

Museum of Zoology, University of Michigan, Ann Arbor, Michigan,
and Museum of Vertebrate Zoology, University of California, Berkeley,
California

Sexual Behavior, Sexual Interference and Sexual Defense in the Salamanders *Ambystoma maculatum*, *Ambystoma tigrinum* and *Plethodon jordani*

By STEVAN J. ARNOLD

With 32 figures

Received: 3 March 1975

Abstract

Courtship behavior and ♂-♂ interactions in the salamanders *Ambystoma maculatum*, *Ambystoma tigrinum* and *Plethodon jordani* were investigated with the aim of determining their adaptive significance. The courtship behavior of approximately 30 pairs of *A. maculatum*, 10 pairs of *A. tigrinum* and 50 pairs of *P. jordani* was observed. Behaviors were recorded by verbal transcription, motion picture and still photography. *A. maculatum* ♂♂ court rapidly, deposit many spermatophores, have simple behavior and low success with each spermatophore. *P. jordani* ♂♂ court slowly, usually deposit a single spermatophore, have complex behavior and high success with each spermatophore. *A. tigrinum* is intermediate in these respects. ♂♂ of the two *Ambystoma* species actively cover the spermatophores of rival ♂♂. Male *A. tigrinum* and *P. jordani* dupe rival ♂♂ into unprofitable spermatophore depositions by mimicking female behavior. Male *A. tigrinum* actively transport the ♀ away from rivals. Male *P. jordani* chase rivals away from the ♀. A hypothesis is proposed which relates the evolution of courtship speed, spermatophore number and sexual defenses to the length of the courtship season, the probability of intrusion by rival ♂♂ and the sexual receptivity of the ♀.

I. Introduction

There are two distinct ways for an individual to promote its sexual success. An individual may directly increase the success of its gametes by engaging in sexual behavior or, since fitness is relative, the individual may advance its own success by detracting from the sexual success of others of the same sex. Behaviors of the latter kind may be called "sexual interference" and are a special category of what HAMILTON (1970) has termed selfish behavior. In the salamanders discussed here, modes of sexual interference include covering the spermatophores of rival ♂♂ and mimicry of female behavior. A third class of behaviors (e. g. transportation of the ♀ away from other ♂♂, biting and chasing other ♂♂ away from the ♀) may be interpreted as "sexual defenses" against the debilitating effects of sexual interference. Recognition of the last

two kinds of behavior may improve our understanding of the adaptive significance and evolution of sexual behavior.

My general goal in comparing three species is to discover what types of selection modify courtship behavior in salamanders. I have purposefully chosen three species that differ greatly in several aspects of courtship behavior (e. g. in the number of spermatophores deposited and the rapidity of courtship). Since the probability that the ♂ will inseminate the ♀ will often be the most important index of male reproductive success in a courtship encounter, I have focused on aspects of behavior that might influence this probability.

While this paper deals specifically with *Ambystoma maculatum*, *Ambystoma tigrinum*, and *Plethodon jordani*, there are less complete observations on relatives of each of these species that suggest that the discussions apply equally well to these relatives. Thus *Ambystoma annulatum* (SPOTILA and BEUMER 1970), *A. opacum* (NOBLE and BRADY 1933) and *A. texanum* (GARTON 1972) share many courtship traits with *Ambystoma maculatum* (e. g. brief interactions between the ♂ and the ♀, rapid deposition of many spermatophores). The courtship of *Ambystoma dumerili* (BRANDON 1970) and *A. mexicanum* (GASCO 1881) are nearly identical to *A. tigrinum* (e. g. transportation of the ♀ by shoving, spermatophore deposition in front of the ♀ during a tail-nudging walk). *Plethodon caddoensis*, *P. glutinosus* (ORGAN 1960a), *P. ouachitae*, *P. welleri* (ORGAN 1960b), and *P. yonahlossee* differ from *Plethodon jordani* only in certain minor aspects of courtship (ARNOLD 1972). So far as is known all plethodontid salamanders have lengthy courtships and typically deposit a single spermatophore at the end of a tail-straddling walk.

It is remarkable that there have been no additional studies of courtship in *Ambystoma maculatum* since WRIGHT and ALLEN's 1909 description, or of courtship in *A. tigrinum* since KUMPF's brief 1934 outline, since both species are widespread, locally abundant and attract considerable attention during the breeding season. ORGAN 1958 has given an excellent, detailed description of courtship in *Plethodon jordani*, and MACMAHON 1964 provided additional observations of courtship in the field.

II. General Biology

Ambystoma tigrinum is found from coast to coast in the United States with some major discontinuities in distribution. *Ambystoma maculatum* is broadly distributed in eastern United States, and *Plethodon jordani* is restricted to the Appalachian Mountains. Courtship occurs at night in all three species. Both *Ambystoma* species court and lay eggs under water in ponds, while *Plethodon jordani* courts and lays eggs on land.

During the summer and winter adult *Ambystoma maculatum* live underground. In early spring they migrate at night to small woodland ponds to breed. The critical environmental factors affecting the timing of migration appear to be temperatures above freezing in conjunction with moisture provided either by snow melt or rain (WRIGHT and ALLEN 1909; BLANCHARD 1930; BALDAUF 1952; WHITFORD and VINEGAR 1966). Most of the animals arrive at a pond on a very few successive nights or on each of a few nights separated by periods of no migration. Courtship occurs immediately upon arrival, usually in shallow water near the pond margins. The ♀♀ begin laying eggs a few days after migration begins. Egg masses are attached to submerged stems in the deeper portions of the pond. HUSTING 1965 found that 50% of

the population arrives at a pond in the first 5 nights of migration (average of 4 years). From his data we can guess that 50% of the population will have completed their courtship activities in the first 7—10 days of the courtship season, and that courtship in nearly the entire population will have been completed within 2—3 weeks. The adults then disperse from the pond and resume their fossorial lives. The larvae hatch in a few weeks after oviposition and transform after a period of 57 to 144 days (WILBUR and COLLINS 1973).

Ambystoma maculatum adults are surprisingly long-lived. Of 72 ♂♂ marked in a breeding population in Rhode Island, at least 87.5% survived to the next breeding season (WHITFORD and VINEGAR 1966). HUSTING 1965 estimated annual survivorship of ♂♂ in Michigan at 0.76 to 0.94 (depending on the method of estimation), and I was able to capture a few ♂♂ 11 years after HUSTING marked them as sexually mature adults. The time to reach sexual maturity is unknown, since mark and recapture studies have been restricted to breeding adults.

The preponderance of ♂♂ in breeding populations may be the result of higher male survivorship or attainment of sexual maturity at an earlier age than ♀♀ (HUSTING 1965). Only 36% of the ♂♂ and 32% of the ♀♀ in HUSTING's population migrated to the pond and bred in particular years. Since adults are known to be faithful to a particular pond (HUSTING 1965; WHITFORD and VINEGAR 1966) apparently most of the adults do not breed in consecutive years.

The life history of *A. tigrinum* is rather similar to *A. maculatum* but demography has been less studied. Adults are terrestrial during most of the year and apparently reside in mammal burrows (CALEF 1954; STEBBINS 1962). Adults migrate to ponds and breed in the early spring in Michigan. As in *A. maculatum*, the courtship season is brief.

Plethodon jordani completes its entire life cycle on land. ♀♀ probably lay their eggs in May and June and brood them until hatching in the late summer, but this is simply inference from HIGHTON's 1962a study of the closely related *P. glutinosus*. HIGHTON 1962a found that in northern populations of *P. glutinosus* only half of the mature ♀♀ oviposit each year and suggested that individual ♀♀ lay eggs every other year. This is probably the case in *P. jordani* as well. During the late summer courtship season many *P. jordani* ♀♀ of mature size do not have large ovarian eggs. In the laboratory these ♀♀ did not elicit persistent courtship and were not inseminated. These are apparently ♀♀ that have just finished brooding young. ♀♀ with large ovarian eggs were courted persistently and inseminated. Such ♀♀ probably lay their eggs the following spring. In contrast, nearly all mature ♂♂ in the population are capable of courting in a particular courting season (as indicated by their well developed mental glands). As a consequence, sexual ♂♂ outnumber sexual ♀♀ in the population, probably by 2 to 1. Plethodontids comparable in size and sympatric with *Plethodon jordani* reach sexual maturity in 3 to 5 years and then probably survive for no more than 7 years with annual survivorship between .38 and .57 (ORGAN 1961; HIGHTON 1962a).

In salamanders with internal fertilization, insemination is accomplished by means of spermatophores that are attached to the substrate. Salamanders of the genus *Euproctus* constitute the only known exception. In this genus the ♂ passes the spermatophore directly to the ♀ while clasping her (BEDRIAGA 1882, 1883, 1895, 1897; DESPAX 1923; DÄHNE 1926).

Salamander spermatophores are always two-part structures: a clear gelatinous base that is fixed to the substrate and an apical mass of sperm. The

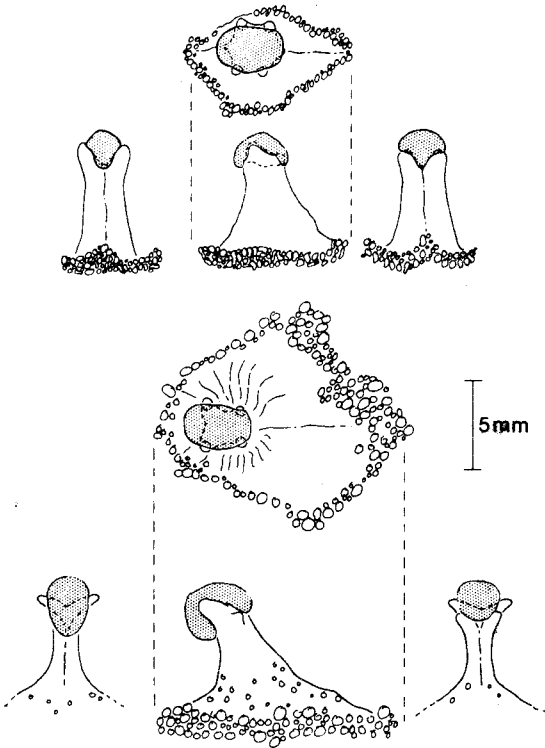


Fig. 1: Spermatophores of *Ambystoma maculatum* (above) and *Ambystoma tigrinum* (below). Anterior is to the left. Sperm masses are stippled

sperm mass is removed by the ♀ and the base remains attached to the substrate. In the genus *Ambystoma* the sperm mass is perched on a platform composed of the four knobs of the spermatophore base (NOBLE and BRADY 1933; UZZELL 1969; ANDERSON 1970, and Fig. 1). Female *Ambystoma* may remove all or only part of the sperm mass. In *Plethodon jordani*, and all other plethodontid salamanders, the spermatophore base has a laterally compressed spike that inserts into the sperm mass (ORGAN 1958 and Fig. 2). The sperm mass of *Plethodon jordani* is actual-

ly a rigid cap-like structure due to the thick capsules that surround it (ORGAN and LOWENTHAL 1963). The cap readily slides off the spike, and the whole sperm mass is removed by the ♀.

GASCO 1881, SMITH 1910, NOBLE and BRADY 1933, and UZZELL 1969 observed multiple spermatophores (Fig. 27) in *Ambystoma*, although apparently only GASCO observed how they were produced. SMITH 1910 considered the formation of multiple spermatophores in *Ambystoma maculatum* to be "a useless and wasteful procedure". Multiple spermatophores are, nevertheless, very common in the field (SMITH 1910; UZZELL 1969, pers. obs.). In salamanders such spermatophores are known only in the genus *Ambystoma*.

The symmetry of the spermatophore is correlated with an important aspect of courtship behavior. The spermatophore is almost radial in symmetry (Fig. 1) in species in which the ♀ may approach the spermatophore from almost any direction, as in *A. maculatum* (Fig. 31). If the ♀ always (*P. jordani*) or

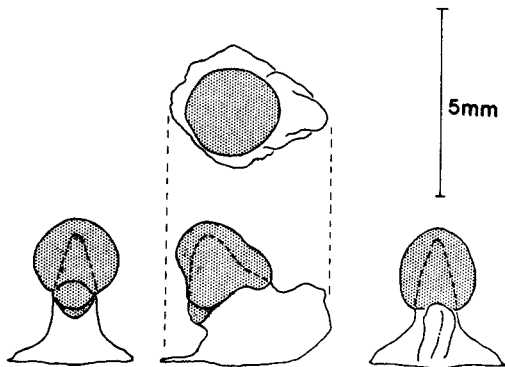


Fig. 2: The spermatophore of *Plethodon jordani*. Anterior is to the left. The sperm mass is stippled

usually (*A. tigrinum*) finds the spermatophore while following behind the ♂ (Fig. 31), the spermatophore is bilaterally symmetrical with the apex of the base pointing anterior. In these species the ♀ moves over the spermatophore from right to left in Fig. 1 and 2 and then settles backward and downward so that her force is distributed down the inclined axis of the base SCHALLER 1971 notes a similar correlation between spermatophore symmetry and uncertainty of ♀ approach in several groups of soil arthropods that deposit spermatophores on the substrate.

III. Material and Methods

Observations on *Ambystoma maculatum* are based on animals from two localities: Goss Pond, near Dixboro (southeast of the intersection of Goss and Earhart roads), Washtenaw Co., Michigan and the Edwin S. George Reserve (University of Michigan, Museum of Zoology), near Pickney, Livingston Co., the E.S.G.R. Most observations on *Plethodon jordani* are based on animals from the south side of Whitetop Mtn. (1280–1460 m), Grayson Co., Virginia. I observed courtship in animals from four other localities: 1.) Standing Indian Recreation Area, 11 km southeast of Hwy. 64, Macon Co., North Carolina (Fig. 18 is based on animals from this locality), 2.) 2.4 km north of Deep Gap, along the road to Deep Gap on Standing Indian Mtn., 1295 m, Macon Co., North Carolina (Figs. 15, 16, and 19 are based on animals from this locality), 3.) junction of Hwy. 441 and the road to Clingman's Dome, Swain Co., North Carolina, 4.) Highlands, Macon Co., North Carolina.

Ambystoma maculatum and *A. tigrinum* were collected as they migrated to breeding ponds in early spring (1969, 1970, 1971, 1972) either by patrolling the margins of the pond at night (Goss Pond) or by trapping animals at drift fences around ponds (E.S.G.R., through the courtesy of H. WILBUR and J. COLLINS). Consequently ♂♂ had not yet courted and ♀♀ had not yet been inseminated. All courtship observations were made within 10 days (and usually much less) of the date of capture. The animals were not fed during this time. From soon after capture until observations were made the animals were housed in damp leaf litter in a dark refrigerator at 4 °C.

Plethodon jordani were collected during the courtship season (1967, 1968, 1969) by turning surface objects during the day. Pairs were housed together continuously at a temperature of 13–16 °C on a natural photo-period. Animals kept for two weeks or less were not fed. Animals maintained for longer periods were fed houseflies (*Musca domestica*), blowflies (calliphorids) or crickets (*Acheta domestica*) once a week.

Courtship in the two *Ambystoma* species was observed with three laboratory or experimental setups and in the field. All observations were made at night.

1.) Animals were placed overnight in cylinders of aluminium window screen (50.8 cm in diameter by 30.5 cm high) with tops and bottoms of fiberglass window screen. The cylinders were half submerged in Burt Pond on the E.S.G.R. and the spermatophores were removed and counted the next morning (Tables 1, 2, 5). In one experiment (Table 1) the ♀ was separated within the cylinder by enclosing her in a smaller screen cylinder (10.2 cm in diameter by 25.4 cm high).

2.) Large aquaria (80 × 100 × 70 cm) with wooden floors were used for motion picture photography. Water temperatures were 13–16 °C. Motion pictures were made at night with illumination provided by one or two 500 watt flood lamps. The onset of continuous bright light tended to interrupt behavior during preliminary phases of courtship. *Ambystoma* continued to court under bright illumination, if light was increased gradually or courtship had progressed to spermatophore deposition or sperm transfer. I noticed no changes in courtship activities under these circumstances compared with activities in very dim light. Motion pictures were made with 16 mm film (Kodak 4X Reversal) at 18 or 22 fps with Bolex or Pathe cameras. Relatively long sequences (12–15 min) were filmed with a 400 ft. magazine.

3.) Observations of interactions between rival ♂♂ were made with animals courting in a metal washtub with a corrugated bottom (53.5 cm in diameter) and a water depth of 20 cm. Three pairs and three trios of *Ambystoma maculatum* (all ♂♂ were captured on the same night at E.S.G.R. while the ♀♀ were from Goss Pond) and one trio of *A. tigrinum* were observed under the same conditions within 5 days of the date of capture. During an observation session I verbally noted into a tape recorder whenever a ♂ began spermatophore deposition, whenever the ♀ found and squatted upon a spermatophore, and whenever interference occurred between animals. When two ♂♂ were courting a ♀ I was able to recognize each ♂ by differences in size or in the pattern of dorsal spots. I could therefore tell which ♂ was

depositing a spermatophore, but due to the number of spermatophores I could not tell whose spermatophore the ♀ had found. At the end of 90 min I separated the animals, examined the ♀'s cloaca for sperm and counted the number of spermatophores. For *Ambystoma maculatum* I later made paper records of the observations on an event recorder while listening to the magnetic tapes. The times between various behavioral events (Table 5) were then measured directly from these paper records. Water temperature was 8 °C.

4.) Field observations of courtship were made at night with white light illumination at several ponds in the vicinity of Ann Arbor, Michigan (*Ambystoma maculatum*) and at Ellicott Pond, Santa Cruz Co., California (*A. tigrinum*).

Most observations of courtship in *Plethodon jordani* were made by housing pairs in clear plastic shoe boxes (15 × 29 × 8.5 cm) with a substrate of damp paper towels. Interactions between ♂♂ were made by housing a few ♂♂ and ♀♀ in glass aquaria (25 × 50 × 30 cm or 30 × 60 × 30 cm) with the same substrate. All observations were made at night and the containers were illuminated with dim white light. Ongoing courtship was described in detail into a tape recorder and later transcribed. Still photographs of courting animals were made at night with a single lens reflex camera equipped with an electronic flash. The flash did not appear to affect courtship in any way. Some 16 mm motion pictures were made as described for *Ambystoma*.

The number of spermatophores per courtship in *Ambystoma* (Tables 1 and 2) was determined by removing the spermatophores from screen cylinders after one night of courtship and counting them under a binocular dissecting microscope. Counts of *Plethodon jordani* spermatophores were made each morning in each container housing a single pair and the spermatophores were then removed.

The probability that the ♀ will find a spermatophore per spermatophore deposition (Table 3) was estimated from motion pictures and tape recorded observations for *Ambystoma* and from tape recorded observations for *Plethodon jordani*. The total number of times the

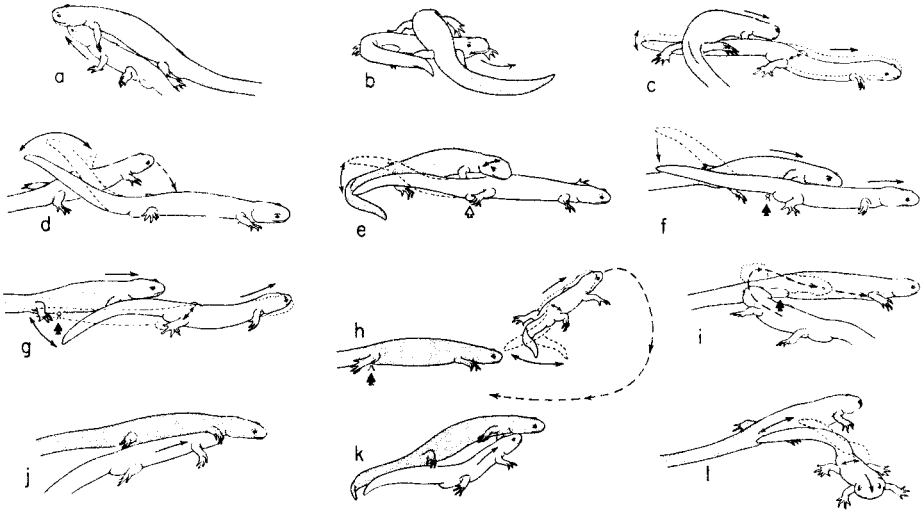


Fig. 3: Courtship activities in *Ambystoma maculatum*. a) The ♂ lifts his head under the ♀ (stippled) while moving forward (0 s). b) The ♂ turns under the ♀ and begins to move away (1.0 s). c) The ♂ moves away while shuffling his vent (3.0 s). d) The ♂ begins to deposit a spermatophore (7.4 s). e) The ♀ has turned towards the ♂ and rubs her snout on his dorsum (11.4 s). f) The ♂ moves away from the spermatophore (indicated with solid arrow head) and the ♀ moves forward parallel to him (15.7 s). g) The ♂ shuffles his vent as he moves forward. The spermatophore contacts the ♀'s body as she moves forward (18.5 s). h) The ♂ continues to shuffle his vent as he moves forward. The ♀ has found the spermatophore with her vent and pauses with the spermatophore inserted into her cloaca (23.1 s). i) The ♂ has approached the ♀ again and now nudges her with his snout along the path indicated by the dotted line. The ♀ remains over the spermatophore (39.9 s). j) The ♂ pushes his head under the ♀'s chest (41.7 s). k) The ♂ lifts under the ♀'s chin and dislodges her from the spermatophore (34.9 s). l) The ♂ again moves away from the ♀ and shuffles his vent (47.9 s). From a 16 mm movie; total elapsed time is 47.9 s

♀♀ found and squatted upon spermatophores was divided by the total number of spermatophores deposited by the ♂♂.

The success of the ♀ in actually retrieving sperm from each spermatophore could be accurately determined only for *Plethodon jordani* (Table 4). A sperm transfer was considered successful only if the ♀ departed with the sperm cap held in her cloaca. Sperm transfer was considered unsuccessful if the ♀ failed to find the spermatophore with her vent, if she did find it but then failed to remove the sperm cap, or if she removed the sperm cap but then lost it as she moved away from the spermatophore.

The courtship time per spermatophore deposition (Table 8) was estimated for each species by dividing the total elapsed time during which animals were observed courting by the total number of spermatophores deposited.

The sites of spermatophore deposition in relation to the ♀ were compiled from motion pictures for the two *Ambystoma* species and from tape recorded observations for *Plethodon jordani* (Fig. 31). The angle of the ♂ and his relative distance from the ♀ were noted at the onset of spermatophore deposition.

Measurements of tail and body length in *Ambystoma* were made on living, sexually mature animals recently captured during the courtship season. Body length was measured from the tip of the snout to the posterior margin of the vent. Tail length was measured from the posterior margin of the vent to the tip of the tail (Figs. 13, 14).

Drawings from motion pictures were made with the aid of a Vanguard motion picture analyzer. Spermatophores were preserved in 10% formalin and stained for observation with methylene blue. Drawings of spermatophores were made with a camera lucida.

I observed courtship in approximately 10 pairs of *Ambystoma tigrinum*, 30 pairs of *A. maculatum* and 50 pairs of *Plethodon jordani*.

IV. Descriptions of Courtship Behavior

A. *Ambystoma maculatum*

1. Catalog of Male Actions

Nudging. The ♂ contacts his snout to the lateral portions of the ♀'s body (Fig. 3i).

