these precipitates were formed in vivo during the uptake of I^- (*13*). As Tl, after saturation, can be presumed to be present all over the cytoplasm, the most obvious interpretation is that these precipitates mark the routes of I^- transport.

This first direct evidence for significant amounts of ions in the endoplasmic reticulum suggests that this membrane system plays a role in intracellular ion transport.

At present the role of compartmentation in ion transport in plant cells is being seriously discussed. Many authors have used the simple serial model advocated by Pitman (*14*), which implies that ions enter the vacuole via the bulk of the cytoplasmic content. However, kinetic evidence has suggested a more or less parallel relation between uptake into the cytoplasm and into the vacuole (*15*). Such a model requires a direct connection between plasmalemma and tonoplast, bypassing the cytoplasmic ion pool. It has been suggested that the endoplasmic reticulum and pinocytotic vesicles provide such a connection. Evidence for a close association between endoplasmic reticulum and tonoplast over large areas is of interest in this respect (*16*). Our findings lend support to the parallel model.

FRANK VAN IREN
ANTONIE VAN DER SPIEGEL
Ion Transport Research Group,
Botanisch Laboratorium,
Rijksuniversiteit, Leiden,
The Netherlands

References and Notes

1. H. Komnick, *Protoplasma* **55**, 414 (1962).
2. R. E. Bulger, *J. Cell Biol.* **40**, 79 (1969); M. A. Clarck and G. A. Ackerman, *J. Histochem. Cytochem.* **19**, 727 (1971).
3. B. P. Lane and E. Martin, *J. Histochem. Cytochem.* **17**, 102 (1969); A. L. Kierszenbaum, C. M. Libanati, C. J. Tandler, *J. Cell Biol.* **48**, 314 (1971).
4. R. F. M. van Steveninck, A. Chenoweth, M. van Steveninck, in *Ion Transport in Plants: Proceedings of an International Meeting, July 1972, Liverpool*, W. P. Anderson, Ed. (Academic Press, London, 1973), p. 25.
5. J. A. Zadunaisky, *J. Cell. Biol.* **31**, C11 (1966).
6. J. Ochi, *Histochemie* **14**, 300 (1968); M. J. Legato and G. A. Langer, *ibid.* **41**, 401 (1969).
7. G. G. J. Bange and F. van Iren, *Acta Bot. Neerl.* **19**, 664 (1970).
8. W. E. Schmidt, *Trans. Ill. State Acad. Sci.* **60**, 61 (1967).
9. P. J. Gehring and D. B. Hammond, *J. Pharmacol. Exp. Ther.* **155**, 187 (1967); I. Rusznyák, L. György, S. Ormai, T. Millner, *Experientia* **24**, 809 (1968); J. P. Manners, C. K. Morallee, R. J. P. Williams, *Chem. Commun.* **16**, 965 (1970); J. Solt, H. Paschinger, E. Broda, *Planta* **101**, 242 (1971); A. M. Potts and P. C. Au, *Invest. Ophthalmol.* **10**, 925 (1971); C. K. Pallaghy, *Aust. J. Biol. Sci.* **25**, 415 (1972).
10. Z. Böszörményi and E. Cseh, *Plant Physiol.* **14**, 242 (1961); *ibid.* **17**, 81 (1964).
11. Excised roots of 8-day-old seedlings of barley (cultivar Herta) of low salt content were obtained as described in (*7*). After incubation (4 hours) in 0.1 m*M* Tl_2SO_4 + 0.1 m*M* $Ca(HCO_3)_2$ (aerated, *p*H 7) in the dark at 25°C and rinsing (1 minute) in demineralized water, roots were incubated for 30 minutes in 5 m*M* NH_4I, 0.1 m*M* $Ca(HCO_3)_2$, and 5 m*M* $CaSO_4$ (aerated, *p*H 7).
12. Specimens were fixed for 2½ hours in phosphate-buffered OsO_4 (1 percent, *p*H 7.4) at 4°C. They were dehydrated for 2½ hours in dry ethanol vapor as described by P. Sitte [*Naturwissenschaften* **17**, 402 (1962)]. The total fluid volume was less than 0.5 ml per 5 to 20 cm (length) of root. Specimens were embedded in ERL-4206 mixture [composition intermediate between A and B described in A. R. Spurr, *J. Ultrastruct. Res.* **26**, 31 (1968)]. Except for the ERL all media contained 5 m*M* NH_4I to prevent solution of the precipitates. Ultrathin sections were collected on a saturated solution of TlI. Grids were quickly rinsed in isopentane to remove adhering solution.
13. In parallel experiments the amount of I^- taken up within 30 minutes appeared to be less than half the amount of Tl^+ absorbed in 4 hours. Therefore at the end of the I^- absorption period more than half the amount of Tl^+ present in the tissue must still have escaped precipitation. Loss of Tl^+ from root batches during subsequent fixation in the presence of 5 m*M* I^- appeared to be only about 8 percent, however. Consequently considerable amounts of Tl^+ must have been precipitated by the iodide in the fixative. Thus, the fixation we applied after the in vivo precipitation was a Komnick fixation. As stated in the introduction, results of the Komnick method are subject to fundamental criticism so we had to discriminate between precipitates formed in vivo and those formed during fixation. The latter were visualized by omission of the in vivo precipitation step from the whole procedure. Electron microscopic observation then revealed TlI particles in clusters near the cell wall or dispersed in the cytoplasm, but membrane-bound precipitates were never found.
14. M. G. Pitman, *Aust. J. Biol. Sci.* **16**, 647 (1963); *Plant Physiol.* **44**, 1417 (1969).
15. J. J. M. Hooymans, *Acta Bot. Neerl.* **13**, 507 (1964); G. G. J. Bange and J. J. M. Hooymans, in *Isotopes in Plant Nutrition and Physiology* (International Atomic Energy Agency, Vienna, 1967), p. 249; E. A. C. MacRobbie, *J. Exp. Bot.* **20**, 236 (1969); C. K. Pallaghy, U. Lüttge, K. von Willert, *Z. Pflanzenphysiol.* **62**, 51 (1970); J. W. F. Costerton and E. A. C. MacRobbie, *J. Exp. Bot.* **21**, 535 (1970); W. P. Anderson, Ed., *Ion Transport in Plants: Proceedings of an International Meeting, July 1972, Liverpool* (Academic Press, London, 1973), pp. 399–461.
16. B. A. Fineran, *J. Ultrastruct. Res.* **43**, 75 (1973).

