A Double Role of Sperm in Scorpions: The Mating Plug of *Euscorpius italicus* (Scorpiones: Euscorpiidae) Consists of Sperm

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ABSTRACT Mating plugs occluding the female gonopore after mating are a widespread phenomenon. In scorpions, two main types of mating plugs are found: sclerotized mating plugs being parts of the spermatophore that break off during mating, and gel-like mating plugs being gelatinous fluids that harden in the female genital tract. In this study, the gel-like mating plug of *Euscorpius italicus* was investigated with respect to its composition, fine structure, and changes over time. Sperm forms the major component of the mating plug, a phenomenon previously unknown in arachnids. Three parts of the mating plug can be distinguished. The part facing the outside of the female (outer part) contains sperm packages containing inactive spermatozoa. In this state, sperm is transferred. In the median part, the sperm packages get uncoiled to single spermatozoa. In the inner part, free sperm is embedded in a large amount of secretions. Fresh mating plugs are soft gelatinous, later they harden from outside toward inside. This process is completed after 3-5 days. Sperm from artificially triggered spermatophores could be activated by immersion in insect Ringer’s solution indicating that the fluid condition in the females’ genital tract or females’ secretions causes sperm activation. Because of the male origin of the mating plug, it has likely evolved under sperm competition or sexual conflict. As females refused to remate irrespective of the presence or absence of a mating plug, females may have changed their mating behavior in the course of evolution from polyandry to monandry. J. Morphol. 271:383–393, 2010.

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INTRODUCTION Substances that block the female genital opening after mating, so-called mating plugs, genital plugs, or spermatocleutra, are a widespread phenomenon in the animal kingdom and have evolved many times independently (Shine et al., 2000). They are found in some species of scorpions (Polis and Sissom, 1990), in vertebrates and many invertebrates (Colonello and Hartfelder, 2005; Oh and Hankin, 2004; Ramm et al., 2005) including other arachnids (Alberti and Coons, 1999; Fromhage and Schneider, 2006; Masumoto, 1993; Suhm et al., 1995/96). Generally, they are formed by coagulation of ejaculate and/or other male substances or even female substances (Aisenberg and Eberhard, in press; Eberhard, 1996; Wigby and Chapman, 2004). There is an enormous variation in formation, efficiency, and relative size of the mating plugs. For example, in an orb-weaving spider, the whole male body serves as a short-time mating plug after the male’s death during copulation (Foellmer and Fairbairn, 2003) or in some *Drosophila* species, the females occlude the genital opening themselves with their own secretions (Eberhard, 1996). Concerning the function of mating plugs, they have mainly been interpreted as a mechanism of males to prevent direct sperm competition (Birkhead and Møller, 1998; Ramm et al., 2005; Simmons and Siva-Jothy, 1998). The sexual conflict hypothesis (Chapman et al., 2003) proposes the plugs to be a result of the male–female conflict over the control of fertilization. Furthermore, the plug could reduce sperm desiccation (Woyciechowski et al., 1994), ensure safe sperm transfer (Polak et al., 1998), or prevent pathogens from entering the females’ genital tract (Contreras-Garduño et al., 2006).

A mating plug in scorpions was first described by Pavlovsky (1925). Currently, mating plugs are...
known for the following scorpion families: Bothriuridae (Castelvetri and Peretti, 1999; Peretti, 2003), Buthidae (Probst, 1972; Shulov and Amitai, 1958), Chaetidae (Stockwell, 1989), Euscorpiidae (Angermann, 1957; Jacob et al., 2004), Urodacidae (Prendini, 2000; Smith, 1966; Stockwell, 1989), and Vaejovidae (Contreras-Garduño et al., 2006; Hendrixson, 2001; Sissom, 1993; Stockwell, 1989). The mating plugs described so far can be roughly separated into two types according to their composition and origin (Contreras-Garduño et al., 2006; Stockwell, 1989). One type, described for Vaejovidae, Urodacidae, and Bothriuridae, is the sclerotized mating plug (Contreras-Garduño et al., 2006; Mattoni and Peretti, 2004; Peretti, 2003) consisting of parts of the spermatophore that break off at predefined points during mating.

