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MANIPULATION OF FEMALE BEHAVIOUR IN FIELD POPULATIONS OF *MICROTUS TOWNSENDII*

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SUMMARY

(1) The behaviour of female *Microtus townsendii* was manipulated in field populations during a spring decline in numbers. All females were implanted with testosterone to increase aggressiveness in one area, and force-fed the chemosterilant mestranol to reduce aggressiveness in another area.

(2) Testosterone treatment resulted in wounding among females, increased size of female home ranges, a reduction in female survival, and increased female immigration.

(3) Males in the population with testosterone-treated females had the same population dynamics as males in a control population and a population with no females.

(4) Mestranol treatment had no effect on female dynamics. Males with mestranol-treated females survived better than control males.

(5) In spite of the profound affect of testosterone on social strife, the spring decline in numbers was little affected by these experiments. Female spacing behaviour may operate in more subtle ways than overt aggression.

INTRODUCTION

Populations of the vole *Microtus townsendii* Bachman on Westham Island, B.C. decline substantially in number with the onset of breeding in spring (LeDuc & Krebs 1975; Krebs 1979). The repeatability of this event has facilitated experimental investigation into the causes of spring declines (Gipps *et al.* 1981; Taitt *et al.* 1981).

Hormonal manipulation of the behaviour of animals in the field has so far been restricted to males (red grouse (*Lagopus lagopus scoticus*) Watson 1970; sharp-tailed grouse (*Pedioecetes phasianellus*) Trobec & Oring (1972); Townsend's vole (*M. townsendii*) Krebs, Halpin & Smith 1977, and Gipps *et al.* 1981; bank vole (*Clethrionomys glareolus*) Gipps & Jewell 1979), with the exception of Goulet (1979) who manipulated the behaviour of female Richardson's ground squirrels (*Spermophilus richardsonii richardsonii*). The results of the experiments on male *M. townsendii* have been disappointing because they did not substantially change the demography of the spring decline. This and the accumulating evidence (Leuze 1976; Redfield, Taitt & Krebs 1978; Madison 1980; Saitoh 1981) that indicates that female spacing behaviour is important in populations of small mammals lead us to the present experiments.

If female aggressive behaviour contributes to the socially-induced mortality or dispersal which is believed to be the cause of spring declines (Krebs & Boonstra 1978), then manipulation of female behaviour should result in certain predictable outcomes. Large numbers of aggressive females should increase social strife which, in turn, should result in a more rapid population decline. On the other hand, a population with predominantly

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passive females should have less social strife and hence should decline more slowly if at all. The complete removal of females from a population and the simultaneous monitoring of an unmanipulated control should indicate how female spacing behaviour affects spring population dynamics.

METHODS AND EXPERIMENTS

Five trapping grids were established in grassland on Westham Island in the Fraser River Delta south of Vancouver, B.C. (Fig. 1). All grids except C2, were bounded on one side by a 6 mm ($\frac{1}{4}''$) wire-mesh fence, and on the side at right angles by either a road (grid M2) or a water-filled ditch (grids C1, M1, T). Grid C2 was bounded on one side by a road and on the other by a water-filled ditch (Fig. 1). Control grid C1 and experimental grids M1 and T were established in uncut grassland. Both control C2 and experimental grid M2 had less cover because the grass had been cut to 15 cm in July 1979.

Two Longworth live-traps were placed at each of thirty-nine points 7·6 m (25 ft) apart on each grid. Voles were trapped every 2 weeks for 2 nights and the intervening day from November 1979 to May 1980. All traps were locked open for prebaiting between trapping periods. Each new vole captured was given a numbered ear tag and its location, weight, and breeding condition were recorded. Males had either abdominal or scrotal testes; females had open or closed vaginas and small, medium or large nipples. All pregnancies and trap litters were noted. Voles weighing <30 g were classed as juveniles, subadults weighed 30–42 g and all voles > 42 g were called adults. All wounds on the rear of each vole were counted.