17. We thank G. G. J. Bange for his valuable suggestions during the experiments and the preparation of the report, and R. J. Helder, K. R. Libbenga, and A. Quispel for their comments on the manuscript.

9 October 1974; revised 4 December 1974 ■

Male-Induced Pregnancy Termination in the Prairie Vole, Microtus ochrogaster

Abstract. *Postimplantation as well as preimplantation pregnancy in prairie voles can be terminated by replacing the original stud male with an unfamiliar male. The pregnancy is disrupted by the ensuing male-induced estrus. Females spontaneously abort their litters, become receptive, and successfully breed again 4 or 5 days after introduction of the new male.*

Olfactory, endocrine, and social factors involved in pregnancy block (the Bruce effect) have been extensively investigated (*1*). Preimplantation pregnancy block occurs in laboratory mice (*Mus musculus*) (*2*), wild house mice (*Mus musculus*) (*3*), prairie deer mice (*Peromyscus maniculatus bairdii*) (*4*), field voles (*Microtus agrestis*) (*5*), and meadow voles (*Microtus pennsylvanicus*) (*6*). Postimplantation pregnancy block in the prairie vole (*Microtus ochrogaster*) is described in this report; the known period of susceptibility to pregnancy block is thus extended to virtually all stages of pregnancy. This contrasts with the limited period for pregnancy block demonstrated in *Mus* (*7*). Other species in which block of pregnancy has been described have been investigated only during the preimplantation period.

Laboratory-born prairie voles were housed in 41 by 24 by 18 cm solid bottom cages supplied with wood shavings and straw and had free access to rabbit chow and water as described (*8*). All females were nulliparous and initially anestrous. Control females ($N = 28$) were paired with sexually inexperienced males of similar age (60 to 120 days). Experimental females ($N = 61$) were similarly paired but had their original mates replaced by unfamiliar males at intervals ranging from 5 to 19 days after first pairing. Pairing under the normal control breeding conditions results in a conception rate of 87 percent; hence, nearly all females were assumed to be pregnant. Gestation was judged to be the interval from pairing minus the 2 to 3 days required for estrus induction and breeding (*9*). Experimental females were thus divided among five groups assumed to be either 3 to 4, 8 to 9, 12 to 13, 14 to 15, or 16 to 17 days pregnant at introduction of the second male ($N = 10$, 14, 13, 12, and 12, respectively).

