

**RESOURCE SELECTION OF AN ENDANGERED MOUSE *PEROHNATHUS*
LONGIMEMBRIS PACIFICUS: USING PLANT COVER SURVEYS AND SCAT
ANALYSIS**

by

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A professional paper submitted in partial fulfillment
of the requirements for the degree

of

Master of Science

in

Land Resources and Environmental Sciences

MONTANA STATE UNIVERSITY
Bozeman, Montana

December 2024

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ACKNOWLEDGEMENTS

I would like to thank my advisor Anthony Slominski for guidance and expertise in scientific writing and in-depth analysis. If it not for him this project would not have been accomplished. I would also like to thank the great biologists from the U.S. Geological Survey San Diego Field Station for their continuous work studying this amazing little endangered mouse. More specifically Cheryl Brehme and Robert Fisher for their blessing in using valuable data gathered while monitoring for PPM.

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ABSTRACT

Resource selection of the endangered Pacific Pocket Mouse (*Perognathus longimembris pacificus*; PPM) is poorly understood and past limitations in study methods have left a gap in an important part of the life history of the species. Understanding what types of resources they are consuming and when are important for current and future species management decisions regarding habitat enhancement and reintroduction sites. Therefore, I used data obtained by metabarcoding of PPM scat and plant phenology survey data to estimate food availability, to assess diet composition and analyze diet selectivity (diet composition – food availability) at the plant functional group level for the rodent community throughout two PPM populations. The study was performed at two extant populations on MCB Camp Pendleton in 2016 and 2017. Plant phenology surveys show trends in resource availability that matched what was expected in a coastal sage scrub habitat type. Diets for PPM and competitor rodent mimicked each other at both populations across multiple seasons and years. Also, forb use was high by PPM and competitor rodent species at both populations across seasons and years regardless of the percent cover of other functional groups. Understanding PPM resource use from this study will inform land managers how to better manage PPM habitat and make decisions to enhance currently occupied and future reintroduction sites. Future work to understand the specific plant species being used by PPM and their competitors will build on the work done in this project.

CHAPTER ONE

INTRODUCTION

The Pacific Pocket Mouse (PPM) is one of 19 subspecies of the little pocket mouse (*Perognathus longimembris*) in the heteromyid rodent family (Brehme et al. 2010). The PPM is the smallest of the species, weighing approximately 5 to 12 grams (Swei et al. 2003; USFWS 2010) (Figure 1). They are nonsocial, nocturnal granivores with external fur-lined cheek pouches, adapted to warm dry climates (USFWS 1998, 2010). The PPM is endemic to the southern coast of California, historically distributed down the coastal strip from Marina Del Rey & El Segundo in Los Angeles County, south to the Tijuana River Valley (USFWS 1998, 2010). The PPM inhabits sparse coastal sage scrub ecosystems on marine terraces and alluvial plains, particularly in areas with fine-grain, sandy substrates with low silt content such as coastal dune, strand, river alluvium, and tidal flats within 4 kilometers of the ocean (Swei et al. 2003; USFWS 1998, 2010).

PPM were thought to be extinct after the development of “Spyglass Hill” in the San Joaquin Hills in 1972, which resulted in the destruction of habitat for the PPM and a number of other rodent species (Meserve 1976). They were rediscovered in 1993 in the Dana Point Headlands and were federally listed as endangered on 29 September 1994 (59 FR 49752; see Brehme & Fisher 2009; Brylski 1993; USFWS 1994). Researchers estimated that there were approximately 25-36 individuals present in Dana Point Headlands in 1993 (Brylski 1993; Spencer, 2005). Since their rediscovery in 1993, PPM were also discovered at three additional sites, all within the Marine Corps Base Camp Pendleton (MCBCP) (Figure 2): San Mateo North and South (also referred to collectively as San Onofre), and the Edson Range/Oscar One training areas near the Santa Margarita river mouth (Ogden 1995; Spencer 2015; USFWS 2010). The north San Mateo population has since been considered extirpated and no additional populations have been identified (USFWS 2015).

There are several major threats and stressors affecting these small PPM populations. Threats include habitat destruction, degradation (or alteration), and fragmentation mainly due to human activities including development, as well as recreational and military training activities

(such as foot traffic and off-road vehicles) (Brehme et al. 2010; USFWS 1998). Other environmental and ecological stressors include competition for limited resources – specifically by native and nonnative rodent species, and invasive Argentine ants (*Linepithema humile*) (Brehme and Fisher 2009; Brehme et al. 2010). Foraging activities may also be negatively impacted by vibrations, noise, and artificial light (Engel 2007; USFWS 2010, Brehme et al. 2014). Increased competition for limited resources impacts the PPM's ability to adequately forage and cache enough resources, resulting in decreased fitness.

PPM obtains both metabolic water and energy from seeds and they are highly efficient at minimizing evaporative water loss (French 1993). Their diet is comprised primarily of grass and forb seeds, and occasionally includes green vegetation and insects (Meserve 1976; USFWS 2010). Data on the dietary habits of PPM is limited; the most generally cited research study on heteromyid food habits is by Peter Meserve (1976). Through microscopic examination of feces and analysis of cheek contents, researchers identified several species of forbs, shrubs and grasses consumed by PPM including: *Centaurea melitensis* (tocalote), *Corethrogyne filaginifolia* (California aster), *Crysothamnus* sp. (rabbitbrush), *Croton californicus* (croton), *Erodium* sp. (storksbill), *Franseria* sp. (ragweed, bur-sage), *Gnaphalium* sp. (cudweeds), *Heterotheca grandiflora* (telegraph weed), *Hordeum murinum* (barley), *Pluchea sericea* (arrow weed), *Monanthochloe* sp. (shoregrass), *Salvia apiana* (sage), *Eriogonum fasciculatum* (California buckwheat), *Lotus scoparius* (California broom), *Rhus integrifolia* (lemonadeberry), and *Bromus* sp. (brome grasses) (Bailey 1939; Germano 1997; Meserve 1976; von Bloeker 1931).

In 1998 the U.S. Fish and Wildlife Service created the Pacific Pocket Mouse Recovery Plan, with goals for the removal and reclassification of PPM from the endangered species list to threatened status, with the main goal of maintaining sufficient populations and habitat throughout their range. Based on the USFWS PPM Recovery Plan, reclassification to threatened status will be considered when:

1. Ten populations are independently viable and stable or increasing, and their habitats are secure and fully protected.
2. Occupied habitat consists of a minimum of 2,000 hectares that are secure and fully protected.

3. All PPM populations are managed through a program to maintain genetic diversity for future generations.
4. All PPM populations and essential habitat are managed so that current and potential threats are eliminated or minimized to the extent that each population is not at risk of extirpation.

To achieve the USFWS PPM Recovery Plan, understanding PPM habitat requirements is extremely important. Some of the recovery actions USFWS has identified include: Identifying and protecting all extant populations and essential habitat; identifying prospective habitat and population sites; identifying essential habitats and sites; preparing and implementing habitat management plans; developing a species management plan; and enhancing and expanding PPM habitat (USFWS 1998). Based on the recovery plan: “Management of the Pacific pocket mouse habitat has not yet been attempted, and there is limited information on techniques that can be used to create, restore, enhance, or expand Pacific Pocket Mouse habitats” (USFWS 1999). Knowledge of the dietary habits of PPM is essential to establishing a robust management and rehabilitation plan. Understanding resource selection and availability can additionally provide other information on PPM habitat suitability that can further inform habitat management and restoration practices. Existing data on PPM seed preference is limited due to previous research and analysis methods (i.e., small sample size and limited plant species classifications) and potential differences in plant communities between previous and current population locations (Iwanowicz et al. 2016). Most of the previous PPM research studies were conducted in the 1930s prior to the development of “Spyglass Hill” when the species was thought to be extinct.



Figure 1. The Pacific Pocket Mouse (*Perognathus longimembris pacificus*).

