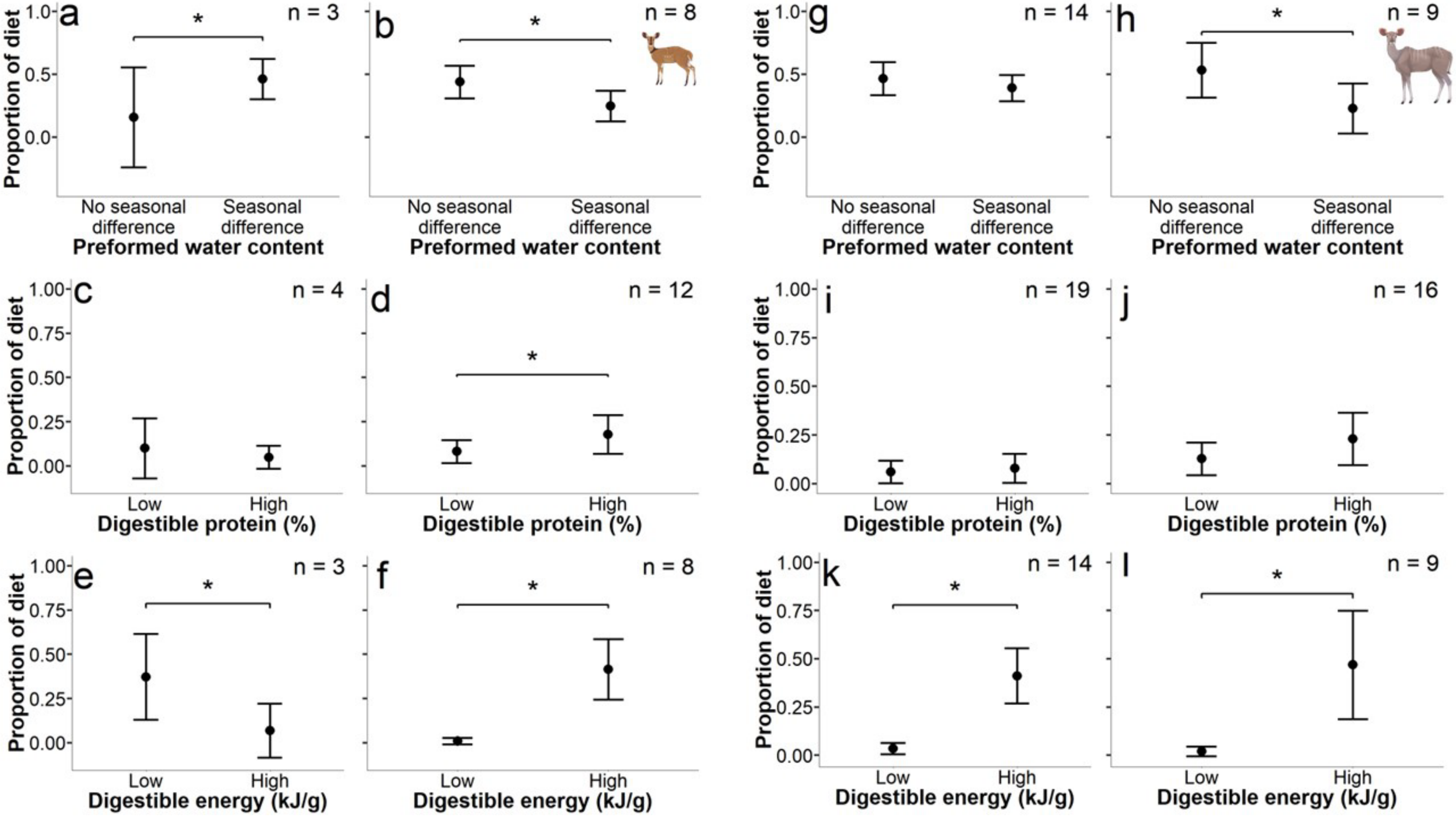

Figure 1.2. – Mean (± 95% CI) proportion of bushbuck (panels a–f) and kudu (panels g–l) diets comprised of plants that (1) did or did not differ significantly in water content between the early and late dry season (first row) and had either (2) digestible protein content (second row) or (3) digestible energy content (third row) in the lower or upper quartile of the respective distributions across forage plant species. Early dry season (May/June 2022) results are presented in the first column and late dry season (September/October 2021) results in the second column of panels for each antelope species. Significant differences (two-sample z-test, α = 0.10) are indicated by an *.

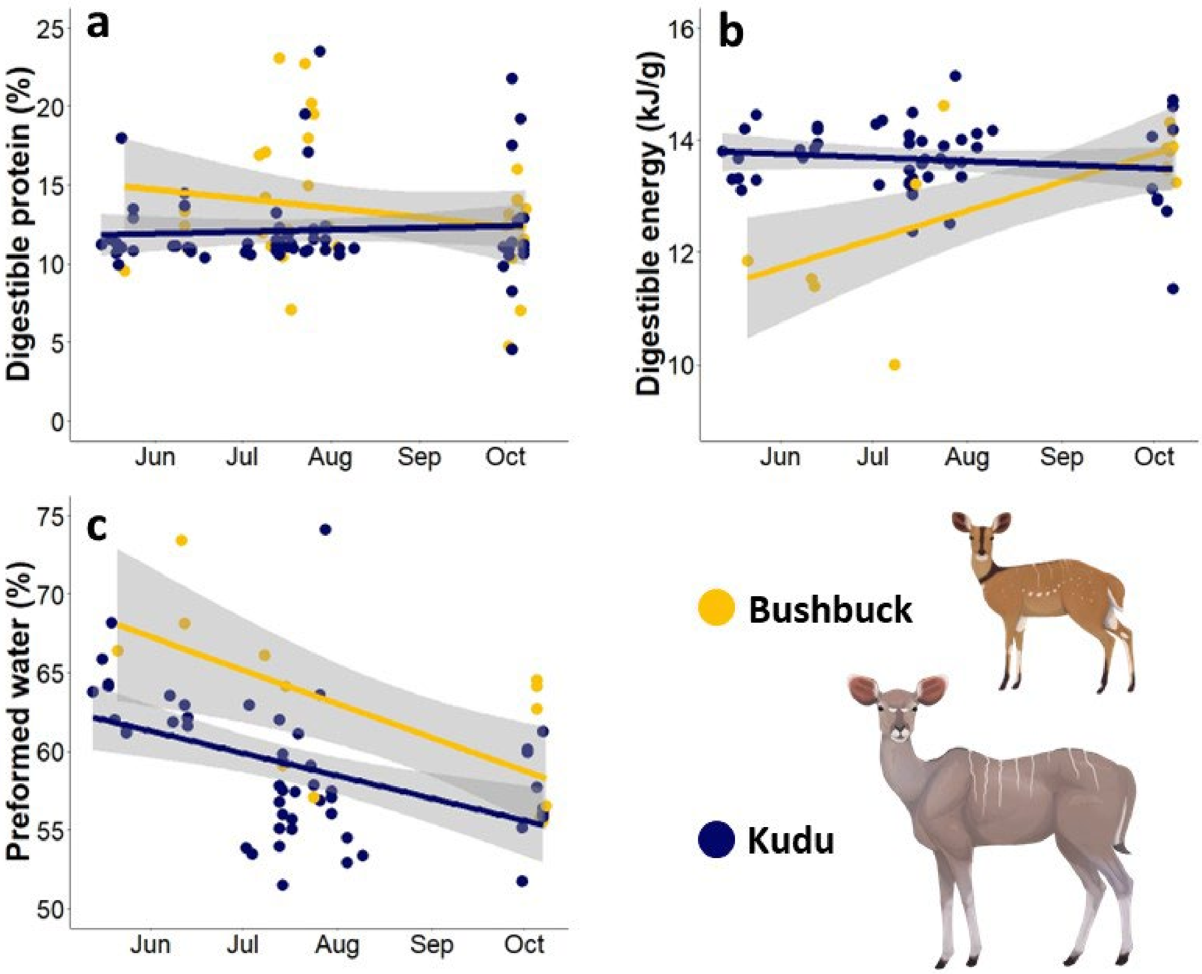

Figure 1.3. – (a) Percent digestible protein in bushbuck (*n* = 33, yellow) and kudu (*n* = 66, blue) diets during the dry season in Gorongosa National Park, Mozambique. Early dry season fecal samples were collected in May and June 2022; mid-dry season samples were collected in July and August 2021; late-dry season samples were collected in September and October 2021. Dietary protein content did not change during the dry season for either species (Julian day: $\beta = -0.019$, $P = 0.158$, adj. $R^2 = 0.022$) but bushbuck diets contained more dietary protein (Species: $\beta = -6.27$, $P = 0.08$) than kudu diets. (b) Dietary digestible energy also did not change during the dry season for kudu (*n* = 51) but increased for bushbuck (*n* = 15; Julian day × species interaction: $\beta = -0.02$, $P < 0.001$, adj. $R^2 = 0.29$). (c) Preformed water content in the diets of bushbuck (*n* = 15) and kudu (*n* = 51) declined significantly during the dry season (Julian day: $\beta = -0.07$, $P = 0.001$, adj. $R^2 = 0.288$) and bushbuck diets contained significantly more preformed water than kudu diets (Species: $\beta = -9.49$, $P = 0.09$).

Changes in diet composition related to plant energy content (DE) were more pronounced. In the early dry season, bushbuck consumed a significantly ($P < 0.001$) greater proportion of low-DE plants, whereas kudu consumed a significantly ($P < 0.001$) greater proportion of plants that were high in DE (Fig. 1.2). During the late dry season, however, the diets of both species were comprised of a significantly ($P < 0.001$) greater proportion of high-DE plants (Fig. 1.2). Unlike dietary water content, shifts in diet composition with respect to plant nutrient content were apparently sufficient to prevent declines in diet quality as the dry season progressed. We found no evidence that DP in the diets of bushbuck and kudu changed during the dry season ($\beta_{\text{Julian}} = -0.019$, $P = 0.158$, adj. $R^2 = 0.022$; Fig. 1.3), although bushbuck diets contained significantly more DP than kudu diets ($\beta_{\text{Species}} = -6.27$, $P = 0.08$). Dietary DE also did not change during the dry season for kudu, and increased as the dry season progressed for bushbuck ($\beta_{\text{Species x Julian}} = -0.02$, $P < 0.001$, adj. $R^2 = 0.29$; Fig. 1.3).

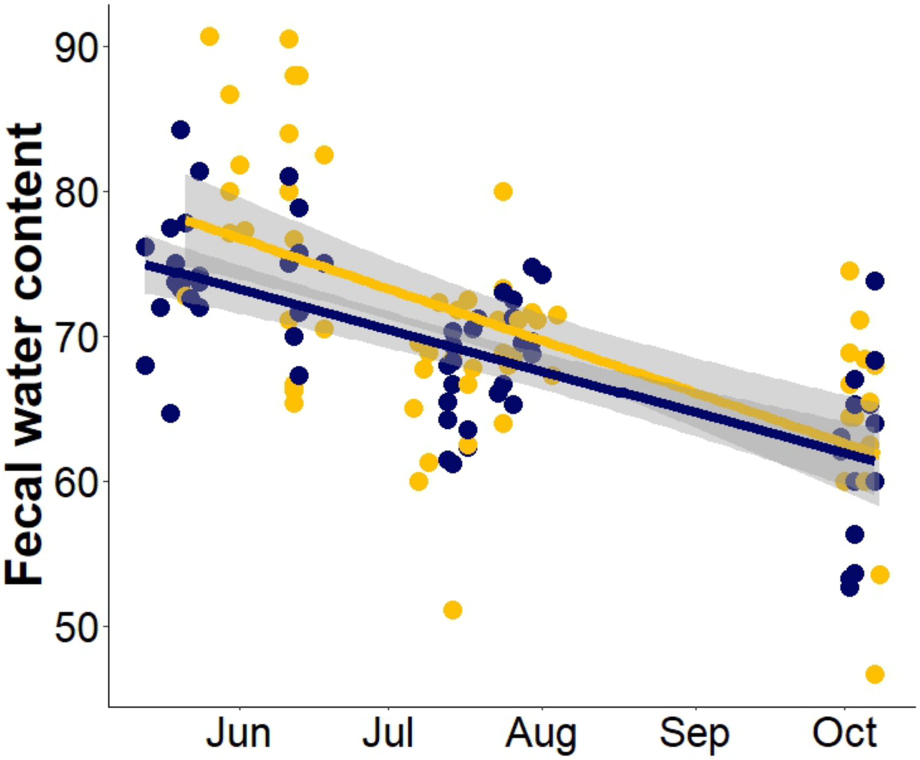

Figure 1.4. – Water content (percentage by mass) of bushbuck (*n* = 62, yellow) and kudu (*n* = 65, blue) fecal samples collected during the dry-season in Gorongosa National Park, Mozambique. Early dry season fecal samples were collected in May and June 2022; mid-dry season samples were collected in July and August 2021; late-dry season samples were collected in September and October 2021. Fecal water content decreased significantly across the dry season for both species ($\beta_{\text{Julian}} = -0.12$, $P < 0.001$, adj. $R^2 = 0.388$).

Plant-trait relationships.–Rate of water loss during the dry season was significantly related to both leaf mass per area and plant height after removal of a single outlier species that was rarely consumed by antelope (the makalani palm, *Hyphaene petersiana*). Plants with larger leaf mass per area and/or taller plants lost less water between the early and late dry season (leaf mass per area: $\beta = -0.086$, $P = 0.055$, adj. $R^2 = 0.10$; plant height: $\beta = -0.65$, $P = 0.042$, adj. $R^2 = 0.11$; Fig. A.4).

Selection for surface water.–Mean distance to surface water during the dry season did not differ between bushbuck and kudu ($P = 0.16$, Fig. 1.5). Moreover, standardized coefficients for distance to water were negative and significant in RSFs for both species throughout the dry season (bushbuck: $\beta = -0.33$, $P = 0.002$, kudu: $\beta = -0.22$, $P < 0.001$), indicating strong and consistent selection for surface water. As the dry season progressed, however, we observed temporal changes in selection for surface water that differed between bushbuck and kudu. Bushbuck selected surface water more strongly as the dry season progressed ($\beta = -0.06$, $P < 0.001$, adj. $R^2 = 0.746$), whereas strength of selection for surface water by kudu declined later in the dry season ($\beta = 0.02$, $P = 0.001$, adj. $R^2 = 0.508$; Fig. 1.6).

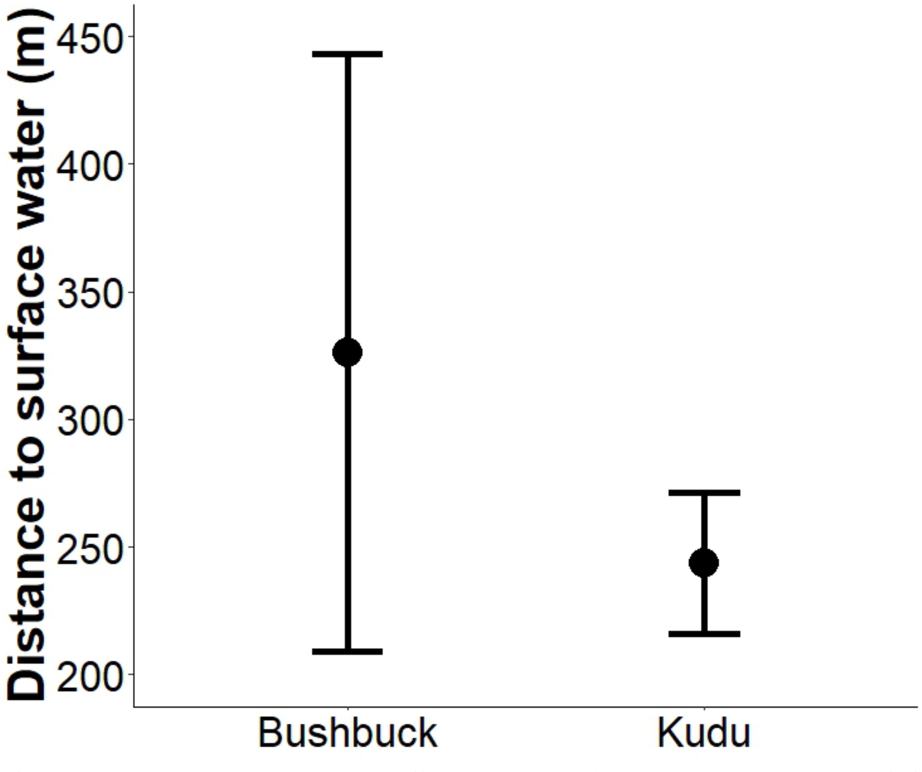

Figure 1.5. – Mean (± 95% CI) distance to surface water (e.g., perennial pans and rivers) of GPS locations obtained for bushbuck ($n = 12$) and kudu ($n = 12$) during the dry season (5 July–31 October 2021) in Gorongosa National Park, Mozambique. Distance to surface water did not differ between species ($P = 0.16$, 2 sample *t*-test).

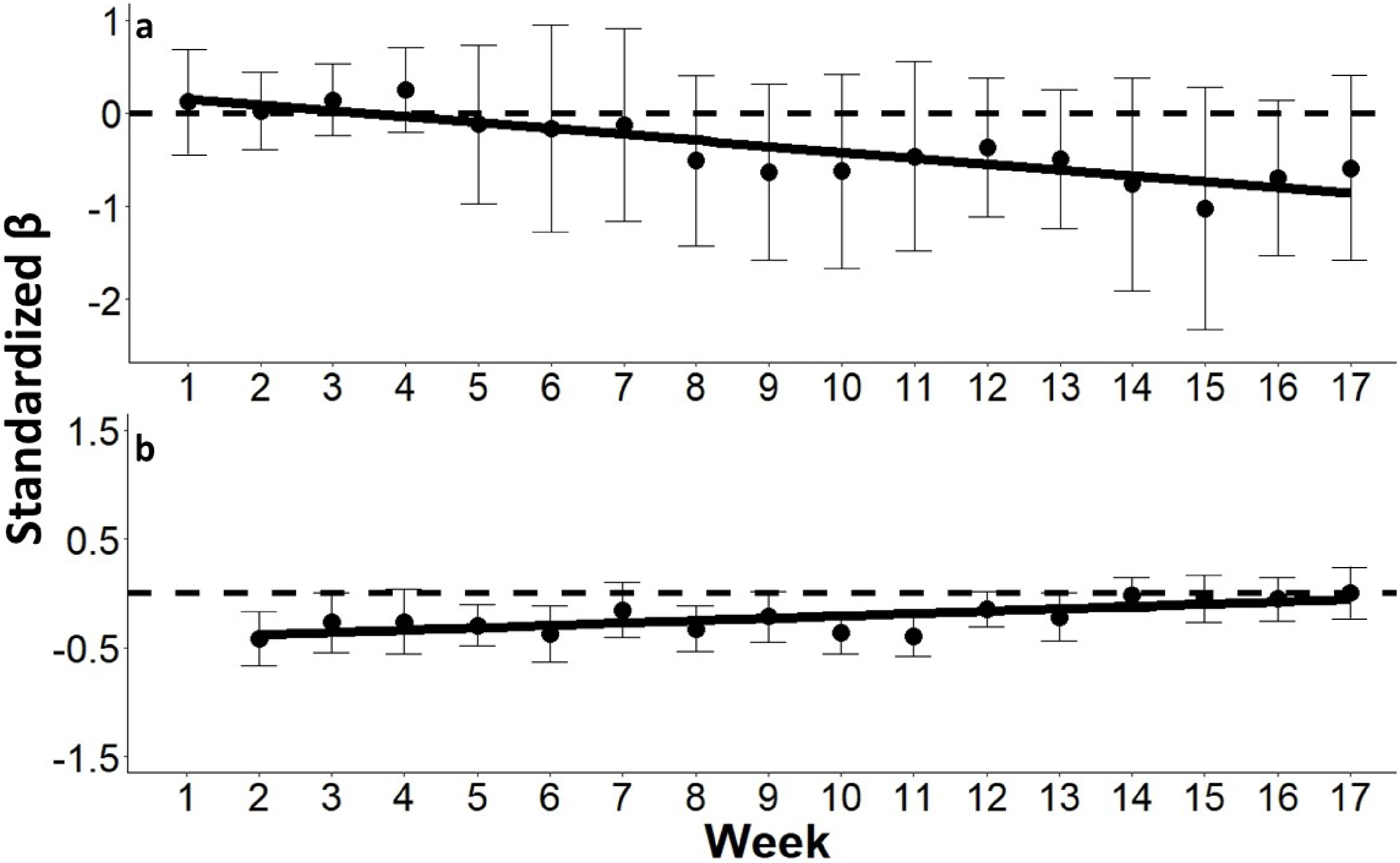

Figure 1.6. – Simple linear regressions of weekly changes in selection for surface water (standardized coefficient from GLMM) in Gorongosa National Park, Mozambique for (a) collared bushbuck and (b) collared kudu during the dry season (5 July–31 October 2021). Strength of selection for surface water changed significantly during the dry season for both species, but in opposite directions (bushbuck: $\beta = -0.06$, $P < 0.001$, adj. $R^2 = 0.746$; kudu: $\beta = 0.02$, $P = 0.001$, adj. $R^2 = 0.508$).

Surface water consumption.–Drinking bout duration did not change during the dry season for either species based on 7 weeks of audio recordings for kudu and 9 weeks of camera trap recordings for bushbuck (kudu: $\beta = -0.02$, $P = 0.974$, adj. $R^2 = 0.00$; bushbuck: $\beta = -2.41$, $P = 0.272$, adj. $R^2 = 0.004$; Fig. 1.7). Mean duration of drinking bouts was significantly longer for bushbuck than kudu (bushbuck: 73.8 ± 12.7 s, kudu: 29.7 ± 2.6 s). Camera recordings also indicated that the weekly number of drinking bouts by bushbuck (7.75 ± 5 drinking bouts/week) did not change during the period of camera deployment ($\beta_{Week} = -0.12$, $P = 0.887$, adj. $R^2 = 0.00$; Fig. A.5). Audio recordings revealed similar results for kudu ($\bar{x} = 3.29 \pm 0.4$ drinking bouts/week; $\beta_{Week} = 0.20$, $P = 0.304$, adj. $R^2 = 0.05$; Fig. A.6). The relationship between mean number of weekly drinking bouts recorded aurally and mean number of weekly drinking bouts estimated from GPS data (Table A.2) during the same period was positive and significant ($\beta = 0.40$, $P = 0.037$, adj. $R^2 = 0.537$) for kudu, suggesting that audio loggers were an effective means of identifying instances of drinking.

Bushbuck spent significantly more time drinking than kudu (bushbuck: 567 ± 185 s/week, kudu: 98 ± 30 s/week; $\beta_{\text{Species}} = -484.2$, $P = 0.005$, adj. $R^2 = 0.65$; Fig. 1.8), but mean time spent drinking did not change across weeks for either species ($\beta_{\text{Julian}} = -0.37$, $P = 0.89$; Fig. A.7). Because data collection for bushbuck and kudu only overlapped for two weeks, we conducted an additional analysis using data just from those two weeks; results were qualitatively similar ($\beta_{\text{Species}} = -565.8$, $P = 0.033$, $\beta_{\text{Julian}} = -4.21$, $P = 0.50$, adj. $R^2 = 0.99$).

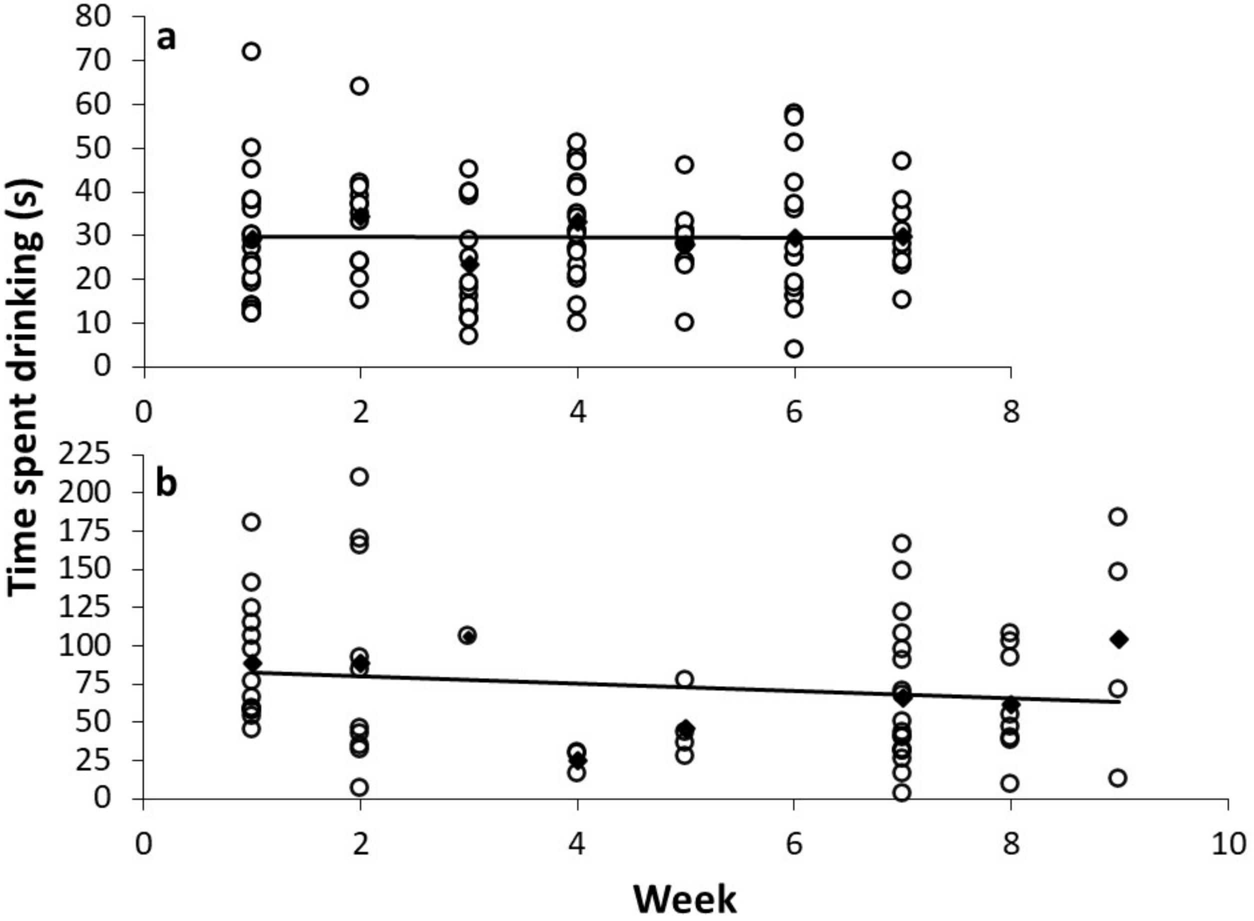

Figure 1.7. – Simple linear regressions of weekly drinking bout duration in Gorongosa National Park, Mozambique for (a) kudu (monitored July–August 2021, *n* = 102 drinking bouts, 3–7 kudu monitored per week), and (b) bushbuck (monitored August–October 2021, *n* = 62 drinking bouts). Open circles represent individual observations and closed diamonds show weekly means. Drinking bout duration did not change during the dry season for either species (kudu: $\beta = -0.02$, $P = 0.974$, adj. $R^2 = 0.00$; bushbuck: $\beta = -2.41$, $P = 0.272$, adj. $R^2 = 0.004$).