We attempted to increase aggressive behaviour of females by giving them subcutaneous implants of testosterone. In the laboratory, albino female mice (*Mus musculus*) showed more fighting after treatment with testosterone (Barkley & Goldman 1977). We placed dry testosterone powder in 1·5-cm lengths of Dow Corning Silastic tubing (No. 602–285). The ends of the tubing were sealed with Silastic glue to form a capsule. The capsules were

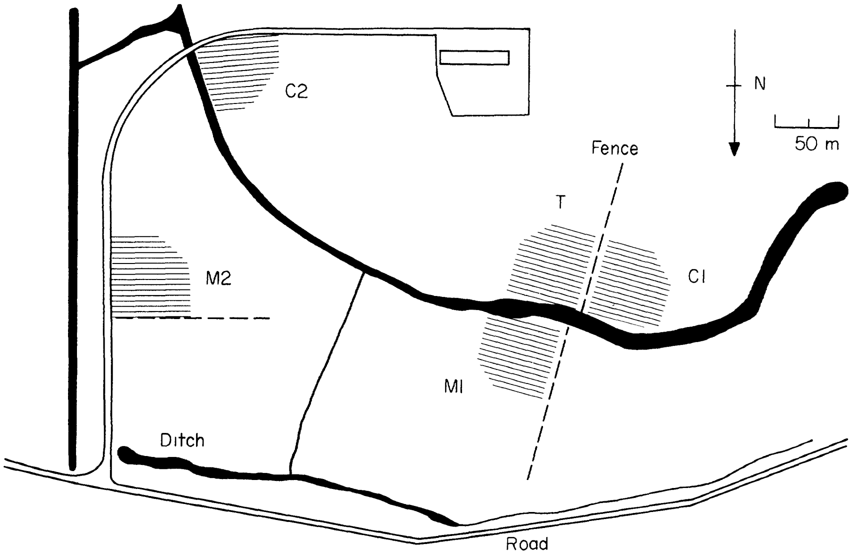

FIG. 1. Map of the study site on Westham Island, British Columbia, showing the locations of the live-trapping areas. Grids C1 and C2 are controls, M1 is the 'female removal', M2 the 'passive female', and T the 'aggressive female' grid.

pushed under the skin through a small slit cut at the back of the neck. One stitch was sufficient to close the slit. All females on grid T were implanted with a capsule of testosterone on 28–30 January 1980. New females that emigrated onto the grid up to 12 April 1980 were implanted, and all females were re-implanted once a month until this date. Since Gipps *et al.* (1981) found no affect of implants on survival in this species we assummed this to be true in 1980.

We tried to reduce female aggression initially by subcutaneous injection of mestranol. Goulet (1979) stopped reproduction and reduced aggression of female Richardson's ground squirrels by administering this chemosterilant. Mestranol has been shown to inhibit reproduction in voles (Howard & Marsh 1969). All females on grid M1 were given a 2 ml injection of sesame seed oil containing approx 0·001 g of mestranol during the period 28–30 January 1980, but after a second injection 2 weeks later most females on grid M1 disappeared. On another grid (M2), we force-fed approx 0·001 g of mestranol powder to all females on 10–12 March 1980. All remaining females were removed from grid M1 on 25 February and the grid was maintained as a 'female removal' from then until 8 May.

RESULTS

Social interaction

Wounding of individuals in a population has been used as an indirect measure of intraspecific strife (Southwick 1958; Christian 1971).

Apart from grid T, males had more wounds and earlier wounds than females on all grids (Fig. 2). The peak of wounding in both sexes was in the month early March to early April. The number of females wounded on grid T increased immediately after females had been

