Ear Length as a Diagnostic Character for Identifying Species of Kangaroo Rats

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ABSTRACT

The following study examines the use of ear length as a diagnostic field character for differentiating between species of Kangaroo rats in San Luis Obispo County. Data was taken from a study of Kangaroo rats in the Guadalupe-Nipomo Dunes National Wildlife Refuge, as well as various collections under the supervision of Francis X. Villablanca, Ph. D. and California Polytechnic State University, San Luis Obispo. An ANOVA was used to compare subspecies within *Dipodomys heermanni* and t-tests were used to compare ear lengths between *Dipodomys heermanni* and *Dipodomys venustus*. The analysis shows a statistically significant difference and supports the hypothesis that ear length can be used to differentiate species. Further testing was completed at the subspecies level within *Dipodomys heermanni* but did not prove to be significant. This experiment is evidence that confirms the identification of *Dipodomys venustus* outside of its known range.

INTRODUCTION

Trying to identify a species in the field among species that have very similar characters can be extremely difficult. Diagnostic field characters can often be misleading and inconclusive. For example, the literature may suggest that individuals of the species in question exhibit a partial hip stripe and specific pelage coloration yet you are unable to confirm either character conclusively.

The genus *Dipodomys* is very speciose and is considered to be very compact and homogenous in comparison to other rodent families and genera (Grinnell 94-97). This makes for difficult identification in the field.

A problem exists with the known range for *Dipodomys venustus* (Narrow-faced Kangaroo Rat). There is little known about the actual range of *D. venustus* that can be found in the literature accept for what has been published by T. Best in 1992 (Figures 1 and 13). More specifically, ranges and distributions of Kangaroo rats in San Luis Obispo County are poorly known. Ear length may be a more definitive approach for differentiating between species of *Dipodomys* than other characters. An exact measurement that falls within a known range for the species is stronger evidence than a subjective character that can be easily misinterpreted. It is possible that testing the utility of ear length performed throughout the experiment will confirm the presence on *D. venustus* several miles from its known or hypothesized range.



Figure 1: The known extant range of *D. venustus* map taken from IUCN website extending from San Francisco to San Luis Obispo County, according to Best (1992).

Further testing may provide significant evidence that the same technique is useful in identifying subspecies. The reasoning for this is that historical literature shows great variability among sampled and measured *D. heermanni*, suggesting a range in ear length of 10mm to 17mm within the species (Grinnell 94-97) and it is possible that subspecies do not have overlapping values. Identifying external differences at subspecies levels is even more cryptic and often requires knowing geographic distributions of the populations, endemism ranges, or may require DNA testing. For example, *Dipodomys heermanni arenae* (Lompoc Kangaroo rat) is known to exhibit an incomplete hip-stripe in about 5% of its population, whereas *Dipodomys heermanni morroensis* (Morro Bay Kangaroo rat) exhibits this same character in 75% of its population (A. I. Roest). Given this scenario, it may be plausible that one subspecies be mistaken for another if all other characters are similar or are not easily determined. As seen in Figure 2,

their geographic ranges are adjacent to each other and may possibly overlap. Mistaking one for the other may mean the difference between identifying and working with a healthy population of animals exemplified by the former subspecies, when in fact the population is greatly endangered or potentially extinct as in the latter.



Figure 2: Distribution of D. heermanni in California as described by Hall (1981) and A. I. Roest (in litt.). 1: D. h. arenae; 2: D. h. berkeleyensis; 3: D. h. dixoni; 4: D. h. goldmani; 5: D. h. heermanni; 6: D. h. jolonensis; 7: D. h. morroensis; 8: D. h. swarthi; D. h. tularensis. (Kelt 1988)

The following explains the methods and results of an experiment carried out to test the hypothesis that measurements taken from the ear notch to the longest point on the ear can be used to differentiate between *D. h. arenae* and *D. venustus*. Further testing may show a significant difference in ear length between subspecies of *D. heermanni*, allowing for a more definitive approach for identifying to a subspecies level. Our sample was inconclusive at the subspecies level.