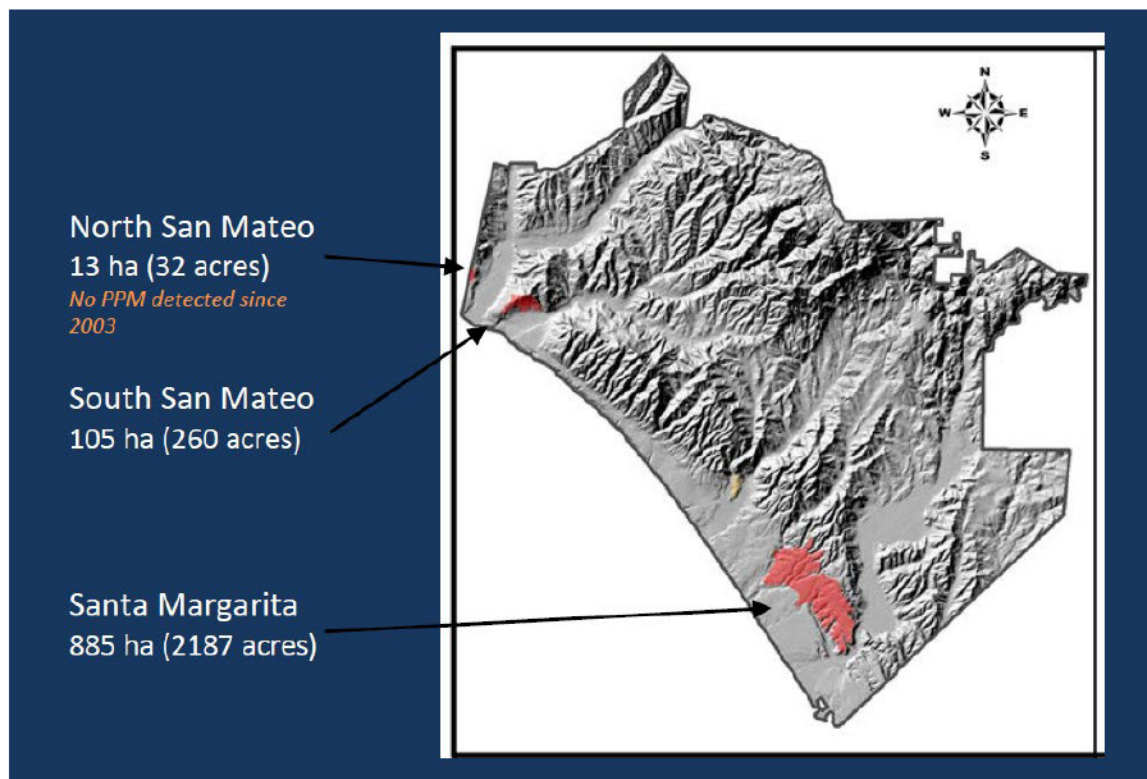


Figure 2. PPM populations on Camp Pendleton. North San Mateo PPM have been extirpated. South San Mateo and Santa Margarita are the two extant populations.

Research Overview and Goals

Knowledge of the dietary habits of PPM is essential to establishing a robust management and rehabilitation plan aimed at providing PPM habitat with adequate forage quality (i.e. nutrient composition) and quantity (i.e. food abundance). Such habitat is imperative for individual growth and survival, and directly influences population growth, stability, resilience, and may promote range expansion through dispersal from source populations (Anderson 1980). Central to these management implications is an understanding of 1) resource utilization relative to the seasonal availability of seed plant species and 2) the degree to which interspecific resource competition influences resource selection. To help address these knowledge gaps, the objectives of this research project are to 1) identify the food preferences of PPM and other native rodent species in the local rodent community, 2) gain a better understanding about food availability for PPM and other rodent species in the habitat, and 3) determine PPM and other rodent species resource selectivity based on resource availability and how it influences PPM resource use. The anticipated results from this project will provide specific information to help identify factors that influence PPM foraging patterns, as well as identify food resources that should be prioritized by land managers and habitat restoration projects to provide food resources for PPM across their entire foraging season. Establishing these priorities is particularly urgent because much of the remaining PPM habitat will continue to be impacted by human development and climate change (Riordan and Rundel 2014).

CHAPTER TWO

MATERIALS AND METHODS

Study Area

This study took place on Marine Corps Base Camp Pendleton in San Deigo, CA. The base is over 50585 hectares, which consists of many different habitat types from beach sand dunes, riparian, and coastal sage scrub among others. The habitat at the study site consists of coastal sage scrub species with early spring forb growth and thick areas of non-native grasses. Vegetation differs between the two populations with Santa Margarita consisting of lower shrub densities and higher non-native grasses, while South San Mateo has higher shrub densities and lower non-native grass densities (Figure 3). Both areas have native and non-native forb plant species present, mainly in the spring season after seasonal rains. Common shrubs at both locations include coastal sagebrush (*Artemisia californica*), white sage (*Salvia apiana*) and California buckwheat (*Eriogonum fasciculatum*). Both populations have non-native grasses including brome species (*Bromis sp.*) and wild oat (*Avena barbata*). Common forbs thought to be important to PPM include California Croton (*Croton californicus*), invasive Filaree species (*Erodium sp.*), and invasive mustards (*Brassica sp.*) The local rodent community (Table 1) is similar at both populations in the study, however their densities are not known and requires further studies to determine.

The study site lies just off the Pacific Ocean which provides cool summer and warmer winters with common night and early morning fog during the winter months. Rainfall is heaviest during the winter months, with most rainfall occurring late November to early February.

Average annual precipitation levels hover around 33.8 centimeters, spring rainfall is relatively minimal with averages around 1.27 centimeters (NOAA 2024). The fire regime on Camp Pendleton can be higher than usual because of the high amount of live fire ranges and overall amount of live fire drills that take place on the base. Fire activity historically has been different at both study populations with Santa Margarita experiencing fire on a more consistent basis than South San Mateo, which has not had a fire the previous twenty years. Santa Margarita recorded fires in 2011, 2012, and 2014. The fire histories at both sites have impacted the habitat cover and helps influence the differences in shrub cover at the two sites.

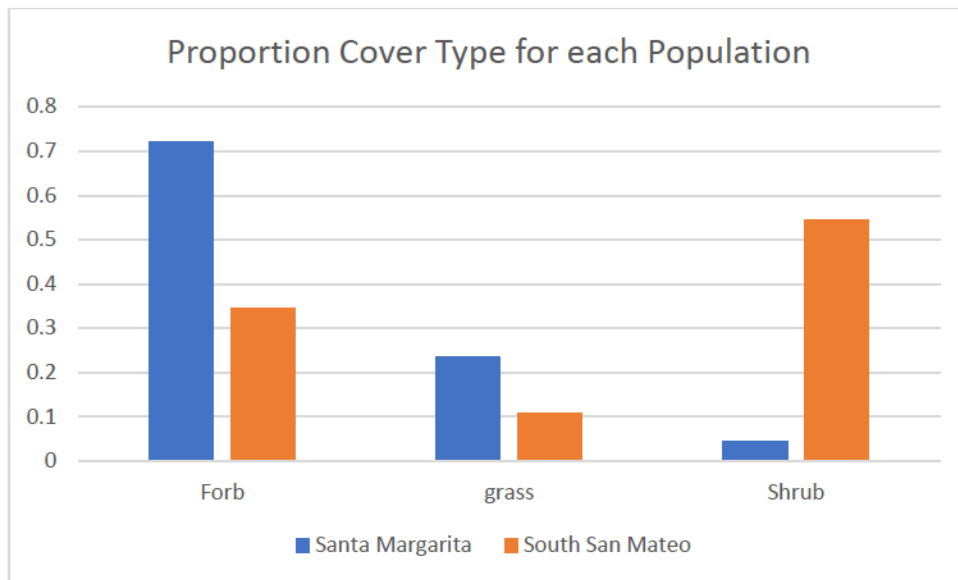


Figure 3. Proportion cover of functional groups at both populations (Santa Margarita in blue and South San Mateo in orange) based on plant phenology surveys.

Table 1. Native rodent species co-occurring with PPM at Santa Margarita and South San Mateo.

Scientific Name	Common Name
<i>Chaetodipus californicus</i> (CHCA)	California Pocket Mouse
<i>Reithrodontomys megalotis</i> (REME)	Western Harvest Mouse
<i>Peromyscus maniculatus</i> (PEMA)	Deer mouse
<i>Peromyscus eremicus</i> (PEER)	Cactus Mouse
<i>Peromyscus californicus</i> (PECA)	California Mouse
<i>Neotoma lepida</i> *	San Diego Desert Woodrat

*Not included in study. PEMA, PEER, and PECA are grouped into PESP hereafter.