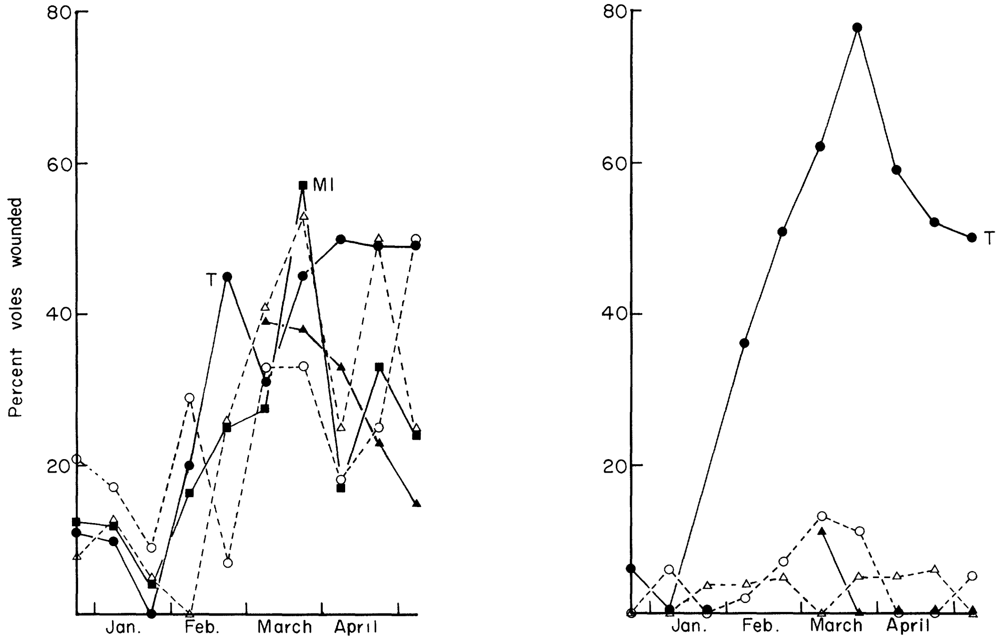

FIG. 2. Percentage of voles with wounds on rear portion of body. (a) Males, (b) females. ●——●, grid T; ○---○, control C1; △---△, control C2; ■······■, grid M1; ▲——▲, grid M2.

implanted with testosterone. The number wounded was not only much higher than the number of females wounded on the controls, but also much higher than the number of males wounded on all grids (Table 1). Males in the 'passive' female population had 0·58 wounds per vole compared with 0·75 wounds per vole on the low cover control in the period of this experiment, while female wounding was the same (0·38 on grid M2 and 0·39 on grid C2).

Home ranges were calculated using the elliptical model of Koeppl, Slade & Hoffman (1975). Male voles had larger home ranges than females on all grids except grid T (Table 2). Males and females on the low cover control (C2) had larger home ranges than C1 controls (males 1·34 times, females 1·29 times). Females with testosterone had significantly larger home ranges than males in the same population and females on the control (C1) ($P < 0{\cdot}05$ ANOVA test).

TABLE 1. The frequency distribution of wounds recorded on each grid between 30 January and 8 May

Number of wounds	Control C1		Control C2		Grid M1	Grid T	
	♂	♀	♂	♀	♂	♂	♀
0	95	206	72	123	125	85	95
1	20	8	14	4	29	25	30
2	4	5	8	1	8	7	20
3	3	0	6	0	9	10	20
4	2	0	2	0	0	3	12
5	1	0	0	0	0	0	9
6	1	0	1	0	1	0	5
7	0	0	0	0	0	0	3
8+	1	0	0	0	0	1	9
Total wounds	64	18	62	6	78	89	346
Mean number of wounds per vole	0·50	0·08	0·60	0·05	0·45	0·68	1·70

TABLE 2. Home range size in m^2 for the period 30 January to 8 May. Sample size in parentheses

	Control C1	Control C2	Grid M1	Grid T
Males	266 (29)	357 (16)	325 (34)	191 (22)
Females	187 (37)	247 (23)	–	297 (39)

Population size

The peak populations on the low cover grids (C2 and M2) were half or less than peak numbers on the other grids (C1, T, M1) (Fig. 3). Numbers declined very slowly initially on these two grids in contrast to the uncut grids. But, by the end of the spring decline in late April, numbers had reached the same low levels as on all grids.

The period of maximum loss (10 March to 11 April) in both sexes coincides with the peak in wounding (Fig. 2) and with the onset of breeding. Over 50% of adult males on Control C1 were breeding, and over 30% of adult males on all other grids had scrotal testes in early March (Table 3). Apart from grid T, the maximum number of females with perforate vaginas was also recorded in this period (Table 3), as was the first pregnancy. (The higher numbers of perforate females later on grid T was a direct response to hormonal treatment.) The end of the period of maximum decline coincides with the appearance of the first juveniles.

The presence of 'aggressive' and 'passive' females or even the 'absence' of females had