Head-swinging. The ♂ swings his head laterally to and fro in short arcs with his mentum or chin in contact with the ♀'s body.

Lifting. The ♂ lifts his head with the dorsum of his head or snout in contact with the ♀'s venter (Fig. 3k). The ♂ may simultaneously turn his head sharply to one side towards the ♀ (Fig. 3b).

Vent-shuffling. The ♂ shuffles his vent laterally by swinging the entire posterior body and tail from side to side. This action may be performed with the ♂ stationary or while moving forward slowly (Fig. 3c).

Begin spermatophore deposition. While stationary the ♂ presses his hindlimbs against his lateral tail base. The hindlimbs are slightly raised from the substrate, and the midbody is arched upwards slightly. The tail is level and may be slowly undulated (Fig. 3d).

Spermatophore deposition. Every postural component during spermatophore deposition promotes stability of the ♂'s vent and helps prevent other animals from dislodging the ♂ during the formation of the spermatophore. The forelimbs are outstretched. The body may be slightly curved laterally, or the body and tail may form an S with shallow inflexion. The midbody is arched slightly so that the vent is forced against the site of spermatophore deposition. Both in the field and in the laboratory I have observed ♂♂ clasping twigs or leaves with the hindlimbs as they deposit spermatophores (see also

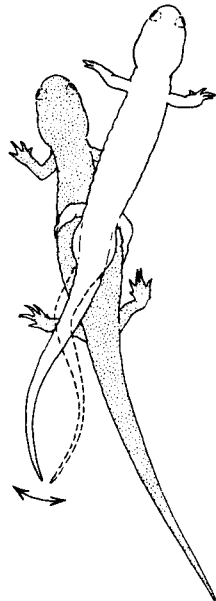


Fig. 4: A male *Ambystoma maculatum* depositing a spermatophore on the dorsum of the ♀. The ♂ is gripping the ♀'s body with his hindlimbs

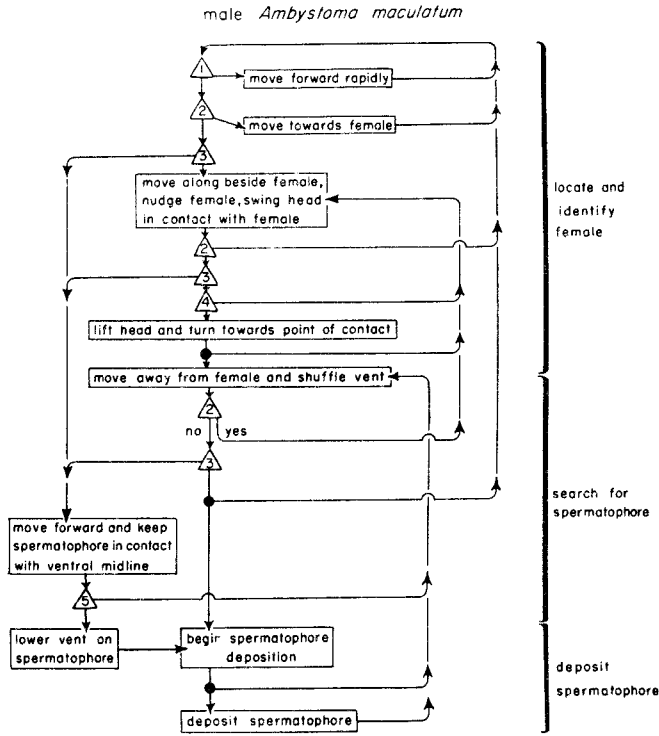


Fig. 5: An algorithm for courtship in male *Ambystoma maculatum*. Triangles represent the following questions: 1) Is the ♀ near? 2) Does the ♂'s snout contact the ♀? 3) Does a spermatophore contact the ♂'s venter? 4) Does the ♀ contact the dorsum of the ♂'s head? 5) Does the spermatophore contact the ♂'s venter? The arrows which exit the triangles on the right represent "no" answers, while arrows which exit on the left represent "yes" answers. The only exception is indicated in the figure. Solid circles represent stochastic branch points; points at which the ♂ may show two or more behaviors and the basis for the decision, if one exists, is unknown

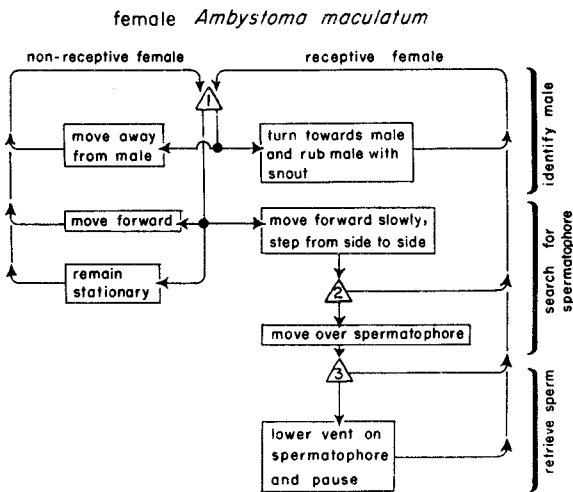


Fig. 6: An algorithm for courtship in female *Ambystoma maculatum*. 1) Does a ♂ contact the ♀ ("yes" exits on the right)? 2) Does a spermatophore contact the venter of the ♀? 3) Does the spermatophore contact the ♀'s venter? For questions 2 and 3 "yes" exits are on the left as in Fig. 5

WRIGHT and ALLEN 1909 and UZZELL 1969). Fig. 4 shows a rare extension of the same tendency. (Such clasping of the ♀ with the hindlimbs was observed only a few times but it provides a clue to the origin of clasping of the ♀ prior to spermatophore deposition. The latter kind of clasping is routine in some *Ambystoma* species.) The ♂ may also clasp a spermatophore with his hindlimbs when depositing a secondary spermatophore. When only a level surface is available for spermatophore deposition, as in my observation tanks, the ♂ presses his hindlimbs backwards towards his tail base as he deposits a spermatophore (UZZELL 1969).

2. Temporal Relations

a. *The Behavior of the ♂*

The temporal organization of the ♂'s behavior is shown diagrammatically in Fig. 5. The ♂'s behavior may be arranged in three broad categories: 1) location and identification of the ♀; 2) search for a spermatophore deposition site; 3) the deposition of new spermatophores. Courtship is exceedingly rapid (Fig. 30).

♂♂ appear to locate ♀♀ and other ♂♂ by the water currents produced by moving animals. For example the ♂ will suddenly turn toward an animal that moves behind him and out of his visual range. When the ♀ is more than a few body lengths away the ♂ appears unable to locate her by this means. Under these circumstances the ♂ moves about rapidly until he comes into the ♀'s proximity.

When the ♂ locates a ♀ he contacts her repeatedly with his head. He nudges her with his snout, swings his head back and forth on her dorsum and lifts his head under her chin (Fig. 3). Apparently the ♂ identifies the ♀ as a conspecific during these activities, probably by chemoreception. Whenever the ♂ encounters the ♀ after a brief absence during courtship he repeats these three activities. When the ♂ is housed with only a conspecific ♂ or a ♀ of the wrong species few if any spermatophores are deposited (Table 1).

The ♂ tends to turn towards an object that contacts the dorsum of his head. As a consequence the ♂ turns under the ♀'s chin as he moves anteriorly beside her (Fig. 3b) and under the ♀'s tail when he moves posteriad. Thus the ♂ may circle around and around the ♀ while contacting her continuously with his head.

Next the ♂ initiates a behavior that may be interpreted as a search for an appropriate site for spermatophore deposition and, in particular, as a search for other spermatophores. He moves away from the ♀ while shuffling his vent in contact with the substrate (Fig. 3c). The shuffling action causes the ♂'s body to contact a larger area of the substrate than does simple forward progress. I was unable to identify any behavior in the ♀ that triggers vent-shuffling in the ♂, but it is quickly abandoned if the ♂ contacts the ♀ with his snout. When this happens the ♂ again performs nudging, head-swinging and lifting.

When the ♂ finds a spermatophore he deposits a new spermatophore upon it (a "secondary" spermatophore). First the ♂ orients actively to the spermatophore that contacts his venter. He shifts his body towards and over the spermatophore as he moves forward so that it remains in contact with the midline of his venter. Sometimes the ♂ loses tactile contact with the spermatophore and reinitiates vent-shuffling. But if the spermatophore contacts his vent, the ♂ rapidly inserts the spermatophore into his cloaca by shifting back and forth while squatting upon it. The ♂ then deposits a new spermatophore that completely sheaths the original spermatophore and renders the original sperm inaccessible to the ♀ (Fig. 27).

The ♂ may deposit a spermatophore directly upon the substrate if he fails to find a spermatophore during vent-shuffling. Spermatophores are deposited upon the substrate ("primary" spermatophores) only after the ♂ has contacted the ♀ with his head, lifted under her chin and then moved away from her. In contrast, the ♂ will interrupt any courtship activity to deposit a secondary spermatophore if a spermatophore contacts his body (Fig. 5). Since the ♂ seems to require less information about the ♀'s position to deposit a secondary spermatophore, it is not surprising that secondary spermatophores tend to be deposited further from the ♀ than primary spermatophores (Fig. 31).

After depositing a spermatophore, the ♂ moves forward and shuffles his vent against the substrate. He may immediately deposit another spermatophore, if a spermatophore contacts his vent, without any intervening contact with the ♀. Otherwise the ♂ moves rapidly back toward the ♀ and resumes nudging and head-swinging (Fig. 3h).

Toward the end of lengthy courtship in which many spermatophores have been deposited the ♂ shows a rather sudden waning in the tendency to complete the events leading to spermatophore deposition (Fig. 23): he fails to begin spermatophore deposition after vent-shuffling, once he begins spermatophore deposition he may move away without depositing a spermatophore and when he finds a spermatophore he may ignore it. All of these tendencies emerge quickly, and the ♂ then stops responding to the ♀ altogether. The cessation of courtship is probably related to a temporary exhaustion of the supply of materials required to construct spermatophores, for when two ♀♀

Table 1: Number of spermatophores deposited under various conditions by *Ambystoma maculatum*

Experiment	Mean	Sample Size	Standard Deviation	Range	Mean per ♂	Statistical Tests ^a
1 ♂ + 1 ♀	40.41	29	18.79	10 - 81		
2 ♂♂ alone	1.33	6	3.27	0 - 8	0.66	F = 33.18 ** sex recognition T = 10.46 **
1 ♂ + 1 ♀ <i>A. tremblayi</i> ^b	2.38	13	3.04	0 - 8		F = 38.20 ** species T = 7.22 ** recognition
2 ♂♂ + 1 ♀ ^c	86.66	6	15.23	24 - 68	43.33	F = 1.52 ns effect of rival ♂ T = 0.36 ns
1 ♂ + 2 ♀♀	49.00	8	17.07	31 - 78		F = 1.14 ns effect of T = -1.01 ns additional ♀
1 ♂ + 1 enclosed ♀	5.0	9	6.30	0 - 19		F = 8.78 ** necessity of 8.68 ** contact
Correlation between number of spermatophores and size of ♂ r = 0.28 (n = 23) ns						
Correlation between number of spermatophores and size of ♀ r = 0.37 (n = 23) ns						

^a The statistical tests compare a particular experiment with the number of spermatophores deposited by a single ♂ courting a single ♀. When variances were unequal (F), Student's T (T) was corrected for unequal variances. In this and subsequent tables ** indicates statistical significance at the 0.01 level, * significance at the 0.05 level, and "ns" indicates lack of statistical significance at the 0.05 level.

^b *Ambystoma tremblayi* is a triploid, all female species sympatric with *A. maculatum* in Michigan (UZZELL 1964).

^c Based on actual observation of ♂♂ during courtship. These are the same ♂♂ as in Fig. 23. Thus sample size is 6 ♂♂ (3 trios) and the standard deviation and range are based on the scores of individual ♂♂. However, in the other experiment with two ♂♂ (second line) the animals were not observed and the statistics are for the combined scores of each of 6 pairs.

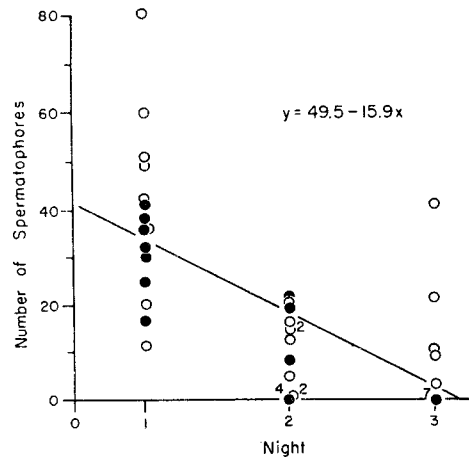
are housed with a ♂ he deposits no more spermatophores than with a single ♀ (Table 1).

Under experimental conditions male *A. maculatum* do not show a smooth adjustment in the number of spermatophores deposited per courtship (Table 1). ♂♂ produce either a full complement of spermatophores (mean of 40) or very few. The ♂ deposits very few or no spermatophores when a conspecific ♀ is replaced by a ♀ of another species, or by another ♂, or when the ♂ cannot actually touch the ♀. In the last case the ♀ was separated from the ♂ by screening, permitting water disturbances and any waterborne pheromones to reach the ♂. Since the ♂ requires actual contact with a conspecific ♀ before he will deposit spermatophores, in the field the ♂ probably does adjust the number of spermatophores he deposits according to the degree of interest shown by the ♀. The ♂ simply continues to deposit spermatophores as long as the ♀ remains close to him.

Surprisingly there is no relationship between the size of the ♂ or the size of the ♀, and the number of spermatophores in a courtship (Table 1).

The ♂'s supply of spermatophores is exhausted rapidly (Fig. 7). When ♂♂ are confined with ♀♀ on successive nights the number of spermatophores decreases at a rate of about 16 spermatophores per night.

Fig. 7: Exhaustion of spermatophore supply in *Ambystoma maculatum*. The numbers of spermatophores deposited by ♂♂ courting with the same ♀♀ on successive nights are shown with solid circles. Open circles show the numbers of spermatophores deposited by ♂♂ courting with non-inseminated ♀♀ on successive nights. The regression equation is for all data combined



An analysis of covariance with linear regressions indicates that the rate of decrease for ♂♂ courting the same ♀♀ on successive nights is not different from ♂♂ courting virgin ♀♀ on successive nights. On the third night a few ♀♀, which had courted the two previous nights, laid eggs. Impending or actual egg laying may have been responsible for fewer spermatophores with inseminated ♀♀ on the third night (Fig. 7). Unfortunately the responses of inseminated ♀♀ were not observed and I made only irregular notes about the timing of egg laying after insemination.

♂♂ apparently have a total supply of materials for less than a hundred spermatophores. A ♂ will invest nearly half of his total supply with the first ♀ he courts.

b. The Behavior of the ♀

The ♀'s activities may be grouped into three categories: (1) identification of the ♂, (2) location of spermatophores and (3) retrieval of sperm (Fig. 6).

A sexually receptive ♀ will turn towards the ♂ and nudge him with her snout whenever he contacts her. Perhaps the ♀ is able to identify the ♂ as a conspecific by nudging him, but I have no experimental evidence to prove this.

The ♀ searches for spermatophores by stepping from side to side with her hindlimbs while moving slowly forward. She shows this behavior whenever she is not in contact with the ♂. Once the ♀ finds a spermatophore she orients to it tactually, just as the ♂ would, and squats upon it. While resting on the spermatophore her posture is identical to the ♂ during spermatophore deposition, but she does not undulate her tail. As the ♀ moves off the spermatophore she arches her tail base.

Sometimes the entire sperm mass seems to be removed by the ♀, sometimes only a portion of it, and frequently the sperm mass of the spermatophore appears just as massive as before the ♀ squatted on it.

The ♀ finds and squats upon several spermatophores during a courtship (Fig. 24). Undoubtedly she removes sperm from more than one of these spermatophores. Like the ♂, the ♀ shows a sudden waning of responsiveness toward the end of a courtship. She pauses on spermatophores only briefly or ignores them altogether, and tends to ignore the ♂ or move away from him. Finally, the ♀ swims away from the ♂ and ignores him even if he locates her again.

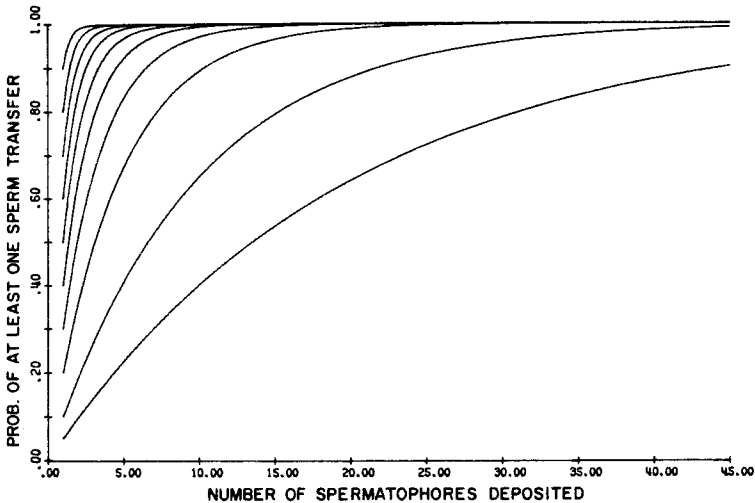


Fig. 8: The probability of sperm transfer as a function of the number of spermatophores deposited and the probability of success with each spermatophore. Each line shows the probability of at least one sperm transfer as a function of the number of spermatophores deposited for a particular probability of success with each spermatophore, p . The lowest line shows the function for $p = 0.05$, the next line for $p = 0.1$, the next for $p = 0.2$, etc.

Female *A. maculatum* find approximately 4 out of 10 spermatophores deposited by the ♂ (Table 4). Because one usually cannot tell by simple observation whether the ♀ removes sperm from a spermatophore, the probability of sperm transfer with each spermatophore found is unknown. Even if the probability of sperm transfer per spermatophore deposited is as low as 0.1, the ♀ will almost certainly be inseminated if she is exposed to 40 spermatophores (Fig. 8). In Fig. 8 the probability of at least one sperm transfer during a courtship was calculated as $1-(1-p)^n$ where n is the number of spermatophores and p is the probability of sperm transfer per spermatophore. This probability function could be tested directly by examining the cloacae of ♀♀ that had located a known number of spermatophores.

The masculine process of depositing spermatophores and the feminine process of finding them are quite independent. The ♀ may find or squat on a spermatophore while the ♂ is nudging her, head-swinging, lifting, vent-shuffling, depositing a spermatophore, or surfacing for air. While the ♂ deposits a spermatophore the ♀ may find a spermatophore, squat on a spermatophore, nudge the ♂, move about, or surface for air.

Frequently the ♀ is several cm or more from the ♂ when she finds a spermatophore. The ♀ may nudge the tail or tail base of the ♂ as he deposits a spermatophore, then move forward behind him and find the spermatophore he has just deposited (Fig. 3h), but this is rather exceptional.

Occasionally the ♂ may dislodge a ♀ from a spermatophore or the ♀ may dislodge the ♂ during spermatophore deposition.

When one animal has separated from the other sexual partner, it tends to turn and move back towards it. As a consequence the activities of courtship take place within an area 50 cm or less in diameter, even in a large container. When an animal surfaces for air during courtship, it tends to turn after gulping air and swim back down to the site of departure with great accuracy.

B. *Ambystoma tigrinum*

1. Catalog of Male Actions

The following actions are performed by the ♂ while facing towards the ♀.

Nudging. As in *A. maculatum*.

Head-Swinging. As in *A. maculatum*.

Lifting. As in *A. maculatum*.

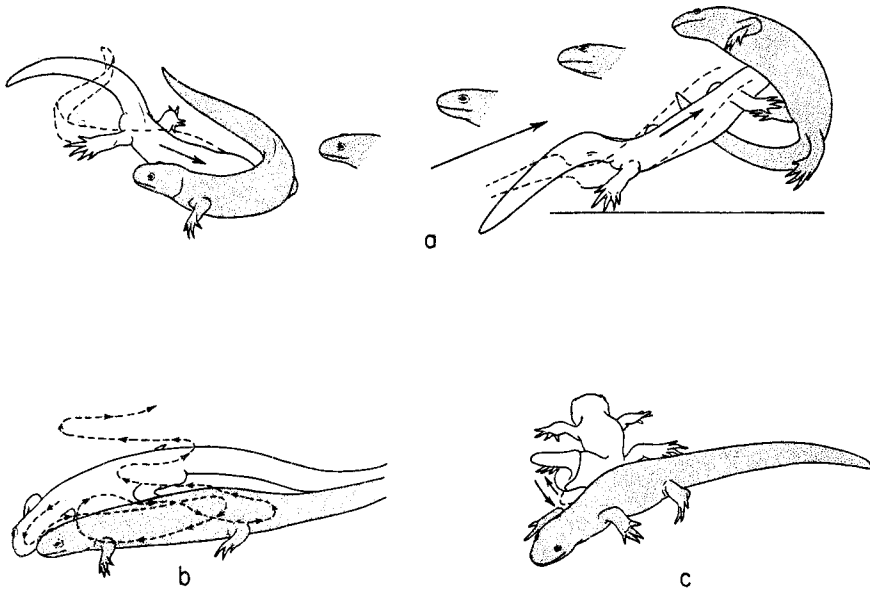


Fig. 9: Initiation of a tail-nudging walk in *Ambystoma tigrinum*. a) The ♂ transports the ♀ (stippled) by shoving her through the water (0—0.9 s). b) The ♂ rapidly slides his snout over the ♀'s dorsum and then moves away from her along the path indicated by the dotted line (43.3—56.2 s). c) The ♂ pauses while facing away from the ♀ and repeatedly taps his tail on her dorsum (60.6 s). From a 16 mm movie; total elapsed time is 60.6 s

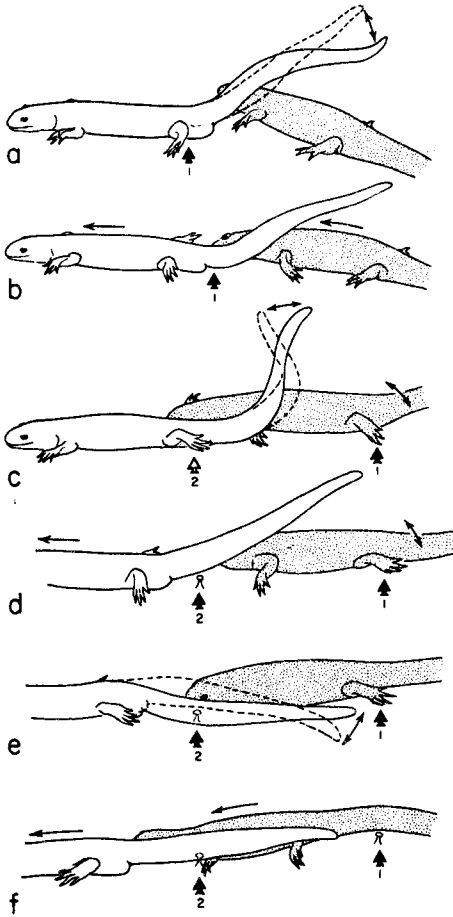


Fig. 10: Sperm transfer during the tail-nudging walk in *Ambystoma tigrinum*. a) The ♂ pauses and taps his tail on the dorsum of the ♀ (stippled). A spermatophore which was deposited earlier lies out of view behind the ♂ as indicated by solid arrow head 1 (0 s). b) The ♀ and ♂ are moving forward (2.4 s). c) The ♀ has moved over spermatophore 1, found it with her vent and now pauses with the spermatophore inserted into her cloaca. The ♂ is depositing another spermatophore (14.9 s). d) The ♂ moves away from spermatophore 2, while the ♀ remains stationary with spermatophore 1 still in her cloaca (22.8 s). e) The ♂ passes in front of the ♀ (26.0 s). f) The ♀ has moved off spermatophore 1 and nudges the ♂'s tail base. The ♂ moves forward (29.1 s). From a 16 mm movie; total elapsed time is 29.1 s

Shoving. The ♂ swims vigorously while pushing his snout against the lateral or ventral portions of the ♀'s body and consequently shoves the ♀ through the water for a distance of several cm to a m or more. Frequently the ♀'s entire body is suspended in the water as he shoves her rapidly forward and upward (Fig. 9).