METHODS

Ear length data was collected from multiple locations throughout central California. The most recent data set of *D. h. arenae* was acquired from the Guadalupe-Nipomo Dunes National Wildlife Refuge (GNDNWR), a few miles north-west of Guadalupe, California (Table 2). Data was collected as part of an ongoing, long term demographic study of small mammals directed by Francis X. Villablanca, Ph. D. The twelve hundred meter sampling transect (Figure 5) was established for sampling and has been returned to quarterly over the duration of the four year project (see figure 3 and 4 for map of project boundary and location).



a few miles

es located a few 2, CA.



Figure 5: Sample transect line located in GNDNWR, CA.

Mammals were caught in Sherman extra-long livetraps that were placed along the transect every twenty meters. Two traps were set at dusk within a two meter radius of each interval for three straight nights. There were 62 stations; two traps per station over a total of three nights equaling 372 trap-nights. Old fashioned rolled oats were used as bait for the traps and sand was used to cover the outer portion of the traps to provide insulation for any animal caught while also providing camouflage to avoid potential trap shyness.

Traps were then checked around dawn the following morning as to avoid mortality and any mammal caught was emptied into a bag, identified, and sexed. Further demographic information was taken from species of interest (ex. Kangaroo rat, Deer mouse (*Peromyscus maniculatus*)) in order to fulfill research guidelines. These measurements included weight taken with a pesola, age determination, ear measurements for all Kangaroo rats, reproductive status for all Kangaroo rats, and marking with an ear tag or permanent marker if determined to be a new capture. Animal identification by ear tags was also used to average measurements made on the same individual over the course of the experiment (Table2). All information was recorded on a standardized form as shown in Table 1. Animals were released at the site of capture after desired information was gathered.

Table 1: Standardized field form for field data collection at GNDNWR.

Date:			Names:						[]
Site: Gu	adalupe-l	Nipomo Dunes I	National Wildlife Refu	ıge - i	f other e	nter:			
Station	Species	Tag# and ear*	Side marked today@	New	Recap^	Sex	Breed. Cond	Age Notes	Weight (gm)

The second source of ear length data was taken from *D. venustus* data collected at Hi Mountain the previous year (Table 2). Measurements were taken using the same technique as were used in GNDNWR. The final ear length dataset was collected off of tags from previously measured specimens located in the Aryan Roest Vertebrate Zoology Collection at Cal Poly (Table 2).

Before any statistical analysis could be performed, the dataset needed to go through a process of sorting and editing to clean up and remove any duplicate or erroneous data. Any specimen that did not have ear measurements or ones which had ear measurements taken from the crown instead of the notch were removed from the dataset. Because there were less than five sampled specimens identified as being sub adult aged, only those considered adult aged were included in the analysis. Adequate testing could not be performed in order to determine if there is a difference between adult and sub adult ear length because of the small sample size of sub adults.

Species	Number	Location	Sex	Ear
dha	18	GNDNWR	f	15
dha	63	GNDNWR	f	15
dha	68	GNDNWR	m	15
dha	74	GNDNWR	m	14.5
dha	80	GNDNWR	m	15
dha	82	GNDNWR	m	15
dha	88	GNDNWR	f	15.5
dha	89	GNDNWR	m	15
dha	110	GNDNWR	m	15.5
dha	113	GNDNWR	m	16
dha	115	GNDNWR	f	14
dha	120	GNDNWR	m	15.75
dha	124	GNDNWR	m	15

Kangaroo rat ear measurements.