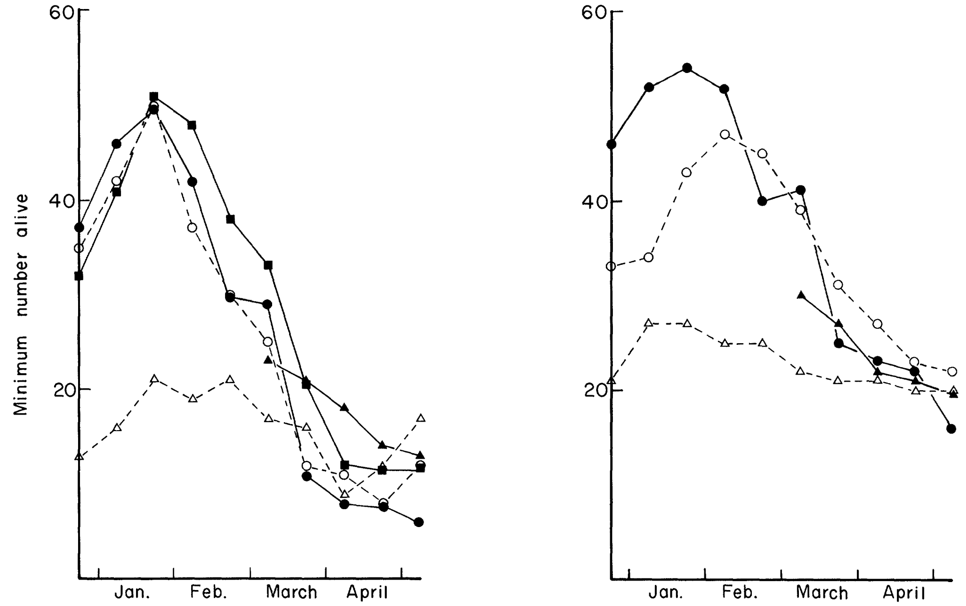

FIG. 3. Minimum number of individuals known to be alive during the spring decline. (a) Males, (b) females. ●——●, grid T; ○---○, control C1; △---△, control C2; ■——■, grid M1; l; ▲——▲, grid M2.

TABLE 3. Percentage of male voles with scrotal testes, number of females with perforate vaginae, and, in parentheses, the number of pregnant females in each trapping period

	Control C1		Control C2		Grid M1	Grid M2		Grid T	
Date in 1980	♂	♀	♂	♀	♂	♂	♀	♂	♀
12–14 Feb.	16	12	0	3	12	–	–	5	37
25–27 Feb.	38	17	6	6	24	–	–	4	28
10–12 Mar.	75	20 (1)	35	9	35	41	23	31	37
24–26 Mar.	33	13	43	6 (1)	53	79	16 (1)	100	16
9–11 Apr.	89	10 (4)	71	8 (4)	92	94	11 (1)	71	18
22–24 Apr.	100	12 (3)	88	7 (4)	83	100	13 (2)	88	14 (1)
6–8 May	100	13 (4)	90	8 (4)	92	100	8 (2)	100	11
14–16 May	75	12 (2)	92	10 (6)	100	93	11 (4)	86	12

no significant effect on the decline in male numbers (Fig. 3). Males in the presence of 'passive females' had a lower rate of decline than males on all other grids during the periods of maximum decline. Female population density was also not affected by the behaviour treatments.

Immigration

The number of new adult and subadult males captured on the control C1 and the experimental grids (M1, T) was very similar over the experimental period (30 January to 8 May) (Table 4).

More females immigrated onto the 'aggressive-female' and 'female-removal' grid than to either control. This was particularly marked in the period of maximum decline when twenty-six new females were captured on the 'female-removal' grid, and seventeen new

TABLE 4. New voles captured on each grid

Date in 1980	Control C1		Control C2		Grid M1		Grid M2		Grid T	
	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀
28–30 Jan.	11	12	5	2	11	9	–	–	8	8
12–14 Feb.	4	7	3	4	0	0	–	–	6	7
25–27 Feb.	0	3	4	2	4	3	–	–	1	3
10–12 Mar.	5	3	3	2	4	5	–	–	1	11
24–26 Mar.	0	1	1	1	2	8	0	1	0	1
9–11 Apr.	1	2	1	1	1	13	0	1	0	6
22–24 Apr.	0	1	2	0	1	3	0	2	1	7
6–8 May	0	3	4	2	1	2	2	2	3	0
14–16 May	2	1	6	3	0	2	4	4	1	5
Total for experimental period	10	20	18	13	13	34	–	–	12	35

TABLE 5. Average survival of 'residents'. Number of animals in parentheses

	30 January–11 April		10 March–11 April	
	♂	♀	♂	♀
Control C1	0·05 (43)	0·41 (39)	0·30 (23)	0·57 (37)
Control C2	0·16 (19)	0·60 (25)	0·50 (16)	0·91 (21)
Grid M1	0·15 (40)	–	0·29 (31)	–
Grid M2	–	–	0·86 (21)	0·67 (27)
Grid T	0·12 (43)	0·13 (47)	0·26 (27)	0·36 (36)

females on the testosterone grid, compared with six and four females respectively on the controls C1 and C2. Unfortunately the oral administration of mestranol to females on grid M2 was too late for direct comparison of this population, but the level of immigration was similar to the controls.