To verify actual breeding dates and to determine more precisely the duration of gestation before introduction of the second male, 36 percent of females were examined daily for a copulation plug or sperm in a vaginal lavage. The breeding performance and time to parturition in these handled females was the same as in those females not handled or examined. To further minimize possible handling effects (*9*), only a few females from each experimental group were inspected daily for vaginal bleeding and indication of breeding following introduction of the second male. Remaining females in each group and the controls were not handled except for brief daily inspection during late pregnancy to determine parturition date and litter size.

The distribution of intervals from pairing to birth of the first litter for controls and the five groups of experimental females is shown in Fig. 1. Intervals of less than 21 days after introduction of the second male result if the original pregnancy is maintained. Intervals equal to or longer than the normal gestation period of 21 to 23 days (*9*) result from the establishment of a second pregnancy after termination of the original pregnancy. Two control and two experimental females showed much longer intervals (33 to 40 days) than other females. These data are not included in the figure or in further analysis and are presumed to result from rebreeding with the same male after pseudopregnancy (*10*).

Of 26 control females, 22 produced litters 24.8 ± 0.3 days (mean ± standard error) after pairing. Litter size (counted within 24 hours of birth) averaged 3.3 ± 0.2 young.

Of 59 experimental females, 54 successfully littered; 15 maintained their first pregnancy while 39 produced litters after disruption of the original pregnancy. Unfamiliar males caused termination of the existing pregnancy in 81 percent of the females ranging from 3 to 15 days pregnant and in 36 percent of the females 16 to 17 days pregnant. In the females whose first pregnancy was disrupted, the interval from introduction of the unfamiliar male to birth of the litter was 26.1 ± 0.3 days and averaged 1.3 days longer than the interval in controls ($t = 3.30$, $P < .01$). The number of young (3.3 ± 0.2) in the litters of these experimental females did not significantly differ from that of controls ($t = 0.19$, $P > .50$).

Among females maintaining their first pregnancy in spite of introduction of an unfamiliar male, the interval from first pairing to birth averaged 23.9 ± 0.2 days, significantly shorter ($t = 2.27$, $P < .05$) than that of control females. Apparently, the introduction of unfamiliar males slightly shortens

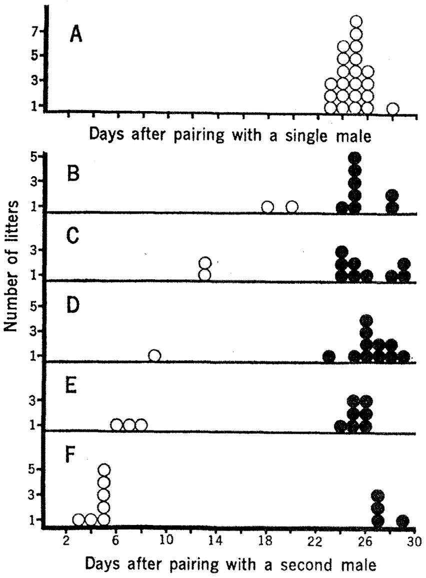

Fig. 1. Frequency of intervals from pairing until parturition in female *Microtus* after replacement of the original stud male by an unfamiliar male at various stages of pregnancy. Data for control females (exposed to only one male) are shown in (A). For experimental females, the original stud was replaced by an unfamiliar one when the females were 3 to 4 days pregnant (B), 8 to 9 days pregnant (C), 12 to 13 days pregnant (D), 14 to 15 days pregnant (E), and 16 to 17 days pregnant (F). Open circles represent first litters maintained; closed circles represent litters conceived after termination of the first pregnancy.

gestation if pregnancy is not interrupted. Litter size in these females (2.7 ± 0.3) tended to be smaller than that of controls, but the difference was not significant ($t = 1.70$, $P \simeq .10$). The proportion of young surviving until weaning was also reduced (75 percent compared to 86 percent weaned by control females) although the difference was not significant ($\chi^2 = 2.14$, $P \simeq .16$).