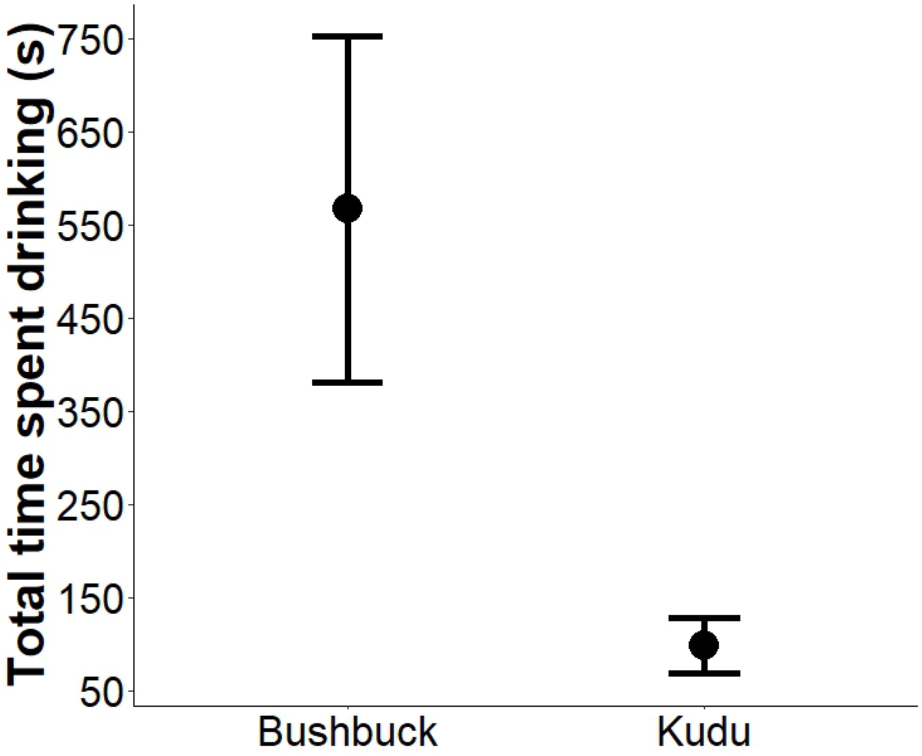

Figure 1.8. – Mean (± 95% CI) predicted total time spent drinking per week during the dry season by bushbuck (monitored August–October) and kudu (monitored July–August) in Gorongosa National Park, Mozambique. Predicted values for bushbuck were calculated by multiplying the mean number of drinking bouts per week (calculated across all 9 weeks of camera deployment) by the mean bout length per week. Predicted values for kudu were calculated by multiplying the mean number of drinking bouts per week (calculated across monitored individuals within each week) by the mean bout length per week.

DISCUSSION

Surface water dependence can dictate the distributions of large-herbivore populations in savanna ecosystems and can drive corresponding patterns of competition and predation during the dry season (Harrington et al. 1999, Ogutu and Owen-smith 2003, Mosser et al. 2009). This trait is generally considered to scale positively with body size (Woodall et al. 1999, Hempson et al. 2015). However, Veldhuis et al. (2019) argued that the positive relationship between dung moisture content (a common metric of surface water dependence) and body size was driven primarily by a few outliers at the extreme ends of the size continuum (e.g., Kirk's dik dik, *Madoqua kirkii*, on the lower end and elephant, *Loxodonta africana*, on the upper), and that surface water dependence is more strongly influenced by nuanced interspecific variation in morphology, physiology, and behavioral plasticity. We used a diverse suite of data streams to test the prediction that surface water dependence scales positively with body mass in two closely related species of browsing antelope, and this prediction was not supported. Kudu, the larger of the two species by ~4×, consumed less preformed water, spent less time drinking, and relaxed their selection for surface water as the dry season progressed. In contrast, bushbuck consumed more preformed water, spent more time drinking, and strengthened their selection for surface water later in the dry season. These results support the contention of Veldhuis et al. (2019) that body size alone is likely insufficient for predicting surface water dependence of species in diverse large-herbivore communities.

Proximity to surface water (Redfern et al. 2003, Valeix et al. 2009, Smit 2011) and dung moisture content (Woodall et al. 1999, Kihwele et al. 2020) are commonly used proxies of surface water dependence in large herbivores. Our results suggest, however, that these metrics may sometimes be misleading. Fecal water content did not differ between bushbuck and kudu in our study, nor did mean proximity to surface water. Data on dietary water intake, selection for surface water, and time spent actively drinking told a different story, however, and strongly indicated that bushbuck are more dependent on surface water than kudu. These results underscore the need for more mechanistic research on the drivers of surface water dependence in large herbivores, and suggest that capitalizing on recent advances in methods for monitoring herbivore behavior and physiology (e.g., bio-logging technology; Ditmer et al. 2015, Wilmers et al. 2015) may provide novel insights into herbivore ecology across levels of organization.

As the dry season progressed, both bushbuck and kudu consumed a greater proportion of plant species that retained their water content, ostensibly in an effort to maintain dietary water intake as surface water became increasingly scarce. This shift in diet, however, did not appear to carry a nutritional cost. Dietary protein content did not change for either species as the dry season progressed, and bushbuck actually increased dietary energy intake later in the season. These temporal patterns (or lack thereof) in diet quality were attributable to changes in diet composition for bushbuck, whose diets contained a greater proportion of high-quality (with respect to both DP and DE) plants later in the dry season, but not for kudu. Interspecifically, our results are broadly consistent with Daskin et al. (2023): bushbuck diets generally contained more DP than kudu diets, whereas kudu diets were higher in DE. The opposing direction of the scaling relationships between these two metrics of diet quality and body size is intriguing, and adds further support to the contention of Daskin et al. (2023) that DP may be a more useful metric of diet quality in the tropics, where large herbivores do not accumulate large fat stores (influenced more by DE than DP) to support reproduction (Jönsson 1997, Sainmont et al. 2014, Stephens et al. 2014).

One constraint on comparative inferences about time spent drinking by bushbuck and kudu in our study stems from the limited temporal overlap of audio and video data (2 weeks) for the two species. This limitation resulted from the differential timing of collaring operations, during which bio-loggers were deployed, and establishment of the camera trap array. Prior to deploying camera traps at perennial water sources within bushbuck home ranges, it was necessary to collect several weeks of GPS-collar data to estimate those home ranges, and during that time bio-loggers placed on kudu were recording continuous audio data. We attempted to overcome this constraint in several ways. First, because GPS-collar data for both species overlapped for the entirety of the dry season, we used resource selection functions to evaluate whether patterns of surface water selection by either species changed markedly outside the 2-week period of overlap in audio/video data. This analysis was based on the premise that any marked changes in time spent drinking would likely be mirrored to some degree by changes in selection for surface water. In contrast to this, temporal trends in surface water selection by both species appeared comparable within and outside the period of audio/video overlap. We also compared results of our analyses of total time spent drinking for the entirety of each data stream to results of similar analyses limited to the two weeks of audio/video overlap, and our conclusions did not differ qualitatively.

Another limitation of our study stemmed from the shifting diets of antelope, and of bushbuck in particular, as the dry season progressed. Previous data on *Tragelaphus* spp. diets in Gorongosa have been collected largely during the mid- to late-dry season. One consequence of using those data to generate our plant species list for sampling DP, DE, and preformed water content was that those species often comprised <50% (our threshold for inclusion in diet-based analyses) of sampled diets during the early dry season. This limited our early dry season sample size for bushbuck, reducing our ability to detect temporal changes in diet characteristics of bushbuck during the dry season. Nevertheless, that we did still detect significant changes in diet quality and water content in bushbuck suggests that our results are robust to this limitation. More broadly, our results suggest that future studies should not take for granted that diet composition and associated dietary traits are invariant even with a season, and that longitudinal sampling of diets (e.g., Walker et al. 2023) has considerable potential to shed new light on the nuanced causes and consequences of dietary variation within and among species.

Africa is one of the most vulnerable parts of the world to climate change (Serdeczny et al. 2017), and a thorough understanding of how large herbivores are likely to cope with such changes is imperative for predicting population- and community-level responses. Like much of the continent, Gorongosa is projected to become hotter and drier over the next century, and changes in the distribution and availability of surface water will likely have strong effects on Africa's diverse large-herbivores communities. As obligate browsers, bushbuck and kudu are often considered to be surface-water independent (Kihwele et al. 2020). Our results demonstrate, however, that considerable variation in surface water dependence exists even within this guild, let alone across the browser-grazer continuum. This variation has likely been underappreciated owing to the technical challenges of quantifying drinking behavior in free-ranging large herbivores. Nevertheless, interspecific differences in surface water dependence could have important effects on the distribution and composition of large-herbivore communities as temperatures warm and droughts become more frequent and severe. For example, a hotter, drier climate is likely to intensify the tradeoff between water requirements and predation risk for species like bushbuck that are both smaller and more water dependent. Body size scales negatively with predation risk (Sinclair et al. 2003, Hopcraft et al. 2010), and thus smaller species that also depend more heavily on surface water are likely to experience increased predation as surface water becomes less available and individuals are forced to concentrate their activity around remaining water sources (Mosser et al. 2009, Veldhuis et al. 2019). Alternatively, heat sensitivity (i.e., the ability to dissipate excess heat) scales positively with body size (Porter and Gates 1969), and thus warming temperatures could increase the hydric costs of thermoregulation disproportionately more in larger-bodied species. This could, in turn, increase surface water dependence of larger species, a trend that would be particularly consequential for large grazers that are already heavily dependent on surface water. Disentangling and predicting such effects at the population and community levels will require mechanistic approaches that are grounded in the principles and concepts of physiology and behavior and that take full advantage of current tools and techniques for measuring animal responses to environmental variation. Our study takes an important step in that direction and provides both a conceptual and methodological basis for future efforts to more fully characterize variation in surface water dependence among large-herbivore species.

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Chapter 2: Sex-dependent variation in diet and space use of congeneric African antelopes

ABSTRACT

Sex-dependent variation in behavior is common in the animal kingdom, and sexual segregation, defined as the differential use of space (e.g., habitat and forage) and/or separate social aggregation of the sexes outside the mating season, is particularly common among ungulates. Yet, most research exploring the causes and consequences of sexual segregation in ungulates has focused on temperate species in the northern hemisphere; the more diverse herbivore assemblages found in the tropics and subtropics have received comparatively little attention. Many current hypotheses for explaining why the sexes of ungulates often live separately for much of the year are based on the tenet that a polygynous mating system resulted in increased sexual size dimorphism, which in turn perpetuated sexual segregation. An alternative hypothesis, recently based on phylogenetic path analyses, posits that sexual segregation evolved prior to sexual size dimorphism as a means of reducing intraspecific competition for food. Empirical tests of this hypothesis, however, are currently lacking. We tracked movements and analyzed diets of three closely related antelope species—greater kudu, (*Tragelaphus strepsiceros*), nyala (*Tragelaphus angasii*), and bushbuck (*Tragelaphus sylvaticus*)—to quantify patterns of sex-dependent niche partitioning and shed new light on the causes and consequences of sexual segregation. We hypothesized that key components of individuals' niche space differ between males and females, and predicted that the magnitude of those differences across species would be a function of the degree of size dimorphism between the sexes. Our results did not support this prediction, and although the nature and magnitude of differences in behavior between the sexes varied considerably among species, this variation appeared to be driven by factors other than size dimorphism. Bushbuck, the smallest species, showed the greatest sex-dependent difference in diet quality. Nyala showed the greatest difference in habitat use, and kudu, the largest species (~4× larger than bushbuck), showed the greatest difference in movement behavior. This variation suggests that sex-dependent niche partitioning, and ultimately sexual segregation, is influenced by a suite of ecological factors that do not all covary with sexual size dimorphism in ungulates.

Moreover, such variation also highlights the potential for intraspecific behavioral variation to contribute to patterns of coexistence in large-herbivore communities.

INTRODUCTION

Sex-dependent variation in behavior is a recurrent theme in the animal kingdom, and one manifestation of this fundamental pattern is sexual segregation—the differential use of space (e.g., habitat and forage) and/or separate social aggregation of the sexes outside the mating season (Conradt 1998; Ruckstuhl 1998, 2007; Barboza and Bowyer 2000; Bowyer 2004; Wearmouth and Sims 2008). A combination of physiological and ecological factors related to nutrition and/or predation risk, as well as social factors tied to reproductive success (e.g., development of fighting skills in males and the establishment of pre-rut dominance hierarchies), are theorized to have driven the evolution of sexual segregation (Main et al. 1996, Bowyer 2004), which has now been documented across a wide variety of taxa. For example, in blue tits (*Cyanistes caeruleus*), migratory behavior is biased toward females because they are less able to compete for food on breeding grounds during the nonbreeding season (Goodenough et al. 2010). In contrast, adult male grey seals (*Halichoerus grypus*) are more likely than females to display directed, long-distance movements outside of the breeding season owing to differences in diet and foraging strategies (Austin et al. 2004).

For several reasons, ungulates have often been the focus of research on the causes and consequences of sexual segregation (Bowyer 2022), and indeed, the number of publications on sexual segregation in ungulates has increased at twice the rate of any other mammalian taxon since the 1970s (Bowyer 2004). Many ungulates exhibit considerable sexual dimorphism (the proximate mechanism thought to drive sexual segregation) and segregate strongly apart from breeding. In addition, ungulates have pronounced effects on fundamental ecosystem processes that are likely modulated by how the sexes partition space, habitat, and forage (Bowyer 2004). Importantly, however, the overwhelming majority of work has centered on temperate ungulates in the northern hemisphere (Main et al. 1996, Bleich et al. 1997, Kie and Bowyer 1999, Mooring et al. 2005, Long et al. 2009, Bowyer 2022). Many tropical and subtropical ungulates exhibit significant sexual size dimorphism but do not segregate as strongly. Accordingly, extending this line of inquiry to the more diverse herbivore assemblages found closer to the equator holds potential to shed new light on the causes and consequences of sexual segregation.

Many current hypotheses for explaining why the sexes of ungulates often live separately for much of the year are based on the tenet that a polygynous mating system resulted in increased sexual size dimorphism, which in turn perpetuated sexual segregation (Bowyer 2004). Male-male competition in polygynous ungulates, coupled with female selectivity, has rendered males 20–70% larger than females on average (Barboza and Bowyer 2001). Differences in body size between the sexes have resulted in associated differences in physiological and morphological traits that scale (often allometrically) with body size, which can profoundly influence the ecology and life-history of the sexes (Jenks et al. 1994; Barboza and Bowyer 2000, 2001).

An alternative hypothesis that has received less attention suggests that sexual size dimorphism arose from natural selection as a result of niche separation that favored different optimal sizes for males and females (Shine 1989, Blanckenhorn 2005). Slatkin (1984) used theoretical modeling to show that sexual dimorphism can evolve purely from ecological selective pressures. The intersexual niche divergence or 'ecological sexual dimorphism' hypothesis posits that sexual size dimorphism emerged as a consequence of sexual segregation, which evolved to reduce intraspecific competition for food. Empirical tests of this hypothesis are limited and have produced mixed results (Fairbairn 1997). Moreover, even when results have been supportive, it has often been difficult to exclude the hypothesis that niche differentiation evolved as a consequence of preexisting sexual dimorphism (Ralls 1976, Shine 1989, Fairbairn 1997).

For many decades, Jarman's (1974) model of ungulate evolution, which posits that polygyny evolved as a consequence of African ungulates' progression toward larger body size, diet specialization, and social living as they moved into open habitats, played a central role in ungulate ecology (Janis 1982, Bowyer et al. 2020, Szemán et al. 2021). Jarman (1983) subsequently built on his original model by combining the effects of natural and sexual selection, and hypothesized that polygyny and sexual size dimorphism evolved via directional selection as a result of male-male combat and the monopolization of reproductive opportunities by larger-bodied males (Jarman 1983). Phylogenetic testing of this model confirmed that polygyny did evolve after ungulates moved into open habitats, and that sexual size dimorphism is strongly and positively correlated with a polygynous mating system (Pérez-Barbería et al. 2002). Yet, it was not possible in that study to rule out an alternative explanation in which competition for food resources resulted in larger male body size. More recent work by Cassini (2022) supported an ‘ecological’ version of Jarman’s model based on phylogenetic confirmatory path analysis (Shipley 2009, van der Bijl 2018), and in doing so provided an alternative explanation for the evolution of size dimorphism in artiodactyls. Cassini’s (2022) ecological model overcomes some of the main limitations of Jarman’s original model. For example, the ecological model explains the existence of mammalian families that have polygynous mating systems and sexually segregate, but are monomorphic. And, importantly, Cassini’s ecological model predicts that niche partitioning produced by natural selection, not sexual selection, led to the evolution of sexual size dimorphism (Cassini 2022). In other words, sexual segregation evolved prior to sexual size dimorphism.

Mozambique’s Gorongosa National Park is an ideal system for assessing the relative merit of the intersexual niche divergence hypothesis as a general explanation for sexual segregation in ungulates. Gorongosa’s mosaic of rich habitats was formerly home to 44 species of large mammals (≥5 kg mass; Tinley 1977), but the Mozambican Civil War (1977–1992) reduced the park’s large-mammal populations by >90% (Stalmans et al. 2019). Restoration efforts in the subsequent decades have largely been successful, and many herbivore populations have returned to their prewar numbers. In contrast, large-carnivore (i.e., lion (*Panthera leo*), wild dog (*Lycaon pictus*), leopard (*Panthera pardus*), and spotted hyena (*Crocuta crocuta*)) recovery has progressed much more slowly (Stalmans et al. 2019). Lions were the only large carnivore to persist postwar, albeit in low numbers (≈100 individuals; Bouley et al. 2018), and wild dog, leopard, and hyena reintroductions only began in 2018, 2019, and 2022, respectively. This ecological asymmetry has, until recently, provided herbivores with a unique opportunity to optimize their behavior energetically in the face of limited predation pressure, and allowed us to explore mechanisms of intersexual niche differences in the context of ostensibly diminished predation risk.

Of the many large-herbivore species that occupy Gorongosa, three congeneric antelope—greater kudu, (*Tragelaphus strepsiceros*), nyala (*Tragelaphus angasii*), and bushbuck (*Tragelaphus sylvaticus*)—are ideal for studying relationships among intersexual niche differences, sexual size dimorphism, and patterns of sexual segregation. These three related antelope species are sympatric within woodland and grassland habitats and all strongly select for resources associated with termite mounds, which provide high-quality forage and shade but make up <2% of the landscape (Daskin et al. 2023). All three species have similar life histories, are predominately browsers, and outside of breeding, females and mature males typically segregate (though importantly, bushbuck are non-seasonal breeders, whereas nyala and kudu are seasonal breeders; Lobão Tello and Van Gelder 1975, Tinley 1977, Estes 2012, Hempson et al. 2015). Furthermore, all three species exhibit considerable sexual size dimorphism, with males being approximately 1.4 to 1.9 times larger than females. Despite these similarities, the 3 species differ four-fold in body mass (based on average mass of adult females; Kingdon 2015). These attributes provide a unique opportunity to assess how size dimorphism contributes to niche differentiation and sexual segregation in ungulates.

We tracked the movements of male and female bushbuck, nyala, and kudu using GPS collars and collected fecal samples for analysis of diet composition and quality to better understand the nature and magnitude of niche partitioning between the sexes. In the context of Cassini's (2022) ecological model, we used those data to test the hypothesis that key components of individuals' niche space (e.g., diet composition, habitat use and movement patterns) differ between male and female Tragelaphine antelope because (1) diet quality scales inversely with body size, and thus (2) sexual dimorphism that evolved in response to intraspecific competition for food resources should be correlated with sex-dependent differences in foraging behaviors. We predicted that males and females of all three species would consume significantly different diets, and that the quality of those diets (i.e., digestible energy and/or protein content, DE and DP, respectively) would be higher for females (Bell 1971, Jarman 1974, Bowyer 2004). Furthermore, we predicted that females of all 3 species would spend relatively more time in high-quality foraging habitat (e.g., termite mounds and floodplain grasslands; Atkins et al. 2019, Daskin et al. 2023), and that females would be more selective while foraging, which would manifest in more tortuous movements (i.e., taking more steps across a wider range of turning angles; Mooring et al. 2005). Conversely, we predicted that males would spend more time in habitats that provided higher biomass of lower-quality forage. We also predicted that males of all 3 species would have larger home ranges than females (Ofstad et al. 2016, Noonan et al. 2020, Daskin et al. 2023), and that step length should scale positively with both body size and home range size because larger individuals require greater biomass of forage and often travel further to obtain it (Harestad and Bunnel 1979, Noonan et al. 2020, Daskin et al. 2023). Finally, we predicted that the magnitude of differences in diet, habitat use, and movements across species would be a direct function of the degree of size dimorphism between males and females. Specifically, we predicted that because adult nyala show the greatest magnitude of sexual size dimorphism (males 1.9 times larger than females), differences in diet, habitat use, and movement between male and female nyala would be greater than differences between male and female kudu and bushbuck (males 1.4 and 1.5 times larger than females, respectively).

MATERIALS AND METHODS

Study area.–Mozambique's Gorongosa National Park is situated at the southern end of the Great Rift Valley (18.96°S, 34.36°E) and supports a diverse array of large mammals (Fig. 2.1). Our study area in the core of the park included Lake Urema and its expansive floodplain grassland, as well as *Acacia*, palm, and broadleaf savanna woodland habitats (Pansu et al. 2019, Stalmans et al. 2019). Gorongosa is highly seasonal and receives 700–900 mm of rainfall annually, mostly during the wet season (November to April).

Within woodland habitats, fungus-farming termites (Macrotermitinae) concentrate nutrients and moisture within their nests ('mounds'), which in turn supports the growth of dense, high-quality woody plant communities (Sileshi et al. 2010, Seymour et al. 2014, Daskin et al. 2023). As a result, termite mounds often represent 'islands' of nutrient-rich food that attract browsing ungulates. Daskin et al. (2023) reported that bushbuck, nyala, and kudu in Gorongosa all selected habitat near mature termite mounds and consumed diets dominated by woody plants that occurred at higher densities on mounds than in the surrounding matrix. The floodplain grassland surrounding Lake Urema also provides a source of high-quality forage for Gorongosa's large herbivores. The floodplain supports leguminous shrubs and forbs that contain relatively high levels of digestible energy and protein, and although browsing herbivores rarely venture into open floodplain habitat, previous work in the park demonstrated that bushbuck in the floodplain consumed diets richer in energy and protein and were in better nutritional condition than woodland bushbuck (Atkins et al. 2019, Walker et al. 2023).

Animal capture and handling.–During the dry seasons of 2018, 2019, and 2021 we captured adult bushbuck (*n* = 24 females; 13 males), nyala (*n* = 5 females; 7 males), and kudu (*n* = 33 females; 6 males) as part of the long-term Allometry of Spiral-Horned Antelopes: Movement Ecology & Diet (ASHAMED) project (Daskin et al. 2023). We

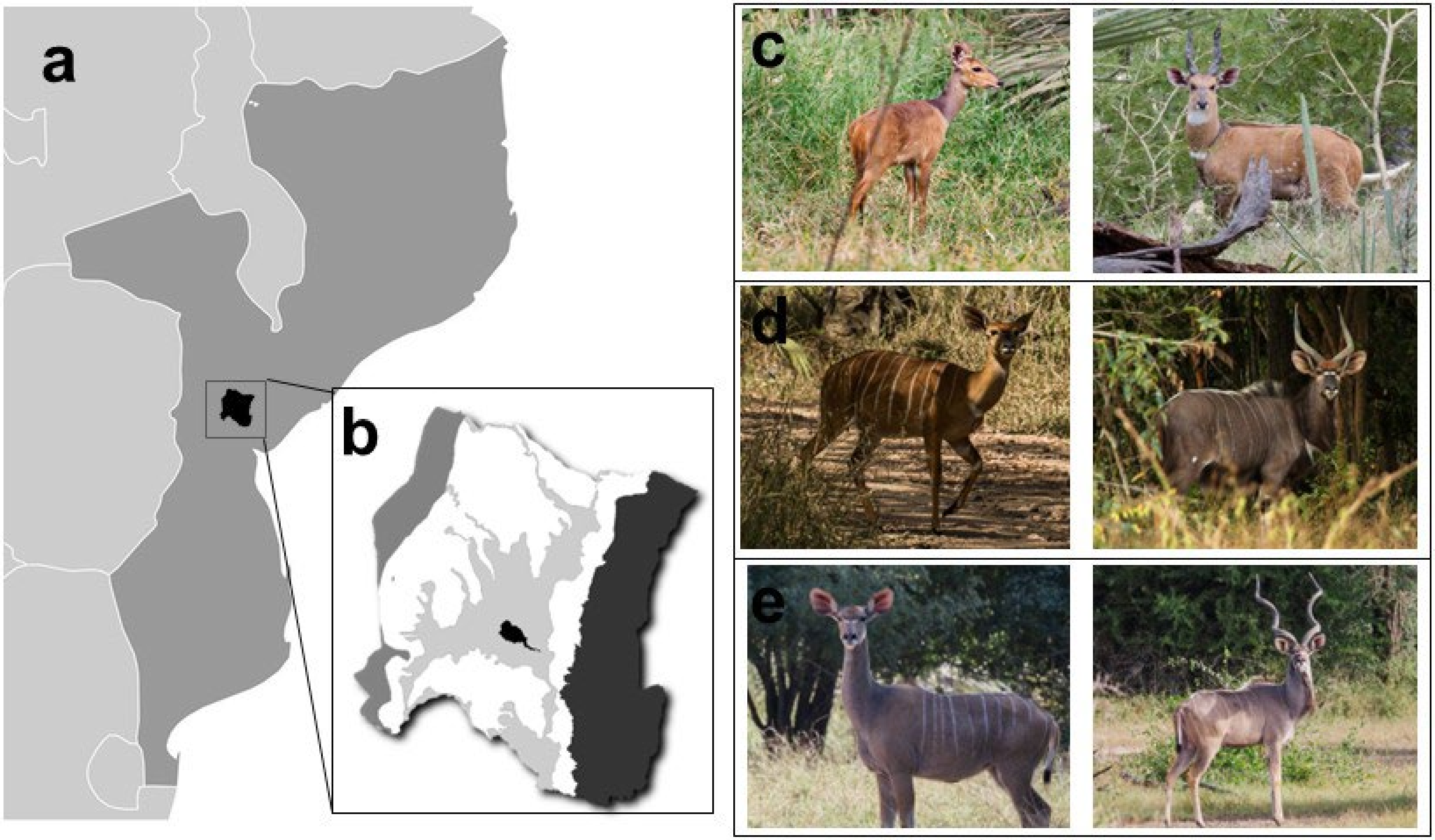

Figure 2.1. – (a) Gorongosa National Park is located in central Mozambique and (b) consists of four major habitat zones (from left: western escarpment [medium gray], woodland [white], floodplain [light gray], and eastern escarpment [dark gray], as well as Lake Urema [black]). (c) Female (left) and male (right) bushbuck. (d) Female (left) and male (right) nyala. (e) Female (left) and male (right) greater kudu. Nyala photos used with permission from Bart Wursten.

chemically immobilized all 3 species via remote injection of a combination of thiafentanil, ketamine, and azaperone. Each individual was fitted with an iridium satellite GPS collar (VERTEX Lite, Vectronic Aerospace) programmed to record locations every 30 min for nyala and kudu and every 60 min for bushbuck. GPS collars were remotely triggered to drop off 1 year after deployment. All animal-handling procedures were approved by the Animal Care and Use Committees of the University of Idaho (IACUC-2019-32) and Princeton University (2075F-16) and were in accordance with guidelines established by the American Society of Mammologists (Sikes & The Animal Care and Use Committee of the American Society of Mammologists 2016).