The following actions are performed while facing away from the ♀.

Tail-tapping. While stationary with the tail directed straight backwards, the ♂ repeatedly raises and lowers his tail, tapping its ventral surface on the ♀'s body, or, less frequently, on the substrate. In addition to the tapping motion, the tail is undulated slightly (Fig. 10a).

Backing-up. The ♂ backs up rapidly a few cm with the tail level, or nearly so, directed straight backward.

Moving forward. The ♂ moves forward with the tail straight and directed backward, and may simultaneously raise and lower his tail.

Begin spermatophore deposition. The ♂ presses his hindlimbs towards his lateral tail base while arching the midbody slightly. The tail is raised 45–90 degrees and is undulated very rapidly.

Spermatophore deposition. The ♂ continues in the same posture with the forelimbs outstretched and raised above the substrate (Fig. 10). Tail undulations cease, the ♂ lifts his vent free of the spermatophore and moves forward slightly.

2. Temporal Relations

a. The Behavior of the ♂

The ♂'s behaviors may be categorized as follows: (1) location and identification of the ♀ as conspecific, (2) forceful transporting of the ♀, (3) a tail-nudging walk, during which the ♂ leads the ♀ as he (4) searches for an appropriate spermatophore deposition site and (5) deposits new spermatophores. The temporal organization of the ♂'s courtship is shown in Fig. 11.

Of these categories, forceful transportation of the ♀ and the tail-nudging walk are absent in *A. maculatum*, but just as in *A. maculatum*, *A. tigrinum* ♂♂ appear to use disturbances in the water, and possibly chemoreception, to locate the ♀ and to stay in close proximity to her. Thus, whenever the ♀ moves away from the ♂, he follows rapidly behind her and nudges her tail

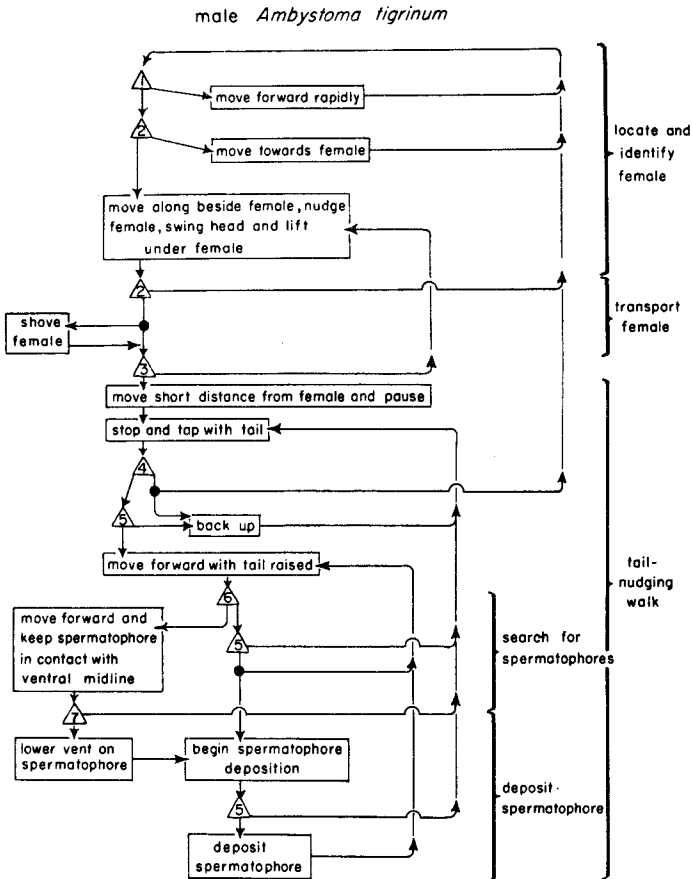


Fig. 11: An algorithm for courtship in male *Ambystoma tigrinum*. For all questions "yes" exits on the left, "no" exits on the right. All other conventions as in Fig. 5. 1) Is the ♀ near? 2) Does the ♂'s snout contact the ♀? 3) Does the ♀ contact the ♂'s head or anterior body? 4) Is there contact to the ♂'s tail? 5) Is there contact to the ♂'s tail base or cloaca? 6) Does a spermatophore contact the ♂'s venter? 7) Does the spermatophore contact the ♂'s vent?

intermittently. When she stops, the ♂ moves rapidly along her length and rubs her with his head (nudging, head-swinging and lifting).

Apparently the ♂ identifies his partner as a conspecific ♀ at this time. This is supported by two types of observations. First, the ♂ deposits few if any spermatophores with ♀♀ of the wrong species (see Table 2). Second, the ♂ can clearly distinguish a ♀ from a ♂. The ♂ will shove ♀♀ with his snout, but he does not shove and transport other ♂♂. Furthermore, when two ♂♂ are housed together in the absence of a ♀ no spermatophores are deposited (see Table 2).

Usually the ♂ shoves the ♀ just before he initiates a tail-nudging walk, but sometimes shoving is deleted.

The ♂ initiates the tail-nudging walk (Fig. 10) by suddenly departing from the ♀ and pausing with his vent less than one tail length from her. This departure only occurs if the ♀ is stationary. Next the ♂ monitors the ♀'s position with his tail. Tail-tapping apparently tells the ♀ where the ♂ lies, and the ♂ uses this behavior to elicit a following response from the ♀.

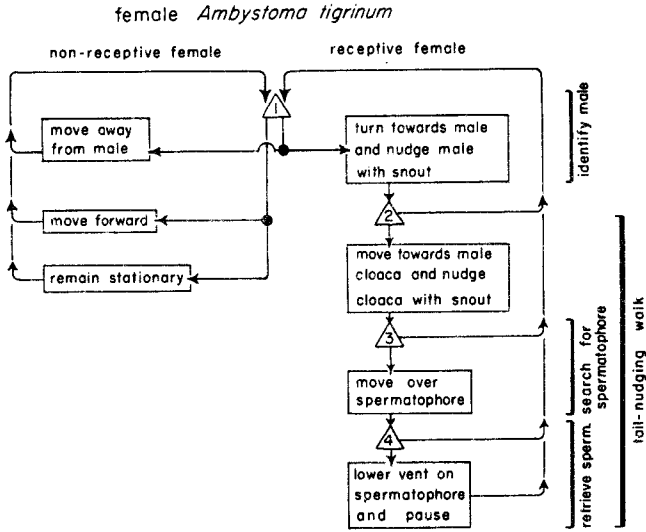


Fig. 12: An algorithm for courtship in female *Ambystoma tigrinum*. 1) Does a ♂ contact the ♀? (“yes” exits on the right) 2) Does the ♂’s tail tap on the ♀? 3) Does a spermatophore contact the ♀’s venter? 4) Does the spermatophore contact the ♀’s vent? For questions 2, 3 and 4 “yes” exits on left, “no” exits on the right

Table 2: Numbers of spermatophores deposited under various conditions by *Ambystoma tigrinum*

Experiment	Mean	Sample Size	Standard Deviation	Range	Mean per ♂	Statistical Tests ^a
1 ♂ + 1 ♀	20.59	32	6.44	8 - 37		F = 8.50 ** <i>A. maculatum</i> T = 5.40 ** vs <i>A. tigrinum</i>
2 ♂♂ alone	0.0	5				sex recognition
1 ♂ + 1 ♀ <i>A. maculatum</i>	3.71	7	6.47	0 - 18		F = 1.01 ns species T = 6.27 ** recognition
2 ♂♂ + 1 ♀ ^b	23.83	12	8.27	12 - 37	11.92	F = 1.65 ns effect of rival ♂ T = -1.37 ns
Correlation between number of spermatophores and size of ♂						r = -0.02 (n = 31) ns
Correlation between number of spermatophores and size of ♀						r = 0.12 (n = 31) ns

^a The statistical tests compare a particular experiment with the number of spermatophores deposited by a single ♂ courting a single ♀. All other conventions are as in Table 1.

^b The ♂♂ were not observed during courtship. Thus the sample size is 12 trios and the statistics and tests are for the combined scores of two ♂♂ from each trio. The tests show that the total number of spermatophores from two ♂♂ is no different than the score for one ♂. Thus ♂♂ inhibit each other and probably each ♂ deposits only half as many spermatophores as when alone with the ♀.

b. The Behavior of the ♀

A receptive ♀ will turn in the direction of the ♂’s tail during tail-tapping and move towards his cloaca. Whenever she contacts the vicinity of the ♂’s cloaca with her snout, the ♂ moves forward. The upward flexure of his tail exposes the papillae of his cloaca¹.

1) There is a dramatic difference between the cloacae of male *Ambystoma tigrinum* and male *Ambystoma maculatum*. Male *A. tigrinum* have a narrow band of finger-like papillae just inside the cloacal lips (i. e. along the entire ventro-lateral margin of the cloacal opening),

Secretions from these papillae may be wafted back towards the ♀ by the undulations of the ♂'s tail. At close range the ♀ could orient to pheromones from these papillae and at greater distances she could orient to the contact and water disturbances produced by the ♂'s tail. The persistent nudging of the ♂'s tail by a receptive ♀ causes the ♂ to continue forward and then stop and deposit a spermatophore. If the ♀ continues to follow and nudge the ♂, he continues forward and deposits additional spermatophores.

Simple contact with the ♂'s tail is sufficient to elicit the entire process of spermatophore deposition. Once, when a ♀ failed to follow a ♂, I reached into the aquarium and gently touched his tail base with the bulb of a thermometer. Repeated touching of the ♂'s tail base caused him to move forward and the absence of contact caused him to pause. After intermittent forward progress the ♂ stopped and deposited a spermatophore as I held the thermometer in contact with his tail base. The ♂'s behavior was indistinguishable from his behavior in the presence of a cooperative ♀. ♂♂ can be fooled in this way only if they have just turned away from the ♀ and are tail-tapping. If the ♂'s tail base is touched in the same fashion at any other time, he simply turns back towards the object that is touching him.

A non-receptive ♀ will not follow the ♂ and consequently the ♂ does not deposit spermatophores. If the ♀ fails to follow the ♂, either because she is non-receptive or has strayed from the ♂, the ♂ will back up slightly and resume tail-tapping. If she still fails to touch his tail or cloaca, the ♂ turns around and searches for her. When he locates her he renews the courtship by rubbing the ♀ or shoving her. Thus a non-receptive ♀ will cause the ♂ to cycle through his rubbing, shoving and tail-tapping behaviors. After several such cycles the ♂'s interest in the ♀ wanes and courtship terminates with no, or very few, spermatophores having been deposited. This system for elimi-

whereas male *A. maculatum* lack these papillae. Gross dissection reveals that these papillae serve as the exit points for long tubular glands that are housed in the ventral portion of the swollen cloacal lips. The morphological identity and function of these glands is open to question in the absence of a thorough histochemical survey of cloacal glands in ambystomatid salamanders. In *A. tigrinum* the glands that open on the papillae fringing the cloaca are not differentiated in gross structure from the cloacal glands that exit on ridges within the cloaca and form the spermatophore base. NOBLE and BRADY (1933) distinguished four types of glands in the cloaca of *A. opacum* on the basis of histochemical differences in secretions: the *pelvic glands* on the cloacal roof, that produce secretions which appear to cement the sperm mass to the apex of the spermatophore base (these glands are easily observed in *A. maculatum* and *A. tigrinum*); the *scaffolding glands*, on either side of the pelvic glands, participate in the formation of the spermatophore base; the *cloacal glands* (sensu stricto) line the cloacal walls and are responsible for the bulk of the spermatophore base; and the *abdominal glands*, which empty on the cloacal lips distal to the cloacal glands. In site of exit these "abdominal glands" correspond to the glandular, finger-like papillae of *A. tigrinum*, but *A. opacum*, like *A. maculatum*, lacks such papillae. In some salamandrids the abdominal glands exit on long papillae on the posterior lip of the cloaca (ZELLER 1905). Experiments with aquatic salamandrids indicate that the ♀ orients the pheromones from the ♂'s abdominal glands as well as to the water current produced by his undulating tail (ROGOFF 1927; PRECHTL 1951). If NOBLE and BRADY were correct in their identification of abdominal glands in *Ambystoma opacum*, then the finger-like papillae of *A. tigrinum* ♂♂ may release a pheromone that serves to orient the ♀ during the tail-nudging walk. This interpretation is supported by the tail raising behavior of male *A. tigrinum*, which exposes the papillae, as well as by the accompanying tail undulation that might serve to waft the pheromone posteriorly toward the ♀. In the absence of either experiments that would test the pheromone hypothesis or a histochemical survey that would establish the homology of glands in ambystomatids and salamandrids, at least two other functions are possible. The fringing papillae in *A. tigrinum* may simply form the basal part of the spermatophore or they may serve a tactual function as the ♀ follows during the tail-nudging walk.

nating spermatophore depositions with non-receptive ♀♀ is not completely fool-proof, however. When the path of a non-receptive ♀ is constrained, as for example by the side of the aquarium, she may push along behind the ♂ and cause him to deposit one or more spermatophores, but show no interest in them.

The tail-nudging walk also coordinates the positions of the ♂ and a receptive ♀ before and during spermatophore deposition. The ♂ monitors the ♀'s position with his tail and does not deposit spermatophores unless the ♀ is directly behind him. *Ambystoma maculatum* ♂♂ do not require following by the ♀ and deposit spermatophores in a much wider variety of sites (Fig. 31).

The special importance of the ♂'s tail in courtship is reflected in the size and shape of the tail. Male *A. tigrinum* have proportionately longer tails than

♀♀, but this is not the case in *A. maculatum* (Fig. 13, 14). In addition, the tail tip is spatulate in lateral outline and the whole tail is higher and more compressed than the ♀'s tail. *A. maculatum* shows no such sexual dimorphism in tail shape. These structural modifications of the ♂'s tail in *A. tigrinum* are undoubtedly related to its use both in the tail-nudging walk and in shoving the ♀.

When following the ♂, the ♀ moves forward slowly and steps from side to side with her hindlimbs. When

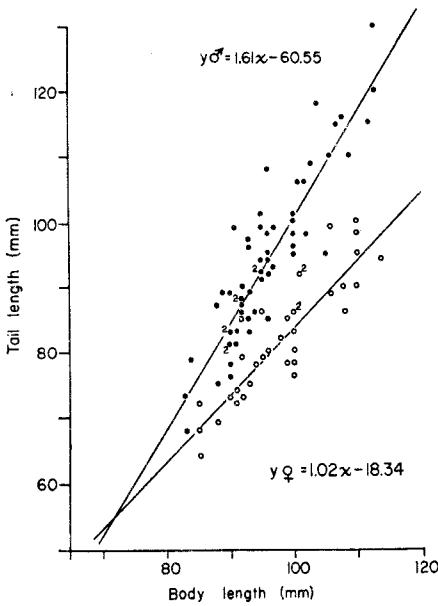
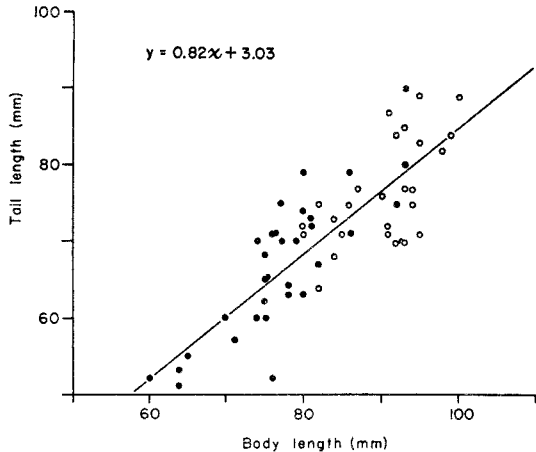


Fig. 13: Sexual dimorphism of tail length in *Ambystoma tigrinum*. Tail length as a function of body length for 61 ♂♂ (solid circles) and 35 ♀♀ (open circles) from Livingston Co., Michigan. Analysis of covariance shows that the slopes for ♂♂ and ♀♀ are significantly different; $F_{(1,92)} = 15.78, p < 0.01$

Fig. 14: Lack of sexual dimorphism of tail length in *Ambystoma maculatum*. Tail length as a function of body length for 32 ♂♂ (solid circles) and 27 ♀♀ (open circles) from Livingston Co., Michigan. Analysis of covariance shows no significant difference in slope between ♂♂ and ♀♀, $F_{(1,55)} = 1.25, p > 0.1$, and no difference in elevation, $F_{(1,56)} = 1.95, p > 0.05$



the ♂ pauses and begins spermatophore deposition, the ♀ usually stops behind him with her snout in contact with his tail base or cloaca.

As in *A. maculatum* the ♀ appears to find spermatophores by touch. If a spermatophore contacts her vent, she shuffles her vent laterally over the top of it, then squats upon it and inserts the spermatophore into her cloaca. She then assumes the postures taken by the ♂ during spermatophore deposition. She presses her hindlimbs against her tail base, arches her body slightly and undulates her raised tail rapidly (Fig. 10c). After several seconds she ceases tail undulations, moves forward off the spermatophore, and nudges the ♂'s tail base or cloaca, if he is still in front of her.

Although the ♀ usually finds spermatophores while following behind the ♂, the spermatophore she finds may not be the one just deposited by the ♂. The ♀ may find a spermatophore deposited much earlier by the ♂, if she passes over it while following behind him (Fig. 10). The ♀ may also find spermatophores as the ♂ performs nudging, head-swinging or lifting, but she cannot find spermatophores while the ♂ is shoving her.

Like male *A. maculatum*, male *A. tigrinum* will orient to and deposit a new spermatophore on a spermatophore that contacts the ventral or lateral portions of the body, but the ♂ will not orient to spermatophores while performing the actions of nudging, head-swinging, lifting or shoving.

C. *Plethodon jordani*

Despite its rather restricted geographic range, *Plethodon jordani* shows marked geographic variation in morphology, particularly in various aspects of coloration (HIGHTON 1962b, 1970). I have observed courtship in animals from four of the geographic isolates recognized by HIGHTON but was unable to detect any geographic variation in courtship behavior.

1. Catalog of Male Actions

The following actions are performed by the ♂ while facing towards the ♀.

Tapping. The ♂ rapidly lowers and raises his head, and consequently his nasolabial cirri tap the dorsum of the ♀ or the substrate.

Head-sliding. The ♂ moves forward while sliding his mentum in contact with the ♀'s dorsum.

Nudging. The ♂ contacts his snout to the lateral parts of the ♀'s body.

Foot Dance. The ♂ raises and lowers his forelimbs and hindlimbs. Only one limb is raised and lowered at a time. There is no obvious patterning to the sequences of limb movements. This action is performed when the animal is stationary or moving forward very slowly. ORGAN (1958) termed this action "foot dance".

Lifting. While turning his head towards the ♀, the ♂ lifts his head and contacts his snout or the dorsum of his head to the ♀'s chin. Lifting is usually performed with the ♂'s body parallel to the ♀ and facing in the same direction (Fig. 15) but it may also be performed with the ♂ facing the ♀ head on.

The following actions are performed by the ♂ while facing away from the ♀.

Stationary with tail arched and undulating. While stationary, the ♂ arches the proximal part of his tail. His body and vent are raised above the substrate (Fig. 16). The ♂ may or may not undulate his tail slowly.

Turning back towards the ♀. This is actually a complex series of actions (Fig. 18). The action begins with the ♀'s chin resting on the proximal part of the ♂'s tail. The ♂ faces straight away from the ♀ while arching and undulating his tail. Abruptly the ♂ turns his head and forebody back towards the ♀'s head by slowly stepping to one side with his forelimbs (Fig. 18, arrow 1). The ♂ maintains this posture with his body axis in a

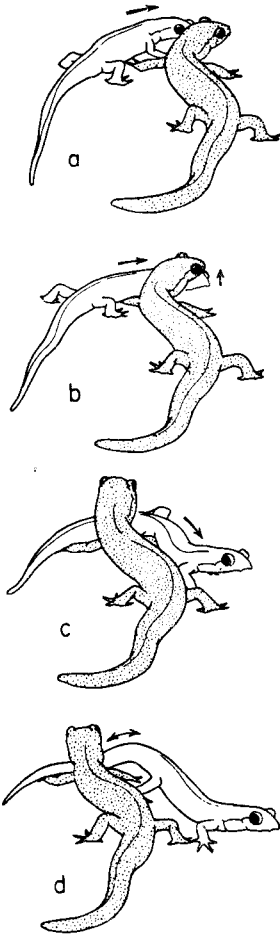


Fig. 15: An attempt to initiate a tail-straddling walk in *Plethodon jordani*. a) The ♂ touches his snout to the ♀'s cheek. The ♀ (stippled) has turned her head slightly away from the ♂. b) The ♂ lifts his head in contact with the ♀'s chin. c) The ♂ continues forward under the ♀'s chin. d) The ♂ undulates his arched tail in contact with the ♀'s chin. This attempt to initiate a tail-straddling walk was unsuccessful. From a series of photographs of a courting pair

U-shape and with his head held high (Fig. 18). He accentuates the arch of his tail, and the undulations of his tail increase in amplitude. While maintaining this U-shaped posture, the ♂ may pivot forward about a point at the center of the U by stepping forward with both forelimbs and hindlimbs. After maintaining this U-shaped posture, the ♂ gradually turns his head even further towards the ♀ so that his head is high and his mentum lies directly above the ♀'s head. The ♂'s body axis now forms a complete circle rather than a U. The ♂ pauses momentarily in this posture and then suddenly and rapidly he drops his head and slaps his mental gland on the dorsum of the ♀'s head or snout. The slapping motion is very rapid, and the ♂ pulls his head back as it moves downward (Fig. 18, arrow 2). The ♂ again pauses momentarily, with his head level, and then turns slightly away from the ♀, so that his body axis again forms a U. He pauses and then turns fully away from the ♀, straightening his body (Fig. 18, arrow 3).

Vent-sliding. The ♂ moves forward and slides his vent in contact with the substrate. The tail is level and may undulate slightly. The venter of the body is in contact with the substrate.

Begin spermatophore deposition. The ♂ pauses with his vent and ventral body in contact with the substrate. The ♂'s tail undulates slightly. Usually the tail is level, but the ♂ may arch the proximal part of his tail.

Spermatophore deposition. The ♂ continues in the posture just described. Undulations of relatively large amplitude proceed along the entire length of the tail (Fig. 19a) and increase in frequency and amplitude during the course of spermatophore deposition. Finally the ♂ stops undulating his tail and lifts his vent free of the spermatophore (Fig. 19b).

