

Fine-scale genetic structure in (*Spermophilus beldingi*), a sexually promiscuous ground squirrel in the High Sierra

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Abstract

Spermophilus beldingi (Belding's ground squirrel) is a social montane rodent. The Tioga Pass Meadow (Mono Co. CA) population has been studied for several decades, but little is known about the fine scale genetic structure of the population. Maternity was deduced by observing both female burrow usage during gestation and the first emergence of offspring from their natal burrows, but paternity can only be inferred by mating, and multiple males may mate with a given female in one season. Polymerase chain reaction (PCR) was utilized on eight microsatellite loci to elucidate fine scale genetic structure. Microsatellites were used to verify maternity and determine paternity, thus determining sibship within litters, which was correlated with previously collected behavioral data. These findings will allow for a better understanding of the population in this area and how kinship affects behavior between full and half siblings both as pups and adults.

Introduction

Belding's ground squirrel (Fig. 1) is a social rodent that lives in the Sierra Nevada. The population at Tioga Pass Meadow has been studied behaviorally and demographically for four decades (Sherman and Morton 1984, Nunes et al. 2004), but knowledge of kinship and population structure has been limited to observational studies. Using microsatellites, we examined genetic relatedness among squirrels from two sites to determine the genetic diversity and gene flow within this population, components critical to the overall fitness of the population. Because *S. beldingi* exhibits promiscuity in both sexes, multiply sired litters are common (Hanken and Sherman 1981). We investigated paternity and sibship in these squirrels.

Objectives:

- Determine the population structure and investigate its genetic diversity.
- Investigate parentage patterns and sibling interaction on a genetic level.



Fig.1 A female squirrel exhibiting posting behavior next to a live trap baited with peanut butter.

Methods

- We sampled 125 squirrels in May-July 2010, all of which were used. We used another 160 samples collected between 2002-2005 (Table 1).
- Squirrels were sampled at the Tioga Pass meadow, a field north of Tioga Lake, and further down the valley in the Aspen and Lower Lee Vining Campgrounds (Fig. 2).
- Samples consisted of a 1mm x 3mm sliver of tissue removed from the edge of one ear. The squirrels were tagged with metal ear tags and released at their point of capture (Fig. 3).
- The tissue was digested, DNA was extracted and purified, and then amplified via PCR, focusing on eight polymorphic loci (Table 2).
- Arlequin (Excoffier et al. 2005) and Colony (Jones and Wang 2009) were used to analyze fine scale genetic structure and the sibship, respectively. F_{ST} was used in lieu of R_{ST} because of deviations from the standard stepwise mutation model exhibited by our microsatellites (Balloux et al. 2000).
- Previously collected play data was compared to our sibship findings.



Fig. 2 Satellite image of the field sites in Mono Co. CA. The sites indicated in red are at approx. 3000m in elevation, while the sites indicated in yellow are at approx. 2200m. The road pictured is CA Hwy 120.

Table 1. Squirrels sampled per location and year.

	2002-2005	2010	Total
Lower Elevations	38*	13	51
Upper Elevations	122	112	234
Total	160	125	

*All of these squirrels were sampled in 2004.



Fig.3 Squirrel moments after receiving a unique ear tag marker and an ear clip.

Table 2. Microsatellite locus, repeat motif, number of alleles, and size range of alleles.

	A120 (CA)	B6 (AAC)	B12 (AAC)	B108 (AAC)	C4 (AAAG)	D4 (TAGA)	D106 (TAGA)	D108 (TAGA)
Number of alleles	13	4	4	7	15	14	4	7
Size range	39	18	12	19	56	72	12	24

Results and Discussion: Population Structure

- Limited gene flow (as indicated by the observed F_{ST} value) occurs between the Tioga meadows and the campgrounds, which are separated by a change in elevation of approximately 800m (Table 3).
- Sex-biased spatial patterns were not found (Table 3)
- Effective population size estimates and heterozygosity estimates indicate high levels of genetic diversity of the high elevation population, whereas the lower elevation population exhibited lower diversity (Tables 4 and 5).
- No significant divisions were found within Tioga Pass meadow, indicating that squirrels move freely from one end of the meadow to the other.
- Heterozygosity was overall somewhat lower than expected (Table 5).

Table 3. Differences between populations and migration rates

	elevation	sex
F_{ST}	0.06813*	0.00020
N_m	6.84*	22518.81

* indicates statistical significance at $p < 0.05$.

