Effect of limited mating on the corpora lutea in montane voles, *Microtus montanus*

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Conaway (1971) has distinguished 3 basic types of reproductive cycles displayed by nonpregnant female mammals. In the first type (e.g. in some hystricomorph rodents and higher primates), both ovulation and pseudopregnancy, defined as a functional luteal phase in a nonpregnant cycle, occur spontaneously. In animals with the second type of cycle (e.g. cats, rabbits) ovulation is induced and pseudopregnancy is spontaneous; the luteal phase following hormone-induced ovulation is indistinguishable from that following copulation. Rats and mice typify a third type of cycle in that ovulation is spontaneous, but the luteal phase is dependent upon copulatory stimulation. Recently, Milligan (1974, 1975) has reported a fourth variation: in *Microtus agrestis* low levels of copulatory stimulation induce ovulation, but the resulting corpora lutea (CL) degenerate about the 3rd day after mating, i.e. ovulation and activation of functional CL are dependent on copulation.

Induced ovulation has been demonstrated for several species of *Microtus* (Greenwald, 1956; Austin, 1957; Richmond & Conaway, 1969; Clulow & Mallory, 1970; Cross, 1972; Gray, Davis, Zerylnick & Dewsbury, 1974a; Gray, Zerylnick, Davis & Dewsbury, 1974b), but the precise requirements for the development of luteal function have been determined only for *M. agrestis*. No dissociation between the occurrence of ovulation and of functional CL has been found in *M. ochrogaster* (Kenney, Lanier & Dewsbury, 1977).

Because *M. agrestis* and *M. montanus* are reproductively similar in many other ways [e.g. large amounts of copulatory stimulation (Davis, Gray, Zerylnick & Dewsbury, 1974; Milligan, 1975); high LH levels after copulation (Charlton, Naftolin, Sood & Worth, 1975; Gray, Davis, Kenney & Dewsbury, 1976); occasional ovulation when separated from the male by a wire-mesh barrier (Gray *et al.*, 1974a; Milligan, 1974)], the influence of a limited amount of mating on luteal development of montane voles was examined.

All animals were laboratory-bred and had produced at least one litter. They were maintained on a reversed 16 h light:8 h dark cycle, and were given free access to Purina rabbit chow and water. Twenty female *M. montanus*, 3–6 months of age, were used. Fourteen males served as mating partners, and no male was used more than once in a group. Females were brought into oestrus by being placed opposite a male in a 48 × 27 × 13 cm divided cage. Daily vaginal smears were taken with a wire loop. Mating tests were initiated by placing the female in the 29 × 19 × 13 cm home cage of the male. If the female was judged unreceptive or the male failed to achieve intromission within 15 min the test was discontinued and discounted. Mating occurred about 2 h into the dark phase of the light cycle, on the 2nd day of vaginal oestrus (more than 50% cornified cells). After a positive mating test, the female remained in the male's cage, and the male was removed. *Microtus montanus* males have a mean ejaculation frequency of 5.0 preceding attainment of a satiety criterion (Dewsbury, 1973), and the proportion of females ovulating increases with the number of ejaculatory series received (100% ovulation following 3 or more ejaculations). Because one ejaculatory series does not consistently result in the production of CL that are maintained until Day 8 (Davis *et al.*, 1974), the mating stimulus was limited to one ejaculatory series. Females were killed on the 3rd (Group 1) or 8th (Group 2) day after mating. Ovaries and uteri were examined under a dissecting microscope and the number of CL in each ovary was recorded. To verify accuracy of CL counts, several ovaries were embedded in paraffin wax, serially sectioned at 10 µm, stained with haematoxylin and eosin and examined microscopically.

Of the 10 females killed on Day 3 (Group 1), 7 had well-developed CL (mean 4.3 ± 0.6 (S.E.M.)). In contrast CL (6) and implanted embryos (5) were present 8 days after copulation in only 1 of the

10 females in Group 2, and this difference in CL frequency was significant ($\chi^2 = 7.5$, $P < 0.01$). There was no difference in the amount of stimulation received by females in the two groups, the mean no. of intravaginal thrusts/test being 58.4 ± 4.2 for Day 3 and 62.1 ± 6.4 for Day 8. Similarly, there was no difference in number of thrusts received by females exhibiting CL at examination (59.5 ± 5.0) and those in which no CL were present (60.8 ± 5.4).

Montane voles are therefore like field voles in that limited amounts of mating produce ovulation and the resulting CL are short-lived. It is important to know which reflexly ovulating mammals show independence of ovulation and the formation of functional CL, as well as the correlates and adaptive significance of this dissociation.

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