Moving forward with tail flexed. The ♂ moves forward with the proximal part of his tail flexed 45–90 degrees to one side. The ♂ holds his body in a sigmoid lateral flexure and keeps his vent above the substrate (Fig. 19c).

Stationary with tail flexed. The ♂ executes the following series of actions as the ♀ stops with her vent near or over the spermatophore. The ♂ stops and extends upward and backward on his hindlimbs and consequently straightens the sigmoid flexure of his body. The ♂ then straightens and flexes his hindlimbs rhythmically, that is he does push-ups with his hindlimbs (Fig. 19e). Simultaneously his forelimbs are inclined backward, but he does not do push-ups with his forelimbs. The ♂'s tail remains flexed to one side. Finally the ♂ stops flexing and extending his hindlimbs and remains motionless with his hindlimbs extended.

2. Temporal Relations

a. The Behavior of the ♂

When the ♂ first contacts a ♀ he moves the length of her body and performs tapping, nudging, or head-sliding. The ♂ may move antieriad or posteriad. He may perform a foot dance with any of these modes of head con-

tact. Usually the δ moves along beside the ♀ , but less frequently he moves along on top of the ♀ 's body and slides his mentum in contact with her dorsum.

The δ apparently identifies the sex and species of his sexual partner by chemoreception during these tapping, nudging and head-sliding activities. In addition the δ probably uses chemoreception to tell if the ♀ is gravid. Only ♀ with large ova elicited persistent courtship from the $\delta\delta$.

Once the δ has contacted a conspecific ♀ , he locates her head. This is accomplished by trial and error if the ♀ remains stationary. Apparently the δ has no means of distinguishing anterior from posterior as he moves along the length of the ♀ . If he moves posteriorly he proceeds to the tip of the ♀ 's tail, turns around and moves anterior to her head. The δ readily locates the ♀ 's head as she is moving forward. Whenever she moves forward, he moves towards her rapidly. When she stops in close proximity, the δ approaches very slowly with his body and head held low to the ground (STEBBINS 1949 provides a figure of this same cautious approach in the related salamander *Ensatina*). In some instances it appeared that the δ was visually orienting to the ♀ 's gular fluttering.

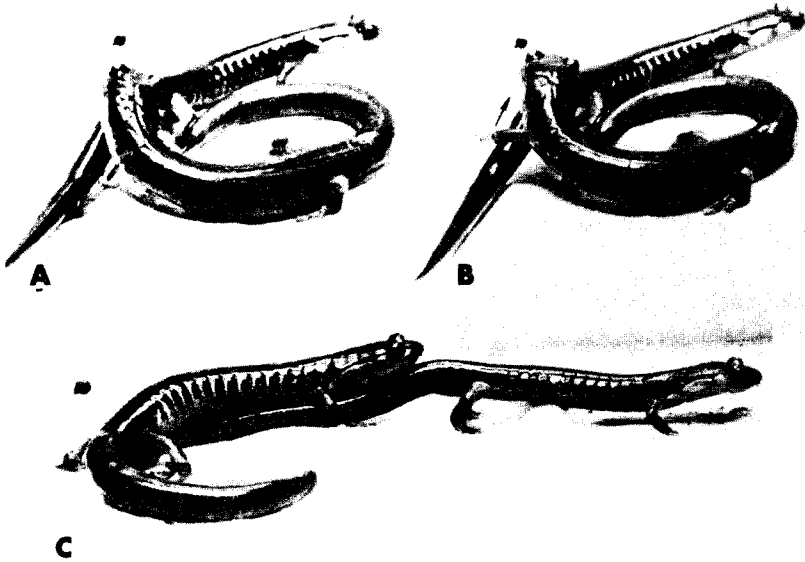


Fig. 16: Initiation of a tail-straddling walk in *Plethodon jordani*. a) The ♀ has turned her head from perpendicular to axial on the δ 's arched tail as it undulates under her chin. b) The ♀ steps astride the δ 's undulating tail. c) The ♀ (left) and the δ in a tail-straddling walk. The δ is arching and undulating his tail under the ♀ 's chin. The δ 's mental gland can be seen as a low projection from his chin just below his eye. These animals are from Standing Indian Recreation Area, Macon Co., North Carolina

When the δ reaches the ♀ 's head he attempts to initiate the tail-straddling walk (Fig. 16). First he nudges or slides his mental gland along her cheek or snout while tilting his head slightly to one side. Next he turns his head under the ♀ 's chin and lifts. He may rub the ♀ 's chin rhythmically by sliding his snout or the dorsum of his head laterally or forward and backward. Finally the δ crawls forward under the ♀ 's chin. Once the δ has turned away from the ♀ , any light contact to the dorsum of his tail (e. g. by the venter of the ♀ ,

inanimate objects or another ♂) causes him to arch and undulate his tail. Usually, however, it is the ♀'s chin that elicits tail undulation. When the ♀'s chin contacts the dorsum of the ♂'s body, the ♂ frequently turns away from the ♀, so that he follows a sigmoid path with the inflexion point beneath the ♀'s chin. In this way the ♂ aligns his body with that of the ♀ and pauses with her chin in contact with the proximal part of his tail.

b. The Behavior of the ♀

When the ♀ is refractory, courtship seldom proceeds beyond this point. A refractory ♀ will repeatedly flee from the ♂ as he contacts her and moves toward her chin. If the ♂ succeeds in crawling under her chin, the ♀ will either move away from his undulating tail or will remain motionless. In the latter case the ♂ stops undulating his tail after a few min, turns around and approaches her anew. In a small container a ♂ will follow a refractory ♀ and attempt to initiate a tail-straddling walk for several h without success. In larger containers a refractory ♀ will eventually escape from the ♂. This is undoubtedly the outcome in the field.

A responsive ♀ uses the undulating contact of the ♂'s tail to her chin as a directional cue and will initiate the tail-straddling walk. She turns toward the ♂ and steps astride his tail (Fig. 16). An exceptionally responsive ♀ will actually approach the ♂ even before he has crawled under her chin, step astride his tail and slide forward along it. In such cases the ♂ immediately moves forward and begins the tail-straddling walk.

c. The Tail-straddling Walk

The tail-straddling walk may last for a few min to more than an h (Fig. 30). During this time the ♂ apparently relies solely upon tactile cues to

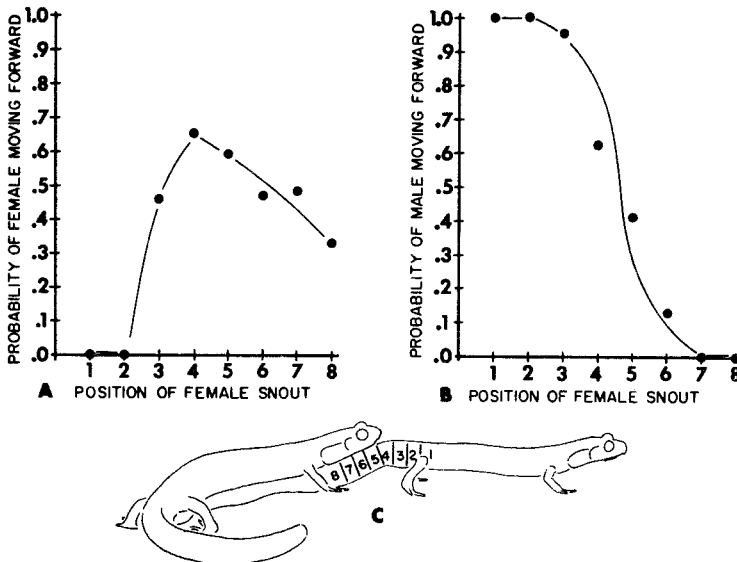


Fig. 17: Forward progress as a function of the position of the ♀'s snout during the tail-straddling walk of *Plethodon jordani*. A) The probability that the ♀ is moving forward as a function of the position of her snout on the ♂'s tail. B) The probability that the ♂ is moving forward as a function of the position of the ♀'s snout on his tail. C) A ♀ (left) and a ♂ in a tail-straddling walk, showing the 8 regions used to describe the position of the ♀'s snout

monitor and regulate the ♀'s position. This interpretation is supported by an experimental manipulation of a single ♂ and by accumulated records of the positions of the sexes and their progress during the tail-straddling walk. The regulatory system is very similar to that of *Ambystoma tigrinum*, but the *Plethodon* system appears to have much finer tuning.

On one occasion I was able to manipulate the behavior of the ♂ by simulating the presence of a ♀ during the tail-straddling walk. As a ♂ was approaching a ♀, and contacting her with his snout, I reached into the container and placed the stainless steel handle of a scalpel in contact with the proximal portion of the ♂'s tail. I held the handle perpendicular to his tail so that the length of contact was only about 1 cm and touched my fingers to the ground on either side of the ♂'s tail, so that the contact to his tail was very light. For the next 20 min I was able to govern the ♂'s forward progress by changing the position of the handle on his tail. The ♂ moved forward whenever the forward edge of the handle was moved anterior to the ♂'s vent. He moved forward very rapidly whenever the handle was moved anterior to his sacrum. Each time I held the handle stationary as the ♂ moved forward he would continue forward and then stop when the contact slipped posterior along his tail. After approximately 20 min of "tail-straddling walk" the ♂ stopped and deposited a spermatophore. During a tail-straddling walk with a real ♀ it is apparently only the anterior contact of the ♀'s chin on the ♂'s tail that affects the ♂'s progress.

The results of compiled records of the positions of the sexes and their progress during the tail-straddling walk are shown in Fig. 17 and Table 3. As in the experiment just described, these data indicate that the ♂ moves for-

Table 3: The effects of the ♀'s position upon forward progress by the ♂ and ♀ during the tail-straddling walk of *Plethodon jordani*

a. Hypothesis: The male tendency to move forward is independent of the female snout position.

♀ snout position	1	2	3	4	5	6	7	8	Total
Total No. of observations of male progress.	6	11	22	56	63	72	16	13	259
No. of observed cases in which ♂ was moving forward.	6	11	21	35	26	9	0	0	108
						9			
Expected No. of cases	2.5	4.6	9.2	23.3	26.2	42.0			
Expected probability of forward progress = 108 / 259 = 0.416									
Chi Square = 60.7, d. f. = 5, p < 0.001									

b. Hypothesis: The female tendency to move forward is independent of the female snout position.

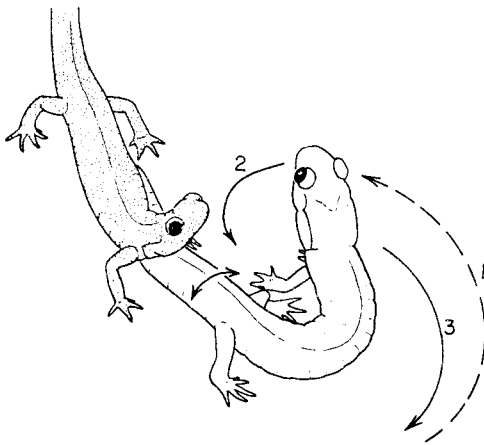
♀ snout position	1	2	3	4	5	6	7	8	Total
Total No. of observations of female progress.	8	10	22	63	81	107	25	18	334
No. of observed cases in which ♀ was moving forward.	0	0	10	41	48	50	12	6	167
		10							
Expected No. of cases.		20.0		31.5	40.5	53.5	12.5	9.0	
Expected probability of forward progress = 167 / 334 = 0.500									
Chi Square = 10.50, d. f. = 5, p > 0.05									

ward when the ♀'s chin is in contact with the proximal part of his tail, but stops and accentuates the arch of his tail when the ♀ slips even slightly posteriad. He moves forward again when the ♀ slides up to the proximal part of his tail. Forward progress by the ♀ is independent of her position on the ♂'s tail (Table 3). Nevertheless, the ♀ remains very close to the ♂'s vent throughout the tail-straddling walk as a consequence of the ♂'s regulatory behavior.

Precision in site of spermatophore deposition is the main outcome of the regulatory process just described. When the ♂ deposits a spermatophore at the end of the tail-straddling walk, it lies directly in front of the ♀'s snout (Fig. 31).

d. The Function of the Mental Gland

In some courtships the ♂ remains on a straight course during the tail-straddling walk. In other courtships the ♂ repeatedly alternates between progress on a straight course and turning-back towards the ♀. Male *Plethodon jordani* possess a large disc-shaped gland on the chin, the mental gland, which is slapped across the ♀'s nares when the ♂ turns back towards the ♀ (Fig. 18). It seems likely that secretions from the mental gland are taken in through the ♀'s nares.



An explanation for the function of mental gland slapping in *Plethodon* must account for the following additional facts. 1) The ♀'s response after slapping is usually to remain in place astride the ♂'s tail and then to continue in the tail-straddling walk. Sometimes the ♀ may raise her head after she has been slapped, and sometimes she may depart from the ♂ and thereby terminate the

Fig. 18: Slapping of the mental gland on the ♀'s snout during the tail-straddling walk in *Plethodon jordani*. The secretions of the mental gland may be an aphrodisiac. From a photograph

courtship. 2) The same ♀ may receive different treatment on different occasions. For example, on August 11, 1968, a ♂ repeatedly slapped a ♀ during the tail-straddling walk, and the subsequent sperm transfer was successful. On August 14th, the same ♀ was courted by another ♂. This ♂ did not slap the ♀ and the sperm transfer was unsuccessful. On August 10, 1969, a ♂ did not perform slapping and the ♀ subsequently removed the sperm cap from the spermatophore. On August 16th this ♀ was again successfully inseminated by the same ♂, but this time he slapped her 4 times during the tail-straddling walk. 3) Thus there appears to be no simple polymorphism for slapping among ♂♂, since the same ♂ may or may not slap the ♀. 4) Tail-straddling walks during which the ♂ slaps the ♀ are of longer duration than tail-straddling walks in which mental gland slapping is absent; when slapping occurs, it occurs repeatedly (Fig. 30). This suggests two distinct classes of tail-straddling walks (those with and those without slapping) rather than a ran-

dom performance of slapping with an unchanging probability of occurrence.

The following functions of mental gland slapping are conceivable. 1) The act enables the ♂ to identify the sex or species of his partner. 2) The act serves to orient the ♀ during the tail-straddling walk. 3) The act tests the sexual receptivity of the ♀. 4) The secretions of the mental gland increase the sexual receptivity of the ♀ such that she becomes more likely to retrieve sperm from the spermatophore.

Of these four possibilities, the first two are unlikely for several reasons. Since the ♂ does not perform slapping during sexual behavior with other ♂♂, it is unlikely that he uses this act for sex-identification. In addition, sex recognition is known to occur at the initiation of courtship and thus long before the tail-straddling walk takes place (ORGAN 1958, 1960a, 1960b). Species recognition by the ♂ also takes place at the onset of courtship, and heterospecific courtship never progress to the stage of the tail-straddling walk. There is no indication that slapping helps the ♀ orient during the tail-straddling walk, since the ♀ does not turn in the direction of the slap, and the ♂ does not slap before changing direction.

Testing and enhancing of female receptivity seem to be more likely explanations for the function of mental gland slapping. These explanations account for all the peculiarities in the occurrence of the act. When the courtship season is long, as in *Plethodon jordani*, ♀♀ are likely to have differing sexual receptivities depending on whether or not they have been inseminated. Long lasting tail-straddling walks with mental gland slapping and brief tail-straddling walks without mental gland slapping may represent adaptive male responses to nonreceptive and receptive ♀♀ respectively. Perhaps the mental gland secretions have an aphrodisiac effect upon the ♀ and increase the probability that she will later retrieve sperm from the spermatophore. Thus the secretions could have a *priming* effect (WILSON and BOSSERT 1963) upon the ♀'s subsequent behavior toward the spermatophore, rather than an immediate releasing effect. The repeated application of the mental gland secretions is compatible with such a priming action, and at the same time might serve to test the ♀'s current state of receptivity. Sometimes ♀♀ depart after being slapped with the mental gland. Such departures may indicate nonreceptivity. SALTHER and SALTHER (1964) have proposed a similar testing function for an act in *Pseudoeurycea belli* that is apparently homologous with mental gland slapping (ARNOLD 1972). I was unable to detect any behavior in the ♀ (e. g. manifestations of sexual reluctance) that elicited slapping by the ♂, or any change in the behavior of the ♀ after she had been slapped repeatedly.

More extensive observation and well designed experiments are needed before the aphrodisiac function of the mental gland can be considered established. In particular we need to know what determines whether slapping will occur and the physiological and behavioral effects of the mental gland secretions upon the ♀. Delivery of the mental gland secretions to the ♀'s nares is apparently a recent evolutionary innovation in the large eastern *Plethodon*. The recent ancestors of these salamanders, as well as all but the most primitive plethodontids, deliver the mental gland secretions directly into the superficial circulation of the ♀'s back by scratching with the elongate premaxillary teeth (ORGAN 1961; ARNOLD 1972).

The principal use of the mental gland is employment during the tail-straddling walk, but the mental gland is used in other contexts as well. When moving towards the ♀'s head along her dorsum, the ♂ may press his mental gland down over the ♀'s snout (ORGAN 1958, Fig. 1). Usually, however, he

reaches her head by moving along beside her and does not touch her head with his mental gland. ORGAN (1958, 1960a) also noted that during the approach phase the ♂ may lift his head slowly from the ♀'s dorsum. As the adhesion breaks between his mental gland and her dorsum, the ♂'s head may snap upward. I observed this only rarely. It is apparently a fortuitous consequence of a rare and momentary adhesion of the mental gland, and not, as ORGAN suggests, a behavior designed to stimulate the ♀. On three occasions I observed ♂♂ slapping the mental gland on the ♀ during the approach phase; the similarities with slapping in the more usual context (Fig. 18) were unmistakable. I was at first puzzled by slapping in this unusual context. Later I discovered that in more primitive plethodontids the mental gland is forcefully applied to the ♀ during the approach phase as well as during the tail-straddling walk (ORGAN 1961; ARNOLD 1972). The rare and unpredictable occurrence of slapping during the approach phase in *Plethodon jordani* suggests a holdover from an ancestral courting mode.

e. Spermatophore Deposition and Sperm Transfer

If the ♀ departs from the ♂ at any time during the tail-straddling walk, the ♂ lowers his tail, ceases tail undulations and approaches the ♀. If, however, the ♀ continues to slide forward intermittently along the ♂'s tail, the ♂ lowers his vent and slides it in contact with the substrate. Vent-sliding apparently represents the actual search for a site on which to deposit the spermatophore and lasts for a min at most. The ♂ may immediately pause and deposit a spermatophore or he may vacillate between pressing his vent to the substrate and holding his vent above the substrate before finally depositing the spermatophore.

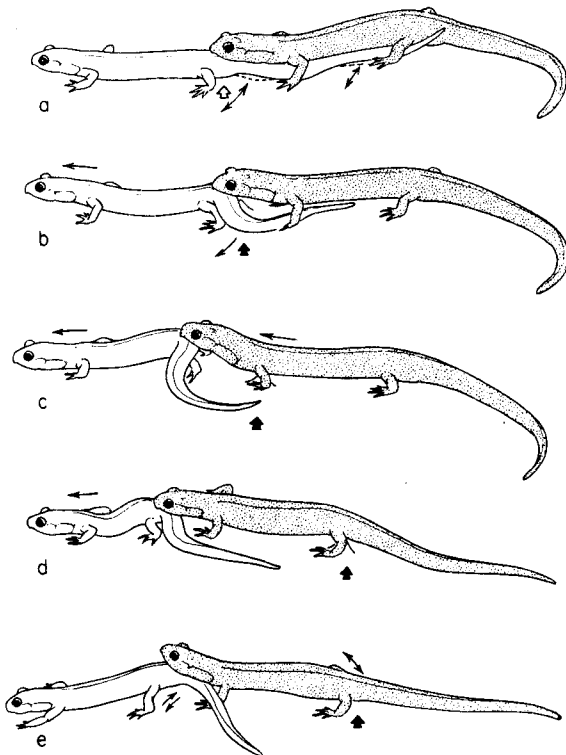


Fig. 19: Sperm transfer in *Plethodon jordani*. a) The ♂ deposits a spermatophore as the ♀ (stippled) straddles his tail. b) The ♂ begins to move away from the spermatophore and flexes his tail to one side and out from under the ♀. c) The ♂ continues forward. The ♀ is now moving forward with the spermatophore in contact with her chest. d) The ♀ has stopped with the spermatophore in contact with her vent. The ♂ continues forward with his body held in sigmoid flexure. e) The ♀ has lowered her vent upon the spermatophore and inserted it into her cloaca. The ♂ extends and flexes his hindlimbs and thereby forces his tail base under the ♀'s chin. From a series of photographs of a courting pair

Should the ♀ depart soon after spermatophore deposition begins, the ♂ approaches her without depositing a spermatophore. If, however, the ♀ departs a few min after spermatophore deposition begins, the ♂ continues just as if the ♀ were present. In such cases, however, the ♂ does not flex his tail to one side when he moves away from the spermatophore.

Usually the ♀ remains astride the ♂'s tail as he deposits the spermatophore. She remains essentially motionless but may slide anteriorly or posteriorly a few mm along his tail. Usually the ♀ does not move her head, but she occasionally appears to swing it rhythmically in short arcs over the ♂'s

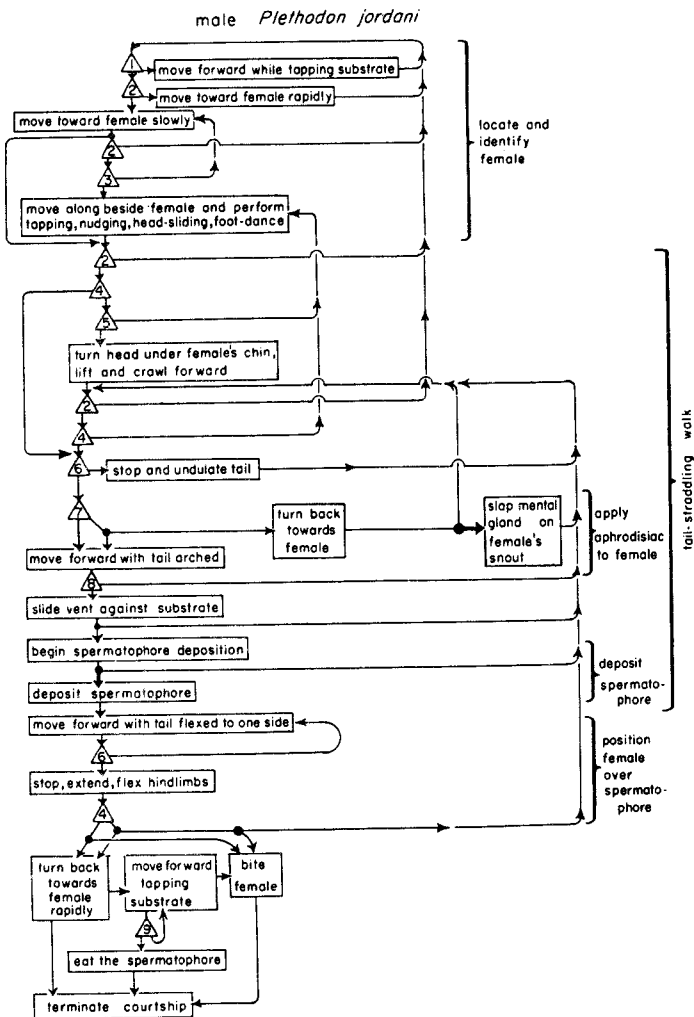


Fig. 20: An algorithm for courtship in male *Plethodon jordani*. For all questions, "yes" exits on the left and "no" exits on the right. 1) Is the ♀ present in the visual field? 2) Is the ♀ very near? 3) Does the ♂'s snout contact the ♀? 4) Does the ♀ contact the dorsum of the ♂'s body or tail? 5) Is the ♂'s head opposite the ♀'s chin? 6) Is the contact anterior to region 4 on the ♂'s tail? 7) Does the ♀ require an aphrodisiac? 8) Will the ♀ retrieve sperm if a spermatophore is deposited? 9) Does the ♂ contact the spermatophore with his nasobial cirri?

undulating tail. As ORGAN 1961 suggested, the pronounced tail undulations during spermatophore deposition probably lubricate the ♀'s venter and thus prevent premature dislodging of the sperm cap when the ♀ slides over the spermatophore. The ♂'s tail undulates in contact with the entire length of the ♀'s abdomen, and frequently a film of mucus can be seen between the ♂'s undulating tail and the ♀'s body.