Table 2: Sorted data set of GNDNWR, Aryan Roest Vertebrate Zoology Collection, and Hi Mountain

dha	131	GNDNWR	f	16
dha	168	GNDNWR	m	15.5
dha	205	GNDNWR	f	15
dha	209	GNDNWR	m	15
dha	230	GNDNWR	f	15
dha	237	GNDNWR	m	15
dha	239	GNDNWR	m	14.5
dha	242	GNDNWR	m	14.75
dha	244	GNDNWR	m	15.5
dha	263	GNDNWR	m	16
dha	267	GNDNWR	m	16.5
dha	273	GNDNWR	f	16
dha	280	GNDNWR	m	15
dha	284	GNDNWR	m	17
dha	286	GNDNWR	m	16
dha	288	GNDNWR	f	15
dha	295	GNDNWR	f	14.5
dha	303	GNDNWR	m	15
dha	304	GNDNWR	f	15
dha	327	GNDNWR	m	15.5
dha	331	GNDNWR	m	15.5
dha	350	GNDNWR	f	15.5
dha	360	GNDNWR	f	16.5
dha	361	GNDNWR	m	16
dha	365	GNDNWR	f	16
dha	371	GNDNWR	m	13
dha	373	GNDNWR	m	13
dha	376	GNDNWR	m	13
dha	380	GNDNWR	f	15
dha	401	GNDNWR	f	15.5
dha	405	GNDNWR	m	15.5

dha	408	GNDNWR	m	15
dha	409	GNDNWR	m	16
dha	410	GNDNWR	m	15
dha	413	GNDNWR	f	15.5
dha	426	GNDNWR	f	15
dha	451	GNDNWR	f	15
dha	456	GNDNWR	m	14.5
dha	458	GNDNWR	m	14.5
dha	459	GNDNWR	m	14
dha	463	GNDNWR	m	15
dha	464	GNDNWR	f	15
dha	474	GNDNWR	f	16.5
dha	498	GNDNWR	f	15
dha	681	GNDNWR	f	15
dha	791	GNDNWR	f	16
dha	865	GNDNWR	f	15
dha	892	GNDNWR	m	16.5
dha	958	GNDNWR	m	15
dha	971	GNDNWR	f	16
dha	974	GNDNWR	f	13
dha	992	GNDNWR	m	15.5
dha	3001	GNDNWR	f	15
dha	3002	GNDNWR	f	14.5
dha	3003	GNDNWR	f	14
dha	3004	GNDNWR	f	15
dha	3005	GNDNWR	m	14.5
dha	3006	GNDNWR	f	14
dha	3007	GNDNWR	f	17
dha	3026	GNDNWR	f	15
dha	3028	GNDNWR	m	15
dha	3051	GNDNWR	m	15

dha	3052	GNDNWR	m	15
dha	3053	GNDNWR	m	15
dha	683 R	High Mountain	m	16
dha	m1473	arroyo grande	m	11
dha	m1721	7 mi west of nipomo	m	17
dha	m2649	dunes lake, slo county	f	14
dha	m2739	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	f	15
dha	m2740	huasna river at huasna road	f	14
dha	m2741	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	m	14
dha	m2742	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	m	16
dha	m2743	huasna river at huasna road	m	16
dha	m2745	guadalupe dunes at oso flaco lake rd .5 m s oso flaco	m	14
dha	m2748	guadalupe dunes at oso flaco lake rd .5 m s oso flaco	m	13
dha	m2749	santa maria river at hwy 1	f	15
dha	m2751	santa maria river at hwy 1	f	14
dha	m2752	santa maria river at hwy 1	f	15
dha	m2756	santa maria river at hwy 1	f	14
dha	m2757	santa maria river at hwy 1		15
dha	m2758	santa maria river at hwy 1	m	14
dha	m2759	santa maria river at hwy 1	f	15
dha	m2760	santa maria river at hwy 1	m	15
dha	m2762	huasna river at huasna road	m	15
dha	m2763	huasna river at huasna road	f	14
dha	m2764	huasna river at huasna road	f	16
dha	m2765	huasna river at huasna road	f	16
dha	m2766	huasna river at huasna road	m	15
dha	m2767	huasna river at huasna road	f	16
dha	m2768	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	f	15
dha	m2769	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	m	15
dha	m2770	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	m	15
dha	m2777	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	f	15