Study Design

I collaborated with USGS scientists from the Western Ecological Research Center at the San Diego field station for this study. To include both population sites that PPM occupy, we implemented a large-scale study design to account for the varying conditions at each site. Diet analysis and plant phenology surveys were conducted from 2016 through 2017 at both PPM population sites: (1) Edson Range/Oscar One training areas (Santa Margarita) (33°15'32.70"N, 117°22'55.04"W) consisting of 2187 acres, and (2) South San Mateo (33°23'29.51"N, 117°33'49.61"W) at 260 acres. Diet analysis was completed through scat metabarcoding using scat collected during live-trapping sessions conducted by U.S. Geological Survey (USGS) biologists associated with a long-term monitoring research program for MCB Camp Pendleton (see Brehme and Fisher 2009). A total of 7 grids were monitored across both populations with 4 grids in the Santa Margarita population and 3 grids in the South San Mateo population. The

difference in grids per population corresponds to the total size of each populations potential PPM habitat, where Santa Margarita is over 700 hectares larger than South San Mateo.

Plant Resource Availability

I conducted plant phenology surveys during each live trapping session in conjunction with the USGS. Plant phenology surveys consisted of standardized visual vegetation estimates of cover type including trees, shrubs, forbs, native grasses, non-native grasses, and open ground (Table 2). For this study, functional plant cover, represents availability based on plant growth form which has been found to be the most important predictor for both vegetative and flowering phenology of plants (Iversen et al 2009). Each live trapping location had a pin flag marking the center of a 12.5 x 12.5 meter grid, which included a total of 16 subplots (Figure 4 & 5). Visual estimates were performed within a 6-meter radius of the central location on 8 of the trapping subplots, alternating between subplots (Figure 6). For each location, the total percent cover of each cover type was equal to at least 100 percent but could be > 100 percent if a cover type was overlapped with another cover type; for example, grass under a shrub would increase the cover at that location. Next, each plant in that plot was identified to the lowest taxonomic level possible, and a visual estimate of percent cover of each taxon was made in relation to the total specific cover type. For example, *Salvia apiana* (White sage) could be 50 percent of the total shrub cover of the plot. Lastly, current phenology (new growth, budding, flowering, set seed, seeded, dead) was identified for each plant species found on the plot. New growth was defined as any plant in the early stages of development that did not fit the criteria for other phenological categories. Budding was defined as having an undeveloped or rudimentary stem, branch, or flower of

a plant. Flowering was defined as having the presence of flowers on any plant (or part). Set seed was defined as the formation of (observable) seeds. Seeded was defined as having seeds spread or dropped from the plant. Each plant species could have multiple phenology states in a single plot; for example, White sage could be flowering and have set seed on the same subplot. To maintain consistent estimates throughout the season, all plant phenology surveys were conducted by myself at the same plot every trapping session.

Table 2. Cover type descriptions used to assess habitat cover at both Sana Margarita and South San Mateo, 2016-2018.

Cover Types	Description	Example Species	Example Common Name
Open Ground	Open ground has no other cover type, ppm can move freely.	None	None
Native Grass	Native grass species, generally low total cover	<i>Stipa cernua</i>	Native bunch grass
Non-Native grass	Non-native or naturalized grass species, generally thick cover, difficult for ppm to move through	<i>Bromus diandrus</i>	Ripgut Brome
Forbs	generally small perennial plant species	<i>Croton californicus</i>	California Croton
Shrubs	generally woody at least at the base, of larger size and live several years	<i>Salvia apiana</i>	White Sage
Tree		<i>Sambucus nigra</i>	Blue Elderberry



Figure 4. Habitat and center pin flag at a Santa Margarita subplot where plant phenology surveys were performed. Pink pin flag marks the center of the subplot and the location the survey was performed.



Figure 5. Habitat and center pin flag at a South San Mateo subplot where plant phenology surveys were performed. Blue flag marks the center of the subplot and the location the survey was performed.

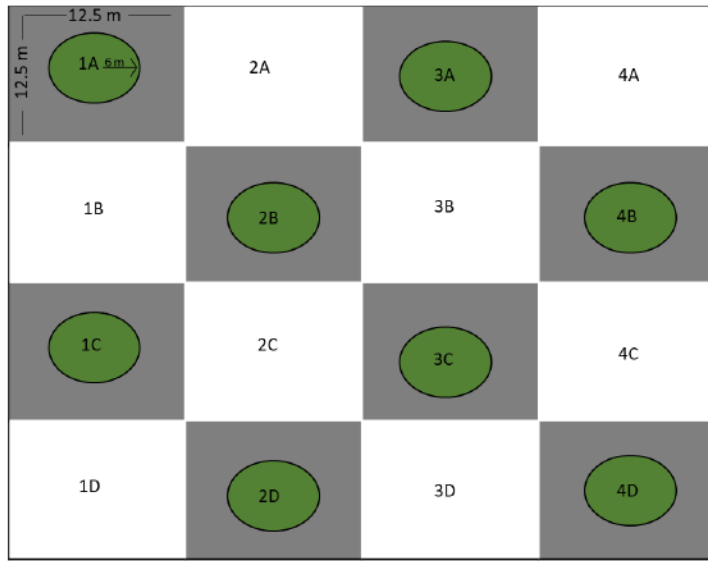


Figure 6. Grid and subplot formation, where each subplot was trapped and each subplot with a green circle a plant phenology survey was performed.

Small Mammal Trapping and Scat Samples

Specific live trapping protocol can be found in the PPM monitoring reports produced by Brehme et al. (2010, 2012, 2014, 2016). Live trapping locations were chosen based on USGS study protocol and conducted during 5 sessions from April-August for three consecutive nights at the same core grids each time (highlighted orange in Figure 7 & 8): four core grids from Santa Margarita (Figure 7) and three core grids from South San Mateo (Figure 8) and for a total of 1920 trap nights. Live trapping was conducted on 16, 12.5-square-meter plots within the 1-ha USGS core grids, with four traps placed within each 12.5-square-meter subplot (information on

tracking tube monitoring (Brehme et al. 2009, 2010, 2012, 2014, 2016). Scat was collected by USGS permitted biologists from live, in-hand PPM if they defecated during processing, which involved measuring, weighing, sex identification, and individual marking for recapture. Each scat sample was placed in a 1.5 mL microcentrifuge tube and labeled with the individual animal ID, location, and date for future reference.

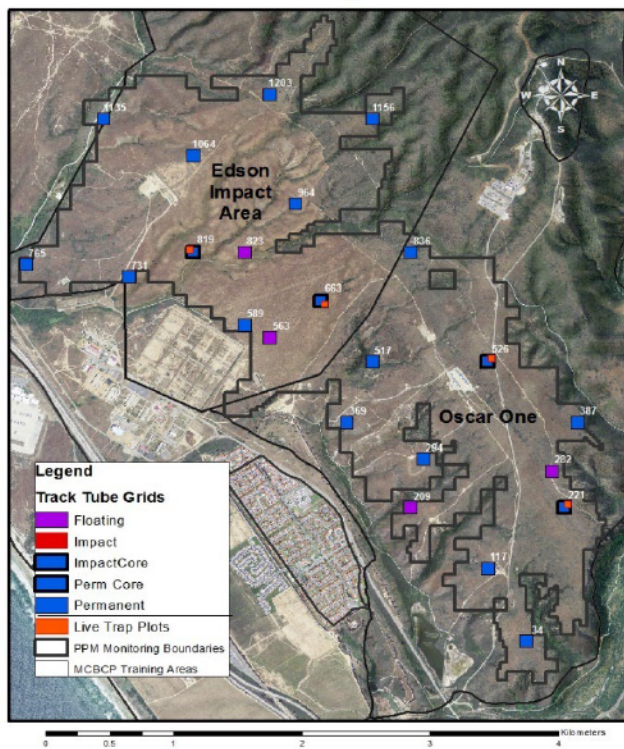


Figure 7. Live trapping grids in Santa Margarita, 2016-2017.

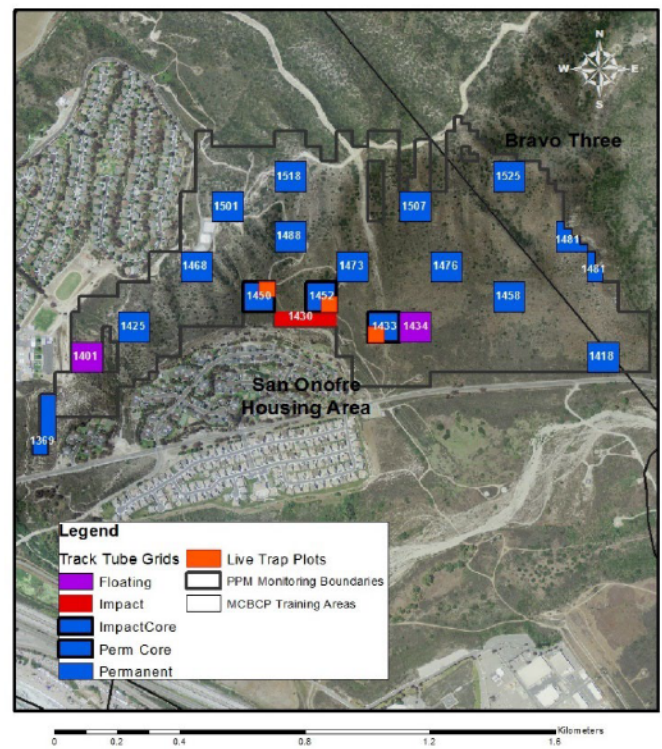


Figure 8. Live trapping grids in South San Mateo, 2016-2017.

