significant associations are likely to be disguised by imprecise knowledge of conception dates, termination periods (*7*), and the environmental characteristics of the putative teratogen. Care must be taken in interpreting any large number of statistical tests such as those of Table 2. Even if there is no correlation between malformation and exposure, it is to be expected that 5 percent of the tests will be significant at the 5 percent level. In Table 2, 25 percent of the tests are significant at the 5 percent level.

Within the terms of the chosen criterion, the results of Table 2 show (i) no identifiable association for anencephaly or spina bifida, (ii) a significant association for talipes when f equals 0.25 but not when f equals 1.00, and (iii) no identifiable association for cleft lip with or without cleft palate, for isolated cleft palate, for malformations of the heart as a group, or for malformations of the male genitalia.

To clarify the nature of the correlations in Table 2, all the tests were recalculated with the contribution of the Whangarei hospital area omitted. Whangarei City (population 35,000) is the only urban area in Northland (population 107,000), and the Whangarei area is responsible for about half the total births. The pattern of correlations obtained was almost unchanged except for some loss of significance. For hypospadias and epispadias the termination period may extend to the end of the first trimester of pregnancy, and for cleft lip, cleft palate, and talipes, to the end of the second trimester. Test statistics for these malformations were recalculated with the exposure values being the average environmental concentration of putative teratogen taken over the second to fourth months and second to seventh months of pregnancy, respectively, for the two groups of malformations. The recalculated statistics showed increases in significance over those in Table 2 for hypospadias and epispadias and for talipes, no change for cleft lip, and a decrease in the case of isolated cleft palate.

The incidence rate of talipes is significantly higher among New Zealand Maoris than among Pakehas (New Zealand Caucasians) (*2*, *8*). Therefore, Maoris and non-Maoris (all those not specified as New Zealand Maori) were analyzed separately. These analyses were then combined by treating race as a partition of the data in the same way as Y, M, and A. The combined analyses, which allow for different malformation rates for Maoris and non-Maoris, produced results almost identical with those of Table 2, indicating that race is not primarily responsible for the correlations obtained in this case.

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9. We thank Barr Bros. Ltd., James Aviation Ltd., Marine Helicopters Ltd., Thames Aerial Topdressing Ltd., and Wishart Helicopters Ltd. for making their records on spraying available for this study. We also thank C. Garlick, superintendent of Northland Base Hospital, Whangarei; the superintendents and staffs of the other hospitals used in the investigation; A. J. Scott, R. B. Elliott, and R. H. Briant of the University of Auckland; A. H. Smith of the Wellington Clinical School, University of Otago; P. Talagaspitiya and R. Talagaspitiya and D. Ware and S. Ware of Whangarei; and G. Barclay, A. Tasnadi, and D. Arrowsmith of Auckland. Communicate with the authors for a copy of the complete report at cost ($13.50 New Zealand including postage).

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Multiple Paternity in Belding's Ground Squirrel Litters

Abstract. *Sexually receptive female* Spermophilus beldingi *(Rodentia: Sciuridae) usually mate with several different males. The paternity of 27 litters born in 1977 and 1978 was ascertained by combining field observations of mating with laboratory paternity exclusion analyses. Most of the litters (78 percent) were multiply sired, usually by two or three males. This may be the highest frequency of multiple paternity ever directly demonstrated in a natural population.*

Multiple paternity in a litter or brood occurs in natural populations of mammals (*1*), snakes (*2*), salamanders (*3*), fish (*4*, *5*), and arthropods (*6-9*); it has also been reported in laboratory populations of mammals (*10*), birds (*11*), fish (*12*), and insects (*8*, *13*). In most field studies, multiple paternity is inferred from the presence, in a single brood, of at least three electrophoretically detectable paternal alleles at a given protein locus (*14*); that is, multiple mating is deduced on the basis of genetic information from females and their broods or from broods alone. This method underestimates the frequency of multiple paternity in a population, because multiply sired broods go undetected when mates share the same biochemical phenotypes at the loci considered. To circumvent this problem, probabilistic formulas have been proposed (*1*, *15*) for estimating population values from the observed frequency of broods containing three paternal alleles.

In this report we quantify directly the occurrence of multiple paternity in a free-living population. Our analysis rests on field observations of mating in Belding's ground squirrels, *Spermophilus beldingi*, and, as in several recent investigations of genetic variation and vertebrate population structure (*16*, *17*), on paternity exclusion studies in which allozymes in the blood are assayed by gel electrophoresis.

Belding's ground squirrels are diurnal rodents that inhabit subalpine meadows in western North America. We observed them in the central Sierra Nevada at the 3040-m summit of Tioga Pass, California (*18*). There ground squirrels are active from May through September; the rest of the year they hibernate. The population has been studied since 1969, and most of

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Table 1. Blood protein loci used in the paternity exclusion analysis.