Diet composition.– We quantified composition of bushbuck, nyala, and kudu diets using fecal DNA metabarcoding following protocols previously established in Gorongosa (Atkins et al. 2019, Branco et al. 2019, Guyton et al. 2020, Potter et al. 2022, Daskin et al. 2023, Walker et al. 2023). At the time of collaring, we collected a fecal sample (>5 pellets) directly from the rectum of immobilized individuals for subsequent diet analysis. To adequately capture the range of variation in dry-season diets, we collected additional fresh fecal samples from all 3 species across 4 dry seasons (2018–2019 and 2021–2022; *n* = 257 bushbuck, 37 nyala, and 132 kudu). We used GPS data and radiotelemetry to locate collared individuals, and we observed them and any conspecifics from a distance of 10–100 m until defecation occurred. At the time of defecation, we noted the individual's distance from the observer and the nearest landmarks to the site of defecation. We then searched the area for the fecal sample and collected a minimum of 5 fecal pellets that were clear of debris using nitrile gloves and an unused, plastic zip top bag. Samples were stored on ice in a portable cooler for less than 5 hrs until they were processed and preserved at the Gorongosa laboratory. Each sample was homogenized, and a pea-sized sub-sample was transferred to a labeled tube containing silica beads and DNA preservation buffer (Xpedition Stabilization/Lysis Solution, Zymo Research Corporation). To lyse cells, tubed samples were vortexed for 1 minute and were then frozen at -80°C until transport to the U.S. Prior to transport, each sample was subjected to an anti-viral heat treatment (72°C for 30 minutes) and re-frozen in accordance with the requirements of the United States Department of Agriculture (Permit 130123 to Robert M. Pringle).

Analyses of diet composition were conducted at Princeton University following protocols used previously for DNA metabarcoding of ungulate diets in Gorongosa (Atkins et al. 2019, Branco et al. 2019, Pansu et al. 2019, Guyton et al. 2020, Becker et al. 2021) and Kenya (Kartzinel et al. 2015, 2019, Kartzinel and Pringle 2020). Briefly, we amplified the P6 loop of the chloroplast *trn*L(UAA) intron, a region commonly used to metabarcode an array of plant taxa, using primers that contain a unique 8-nt tag at the 5' end, enabling pooling of uniquely identifiable PCR products for sequencing in a single high-throughput run (Taberlet et al. 2007). Extraction and sequencing were performed on an Illumina HiSeq 2500 and processed via the OBITools pipeline (Boyer et al. 2016). We discarded low-quality sequences, and the remaining sequences were considered molecular operational taxonomic units (mOTUs). After rarefying to the minimum number of sequence reads per sample, the dataset included 609 dietary mOTUs (172 in 2018; 165 in 2019; 136 in 2021; 136 in 2022). Post bioinformatic filtering, we identified plant sequences by matching them to an extensive reference library of DNA from plant specimens collected in Gorongosa (Pansu et al. 2019), and secondarily to a global reference library derived from the European Molecular Biology Laboratory database. Guyton et al. (2020) provide a detailed description of these protocols. From these data, we determined presence/absence and relative read abundance (RRA) of each plant species contained in each sample. Our analyses are based on RRA, the proportional representation of each mOTU per sample (Deagle et al. 2019, Daskin et al. 2023), and we only included samples in subsequent analyses when the plants for which we had diet quality (i.e., DP and DE) data accounted for ≥70% of the RRA in the sample (n = 176 of 426 for DE and n = 329 of 426 for DP). Sample sizes differed between the two nutritional metrics because we had estimates of DP for a much larger number of plant species than DE, and were therefore able to include more sampled diets in the DP analysis based on our established RRA threshold. Mean RRA of samples analyzed for DE was 84%, 89%, and 90% for bushbuck, nyala, and kudu respectively, and mean RRA of samples analyzed for DP was 90%, 92%, and 93% for bushbuck, nyala, and kudu respectively.

Diet quality.–We combined data on diet composition with detailed, plant species-specific data on nutritional quality obtained during previous studies (Atkins et al. 2019, Potter et al. 2022, Daskin et al. 2023) to estimate DE and DP content of sampled bushbuck, nyala, and kudu diets during the dry season. We calculated weighted averages of DE and DP for each sampled diet by using the RRA of each plant species in the diet (determined from the metabarcoding analysis) as the weighting factor (Atkins et al. 2019, Branco et al. 2019, Pansu et al. 2022, Walker et al. 2023). We then estimated mean DE and DP of each species' dry-season diet as the weighted average of DE and DP estimates across individuals using the total proportion of the diet accounted for (based on RRA, minimum of 70% required for inclusion) in each sample as the weighting factor.

Habitat distribution.–We mapped the distribution of termite mounds in Gorongosa using a digital terrain model (DTM) of the park derived from airborne light detection and ranging (LiDAR) data collected in August 2019 (mid-dry season). We used the hillshade tool to manually digitize mound locations and sizes based on differences in slope and shape (Daskin et al. 2023, Walker et al. 2023). The floodplain boundary was previously mapped by Atkins et al. (2019). Although spiral-horned antelope in Gorongosa rarely use the floodplain, we considered it important to account for that use when it occurred because of the high nutritional quality of the plants available in the floodplain. To quantify use of high-quality foraging habitat (i.e., termite mounds and/or floodplain grassland) by antelope during the dry season, we estimated 95% fixed-kernel utilization distributions (UDs) from each individual's GPS locations using the *adehabitatHR* package in R (Calenge 2019). We then calculated the proportion of the volume of each individual's UD that overlapped high-quality foraging habitat.

Movement metrics.– Tortuosity—a measure of how straight an animal's movement path is—is a useful metric for quantifying searching behavior (Benhamou 2004, Ihwagi et al. 2019). When an animal takes more steps across a wider range of turning angles while foraging, it is typically interpreted to indicate greater foraging selectivity (Mooring et al. 2005). The tortuosity of a search path can be reliably estimated by a sinuosity index that combines the mean cosine of changes in direction with the mean step length. Benhamou (2004) provided a general equation (2.1) for calculating sinuosity, S, where p is step length, c is the mean cosine of turning angles, and b is the coefficient of variation in step length:

$$S = 2\left[p\left(\frac{1+c}{1-c} + b^2\right)\right]^{-0.5}$$

This equation is suitable for a wide range of turning angle distributions and does not require a constant step length (Benhamou 2004). We estimated sinuosity of collared individuals' movement paths using continuous GPS locations obtained for all three species during the dry seasons of 2018, 2019, and 2021, and the *trajr* package (McLean 2020) in R.

We used the *adehabitatHR* package (Calenge 2019) in R to estimate 95% fixed-kernel home ranges during the dry season for the same sample of collared individuals. We estimated mean step lengths during the dry season using the *amt* package (Signer et al. 2023) in R.

Statistical analyses.–We visualized differences in dry-season dietary dissimilarity between sexes and among species using nonmetric multidimensional scaling (NMDS; Daskin et al. 2023; Walker et al. 2023). To test whether diet composition differed between the sexes of each species, we used permutational multivariate analysis of variance (perMANOVA; based on RRA, minimum of 70% required for inclusion). We ran 9999 permutations with sex as a blocking factor in the *vegan* package in R (Oksanen et al. 2022). We used 2-sample *t*-tests to test for significant differences in diet quality (DE and DP), use of high-quality foraging habitat, sinuosity, home range size, and mean step length between males and females of the same species. Finally, we separated females by lactation status (i.e., lactating, non-lactating) and used one-way analysis of variance (ANOVA) with Tukey's honestly significant difference (HSD) post hoc contrasts to test for significant differences in movement metrics among reproductive categories (i.e., lactating, non-lactating, male).

RESULTS

Diet composition and quality.–Male and female antelope consumed significantly different diets during the dry season (bushbuck: pseudo-$F_{1,255} = 8.22$, $P = 0.001$, $R^2 = 0.031$; nyala: pseudo-$F_{1,35} = 3.56$, $P = 0.002$, $R^2 = 0.092$; kudu: pseudo-$F_{1,130} = 2.03$, $P = 0.042$, $R^2 = 0.015$; Fig. B.1). Of the top 10 plant taxa (based on RRA) consumed by males and females of each species, 6 were consumed by both sexes (Tables B.1–B.3). However, the mean RRA of those plant species often differed markedly between the sexes (Fig. 2.2). DE was higher in male than female diets for bushbuck and kudu (bushbuck $P < 0.001$; kudu $P = 0.035$), but not for nyala ($P = 0.225$; Fig. 2.3). DP was higher in the diets of male than female bushbuck ($P < 0.001$) but did not differ between male and female nyala ($P = 0.76$) or kudu ($P = 0.36$; Fig. 2.4).

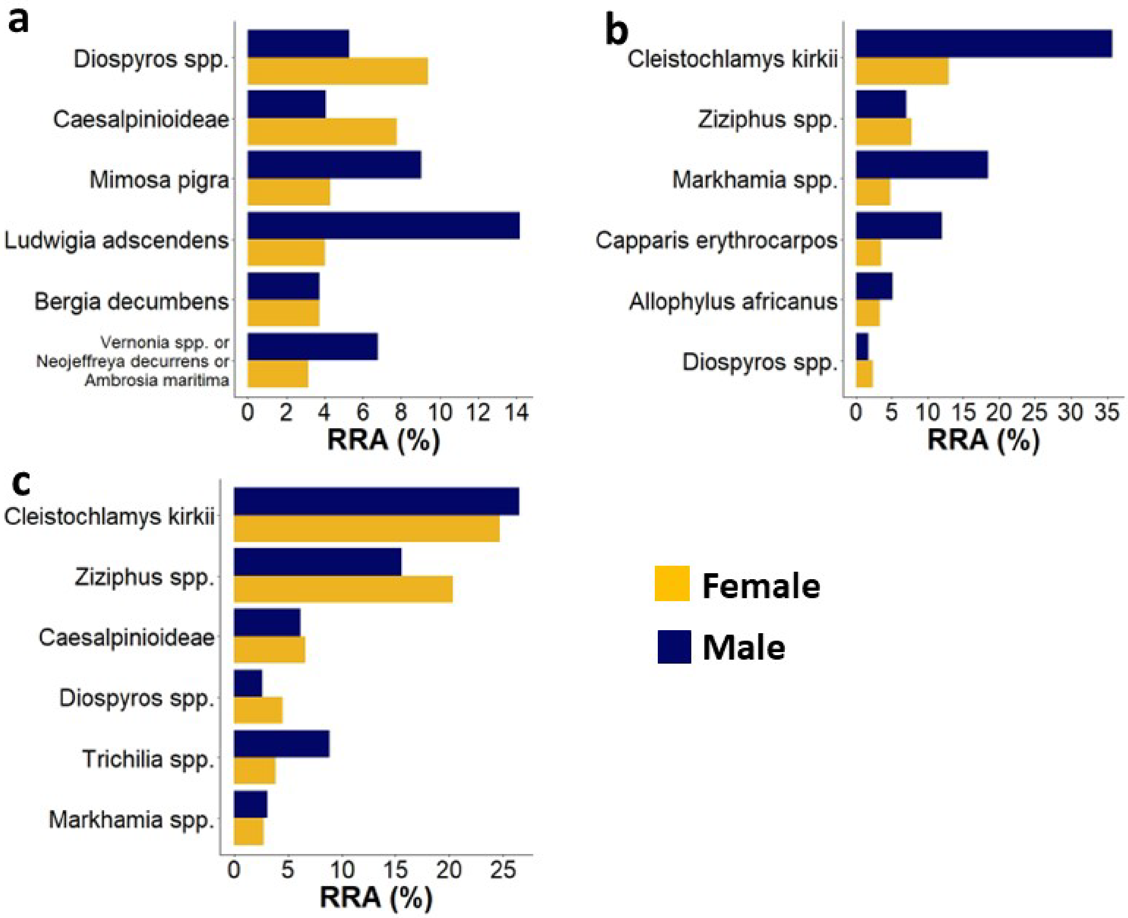

Figure 2.2. –Mean relative read abundance (RRA, determined from metabarcoding analysis of fecal samples) of the top 6 plant taxa consumed by female (yellow) and male (blue) bushbuck (a), nyala (b), and kudu (c) during the dry season (May–October) of 2018–2019 and 2021–2022 in Gorongosa National Park Mozambique.

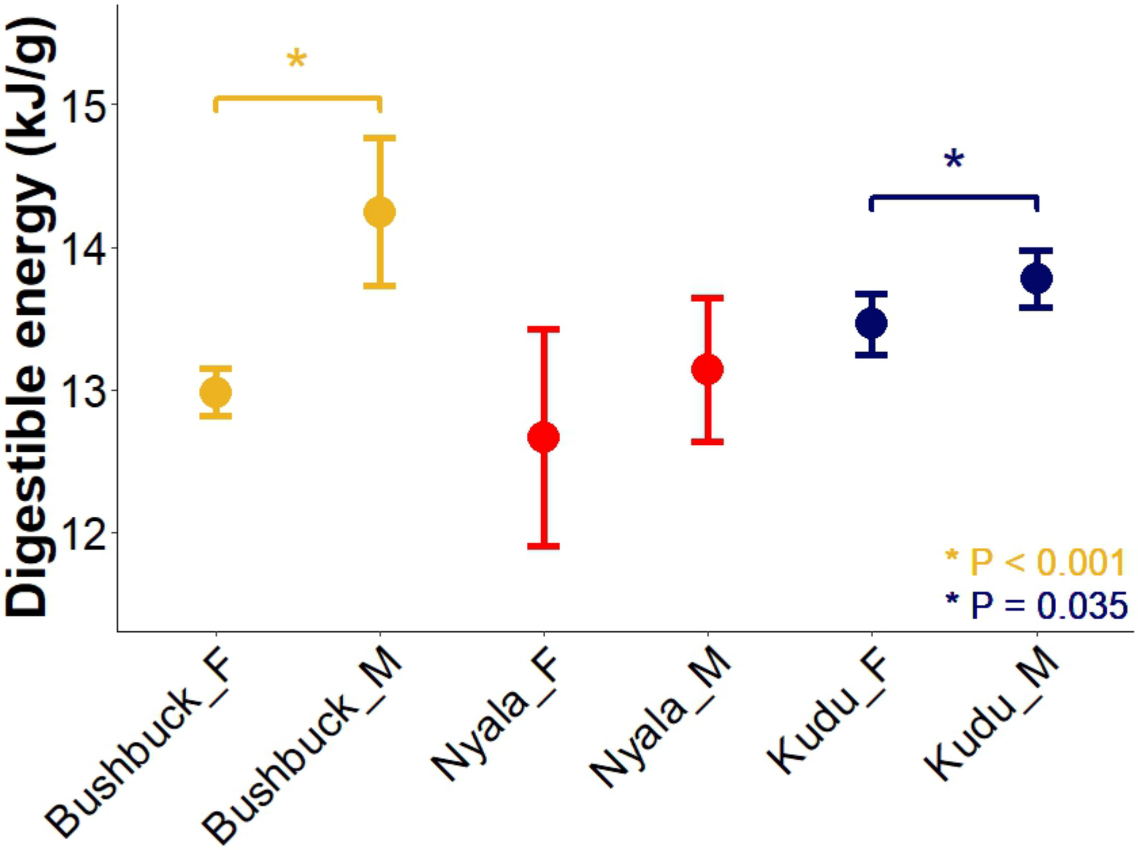

Figure 2.3. – Mean (± 95% CI) digestible energy in the diets of female and male bushbuck, nyala and kudu during dry seasons (May–October) of 2018–2019 and 2021–2022 in Gorongosa National Park, Mozambique (bushbuck females, *n* = 56; bushbuck males, *n* = 27; nyala females, *n* = 6; nyala males, *n* = 11; kudu females, *n* = 47; kudu males, *n* = 29). Means were compared using 2-sample *t*-tests; asterisks indicate significant pairwise differences between sexes.

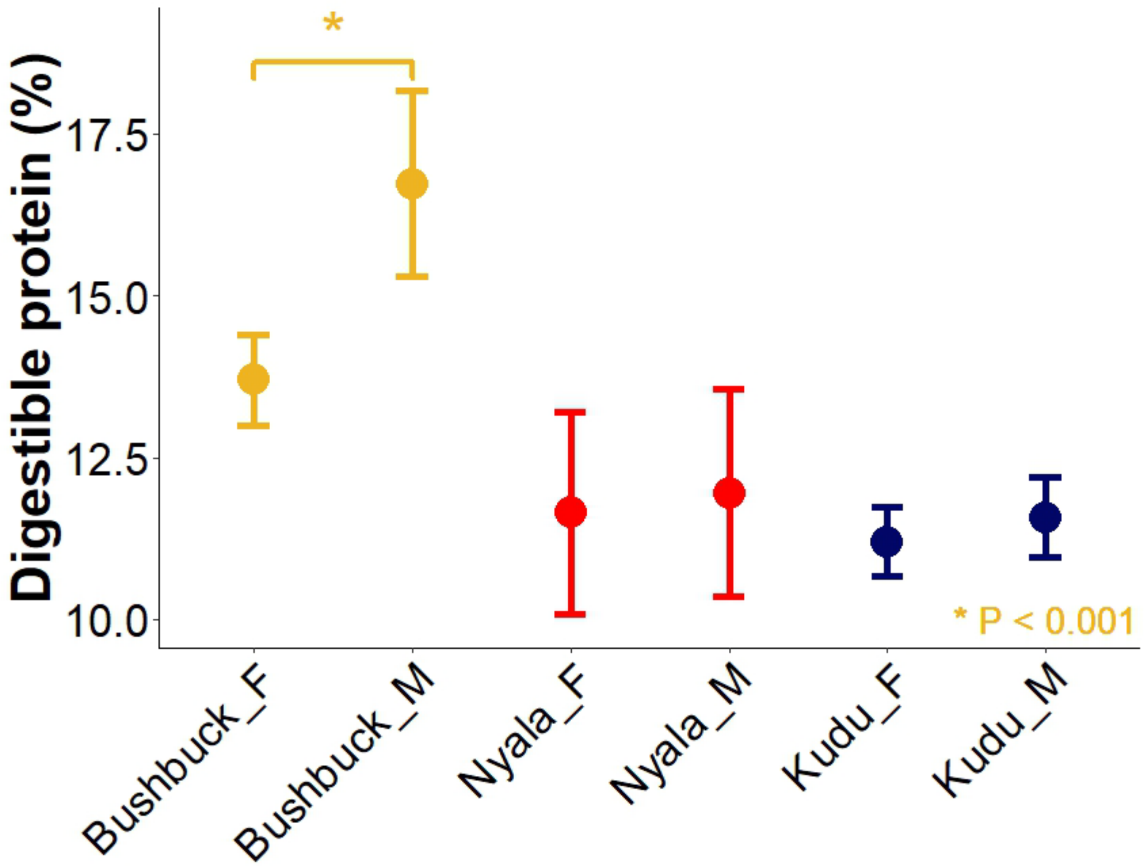

Figure 2.4. – Mean (± 95% CI) digestible protein in the diets of female and male bushbuck, nyala and kudu during dry seasons (May–October) of 2018–2019 and 2021–2022 in Gorongosa National Park, Mozambique (bushbuck females, *n* = 128; bushbuck males, *n* = 68; nyala females, *n* = 12; nyala males, *n* = 12; kudu females, *n* = 62; kudu males, *n* = 47). Means were compared using 2-sample *t*-tests; asterisks indicate significant pairwise differences between sexes.

Movement metrics.–Bushbuck spent more time in high-quality foraging habitats (termite mounds and floodplain grassland) than nyala or kudu, but there was no difference in use of those habitats between male and female bushbuck ($P = 0.94$; Fig. 2.5) or kudu ($P = 0.12$; Fig. 2.5). In contrast, male nyala spent more time than females in high-quality foraging habitat ($P = 0.01$; Fig. 2.5). This pattern persisted for nyala when we distinguished between lactating and non-lactating females; males exhibited significantly ($P = 0.095$; Fig. B.2) greater use of high-quality foraging habitat than lactating females. Use of high-quality foraging habitat did not differ among males, lactating females, and nonlactating females for either bushbuck or kudu ($P \geq 0.11$; Fig. B.2).

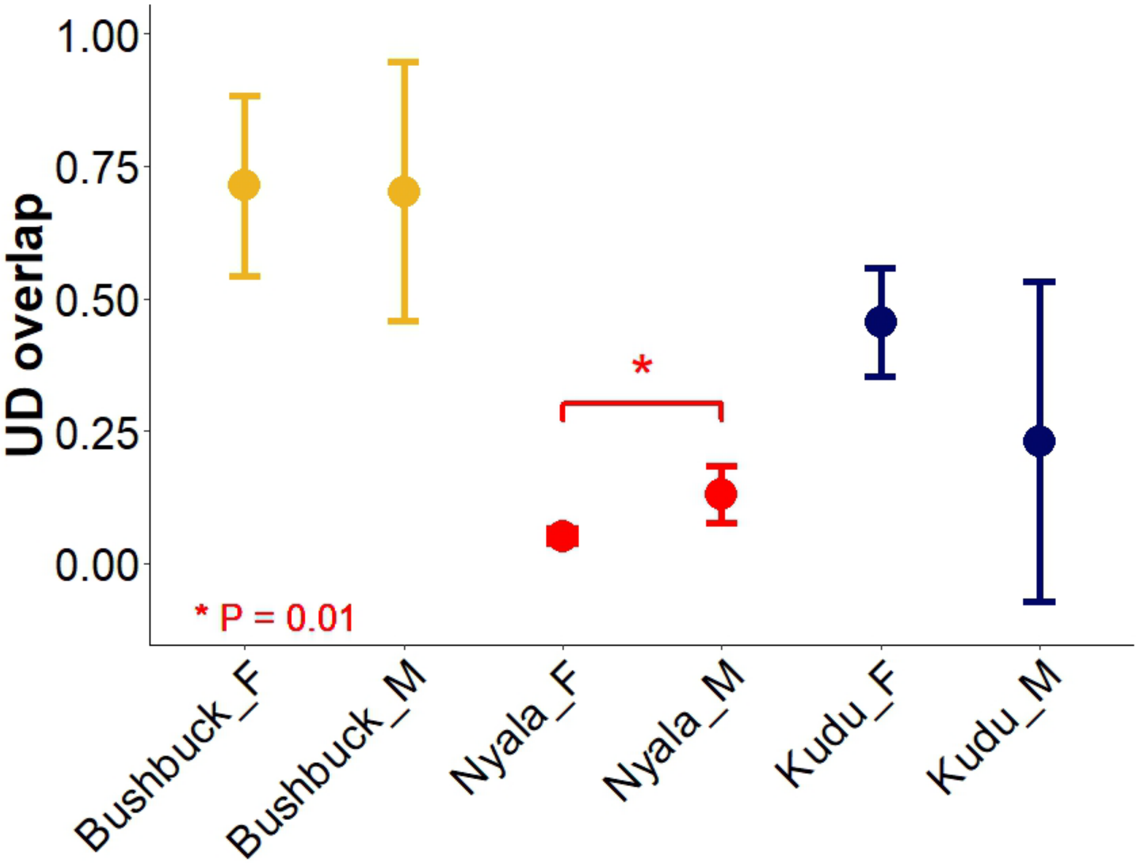

Figure 2.5. – Mean (± 95% CI) proportion (volumetric) of antelope (female and male bushbuck, nyala and kudu) utilization distributions (UD) that overlapped high-quality foraging habitats (i.e., termite mounds and floodplain grassland Atkins et al. 2019, Daskin et al. 2023) during the dry seasons (May–October) of 2018–2019 and 2021 in Gorongosa National Park, Mozambique (bushbuck females, *n* = 24; bushbuck males, *n* = 13; nyala females, *n* = 5; nyala males, *n* = 7; kudu females, *n* = 33; kudu males, *n* = 6). Means were compared using 2-sample *Z*-tests; asterisks indicate significant pairwise differences between sexes.

Home range area (km^2) scaled positively with body size across species, and for all 3 species male home ranges were significantly larger than females (bushbuck $P = 0.025$; nyala $P = 0.017$; kudu $P = 0.024$; Fig. 2.6). These differences largely persisted for bushbuck and kudu when females were separated by reproductive status, but did not persist for nyala (Fig. B.3). Step length (m) also scaled positively with body size, and lengths were significantly greater for male than for female bushbuck ($P = 0.025$) and kudu ($P = 0.036$), but not for nyala ($P = 0.18$; Fig. 2.7). Again, this pattern persisted when females were separated by reproductive status (Fig. B.4). Female kudu exhibited more tortuous movements than males

(*P* = 0.001; Fig. 2.8), and magnitude of the difference was more pronounced for lactating than non-lactating females (Fig. B.5).