dha	m2778	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	m	15
dha	m2779	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	m	15
dha	m2780	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	f	15
dha	m2782	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	f	14
dha	m2783	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	m	15.5
dha	m2784	nipomo mesa 5.4 mi S 3.5 mi E oceano airport	m	15
dha	m402	dune lakes, 2 mi s oceano	f	15
dha	m403	1 mile west of edna	m	16
dha	m404	dune lakes, 2 mi s oceano	m	15
dha	m405	1 mile west of edna	f	16.5
dha	m406	dune lakes, 2 mi s oceano	f	14
dha	m412	1 mile west of edna	f	16
dha	m413	8.5 miles NE arroyo grande, mouth of pheonix river	f	16.5
dha	m414	1 mile west of edna	f	16.5
dha	m417	2 mi west nipomo	m	17
dha	m419	1 mile west of edna	f	15.5
dha	m420	1 mile west of edna		17
dha	m421	1 mile west of edna		16.5
dha	m435	1 mile west of edna		15
dha	m438	1 mile west of edna	f	12
dha	m444	1 mile west of edna	m	17
dha	m469	dune lakes, 2 mi s oceano	m	17
dha	m53	arroyo grande	m	11
dhj	m2478	creston	m	16
dhj	m2539	creston	f	17
dhj	m2746	big sandy creek at salinas river	f	15
dhj	m2747	big sandy creek at salinas river	f	16
dhj	m2750	big sandy creek at salinas river	m	15
dhj	m2753	big sandy creek at salinas river	f	16
dhj	m2754	big sandy creek at salinas river	f	14
dhj	m2755	big sandy creek at salinas river	f	16

dhj	m2785	big sandy creek at salinas river		16
dhj	m387	monterey co. 4 mi se bradley		14.5
dhj	m870	king city	m	14
dhm	m1408	irish hills	f	17.5
dhm	m1723	CPSU union oil stock	f	15
dhm	m1725	los osos e end of santa ysabel	m	13
dhm	m2123	baywood heights	m	14
dhm	m2479	.5 mi s palisades dr	f	15
dhm	m2486	jr high site, baywood	m	15
dhm	m2487	bayview site	f	17
dhm	m2544	CPSU	f	18
dhm	m774	baywood park	f	12
dhs	m1660	carrizo plain	f	12
dhs	m1662	soda lake rd 2 mi nw simmler	m	10.4
dhs	m1663	soda lake rd 2 mi nw simmler	m	16
dhs	m1724	north end of soda lake	f	11
dhs	m2142	carrizo plain, 2 mi s. crocker grade		17
dhs	m2145	carrizo plain, foot of crocker grade		18
dhs	m2280	elkhorn plains, panorama hills	m	16.9
dhs	m2290	soda lake rd	f	15
dhs	m2293	elkhorn rd		13
dhs	m2294	elkhorn rd	m	17
dhs	m2295	elkhorn plains, panorama hills	m	16
dhs	m2296	elkhorn plains, panorama hills	m	15
dhs	m2761	crest of hwy 58, temblor range	f	15
dhs	m2771	crest of hwy 58, temblor range	f	16
dhs	m2772	crest of hwy 58, temblor range	f	16
dhs	m2773	crest of hwy 58, temblor range	f	16
dhs	m2774	crest of hwy 58, temblor range	f	16
dhs	m2775	crest of hwy 58, temblor range	m	17
dhs	m2776	crest of hwy 58, temblor range	f	14