Termination of pregnancy is accompanied by a mucilaginous bloody discharge from the vaginal opening usually within 36 hours after introduction of the new male. Bleeding is observed even in females only 6 or 7 days pregnant due to the collapse of deciduae (*7*). Females in later stages of pregnancy often discharge amorphic pieces of embryonic, placental, or uterine debris from the vagina. Abortion of the litter is complete within 3 days after pairing with the second male and rebreeding occurs within 1 or 2 days after abortion.

The susceptibility of female *Microtus ochrogaster* during most of gestation to male-induced termination of pregnancy is in marked contrast to the limited period of susceptibility shown by *Mus*. At least 60 percent of *Mus* females up to 4 days pregnant show pregnancy block after 2-day exposure to strange males separated from the females by wire mesh. In females 5, 6, or 7 days pregnant the percent of blocked pregnancies drops to 38 percent, 18 percent, and 3 percent, respectively (*7*). Apparently, either the maintenance of pregnancy in female *Mus* becomes increasingly independent of any male-induced endocrine changes, or the female becomes unresponsive to olfactory stimuli from unfamiliar males. The decreased tendency of female *Mus* to show pregnancy block coincides with completion of endocrine events required for the decidual cell reaction 4 to 5 days after mating (*11*).

In *Microtus ochrogaster* 15 days of gestation must be completed before any reduction in the incidence of pregnancy termination is apparent. This suggests a marked difference between *Mus* and *Microtus* in the control of the endocrine balance necessary for the maintenance of gestation. Prolonged sensitivity or increased responsiveness of females to male stimuli may be a consequence of selection for the noncyclic, male-induced estrus period typical of *Microtus*. Pregnancy termination in *Microtus ochrogaster* apparently is an unavoidable side effect of the endocrine events associated with estrus induction. The introduction of a strange male has an overriding influence on the female regardless of the reproductive condition of the female.

Any comparisons between *Microtus* and *Mus* must be made cautiously. Experiments with *Mus* have been designed primarily to determine the role of olfactory stimuli; therefore, behavioral interactions allowing direct contact have been avoided. It is possible that the addition of tactile and other cues would change the response of pregnant *Mus* to a strange male. Should this be the case, the occurrence of male-induced pregnancy termination in somewhat more natural breeding situations may be more widespread than reported.

Male-induced termination of pregnancy remains to be demonstrated in wild populations. This is not unexpected because the frequency of abortion as a cause of prenatal mortality cannot be reliably estimated from necropsy of wild-trapped animals (*12*). Nevertheless, the rapid and consistent occurrence of abortion in the laboratory under relatively undisturbed and otherwise normal breeding conditions makes the occurrence in the wild at least plausible. Stable social relationships, as demonstrated by the family structure of *Microtus arvalis* (*13*), if disrupted, could lead to pregnancy termination in natural populations. Likewise, excessive immigration into resident populations could lead to increased occurrence of pregnancy termination. A continuously disrupted, shifting population probably would produce very few juveniles although the incidence of breeding and early stages of pregnancy might remain high. Thus, the establishment and maintenance of stable social and spatial relationships within a population of voles would be an important influence on birthrate. The implications of these findings relative to demographic change and gene flow in microtine populations remain to be investigated.

ROBERT A. STEHN
Field of Ecology and Evolutionary Biology, Cornell University, Ithaca, New York 14853
MILO E. RICHMOND
Cooperative Wildlife Research Unit, Department of Natural Resources, Cornell University