Locus	Source	Buffer (*20*)	Allele	Frequency* 1977	Frequency* 1978
Mannose phosphate isomerase	Hemolyzate	Continuous tris-citrate (*p*H 8.0)	M	0.97	0.95
			S	0.03	0.05
Adenosine deaminase	Hemolyzate	Phosphate (*p*H 6.7)	F	0.75	0.75
			M	0.13	0.20
			S	0.12	0.05
Adenylate kinase	Hemolyzate	Continuous tris-citrate (*p*H 8.0)	M	0.47	0.37
			S	0.53	0.63
Phosphoglucomutase 2	Hemolyzate	Phosphate (*p*H 6.7)	F	0.18	0.27
			M	0.82	0.73
Protein 1	Plasma	Tris-EDTA-borate (*p*H 8.0)	F	0.38	0.17
			M	0.17	0.25
			S	0.45	0.58
Protein 3	Plasma	Tris-HCl (*p*H 8.2)	M	0.99	0.93
			S	0.01	0.07

*Frequency estimates are based on 186 animals in 1977 and on 64 in 1978.

the animals have been marked with hair dye for visual recognition and have had their ears tagged or their toes clipped for permanent identification.

We observed matings on 16 days in 1977 (5 to 8 May and 30 May to 11 June) and on 18 days in 1978 (8 to 25 June)—a total of 603 man-hours. The females are sexually receptive for 4.7 ± 0.3 hours (standard deviation; $N = 54$) on a single afternoon each year. Copulation takes place above ground, where the females mate with one to five different males (Fig. 1). By watching almost continuously, we recorded all the matings of 19 females and most of the matings of 14 others (*19*). We then livetrapped all 33 (plus five females that we did not see mate) and removed 1 to 2 ml of blood from each by pipetting from the suborbital sinus. We also took blood from all 33 potential sires in the 1977 and 1978 populations (the 28 males that we saw mate, plus five adult residents that we did not see mate). Finally, all 175 young weaned by the females (135 in 1977, 40 in 1978) were captured and sampled within 2 days of emerging from their natal burrows.

In the field, blood samples were kept on ice until centrifugation (2 to 4 hours); after separation, the plasma and cellular fractions were stored separately in liquid nitrogen (-196°C). Within 3 weeks the samples were transferred to a laboratory freezer (-76°C), where they remained for 2 to 5 months until analysis. Proteins were studied with standard horizontal starch-gel electrophoresis and histochemical staining procedures (*20*). Before beginning this study, we surveyed 30 loci ($N \geq 19$ animals) to identify polymorphic systems (*21*). We found six polymorphic loci in blood (Table 1) for use in our paternity analysis.

The phenotype at each of the six loci was determined for every animal. Then, for each litter, the phenotype of every juvenile and its dam was used to implicate or eliminate one of the observed mates as the sire. In some litters, all offspring could have been sired by only one male (single paternity). In others, a given mate must have sired some offspring, but could not have sired them all (multiple paternity).

We evaluated paternity indirectly when mating observations were incomplete or nonexistent. Genetic information from the female and her young was used to postulate the phenotype of a male that could have sired the whole litter. If none of the males in the population had the appropriate phenotype, multiple paternity was inferred. The presence in a litter of at least three paternal alleles at the adenosine deaminase or protein 1 locus also indicated multiple paternity (*1*, *3*, *5*, *8*).

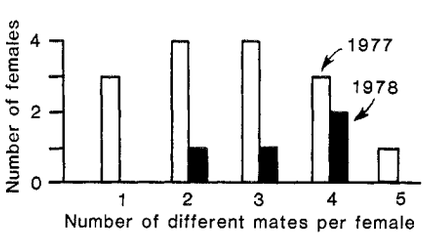

Fig. 1. Mating frequency of female *S. beldingi* at Tioga Pass. The mean number of mates per female was 2.7 in 1977 and 3.3 in 1978.

Table 2. Results of the paternity exclusion analysis.

Female's matings observed	*N*	Litters with one sire	Litters with multiple sires	Litters unresolved*
All	19	6†	7	6
Most	14	0	10	4
None	5	0	4	1
Total	38	6	21	11

*These litters could have been sired by at least two mates; due to phenotypic (allelic) similarities among mates, we could not determine whether the litter was actually sired by one or both. †Three of these six females mated monogamously.

In all, the paternity of 27 of the 38 study litters was resolved (Table 2). Of these, 21 (78 percent) were multiply sired. We believe that this is the highest frequency of multiple paternity ever demonstrated in a natural population. Even if all 11 unresolved litters were singly sired, the minimum frequency of multiple paternity would be 55 percent. The implication is that most *S. beldingi* litters comprise full siblings and maternal half-siblings.

Of the 19 litters for which there are complete data (Table 2), six were singly sired; three of these were born to females that had mated only once. Thus seven of ten litters born to multiply mating females were multiply sired. We conclude that in this species multiple mating usually results in multiple paternity.