Diet Analysis

In general, diet analyses on rodent species are performed in a variety of ways including morphological analysis of scat and the collection of seed from seed pouches or stomachs (Iwanowicz et al. 2016). These are invasive, labor intensive and time-consuming methods. Metabarcoding can be used as a non-invasive method to determine PPM diet. Metabarcoding requires comparing DNA sequences identified from scat samples to a DNA reference library (Kartzinel et al. 2015). Metabarcoding is an attractive approach because it works with small amounts of input material, is minimally invasive, has a high taxonomic resolution (compared to previous methods), and recovers a high proportion of total dietary consumption. PPM fecal pellets are extremely small (~ 2 mm in length and < 1 mg in weight) and contents are difficult to identify by morphological analyses, thus a metabarcoding approach is particularly appealing (Iwanowicz et al. 2016). Metabarcoding was performed and analyzed by USGS geneticists using protocol published by Iwanowicz et al. (2016). The resulting data is semi-quantitative, with consumed plant genera (and species) identified from each PPM and other species scat. Additionally, analysis was conducted to determine the proportion of plant functional group types in each scat. Once the proportion of each plant type was identified in each scat, the data were combined by small mammal species to determine the overall functional group proportions.

Diet Composition and Diet Selectivity

To determine diet composition at the functional group level (forb, grass, shrub/tree; non-native and native grass was combined), the proportion of each functional group in the diet was calculated for each individual mouse in the study. To assess the influence of food availability on diet composition, we calculated diet selectivity by subtracting the average percent cover of each functional group (forb, grass, and shrub/tree) at each trapping location from the proportion of each functional group in the diets of each trapped mouse (proportion in diet – proportion of percent cover). Average proportions of percent cover for each functional group were calculated along vertically oriented subplots in each grid. For example, percent cover of vegetation observed in subplots 1B and 1D in the trapping grid (figure 6) were used to calculate average percent cover for that vertical column of subplots (subplots 1A-1D; hereafter ‘plot’). Given the known foraging range of rodents in these communities, average percent cover values calculated at the plot level were representative of the food resources available to foraging mice trapped in the subplots of each plot. Diet composition and diet selectivity were compared between PPM and its competitor species across seasons and years, and between 2016 and 2017. Due to limited available data at both populations across all seasons and years non-PPM species (PPM competitor species) were combined into a single group (hereafter referred to as “other” rodent species).

Statistical analysis

All analyses were conducted using R statistical software version 4.3.1. To statistically assess the effects of food availability (percent cover), diet composition, and diet selectivity across populations, seasons, and years, we used linear mixed-effects models. Diet selectivity was set as the response variable, with average percent cover set as a fixed effect, following Soininen et al (2013). To assess differences in diet composition and diet selectivity between the PPM and its competitors, mouse species (PPM or other) was included as a fixed effect in each model. Grid and plot were included in each model as nested random effects. Models were fit separately to each functional group (forb, gras, shrub/tree) and separately by season (spring and summer) and year (2016 and 2017). The spring season was represented by data collected in March-May and the summer season was represented by data collected in June-August. All mixed effects models were fit using the lme4 package (Bates et al, 2014). Post hoc pairwise comparisons of average percent cover, diet composition, and diet selectivity were conducted using the emmeans package (Length 2024).

CHAPTER THREE

RESULTS

Diet Availability

Functional group cover, which is used here as a proxy for plant food availability, was consistent within populations between years, but differed across populations. For example, Santa Margarita had higher forb and grass cover (Table 3) compared to South San Mateo in both 2016 and 2017 (Figure 9 & 10), whereas South San Mateo had much higher average shrub cover (exceeding 90 percent) compared to Santa Margarita (2 percent). Seasonally, and within populations, the trends in functional group percent cover were similar between 2016 and 2017. For instance, forb cover increased from spring to summer, rising from 0.57 to 0.65 in 2016 and 0.56 to 0.69 in 2017 at Santa Margarita. Grass cover at Santa Margarita was the lone exception, where spring grass cover in 2017 (0.48) was lower than 2016 (0.60). Grass cover differs from the general trend, with all other cover types exhibiting changes of 0.10 percent in both years at each population.

Table 3. Functional group cover proportion for both populations in 2016 and 2017.

	Forb Spring	Forb Summer	Grass Spring	Grass Summer	Shrub/Tree Spring	Shrub/Tree Summer
Santa Margarita (2016)	0.57	0.65	0.60	0.72	0.01	0.01
South San Mateo (2016)	0.43	0.35	0.40	0.28	0.99	0.99
Santa Margarita (2017)	0.56	0.69	0.48	0.71	0.01	0.02
South San Mateo (2017)	0.44	0.31	0.52	0.29	0.99	0.98

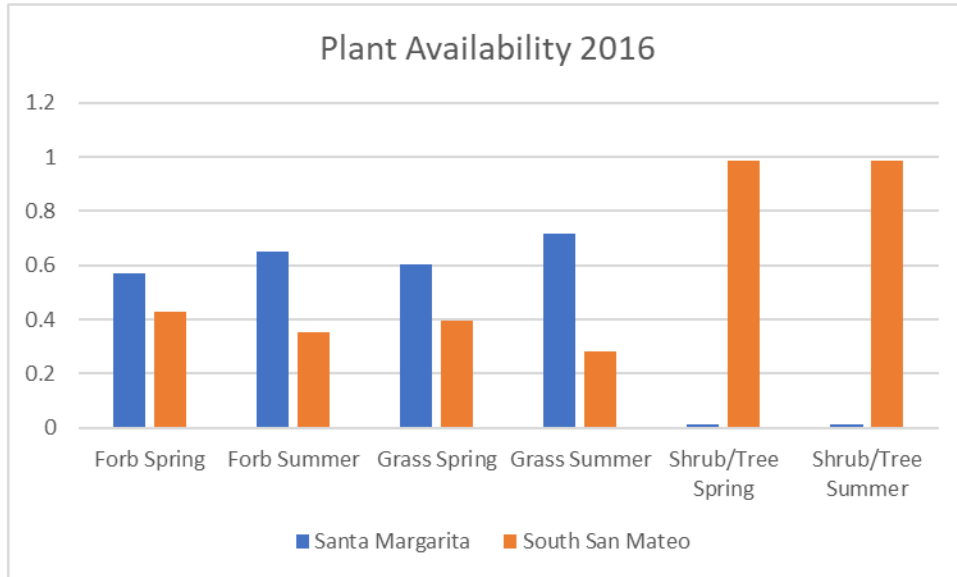


Figure 9. Plant functional group proportion available in 2016 for both populations.

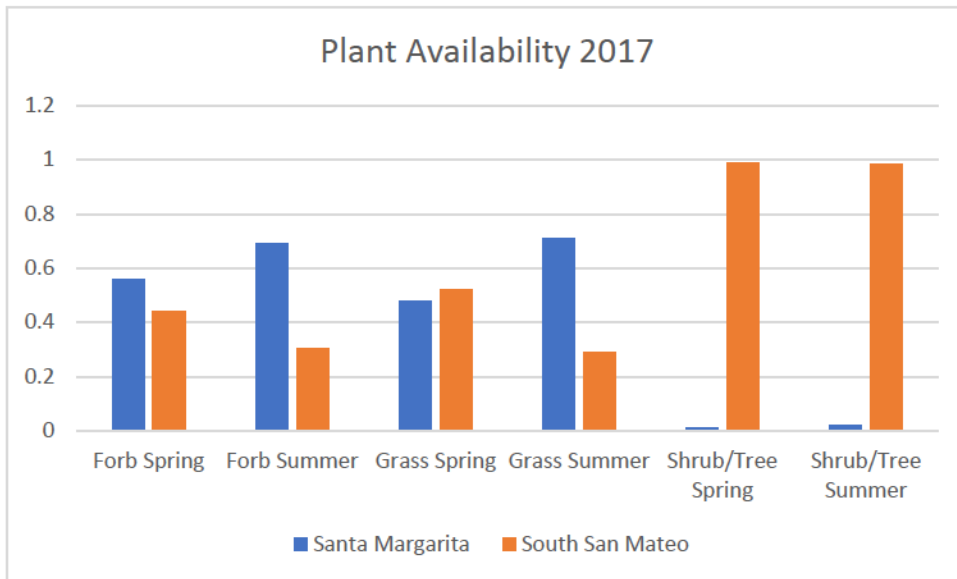


Figure 10. Plant functional group proportion available in 2017 for both populations.