Emigration and death

It is not possible to differentiate between these two components of demography when a vole ceases being trapped. In an attempt to overcome this problem, Taitt *et al.* (1981) suggested that voles could be divided into two groups on the basis of their trapping records. 'Emigrants' were voles that were captured once only while 'residents' were captured at least twice. When 'residents' cease being trapped they may still have emigrated though more of them probably died or were killed *in situ*. 'Residents' were used to calculate average survival of voles in each population.

Only 16% or less of males that were present on all grids when experiments began in January were alive at the end of the period of maximum decline (Table 5). Female survival on the two control grids was higher than male survival. But females with testosterone had significantly poorer survival than control females ($P < 0\cdot05$, χ^2 test), and, in fact, had as poor survival as males. Males in the presence of mestranol-treated females had much better survival through the month of maximum loss (10 March to 11 April) than males on the controls and males with no females (grid M1) ($P < 0\cdot05$, χ^2 test), while males with 'aggressive' females had the lowest survival in this period.

DISCUSSION

The results of a number of experimental and descriptive studies done in the last decade indicate that female small mammals may be regulating population size. Redfield began sex-specific removals from field populations of *M. townsendii* in 1972. After 2 yr of maintaining a predominantly 'male' and 'female' population, he concluded that recruitment was dependent on the density of female voles not male voles (Redfield *et al.* 1978). In particular, he found an inverse relationship between the number of young voles recruited and the density of females. Boonstra (1978) and Taitt (1981) established that the survival of young voles (*M. townsendii*) and deermice (*Peromyscus maniculatus*) was also dependent on female not male density. Studies have also shown that breeding females occupy areas free of other females (Leuze 1976 for water voles (*Arvicola terrestris*), Madison 1980 for meadow voles (*M. pennsylvanicus*), Saitoh 1981 for red-backed voles (*Clethrionomys rufocanus bedfordiae*), and Boutin 1980 for snowshoe hares (*Lepus americanus*)). Spacing behaviour by females may be the mechanism which gives rise to the stability in numbers of reproductive females per unit area observed in bank voles (Bujalska 1973), water voles (Leuze 1976), and red-backed voles (Saitoh 1981).

Populations of Townsend's vole on Westham Island, B.C. show marked spring declines in density at the onset of breeding. The breeding population thus established is remarkably constant from year to year. We have attempted to test whether female behaviour, manipulated by hormone treatment, could produce predictable changes in the spring population. Our tests of the effects of the hormones on laboratory-housed voles were disappointing (Appendix). But the high level of wounding that we observed in the field population of testosterone-treated females indicated that we were successful at increasing social strife. Such a dichotomous result between laboratory and field was also obtained by Turner, Iversen & Severson (1980) with castrated meadow voles. We agree with their suggestion that the laboratory environment was probably so unnatural that it masked the experimental results.

In spite of the high level of wounding in the aggressive female population, the number of voles at the end of the decline was only slightly lower (at 22) than the controls (at 30 and 37). 'Aggressive' females had lower survival than control females, but, immigration of females to the 'aggressive' female population was as high as to a female removal area. The dynamics of male voles was unaffected by either 'aggressive' females or by the absence of females. So, like Redfield, Taitt & Krebs (1978), we conclude that the dynamics of male and female voles of this species seem to be largely independent of each other. Although the higher survival of males in the population of 'passive' females, and higher male survival in a population of scopolamine-treated (more passive) adult males (Gipps *et al.* 1981) does suggest that the presence of non-aggressive individuals of either sex may be important to male survival.