Following spermatophore deposition, the ♂ withdraws his tail from under the ♀'s body (thereby exposing the spermatophore to the ♀'s venter) and moves forward with his tail flexed to one side. The ♀ follows behind him with her chin resting on the dorsum of his tail base. As the ♀ moves forward the spermatophore contacts her venter and she slides lightly over the top of it. If the spermatophore contacts her vent, the ♀ lowers her vent upon it and inserts the entire spermatophore into her cloaca. She undulates her tail base

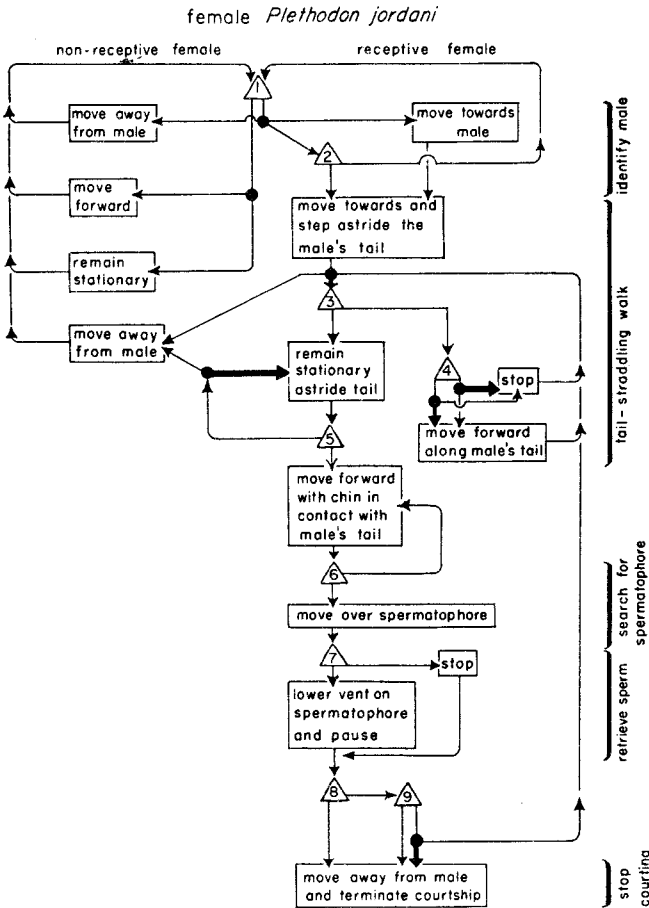


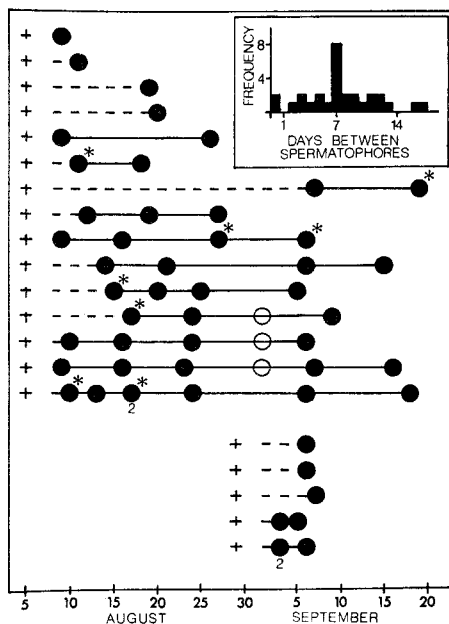
Fig. 21: An algorithm for courtship in female *Plethodon jordani*. For all questions, "yes" exits on the left and "no" exits on the right unless indicated otherwise. 1) Does the ♂ contact the ♀? ("yes" exits on the right). 2) Is the ♂'s tail undulating in contact with the ♀'s body? 3) Is the ♂ depositing a spermatophore? 4) Is the ♀'s snout in region 4 on the ♂'s tail? 5) Is the ♂'s tail flexed to one side? ("yes" exits on the right). 6) Does the spermatophore contact the ♀'s venter? 7) Does the spermatophore contact the ♀'s vent? 8) Does the ♂ bite the ♀? 9) Is the ♀ inseminated?

slightly while resting on the spermatophore but is otherwise quite motionless. The ♂ meanwhile stops in front of the ♀ and executes the series of actions described above in the catalog of male actions. The extensions and flexions of his hindlimbs move the ♀'s head up and down rhythmically. This male activity varies little from courtship to courtship. It probably serves to position and maintain the ♀ over the spermatophore.

The actions that occur next are remarkably variable from courtship to courtship (Figs. 20, 21). The ♀ may remain motionless over the spermatophore or she may suddenly depart. The ♂ may remain motionless, or he may suddenly swing around and back towards the ♀. After swinging around, the ♂ may remain motionless, he may briefly tap or nudge the ♀, or he may lunge and bite the ♀. ♀♀ flee immediately when bitten by the ♂. After the ♀ has departed the ♂ may tap the substrate and circle about the site of spermatophore deposition. If he contacts the spermatophore with his nasolabial cirri, the ♂ usually grasps the spermatophore repeatedly with his mouth and eats part or all of it. The sudden increase in the variability of behavioral sequences after the ♀ has made her attempt to retrieve sperm is probably the result of relaxed selective pressures. Since the ♂ usually deposits only one spermatophore in an evening, once the ♀ has attempted to retrieve sperm from this spermatophore, natural selection will have little effect upon the ♂'s subsequent behavior. This argument is an analog to FISHER'S (1958) insight that the direct action of natural selection will be proportional to reproductive value.

Once the ♀ attempts to retrieve sperm she usually flees and repeatedly moves away from the ♂ whether or not she has successfully removed the sperm cap. If the ♂ does resume contact with the ♀ after spermatophore deposition, he usually moves away after a very short time. Usually within 5 min after spermatophore deposition, the ♂ no longer responds to movement by the ♀. ♂♂ deposit a mean of 1.04 spermatophores per courtship (49 observations of 1 spermatophore and 2 observations of 2 spermatophores; only

Fig. 22: Long term courtship records for male *Plethodon jordani*. Each horizontal line represents the courtship record for an individual ♂ housed with a single ♀. Crosses (+) indicate the date each pair was collected. The onset of the dashed lines indicate the date on which the pair was established in the laboratory and observations begun. Solid circles indicate the dates on which spermatophores were deposited. A subscript 2 indicates a ♂ deposited two spermatophores in an evening. Observations were interrupted on 29–30 August 1969 for field work. Three spermatophores were found on 31 August (open circles). Since these spermatophores might have been deposited on 29–31 August the intervals on either side of these spermatophores are not included in the histogram. Stars indicate sperm transfers which were known to be successful. Other spermatophores were either unsuccessful or transfer was not observed. No spermatophores were deposited from 20 September to 10 November 1969, when the animals were preserved



one of the courtship with two spermatophores was actually observed, Fig. 30). The mean time between spermatophore depositions for a single ♂ housed continually with a single ♀ is 7.59 days (standard deviation = 4.15, n = 29; Fig. 22).

Female *Plethodon jordani* are more likely to find a particular spermatophore than female *A. maculatum*, and *A. tigrinum* ♀♀ have intermediate success (Table 4). The probability that a *Plethodon jordani* ♀ will retrieve

Table 4: Species differences in the probability of finding a spermatophore

	<i>Ambystoma maculatum</i>	<i>Ambystoma tigrinum</i>	<i>Plethodon jordani</i>
Number of spermatophores deposited	131	28	35
Number of spermatophores found by ♀	51	17	33
Probability of female finding a particular spermatophore	0.39	0.61	0.94
Statistical tests:			
<i>A. tigrinum</i> versus <i>P. jordani</i>		Chi Square = 10.71 **	
<i>A. maculatum</i> versus <i>A. tigrinum</i>		Chi Square = 4.47 *	
<i>A. maculatum</i> versus <i>P. jordani</i>		Chi Square = 33.86 **	
Total number of successful transfers	Unknown	Unknown	19

sperm from a spermatophore is 0.54 (Table 4). If the ♀ is exposed to 5 spermatophores she will almost certainly be inseminated at least once (Fig. 8). ♀♀ are probably inseminated more than once during a courtship season. In captivity ♀♀ will continue to court, and be inseminated, after the first insemination. When more than one ♂ was housed with a ♀, she was sometimes inseminated by more than one ♂, but always on different nights. ORGAN and LOWENTHAL (1963) found that each sperm cap of *Plethodon jordani* is covered with thick capsules. Consequently, once the ♀ is inseminated, a few days are probably required to break down these capsules and during this time the ♀ cannot accommodate additional sperm caps in her cloaca.

On a very few occasions I observed ♀♀ performing each of the following "male" actions: foot dance, arching and undulating the tail in contact with the ♂'s chin, and turning-back towards the ♂. In the latter case the ♀ never completed the action by slapping her chin at the ♂'s head. The rare occurrence of these behaviors in ♀♀ suggests that at least some of the loci that govern them are not sex-linked.

V. Dispersion of Courting Animals, Length of the Courtship Season, Sexual Interference and Sexual Defense

A. *Ambystoma maculatum*

1. The Courtship Setting

In *Ambystoma maculatum* several factors act in concert to produce extreme sexual competition among ♂♂. These factors promote sexual competition because each ♂ is capable of inseminating more than one ♀, while each ♀ can be effectively inseminated by just one ♂ (this is probably true of

all three species). Sexual ♂♂ are crowded in both time and space, since the courtship season is short and courtship occurs in ponds that are much smaller than the area occupied outside of the courtship season. This temporal and spatial crowding of ♂♂ increases the opportunity for sexual interference since it allows a ♂ to interfere with several other ♂♂ almost simultaneously (e. g. by covering spermatophores in an aggregation of competitors). Sexual competition is also increased by the clumping of ♀♀ in space, since this increases the opportunity of individual ♂♂ to inseminate several ♀♀. (The chances of sexual monopoly are, however, lessened by the clumping of ♀♀ in time.) Finally competition among ♂♂ is enhanced by the tendency of ♂♂ to outnumber breeding ♀♀ in at least some populations (HUSTING 1965).

In some ponds courtship tends to occur in specific areas year after year. There is *no* indication, however, that ♂♂ maintain permanent stations within these areas. In fact, the ♂♂ constantly mill about. The areas are invariably in shallow water at the pond margin and may simply represent major points of arrival during migration. At Goss Pond, for example, most animals arrive at the pond along a shallow gully that drains runoff. Most of the courting animals are found in the pond just beyond the mouth of this migration funnel. Due to the aggregation of ♂♂ at this point, ♀♀ are engaged in courtship very soon after they reach the pond. A few even court in small pools on the approach to the pond.

As a consequence of these factors several ♂♂ are usually found courting a ♀ simultaneously. Occasionally a single ♂ courts a ♀, but the density of ♂♂ is so high that additional ♂♂ quickly locate and join a courting pair. Frequently, aggregations of 10 to 50 ♂♂ can be found rapidly milling around one or a very few ♀♀. The ♂♂ closest to the ♀ actively court her and deposit spermatophores. BLANCHARD (1930) and BREDER (1927) report similar observations. Studies on animals in the laboratory suggest that ♀♀ are inseminated within these aggregations in less than an h and then depart and lay their eggs within 24—48 h. In contrast the ♂♂ probably continue to court night after night until their supply of spermatophores is exhausted (see Fig. 7). The normal setting for courtship in *Ambystoma maculatum* is a polyandrous frenzy.

2. Sexual Interference

When several ♂♂ court a ♀ simultaneously, they rub and nudge each other as well as the ♀, but the ♂♂ never attempt to bite one another or to transport the ♀ away. Each of the courting ♂♂ may deposit spermatophores, either on the substrate or on other spermatophores (Figs. 23, 25).

The ♂♂ do *not* exert a synergistic effect upon the ♀ such that each ♂ profits from the participation of other ♂♂. In the presence of multiple ♂♂ the ♀ finds spermatophores faster than with a single ♂ simply because more spermatophores are present, but the ♀ stops responding to spermatophores and swims away once she has found about 20 spermatophores regardless of how many ♂♂ are participating (Fig. 24, Table 5). The ♀ probably terminates courtship once her spermatheca is filled. Each ♂ experiences a lowered reproductive success due to the presence of other ♂♂.

The success of each ♂ is lowered by a variety of effects. First, since each ♂ deposits as many spermatophores as if he were alone with the ♀, each ♂'s spermatophores are diluted by the spermatophores of others. Second, some of his spermatophores will be covered by the spermatophores of other ♂♂. Finally, the nudgings of competitors can disrupt a ♂ in the act of depositing a spermatophore, dislodge the ♀ from his spermatophore, or in general, inter-

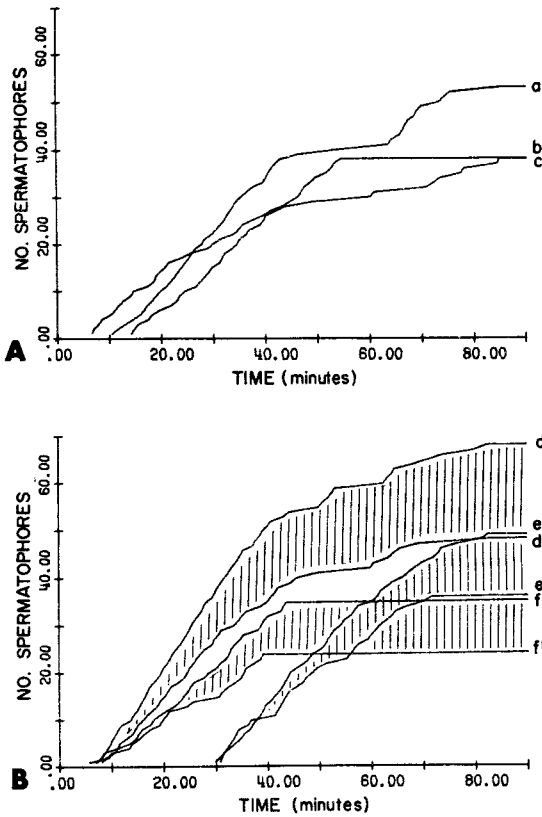


Fig. 23: Spermatophore deposition rates for single (above) and rival (below) ♂♂ of *Ambystoma maculatum*. The cumulative number of spermatophores is shown as a function of time. The animals were introduced at time 0. Vertical lines in the lower figure connect the ♂♂ which courted the same ♀.

These effects tend to reduce the probability that a rival ♂ will inseminate the ♀. Thus these effects constitute examples of sexual interference.

Despite the nearly constant nudging and pushing interactions between competitors, individual ♂♂ are able to slightly increase their rates of spermatophore deposition under these conditions (Fig. 23, Table 5). It is unclear, however, whether the presence of rivals increases the tendency to deposit spermatophores in each ♂ or whether the larger number of spermatophores re-

quiring covering causes the increased deposition rate. Either mechanism would tend to recoup losses in fitness due to rivals.

The principle defense against sexual interference in *Ambystoma maculatum* seems to be rapid courtship.

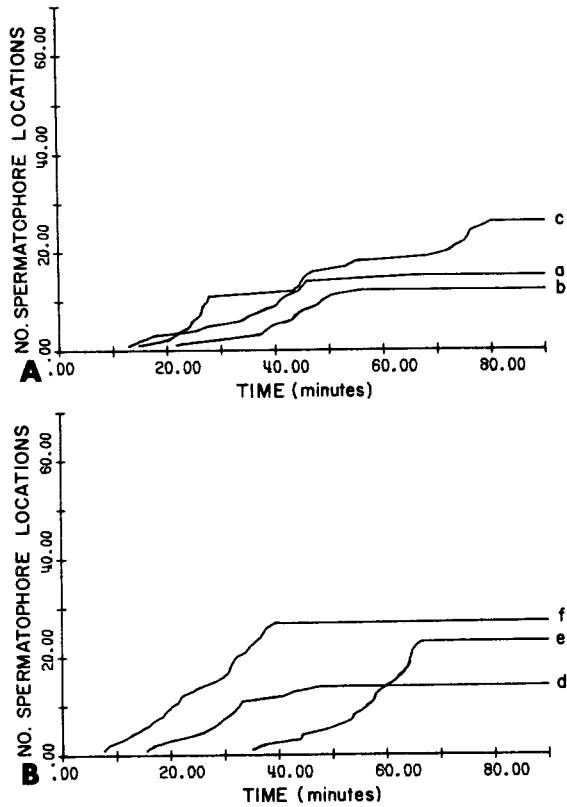
3. The Significance of Multiple Spermatophores

In *Ambystoma maculatum* a third of the spermatophores are deposited on top of other spermatophores (Fig. 27, Table 6). Two lines of evidence suggest that the behavior of depositing spermatophores on top of others is pre-eminently a kind of sexual interference and not simply a fortuitous by-product of courtship. The first argument is theoretical. It is easy to imagine that it is advantageous for a ♂ to deposit spermatophores on other spermatophores rather than on the substrate if the spermatophores of competitors are more abundant than his own. In fact the advantage is more universal. Suppose that ♂ A has n functional spermatophores while his competitor(s) has m spermatophores. The probability that the ♀ will find one of A's spermatophores is:

$$p = n / (m + n) \tag{1}$$

If the ♂ deposits the next spermatophore on the substrate this probability increases (Table 7). On the other hand, the ♂ may place his spermatophore on top of another. His chance of reproductive success remains unchanged if he covers one of his own spermatophores but increases if he eliminates a compe-

Fig. 24: Rates of spermatophore discovery by female *Ambystoma maculatum* courting with one (above) and with two (below) ♂♂. The cumulative number of spermatophores located by the ♀ is shown as a function of time. These ♀♀ were courted by the ♂♂ in Fig. 23 and letters correspond to the names of ♂♂ in Fig. 23



titor's spermatophore while depositing one of his own (Table 7). The ♂'s expected chance of success, p_s , is the sum of the probabilities of these two events:

$$p_s = (p)p_c + (1-p)p_d \quad (2)$$

Substituting the appropriate functions in Table 7 into equation 2, we find that:

$$p_s > p_b$$

for all n and m greater than zero. In other words if a ♂ encounters a spermatophore of unknown identity, it is better to cover the sperma-

Table 5: A comparison of courtship in *Ambystoma maculatum* when single and when rival ♂♂ court a ♀

Variable	Sample Size =	One ♂ (3 pairs)	Two ♂♂ (3 trios)	Statistical Test
Total number of spermatophores		129	260	
Total number of spermatophores found by ♀♀		53	64	Chi Square = 11.12, d. f. = 1, $p < 0.01^{**}$
Number of spermatophores deposited per ♂	Mean = Standard Deviation =	43.00 8.66	43.33 15.23	$U = 7.0, p > 0.05$ ns
Number of spermatophores deposited per ♀	Mean =	43.00	86.67	
Number of spermatophores found by ♀	Mean = Standard Deviation =	17.67 7.37	21.33 6.66	$U = 3.0, p > 0.05$ ns
Time between spermatophore depositions	Mean = Variance = n =	1.53 min 0.106 126	1.24 min 0.087 254	$T_{(378)} = 2.05, p = 0.02$ (one tailed test)* $F_{(125, 253)} = 1.22, p = 0.095$ ns
Time between findings of spermatophores by ♀ ^b	Mean = Variance = n =	3.05 min 0.221 50	1.56 min 0.136 61	$T_{(193.5)} = 2.27, p = 0.013$ (one tailed test)* $F_{(49, 60)} = 1.62, p = 0.019$ (one tailed test)*

^a A test of the hypothesis that the ♀ finds the same proportion of spermatophores when courting with one or two ♀♀. The hypothesis is rejected.

^b Statistical tests were applied to the logarithm to the base 10 of this variable. The variance indicated is for the transformed distribution.

Table 6: Occurrence of multiple spermatophores in *Ambystoma maculatum* and *Ambystoma tigrinum*^a

Number of spermatophores deposited per structure	<i>Ambystoma maculatum</i> (26 courtships by single ♂♂)			<i>Ambystoma tigrinum</i> (26 courtships by single ♂♂)		
	Frequency predicted by truncated Poisson distribution	Observed frequency	Deviation from expectation	Frequency predicted by truncated Poisson distribution ^b	Observed frequency	Deviation from expectation
1	431.8	470	+	240.1	248	+
2	198.9	145	-	121.7	107	-
3	61.1	65	+	41.2	44	+
4	14.1	19	+	10.4	17	+
5	2.6	6	+	2.1	0	-
6	0.4	4	+	0.4	0	-
7	0.2	0	-	0.2	0	-
Total	709.1	709		416.1	416	
Mean		1.53			1.59	
Variance		0.81			0.70	
Goodness of fit:						
Chi Square		26.15			3.39	
Degrees of freedom		2			2	
		p < 0.005			0.5 > p < 0.1	

^a From courtships in screen cylinders. All animals from the E.S.G.R.

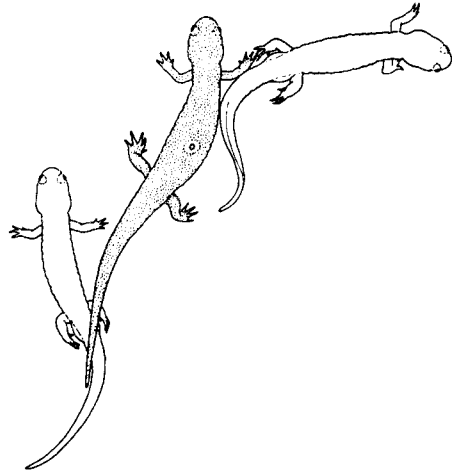
^b Fitted using the method of COHEN 1960.

Table 7: Sexual success as a function of the ♂'s behaviour towards the spermatophores he encounters

	Absolute fitness of ♂ A	Absolute fitness of rival (s)	Relative fitness of ♂ A		Type of spermatophore deposition or other action by ♂ A.
	Number of spermatophores of ♂ A.	Number of functional spermatophores of rival ♂♂.	Probability that the ♀ will find one of A's spermatophores.		
n^{th} spermatophore deposition	n	m	$n / (n + m)$	p	
$n + 1^{\text{th}}$ spermatophore deposition or behavior	n + 1	m	$(n + 1) / (n + m + 1)$	p_b	Deposit spermatophore on substrate.
	n	m	$n / (n + m)$	p_c	Deposit spermatophore on own spermatophore.
	n + 1	m - 1	$(n + 1) / (n + m)$	p_d	Deposit spermatophore on rival's spermatophore.
	n - 1	m	$(n - 1) / (n + m - 1)$	p_e	Destroy own spermatophore.
	n	m - 1	$n / (n + m - 1)$	p_f	Destroy rival's spermatophore.

tophore with one of his own, *regardless* of how rare or common the competitor's spermatophores are, than to deposit a spermatophore on the substrate. The advantage over spermatophore deposition on the substrate is greatest when only a few spermatophores have been deposited by the ♂ and his competitor(s), because under these circumstances the elimination of one of the competitor's spermatophores will greatly reduce the competitor's chance of inseminating the ♀.