Diet Composition

At the functional group level, forbs made up the majority of diets in both populations across seasons and years for both PPM and “other” rodents. However, the proportions of functional groups varied between the diets of PPM and other rodents across seasons and years.

Spring 2016: diet comparisons between populations

During the spring of 2016, PPM were highly dependent on forbs at both populations, which comprised over 75 percent of their diet (Figure 11). For the other species, forbs comprised 80 percent of diets at South San Mateo and they made up 66 percent of their diets at Santa Margarita (Figure 11). The proportion of grass in the diets of PPM and other species was higher at Santa Margarita (PPM= 0.18, other= 0.02) compared to South San Mateo (PPM= 0.1, other= 0) (Figure 11). Shrub consumption was highest at South San Mateo at over 10 percent for both PPM and PPM competitors, whereas at Santa Margarita shrubs comprised less than 1 percent of both PPM and other rodent species diets (Figure 11).

Summer 2016: diet comparisons between populations

In summer, the primary functional group in both PPM and other rodent species diet was forbs. Forbs represented 85 percent of diets in PPM and 82 percent of diets in other species at Santa Margarita, and consisted of 70 percent of diets in PPM and 82 percent of diets in other species at South San Mateo (Figure 12). The proportion of grass in the diets of both PPM and other species was minimal at South San Mateo (0.1) but was greater at Santa Margarita for only the other species (0.1) (Figure 12). Additionally, PPM consumed more shrubs than their

competitors at South San Mateo (PPM= 0.18, other= 0.1) while the opposite was true at Santa Margarita (PPM= 0.05, other= 0.1) (Figure 12).

2016: diet comparisons between spring and summer

In summer 2016, the proportion of forbs in PPM diets decreased at South San Mateo compared to spring, dropping from 78 percent to 68 percent (Figure 11 &12). The proportion of forbs in the diets of other rodent species remained above 80 percent for both seasons (Figure 11 &12). The proportion of grass in both PPM and other rodents' diet was less than 0.05 percent at South San Mateo for both seasons (Figure 11 &12). Shrub proportion in diet in the spring (PPM= 0.1, other= 0.08) was lower than the summer (PPM= 0.18, other= 0.1) season for both PPM and the other rodent species (Figure 11 &12).

At Santa Margarita forb consumption was high for PPM across both seasons, consisting of over 80 percent of their diet (Figure 11 &12). While other rodent species experienced an increase in forb consumption from spring (65 percent) to summer (80 percent) (Figure 11 &12). The proportion of grass in the diets of both PPM and other rodents decreased from spring (PPM= 0.18, other= 0.2) to summer (PPM= 0.02, other= 0.01), whereas shrub consumption increased slightly for the other rodent species from spring (0.02) to summer (0.1), while PPM remained similar for both seasons (0.02) (Figure 11 &12).

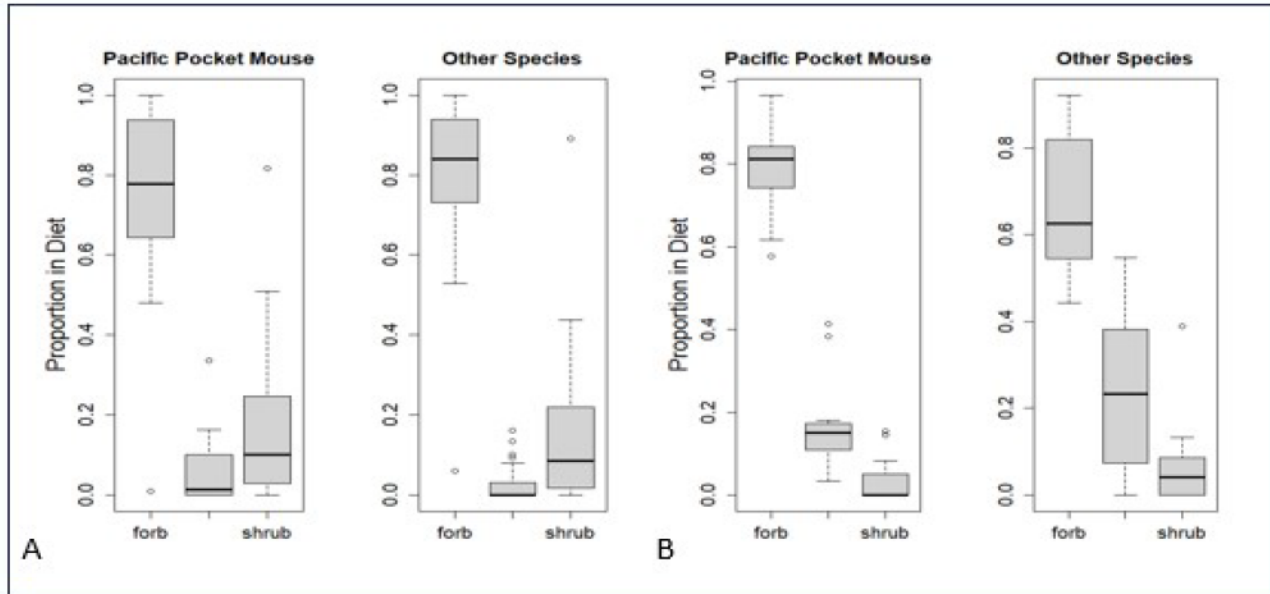


Figure 11. Proportion in diet for PPM and other rodent species at A) South San Mateo and B) Santa Margarita for spring 2016.

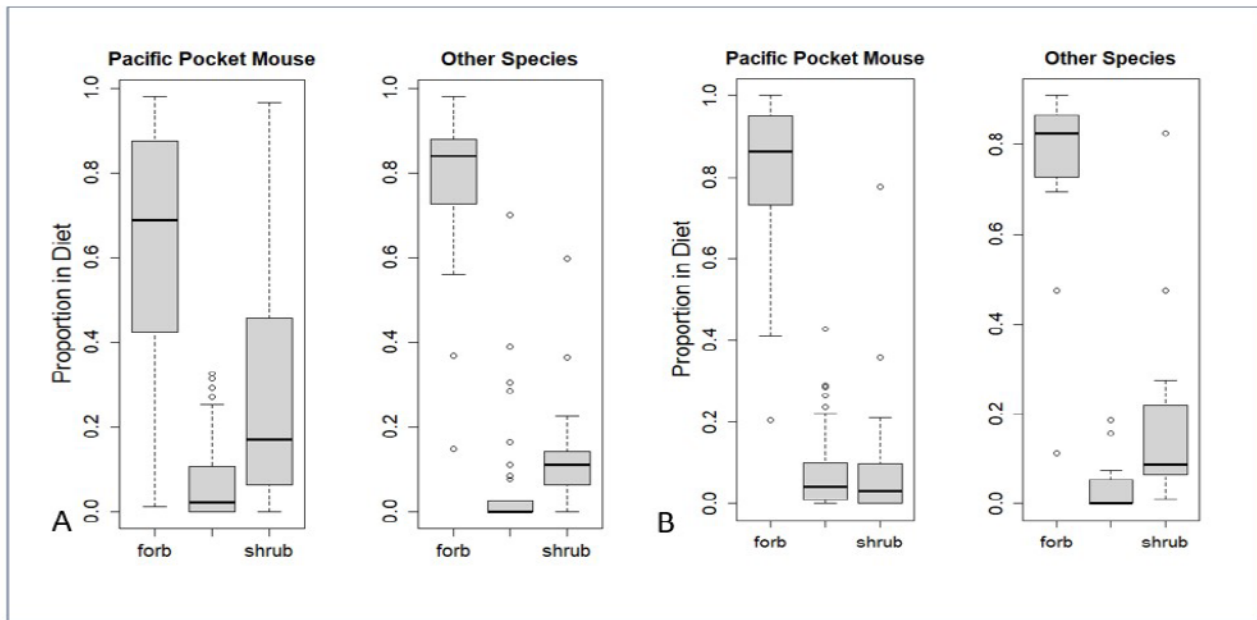


Figure 12. Functional group proportion in diet for PPM and other rodent species at A) South San Mateo and B) Santa Margarita for summer 2016.