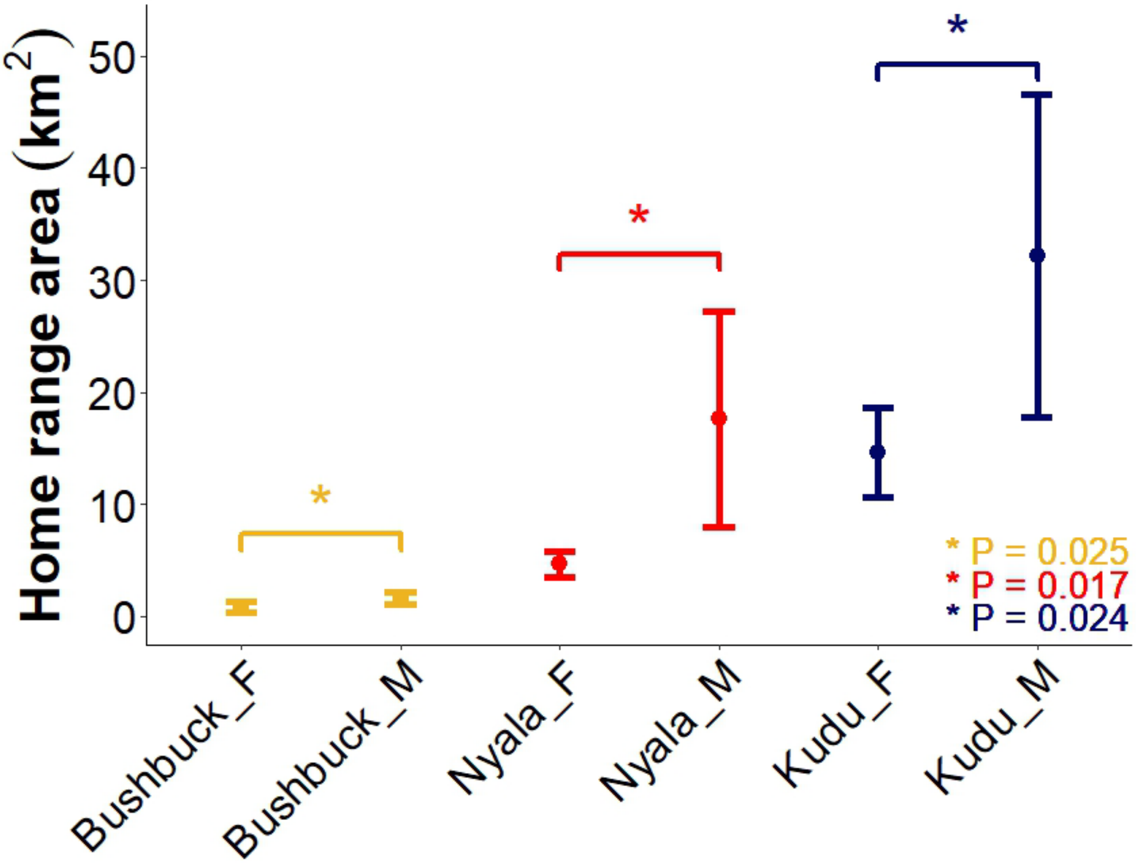

Figure 2.6. – Mean (± 95% CI) home range area (based on 95% fixed-kernel UD) of female and male bushbuck, nyala, and kudu during the dry seasons (May–October) of 2018–2019 and 2021 in Gorongosa National Park, Mozambique (bushbuck females, *n* = 24; bushbuck males, *n* = 13; nyala females, *n* = 5; nyala males, *n* = 7; kudu females, *n* = 33; kudu males, *n* = 6). Means were compared using 2-sample *t*-tests; asterisks indicate significant pairwise differences between sexes.

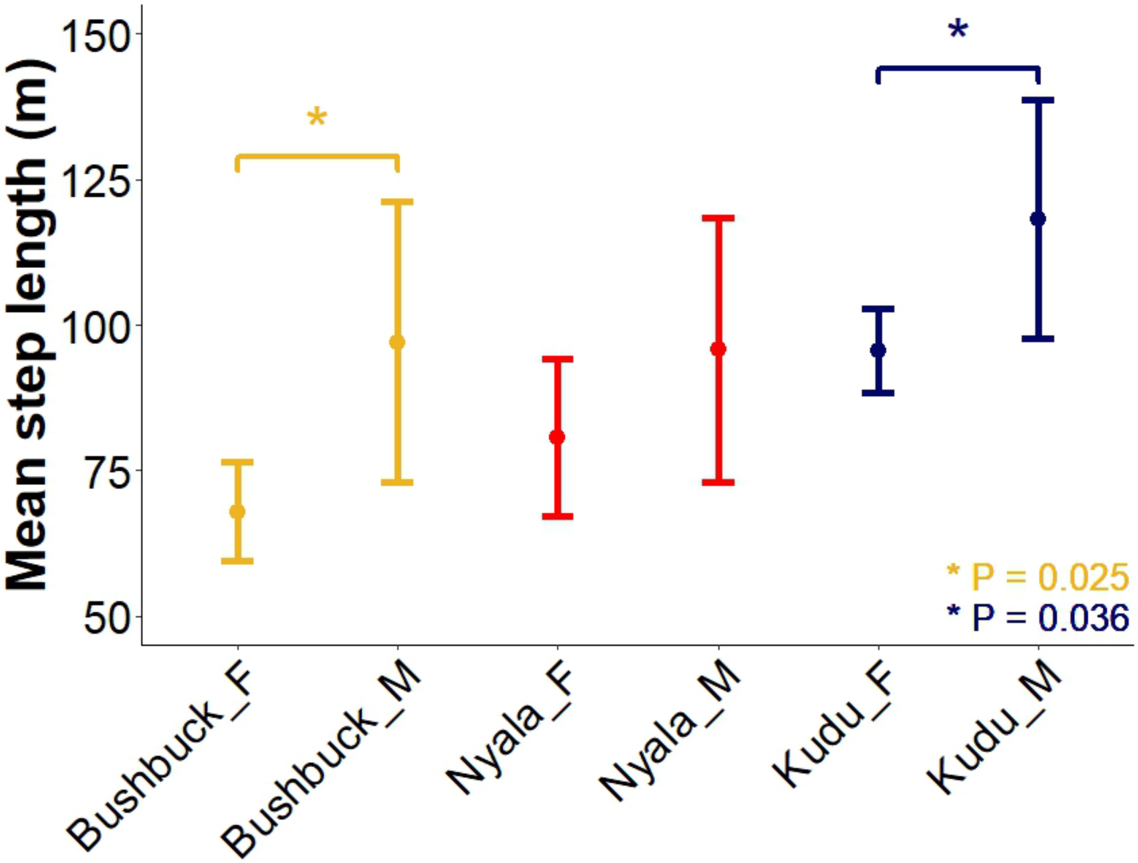

Figure 2.7. – Mean (± 95% CI) step length of female and male bushbuck, nyala, and kudu during the dry seasons (May–October) of 2018–2019 and 2021 in Gorongosa National Park, Mozambique (bushbuck females, *n* = 23; bushbuck males, *n* = 8; nyala females, *n* = 5; nyala males, *n* = 7; kudu females, *n* = 33; kudu males, *n* = 6). Means were compared using 2-sample *t*-tests; asterisks indicate significant pairwise differences between sexes.

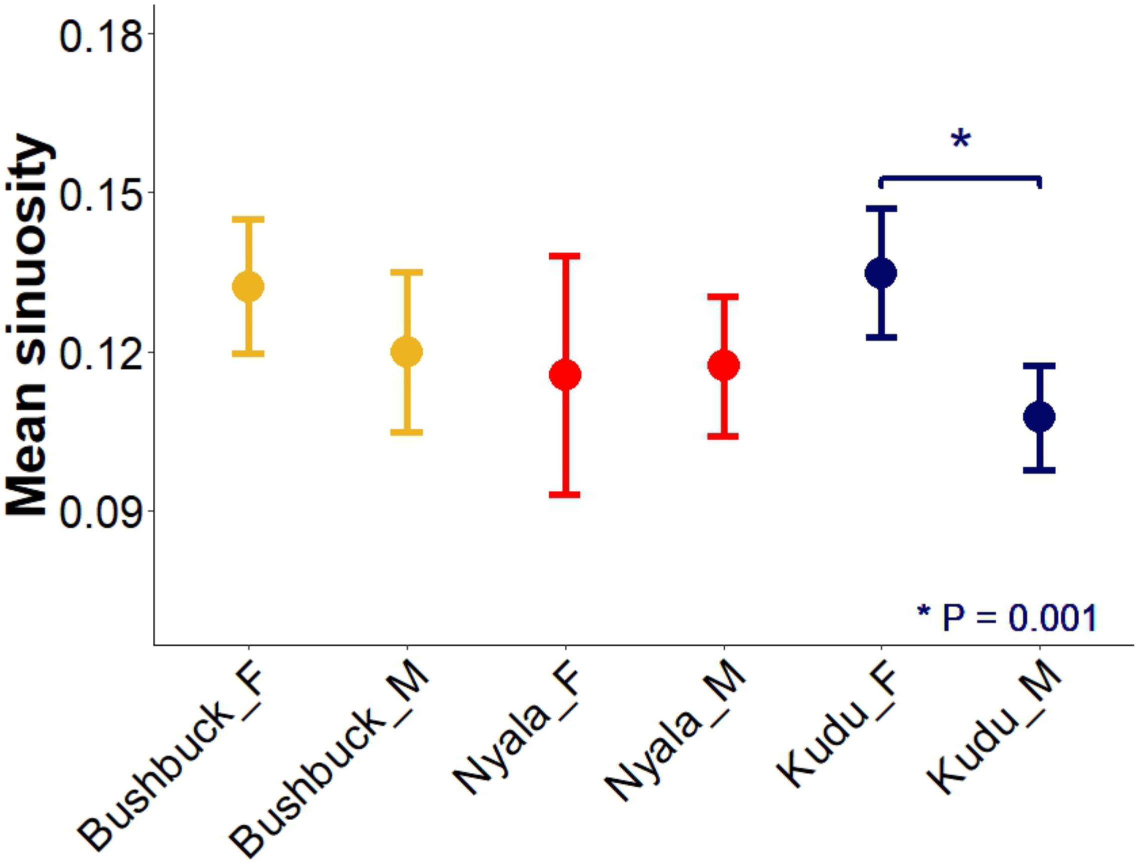

Figure 2.8. – Mean (± 95% CI) sinuosity of the movement paths of female and male bushbuck, nyala, and kudu during the dry seasons (May–October) of 2018–2019 and 2021 in Gorongosa National Park, Mozambique (bushbuck females, *n* = 24; bushbuck males, *n* = 13; nyala females, *n* = 5; nyala males, *n* = 7; kudu females, *n* = 33; kudu males, *n* = 6). Higher sinuosity values indicate more tortuous movements, potentially indicating greater selectivity while foraging (Mooring et al. 2005). Means were compared using 2-sample *t*-tests; asterisks indicate significant pairwise differences between sexes.

Discussion

The Jarman-Bell Principle (JBP; Bell 1971, Geist 1974, Jarman 1974) was originally proposed to explain the coexistence of different sized African ungulates through the spatiotemporal partitioning of food resources, and has since been extended to explain intraspecific patterns of sexual segregation (Barboza and Bowyer 2000). The principle posits that smaller-bodied mammalian herbivores require higher-quality diets because of their high mass-specific metabolic rates, whereas larger-bodied herbivores can subsist on lower-quality diets because their larger gut capacity leads to greater retention times and more complete digestion of forage (Bell 1971, Geist 1974, Jarman 1974). Barboza and Bowyer's (2000) gastrocentric model of sexual segregation is grounded in JBP, and predicts that females must consume higher-quality forage due to their smaller body size and lower digestive efficiency. In contrast, the digestive morphology and physiology of larger-bodied males is better adapted to higher volumes of lower-quality forage. More recently, however, mechanisms underpinning the inverse relationship between body size and diet quality have been debated, and increasing evidence suggests that larger herbivores might consume lower-quality diets not because of increased digestive efficiency, but simply because they consume more biomass, and high-quality forage is typically scarce (Clauss et al. 2013, Müller et al. 2013).

Although we observed marked intersexual differences in diet composition and quality, those differences were opposite of predictions from JBP; larger-bodied male antelope generally consumed higher-quality diets than females. These results are not consistent with the gastrocentric model (Barboza and Bowyer 2000) and support the hypothesis that, given the opportunity, males of at least some ungulate species are capable of consuming enough high-quality forage to meet or exceed the level of diet quality attained by females. There are several potential explanations for these results. First, larger body size confers greater mobility (a generality supported by our results; males of all three species had greater step lengths and larger home ranges), and in a heterogeneous landscape with relatively low predation risk, increased mobility could have afforded males the opportunity to find and consume high-quality forage more readily than females. One assumption of this hypothesis is that availability of high-quality forage is sufficient to preclude the need for males to shift to less nutritious but more abundant plants to meet their intake requirements. This is a plausible scenario in Gorongosa, where all Tragelaphus populations appear to be well below carrying capacity based on population growth rates and measures of individual condition (Daskin et al. 2023).

A complementary explanation relates to the differential costs of reproduction incurred by female vs. male herbivores. In addition to being less mobile in general, movements of females with offspring at heel are even more constrained, which could help explain why females appear to consume lower-quality diets. Many of the females in our study were lactating, and although we found no significant differences in movement metrics between lactating and non-lactating females, sample sizes were relatively small when we parsed females by reproductive status. In addition, lactation is the most energetically expensive life stage experienced by mammals (Urison and Buffenstein 1995, Speakman and McQueenie 1996, McNab 2002), and mature male antelope in our study experienced no analogous cost of reproduction. As a result, males were typically in better nutritional condition than females (Table B.4). Walker et al. (2023) recently reported that nutritional condition of bushbuck in Gorongosa was positively correlated with diet quality. The precise mechanisms underpinning that relationship were uncertain, but if it holds across species and sexes of Tragelaphus antelope then it would support the hypothesis that males, which were generally in better condition, consumed higher-quality diets because they could. This again contrasts with predictions of the gastrocentric model of sexual segregation, which posits that male ungulates are unable to consume high-quality diets owing to their greater digestive efficiency and the consequences that would ensue from processing highly digestible forage (e.g., rumen acidosis and bloat; Barboza and Bowyer 2000).

One surprising result of our analyses was that although diet quality differed significantly between male and female bushbuck, their use of high-quality foraging habitat (i.e., termite mounds and floodplain grassland) did not. This contrast was even more pronounced for kudu in that females exhibited more tortuous movements (potentially indicative of more selective foraging) but male diets contained more digestible energy. Recent studies have revealed that nuanced patterns of dietary resource partitioning can be an important mechanism for reducing interspecific competition and facilitating coexistence (Kartzinel et al. 2015, Pansu et al. 2022). For example, even within a feeding guild, sympatric herbivore species consume different forage plants in different proportions, underscoring how interspecific niche differences and the partitioning of forage plants can stabilize coexistence within large-herbivore communities (Kartzinel et al. 2015, Daskin et al. 2023). Our results build on this foundation and suggest that even at the intraspecific level, males and females that utilize the same habitat types may often consume different diets, reducing intraspecific competition and potentially helping to stabilize population performance.

Although our study demonstrated patterns of niche differentiation between male and female antelope, our prediction that the magnitude of differences in diet, habitat use, and movement across species would be a function of the degree of sexual size dimorphism was not supported (Fig. B.6). Instead, we documented considerable variation among species in the axes along which males and females partitioned their behaviors. The smallest species, bushbuck, showed the greatest sex-dependent difference in diet quality. Nyala showed the greatest difference in habitat use, and kudu, the largest species, showed the greatest difference in movement behavior. This variation suggests that sex-dependent niche partitioning, and ultimately sexual segregation, is influenced by a suite of ecological factors that do not all covary with sexual size dimorphism in ungulates. Such variation also highlights the potential for intraspecific behavioral variation to contribute to patterns of coexistence in large-herbivore communities, a line of inquiry that warrants further investigation.

Our study lies at the interface of theory and application and has important implications for how male and female large herbivores might be conserved and managed differently. Some previous authors (e.g., Long et al. 2009, Schroeder et al. 2010) have argued that sex-dependent differences in behavior and ecology of large herbivores are often sufficient to warrant managing them as if they were different species. Although results of our study alone do not necessarily warrant such an approach in Gorongosa or other savanna systems, they do suggest that ignoring intraspecific niche differentiation could undermine conservation or management efforts. For example, males and females may experience differing levels of interspecific competition and associated feedbacks on condition and performance, which could have important impacts on population growth rates. Furthermore, the recent reintroduction of large carnivores also has the potential to impact the sexes differently. For example, the nature and magnitude of tradeoffs between predation risk and forage or other factors could differ markedly between the sexes, and management efforts to alleviate such tradeoffs may benefit one sex more than other. Large herbivores also affect ecosystem structure and function (e.g., nutrient cycling, rates of plant succession, and diversity of other species; (Molvar et al. 1993, Bowyer et al. 1997, Kie et al. 2003, Ellis-Soto et al. 2021, Pringle et al. 2023), and a more nuanced understanding of how males and females differentially contribute to such effects could shed new light on the suite of ecosystem processes that are modulated by large herbivores.

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Chapter 3: The influence of tusklessness on foraging behavior of female African elephants

ABSTRACT

Anthropogenic disturbance in the form of human harvest can exert tremendous pressure on wildlife populations. For example, harvest of individuals with specific traits in ways that do not mimic natural predation can lead to patterns of phenotypic change even in the absence of rapid population decline. Although animals can adjust their behavior in response to human harvest pressure, whether and to what degree individuals can compensate behaviorally for harvest-induced morphological or physiological changes is not well understood. Recent work demonstrated that poaching for ivory led to an increase in tusklessness among female African elephants (*Loxodonta africana*) in Mozambique. In female elephants, tusks are believed to play an important role in foraging by helping to facilitate bark stripping, tree toppling, and other destructive behaviors that define elephants' role as a keystone species in savanna ecosystems. Accordingly, if increased frequency of tusklessness alters these behaviors, it could compromise the vital role played by elephants in maintaining ecosystem structure and function. We used GPS tracking data and remotely sensed vegetation layers to investigate whether foraging behavior differs between tusked and tuskless female elephants. We hypothesized that because tuskless females may not browse as efficiently as their tusked counterparts, they would heavily utilize grassland habitats throughout the year, whereas females with tusks would rely more heavily on woodland habitats. Tuskless females showed stronger selection for grassland habitat than tusked females, supporting our hypothesis. However, we also found that both tusk morphs selected woodland habitats more strongly than grassland regardless of season. Our results suggest that intensive poaching for ivory could have consequences for savanna ecosystems that are mediated by elephant behavior, and that additional work on the strength and mechanisms underpinning such effects is warranted.

INTRODUCTION

Human harvest, legal or otherwise, can exert tremendous pressure on fish and wildlife populations (Sih 2013). For example, the crash of the northern Atlantic cod (*Gardus morhua*) population in Newfoundland and Labrador, Canada in 1992 was attributed to overexploitation (Hutchings and Myers 1994). Likewise, in less than a century, market hunting led to near extinction of the American bison (*Bison bison*) throughout its entire range (Bolger et al. 2008). The effects of human harvest are not always so dramatic, however, and over time, selective harvest of individuals with discernible traits (e.g., large horns or tusks) can lead to patterns of phenotypic change even in the absence of rapid population decline (Allendorf et al. 2008, Allendorf and Hard 2009, Darimont et al. 2009, Dirzo et al. 2014). For example, trophy hunting contributed to a decrease in horn size of bighorn sheep (*Ovis canadensis*; Coltman et al. 2003, Pigeon et al. 2016) in Alberta, Canada, and poaching for ivory led to an increase in tusklessness among female African elephants in Mozambique (*Loxodonta africana*; Campbell-Staton et al. 2021).

Animals can adjust their behavior in response to anthropogenic disturbance (Crick et al. 1997, Walther et al. 2002), including harvest pressure (Lone et al. 2015, Ihwagi et al. 2019), and behavioral plasticity often buffers animals against environmental variation (Huey et al. 2003). Yet, whether and to what degree individuals can compensate behaviorally for harvest-induced morphological or physiological changes is not well understood. African elephants are a keystone species in savanna habitats, and their behaviors can have dramatic impacts on whole ecosystems (Midgley et al. 2005, Daskin et al. 2016). Elephants spend approximately 75% of the day foraging (Wyatt and Eltringham 1974) and act as ecosystem engineers through their destructive foraging behavior (Haynes 2012, Coverdale et al. 2016). The toppling and crushing of trees and shrubs makes previously inaccessible forage and concealment cover available to numerous smaller species (Pringle 2008, Pringle et al. 2015, Daskin et al. 2016). Although the precise function of tusks in female elephants is uncertain, they are believed to play an important function in elephant foraging behavior. Female elephants have been observed using their tusks for various destructive behaviors, including stripping bark from trees and digging for water and minerals (Whitehouse 2002, Midgley et al. 2005). Thus, if the poaching of elephants for ivory leads to a phenotypic shift toward tusklessness, it could compromise behaviors that define elephants' keystone role and have cascading impacts on the structure and function of savanna ecosystems.

Elephants are generalist foragers with diverse diets and are considerably larger than most other species. As a result, elephants have a much greater requirement for forage biomass than other species. Indeed, recent work reported that elephants foraging in natural landscapes closely tracked precipitation-driven patterns of plant phenology and consistently selected plants that were at or near peak greenness (i.e., peak biomass; Bohrer et al. 2014; Branco et al. 2019). In seasonal African savannas, grasses comprise a large proportion of the green biomass during the wet season, whereas most green forage during the dry season is woody. This raises the question of whether tuskless elephants, which may browse less efficiently, track spatiotemporal changes in peak forage biomass to the same degree as their tusked counterparts, and if not, what alternative behavioral strategies they may use for acquiring sufficient nutrients when grasses senesce during the dry season.

Home range size scales positively with body size (Ofstad et al. 2016, Daskin et al. 2023), whereas predation risk scales negatively (Sinclair et al. 2003, Hopcraft et al. 2010). These relationships afford elephants the ability to search for habitats that provide high forage biomass with little concern for predators (Bohrer et al. 2014, Gaynor et al. 2018, Branco et al. 2019). Moreover, elephants are highly intelligent and have the ability to remember the locations of resources across large spatiotemporal scales (Byrne et al. 2009). These characteristics make African elephants an ideal species for investigating the effects of phenotypic change on behavior. More specifically, Mozambique’s Gorongosa National Park is an ideal system for assessing the effects of harvest-induced tusklessness on seasonal changes in foraging behavior of female African elephants. From 1968–1972 the elephant population in Gorongosa was estimated at roughly 1,900 individuals (Tinley 1977). However, during Mozambique's civil war (1977–1992), the poaching of elephants for meat and ivory resulted in a dramatic decline in the population (Stalmans et al. 2019, Campbell-Staton et al. 2021). Indeed, only 163 elephants were observed in the first aerial wildlife survey conducted after the war (Stalmans 2012). In subsequent decades, restoration efforts have been largely successful, and 620 elephants were counted during the 2022 aerial survey (Stalmans et al. 2022). Importantly however, frequency of the tuskless phenotype has also increased in the population. Across their range, 2–4% of female African elephants are tuskless, but in Gorongosa approximately 50% of female elephants that survived the war were tuskless, and 33% of their daughters also lacked tusks (Campbell-Staton et al. 2021). This sharp rise in tusklessness among female elephants in Gorongosa, coupled with strong seasonal changes in the availability of key forage resources (especially grasses), presents a unique opportunity to understand how human harvest may influence the foraging behavior of this iconic species.

We investigated whether foraging behavior differs between tusked and tuskless female elephants (Fig. 3.1). We hypothesized that because they lack the morphological structures (i.e., tusks) to browse as efficiently as their tusked counterparts, tuskless females heavily utilize grassland habitats throughout the year, and thus do not exhibit a strong response to seasonal changes in grass biomass (i.e., grassland greenness). In contrast, we hypothesized that females with tusks consistently select areas of peak forage biomass throughout the year, utilizing grasslands more heavily during the wet season and woody habitats more heavily during the dry season.

Figure 3.1.– Tuskless (left) and tusked (right) female elephants in Gorongosa National Park, Mozambique. Photo used with permission from Miguel Lajas.

MATERIALS AND METHODS

Study area.–Mozambique's Gorongosa National Park is situated at the southern end of the Great Rift Valley (18.96°S, 34.36°E) and supports a diverse array of large mammals, with the highest densities occurring within the Rift Valley portion of the park. This 40-km expanse of the valley floor slices through the center of the park and includes Lake Urema and its expansive floodplain grassland, as well as *Acacia*, palm, and broadleaf savanna woodland habitats (Pansu et al. 2019, Stalmans et al. 2019). There are two primary seasons in Gorongosa: the wet season, which occurs from November to April, and the dry season, which occurs from May to October (Fig. 3.2). Annually the park receives 700–900 mm of rainfall, with peak precipitation occurring from December to February (Tinley 1977). Monthly precipitation during the dry season averages <50 mm, and both water and high-quality forage become increasingly scarce as the dry season progresses.

Rampant poaching during the Mozambican Civil War (1977-1992) decimated wildlife populations in Gorongosa, reducing the Park's wildlife biomass by >90% (Stalmans et al. 2019). In the ensuing decades, researchers sought to restore the Park's large mammal community and to understand the cascading effects of its near extirpation. Restoration efforts have been largely successful, and many large herbivore populations have rebounded to pre-war levels (Stalmans et al. 2022). Although the elephant population has not yet reached half of its pre-war size, numbers have been steadily increasing for more than a decade, and this trend appears likely to continue.

