Table 2 Role of mating plug in mating-induced decline in female attractivity

Treatment	*N*	No. of females courted
Pre-test:		
Untreated	18	18
Test:		
Cloacal wash		
mated female	7	1*
unmated female	7	7
Distilled water	4	4
Post-test:		
Alcohol–water wash	18	15†

Females were tested for sexual attractivity using protocol described in Table 1. In the first of three consecutive courtship tests, the sexual attractivity of the females was established. Cloacal material from recently mated females or unmated females, or distilled water, was then applied to the dorsal surface and the females were retested for attractivity. Females were then washed with alcohol (90%) followed by distilled water to remove the applied substance before being retested for attractivity. *N*, no. of animals tested.

*Male courtship weak.

†Of the three non-courted females, two were not courted during the post-test period because of the decline in male courtship behaviour as a result of the exposure to mating plug material (see text).

These field observations of male dispersal from the mating pair suggest that the mating male deposits an odour(s) during copulation which communicates to other males the mated status, and hence non-receptivity, of the female. Thus, it is possible that the mating plug may, in addition to aiding in sperm retention, enable other sexually active males to determine whether a particular female has mated recently.

The experimental finding that exposure to odour(s) secreted during copulation (presumably by the male's urogenital system) results in sexually active males 'losing interest' in unmated, sexually attractive females, however, suggests an alternate explanation for male dispersal from the mating ball. In those insects having dense mating aggregations in which the probability of multiple inseminations is high (intense sperm competition), mechanisms have evolved which reduce the likelihood of multiple inseminations[19]. It is possible that, in *Thamnophis*, the odour(s) deposited by the mating male maximise the male's reproductive success by subsequently decreasing female attractivity as well as temporarily removing other males from the breeding population (that is, while the recently mated male is sexually refractory and unable to mate). Male dispersal from the mating ball, therefore, could be a consequence of unsuccessful males avoiding these odour(s). The demonstration and clarification of either or both of these postcopulatory adaptations must await detailed field studies of *Thamnophis* sexual behaviour.

Field observations also indicate that females leave the vicinity of the hibernaculum soon after emergence and copulation[17]. Since copulation is prolonged and conspicuous in *Thamnophis*, the mating pair is presumably very vulnerable to predation at this time. Mating-inhibition of further female sexual attractivity and receptivity, therefore, may act along with the female's departure from the hibernaculum area to minimise the female's vulnerability to predation (see also refs 15, 17).

Although our findings may be due to the restricted testing conditions, they demonstrate that sexually active males do not court sexually attractive females after prolonged or forced exposure to copulation odour(s). The specificity of this decline in male courtship behaviour as a consequence of exposure to a mated female is indicated by the observation that such males continue to investigate introduced snakes and trail-follow. It would thus be of interest to know whether this cessation of courting behaviour is due to a change in sensitivity at the peripheral or central level. In any case, the demonstration in vertebrates that the mating plug of one male can influence the behaviour of other males toward other females is, to the best of our knowledge, unique.

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Copulatory plugs, restricted mating opportunities and reproductive competition among male garter snakes

REPRODUCTIVE competition for females among males of a given species can take many forms, some of which (like mating plugs) may place constraints on the mating opportunities of males and intensify competition for available females. I have studied the reproductive behaviour of two garter snake species (*Thamnophis sirtalis* and *T. butleri*) in the field for 5 years. They do not exhibit pair-bonding, paternal care of offspring, territoriality, or intrasexual combat. Sexual selection in this mating system should favour males which compete to locate, court and mate with the maximum number of females[1]. Yet I have observed numerous solitary female *Thamnophis* within the same time periods and areas in which other females were being vigorously courted, often by several males. I have previously observed that the anterior cloaca of any recently mated female *Thamnophis* contains a copulatory plug which occludes the oviductal orifices[2]. The plug evidently is formed by the copulating male after sperm transfer. I interpreted this as a form of intrasexual competition in which the successfully copulating male makes the female temporarily unavailable to other males and reduces the likelihood of multiple inseminations. Some reports[3,4] indicate that multiple inseminations are possible. I present here evidence from the field that male garter snakes recognise females with a copulatory plug and behave as if these females were unavailable.

The pattern of emergence from hibernation and mating has been described for *T. sirtalis* in Canada[5–8], but my own observations differ in several important respects. Mating activity in southeastern Michigan is most intense on warm days in March and April immediately following the emergence of these snakes from hibernation, but before they disperse from the denning area. A denning area in Michigan is a collection of separate refuges, such as crayfish or rodent burrows, no one of which is used by all of the snakes at

that area. Males are conspicuous throughout the mating season, either courting or actively moving through the denning area evidently searching for females which might be emerging from any of the refuges. The emergence of females from hibernation is sporadic with individuals appearing on different dates. Females showing evidence (the copulatory plug) of recent mating do not immediately disperse from the denning area. These females, however, were never being courted when I encountered them. By contrast, those females which did not have a plug were usually being courted (Table 1).