Spring 2017: diet comparisons between populations

For the spring season, data was limited for both PPM and other rodent species at South San Mateo and only available for PPM at Santa Margarita, therefore only PPM will be compared for 2017. In spring 2017, the proportion of forbs in the diets of PPM was over 80 percent in both populations (Figure 13). Grass was the least consumed functional group in the diets of PPM at both populations for spring 2017 (Figure 13). The proportion of shrub in the diet of PPM was only slightly higher at South San Mateo (0.02) than Santa Margarita (0) (Figure 13).

Summer 2017 diet comparison between populations

In 2017, the proportion of forbs in the diets of PPM was over 80 percent in both populations during the summer. The proportion of grass in PPM diets at South San Mateo was higher (0.02) than at Santa Margarita (0.1) during the summer season (Figure 14). The proportion of shrub in PPM diets at South San Mateo (0.05) was slightly higher than at Santa Margarita (0.02) (Figure 14).

2017: Diet comparison between spring and summer

Temporal data for 2017 is exclusively for PPM, not enough data for other rodent species was available for analysis. Forb consumption at South San Mateo was higher in the spring (90 percent) compared to the summer (80 percent) (Figure 13 & 14). The proportion of grass in the diets of PPM at South San Mateo increased slightly from spring (0.01) to the summer (0.02), while shrub proportions slightly increased from spring (0.02) to summer (0.05) (Figure 13 & 14).

Santa Margarita exhibited very high proportions of forb in rodent diets for both seasons. PPM grass consumption increased from spring (0) to summer (0.1) and the proportion of shrub in the diets of PPM remained consistently low for spring (0) and summer (0.01) (Figure 13 & 14).

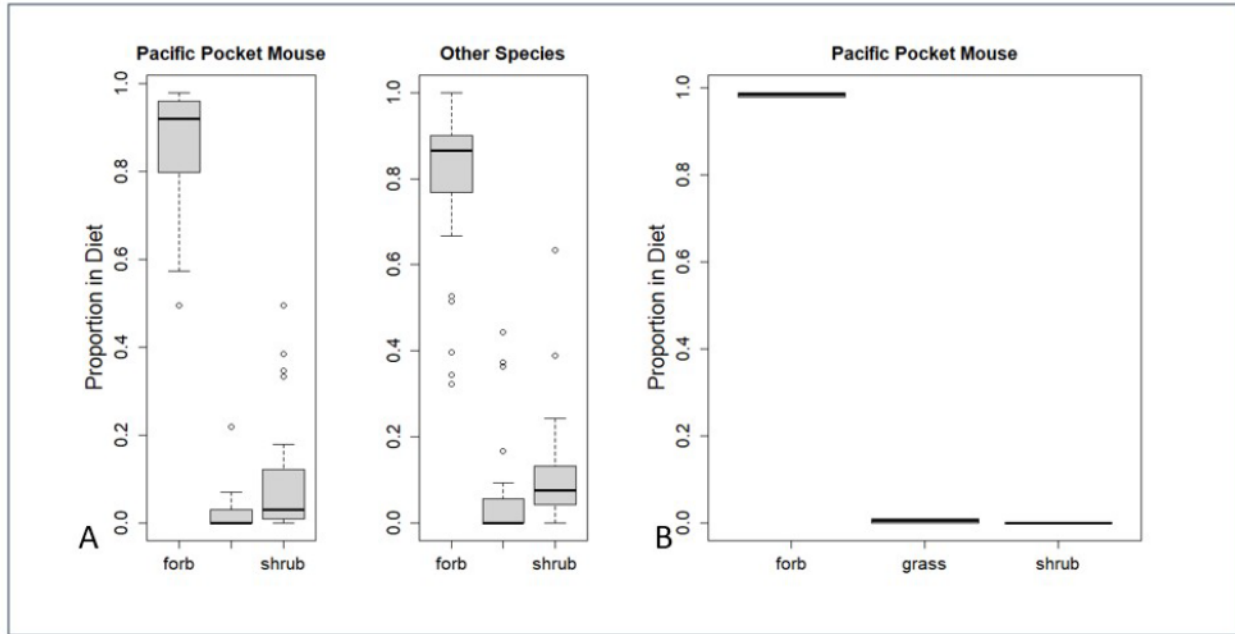


Figure 13. Functional group proportion for spring at A) South San Mateo and B) Santa Margarita in 2017 (only PPM data was available).

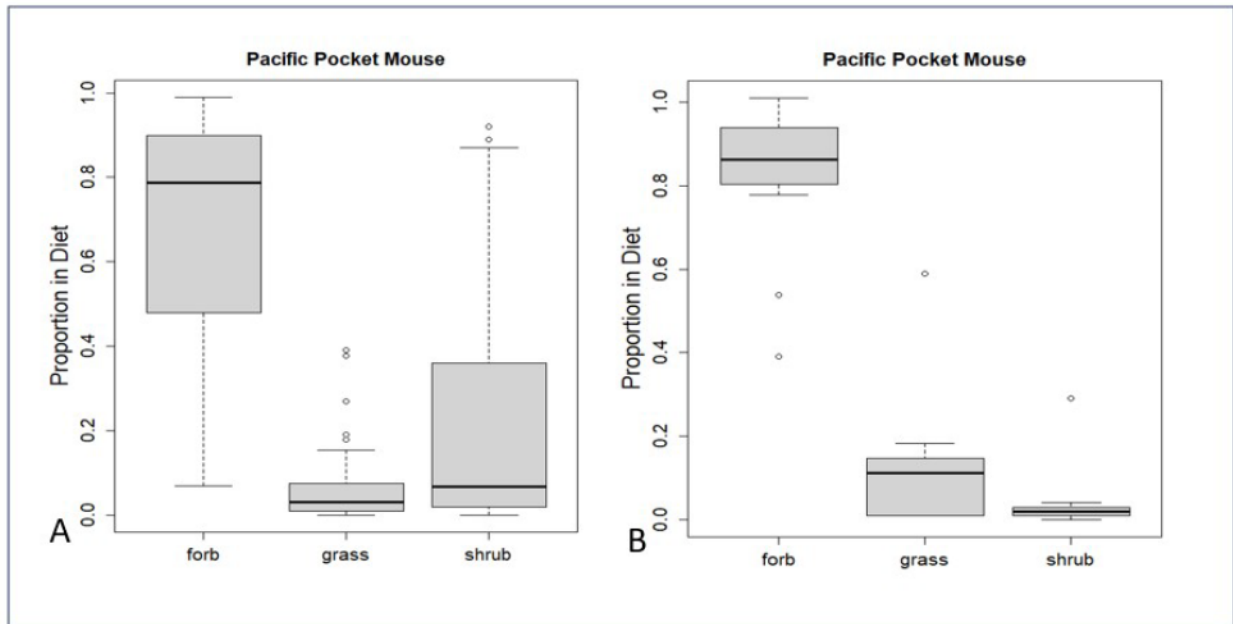


Figure 14. Functional group proportion in diet for only PPM (no other rodent data available) in summer at A) South San Mateo and B) Santa Margarita in 2017.

Diet Selectivity

Due to lack of data for PPM and other rodent species in 2017, analysis of selectivity will only be done for 2016.

Spring 2016

During spring 2016 at South San Mateo, PPM showed positive selectivity values for forbs (mean selectivity= 0.423; Figure 15). Selectivity decreased slightly as forb availability increased (0.0085 decrease per each one percent increase in forb percent cover, $t = 8.293$, $p = 0.0036$; Table 4). Selectivity did not differ between PPM and PPM competitors during spring 2016 ($p = 0.453$). PPM showed a very slight positive selectivity value for grass (mean

selectivity= 0.0053). Selectivity decreased as grass availability increased (0.0108 decrease per each one percent increase in grass percent cover, $t = -3.782$, $p = 0.00049$; table 4), and selectivity did not differ between PPM and PPM competitors during the spring season ($p = 0.092$). In the spring at South San Mateo, shrub showed a negative selectivity value for PPM (mean selectivity= -0.356). Selectivity decreased slightly as shrub availability increased (0.00812 decrease per each one percent increase in fob percent cover, $t = -3.552$, $p = 0.00349$; table 4). While selectivity did not differ between PPM and its competitors during the season ($p = 0.773$).