Fig. 25: Rival ♂♂ depositing spermatophores near a ♀ *Ambystoma maculatum*.
From a photograph



The second kind of evidence is the behavior of interacting ♂♂. ♂♂ actively follow behind competitors and cover their spermatophores. By so doing the ♂ is assured of covering the competitor's, and not his own, spermatophore. In two instances I watched the first ♂ circle around and deposit a third spermatophore as in Fig. 26.

The absence of rapid tail undulations during spermatophore deposition is probably related to this male tendency. In *A. tigrinum* the ♂ apparently uses rapid tail undulations to waft cloacal secretions to the ♀. He thereby notifies her of his position and the site of his spermatophore. Under the crowded courtship settings of *A. maculatum* rapid tail undulations would be more likely to attract an interfering ♂ than a ♀.

The probability of depositing secondary spermatophores does not change with the number of courting ♂♂ in *Ambystoma maculatum*. Apparently ♂♂ use every available opportunity to cover spermatophores even when they court alone with a ♀.

The distribution of multiple spermatophores shows a significant deviation from a Poisson distribution in *A. maculatum* (Table 6). This suggests that the

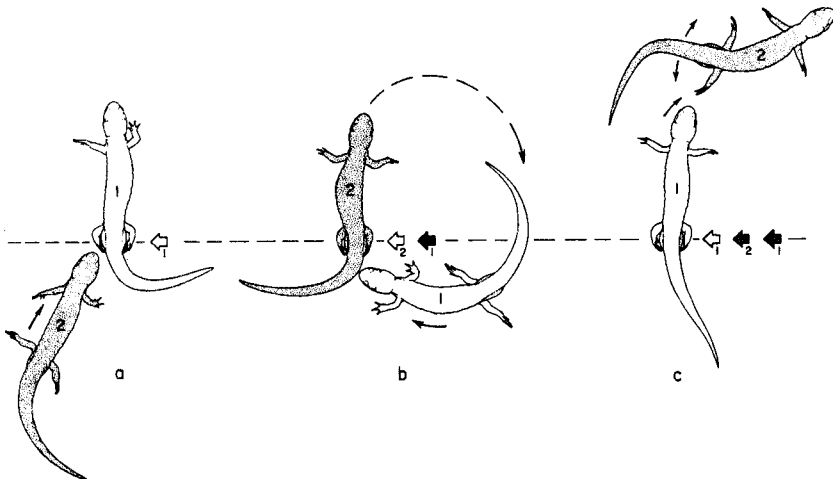


Fig. 26: Sexual interference in *Ambystoma maculatum*. a) ♂2 approaches as ♂1 deposits a spermatophore (indicated with open arrow 1). b) ♂1 has circled around as ♂2 now covers the spermatophore of ♂1 with one of his own. c) ♂1 has moved behind ♂2 and now covers the spermatophore of ♂2 with his spermatophore. The spermatophore is now a triple structure as in Fig. 27. Reconstructed from taped observations

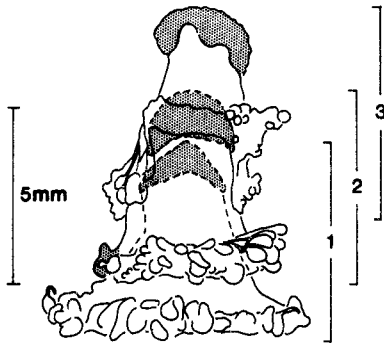


Fig. 27: A triple spermatophore of *Ambystoma maculatum*. Notice that the sperm masses of the two lower spermatophores are completely enclosed by the spermatophore bases on top of them

probability of spermatophore covering varies with the number of spermatophores already in the stack. The deviation consists of a deficiency of double spermatophores and an overabundance of other types. Observation of courting $\delta\delta$ indicates that spermatophores are found by touch. Since double spermatophores are taller than single spermatophores, perhaps they are found more readily by courting $\delta\delta$. Thus double spermatophores may be underrepresented because they are found more readily than single spermatophores and are converted into triples. No such argument is necessary for *A. tigrinum*. This species does not show a significant deviation from a Poisson distribution of multiple spermatophores (Table 6).

Covering of spermatophores probably arose from a tendency to secure spermatophores on projections from the substrate. In the field, leaf edges and sticks are particularly favored as sites of spermatophore deposition in *A. maculatum* and other *Ambystoma* species (UZZELL pers. comm.). The δ clasps

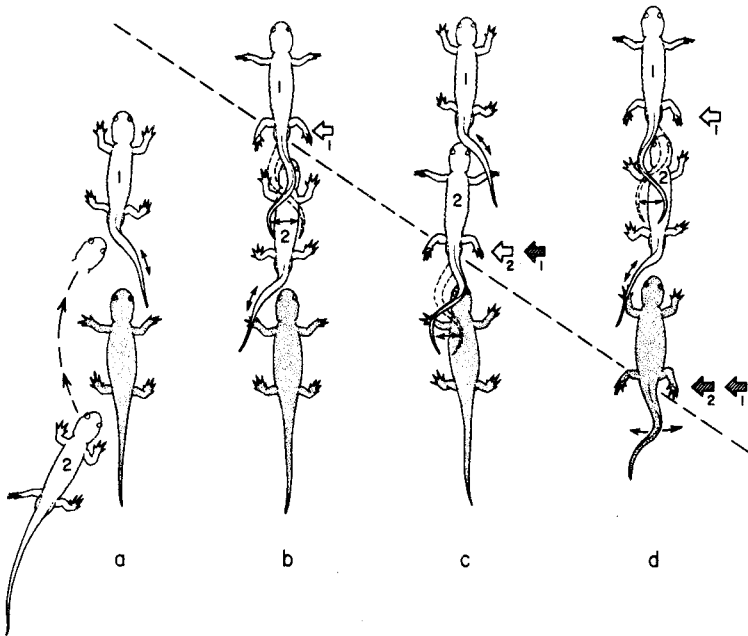


Fig. 28: Sexual interference in *Ambystoma tigrinum*. a) $\delta 2$ intrudes as $\delta 1$ leads the ♀ (stippled) in a tail-nudging walk. b) $\delta 2$ has now moved between $\delta 1$ and the ♀ and monitors the ♀ 's position by tapping with his tail. $\delta 1$ deposits a spermatophore. c) $\delta 2$ deposits a spermatophore on top of the spermatophore of $\delta 1$. The ♀ continues to follow $\delta 2$. d) The ♀ attempts to retrieve sperm from the double spermatophore. Only the sperm of $\delta 2$ is accessible, however. $\delta 1$ deposits another spermatophore. Reconstructed from taped observations

such projecting objects with one or both hindfeet during spermatophore deposition (UZZELL 1969 and personal observations). The same clasping behavior is employed during spermatophore covering.

B. *Ambystoma tigrinum*

1. Sexual Interference and Sexual Defense

Male *Ambystoma tigrinum* show two remarkable behaviors in the presence of other courting ♂♂. By shoving the ♀ (Fig. 9) the ♂ can transport her away from another ♂ and thereby insure his monopoly of her. When I confined two ♂♂ with a ♀ in a washtub 54 cm in diameter, each ♂ continually interrupted the courtship of the other by shoving the ♀ to the other side of the container. Thus shoving behavior may be used as a sexual defense (when a ♂ shoves the ♀ away from the intruder) or as a kind of sexual interference (when the intruder shoves the ♀ away from the original courting ♂). Because of these continual interruptions each ♂ deposits fewer spermatophores than he would if he courted the ♀ alone (Table 2). In the field ♂♂ are undoubtedly able to transport the ♀ completely away from intruding ♂♂.

During the tail-nudging walk, a male *A. tigrinum* relies completely upon touch to keep track of the ♀ behind him. This simple reliance makes the ♂ very susceptible to sexual interference. If a ♂ intrudes during the tail-nudging walk he may move between the ♂ and ♀ (Fig. 28). In this position the intruding ♂ actively nudges the cloaca of the ♂ in front of him and at the same time monitors the position of the ♀ by tapping with his tail. On two occasions I watched the intruding ♂ cover the spermatophores of the other ♂ with his own spermatophores as he followed behind him.

The ability of rivals to mimic female behavior places an additional premium on transportation of the ♀. This is probably the selective basis for the male trait of shoving the ♀ just before the initiation of the tail-nudging walk. By transporting the ♀ away from a competitor the ♂ lessens the chance of sexual interference during the tail-nudging walk.

2. The Courtship Setting

The absence of courtship aggregations in *A. tigrinum* is undoubtedly due to the ability of the ♂ to transport the ♀ away from rival ♂♂. HENRY WILBUR (pers. com.) observed courting pairs widely spaced in Burt Pond. It seems clear from laboratory observations that this spacing in the field is a simple consequence of ♂♂ shoving the ♀♀ away from the rival ♂♂, but additional field observations are needed to fully substantiate this point.

The ability of the ♂ to transport the ♀ away from rival ♂♂ is particularly significant when we consider the capacity of rival ♂♂ to find conspecifics. Male *A. tigrinum* apparently locate other animals by water disturbances and perhaps by pheromones. In open water ♂♂ can locate other animals readily. On one night, 17 male *A. tigrinum* were trapped in a single unbaited minnow trap that floated on the surface of Burt Pond (HENRY WILBUR, pers. com.). Apparently each successive ♂ was attracted by the movements of the other ♂♂ within the trap. The two openings of the trap were so small that each ♂ had to force his way inside. Early one morning at Burt Pond I observed a group of 8 ♂♂ milling about the screen cylinders that housed pairs of *A. tigrinum*. These ♂♂ had probably been attracted to the cylinders by the

activities of the courting animals during the night, but no spermatophores were deposited outside the cylinders. The ♂♂ repeatedly turned towards and nudged one another as they moved about. It seemed that they responded to shock waves produced by the movements of the other ♂♂. At Ellicott Pond in California, however, I watched a ♂ court successfully with a ♀ while a group of 6 ♂♂ nudged one another only 1 m away. Apparently the intervening pond vegetation impeded the propagation of waves and rendered the pair undetectable to the other ♂♂. In light of the ♂'s ability to find conspecifics, particularly in unobstructed habitats, *A. tigrinum* would undoubtedly form courtship aggregations like *A. maculatum* if *A. tigrinum* ♂♂ were not able to transport the ♀ away from rivals.

The courtship season of *A. tigrinum* is very brief and probably all courtships in a pond are completed within two weeks at most. At Burt Pond in Michigan, courtships seemed to occur on only 1 to 3 nights in each of two years.

C. Plethodon jordani

1. The Courtship Setting

The courtship season of the *Plethodon jordani* population in the White-top-Mt. Rodgers area is at least 10 weeks long (mid July to early October). ♂♂ collected in early July show poorly developed mental glands and do not court when brought into the laboratory. I did observe preliminary courtship stages on 7–8 July 1968, but the earliest spermatophore was deposited on 15 July. ♂♂ collected from the latter part of July to at least late September show well developed mental glands and court in the laboratory. I made only one small collection in early October and do not know how long courtship continues into the fall, but one ♂ collected on 2 October 1969 deposited a spermatophore on 4 October. ORGAN (1958) observed courtships in the field from 3 August to 27 August.

In the laboratory ♂♂ housed with a single ♀ will continue to court and deposit spermatophores for several weeks (Fig. 22). It is possible that in the field individual ♂♂ continue to court throughout the entire 10 week courtship season. We do not know, however, whether the courtship season of individual ♂♂ is curtailed by exhaustion of spermatophore supply, lack of receptive ♀♀ or drying of forest litter in the fall. In any case, the courtship season for individual male *Plethodon jordani* is considerably longer than for *Ambystoma maculatum* (compare Figs. 7 and 22) or *Ambystoma tigrinum*.

Adults of the same sex do not congregate during the courtship season. A ♂ and ♀ may be found close together under the same surface object, which suggests that a ♂ may court more than once with a particular ♀, but such pairs are widely separated.

MADISON (1969) marked and recaptured *Plethodon jordani* during the courtship season in North Carolina and found that individuals are very sedentary. 80% of recaptures occurred within a 6.10 m radius of the initial point of capture in ♂♂ and within a 3.05 m radius in ♀♀. In a similar study MERCHANT (1972) estimated the home range to be 11.5 m² for ♂♂ and 2.8 m² for ♀♀. When animals are displaced they return to the initial area of capture with surprising accuracy (MADISON 1969; MADISON and SHOOP 1970).

The density of courting ♂♂ in *A. maculatum* populations is 7 (0.22 ♂♂/m², HUSTING 1965) to 100 times (3.03 ♂♂/m², WHITFORD and VINEGAR 1966) greater than the comparable density in *Plethodon jordani* (MADISON and

SHOOP 1970). The effective density of courting *A. maculatum* ♂♂ is actually much greater because ♂♂ tend to form aggregations. There is no published information on the density of courting *A. tigrinum*, but casual observations suggest that their densities are greater than *P. jordani* and may approach the uniform densities reported for *A. maculatum*.

Because of the lower density, more even dispersion and sedentary nature of male *Plethodon jordani* intrusions by rival ♂♂ during courtship encounters are much less likely than in *Ambystoma maculatum* or *A. tigrinum*.

2. Sexual Interference and Sexual Defense

ORGAN (1958) observed two different types of interactions between *Plethodon jordani* ♂♂ during the courtship season. First, one ♂ repeatedly interrupted his courtship with a ♀ and bit other ♂♂ in the container. Second, ORGAN observed two ♂♂ engaged in a homosexual courtship. These two ♂♂ began courting a ♀ simultaneously and then began courting each other. One ♂ then deposited a spermatophore as the second ♂ straddled his tail, just as a ♀ would. ORGAN also found a pair of animals engaged in tail-straddling walk in the field and both proved to be ♂♂. I found that both of these types of behaviors were common when two or more ♂♂ were housed with one or more ♀♀.

When a courting ♂ pursues and bites another ♂, the pursued ♂ flees. Sometimes, as ORGAN (1958) observed, an attacking ♂ may dash forward and bite a ♀. Frequently, however, the ♂ resumes courtship with a ♀ after attacking another ♂. Interactions of this kind are not restricted to captive specimens. HUTCHISON (1959) observed two ♂♂ pursuing and biting each other in the field. Aggressive interactions between courting ♂♂ have been observed in several other species of *Plethodon*, as well as in *Desmognathus*, *Eurycea*, and *Pseudotriton* (ORGAN 1961, ARNOLD 1972). These aggressive interactions between ♂♂ are probably responsible, in part, for the observed spacing of *Plethodon jordani* ♂♂ during the courtship season and serve to reduce the probability of ♂ intrusion during courtship. The importance of spacing can be more fully appreciated when we consider the second type of male interaction in detail.

Homosexual courtships between ♂♂ were so frequent during my observations in 1968 that I subsequently housed only one ♂ and one ♀ together so that I could confine my observations to heterosexual interactions. Interactions between rival ♂♂ take a rather predictable course, if biting does not occur. When several ♂♂ are housed together with a ♀, frequently an intruding ♂ will approach a courting pair. The intruding ♂ then nudges and slides his mental gland along the ♀ as he periodically performs a foot dance. When the intruder contacts the first ♂ with his snout, he may bite the first ♂ and chase him away. In other instances, and I was unable to determine what factors govern the outcome, the intruder simply slides his mental gland along the other ♂'s tail, and the ♂♂ move forward in a tail-straddling walk away from the ♀. The intruding ♂ behaves very much like a ♀, for he pauses periodically as the other ♂ leads him forward. Tail-straddling walks with an intruding ♂ are of shorter duration than walks with a ♀, and I never observed mental gland slapping. An intruder ♂ apparently behaves as a very receptive ♀ during the tail-straddling walk. During the 3–4 min required for spermatophore deposition the intruding ♂ remains essentially motionless astride the leading ♂'s tail (Fig. 29a) just as the ♀ would. After spermatophore deposition the intruding ♂ moves forward behind the leading ♂ (Fig. 29b), but he does

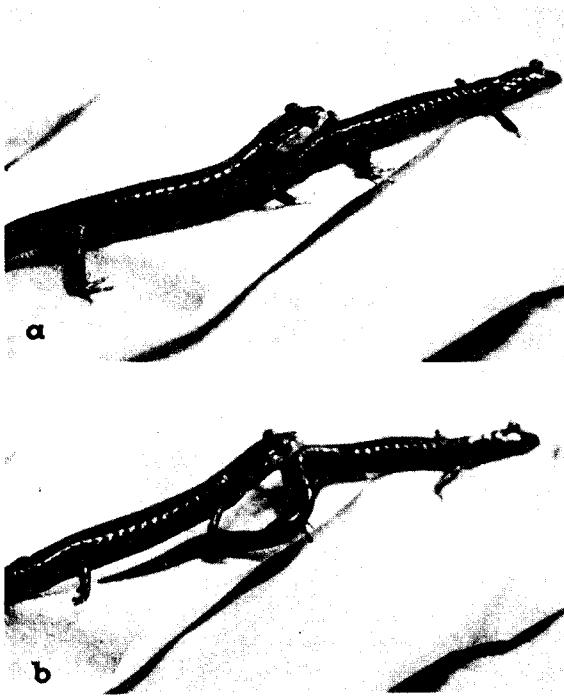


Fig. 29: Sexual interference in *Plethodon jordani*. a) One ♂ behaves as a ♀ and straddles the tail of another ♂ while he deposits a spermatophore. b) The leading ♂ moves forward away from his spermatophore and the other ♂ follows

not stop over the spermatophore. At this point the intruding ♂ may either depart from the leading ♂, or the leading ♂ may suddenly swing around and bite the intruding ♂, which flees rapidly.

The following points suggest that homosexual courtship in *Plethodon jordani* is not simply a sexual pathology peculiar to captivity, but rather a technique for sexual interference. First, intruding ♂♂, which induced another ♂ to deposit a spermatophore, were later observed to court and deposit spermatophores with ♀♀ in the same evening. Intruding ♂♂ are therefore not obligate homosexuals. Second, once a ♂ has deposited a spermatophore, he usually does not court and deposit spermatophores for several days (Fig. 22). Thus a ♂ that is induced to deposit a spermatophore by a rival ♂ is unable to inseminate a ♀ for several days. Third, ♀♀ only find a spermatophore when they follow a ♂ after spermatophore deposition. Spermatophores deposited during homosexual courtship are reproductive failures. They are never found by and never inseminate ♀♀. Fourth, homosexual courtship do not occur, and spermatophores are not deposited, unless ♀♀ are present in the container. Homosexual courtships only occur when two courting ♂♂ converge on and initiate courtship with the same ♀. Fifth, the behavior of the intruding ♂, once the other ♂ begins a tail-straddling walk, is remarkably similar to a ♀. Sixth, homosexual courtships between ♂♂ occur in the field (ORGAN 1958) as well as in captivity. Finally, homosexual courtships are not peculiar to *Plethodon jordani*. They also occur in *Plethodon glutinosus*, *P. ouachitae*, *P. yonahlossee* (ORGAN, pers. comm.) and in the closely related genus *Ensatina*. All of these observations are consistent with the view that there has been selection for ♂♂ with the capacity to distract other ♂♂ from a ♀ and then induce them to deposit spermatophores that serve no reproductive function. Such distracting ♂♂ depress the fitness of the other ♂ and at the same time are able to return to the ♀ and inseminate her.

♀-mimicry in *Plethodon* constitutes a temporary loss in courtship time for the pseudo-♀ (unlike the comparable behavior in *Ambystoma tigrinum* in

which the ♂ may inseminate a ♀ as he dupes a rival). Such mimicry can only evolve when there is a very small chance that the pseudo-♀ would find and inseminate a ♀ during the time required to dupe a rival ♂. This is, of course, the case in *Plethodon jordani*. Since adults are so widely spaced it is unlikely that a ♂ could find a ♀ in the small amount of time it takes to depress the fitness of a rival.

VI. Selection for Aspects of Courtship Behavior

A. Crowding and Length of Courtship Season as Selective Pressures

The data in Table 8 suggest that ♂♂ pay a price for rapid courtship. If the ♂ courts rapidly, as in *Ambystoma maculatum*, the sites of spermatophore deposition will be relatively unpredictable (Fig. 31) and it will be correspondingly difficult for the ♀ to find any particular spermatophore. If the ♂ courts slowly, as in *Plethodon jordani*, he will be able to coordinate the placement of his spermatophore with the position of the ♀ and so the ♀ will be more likely to find it. This functional relationship between courtship time expended per spermatophore and the probability that the ♀ will find it is shown diagrammatically in Fig. 32. HALLIDAY 1974 has confirmed this relationship in the salamander *Triturus vulgaris*; the longer the ♂ courts a ♀, the greater the probability of successful sperm transfer per spermatophore.

Table 8: Summary of courtship variables

	<i>Ambystoma maculatum</i>	<i>Ambystoma tigrinum</i>	<i>Plethodon jordani</i>
Total elapsed time (min)	178.7	126.4	1398.0
Total number of spermatophores	131	28	25
Courtship time per spermatophore (min)	1.4	4.5	55.9
Complexity of male behavior	+	++	+++
Variability in site of spermatophore	+++	++	+
Probability of ♀ finding a spermatophore	0.39	0.61	0.94
Mean Number of spermatophores per courtship	40.41	20.59	1.04
Length of courtship season (weeks)	1 - 2	1 - 2	10
	Maximum	Maximum	
Probability of a rival ♂ intruding during courtship.	+++	++	+

Note: The series +, ++, +++ indicates increasing values of a particular variable.

Why do *Ambystoma maculatum* court rapidly and deposit so many spermatophores? Both of these characteristics seem to be adaptive responses to crowded courtship situations and a short courtship season. When rival ♂♂ are simultaneously courting a ♀, the ♂ that deposits spermatophores at a slow rate will find that the ♀ will be inseminated by other ♂♂. Hence when courtship occurs in crowded circumstances, selection will favor ♂♂ that deposit spermatophores rapidly. If a ♂ is able to deposit many spermatophores he will be able to compensate for the reduction in his success caused by competitor spermatophores and elimination of his spermatophores by rivals.