Animal capture and handling.–In July 2018, we chemically immobilized 6 female elephants (3 with tusks, 3 without; paired sampling across 3 separate family groups) via remote injection with a combination of thiafentanil oxalate (9–15 mg) and azaperone (40–60 mg), with the dosage based on the approximate age and size of the individual. Elephants were carefully monitored during handling, and the following parameters were measured at 5-min intervals: cardiac rate (normal: 25–30 bpm), respiratory rate (normal: 4–6 breaths/minute), and rectal temperature (normal: 36–37°C). When handling was complete, thiafentanil was reversed with naltrexone and we observed each elephant from a safe distance until it regained footing and walked away. All animal handling procedures were approved by the Animal Care and Use Committee at the University of Idaho (protocol #2015- 39) and

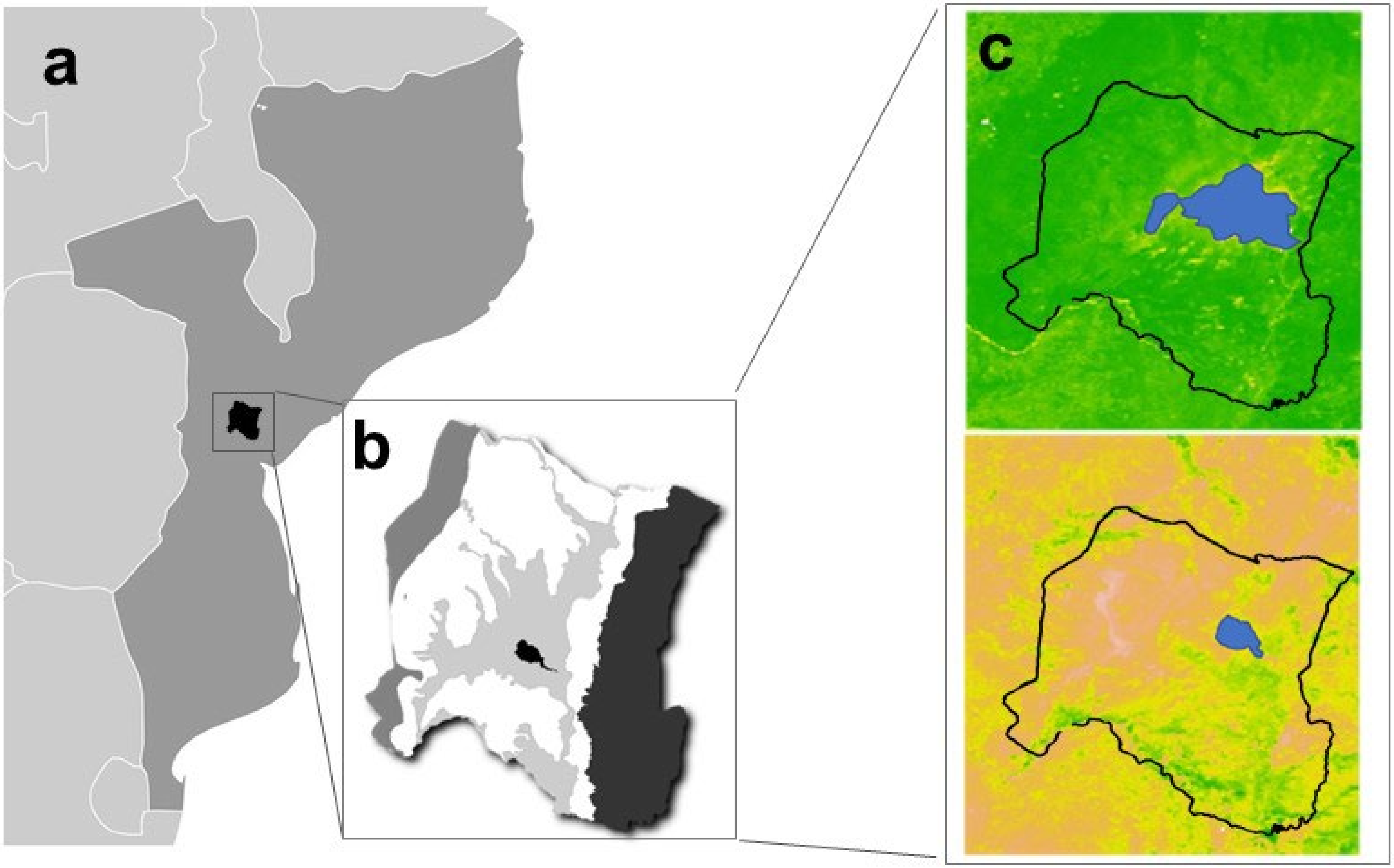

Figure 3.2.– (a) Gorongosa National Park is located in central Mozambique and (b) consists of four major habitat zones (from left: western escarpment [medium gray], woodland [white], floodplain [light gray], and eastern escarpment [dark gray], as well as Lake Urema [black]). (c) Normalized difference vegetation index (NDVI) images of Gorongosa during the wet season (top) and the dry season (bottom). NDVI quantifies ‘greenness’ and is widely used as a proxy for vegetation phenology and standing plant biomass. Lake Urema is shown in blue.

were in accordance with guidelines established by the American Society of Mammalogists (Sikes and The Care and Use Committee 2016).

We equipped each elephant with an iridium satellite global positioning system (GPS) collar (Savannah model, Savannah Tracking Ltd, Kilifi, Kenya; weight = 14 kg) in July 2018. Collars were programmed to transmit fixes every 15 min through the iridium satellite system. Two collars stopped transmitting fixes in July (tuskless) and October (tusked) 2020, respectively, whereas the four remaining collars transmitted as scheduled for the duration of the collar's battery life (i.e., until August 2021).

Landscape greenness.–To track green-up and brown-down of the grassland and woodland across seasons, we calculated mean values of the Normalized Difference Vegetation Index (NDVI) using surface reflectance bands of the MODIS terra satellite (product MODIStsp, version 2.0.6.9002, resolution 250 m, every 16 days) from July 2018 through August 2021. We limited our analysis to grassland or woodland pixels that were within the portion of the park used by GPS-collared elephants during our study (based on 100% minimum convex polygon [MCP] calculated from all elephant locations). NDVI quantifies the 'greenness' of each pixel in a landscape and is widely used as a proxy for vegetation phenology (Bischof et al. 2012, Aikens et al. 2017, Branco et al. 2019), net primary production (Pettorelli et al. 2005), and standing plant biomass (Dancose et al. 2011). We set to 'no data' pixels that were categorized as snow/ice or cloud (3% of total pixels) by the classification algorithm. We used the Classes and Methods for Spatial Data (*sp*) package (Pebesma et al. 2021) in R to calculate mean NDVI of both habitat types separately every 16 days.

Grassland selection and space-use analysis.–We quantified resource selection by tusked and tuskless female elephants using resource selection functions (RSFs) and a use-availability design (Boyce et al. 2002, Johnson et al. 2006). We delimited available habitat based on the 100% MCP derived from all elephant locations. Using the methods of Long et al. (2014) we determined that 7,500 random locations was sufficient to accurately represent habitat availability within the elephant MCP, and we generated that number of random points (coded 0 for analysis) for each unique combination of individual and month (n = 199 individual-months) during the study period; we stratified GPS locations (coded 1 for analysis) similarly across individuals and months. All locations, used and random, were then spatially joined to the underlying habitat layers (grassland versus non-grassland, and NDVI). NDVI is updated every 16 days, therefore we used the *raster calculator* function in ArcGIS 10.8.1 to calculate monthly average NDVI values. We then extracted these monthly values at each used and random location.

For each tusk morph, we fit separate models to the full dataset and to the data from each month (*n* = 37 months). We fit generalized linear mixed models (Gillies et al. 2006, Bolker et al. 2009, Zuur et al. 2009) with a binomial error distribution and logit link function to the used and random locations for (1) tuskless females and (2) tusked females using the glmer function in the *lme4* package in R (Bates et al. 2022). Each model included a random intercept and a random slope, grouped by individual, and habitat type and NDVI (standardized by subtracting the mean and dividing by the *SD*; Kutner et al. 2004, Cade 2015) were included as fixed effects. Marginal coefficients represented the relative probability of use of grassland relative to non-grassland habitat, and the change in relative probability of use as a function of habitat greenness (NDVI). Accordingly, we used these coefficients as the response variables in two ANOVAs to evaluate the influence of season and tusk morph on resource selection by female elephants. We included season (dry or wet), tusk morphology (tuskless or tusked), and the corresponding 2-way interaction as independent variables. We also weighted the analyses by the inverse of the *SE* associated with each GLMM coefficient to account for uncertainty in estimates of those coefficients.

To quantify use of grassland habitat by elephants independent of availability (i.e., use rather than selection; Millspaugh et al. 2006) and as a function of NDVI, we estimated 95% fixed-kernel utilization distributions (UDs) every 16 days for each individual using the *adehabitatHR* package (Calenge 2019) in R, and calculated the proportion of the volume of each UD that overlapped grassland and woodland habitats. We then used ANOVA with season, tusk morph, and their interaction to determine whether UD overlap with grassland or woodland habitats differed between females with and without tusks, and if so whether that difference varied seasonally. We evaluated the influence of greenness (NDVI) on grassland and woodland use by tusked and tuskless elephants using linear mixed-effects model that included NDVI, tusk morph, and their interaction as fixed effects; we included a random intercept and a random slope for NDVI, grouped by individual animal.

RESULTS

Tuskless female elephants showed strong and consistent selection for grassland habitat throughout the study period ($\beta = 0.33$, $P < 0.001$), whereas their tusked counterparts did not ($\beta = 0.18$, $P = 0.17$). In contrast, both tusked and tuskless elephants showed strong selection for NDVI (and thus, ostensibly, for forage biomass) throughout the study (tusked: $\beta = 0.36$, $P < 0.001$; tuskless: $\beta = 0.32$, $P < 0.001$). During the dry season, NDVI values were comparable between the grassland and woodland. During the wet season, however, mean NDVI was markedly higher in the woodland (Fig. 3.3), indicating that woodland habitats likely provided greater forage biomass during the wet season. Monthly models of habitat selection produced similar results. Tuskless female elephants showed stronger selection for grassland habitat (based on monthly GLMM coefficients) than tusked females throughout the

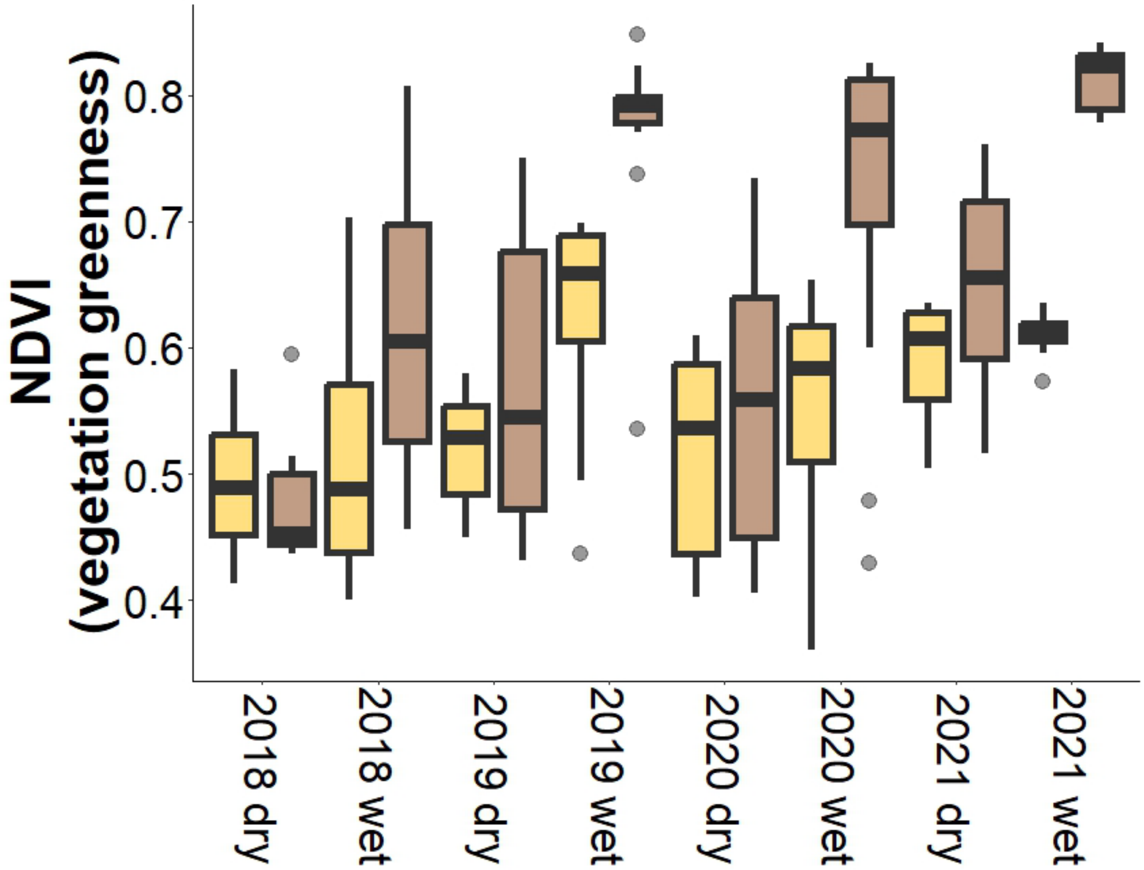

Figure 3.3.– Mean NDVI of grassland (yellow) and woodland (brown) habitats from July 2018 until August 2021 in Gorongosa National Park, Mozambique. Boxplots show median (central bar) and quartiles; whiskers show ± 1.5 × interquartile range; dots are outliers. Dry season months: May–October; wet season months: November–April.

study period (tusk morph: $F_{1,70} = 5.06$, $P = 0.028$; Fig. 3.4), and both tusk morphs showed stronger selection for grasslands during the wet season than the dry season (season: $F_{1,70} = 6.94$, $P = 0.01$; Fig. 3.4); the tusk morph × season interaction was not significant (season × tusk morph: $F_{1,70} = 0.204$, $P = 0.653$; Fig. 3.4). Tusked and tuskless elephants did not differ in their selection for vegetation greenness (based on monthly GLMM coefficients; tusk morph: $F_{1,70} = 0.59$, $P = 0.45$; season: $F_{1,70} = 1$, $P = 0.321$; season × tusk morph: $F_{1,70} = 1.72$, $P = 0.194$; Fig 3.4).

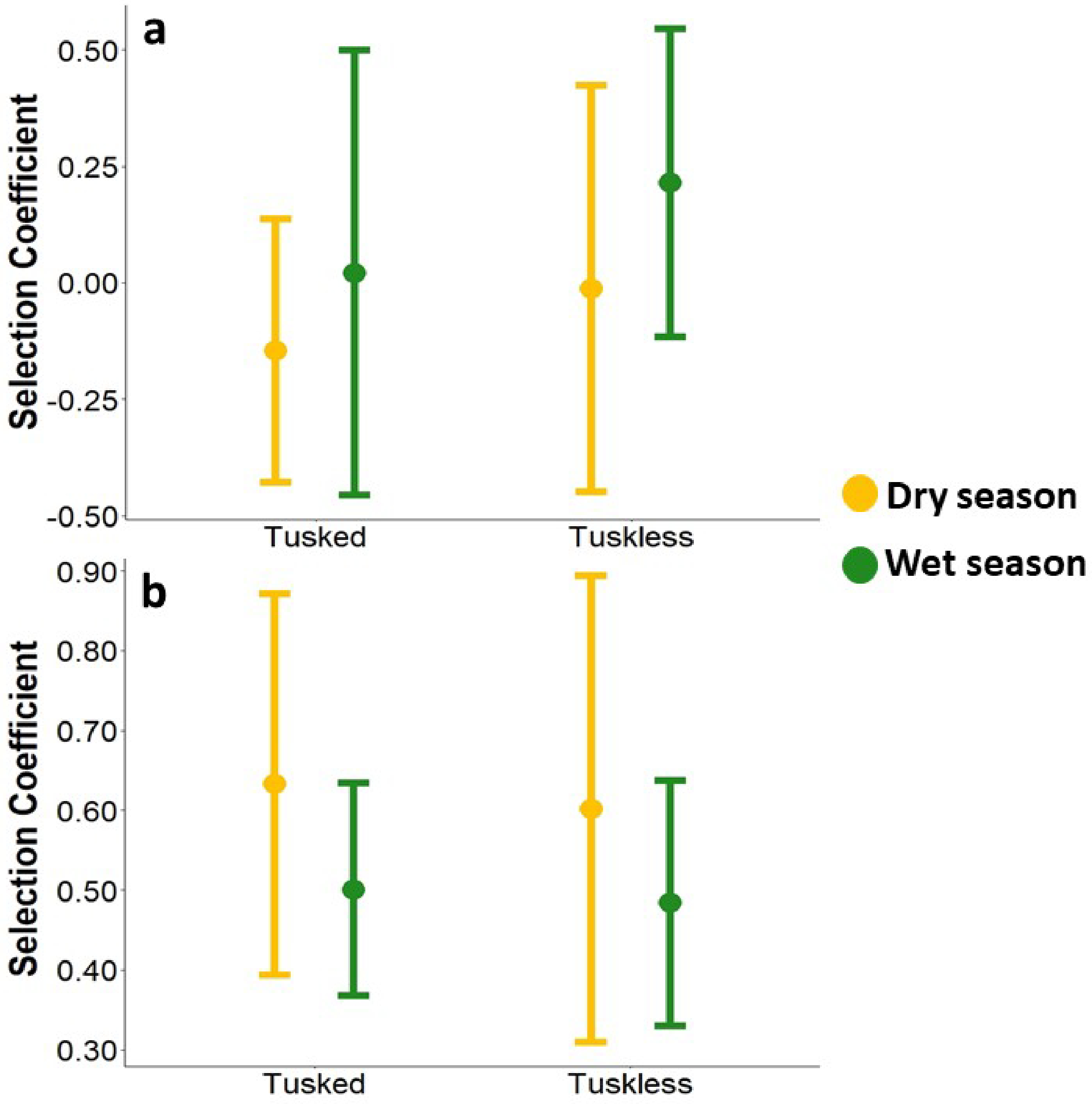

Figure 3.4.–Mean (± 95% CI) selection (based on standardized GLMM coefficients) of (a) grassland habitat and (b) vegetation greenness (NDVI) by tusked and tuskless female elephants during the dry season (yellow) and wet season (green) from July 2018 to August 2021 in Gorongosa National Park, Mozambique. Dry season months: May–October; wet

season months: November–April. Means were compared using ANOVA (Habitat: tusk morph, $F_{1,70} = 5.06$, $P = 0.028$; season, $F_{1,70} = 6.94$, $P = 0.01$; season × tusk morph, $F_{1,70} = 0.204$, $P = 0.653$; NDVI: tusk morph, $F_{1,70} = 0.59$, $P = 0.45$; season, $F_{1,70} = 1$, $P = 0.321$; season × tusk morph, $F_{1,70} = 1.72$, $P = 0.194$).

Analyses of grassland use based on UD overlap produced similar results to analyses of selection. Tuskless female elephants used grassland habitat significantly more than tusked females across seasons (tusk morph: $F_{1,365} = 7.245$, $P = 0.007$; Fig 3.5), and both tusk morphs used grassland more during the dry season than the wet season ($F_{1,365} = 3.923$, $P = 0.048$; Fig. 3.5). Use of woodland habitat did not differ between tusk morphs or seasons (tusk morph: $F_{1,365} = 1.288$, $P = 0.257$; season: $F_{1,365} = 2.1$, $P = 0.148$; season × tusk morph: $F_{1,365} = 0.943$, $P = 0.332$; Fig. 3.5). Use of grassland habitat also declined as a function of greenness for both tusk morphs ($\beta_{\mathrm{NDVI}} = -0.334$, $P = 0.004$; Fig. 3.6), whereas woodland use did not change as a function of greenness ($\beta_{\mathrm{NDVI}} = 0.071$, $P = 0.61$; Fig. 3.6).

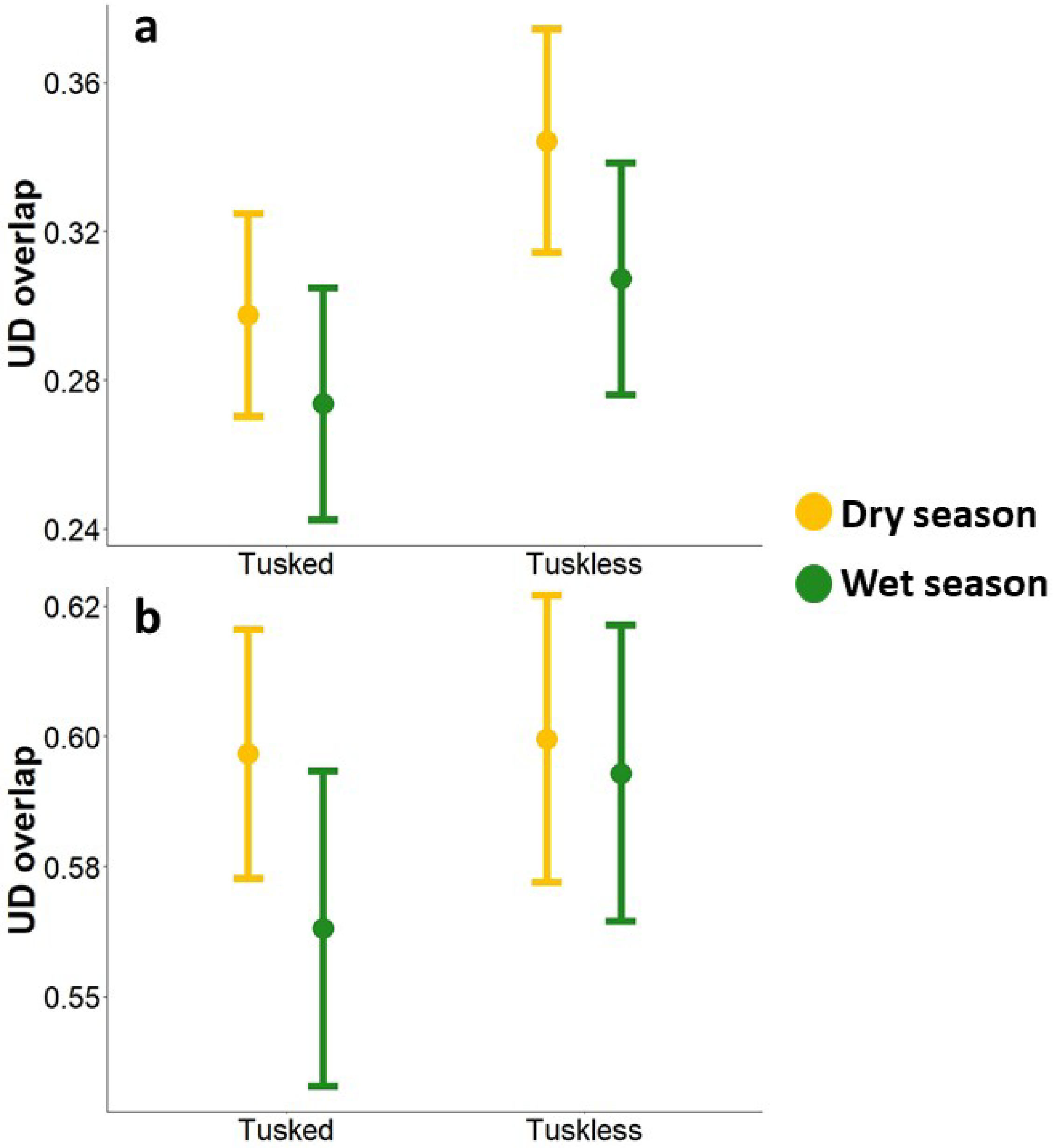

Figure 3.5.–Mean (± 95% CI) overlap (volumetric) of elephant utilization distributions (UD) with (a) grassland and (b) woodland habitats during the dry (yellow) and wet (green) seasons of July 2018 to August 2021 in Gorongosa National Park, Mozambique. We estimated 95% fixed-kernel utilization distributions (UDs) every 16 days for each collared individual. Dry season months: May–October; wet season months: November–April. Means were compared using ANOVA (Grassland: tusk morph, $F_{1,365} = 7.245$, $P = 0.007$; season, $F_{1,365} = 3.923$, $P = 0.048$; season × tusk morph, $F_{1,70} = 0.193$, $P = 0.661$; Woodland: tusk morph, $F_{1,365} = 1.288$, $P = 0.257$; season, $F_{1,365} = 2.1$, $P = 0.148$; season × tusk morph, $F_{1,365} = 0.943$, $P = 0.332$).

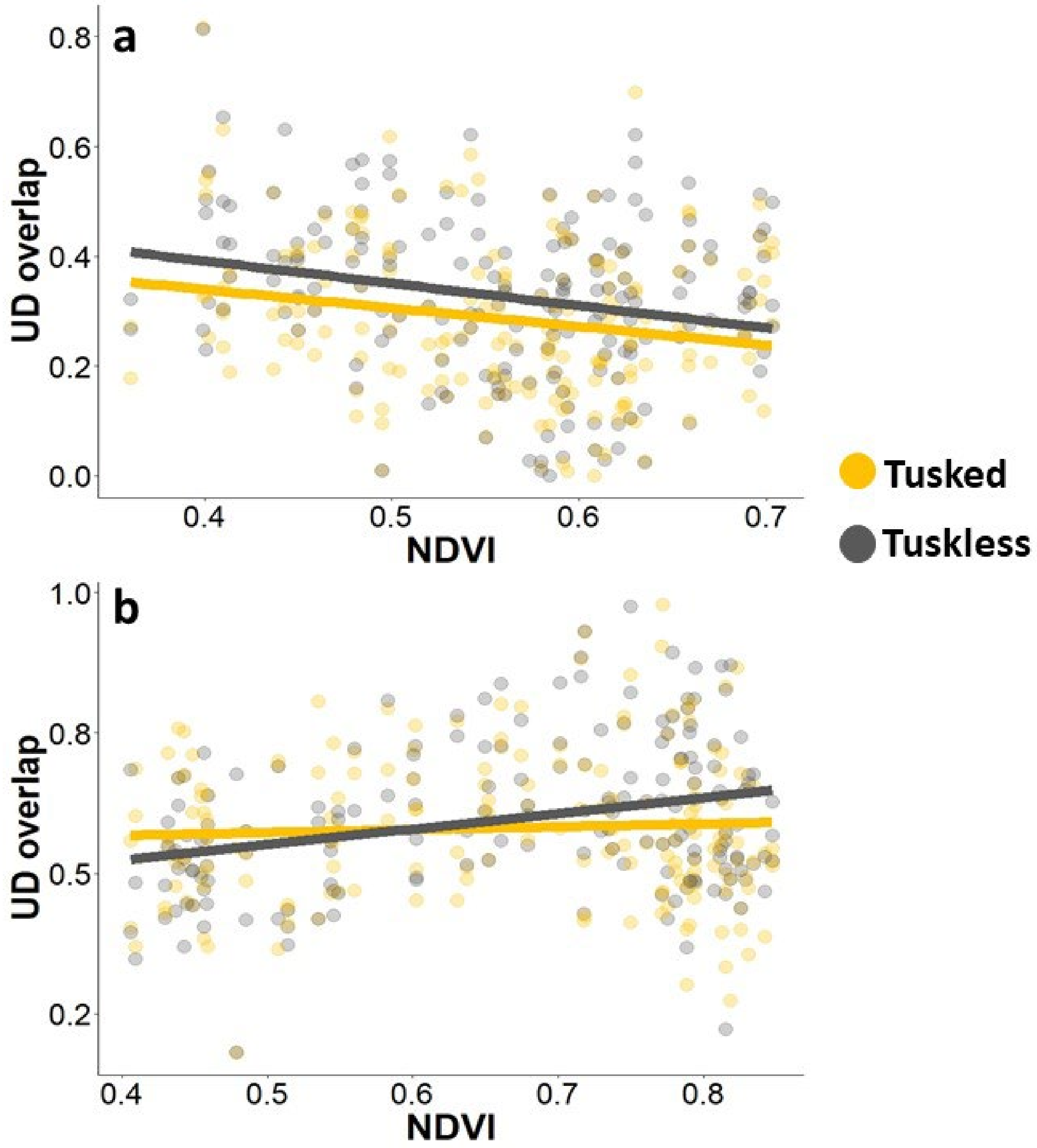

Figure 3.6.– Relationship between NDVI (i.e., greenness) of (a) grassland and (b) woodland habitats and use of those habitats (quantified based on volumetric overlap of elephant UDs) by tusked (yellow) and tuskless (gray) female elephants from July 2018 to August 2021 in Gorongosa National Park, Mozambique. Both tusk morphs used grassland habitat less as it became greener (NDVI: $\beta = -0.334$, $P = 0.004$), and there was no difference in this relationship between the two tusk morphs (NDVI × tusk morph: $\beta = -0.081$, $P = 0.625$). Use of woodland habitat by elephants did not change as a function of NDVI (NDVI: $\beta = 0.071$, $P = 0.61$), and there was no difference in this relationship between the two tusk morphs (NDVI × tusk morph: $\beta = 0.217$, $P = 0.312$).