dhs	m467	slo co. 1.75 mi e. cuyama	f	17.5
dhs	m847	ventura co., cuyama river, 2 mi nw ozena guard sta	m	20
dhs	XXXXX	4 mi north soda lake	f	15.5
dv	m2744	hi mnt rd 2.5 m s pozo	m	17
dv	695 L	High Mountain		20
dv	714 L	High Mountain		16
dv	714 L	High Mountain		18
dv	715 L	High Mountain		17
dv	715 L	High Mountain		18
dv	930 R	High Mountain		18
dv	930 R	High Mountain		19
dv	930 R	High Mountain		19
dv	935 L	High Mountain		19
dv	935 L	High Mountain		19
dv	938 L	High Mountain		18
dv	938 L	High Mountain		18
dv	944 R	High Mountain		19
dv	944 R	High Mountain		19

The next step in the data sorting process involved averaging repeated measurements taken from the same sampled individual over the course of the studies in GNDNWR and High Mountain Lookout. Variability taken from repeated measurements showed a range of 0mm to 3mm with an abnormal range of 10 to 18mm recorded on individual 157. This data was removed from the analysis because of obvious error in sampling. Multiple tests were then carried out to determine the usefulness of comparing ear lengths.

Before comparing any data sets to each other, they were all run through a diagnostic in "R" using the Kolmogorov-Smirnov test for equal variances and normality. If the data set fulfilled the requirements of normality, it was run through a normal t-test, and if not, it was run through a Welch's t-test which assumes unequal variances. The first test involved comparing the data sets collected by students from GNDNWR and High Mountain Lookout, to the stuffed specimens that were measured by Dr. Villablanca and his colleagues which were stored in the Aryan Roest Vertebrate Zoology Collection. *D. h. arenae* was specifically chosen for this test because of its high amount of representatives from

GNDNWR and the mammalogy specimen collection at Cal Poly. The next step included comparing sex within *D. h. arenae* to see if there was any dimorphism between male and females. According to T. L. Best, significant sexual dimorphism was found in all external measurements of D. heermanni with large sample sizes of 474 adult males and 355 adult females (Best, In press). Being that this experiment does not have such large sample sizes, sexual dimorphism may not be identified. If no difference is found, sex of Kangaroo rats will not be included in the comparison of species to species and the data for both sexes will be pooled. Testing of multiple subspecies will be run through an ANOVA test and if it passes with significance, a mean separation will be completed in order to determine the differences in the means.

The independent variable in the experiment is a qualitative, nominal variable because the different species (*D. heermanni*, *D. venustus*, and *D. heermanni* ssp.) are being measured and compared. This grouping can also be considered the treatment of the experiment because it potentially explains differences in the response variable. The dependent variable (or response variable) is ear length, measured in mm. This is the variable we measure in order to identify a significant difference between the two species.

RESULTS

The initial test, comparing *D. heermanni arenae* from the student study at GNDNWR to the Aryan Roest Vertebrate Zoology Collection at Cal Poly showed normal distribution graphs. Sample sizes for the GNDNWR and the Aryan Roest data sets are 77 and 52 respectively with their normality distributions shown in Figures 6 and 7.



The results of the F-test show a p-value of 0.05395 and the Kolmogorov-Smirnov test shows a pvalue of 0.9981. Because of these results, we use a Welch's Two Sample t-test assuming unequal variances. Results of the Welch test show a p-value of 0.9033. Therefore, the comparison is not statistically significant. The hypothesis that the two sample sets are different cannot be confirmed. There is no significant difference between samples taken by students at GNDNWR and those taken from the Aryan Roest Vertebrate Zoology Collection.

The following shows the results of the test for sexual dimorphism between male and female *D. heermanni*, after combining the data from GNDNWR and the Aryan Roest Collection. Sample sizes for male and female D. heermanni are 70 and 59 respectively. Normality distributions are shown in Figures 8 and 9.