The probability of intrusion during courtship is actually a more proximate selective pressure than crowding per se. *Ambystoma tigrinum* court in

relatively crowded ponds, but ♂♂ have an effective means of transporting the ♀ and can thereby reduce the probability of intrusion by rivals. As a consequence selection has favored an elaboration of the communication be-

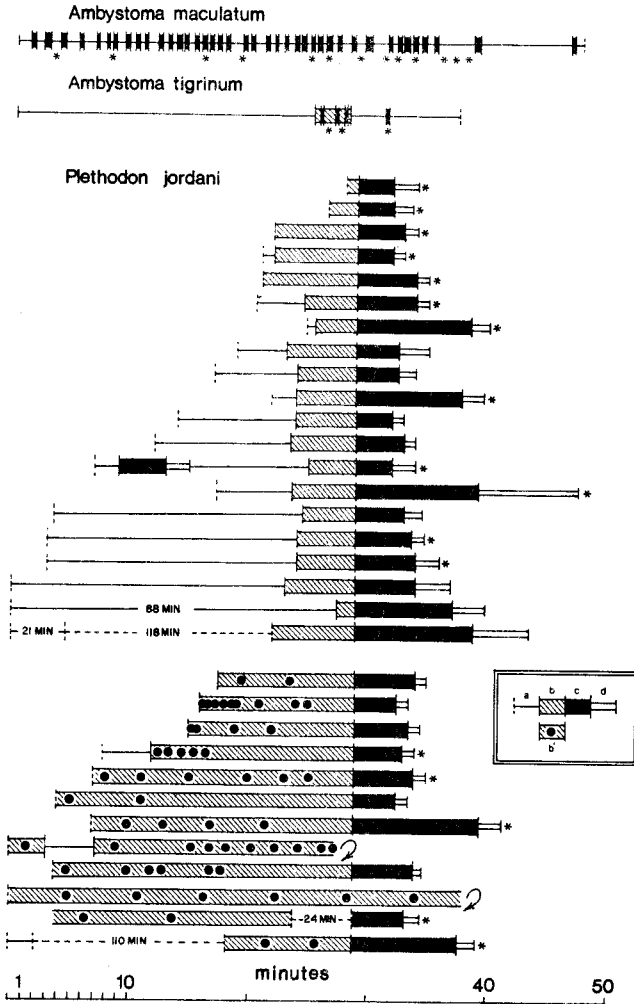


Fig. 30: Temporal patterns of courtship behavior. Each horizontal line represents the record of a courtship. Lengths of each segment on a line correspond to the duration of each activity (see legend on figure). The two *Ambystoma* courtships are shown from the time the ♂ first contacted the ♀ after the pairs were introduced in their tanks. Each of the 30 *Plethodon* courtships (23 different pairs) were already in progress when observations began. These courtships are arbitrarily aligned at the same time of onset of spermatophore deposition. Only courtships which lead to spermatophore deposition are shown. Many *Plethodon* courtship sequences did not lead to spermatophore deposition (see text). a) Indicates approach to and nudging of the ♀. b) Indicates tail-nudging walk (*A. tigrinum*) or tail-straddling walk (*P. jordani*). b') Indicates tail-straddling walk with mental gland slapping; each slap of the mental gland on the ♀'s head is indicated with a solid circle. c) Indicates spermatophore deposition. d) Positioning of the ♀ over the spermatophore. Stars indicate either the time at which the ♀ found a spermatophore and squatted on it (both *Ambystoma*) or that sperm transfer was successful (*Plethodon*). Vertical dashed lines represent the onset or termination of observations. Horizontal dashed lines represent periods of no observation

tween the ♂ and ♀ in *Ambystoma tigrinum*. The ♂ invests more courtship time in each spermatophore and deposits fewer of them.

Intrusions by rival ♂♂ are even less likely in *Plethodon jordani*. Courtship does not occur in restricted areas as it does in the pond-breeding *Ambystoma*. Instead animals are widely spaced during the courtship season, and this dispersion is further enhanced by the aggressive interactions between courting ♂♂. Since intrusion by a rival is relatively rare, selection has favored a large courtship investment in each spermatophore.

When the courtship season is long, a ♂ that conserves spermatophores early in the season will be able to inseminate ♀♀ encountered later in the season. A *Plethodon* ♂ deposits only one spermatophore per courtship but he can continue to court throughout the 10 week courtship season. When the courtship season is extremely short there is little selection to conserve spermatophores for later courtship. Thus a male *Ambystoma maculatum* will use a large portion of his spermatophore supply with a single ♀.

These arguments for the evolution of courtship variables in ♂♂ are quite analogous to LACK's (1954) arguments for the evolution of clutch size. Number of spermatophores per courtship, courtship time per spermatophore, and the

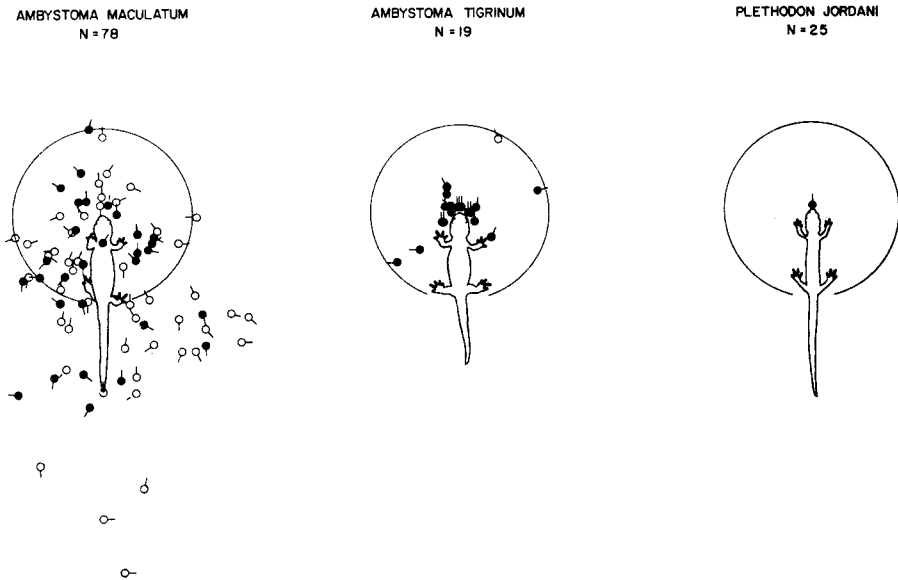


Fig. 31: Sites of spermatophore deposition in relation to the ♀'s body. Small solid circles show the sites of spermatophores deposited on the substrate. Small open circles show spermatophores deposited on other spermatophores. Small lines indicate the direction the ♂ was facing at the onset of spermatophore deposition. Large circles indicate a radius of one body length about the ♀'s snout

probability that the ♀ will find a particular spermatophore are the male analogs of clutch size, female investment per ovum (e. g. amount of yolk, retention time in the ♀, brooding behavior, etc.) and survivorship of the young. LACK argues that an increase in clutch size will normally imply a decrease in survivorship of the young since the ♀ must necessarily lower her investment in each offspring. Likewise when ♂♂ deposit many spermatophores rapidly, there is a smaller chance of success with each spermatophore than when the ♂ takes a longer time with fewer spermatophores. A major constraint on the ♀'s

total investment is her body size since a particular body volume may accommodate either many small eggs or a few large ones. It is more difficult to specify the comparable constraint in ♂♂. There seems to be such a constraint since I can find no instances in salamanders in which the ♂ deposits many spermatophores with each ♀ as in *Ambystoma* and also makes a large courtship investment with each spermatophore as in *Plethodon* (ARNOLD 1972). It is clear that such a ♂ could not court as many ♀♀ as a ♂ with a smaller total investment with each ♀. The constraint on the ♂'s total investment with each ♀ is probably then the reduction in the success of future courtships. Likewise a large investment by the ♀ in a clutch may lessen her subsequent survivorship and fecundity.

B. Female Behavior as a Selective Pressure

The preceding discussions have viewed two male courtship variables (courtship time per spermatophore and number of spermatophores per courtship) as adaptations to the length of the courtship season and the interference behaviors of rival ♂♂. These variables may be adaptations to the ♀'s behavior, however. In particular, the longer courtship time in *Plethodon* may be an indication that *Plethodon* ♀♀ are less receptive than *Ambystoma* ♀♀. Let us suppose for the moment that this is the case even though we have no experimental evidence for differences in female sexual receptivity². Such a difference in female receptivity might be anticipated from differences in length of courtship season. There is a distinct advantage to early mating, and hence high sexual receptivity, in *Ambystoma* ♀♀. Early mating and egg deposition will yield larvae that begin growth sooner than larvae from late matings. Larger, early larvae are advantageous for several reasons since larvae must race to metamorphose in temporary ponds (WILBUR 1972 analyzed larval competition). In addition, a female *Ambystoma* with low sexual receptivity runs the risk of not being inseminated during the short courtship season. In *Plethodon* ♀♀, however, there is little or no advantage to early mating or high receptivity. A ♀ that mates early in the courtship season (late July) must still wait until the following spring to lay her eggs. There may even be an advantage to low receptivity. Such ♀♀ can review several ♂♂ before insemination. I have included these arguments in Fig. 32 (vertical arrow from length of courtship season to sexual receptivity of ♀) because of their heuristic value, despite the lack of evidence for them. Fig. 32 also suggests that low sexual receptivity in the ♀ will favor ♂♂ with lengthy and elaborate courtships. Thus male courtship time per spermatophore may represent adaptation to either female receptivity or to the likelihood of intrusions by rival ♂♂.

On the other hand, I see no way to argue that the intrinsic sexual receptivity of the ♀ affects the evolution of spermatophore number per courtship. In particular, ♂♂ do not compensate for low sexual receptivity by depositing more spermatophores. Rather, in each species ♂♂ deposit few or no spermatophores with non-receptive ♀♀. The species with the lowest female sexual receptivity as inferred from courtship time (*P. jordani*) deposits fewer spermatophores per courtship than the other species.

²) I am supposing here that there are intrinsic differences between the species in sexual receptivity. During courtship the sexual receptivity of the ♀ may be modified by the ♂'s courtship (e.g. by the secretions of the mental gland), but the arguments deal with the intrinsic differences among species before insemination or courtship.

C. Other Possible Explanations

I find no evidence that selection for speed of courtship or number of spermatophores per courtship in these salamanders is related to the medium in which courtship occurs or to species identification.

NOBLE (1931) noted that the courtship patterns of salamanders evolve very slowly and that they are little affected by the habitat in which courtship occurs. Subsequent observations have substantiated both points. Although *Ambystoma opacum* courts on land, its courtship behavior is very similar to *Ambystoma maculatum*, which courts under water (NOBLE and BRADY 1933;

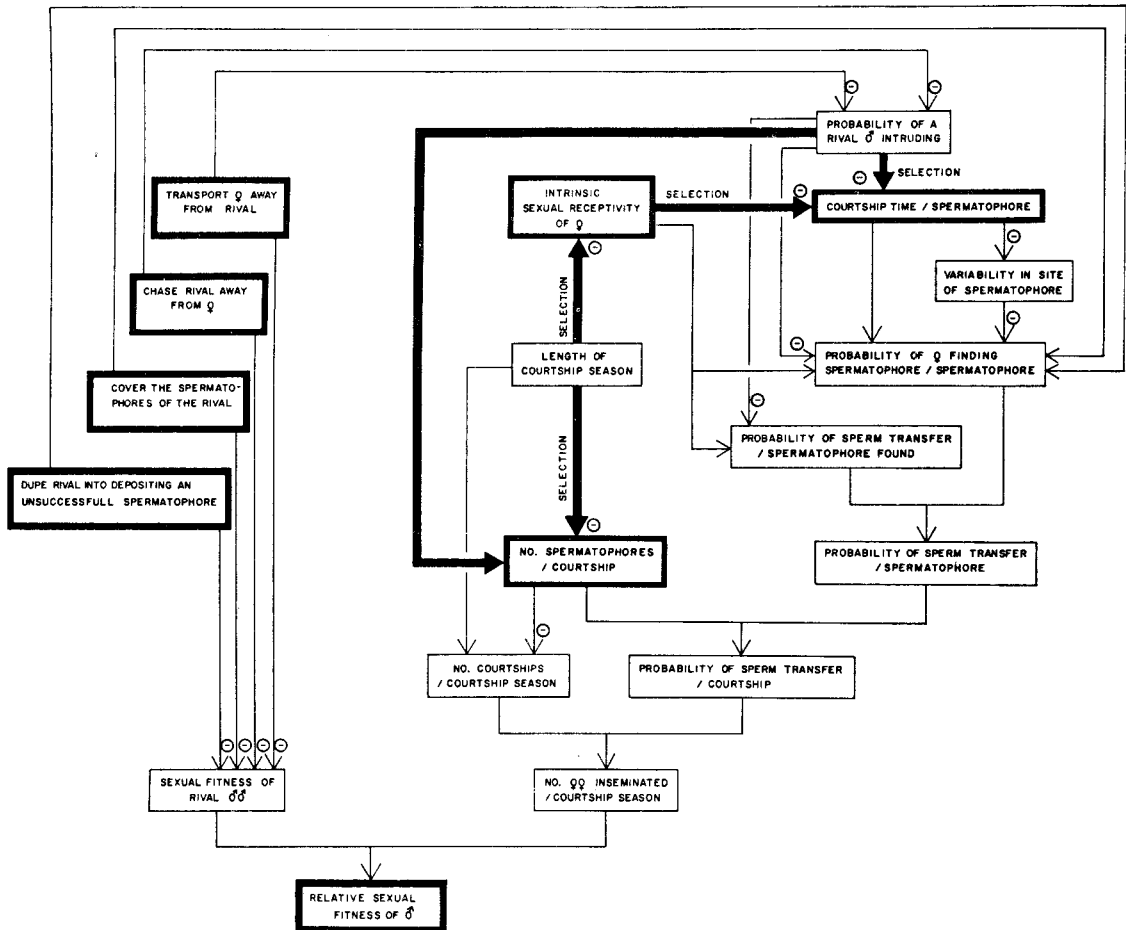


Fig. 32: Relationship of various behaviors to male fitness and some effects of selection. Two aspects of male courtship and four kinds of selfish behavior are shown in heavy boxes. The functional relationships of these variable to various components of male fitness and ultimately to the relative fitness of the performing ♂ are shown with light arrows. Selection will tend to maximize the relative fitness of the ♂. Three variables extrinsic to the ♂ (rival ♂♂, female receptivity, and length of the courtship season) are expected to act as selective pressures as indicated by heavy arrows. All arrows represent positive influences unless otherwise indicated (as the variable at the start of the arrow increases, so will the other variable). Arrows with negative sign indicate negative influences (as the variable at the start of the arrow increases, the variable at the end of the arrow will decrease). See text for further explanation

ARNOLD 1972). Like *A. maculatum*, *A. opacum* courts rapidly with little coordination of ♂ and ♀ activities, deposits many spermatophores in a wide variety of sites and, predictably, has a spermatophore with radial symmetry. Like aquatic-courting *Ambystoma*, *A. opacum* will deposit spermatophores on top of other spermatophores. Conversely, the constellation of courtship

Table 9: A sample of behaviors which accomplish reproductive interference or defense in various animal groups^a

Stage	Reproductive Interference	Reproductive Defense	Reference
Finding mate	destruction of bowers by male satin bower birds	territorial defense	MARSHALL 1954
Courtship	nest destruction by male sticklebacks	territorial defense	ASSEM 1967
	male salamanders dupe rivals into unsuccessful spermatophore deposition. <i>Ambystoma tigrinum</i> <i>Plethodon jordani</i>	♂ transports ♀ away from rival ♂ bites and chases rival away from ♀	
	take-over attempts by male salamanders <i>Ambystoma laterale</i> <i>Taricha</i>	♂ swims away with ♀ ♂ swims away with ♀	STOREZ 1969 ARNOLD 1972
	?	precopulatory monopoly in insects (e. g. sepsids, cicindelids, staphylinids, cerambycids)	PARKER 1970
Release of gametes	interruption of copulation by male lizards <i>Anolis</i> <i>Sceloporus</i>	territorial defense	RAND 1967 CREWS 1975
	male <i>Ambystoma</i> cover the spermatophores of rivals	territorial defense	HUNSAKER 1962
Transfer of gametes	male millipedes, collembola, pseudoscorpions and oribatid mites destroy the spermatophores of rival ♂♂	rapid courtship ?	SCHALLER 1971
	stolen fertilizations in sticklebacks	territorial defense	ASSEM 1967
	take-overs by rival ♂♂ in insects (e. g. orthoptera, scotaphagids, cerambycids) with possible sperm displacement (<i>Drosophila</i>) or simple sperm competition; take-over during oviposition in the odonate <i>Platyhemis</i>	1. post-copulatory monopoly of ♀ in insects (e. g. odonates, orthoptera, reduviids, curculionids) and amphipods. 2. prolonged copulation in houseflies. 3. copulatory plugs in mosquitoes and bees. 4. insemination reaction in <i>Drosophila</i> . 5. spermatophragma of lepidoptera.	PARKER 1970
Fertilization	egg-eating in centrarchid fish	territorial defense	KEENLYSIDE 1972
	stealing of nest materials in birds shag Puerto Rican honeycreeper	territorial defense territorial defense	MURPHY 1936 BIAGGI 1955
	redhead ducks sometimes lay eggs in nests of conspecifics	?	WELLER 1959
	killing of unrelated young by male langurs	exclusion of foreign ♂♂ from troop	SUGIYAMA 1967 HRDY 1974
Offspring			

^a (Behaviors which act before fertilization might be termed *sexual interference* or *defense*, while those that act after fertilization might be termed *parental interference* or *defense*.)

traits in *Plethodon jordani* is not a simple consequence of courtship on land. *Eurycea bislineata*, with courtship much like *Plethodon*, can court both on land and underwater (ARNOLD 1972).

Plethodon jordani is sympatric with more congeneric species than either *Ambystoma maculatum* or *A. tigrinum*. The long time required before spermatophore deposition and the complex programming of ♂ behavior in *Plethodon jordani* might be the result of selection to avoid matings with other species. This appears, however, not to be the case. In the laboratory, male *Plethodon jordani* from Whitetop Mtn. did not even initiate courtships with sympatric female *P. glutinosus* or *P. yonablossee*. In *Plethodon*, as in other salamanders (NOBLE and BRADY 1930; TWITTY 1955), species recognition is apparently accomplished by chemoreception at the very onset of courtship and so before the ♂ uses his complex courtship behaviors.

VII. The Evolution of Reproductive Interference and Reproductive Defense

Sexual selection as proposed by DARWIN depends upon differences in reproductive³ success of members of the same sex that result from 1) competition between rivals of that sex or 2) mating choices exerted by the opposite sex. Considering just the first aspect, it is useful to recognize that contests between rivals can have both a defensive and an offensive component (DARWIN 1859, p. 89—90; DARWIN 1874, p. 238; PARKER 1970). We may refer to these two components as reproductive defense and reproductive interference.

Examples of reproductive interference and defense are familiar to students of animal behavior, but these phenomena are seldom the focus of study. An incomplete survey of such behavior is presented in Table 9; PARKER 1970 discussed these aspects of insect behavior. The table is arranged to indicate that acts of interference and defense can occur at a variety of stages in the cycle of reproduction.

A. Comparisons with salamanders

Some of the examples in Table 9 show striking parallels with behaviors observed in salamanders. Covering of spermatophores is a general phenomenon in the salamander genus *Ambystoma* and can have the double advantage of promoting the sexual success of the ♂ by increasing his spermatophore count while simultaneously eliminating a rival's spermatophore. Many soil arthropods with indirect sperm transfer show analogous behavior. In some millipedes, collembola, pseudoscorpions and oribatid mites, ♂♂ destroy the spermatophores they encounter and replace them with their own spermatophores (SCHALLER 1971). The behavior of a pseudoscorpion in the genus *Serianus* is particularly fascinating (WEYGOLDT 1966). The ♂ deposits a spermatophore *only* when he has just encountered a ♀. He then spins a funnel-shaped structure around it which the ♀ uses to find the spermatophore. When a ♂ finds a spermatophore and funnel he destroys them even if they are fresh. If a ♀ is present, the ♂ deposits a new spermatophore and constructs a new funnel. This appears to be a clear instance of sexual interference and not, as

³) It is clear that DARWIN used the term *sexual* selection to indicate that competition occurs between members of the same sex and not to indicate competition for sexual success as opposed to parental success (DARWIN 1874, p. 236). Thus egg-eating or infanticide can be manifestations of sexual selection as DARWIN conceived it.

has been suggested for other arthropods (SCHALLER 1971), a behavior designed to insure that ♀♀ encounter fresh sperm. *Serianus* presents two unresolved problems, however. First, why does the ♂ destroy the funnel structure of his rival? A second problem is presented by female behavior. Female *Serianus* as well as ♀♀ in other arthropod groups (SCHALLER 1971) sometimes destroy spermatophores just as the ♂♂ do. Is this a non-adaptive behavior or is it a manifestation of sexual competition among ♀♀? It would be useful to know if uniseminated ♀♀ give a sexual response to spermatophores and then change to destructive behavior once they have been inseminated.

In these diverse arthropods, as in *Ambystoma*, the unknown parentage of the spermatophore may present no difficulty to the ♂. Even if the ♂ destroys his own spermatophores in proportion to their abundance, he still profits more from spermatophore destruction and replacement than from simple spermatophore deposition (see Table 7 and the discussion below).

Mimicry of female behavior, as in *Ambystoma tigrinum* and *Plethodon*, seems at first to be an improbable mode of sexual interference. However, the ♂♂ of some fish mimic female coloration and exploit the sexual behavior of their rivals (MORRIS 1958; BARLOW 1967; ASSEM 1967). The phenomenon has been most thoroughly studied in the stickleback, *Gasterosteus aculeatus*, and is called "sneaking" (ASSEM 1967). Once a ♂ begins courtship with a ♀ and leads her to his nest, rival ♂♂ in adjoining territories may suddenly assume the more cryptic coloration of the ♀ and move slowly into the territory of the courting ♂. Once the ♀ enters the nest and begins to spawn, the sneaker dashes toward the nest. In approximately half of the cases sneakers were able to fertilize the clutch *before* the resident ♂. As ASSEM (1967) points out, such stolen fertilizations have a double advantage because the sneaker fathers an extra clutch at the expense of the resident who rears his offspring.

Salamander ♂♂ monopolize the ♀ and thereby guard against interference by rivals by transporting the ♀ just before the vulnerable process of sperm transfer (*Ambystoma tigrinum*) or by biting and chasing away intruders (*Plethodon jordani*, as well as many other plethodontids). Territoriality serves the same function, as well as others, in sticklebacks. ♂♂ actively chase sneakers from their territories if they detect them (ASSEM 1967).

B. Coevolution of Reproductive Interference and Reproductive Defense

"So again, if the chief service rendered to the male by his prehensile organs is to prevent the escape of the female before the arrival of other males, or when assaulted by them these organs will have been perfected through sexual selection, that is, by the advantage acquired by certain individuals over their rivals." (DARWIN 1874, p. 283).

It seems likely, as DARWIN proposed, that sexual defense evolves in response to sexual interference. PARKER (1970) also argues that ♂♂ will tend to evolve mechanisms to usurp fertilizations from sperm previously stored in the ♀ and that this will in turn favor mechanisms in ♂♂ that prevent subsequent insemination of the ♀. In support of this position PARKER noted that fights between ♂♂ for possession of the ♀ are very common in insects, and that copulatory "take-over" may be observed by the patient investigator. PARKER discussed an amazing array of behaviors and other mechanisms by which ♂♂ guard ♀♀ against insemination by rivals. Some of these offensive and defensive mechanisms are shown in Table 11.

For every kind of reproductive defense listed there is either a corresponding kind of reproductive interference or a strong suspicion that it occurs. In some instances modes of defense appear to be recent and specific adaptations

to sexual interference (e. g. transportation of the ♀ in *Ambystoma tigrinum*) while in others the mode of defense has a long evolutionary history related to many aspects of reproduction (e. g. territoriality in fish, birds and lizards).