The exact number of sires was determined for four of the multiply sired litters: three were sired by two males each, and one was sired by three. In two of these litters the sire of each offspring was identified. In the first, a litter of six, one mate sired five young and a second sired one. In the second, a litter of four, two mates sired two young each, while a third mate sired none. Thus, intrabrood paternal representation was unequal—as in multiply inseminated arthropods (*7–9*, *22*), fish (*12*), and laboratory mammals (*10*).

Probabilistic models for estimating the population-wide frequency of multiple paternity (*1*, *15*) yield two extreme estimates: the maximum posits only two sires per brood, and the minimum posits an infinite number (*23*). Using these models and observed allelic frequencies (Table 1), we computed estimates of multiple paternity in *S. beldingi*. For the adenosine deaminase locus, the estimated frequency was between 100 percent (the two-male model) and 32 percent (infinite number of males). For the protein 1 locus, the extreme values were 143 and 66 percent, respectively (*24*). While these estimates indicate that multiple paternity is frequent in *S. beldingi*, they do not permit its precise quantification. In this study both behavioral observations and genetic information were required to obtain a reliable estimate.

Our demonstration of frequent multiple paternity in *S. beldingi* implies that littermates are not all equally related in this species. Should multiple paternity be as common in other social species, studies of intrabrood differences in coop-

eration and competition may prove valuable for evaluating kinship theory (*25*) and for investigating mechanisms of kin recognition (*26*).

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21. Of 30 proteins surveyed in liver, kidney, muscle, and blood, the following were monomorphic (the number of loci assayed is given in parentheses): lactate dehydrogenase (2), peptidase (3), glyceraldehyde phosphate dehydrogenase (1), phosphogluconate dehydrogenase (2), superoxide dismutase (1), diaphorase (1), malate dehydrogenase (2), carbonic anhydrase (1), malic enzyme (1), isocitrate dehydrogenase (2), α-glycerol-3-phosphate dehydrogenase (1), phosphoglucomutase (2), glutamate oxaloacetate transaminase (1), and glucose-6-phosphate dehydrogenase (1). The following loci were polymorphic (the number of alleles is given in parentheses): adenylate kinase (2), sorbitol dehydrogenase (3), mannose phosphate isomerase (2), adenosine deaminase (3), phosphoglucomutase 2 (2), glutamate oxaloacetate transaminase 1 (2), malic enzyme 1 (2), protein 1 (3), and protein 3 (2). There was no evidence of "null" alleles (alleles not detectable by electrophoresis) at any locus. Frequency of polymorphic loci was 33 percent and heterozygosity averaged 10.7 percent. For the six polymorphic loci that were resolvable in blood (Table 1), frequencies of homozygotes and heterozygotes agree with the Hardy-Weinberg expectation, computed from observed allele frequencies ($P \geq .9$ in all cases, χ^2 test).
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24. For the 1977 and 1978 data combined, at the adenosine deaminase locus the two-male and "infinite male" models predicted detection of 2.0 and 6.2 litters containing three paternal alleles, respectively; two litters were observed. At the protein 1 locus, detection of 4.9 and 10.6 litters was expected; seven litters were observed.
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Language-Related Potentials Specific to Human Language Cortex

Abstract. *Event-related potentials following silently named object pictures were recorded directly from the exposed left hemisphere of the human cortex at sites whose relation to naming was subsequently established by electrical stimulation mapping. Two simultaneous potential changes are specific to sites where stimulation disrupts naming: slow potentials at premotor sites and focal desynchronization at posterior sites surrounding the Sylvian fissure. These anatomically specific changes are also specific to the task—present with silent naming and absent in a spatial task with the same visual input. Overt speech is also preceded by slow potentials with earliest onset at premotor sites.*

Only certain areas in the dominant cerebral hemisphere are specialized for language, but neurophysiological correlates of language showing intrahemispheric specificity to these areas have not been demonstrated. Such demonstration requires recording from discrete cortical regions, and the pattern of cortical language sites in each subject must be known, as it varies considerably between individuals (*1*). We now report neurophysiological correlates specific to a language task that are also specific to the language cortex. These are changes in event-related potentials (ERP's) during silent naming, recorded from exposed human cortex that was subsequently shown to be specialized for language by the presence of errors in naming when that cortex was electrically stimulated.

These data were obtained from six adult patients undergoing left-hemisphere craniotomies under local anesthesia for resection of epileptic foci (*2*). In all patients, the left hemisphere was dominant for language, as shown by preoperative intracarotid Amytal testing (*3*). After the epileptic focus was identified, 1-mm silver ball electrocorticographic (ECOG) electrodes were placed at eight to ten arbitrarily selected sites generally outside of the epileptic focus, in the cortex surrounding the Sylvian fissure (four patients) or in the frontal lobe (two patients). During ECOG recording, patients engaged in two tasks; (i) name matching, which elicits silent object

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