DISCUSSION

We found support for our predictions that tuskless female elephants would use grassland habitat more than tusked females, and that tuskless females would not adjust their selection of grasslands seasonally. Our results demonstrate that the rapid evolution of tusklessness in response to intense poaching pressure (Campbell-Staton et al. 2021) likely has behavioral consequences for elephants. We did not link habitat selection to diet composition, richness or quality in this study, and a more complete understanding of behaviorally mediated effects of tusklessness on individual and population performance of elephants will require this step. Nevertheless, given that grass is typically less nutritious than browse (especially during the dry season; Wilson 1969, Owen-Smith 1982, Gagnon and Chew 2000), if tuskless female elephants consume more grass and less woody forage because they are unable to browse efficiently, it could have important impacts on nutrient intake. Although elephants are bulk feeders that are more dependent on forage biomass than quality, small differences in the ratio of energy intake to expenditure can have ‘multiplier’ effects (White 1983, Cook et al. 2004) on fitness parameters such as body mass and condition, and thus shifts in diet induced by tusklessness may still have nontrivial impacts on performance.

Our prediction that selection of grasslands by tusked females would be strongest during the wet season was also supported, and indeed, grassland selection did not change seasonally for either tusk morph. Moreover, selection coefficients for grassland habitat indicated that although tuskless females used grasslands to a greater degree than females with tusks, both tusk morphs preferred woodland habitats (Fig. 3.4). This result is consistent with previous studies (i.e., Loarie et al. 2009, Okello et al. 2015), and makes sense in light of our comparison of NDVI values between grassland and woodland habitats, which showed that woodland habitats in Gorongosa providing greater forage biomass than grasslands, especially during the wet season.

As the largest extant terrestrial mammal, African elephants rely on large home ranges and a bulk, generalist diet to meet their nutritional requirements (Hempson et al. 2015, Kartzinel et al. 2015, Branco et al. 2019). Accordingly, elephants are likely to forage in habitats that provide the greatest amount of forage biomass throughout the year, and elephants are known to track peak NDVI across landscapes (Bohrer et al. 2014, Branco et al. 2019, Tsalyuk et al. 2019). Although our hypotheses were centered more on selection of grassland habitat, savanna woodlands also experience seasonal cycles of green-up and brown-down. In Gorongosa, greenness was comparable between grassland and woodland habitats during the dry season, but during the wet season woodlands experienced a much greater increase in greenness than grasslands. Accordingly, elephants selected woodlands even more strongly during the wet season despite the flush of green biomass that occurred simultaneously in grassland habitat. The result explains the negative relationship between grassland greenness (NDVI) and grassland selection by elephants; when grasslands were green, woodlands were greener.

As engineers of the African savanna, elephant foraging decisions can modify landscapes and affect the structure and function of savanna ecosystems. Although both tusked and tuskless elephants in our study consistently used woodland habits, tuskless females used grasslands more heavily than their tusked counterparts. This suggests that poaching-induced changes in the frequency of tusklessness could have ecosystem-level consequences that are behaviorally mediated. For example, elephants are important seed dispersers (Campos-Arceiz and Blake 2011, Bunney et al. 2017), and thus if tusklessness alters the distribution of foraging elephants on the landscape, it could affect the distribution of plant species that elephants consume. In addition, if tuskless elephants are less prone to engage in tree toppling and other destructive foraging behaviors, then the role they play in providing food and cover for many smaller species could diminish. Our work underscores the importance of understanding the effects of human harvest not only on the evolution of secondary sexual characteristics and other morphological traits, but also on animal behavior, especially in keystone species whose behavioral choices can have cascading effects on community and ecosystem process.

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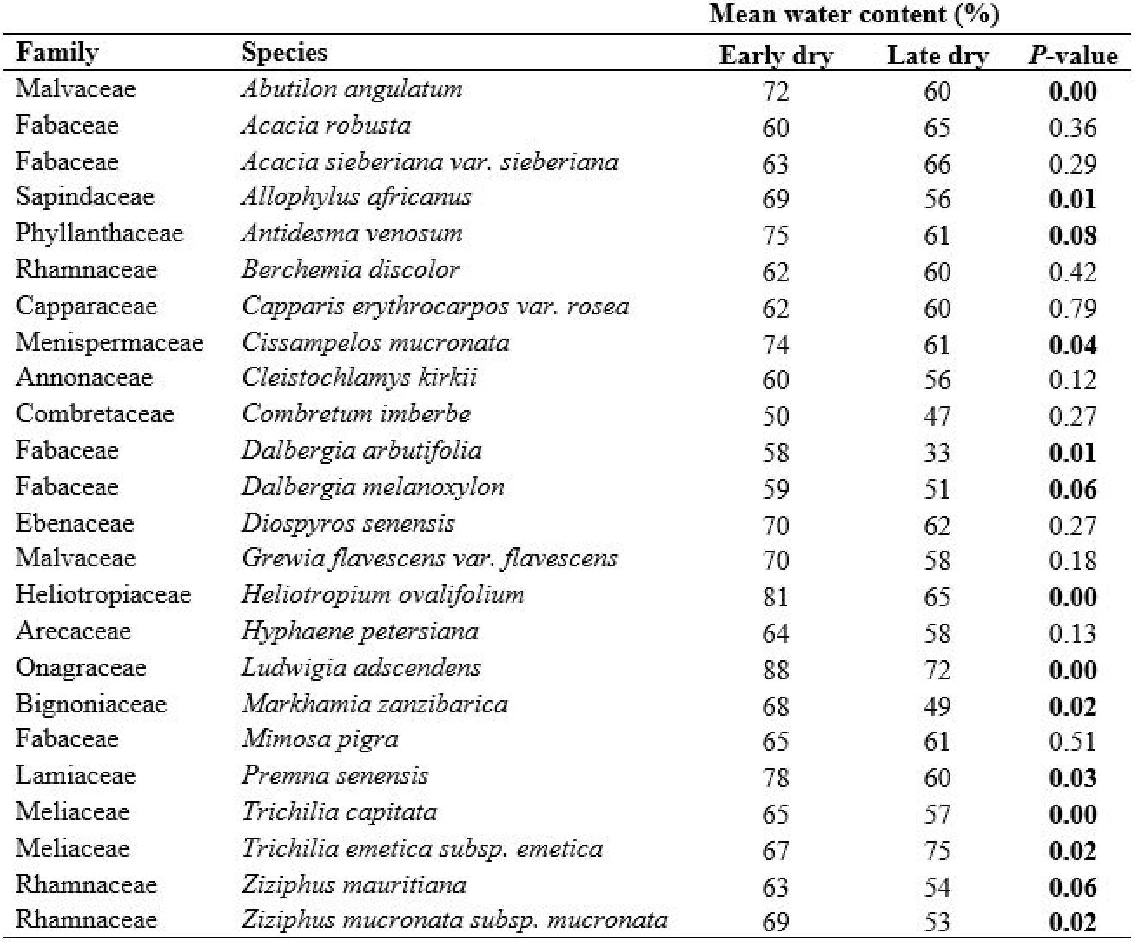
Appendix A: Supplementary materials for “Chapter 1: Decomposing water requirements of savanna ungulates: seasonal and interspecific differences in plant- vs. surface-water intake by browsing antelopes”

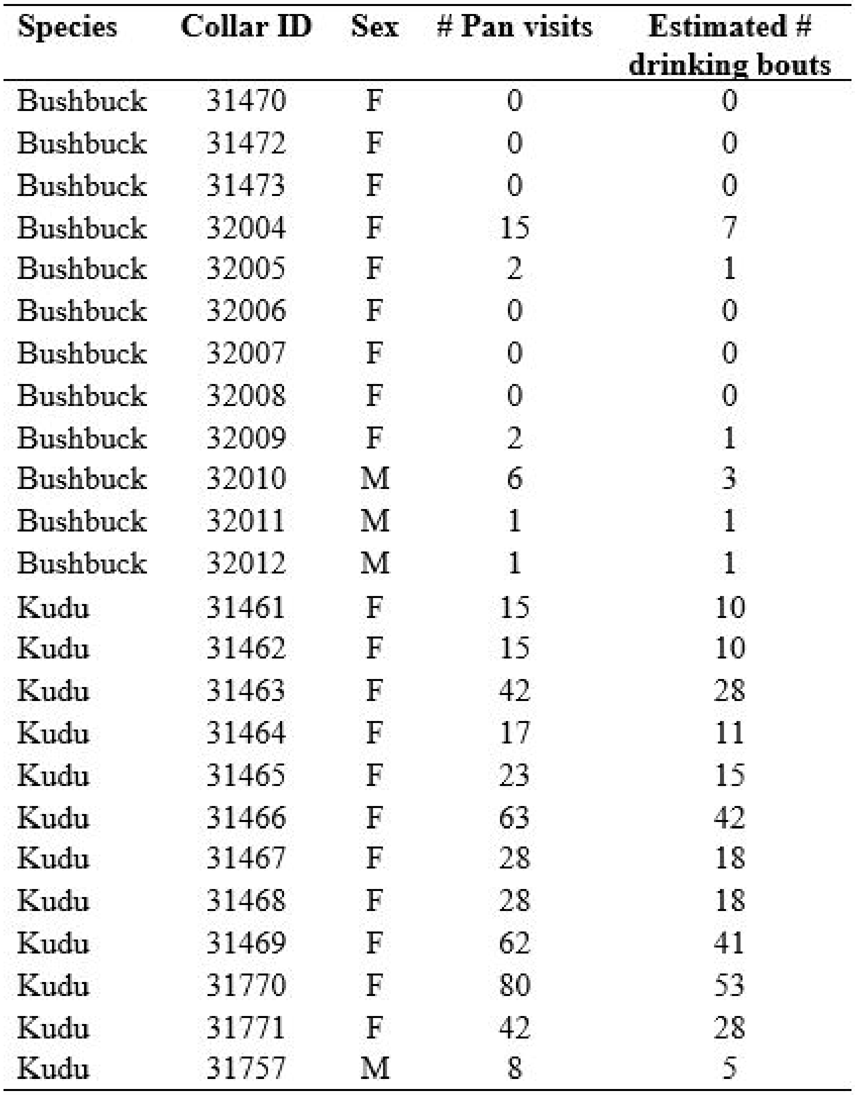
SUPPLEMENTARY TABLES

Table A.1. – Mean preformed water content (percentage) of plant species consumed by bushbuck and/or kudu during the dry season in Gorongosa National Park, Mozambique. Tukey HSD *P*-values signify the effect of season (early versus late) on preformed water content of each species; bold font indicates statistical significance at the α = 0.10 level.



		Mean water content (%)		
Family	**Species**	**Early dry**	**Late dry**	***P*-value**
Malvaceae	*Abutilon angulatum*	72	60	**0.00**
Fabaceae	*Acacia robusta*	60	65	0.36
Fabaceae	*Acacia sieberiana var. sieberiana*	63	66	0.29
Sapindaceae	*Allophylus africanus*	69	56	**0.01**
Phyllanthaceae	*Antidesma venosum*	75	61	**0.08**
Rhamnaceae	*Berchemia discolor*	62	60	0.42
Capparaceae	*Capparis erythrocarpos var. rosea*	62	60	0.79
Menispermaceae	*Cissampelos mucronata*	74	61	**0.04**
Annonaceae	*Cleistochlamys kirkii*	60	56	0.12
Combretaceae	*Combretum imberbe*	50	47	0.27
Fabaceae	*Dalbergia arbutifolia*	58	33	**0.01**
Fabaceae	*Dalbergia melanoxylon*	59	51	**0.06**
Ebenaceae	*Diospyros senensis*	70	62	0.27
Malvaceae	*Grewia flavescens var. flavescens*	70	58	0.18
Heliotropiaceae	*Heliotropium ovalifolium*	81	65	**0.00**
Arecaceae	*Hyphaene petersiana*	64	58	0.13
Onagraceae	*Ludwigia adscendens*	88	72	**0.00**
Bignoniaceae	*Markhamia zanzibarica*	68	49	**0.02**
Fabaceae	*Mimosa pigra*	65	61	0.51
Lamiaceae	*Premna senensis*	78	60	**0.03**
Meliaceae	*Trichilia capitata*	65	57	**0.00**
Meliaceae	*Trichilia emetica subsp. emetica*	67	75	**0.02**
Rhamnaceae	*Ziziphus mauritiana*	63	54	**0.06**
Rhamnaceae	*Ziziphus mucronata subsp. mucronata*	69	53	**0.02**

Table A.2. – Number of visits to perennial pans and estimated number of drinking bouts by bushbuck (*n* = 12) and kudu (*n* = 12) in Gorongosa National Park, Mozambique from 13 July – 31 August 2021. A pan visitation was counted when a GPS-collared individual was within 33 m of a pan (20 m mean pan radius + 13 m collar error). The number of drinking bouts was then estimated by correcting the number of pan visits by the proportion of pan visits recorded by trail cameras during which animals of each species consumed water (49% for bushbuck, 67% for kudu).



Species	Collar ID	Sex	# Pan visits	Estimated # drinking bouts
Bushbuck	31470	F	0	0
Bushbuck	31472	F	0	0
Bushbuck	31473	F	0	0
Bushbuck	32004	F	15	7
Bushbuck	32005	F	2	1
Bushbuck	32006	F	0	0
Bushbuck	32007	F	0	0
Bushbuck	32008	F	0	0
Bushbuck	32009	F	2	1
Bushbuck	32010	M	6	3
Bushbuck	32011	M	1	1
Bushbuck	32012	M	1	1
Kudu	31461	F	15	10
Kudu	31462	F	15	10
Kudu	31463	F	42	28
Kudu	31464	F	17	11
Kudu	31465	F	23	15
Kudu	31466	F	63	42
Kudu	31467	F	28	18
Kudu	31468	F	28	18
Kudu	31469	F	62	41
Kudu	31770	F	80	53
Kudu	31771	F	42	28
Kudu	31757	M	8	5

SUPPLEMENTARY FIGURES

Figure A.1. – Bio-logger attached to a GPS collar fitted to a female kudu. Bio-loggers recorded audio data continuously until the batteries failed 6–51 days after deployment (mean = 33.7 days; 1 logger did not record any usable data).

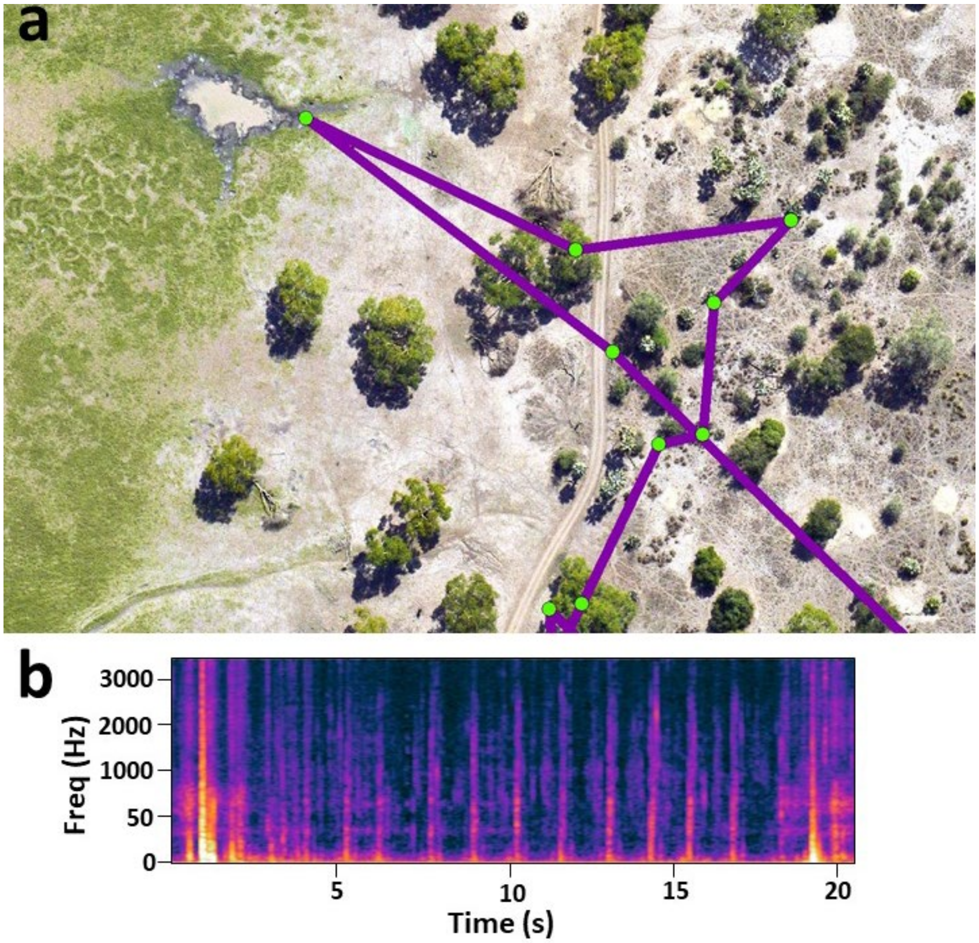


Figure A.2. – (a) Movement path of a collared female kudu in Gorongosa National Park, Mozambique from 1400 to 1424 UTC on August 21, 2021, overlaid on high-resolution LiDAR imagery obtained in August 2019. Green dots show GPS relocations every 3 mins. Fine-scale movement data were used to determine when to listen for the sound of drinking by kudu in audio data recorded by bio-loggers attached to GPS collars. In this example, a collared kudu approaches and then departs from a perennial pan (upper left corner). We reviewed audio data for approximately 15 min before and after all such instances where GPS-collar data indicated an individual was near a water source to minimize the possibility of missing drinking bouts. (b) Spectrogram of a kudu drinking for 16 seconds, identified aurally by listening to the recording during this movement window. Evenly spaced peaks in frequency indicate the rhythmic swallowing of water, and the distinctly elevated peaks in frequency at the beginning and end indicate the sound of the collar moving along the neck as the head is lowered to drink and then raised back up.

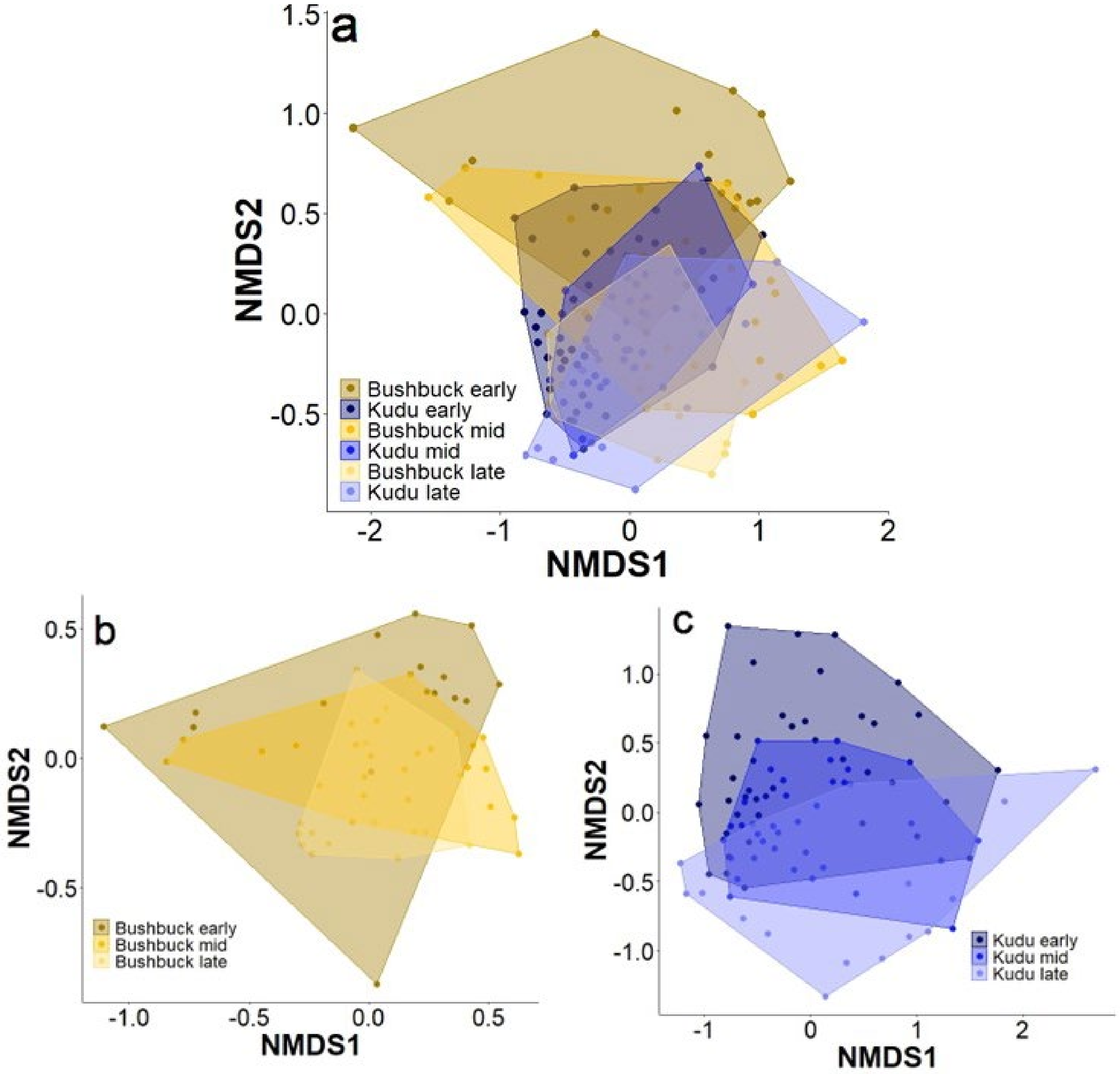


Figure A.3. – Nonmetric multidimensional scaling (NMDS) ordinations showing relative similarity in taxonomic composition of individual fecal samples (points; $n = 148$) and antelope diets (polygons; $n = 6$) in the early (May/June), mid (July/August), and late (September/October) dry season in Gorongosa National Park, Mozambique. Points in closer proximity to one another indicate more similar diets; polygons are convex hulls around all samples from each species grouped by time of the dry season. We observed significant differences in diet among species (perMANOVA: pseudo-$F_{1,146} = 11.194$, $P < 0.001$, $R^2 = 0.071$) and within species (bushbuck, early vs. mid: pseudo-$F_{1,41} = 3.21$, $P = 0.002$, $R^2 = 0.073$; mid vs. late: pseudo-$F_{1,38} = 3.11$, $P = 0.001$, $R^2 = 0.076$; early vs. late: pseudo-$F_{1,31} = 5.2$, $P = 0.001$, $R^2 = 0.144$; kudu, early vs. mid: pseudo-$F_{1,72} = 1.93$, $P = 0.062$, $R^2 = 0.026$; mid vs. late: pseudo-$F_{1,48} = 5.14$, $P = 0.002$, $R^2 = 0.097$; early vs. late: pseudo-$F_{1,54} = 5.35$, $P = 0.001$, $R^2 = 0.09$) across the dry season.

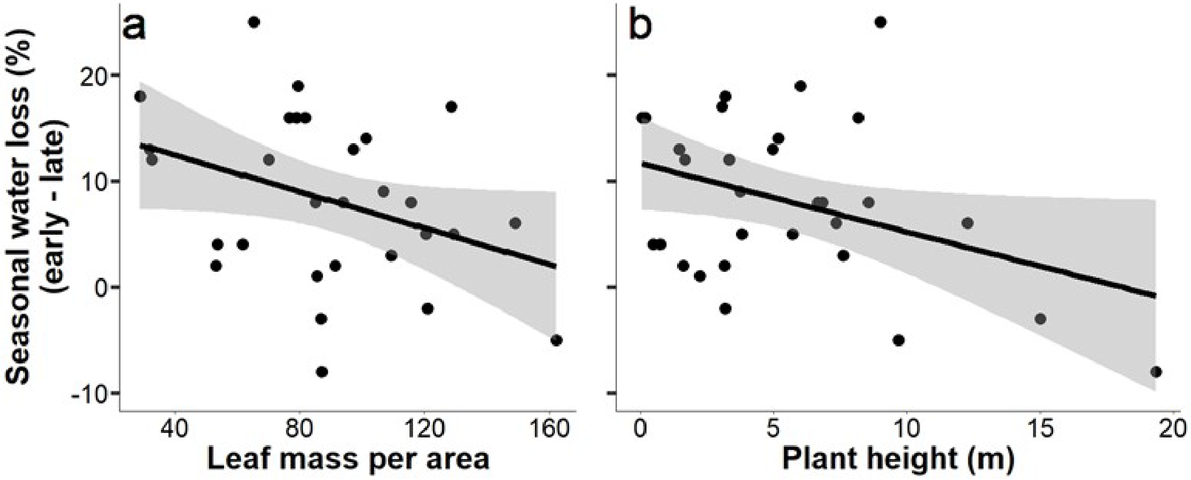


Figure A.4. – Relationship between seasonal water loss of key forage plants (*n* =30) consumed by bushbuck and/or kudu (early minus late dry season values) and (a) leaf mass per area (β = -0.086, P = 0.055, adj. R^2 = 0.10) and (b) plant height (β = -0.65, P = 0.042, adj. R^2 = 0.11) in Gorongosa National Park, Mozambique. A single outlier, *Hyphaene petersiana* (makalani palm), with an LMA of 809, was not included in the analyses.