At Santa Margarita, PPM showed a positive selectivity value for forbs (mean selectivity= 0.388). Forb selectivity decreased as forb availability increased (0.0123 decrease per each one percent increase in fob percent cover, $t = -4.508$, $p = 0.0055$ Table 5). Selectivity values for forbs were greater in PPM compared to other rodents by 0.119 ($p = 0.0336$). PPM selectivity values for grass were slightly positive (mean selectivity= 0.0078). Selectivity decreased as grass availability increased (0.0181 decrease per each one percent increase in grass percent cover, $t = -2.533$ $p = 0.0174$; Table 5). Selectivity did not differ between PPM and PPM competitors for grass during spring 2016 ($p = 0.244$). PPM had a negative selectivity value for shrubs during spring 2016 (mean selectivity= -0.356). Selectivity for shrubs decreased as shrub availability increased (0.00571 decrease per each one percent increase in shrub percent cover, $t = -0.68$, $p = 0.137$; Table 4). There was no difference in selectivity for shrubs between PPM and its competitors ($p = 0.773$).

Table 4. Selectivity at both populations for functional groups in spring 2016.

		estimate	t-value	p-value
South San Mateo 2016	Forb Selectivity	-0.0085	8.293	0.00364
	Grass Selectivity	-0.0108	-3.782	0.000486
	Shrub Selectivity	-0.00812	-3.552	0.00349
Santa Margarita 2016	Forb Selectivity	-0.013	-4.508	0.005508
	Grass Selectivity	-0.0181	-2.533	0.01744
	Shrub Selectivity	-0.022	-0.68	0.5023

Summer 2016

PPM showed a positive selectivity value for forbs during summer of 2016 (mean selectivity = 0.396). Forb selectivity decreased slightly as its availability increased (0.0093 decrease per each one percent increase in forb percent cover, $p = 0.0140$; Table 5). Forb selectivity values in PPM competitors were greater compared to PPM by 0.123 ($p = 0.034$). PPM exhibited a slight positive selectivity value for grass at South San Mateo (mean selectivity = 0.00086). PPM grass selectivity decreased as grass percent cover increased (0.0098 decrease per each one percent increase in grass percent cover, $t = -2.214$, $p = 0.0305$; Table 5). There was no difference in selectivity values between PPM and rodent competitors for grass at South San Mateo ($p = 0.8012$). PPM had a negative selectivity value for shrubs during the summer of 2016 (mean selectivity = -0.242). PPM shrub selectivity decreased as its availability increased (0.0090 decrease per each one percent increase in shrub percent cover, $t = -2.79$, $p = 0.011$; Table 5). Selectivity for shrubs was less in PPM compared to other rodents by 0.119 ($p = 0.0235$).

During the summer at Santa Margarita PPM exhibited a positive selectivity value for forbs (mean selectivity = 0.368). PPM forb selectivity decreased as its availability increased (0.0103 decrease per each one percent increase in forb percent cover, $t = -3.127$, $p = 0.0068$; Table 5). There was no difference in forb selectivity between PPM and its competitors ($p = 0.452$). PPM showed a negative selectivity value for grass during the summer (mean selectivity = -

0.0546). PPM grass selectivity decreased as grass availability increased (0.057 decrease per each one percent increase in forb percent cover, $t = -1.52$, $p = 0.137$; Table 5). There was no difference in grass selectivity between PPM and competitor species ($p = 0.092$). For shrub in the summer PPM showed a slightly positive selectivity value (mean selectivity = 0.0682). Shrub selectivity increased for PPM as shrub availability increase (0.00328 increase per each one percent increase in forb percent cover, $t = 0.054$, $p = 0.957$; Table 5).

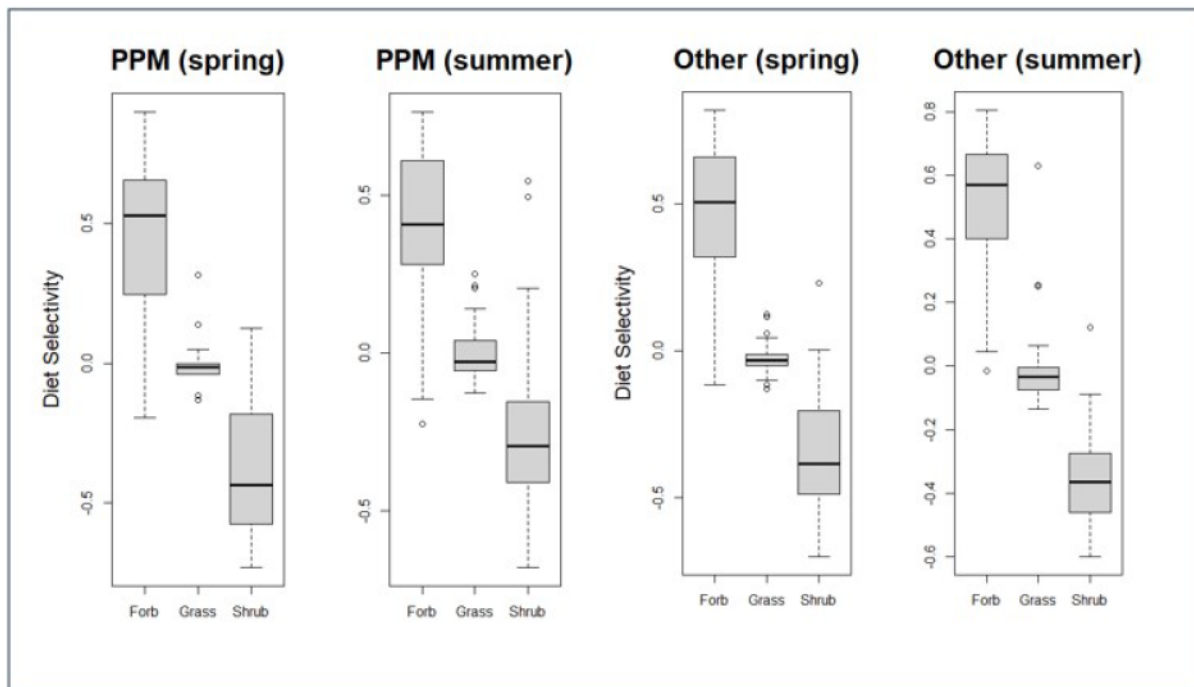


Figure 15. Diet selectivity of PPM and other rodent species at South San Mateo in the spring and summer of 2016.

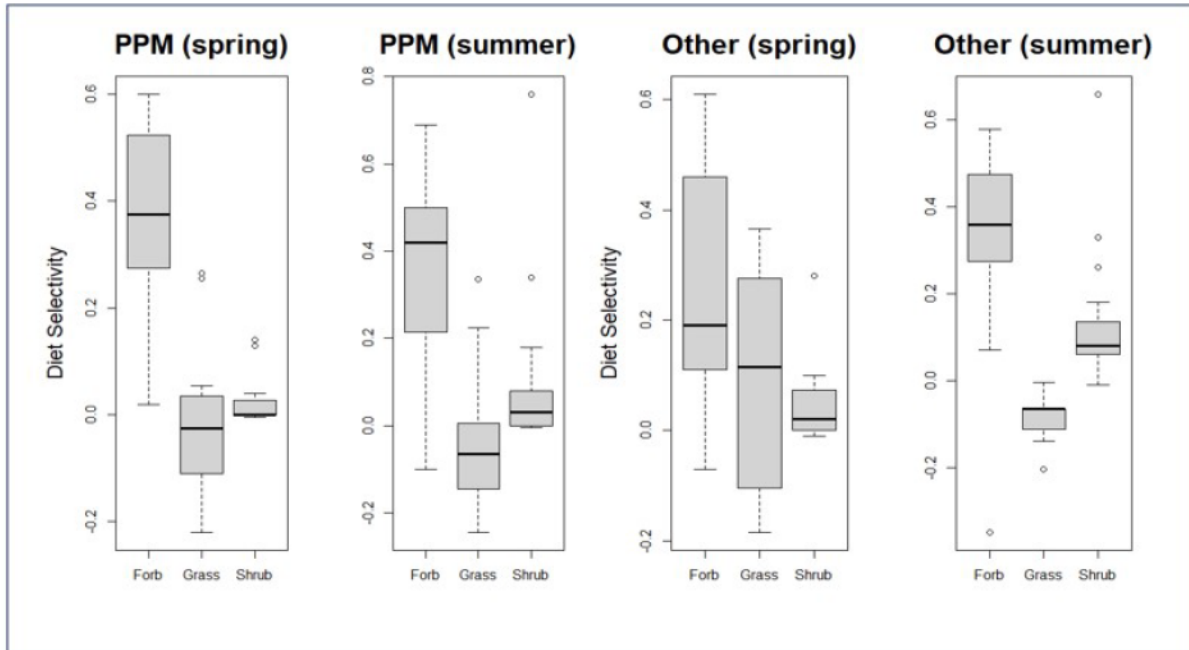


Figure 16. Diet selectivity of PPM and other rodent species at Santa Margarita in the spring and summer of 2016.