Table 4. Effective population size

	N_e
High Elevations	170
Low Elevations	40

Table 5. Heterozygosity of the alleles for Low and High elevations populations

	High H_e	High H_e	P-value	Low H_e	Low H_e	P-value
A120	0.77350	0.83511	0.00000	0.55769	0.61202	0.22234
B6	0.20940	0.21546	0.00573	0.42308	0.39507	0.00000
B12	0.6709	0.66787	0.02780	0.55769	0.60269	0.65705
B108	0.52991	0.53074	0.00048	0.59615	0.66038	0.00626
C4	0.89316	0.88978	0.22610	0.78846	0.80956	0.05609
D4	0.83333	0.83720	0.01855	0.67208	0.83205	0.08032
D106	0.57692	0.59302	0.00000	0.65385	0.68913	0.00707
D108	0.68803	0.71087	0.00529	0.80769	0.75299	0.01109

Results and Discussion: Parentage

- Multiple paternity was found in most litters at an average rate of 1 father per 1.35 pups in a litter.
- Males sired pups in multiple litters in a given season. Females mated with an average of 2.75 males, and males mated with an average of 2.17 females, with no significant difference ($p < 0.05$) (Fig. 4).
- Promiscuous mating results in many more half siblings than full siblings existing in a population (Fig. 5). There was no apparent relationship between full or half sibship and play tendencies (Fig. 6).

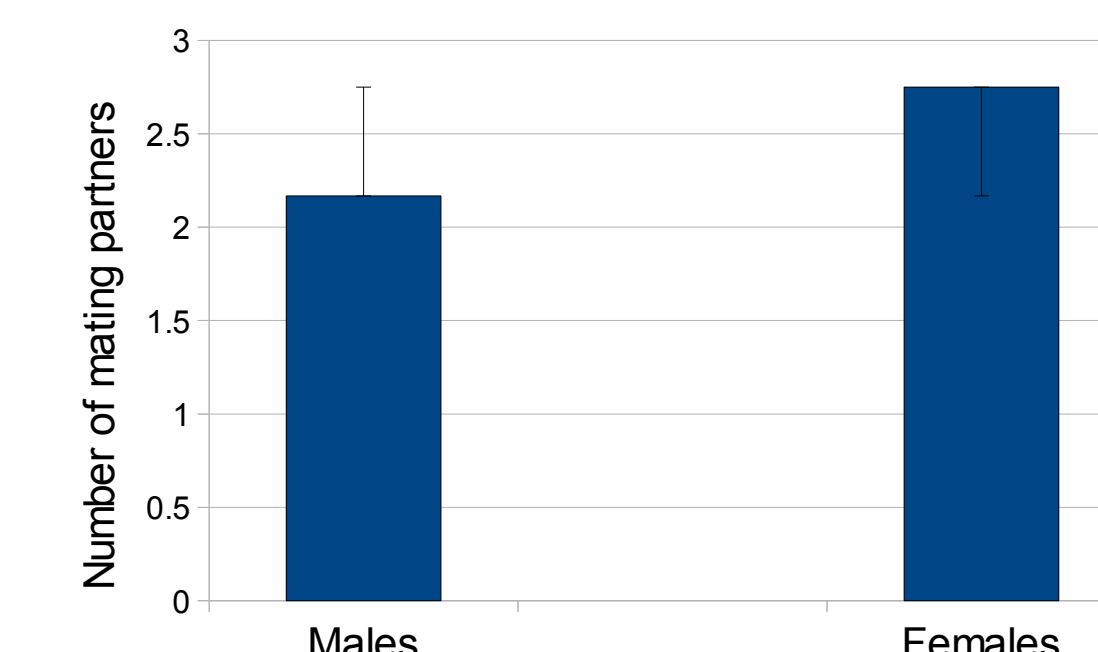


Fig. 4 Comparison of the average number of mating partners exhibited by the sexes.

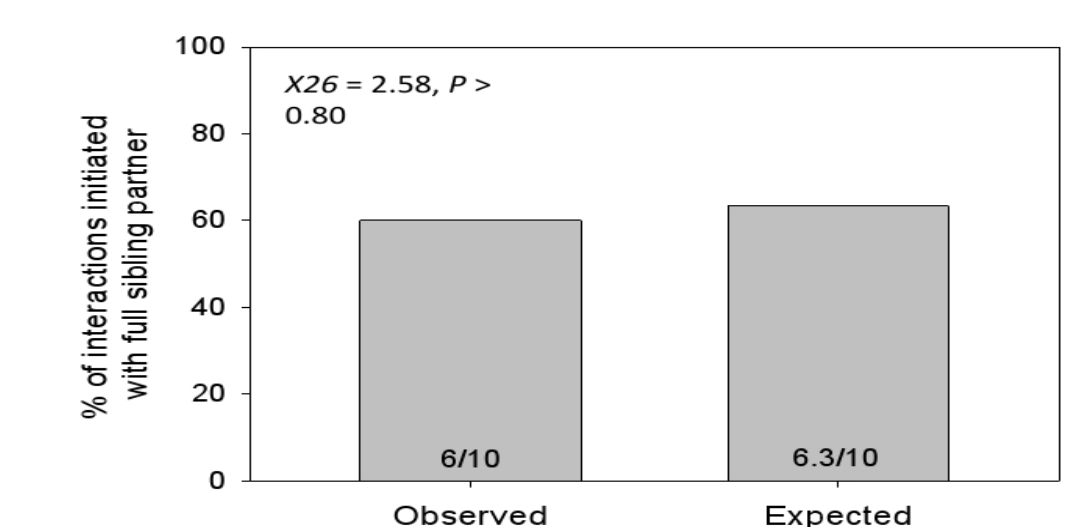


Fig. 6 Proportion of full sibling play partners.