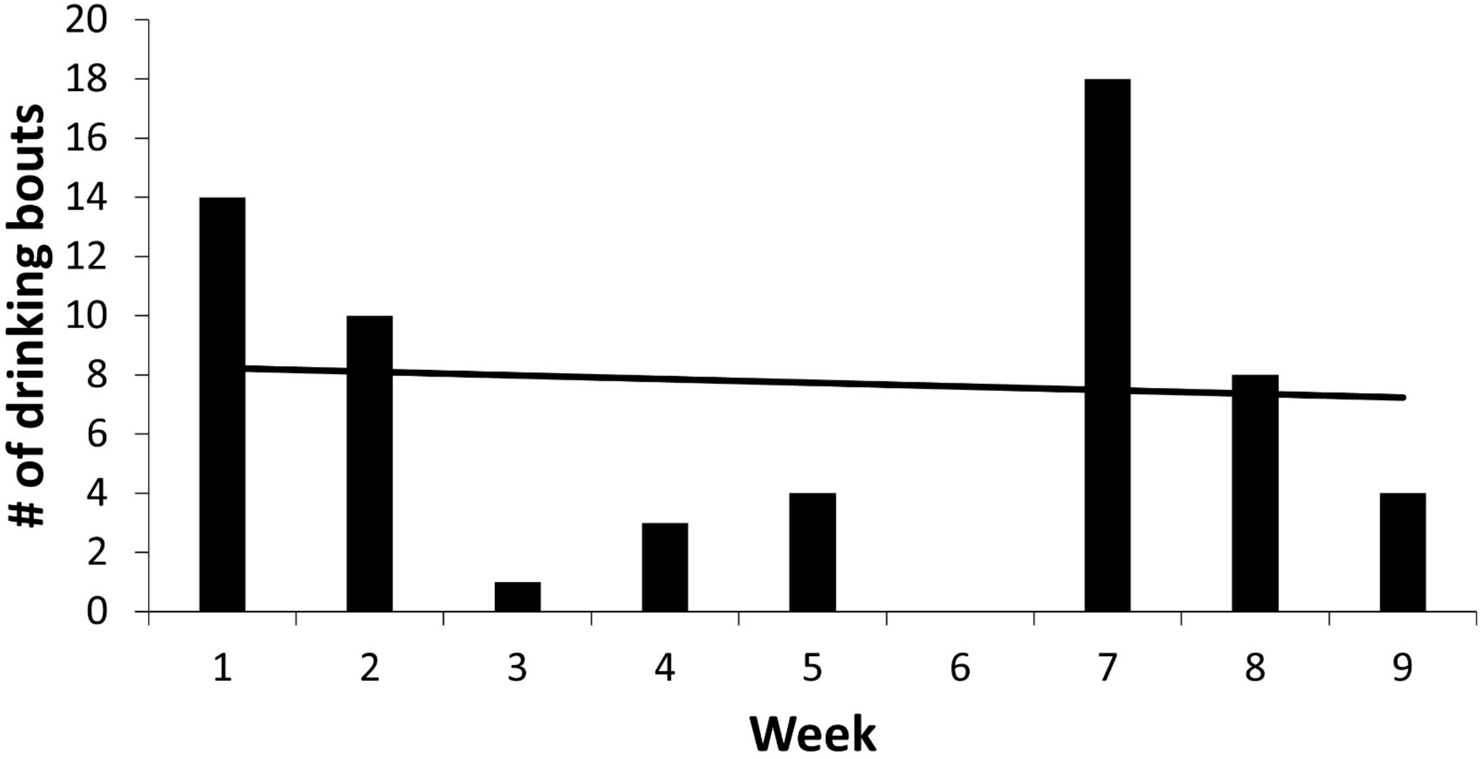


Figure A.5. – Weekly number of drinking bouts by bushbuck (*n* = 62 bouts) recorded using trail cameras (programmed to record videos in 15-s increments) placed on perennial pans used by GPS-collared individuals from 17 August–18 October 2021 (β = -0.12, P = 0.887, adj. R^2 = 0.00).

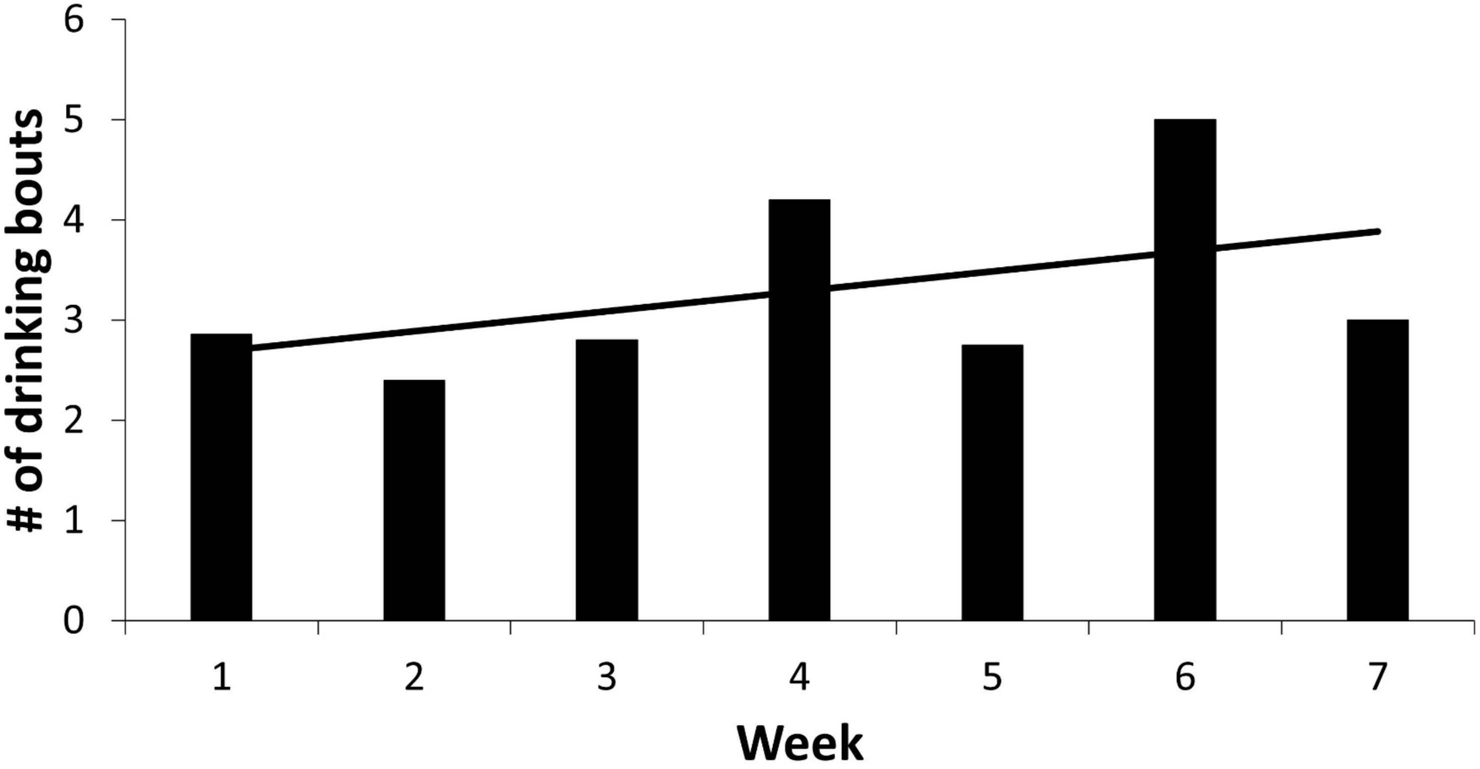


Figure A.6. – Mean number of drinking bouts per week (n = 102 drinking bouts, values averaged across 3–7 kudu monitored per week) recorded using audio loggers mounted to GPS collars from 13 July–28 August 2021 ($\beta = 0.20$, $P = 0.304$, adj. $R^2 = 0.05$).

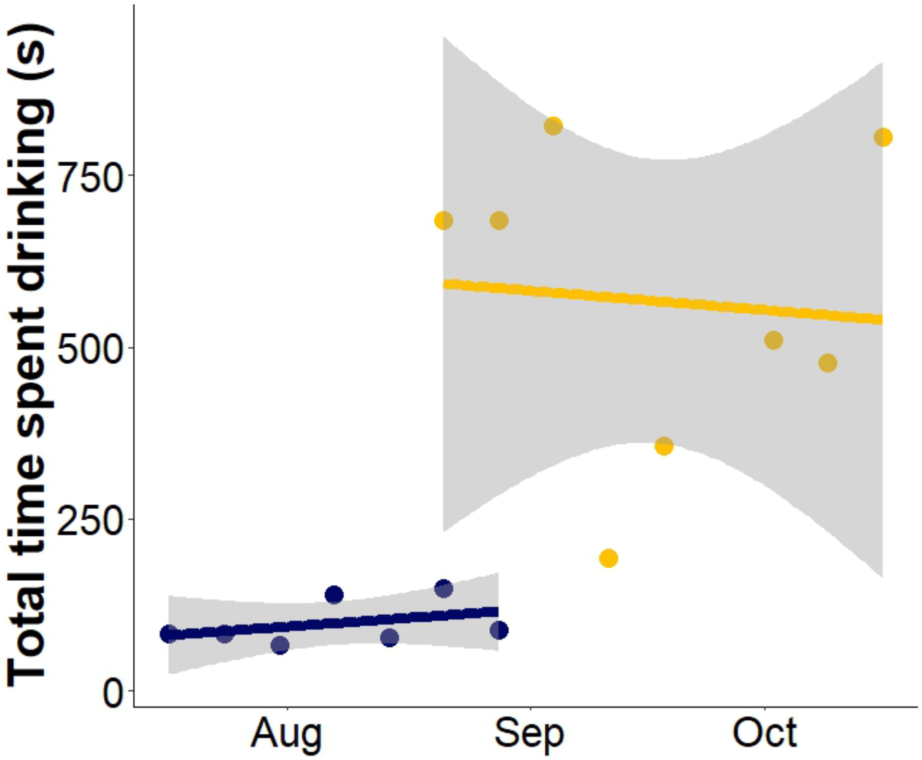
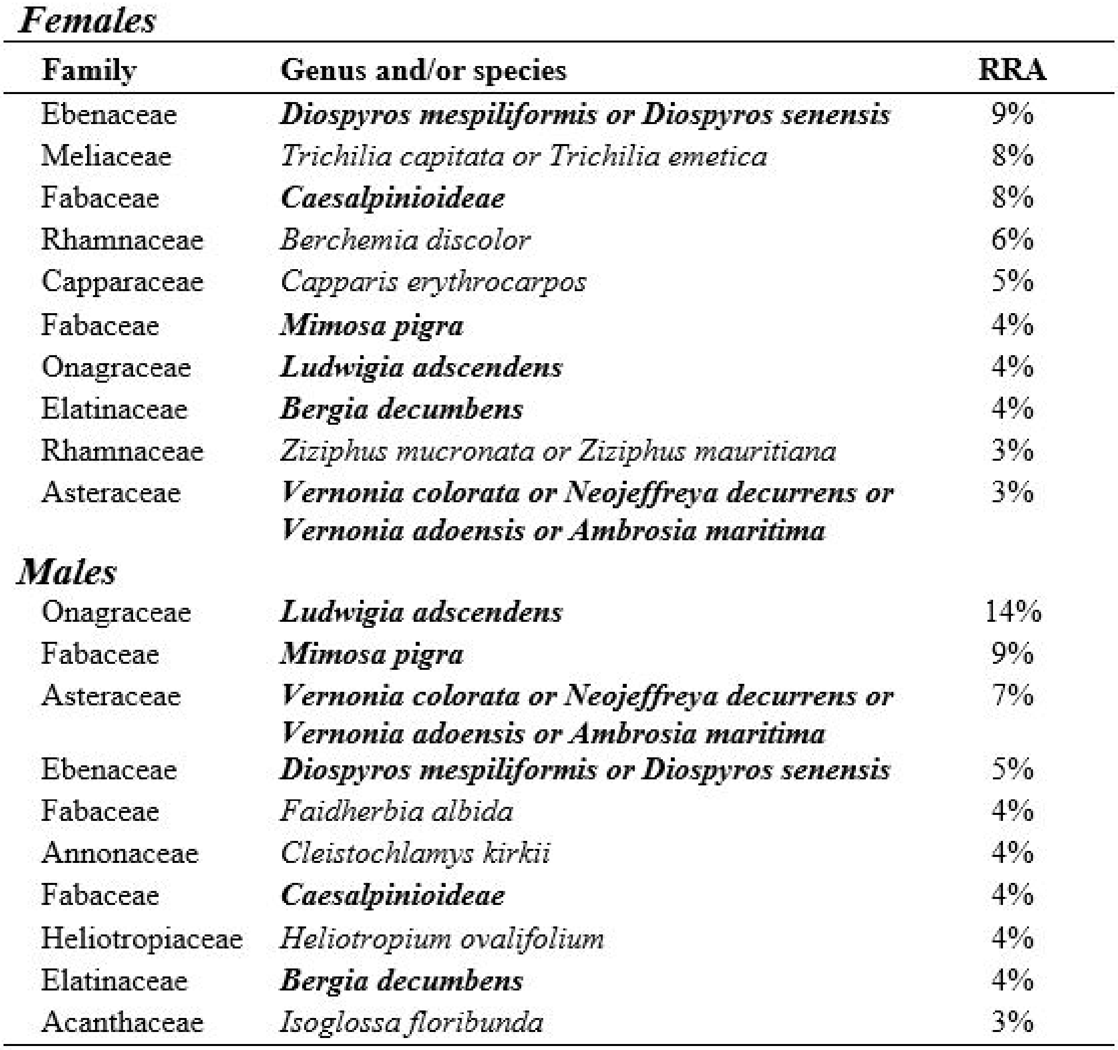


Figure A.7. – Total time spent drinking by bushbuck (*n* = 8, yellow) and kudu (*n* = 7, blue) across the dry season in Gorongosa National Park, Mozambique. Values for bushbuck were calculated by multiplying the mean number of drinking bouts per week (calculated across all 9 weeks) by the mean bout length per week (β_{Julian} = -0.94, P = 0.839, adj. R^2 = 0.00). Values for kudu were calculated by multiplying the mean number of drinking bouts per week (calculated across monitored individuals within each week) by the mean bout length per week (β_{Julian} = 0.82, P = 0.395, adj. R^2 = 0.00).

Appendix B: Supplementary materials for "Chapter 2: Sex-dependent variation in diet and space use of congeneric African antelopes"

SUPPLEMENTARY TABLES

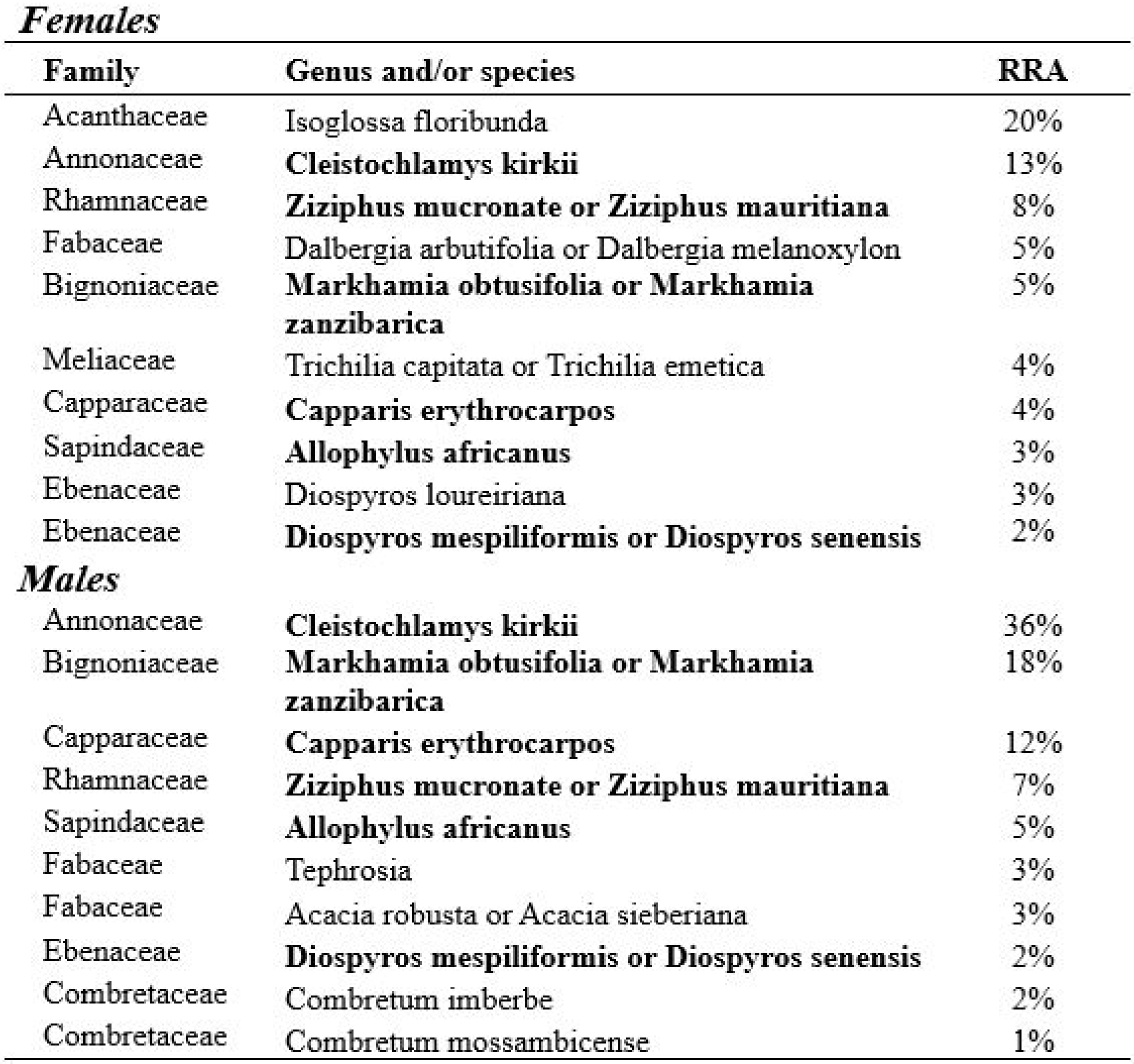
Table B.1. – Top 10 plant taxa (based on relative read abundance, RRA, from metabarcoding analysis of fecal samples) consumed by female and male bushbuck during the dry seasons of 2018–2019 and 2021–2022 in Gorongosa National Park, Mozambique. Bold font indicates species that were consumed by both sexes. These taxa comprised 56% and 60% of the diets of females and males, respectively.



Females

Family	Genus and/or species	RRA
Ebenaceae	***Diospyros mespiliformis or Diospyros senensis***	9%
Meliaceae	*Trichilia capitata or Trichilia emetica*	8%
Fabaceae	***Caesalpinioideae***	8%
Rhamnaceae	*Berchemia discolor*	6%
Capparaceae	*Capparis erythrocarpos*	5%
Fabaceae	***Mimosa pigra***	4%
Onagraceae	***Ludwigia adscendens***	4%
Elatinaceae	***Bergia decumbens***	4%
Rhamnaceae	*Ziziphus mucronata or Ziziphus mauritiana*	3%
Asteraceae	***Vernonia colorata or Neojeffreya decurrens or Vernonia adoensis or Ambrosia maritima***	3%
Males		
Onagraceae	***Ludwigia adscendens***	14%
Fabaceae	***Mimosa pigra***	9%
Asteraceae	***Vernonia colorata or Neojeffreya decurrens or Vernonia adoensis or Ambrosia maritima***	7%
Ebenaceae	***Diospyros mespiliformis or Diospyros senensis***	5%
Fabaceae	*Faidherbia albida*	4%
Annonaceae	*Cleistochlamys kirkii*	4%
Fabaceae	***Caesalpinioideae***	4%
Heliotropiaceae	*Heliotropium ovalifolium*	4%
Elatinaceae	***Bergia decumbens***	4%
Acanthaceae	*Isoglossa floribunda*	3%

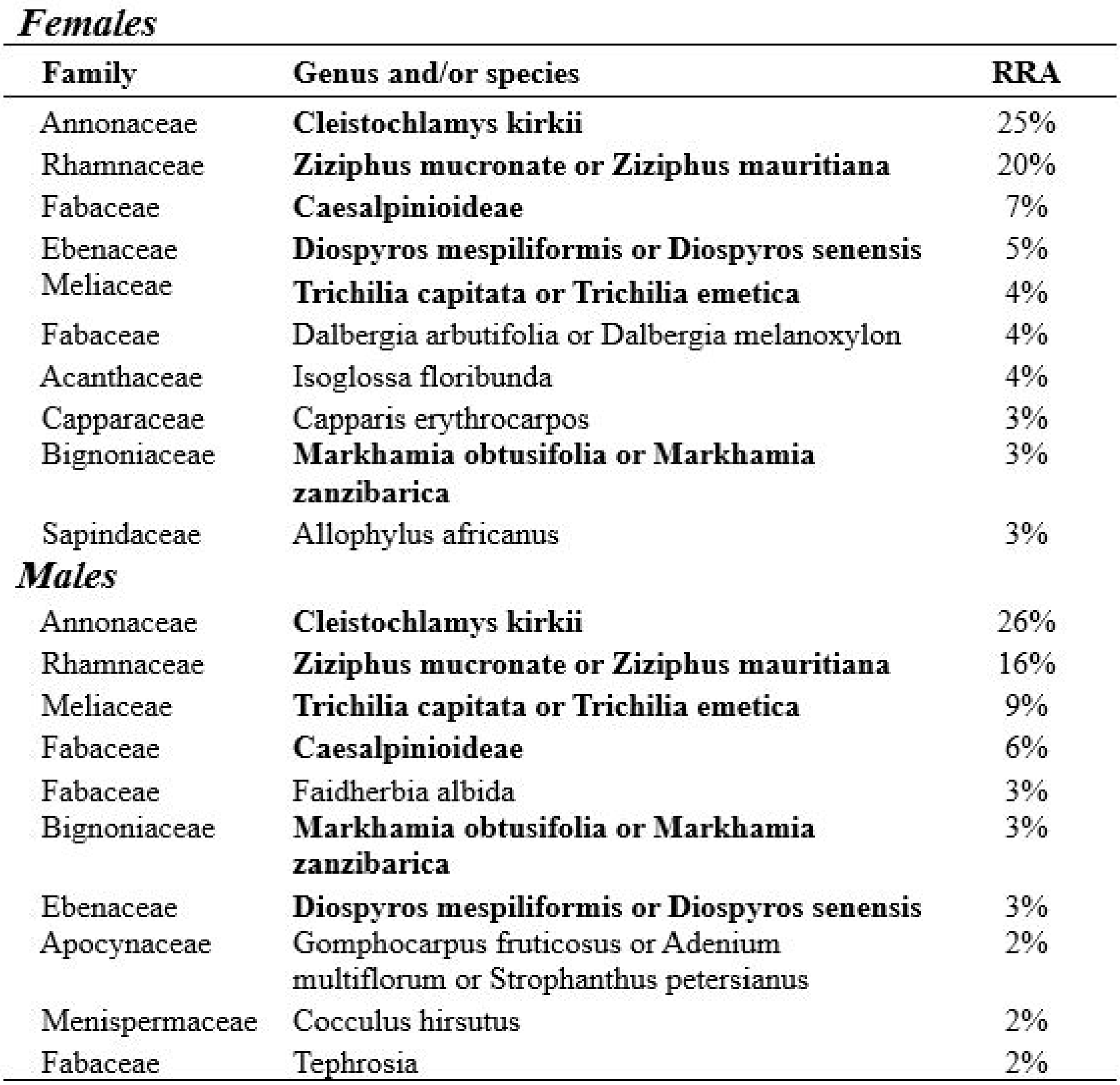
Table B.2. – Top 10 plant taxa (based on relative read abundance, RRA, from metabarcoding analysis of fecal samples) consumed by female and male nyala during the dry seasons of 2018–2019 and 2021–2022 in Gorongosa National Park, Mozambique. These taxa comprised 67% and 89% of the diets of females and males, respectively.



Females

Family	Genus and/or species	RRA
Acanthaceae	Isoglossa floribunda	20%
Annonaceae	**Cleistochlamys kirkii**	13%
Rhamnaceae	**Ziziphus mucronate or Ziziphus mauritiana**	8%
Fabaceae	Dalbergia arbutifolia or Dalbergia melanoxylon	5%
Bignoniaceae	**Markhamia obtusifolia or Markhamia zanzibarica**	5%
Meliaceae	Trichilia capitata or Trichilia emetica	4%
Capparaceae	**Capparis erythrocarpos**	4%
Sapindaceae	**Allophylus africanus**	3%
Ebenaceae	Diospyros loureiriana	3%
Ebenaceae	**Diospyros mespiliformis or Diospyros senensis**	2%
Males		
Annonaceae	**Cleistochlamys kirkii**	36%
Bignoniaceae	**Markhamia obtusifolia or Markhamia zanzibarica**	18%
Capparaceae	**Capparis erythrocarpos**	12%
Rhamnaceae	**Ziziphus mucronate or Ziziphus mauritiana**	7%
Sapindaceae	**Allophylus africanus**	5%
Fabaceae	Tephrosia	3%
Fabaceae	Acacia robusta or Acacia sieberiana	3%
Ebenaceae	**Diospyros mespiliformis or Diospyros senensis**	2%
Combretaceae	Combretum imberbe	2%
Combretaceae	Combretum mossambicense	1%

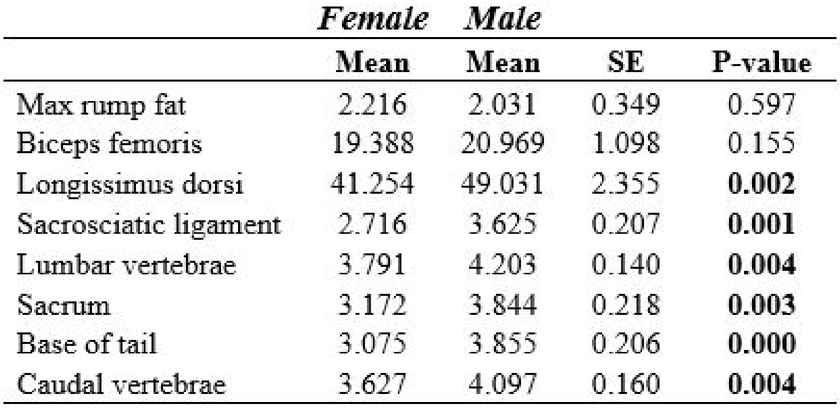
Table B.3. – Top 10 plant taxa (based on relative read abundance, RRA, from metabarcoding analysis of fecal samples) consumed by female and male kudu during the dry seasons of 2018–2019 and 2021–2022 in Gorongosa National Park, Mozambique. Bold font indicates species that were consumed by both sexes. These taxa comprised 75% and 73% of the diets of females and males, respectively.



Females

Family	Genus and/or species	RRA
Annonaceae	**Cleistochlamys kirkii**	25%
Rhamnaceae	**Ziziphus mucronate or Ziziphus mauritiana**	20%
Fabaceae	**Caesalpinioideae**	7%
Ebenaceae	**Diospyros mespiliformis or Diospyros senensis**	5%
Meliaceae	**Trichilia capitata or Trichilia emetica**	4%
Fabaceae	Dalbergia arbutifolia or Dalbergia melanoxylon	4%
Acanthaceae	Isoglossa floribunda	4%
Capparaceae	Capparis erythrocarpos	3%
Bignoniaceae	**Markhamia obtusifolia or Markhamia zanzibarica**	3%
Sapindaceae	Allophylus africanus	3%
Males		
Annonaceae	**Cleistochlamys kirkii**	26%
Rhamnaceae	**Ziziphus mucronate or Ziziphus mauritiana**	16%
Meliaceae	**Trichilia capitata or Trichilia emetica**	9%
Fabaceae	**Caesalpinioideae**	6%
Fabaceae	Faidherbia albida	3%
Bignoniaceae	**Markhamia obtusifolia or Markhamia zanzibarica**	3%
Ebenaceae	**Diospyros mespiliformis or Diospyros senensis**	3%
Apocynaceae	Gomphocarpus fruticosus or Adenium multiflorum or Strophanthus petersianus	2%
Menispermaceae	Cocculus hirsutus	2%
Fabaceae	Tephrosia	2%

Table B.4. – Mean nutritional condition metrics for female and male Tragelaphine antelope. Nutritional condition metrics were collected at the time of capture and included: maximum rump fat depth, thickness of the *biceps femoris* muscle, thickness of the *longissimus dorsi* muscle, palpation score at the sacrosciatic ligament, palpation score at the lumbar vertebrae, palpation score at the sacrum, palpation score at the base of tail, and palpation score at the caudal vertebrae. Rump fat and muscle thickness were collected via ultrasonography and the palpation scores were based on protocols developed for North American ungulates (Cook et al. 2010, Stephenson et al. 2020) and used previously on African ungulates (Atkins et al. 2019, Becker et al. 2021, Walker et al. 2023). Bold font indicates statistical significance at the α = 0.05 level.