Do we have to conclude that the aggressive behaviour of female Townsend's vole is unimportant in setting the spring breeding density? In males, the number of wounds has been correlated with the rate of spring decline (Krebs & Boonstra 1978). Female wounding is typically low and their survival higher than male survival (Krebs & Boonstra 1978). On the 'aggressive' female grid, females were like males (high wounding and low survival), but immigration (replacement of dead/emigrant females) was higher than male immigration. This indicates that overt aggressive behaviour may not be the only factor influencing female spacing behaviour. Females respond more quickly than males to extra food (Fordham 1971; Taitt 1981, and Taitt & Krebs 1981) and cover (Taitt *et al.* 1981), which suggests that their spacing behaviour may be strongly influenced by physical features of the environment. Further, since they achieve spring declines in numbers normally with little, or no wounding, and normally have smaller home ranges than males, they may exhibit more site-specific, defensive spacing behaviour rather than overt aggressive behaviour. Also females may be more responsive in aggressive encounters such that one encounter with a dominant is sufficient to stimulate emigration (observed by Leuze 1976), or avoidance and delayed maturity (as observed by Saitoh 1981). Perhaps by giving females testosterone, we destroyed their 'normal' female behaviour so that they behaved like males, taking larger home ranges, fighting, and wounding one another, and surviving poorly.

In addition to the behavioural manipulations, we monitored the population dynamics of a population with reduced natural cover. This population had a lower winter population peak then declined very little in spring. But the number of breeding males and females was very similar to those on the other grids which underwent substantial spring declines. This result suggests that both sexes are sensitive to reduced cover as well as increased cover (Taitt *et al.* 1981). If densities converge on different areas to a common final density in May (Fig. 3), the spring decline must be density-dependent. This contradicts the finding of Krebs & Boonstra (1978) that spring declines are not density-dependent. But if cover is an intervening variable, the intensity of the spring decline may be related to cover when cover is correlated with the preceding winter peak density.

The experiments that we have tried should be repeated. We were unsuccessful in our initial administration of mestranol, and hence in producing a population of 'passive females' simultaneous with the spring decline. It would be interesting to implant females with testosterone long before the spring decline and attempt to cause a decline in female numbers before the natural decline. It would also be interesting to give only some females in a population testosterone and see how they competed with 'normal' females. Finally, if such experiments are done an attempt should be made to monitor blood steroid levels (I. R. McDonald & M. J. Taitt unpublished) to see if there is any causal relationship between them and survival through the spring decline.

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APPENDIX

We conducted a small-scale laboratory test of the effects of testosterone implants and mestranol injections on female voles. Thirty-two females were removed from the field in November 1979. All were kept separately in plastic cages with *ad lib* food and water. Females were divided randomly into four groups of eight: 'standards', 'controls', 'testosterone', 'mestranol'. After 6 weeks in captivity and before treatment each vole in the last three groups was exposed to a particular 'standard vole' in a neutral arena for 10 min.

Voles in the 'testosterone' group were given a subcutaneous implant of testosterone (as described above for the field). The 'mestranol' group were injected with mestranol (as above); unfortunately two of this group died. A week later, all voles were tested against the same 'standard' vole as in the pre-treatment test.

The tests were repeated on twelve females collected from the field in April 1980. There were four voles in the groups: 'standard', 'mestranol', 'testosterone'. These voles were tested, treated, and re-tested in the same manner as above, except the mestranol was given orally in powder form.

Activity between pre- and post-experimental bouts

	'Control'	'Mestranol'	'Testosterone'
Experiment 1	−18	−73	−37
Experiment 2	–	−85	−13
'Standards' in expt 1 with	−33	−53	−19
'Standards' in expt 2 with	–	−30	+2·5

Paired female voles showed very little behavioural interaction in the neutral arena. Females in all groups had reduced levels of activity (number of 10 cm squares crossed per 10 min bout) in the second (post-experimental) arena test. The movements of individuals during the arena test was the only measurable behaviour that was shown by all voles. Although activity was less in the second test, 'mestranol' females were the least active in both tests. 'Testosterone' females were more active than 'mestranol' females but not as active as 'controls' in the first experiment. Further, 'standard' females when paired with 'mestranol' females showed less activity than when they were paired with 'controls', and most activity when paired with 'testosterone' females.

REFERENCES

Barkley, M. S. & Goldman, B. D. (1977). Testosterone-induced aggression in adult female mice. *Hormones and Behaviour*, **9**, 76–84.

Boonstra, R. (1978). Effect of adult townsend voles (*Microtus townsendii*) on survival of young. *Ecology*, **59**, 242–248.

Boutin, S. (1980). Effect of spring removal experiments on the spacing behaviour of female snowshoe hares. *Canadian Journal of Zoology*, **58**, 2167–2174.

Bujalska, G. (1973). The role of spacing behaviour among females in the regulation of reproduction in the bank vole. *Journal of Reproduction and Fertility*, **19** (suppl.), 465–474.