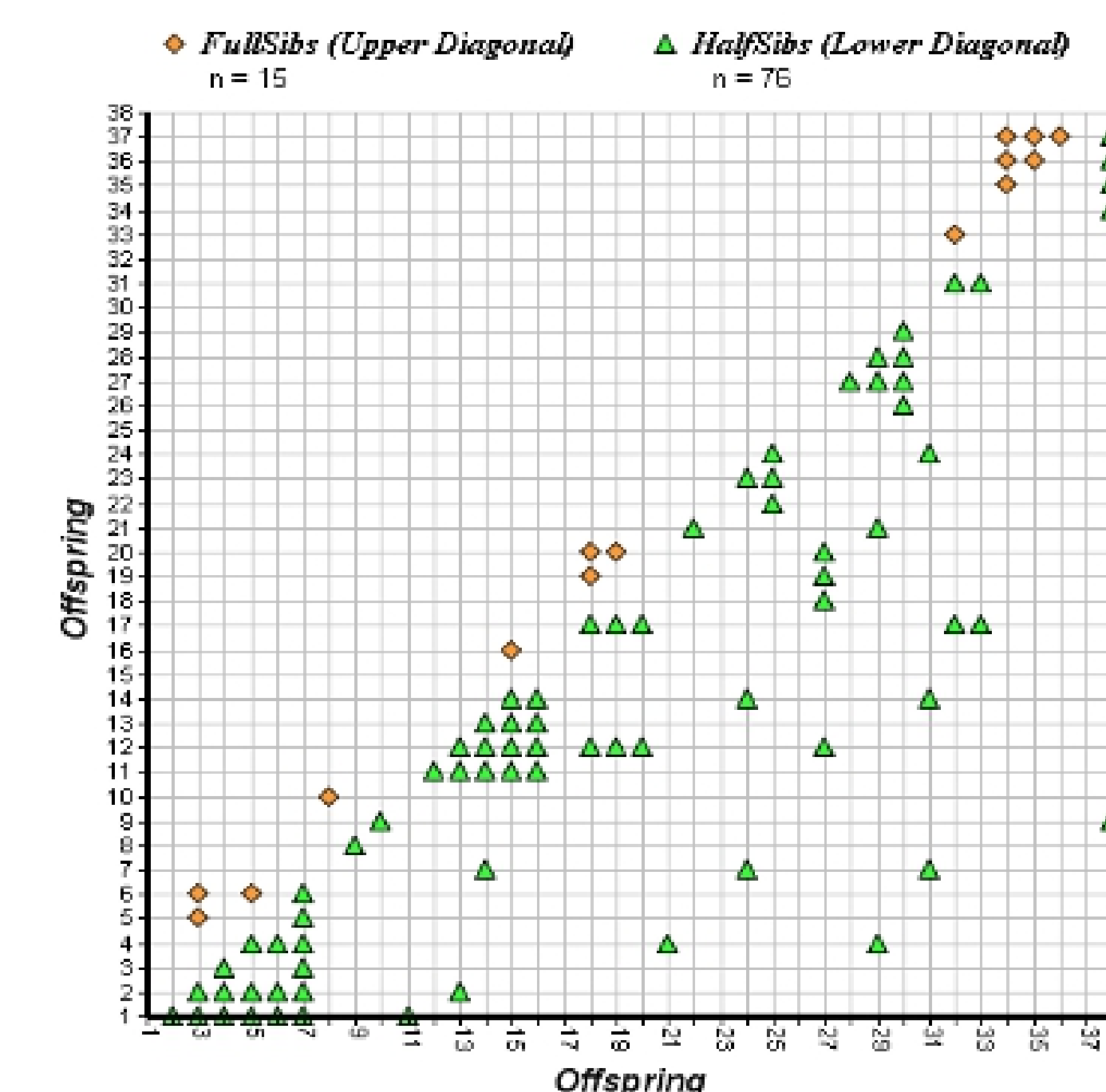


Fig. 5 Full sibs and half sibs in litters with known mothers.

Future research should focus on observing multiple litters and trapping males to verify mating, paternity, and play interactions. Inclement weather constrained the number of squirrels captured in 2010 and a limited number of samples from 2002-2005 were analyzed. Thus, future research should include more extensive sampling including both behavioral and location data collection.

Our findings provide a fuller picture of the natural history of Belding's ground squirrel. Because we found only small untagged males in the North Tioga Lake site, we hypothesize that they were yearling males that dispersed to that location from another site. Female and male dispersal rates are presumed to be similar enough to prevent differentiation between the populations.

The Tioga pass higher elevation population is a large, robust population that can serve as a model for similar populations.

References

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