	Female	***Male***		
	Mean	**Mean**	**SE**	**P-value**
Max rump fat	2.216	2.031	0.349	0.597
Biceps femoris	19.388	20.969	1.098	0.155
Longissimus dorsi	41.254	49.031	2.355	**0.002**
Sacrosciatic ligament	2.716	3.625	0.207	**0.001**
Lumbar vertebrae	3.791	4.203	0.140	**0.004**
Sacrum	3.172	3.844	0.218	**0.003**
Base of tail	3.075	3.855	0.206	**0.000**
Caudal vertebrae	3.627	4.097	0.160	**0.004**

Supplementary Figures

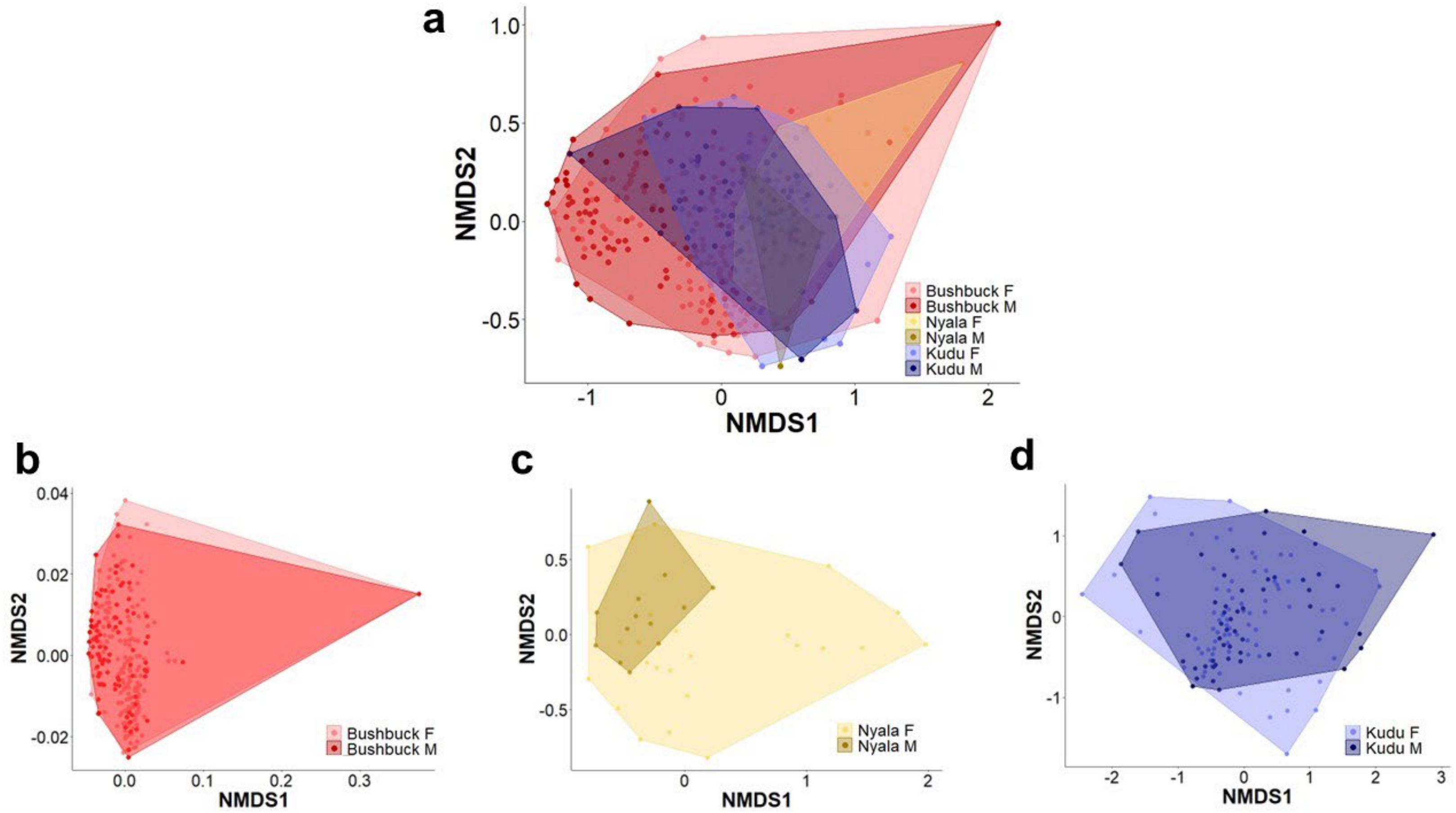


Figure B.1. – Nonmetric multidimensional scaling (NMDS) ordinations showing relative similarity in taxonomic composition of individual fecal samples (points; $n = 425$) and dry-season diets (polygons, $n = 6$; bushbuck shaded in red, nyala in yellow, and kudu in blue) of female and male antelope. Results are shown (a) for all species combined and (b–d) partitioned by species. Points in closer proximity to one another indicate more similar diets; polygons are convex hulls around all samples from each species, grouped by sex.

Diet composition differed significantly between the sexes for all three species (perMANOVA: bushbuck: pseudo-$F_{1,255} = 8.22$, $P = 0.001$, $R^2 = 0.031$; nyala: pseudo-$F_{1,35} = 3.56$, $P = 0.002$, $R^2 = 0.092$; kudu: pseudo-$F_{1,130} = 2.03$, $P = 0.042$, $R^2 = 0.015$).

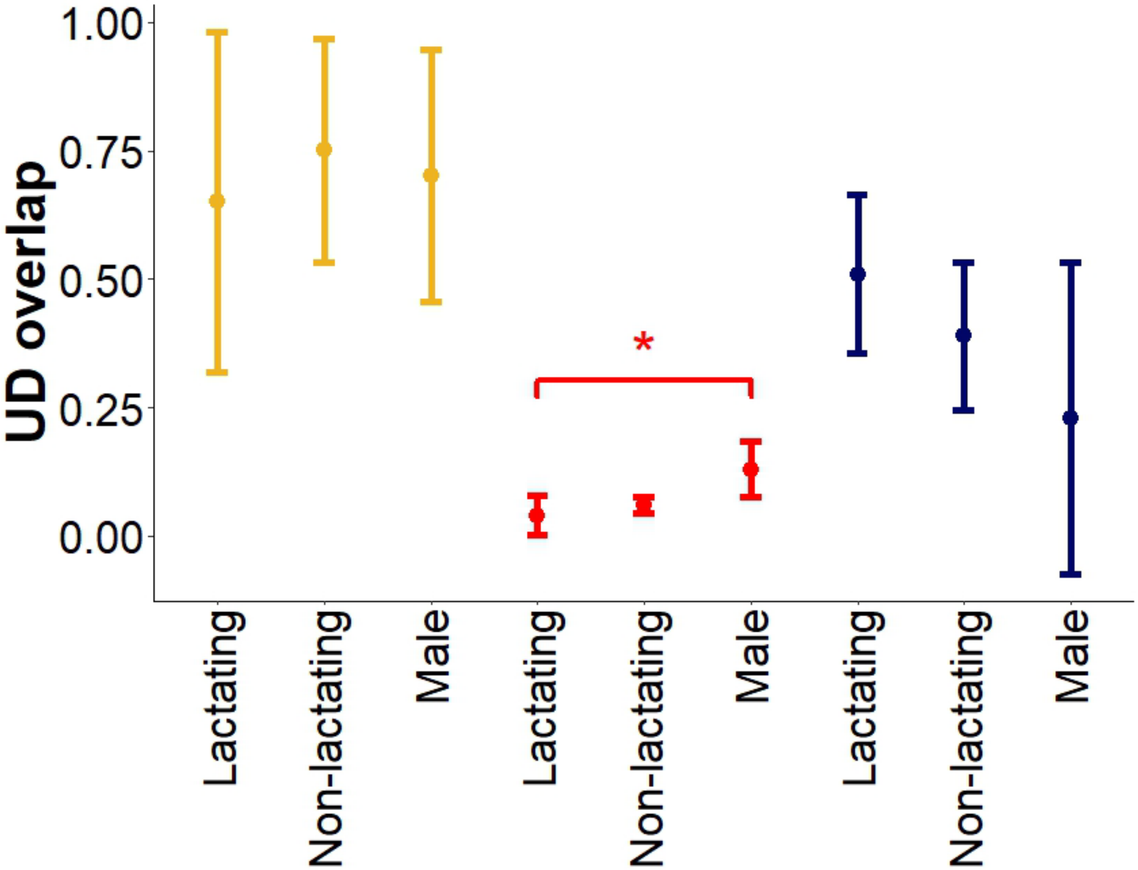


Figure B.2. – Mean (± 95% CI) proportion (volumetric) of antelope (female and male bushbuck (yellow), nyala (red) and kudu (blue)) utilization distributions (UD) that overlapped high-quality foraging habitat during dry seasons (May–October) of 2018–2019, and 2021 in Gorongosa National Park, Mozambique (bushbuck, lactating females n = 9; bushbuck, non-lactating females n = 15; bushbuck males, n = 13; nyala lactating females, n = 2; nyala non-lactating females, n = 3; nyala males, n = 7; kudu lactating females, n = 18; kudu non-lactating females, n = 15; kudu males, n = 6). Means were compared using one-way ANOVA followed by Tukey's HSD; asterisks indicate significant pairwise differences between reproductive category (bushbuck $F_{2,34} = 0.176$, $P = 0.84$; nyala $F_{2,9} = 4.078$, $P = 0.055$; kudu $F_{2,36} = 2.259$, $P = 0.12$; Tukey's HSD contrast for nyala males and lactating females $P = 0.095$).

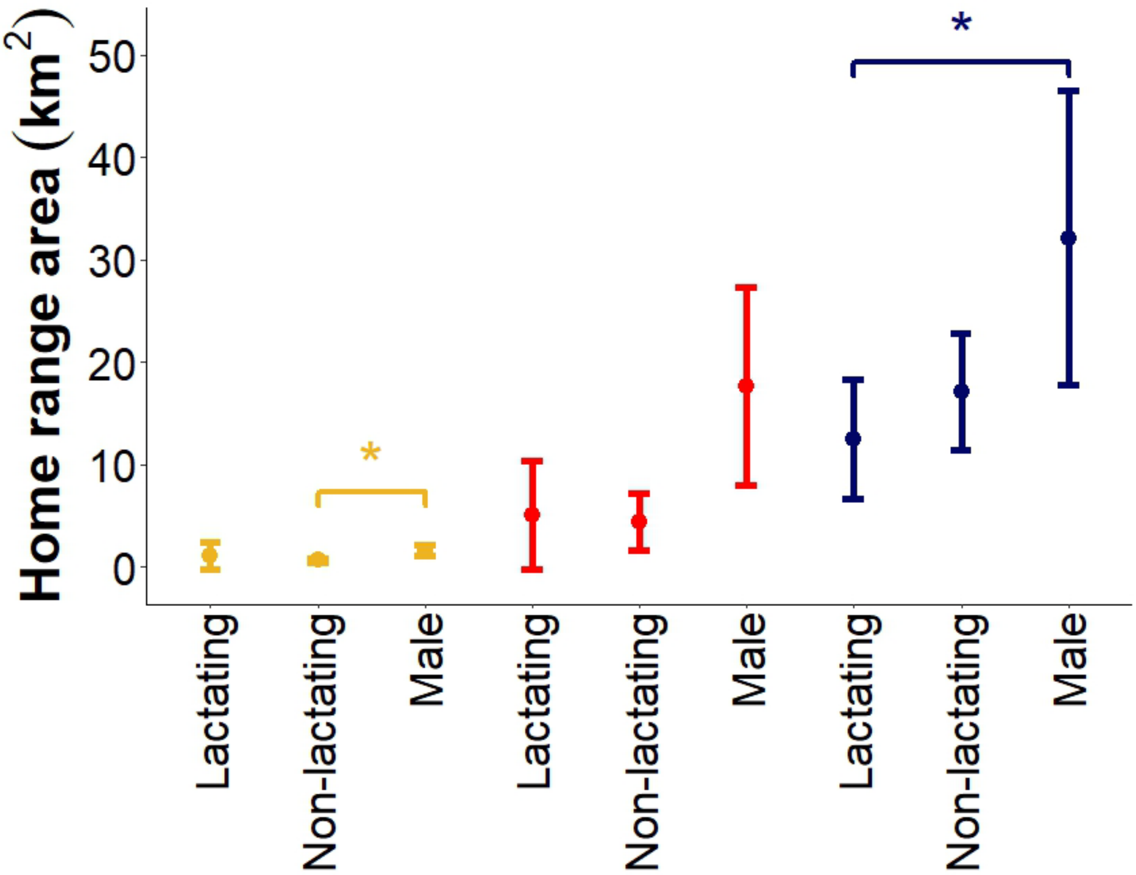


Figure B.3. – Mean (± 95% CI) home range area (95% UD) of female and male bushbuck (yellow), nyala (red) and kudu (blue) during dry seasons (May–October) of 2018–2019, and 2021 in Gorongosa National Park, Mozambique (bushbuck lactating females, *n* = 9; bushbuck non-lactating females, *n* = 15; bushbuck males, *n* = 13; nyala lactating females, *n* = 2; nyala non-lactating females, *n* = 3; nyala males, *n* = 7; kudu lactating females, *n* = 18; kudu non-lactating females, *n* = 15; kudu males, *n* = 6). Means were compared using one-way ANOVA followed by Tukey's HSD; asterisks indicate significant pairwise differences between reproductive category (bushbuck $F_{2,34} = 3.049$, $P = 0.061$; nyala $F_{2,9} = 3.346$, $P = 0.082$; kudu $F_{2,36} = 6.52$, $P = 0.004$; Tukey's HSD contrast for bushbuck males and non-lactating females $P = 0.048$; Tukey's HSD contrast for kudu males and lactating females $P = 0.003$, kudu males and non-lactating females $P = 0.028$).

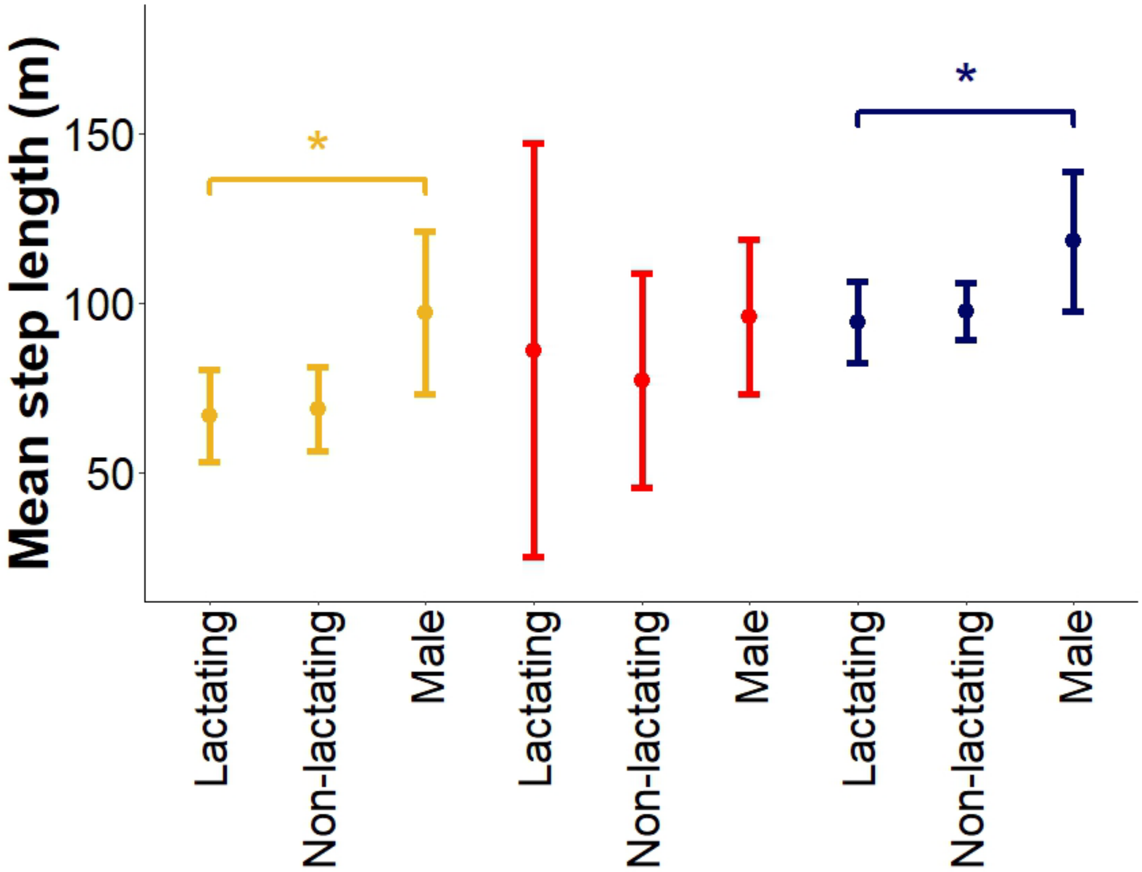


Figure B.4. – Mean (± 95% CI) step length of female and male bushbuck (yellow), nyala (red) and kudu (blue) during dry seasons (May–October) of 2018–2019, and 2021 in Gorongosa National Park, Mozambique (bushbuck lactating females, *n* = 9; bushbuck non-lactating females, *n* = 14; bushbuck males, *n* = 8; nyala lactating females, *n* = 2; nyala non-lactating females, *n* = 3; nyala males, *n* = 7; kudu lactating females, *n* = 18; kudu non-lactating females, *n* = 15; kudu males, *n* = 6). Means were compared using one-way ANOVA followed by Tukey's HSD; asterisks indicate significant pairwise differences between reproductive category (bushbuck: $F_{2,28} = 4.961$, $P = 0.014$; nyala $F_{2,9} = 0.864$, $P = 0.454$; kudu: $F_{2,36} = 3.149$, $P = 0.055$; Tukey's HSD contrast for bushbuck males and lactating females $P = 0.026$, males and non-lactating females $P = 0.023$; Tukey's HSD contrast for kudu males and lactating females $P = 0.047$, males and non-lactating females $P = 0.1$).

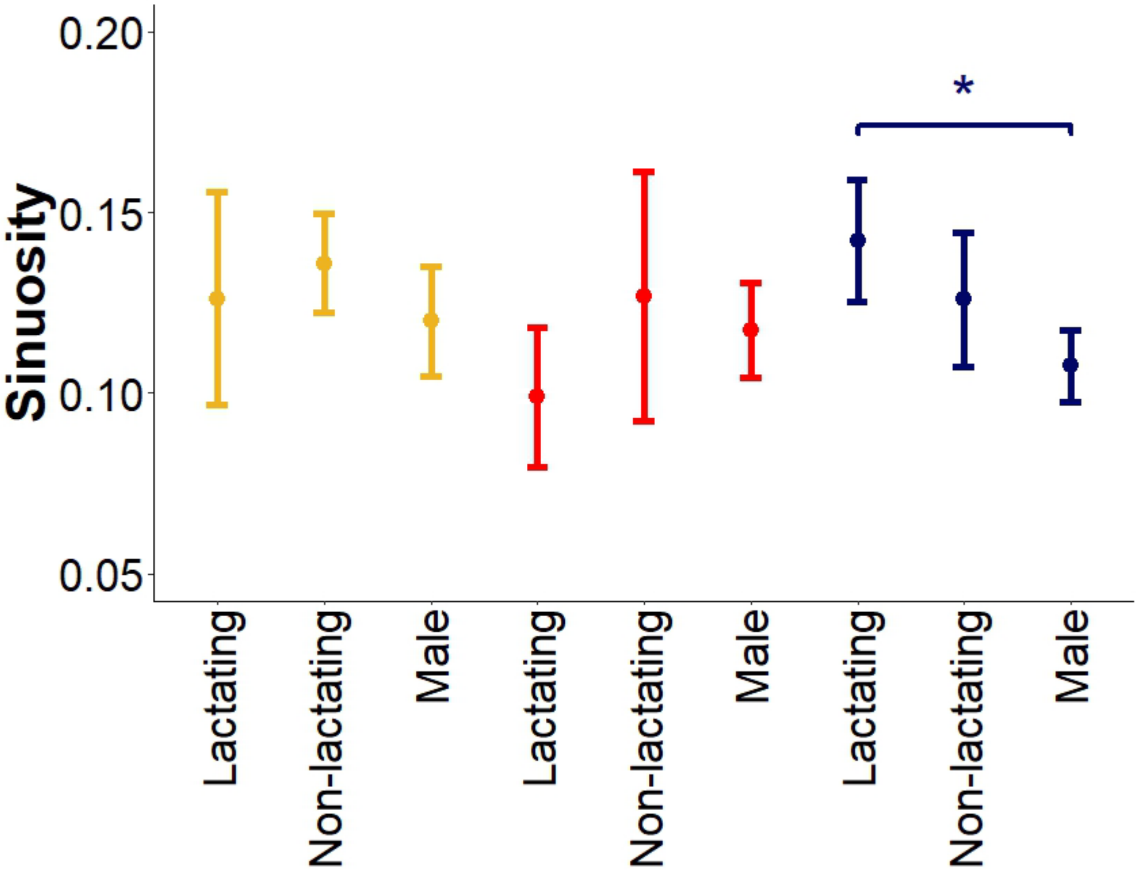


Figure B.5. – Mean (± 95% CI) sinuosity of the movement tracks of female and male bushbuck (yellow), nyala (red) and kudu (blue) during dry seasons (May–October) of 2018–2019, and 2021 in Gorongosa National Park, Mozambique (bushbuck lactating females, *n* = 9; bushbuck non-lactating females, *n* = 15; bushbuck males, *n* = 13; nyala lactating females, *n* = 2; nyala non-lactating females, *n* = 3; nyala males, *n* = 7; kudu lactating females, *n* = 18; kudu non-lactating females, *n* = 15; kudu males, *n* = 6). Greater sinuosity values indicate more tortuous movements, potentially indicative of greater foraging selectivity. Means were compared using one-way ANOVA followed by Tukey's HSD; asterisks indicate significant pairwise differences between reproductive categories (bushbuck: $F_{2,34} = 1.117$, $P = 0.339$; nyala $F_{2,9} = 2.624$, $P = 0.127$; kudu: $F_{2,36} = 3.01$, $P = 0.062$; Tukey's HSD contrast for kudu males and lactating females $P = 0.063$).

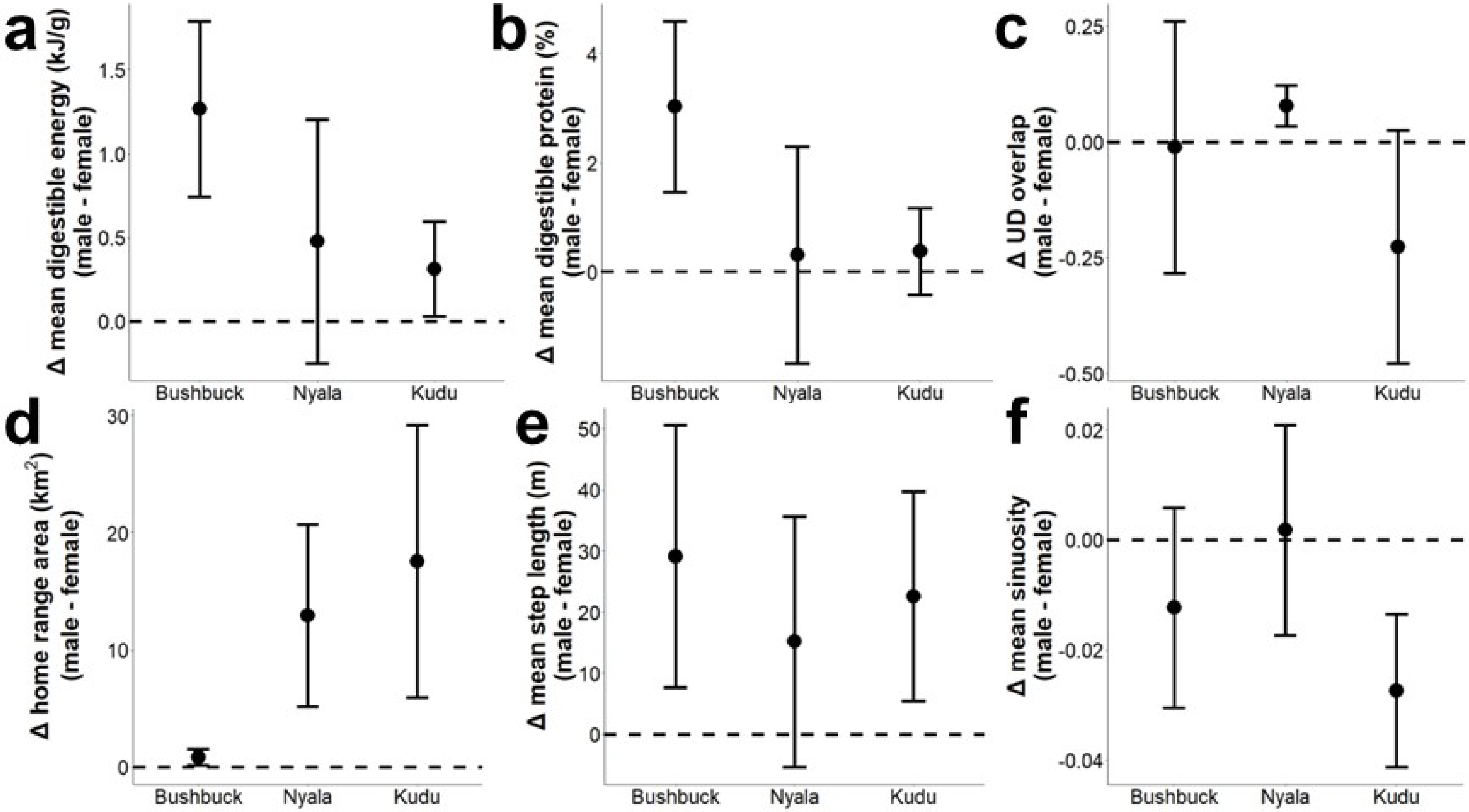


Figure B.6. – Differences (± 95% CI) in (a) digestible energy, (b) digestible protein, (c) use of high-quality foraging habitat, (d) home range area (95% UD), (e) mean step length, and (f) sinuosity between male and female bushbuck, nyala, and kudu during the dry seasons (May–October) of 2018–2019 and 2021 in Gorongosa National Park, Mozambique. Confidence intervals that overlap 0 indicate no difference between the sexes.

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