References and Notes

1. W. K. Whitten, *Adv. Reprod. Physiol.* **1**, 155 (1966); F. H. Bronson, in *Reproduction and Sexual Behavior*, M. Diamond, Ed. (Indiana Univ. Press, Bloomington, 1968), pp. 341; H. M. Bruce, *Br. Med. Bull.* **26**, 10 (1970).
2. H. M. Bruce, *Nature (Lond.)* **184**, 105 (1959); *J. Reprod. Fertil.* **1**, 96 (1960).
3. R. K. Chipman and K. A. Fox, *J. Reprod. Fertil.* **12**, 233 (1966).
4. B. E. Eleftheriou, F. H. Bronson, M. X. Zarrow, *Science* **137**, 764 (196[illegible] F. H. Bronson and B. E. Eleftheriou, *Gen. Comp. Endocrinol.* **3**, 515 (1963).
5. F. V. Clulow and J. R. Clarke, *Nature (Lond.)* **219**, 511 (1968).
6. F. V. Clulow and P. E. Langford, *J. Reprod. Fertil.* **24**, 275 (1971).
7. Vaginal bleeding, observed in *Mus* following exposure to strange males 5, 6, or 7 days after initial breeding, was suggested to be due to decidual collapse by H. M. Bruce [*ibid.* **2**, 138 (1961)]. In *Microtus*, histological examination in our laboratory has confirmed the collapse and sloughing of decidual tissue (R. A. Stehn, K. H. Martin, M. E. Richmond, in preparation).
8. M. E. Richmond and C. H. Conaway, *Lab. Anim. Care* **19**, 80 (1969).
9. ———, *J. Reprod. Fertil.* (Suppl. 6), 357 (1969).
10. M. E. Richmond, thesis, University of Missouri (1967).
11. B. M. Bindon, *J. Reprod. Fertil.* (Suppl. 18), 167 (1973); C. A. Finn and L. Martin, *ibid.* **39**, 195 (1974).
12. F. W. Brambell, *Biol. Rev.* **23**, 370 (1948); J. R. Beer, C. F. MacLeod, L. D. Frenzel, *J. Mammal.* **38**, 392 (1957).
13. F. Frank, *J. Wildl. Manage.* **21**, 113 (1957).
14. We thank K. H. Martin, D. Q. Thompson, and V. T. Harris for reviewing the manuscript. Supported by Department of the Interior award No. 14-16-0008-1037, a Ford Foundation grant to the Cornell Program in Pest Management Ecology and an NSF fellowship to R.A.S.

18 September 1974; revised 26 November 1974 ■

Josephson Junction Detectors: Geophysical Applications

In his review article on Josephson junction detectors (*1*), Clarke comments that the high magnetic field sensitivity of superconducting magnetometers utilizing flux transformers "is of limited use in an unshielded environment because of the noise in the earth's field." With these words the author dismisses, unsuspectingly I am sure, a large branch of geophysics in which this "noise" is of great interest and is studied intensively. In fact, Josephson junction devices are now being used to study fluctuations in the geomagnetic field and there is no doubt that the high sensitivity of the devices can be used to make unique measurements, particularly at frequencies near 5 hertz (*2*).

Even when the high magnetic field sensitivity of the Josephson junction magnetometers is unneeded, their other advantages over conventional systems for geomagnetic field measurement can make their use desirable. These advantages include a flat frequency response from very nearly zero frequency (d-c) up to frequencies in the kilohertz range, compactness (in some applications), and simple shielding of the sensors from external signals.

In addition to making possible studies of the fluctuations in the geomagnetic field, which provide information about the properties of the ionosphere and magnetosphere, Josephson junction magnetometers are also likely to play an important role in rock magnetism measurements (that is, in studies of continental drift) (*3*), in magnetotelluric work (which provides information about the structure of the earth's crust and upper mantle), and in communication systems with receivers that are deeply submerged in the sea (*4*). Two other geophysical applications are discussed by Clarke (*1*). Thus, I would further contest his statement that Josephson junction magnetic field gradiometers are "of far greater practical importance" than the magnetometers.

I do not intend my comments to detract from Clarke's interesting and timely review. Instead, I hope they will further illustrate the diverse and far-reaching application of Josephson junction detectors.

A. C. FRASER-SMITH
Radioscience Laboratory, Stanford Electronics Laboratories, Stanford University, Stanford, California 94305

References

1. J. Clarke, *Science* **184**, 1235 (1974).
2. J. L. Buxton and A. C. Fraser-Smith, *Inst. Elec. Electron. Eng. Trans. Geosci. Electron.* **GE-12**, 109 (1974).
3. W. L. Goodman, V. W. Hesterman, L. H. Rorden, W. S. Goree, *Proc. Inst. Elec. Electron. Eng.* **61**, 20 (1973).
4. S. A. Wolf, J. R. Davis, M. Nisenoff, *Inst. Elec. Electron. Eng. Trans. Commun.* **COM-22**, 549 (1974).

5 July 1974

Fraser-Smith's criticism is, I feel, a fair one. I was trying to make the point that, although a substantial improvement in magnetic field sensitivity can be made with the aid of a flux transformer, this *improvement* is of limited use in an unshielded environment. I share Fraser-Smith's enthusiasm for the use of Josephson junction magnetometers in geophysics.

JOHN CLARKE
Department of Physics, University of California, Berkeley 94720

18 December 1974