The hypothesis that females with cloacal plugs are unattractive to males was tested in the field. Individual male snakes were given the opportunity to court first a female with a plug and then a female without a plug. The results (Table 2) clearly show that sexually active males can recognise and are disinclined to court females with plugs. All of these males subsequently courted the females without plugs.

Table 1 Observed mating status and courting activity of female garter snakes

	No. being courted	No. not being courted	χ^2
T. sirtalis			
without plug	21	6	27.2
with plug	0	19	
T. butleri			
without plug	14	3	9.9
with plug	0	4	

Values are the total number of females observed at the mating areas during the mating seasons from 1974 to 1976. Mating season is defined for each year as the time from the first observed courtship to the last.

A male can increase his opportunities for mating by minimising the time spent in unproductive courtship. It would be advantageous for a male to recognise and disregard females with plugs if plugs effectively prevent further inseminations. Among males in competition to locate unmated females, those that can recognise the altered mating status of a female as soon as possible should be favoured. Frequently, several males court a female simultaneously. When one mates, the unsuccessful males tend to disperse, presumably seeking other females, before their successful rival even finishes mating. I have seen this occur three times in the field and similar reports from the field[9] and the laboratory[10,11] exist. Even the successful male should soon be seeking other females. In at least four cases, males that I had observed mating were courting another female within an hour. More extensive data on males mating more than once in one afternoon have been reported[12].

Table 2 Responses of male garter snakes to females with or without copulatory plugs

Species	*N*	Courted both	Courted neither	Courted only female with plug	Courted only female without plug	Probability
T. sirtalis	11	2	0	0	9	<0.01
T. butleri	13	0	0	0	13	<0.001

The mid-body portion of a female with a plug was carefully set in front of each male. Both the head and anal portions of the female were held by hand to prevent her retreat into the undergrowth. If the male contacted the female with his tongue and initiated normal courtship by aligning his body along the coils of the female's body with his chin pressed tightly against her dorsal skin, his response was scored as positive. A response was scored as negative only after a male contacted the female with his tongue several times and then ignored her. As soon as a score was obtained for a female with plug, she was removed and the procedure was repeated on the same male using a female without a plug. In this matched-pairs design, each male served as his own control. Probabilities are for a one-tailed sign test.

The mechanism by which male garter snakes recognise the altered mating status of females with plugs remains unknown. In my experimental field trials, the males did not seem to behave differently toward either female until tongue contact was made. The males made no attempt to investigate the female's cloacal area (which was within my hand) before courting or rejecting her. This suggests that a chemical cue which is not physically associated with the plug itself may be involved. I doubt that the cue could be from the plug itself inasmuch as that is a male product. If a male were capable of producing any substance which directly caused other males to disregard a female, it would be most advantageous to use it at the beginning of courtship to avoid competition from courting rivals, rather than reserving it for use (in plugs) after mating was completed. This is not observed. If this potential advantage ever existed for some males, competing males that disregarded male odours and courted anyway would be favoured. Alternatively, plugged females could find it advantageous to 'announce' their altered status, since courting aggregations may be just as conspicuous to potential predators as they are to human observers. If fruitful mating is unlikely, females should not risk predation. Therefore, the female is probably responsible for producing the cue.

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Chameleons use accommodation cues to judge distance

ANY animal that adjusts the focus of its eye lens has monocular distance information potentially available from its focusing mechanism. Such accommodation cues are independent of size and context, and thus could provide an objective measure of distance. Ingle has proposed that such information is used by monocular frogs[1] and toads[2], both of which he has shown to be capable of estimating distance with one eye[3]. There is no direct evidence, however, that any animal perceives distance as a function of the plane of focus of the image. The experiments described here demonstrate that chameleons do so in estimating the distance of their prey.

It has not been shown that chameleons judge distance when feeding; however, their method of catching insects seems to be sufficiently exacting to require accurate distance assessment: the tongue is shot out with remarkable speed ($\sim 5\ m\ s^{-1}$) to hit and retrieve insects at distances up to 1.5 times body length[4] (Fig. 1). The eyes of a chameleon normally move independently, each eye scanning (saccadically) over more than a hemisphere. While taking aim at an insect, however, both eyes are swivelled directly towards the target (Fig. 2), often prompting the assumption that these animals acquire distance information by some process of triangulation.