Table 5. Selectivity at both populations for functional groups in summer 2016.

		estimate	t-value	p-value
South San Mateo 2016	Forb Selectivity	-0.0093	-2.822	0.014
	Grass Selectivity	-0.0098	-2.214	0.0305
	Shrub Selectivity	-0.00904	-2.79	0.0109
Santa Margarita 2016	Forb Selectivity	-0.0103	-3.127	0.00683
	Grass Selectivity	-0.00571	-1.516	0.137
	Shrub Selectivity	0.00328	0.054	0.957

CHAPTER FOUR

DISCUSSION

This study is one of a very few studies that identify the diet functional group preferences of PPM, which compares PPM diet data to available resources across spatial and temporal scales. Past studies have used outdated methods to identify PPM diet alone, but not in conjunction with currently available plant resources or co-occurring rodent species diets. This study used both DNA metabarcoding and vegetation surveys in PPM occupied habitat to identify the proportions of functional types in diets for PPM and co-occurring rodent species. As Iwanowicz et al. (2016) summarized: “Metabarcoding effectively identified food sources of an endangered, elusive animal, using minute inputs gathered with minimal harm. Metabarcoding had higher resolution than microscopy and potentially lower observer bias than microscopy, based on comparisons to early work that sacrificed animals to directly examine pouch or gut contents. The data can inform further conservation and restoration of PPM through its application in a wide array of resource-selection studies and can help to better characterize the relationship between habitat/resource availability and PPM demography.” This tool along with plant phenology surveys allows for collection of a robust data set to compare PPM and co-occurring rodent diet composition, to better understand the resource needs of the local rodent community.

Diet Availability

Overall, we found that plant availability varied between populations but was consistent within each population across seasons and years. South San Mateo had more shrub cover compared to Santa Margarita, while Santa Margarita had more forb and grass cover except for spring 2017 when grass was less available at Santa Margarita than South San Mateo. This difference in cover type can have implications on PPM diet composition and overall habitat suitability. Based on a multi-year occupancy model, PPM preferred medium to high open ground and forb cover and avoided areas with high non-native grass cover (Brehme et al. 2016). The observed functional cover type percentages at Santa Margarita appear to present more favorable habitat conditions with high forb cover and lower shrub cover, although the grass cover could be detrimental. South San Mateo exhibits less desirable habitat and resources for PPM because of its extremely high shrub cover, and therefore less forb and grass availability.

A major impact in cover type and diet availability in PPM habitat is habitat management or potential implications of wildfire. Santa Margarita has experienced a semi-occasional fire regime, with wildfires in 2011, 2012 and 2014 (Brehme et al. 2020). These wildfires remove shrub cover and open the habitat for forb growth. A more natural fire pattern could consistently produce more favorable habitat with less shrub cover and more forbs for PPM diet needs (Harper, 2007). South San Mateo has not had a fire in the last 20 years creating a more mature coastal sage scrub habitat, with higher densities of shrub cover. This shrub density can limit non-native grass and forb cover and therefore resources available for PPM consumption, creating less diverse resource opportunities for PPM and competitors.

Diet Composition

We found that the forb functional group was by far the most consumed functional group at both populations, across seasons and years, and across all species. This finding suggests that forb species are the primary food resource for PPM and its competitor species in their local communities. Santa Margarita is comprised primarily of forb cover, which correlates with PPM and other rodents propensities to consume those species at a higher proportion than shrub and grass. In 2016, PPM showed a reduced proportion of forb consumed from spring to summer, with a concurrent increase in shrub consumption. This reduction in forb consumed could be due to forb growth patterns. Many forb species in these study locations are annuals that generally die off as the season gets warmer and time moves further from the last rainfall event. As forb cover decreases, PPM may expand their foraging options to less desirable plant functional groups.

South San Mateo has higher shrub cover compared to Santa Margarita, however the PPM and other rodent species still primarily consumed forbs, while integrating shrubs in their diets at a higher rate than species at Santa Margarita. Since South San Mateo is shrub dominate and has less forb cover, this could cause the rodent community to have to rely more heavily on less preferred shrub species. This shrub reliance is especially true as forbs die off into the summer season, as evidenced by the increase in the proportion in diet of shrub from spring to summer in both 2016 and 2017.

Diet Selectivity

We found that PPM preferentially selected for forbs in their diet across seasons and years in both populations, evidenced by positive selectivity values (i.e., forbs represented a greater proportion in observed diets compared to the proportion available in the habitat). Higher forb cover is a primary predictor of increased probabilities of local PPM occupancy and colonization (Brehme et al., 2018, 2020), which corresponds to their high propensity to select for forbs. At South San Mateo, PPM were selecting for forbs in both the spring and summer seasons, but their selectivity decreased as forb cover increased. The primary difference in PPM selectivity at South San Mateo between seasons was that rodent competition selectivity increased during the summer. The local rodent competitors are all larger in size than PPM which could account for their slight increase in selectivity for those resources, especially during the summer season as forb resources diminish.

Higher non-native grass cover has been a primary predictor of decreased probability of PPM occupancy and increased probability of localized extinction across sites (Brehme et al., In Review). Large contiguous stands of annual grasses impede the growth of other plant species (Flory & Clay, 2010) and may hinder PPM movement and foraging success. Accordingly, it was found at both populations that PPM only slightly selected for grass during both seasons and expressed a decrease in selectivity as grass cover increased. PPM avoided shrubs relative to shrub availability (i.e., showed negative selectivity values) at South San Mateo during both the spring and summer seasons. At South San Mateo as shrub cover increased, PPM selectivity decreased during both seasons, while PPM selected shrub more than its rodent competitors. The

similarities of PPM selectivity for grass and shrubs during both seasons suggests they prefer forb resources and prioritize their availability over the other functional groups. Since South San Mateo is predominantly shrubs and forbs that are early season annuals, during the summer months PPM forb resources decrease so they start to select shrub as an alternative resource over grass. These results can help inform land managers what types of plant species to prioritize for enhancing the habitat of currently PPM populations and future reintroduction sites. It must be noted that climate change, weather, and competitor densities can all impact PPM resource selection and have not been looked at in this study.

Management Implications

The data collected and analyzed in this study will inform land managers how to better manage PPM habitat by understanding PPM diet selectivity and preferences. This information can help land managers reach the recovery goals put forth by the USFWS of establishing 10 resilient populations of PPM in their historical range (USFWS 1998). In order to establish 10 resilient populations, ecosystems will need to be selected based on many factors, such as, historical occupancy, soil type, potential predator densities, and plant species on site to ensure the success of PPM survival: “To maintain their full potential, these ecosystems as a whole must be managed in a way which will support and complement full ecosystem functioning to the extent currently possible. Occupied habitat, potential habitat, and surrounding linkages should be secured and protected so that the ecosystem upon which the Pacific pocket mouse depends is conserved (USFWS 1998)”. Enhancing PPM habitat can be done by informing managers whether future potential reintroduction sites need habitat mitigation, such as: full habitat

restoration; clearance of non-consumed grasses and shrubs to enhance habitat suitability; or establishment of additional plant species to sustain a new population. Since I found that PPM prefer forbs regardless of other functional group cover, for managers who have large densities of shrubs then thinning would be an ideal management action. Thinning shrub cover creates more open space for forb growth which will enhance the habitat for PPM.

Understanding the historical range and resource selection of PPM will establish a restoration/reintroduction habitat minimum to ensure PPM are placed into the best habitat recovery site possible. The USFWS recommends as a recovery action to enhance and expand Pacific Pocket mouse habitat: “regulatory agencies should prepare, or direct the preparation of, a habitat management plan component that provides for optimization of Pacific pocket mouse habitat at managed sites. Techniques that could be employed include revegetation (with or without salvaged materials such as topsoil) and selective habitat enhancement, including experimenting with fire. By confirming that suitable dietary plant species currently occupy a site, this can quicken the recovery efforts by eliminating the need for major landscape changes on potential reintroduction sites.

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