Examples of interference by conspecifics can be found for all steps in the reproductive sequence. In particular taxonomic groups, however, examples of interference are concentrated at particular reproductive stages. This reflects the vulnerabilities peculiar to each group. For example, groups with spermatophores attached to the substrate (most salamanders and many soil arthropods) are particularly vulnerable to intrusion during the intricate process of sperm transfer.

PARKER (1970) noted that the maintenance of interference behaviors will depend upon a small but significant chance of failure in the defenses of rivals. Given imperfect defenses for whatever reason we can expect a rapid co-evolution of interference traits and defenses that cancel their effect. Thus our best evidence for the adaptiveness of interference behaviors will come from studies of closely related species with a "known" phylogeny. Although we lack the latter aid, it is probably significant that *Ambystoma tigrinum* differs from *A. maculatum* in the possession of a vulnerability to interference (the tail-nudging walk), an exploitation of this vulnerability (Fig. 28) and a sexual defense (Fig. 9).

C. Reproductive Interference as Selfish or Spiteful Behavior

The examples of reproductive interference in Table 11 share the characteristic that performance of the act causes a loss in fitness to a reproductive rival. Considering such behaviors, HAMILTON (1970) posed the following question. Is the harm delivered to a rival merely an unfortunate consequence of adaptation for reproduction or is the harm adaptive in itself? HAMILTON (1970, 1971) defined some of the circumstances under which selfish, spiteful and even strongly spiteful⁴ behavior will be adaptive. The conditions for selection of strongly spiteful behavior are very restrictive, usually impractical and promote the extinction of the population (HAMILTON 1971). Selfish behavior, however, will often be selected over its more altruistic alternatives. For such behaviors the harm delivered to the rival constitutes an important contribution to the act's adaptive value; it is not incidental.

For some actual behaviors, however, we can readily dismiss the harm to conspecifics as incidental; the harm has played no role in the selective origin and maintenance of the behavior. For example, during the nesting season ♀♀ of the lizard *Iguana iguana* may reduce the fitness of other ♀♀ in at least three ways: 1) by usurping burrows, 2) by excavating nests and eggs and 3) by occasionally burying other ♀♀ (RAND 1968). All of these effects appear to be simple consequences of typical iguanid nesting behavior. The effects occur because of the high density of ♀♀. The harm is unavoidable.

For other behaviors, however, it is more difficult to suppose that the harm is unavoidable or incidental. In particular, consider the following behaviors: destruction of bowers by male bower birds, destruction of spermatophores

⁴) A selfish behavior harms others while benefiting the self, a spiteful behavior harms others with neither harm nor benefit to the self, and strongly spiteful behavior harms others and also the self. The harm or benefit just specified is to the *absolute* fitness of the recipient or performer. Thus spiteful behavior causes a loss in the absolute fitness of a rival and so can result in a gain in the relative fitness of the performer. The terms are useful ways to signify regions in a two dimensional space where one axis is the change in absolute fitness of a conspecific(s) and the other is the change in absolute fitness of the self.

(male millipedes, Collembola, pseudoscorpions, oribated mites), covering of spermatophores by ambystomatid salamanders, male mimicry of female behavior in sticklebacks and plethodontid salamanders. In each of these cases a less selfish, alternative behavior is feasible but not employed (i. e., the harm is avoidable) and a special behavior is employed (i. e. the harm is *not incidental* to some adaptive activity). These behaviors probably owe their origin, perfection and preservation to the harm they cause to reproductive rivals.

It is possible to strengthen an inference of adaptive harm by comparing the behavior to its more altruistic and spiteful alternatives. Table 7 considers the consequences of a purely sexual behavior (spermatophore deposition on the substrate), a selfish behavior (spermatophore deposition on another spermatophore) and a spiteful behavior (spermatophore destruction). For the last two behaviors we assume that the δ cannot distinguish his spermatophores from those of his rival. We previously showed that there is always an instantaneous advantage in covering a spermatophore rather than depositing a spermatophore on the substrate. From Table 7 we can also find that deposition on the substrate is always superior to spermatophore destruction (i. e. $p_b > p(p_e) + (1-p)p_f$ for all n and $m \geq 0$). Thus for this example, selfish behavior is superior to purely sexual behavior, and sexual behavior is superior to spiteful behavior. These conditions are sufficient to cause skepticism that simple spermatophore destruction (should we observe it) is an adaptive behavior.

The situation is changed somewhat if we suppose that the δ *can* distinguish his spermatophores from those of his rival (i. e. a δ either covers or destroys rival spermatophores when he encounters them but otherwise deposits spermatophores on the substrate). The act of spermatophore destruction is less spiteful than before, since the δ never destroys his own spermatophores. In this case we find, as before, that spermatophore covering is always superior to deposition on the substrate (i. e. for all $n \geq 0$ and all $m \geq 1$). Spermatophore covering is also superior to spermatophore destruction for $n \geq 0$ and $m \geq 1$, and equivalent when $m = 1$. Spermatophore destruction is superior to deposition on the substrate when $m < n + 1$, equivalent when $m = n + 1$, and inferior when $m > n + 1$. This suggests that spermatophore destruction might evolve if the δ can distinguish spermatophores and if his spermatophores usually outnumber his rival's by more than one. In evolutionary time, however, simple spermatophore destruction will be quickly supplanted by spermatophore covering if it is feasible.

The relatedness of rivals will also have an important effect upon the evolution of selfish or spiteful behaviors (HAMILTON 1970, 1971). Alleles for selfish or spiteful sexual interference will not increase in frequency if the phenotype's rivals tend to carry the same alleles. Thus sexual interference is unlikely to evolve if sexual competitors are usually sibs or very close relatives. Similarly, selection for an act of reproductive interference that depresses the fitness of the mate (e. g. infanticide in langurs) can be impeded if mates are relatives and can be severely impeded if both mates and rivals are relatives.

Summary

There are great differences between three species of salamanders in the amount of time the δ courts the ♀ for each spermatophore he deposits and in the number of spermatophores deposited per courtship. A consideration of these differences suggests the following hypotheses for evolution of courtship behavior in salamanders.

Courtship time per spermatophore may represent an evolutionary compromise between success with a particular ♀ and adaptation to rival ♂♂.

- a. Differences among species suggest that the longer the courtship time per spermatophore, the greater the probability that the ♀ will find it and be inseminated.
- b. For this reason it is advantageous for the ♂ to make a large temporal investment in each spermatophore (e. g. *Plethodon*).
- c. When intrusions by rival ♂♂ are likely, however, the advantages of taking a long time can be outweighed by the disadvantage that a rival will intrude and inseminate the ♀. Consequently species which mate in polyandrous aggregations (e. g. *Ambystoma maculatum*) court rapidly and expend little time with each spermatophore.

Number of spermatophores per courtship may represent an evolutionary compromise between success with a particular ♀ and adaptation to length of the courtship season.

- a. ♂♂ have a limited supply of spermatophores.
- b. Consequently ♂♂ which conserve spermatophores in each courtship may be able to inseminate more ♀♀ than ♂♂ which do not conserve spermatophores. This advantage of conservation may outweigh the disadvantage of a lowered success with each ♀ if the courtship season is long (e. g. *Plethodon*).

Male salamanders actively intrude into the courtships of other ♂♂. Such sexual interference may select for sexual defense of the ♀ by the ♂, rapid courtship, and an increase in the number of spermatophores per courtship.

Zusammenfassung

Drei Arten von Salamandern unterscheiden sich deutlich in der Länge der Zeit, die das ♂ vor einem ♀ balzt, ehe es die Spermatophore absetzt, und in der Zahl der Spermatophoren, die es pro Balz absetzt. Daraus lassen sich die folgenden Hypothesen zur Evolution der Salamanderbalz ableiten.

Die Balzdauer pro Spermatophore scheint ein Kompromiß zwischen dem Erfolg bei einem bestimmten ♀ und der Ausschaltung von Rivalen zu sein. Je länger die Balz ist, desto wahrscheinlicher findet das ♀ die Spermatophore. Also sollte das ♂ viel Zeit in jede Spermatophore investieren (z. B. *Plethodon*).

Wenn aber Rivalen hinzukommen, könnten sie in dieser Zeit die Besamung stehlen. Deshalb balzen die polyandrische Ansammlungen bildenden Arten (z. B. *Ambystoma maculatum*) schnell und investieren wenig Zeit in jede Spermatophore.

Die Zahl von Spermatophoren pro Balz scheint ein Kompromiß zwischen dem Erfolg bei einem bestimmten ♀ und der Dauer der Balzzeit. Die ♂♂ haben nur einen beschränkten Vorrat von Spermatophoren. ♂♂, die in der Balz Spermatophoren sparen, könnten mehr ♀♀ begatten als ♂♂, die nicht sparsam sind. Das kann sogar den Nachteil eines geringeren Erfolgs pro ♀ wettmachen, wenn die Fortpflanzungszeit lang ist (z. B. *Plethodon*).

Männliche Salamander drängen sich in die Balz anderer ♂♂ hinein. Solche Störungen können einen Selektionsdruck auf Verteidigung des ♀, schnelle Balz, und auf Vergrößerung der Zahl von Spermatophoren je Balz ausüben.

Acknowledgments

I am very grateful to Richard ALEXANDER, Robert COLWELL, Douglas FUTUYMA, Nelson HAIRSTON, Brian HAZLETT, Lynne HOUCK, Arnold KLUGE, William LIDICKER, James ORGAN, Donald TINKLE, Thomas UZZELL, David WAKE, Charles WALKER, and Henry WILBUR for many useful criticisms and valuable discussions.

Many people generously helped me with field work. In particular I would like to thank Albert BENNETT, James COLLINS, Douglas FUTUYMA, Ronald MARLOW, Della ORGAN, James ORGAN, Mary TILLEY, Stephen TILLEY, Donald TINKLE, Henry WILBUR and Thomas YOCUM.

Research space and facilities were provided by Mountain Lake Biological Station of the University of Virginia (Dr. James MURRAY); the Mathei Botanical Gardens (Dr. Warren WAGNER), the E. S. George Reserve (Dr. Francis EVANS) and the Museum of Zoology (Dr. Nelson HAIRSTON) of the University of Michigan; and by the Museum of Vertebrate Zoology (Dr. David WAKE) of the University of California, Berkeley.

Emily REID and Gene CHRISTMAN prepared most of the illustrations.

This work was supported by a National Science Foundation Predoctoral Fellowship at Mountain Lake Biological Station, a National Institute of Health Traineeship, a National Science Foundation grant for research in systematic and evolutionary biology (GB 8212, to N. G. HAIRSTON), a National Science Foundation Graduate Fellowship, and by the Miller Institute for Basic Research in Science at the University of California, Berkeley. Portions of this work were submitted in partial fulfillment of requirements for the degree of Doctor of Philosophy at the University of Michigan.

Literature cited

- ANDERSON, J. D. (1970): Description of the spermatophore of *Ambystoma tigrinum*. *Herpetologica* 26 (3), 304—308 • ARNOLD, S. J. (1972): The evolution of courtship behavior in salamanders. Ph. D. dissertation, University of Michigan, 570 p. • ASSEM, J. VAN DEN (1967): Territory in the three-spined stickleback, *Gasterosteus aculeatus*. *Behaviour Suppl.* 16, 1—164 • BALDAUF, R. J. (1952): Climatic factors influencing the breeding migration of the spotted salamander, *Ambystoma maculatum*. *Copeia* 1957 (3), 178—181 • BARLOW, G. W. (1967): Social behavior of a South American leaf fish, *Polycentrus schomburgkii*, with an account of recurring pseudofemale behavior. *Amer. Midl. Nat.* 78 (1), 215—234 • BEDRIAGA, J. (1882): Über die Begattung bei einigen geschwänzten Amphibien. *Zool. Anz.* 5, 265—268 • BEDRIAGA, J. (1883): Beiträge zur Kenntniss der Amphibien und Reptilien der Fauna von Korsika. *Arch. f. Naturgesch.* 49, 201—273 • BEDRIAGA, J. (1895): On the Pyrenean newt *Molge aspera*, Dugès. *Proc. Zool. Soc. Lond.* 1895, 150—154 • BEDRIAGA, J. (1897): Die Lurchfauna Europas. II. Urodela, Schwanzlurche. *Bull. Soc. Imp. Nat. Moscou* 10, 699—705 • BIAGGI, V. (1955): The Puerto Rican Honeycreeper, *Coereba flaveola portoricensis* (Bryant). Univ. Puerto Rico Agricultural Station, Special Bulletin • BLANCHARD, F. N. (1930): The stimulus to the breeding migration of the spotted salamander, *Ambystoma maculatum* (Shaw). *Amer. Nat.* 64, 154—167 • BRANDON, R. A. (1970): Courtship, spermatophores and eggs of the Mexican Achoque, *Ambystoma (Bathsiredon) dumerili* (Dugès). *Zool. J. Linn. Soc.* 49, 247—254 • BREDER, R. C. (1927): Courtship of the spotted salamander. *Bull. N. Y. Zool. Soc.* 30, 3 • CALEF, R. T. (1954): The salamander *Ambystoma tigrinum nebulosum* in Southern Arizona. *Copeia* 1954 (3), 223 • COHEN, A. C., Jr. (1960): Estimating the parameter in a conditional Poisson distribution. *Biometrics* 16, 203—211 • CREWS, D. (1975): Psychobiology of reptilian reproduction. *Science* 189, 1059—1065 • DÄHNE, C. (1926): Die Copula bei *Euproctus asper* Dug. *Blätter Aquar.-Terr. Kde.* 37, 198—201 • DARWIN, C. (1859): On the origin of species by means of natural selection or the preservation of favoured races in the struggle for life. John Murray, London, 490 p. • DARWIN, C. (1874): The descent of man and selection in relation to sex. A. L. Burt Co., New York, 797 p. • DESPAX, R. (1923): Contribution à l'étude anatomique et biologique des batraciens urodèles du groupe des *Euproctus*, et spécialement de l'euprocte des Pyrénées. Thèse Facult. Sci. Paris, 258 p. • FISHER, R. A. (1958): The genetical theory of natural selection. Dover Publication, New York, 291 p. • GADGIL, M., and W. H. BOSSERT (1970): Life historical consequences of natural selection. *Amer. Nat.* 104 (935), 1—24 • GARTON, J. S. (1972): Courtship of the small-mouthed salamander *Ambystoma texanum*, in southern Illinois. *Herpetologica* 28, 41—45 • GASCO, F. (1881): Les amours des axolotls. *Bull. Soc. Zool. France* 6, 151—164 • HALLIDAY, T. R. (1974): Sexual behavior of the smooth newt, *Triturus vulgaris* (Urodela, Salamandridae). *J. Herpetology* 8 (4), 277—292 • HAMILTON, W. D. (1970): Selfish and spiteful behavior in an evolutionary model. *Nature* 228 (5277), 1218—1220 • HAMILTON, W. D. (1971): Selection of selfish and altruistic behavior in some

- extreme models; in Man and beast: comparative social behavior (ed. J. F. EISENBERG and W. S. DILLON), pp. 59—91. Smithsonian Annual III, Smith. Inst. Press, Washington • HIGHTON, R. (1962a): Geographic variation in the life history of the slimy salamander. *Copeia* 1962 (3), 597—613 • HIGHTON, R. (1962b): Revision of North American salamanders of the genus *Plethodon*. *Bull. Florida State Museum* 6 (3), 235—367 • HIGHTON, R. (1970): Evolutionary interactions between species of North American salamanders of the genus *Plethodon*. Part 1. Genetic and ecological relationships of *Plethodon jordani* and *P. glutinosus* in the southern Appalachian Mountains. *Evolutionary Biology*, Appleton-Century-Crofts, New York 4, 211—241 • HRDY, S. B. (1974): Male-male competition and infanticide among the langurs (*Presbytis entellus*) of Abu, Rajasthan. *Folia Primatologica* 22, 19—58 • HUNSAKER, D. (1962): Ethological isolating mechanisms in the *Sceloporus torquatus* group of lizards. *Evolution* 16, 62—74 • HUSTING, E. L. (1965): Survival and breeding structure in a population of *Ambystoma maculatum*. *Copeia* 1965 (3), 352—362 • HUTCHISON, V. C. (1959): Aggressive behavior in *Plethodon jordani*. *Copeia* 1959 (1), 72—73 • KEENLEYSIDE, M. H. A. (1972): Intraspecific intrusions into nests of spawning Longear Sunfish (Pisces: Centrarchidae). *Copeia* 1972 (2), 272—278 • KUMPF, K. F. (1934): The courtship of *Ambystoma tigrinum*. *Copeia* 1934 (1), 7—10 • LACK, D. (1954): The evolution of reproductive rates. In: *Evolution as a process*, pp. 172—187 (J. HUXLEY, A. C. HARDY, and E. B. FORD, ed.). Collier Books, New York, N. Y. • MACMAHON, J. A. (1964): Additional observations on the courtship of Metcalf's salamander, *Plethodon jordani* (metcalfi phase). *Herpetologica* 20 (1), 67—69 • MADISON, D. M. (1969): Homing behavior of the red-cheeked salamander *Plethodon jordani*. *Animal Behavior* 17, 25—39 • MADISON, D. M., and C. R. SHOOP (1970): Homing behavior, orientation, and home range of salamanders tagged with tantalum-182. *Science* 168, 1484—1487 • MARSHALL, A. J. (1954): Bowerbirds. Oxford Univ. Press, London • MERCHANT, H. (1972): Estimated population size and home range of the salamanders *Plethodon jordani* and *Plethodon glutinosus*. *J. Wash. Acad. Sci.* 62 (3), 248—257 • MORRIS, D. (1958): The reproductive behavior of the ten-spined stickleback (*Pygosteus pungitius* L.). *Behaviour Suppl.* 6, 1—154 • MURPHY, R. C. (1936): Oceanic birds of South America. Macmillan, New York • NOBLE, G. K. (1931): The biology of the amphibia. McGraw-Hill, New York • NOBLE, G. K., and M. K. BRADY (1930): The courtship of the plethodontid salamanders. *Copeia* 1930 (2), 52—54 • NOBLE, G. K., and M. K. BRADY (1933): Observations on the life history of the marbled salamander, *Ambystoma opacum* Grabenhorst. *Zoological*, New York 11 (8), 89—132 • ORGAN, J. A. (1958): Courtship and spermatophore of *Plethodon jordani* metcalfi. *Copeia* 1958 (4), 251—259 • ORGAN, J. A. (1960a): The courtship and spermatophore of the salamander *Plethodon glutinosus*. *Copeia* 1960 (1), 34—40 • ORGAN, J. A. (1960b): Studies on the life history of the salamander *Plethodon welleri*. *Copeia* 1960 (4), 287—297 • ORGAN, J. A. (1961): Studies on the local distribution, life history, and population dynamics of the salamander genus *Desmognathus* in Virginia. *Ecol. Monog.* 31, 189—200 • ORGAN, J. A., and L. A. LOWENTHAL (1963): Comparative studies of macroscopic and microscopic features of spermatophores of some plethodontid salamanders. *Copeia* 1963 (4), 659—669 • PARKER, G. A. (1970): Sperm competition and its evolutionary consequences in the insects. *Biological Reviews* 45, 525—568 • PRECHTL, H. F. R. (1951): Zur Paarungsbiologie einiger Molcharten. *Z. Tierpsychol.* 8, 337—348 • RAND, A. S. (1967): Ecology and social organization in the iguanid lizard, *Anolis lineatopus*. *Proc. U. S. Nat. Museum* 122 (3595), 1—79 • RAND, A. S. (1968): A nesting aggregation of iguanas. *Copeia* 1968, 552—561 • ROGOFF, J. L. (1927): The hedonic glands of *Triturus viridescens*; a structural and morphological study. *Anat. Rec.* 34, 132—133 • SALTHER, S. N., and B. M. SALTHER (1964): Induced courtship in the salamander *Pseudoeurycea belli*. *Copeia* 1964 (3), 574—576 • SCHALLER, F. (1971): Indirect sperm transfer by soil arthropods. *Ann. Rev. Entomol.* 16, 407—446 • SMITH, B. G. (1910): The structure of the spermatophores of *Ambystoma punctatum*. *Biol. Bull.* 18 (4), 204—211 • SPOTILA, J. R., and R. J. BEUMER (1970): The breeding habits of the ringed salamander, *Ambystoma annulatum* (Cope), in northeastern Arkansas. *Amer. Midl. Nat.* 84, 77—89 • STEBBINS, R. C. (1949): Courtship of the plethodontid salamander *Ensatina eschscholtzii*. *Copeia* 1949 (4), 247—281 • STEBBINS, R. C. (1962): Amphibians of western North America. Univ. of California Press, Berkeley, 539 p. • STOREZ, R. A. (1969): Observations on the courtship of *Ambystoma laterale*. *J. Herpetology* 3 (1—2), 87—95 • SUGIYAMA, Y. (1967): Social organization of Hanuman langurs. In: *Social communication among primates* (S. ALTMANN, ed.). University of Chicago Press, Chicago • TINKLE, D. W. (1969): The concept of reproductive effort and its relation to the evolution of life histories of lizards. *Amer. Nat.* 103 (933), 501—516 • TRIVERS, R. L. (1972): Parental investment and sexual selection. In: *Sexual selection and the descent of man 1871—1971* (B. CAMPBELL, ed.). Aldine, Chicago • TWITTY, V. C. (1955): Field experiments on the biology and genetic relationships of the Californian species of *Triturus*. *J. Exp. Zool.* 129 (1), 129—148 • UZZELL, T. M. (1964): Relations of the diploid and triploid species of the *Ambystoma jeffersonianum* complex (Amphibia, Caudata). *Copeia* 1964 (2), 257—300 • UZZELL, T. M. (1969): Notes on spermatophore production by salamanders of the *Ambystoma jeffersonianum* complex (Amphibia, Caudata). *Copeia* 1969 (3), 602—612 • WELLER, M. W.

(1959): Parasitic egg laying in the redhead (*Aythya americana*) and other North American Anatidae. Ecol. Monographs 29, 333—365 • WEYGOLDT, P. (1966): Spermatophore web formation in a pseudoscorpion. Science 153 (3477), 1647—1649 • WHITFORD, W. G., and A. VINEGAR (1966): Homing, survivorship, and overwintering of larvae in spotted salamanders, *Ambystoma maculatum*. Copeia 1966 (3), 515—519 • WILBUR, H. M. (1972): Competition, predation and the structure of the *Ambystoma-Rana sylvatica* community. Ecology 53 (1), 3—21 • WILBUR, H. M., and J. P. COLLINS (1973): Ecological aspects of amphibian metamorphosis. Science 182, 1305—1314 • WILLIAMS, G. C. (1966): Adaptation and natural selection. Princeton, Univ. Press, Princeton, N. J. • WILSON, E. O., and W. H. BOSSERT (1963): Chemical communication among animals. Recent Prog. in Hormone Res. 19, 673—716 • WRIGHT, A. H., and A. A. ALLEN (1909): Early breeding habits of *Amblystoma punctatum*. Amer. Nat. 43, 687—692 • ZELLER, E. (1905): Untersuchungen über die Samenträger und den Kloakenwulst der Tritonen. Z. f. wiss. Zool. 79, 171—221.

Author's address: S. J. ARNOLD, Department of Biology, University of Chicago, Chicago, Illinois, U.S.A., 60637.