Christian, J. J. (1971). Fighting, maturity, and population density in *Microtus pennsylvanicus*. *Journal of Mammalogy*, **52**, 556–567.

Fordham, R. A. (1971). Field populations of deermice with supplementary food. *Ecology*, **52**, 138–146.

Gipps, J. H. W. & Jewell, P. A. (1979). Maintaining populations of bank voles, *Clethrionomys glareolus*, in large outdoor enclosures, and measuring the response of population variables to the castration of males. *Journal of Animal Ecology*, **48**, 535–555.

Gipps, J. H. W., Taitt, M. J., Krebs, C. J. & Dundjerski, Z. (1981). Male aggression and the population dynamics of the vole, *Microtus townsendii*. *Canadian Journal of Zoology*, **59**, 147–158.

Goulet, L. A. (1979). *Aspects of population dynamics and social behaviour in the Richardson's ground squirrel as modified by a chemosterilant (Mestranol).* Unpublished Ph.D. thesis, Simon Fraser University, B.C.

Howard, W. E. & Marsh, R. E. (1969). Mestranol as a reproductive inhibitor in rats and voles. *Journal of Wildlife Management*, **33**, 403–408.

Koeppl, J. W., Slade, N. A. & Hoffman, R. S. (1975). A bivariate home range model with possible application to ethological data analysis. *Journal of Mammalogy*, **56**, 81–90.

Krebs, C. J. (1979). Dispersal, spacing behaviour, and genetics in relation to population fluctuation in the vole *Microtus townsendii*. *Fortschritte der Zoologie*, **25** (2/3), 61–77.

Krebs, C. J. & Boonstra, R. (1978). Demography of the spring decline in populations of the vole, *Microtus townsendii*. *Journal of Animal Ecology*, **47**, 1007–1015.

Krebs, C. J., Halpin, Z. T. & Smith, J. N. M. (1977). Aggression, testosterone, and the spring decline in populations of the vole, *Microtus townsendii*. *Canadian Journal of Zoology*, **55**, 430–437.

LeDuc, J. & Krebs, C. J. (1975). Demographic consequences of artificial selection at the LAP locus in voles (*Microtus townsendii*). *Canadian Journal of Zoology*, **53**, 1825–1840.

Leuze, C. C. K. (1976). *Social behaviour and dispersion of the water vole*, Arvicola terrestris *Lacepede*. Unpublished Ph.D. thesis, Aberdeen University, Aberdeen, Scotland.

Madison, D. M. (1980). Space use and social structure in meadow voles, *Microtus pennsylvanicus*. *Behavioural Ecology and Sociobiology*, **7**, 65–71.

Redfield, J. A., Taitt, M. J. & Krebs, C. J. (1978). Experimental alteration of sex ratios in populations of *Microtus townsendii*, a field vole. *Canadian Journal of Zoology*, **56**, 17–27.

Saitoh, T. (1981). Control of female maturation in high density populations of the red-backed vole, *Clethrionomys rufocanus bedfordiae*. *Journal of Animal Ecology*, **50**, 79–87.

Southwick, C. H. (1958). Characteristics of house mice living in English corn ricks—density relationships. *Proceedings of the Zoological Society London*, **131**, 163–175.

Taitt, M. J. (1981). The effect of extra food on small rodent populations: 1. Deermice (*Peromyscus maniculatus*). *Journal of Animal Ecology*, **50**, 111–124.

Taitt, M. J. & Krebs, C. J. (1981). The effect of extra food on small rodent populations: 2. Voles (*Microtus townsendii*). *Journal of Animal Ecology*, **50**, 125–137.

Taitt, M. J., Gipps, J. H. W., Krebs, C. J. & Dundjerski, Z. (1981). The effect of extra food and cover on declining populations of *Microtus townsendii*. *Canadian Journal of Zoology*, **59**, 1593–1599.

Trobec, R. J. & Oring, L. W. (1972). Effects of testosterone proprionate implantation on lek behaviour of sharp-tailed grouse. *American Midland Naturalist*, **87**, 531–536.

Turner, B. N., Iverson, S. L. & Severson, K. L. (1980). Effects of castration on open-field behaviour and aggression in male meadow voles (*Microtus pennsylvanicus*). *Canadian Journal of Zoology*, **58**, 1927–1932.

Watson, A. (1970). Territorial and reproductive behaviour of red grouse. *Journal of Reproduction and Fertility*, **11** (Suppl.), 3–14.

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