The p-value for the F test is borderline at 0.05395, meaning that the true variances may or may not have a ratio equal to one. The results of the Kolmogorov-Smirnov test show an insignificant p-value of 0.9981. Results of this test show that the data sets should be analyzed using a Welch's Two Sample ttest assuming unequal variances. Results of the Welch's test show a p-value of 0.9033, with sample mean estimates for males being 15.107 and females 15.085. Therefore, the null hypothesis that sexual dimorphism does not exist cannot be rejected by this test. There is no significant difference between measured male and female ear lengths. Results of this test allow for the pooling of sex data and it can be concluded that sexual dimorphism was not captured in the samples taken.

An ANOVA was then performed on the data collected from different subspecies of *D*. *heermanni*. These species include *D*. *h*. *arenae* (DHA) with a sample size of 129, *D*. *h*. *jolonensis* (DHJ) with a sample size of 11, *D*. *h*. *morroensisl* (DHM) with a sample size of 9, and *D*. *h*. *swarthi* (DHS) with a sample size of 22. Results from the ANOVA are shown with summary statistics in Table 3 and critical values in Table 4.

Table 3: Descriptive statistics for the *D. heermani* subspecies test.

Groups	Count	Sum	Average	Variance
DHA	129	1947.5	15.09689922	1.120419998
DHJ	11	169.5	15.40909091	0.940909091
DHM	9	136.5	15.16666667	4.125
DHS	22	340.3	15.46818182	5.164177489

Table 4: ANOVA results for subspecies analysis of D. heermanni.

Source of						
Variation	SS	df	MS	F	P-value	F crit
Between						
Groups	3.273895812	3	1.091298604	0.619317324	0.603425745	2.658723278
Within Groups	294.2705779	167	1.762099269			
Total	297.5444737	170				

Results show an insignificant p-value of 0.603, meaning that there is not a detected difference between subspecies groups. Therefore, we reject the hypothesis that *D. heermanni* subspecies can be differentiated by ear length alone.

The next step is to combine all of the *D. heermanni* data into one data set and compare it to the *D. venustus* data taken from the Hi Mountain sample. Sample sizes for the pooled *D. heermanni* dataset and *D. venustus* data sets are 171 and 16 respectively. Normality distributions are shown in Figures 10 and 11.



The results of the F-test provide a p-value of 0.9881, meaning the variance ratio is 1 to 1. The Two-Sample Kolmogorov-Smirnov test shows a significant p-value of 1.912e-09. Welch's Two Sample t-test is significant with a p-value of 1.898e-09. *D. heermanni* has a mean of 15.17 with a 95% confidence interval of [14.97, 15.37], while *D. venustus* has a mean of 18.27 with a 95% confidence interval of [17.70, 18.84]. Figure 12 shows the box and whisker plots for *D. heermanni* and *D. venustus* with obvious mean differences.



Figure 12: Box and whisker plot of sampled ear length distribution with *D*. *heermanni* on left and *D. venustus* on the right.

Discussion

Results of this study show that there is no significant sexual dimorphism detected from ear measurements samples taken from D. h. arenae. Because this is the largest sample size of *D. h.* ssp. in the experiment, it can be assumed that there would be no difference detected within each subspecies of *D. heermanni*. Although dimorphism has been suggested in the literature, the sample size is most likely too small to produce a significant difference.

There was also no significant difference found within *D. heermanni* subspecies. Although the literature suggests a large variation within *D. heermanni*, significant differences among subspecies may not actually exist. Because of this, accurate range information is the best indicator of *D. h.* ssp. distributions as well as genetic analysis.

Significant differences were seen between measured ear length of the pooled *D. heermanni* and *D. venustus* data. These results provide striking evidence that support the hypothesis that the *D. venustus* distribution is poorly known because of the location of the sampled *D. venustus* being outside

of that range (Figure 13). Further use of ear length as a diagnostic character for species identification may be useful for determining a more accurate range of *D. venustus* as well as other species of Kangaroo rats.



Figure 13: The southernmost known range of D. venustus, San Luis Obispo County, CA.

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