The second type of mating plugs is the gel-like mating plug (Contreras-Garduño et al., 2006). It is more widespread in scorpions (Mattoni and Peretti, 2004) and consists of different gelatinous substances. They harden in the female genital opening, in this way forming a plug (Stockwell, 1989). They are much less investigated than the sclerotized mating plugs and their origin and formation is only known in very few cases (Polis and Sissom, 1990; Shulov and Amitai, 1958; Smith, 1966).

The function of mating plugs in scorpions remains largely unknown. Solely, large sclerotized mating plugs seem to prevent sperm competition (Contreras-Garduño et al., 2006).

To be able developing hypotheses about the possible function and evolution of a mating plug, it is necessary to find out more about its composition, formation, effectiveness, and for how long it remains in the females’ genital tract. In this study, a gel-like mating plug and its fine structure are investigated in detail. The species studied is Euscorpius italicus. In this article, the changes of the mating plug over time are observed and underlying mechanisms are proposed and tested.

MATERIAL AND METHODS

Collecting Mating Plugs

Living E. italicus (Herbst, 1800) were collected by hand in Tessin (Ticino) county in the south of Switzerland before the breeding season from late July to beginning of August 2006 and in June 2007. The scorpions were kept separately in plastic boxes (10.5 × 8.5 cm) on gypsum ground. They were offered a piece of bark for shelter, water ad libitum, and were fed every week with one house cricket (Gryllodes supplicans F. Walker, 1859) each. Females were allowed to give birth to their young in the laboratory. After the offspring had left the mother, females were used for the matings.

To collect mating plugs, females were allowed to mate in the laboratory. A vertical wall of coarse concrete 40 × 40 cm, surrounded by wood, cling film, and glass to prevent scorpions from escaping, was used as a mating arena. All matings were filmed (Sony digital handycam DCR-TRV900E). In total, 20 complete matings were observed. At different times after mating, mating plugs were removed from the females: after 0.5 h (n = 1), 1 h (n = 1), 3 h (n = 2), 6 h (n = 3), 12 h (n = 1), 1 d (n = 1), 3 d (n = 3), 5 d (n = 2), 7 d (n = 3), 14 d (n = 3). For the removal, females were anesthetized by exposing them to diethyl ether for 8 min and then fixed in a small plastic tube to facilitate handling. The mating plugs were removed under the dissecting microscope with a pair of tweezers. For fixation, mating plugs were immersed in a solution of 2.5% glutaraldehyde in 0.1mol l⁻¹ 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid (HEPES). Pictures of the complete mating plugs were taken with a JVC digital camera KY-F70B. The mating plugs were cut in half with a razor blade. One half was used for the light microscopic investigations, the other one was left for electron microscopic investigations.

Light Microscopy

Seventeen mating plug halves were washed in Na-cacodylate buffer (0.1 mol l⁻¹, pH 7.4) for 1 h, dehydrated in graded ethanol series, and embedded in LR White Resin (Soft Grade Acrylic, London Resin, UK). They were sectioned in semithin series (1-2 µm) with a microtome (Leica RM 2145) using glass knives. The sections were stained with toluidine blue (1%) in an aqueous borax solution (1%) for 30 sec. Light microscopic investigations were performed with a Zeiss Axioplan 2. Pictures were taken with a Zeiss Axiocam MRc.

Transmission Electron Microscopy

Five mating plugs aged 0.5 h, 3 h, 6 h, 12 h, and 3d were studied. They were washed in Na-cacodylate buffer (0.1 mol l⁻¹, pH 7.4) for 15 min, post-fixed in 1% osmium tetroxide in Na-cacodylate buffer (0.1 mol l⁻¹, pH 7.4) for 2 h and washed again in Na-cacodylate buffer overnight. The samples were dehydrated in graded ethanol series followed by propylene oxide and finally embedded in Epon. Ultrathin sections (70 nm) were cut with a diamond knife (Diatome SA, Bienne, Switzerland) on an ultramicrotome (Reichert Ultracut E). The sections were stained with uranyl acetate and lead citrate. Examination was performed with a Philips CM 12 transmission electron microscope.

Analysis of Mating Plug Formation

To investigate the mating plug formation, eight matings were interrupted after spermatophore deposition and the spermatophores were used for different tests.
One preinsemination spermatophore was serially semithin sectioned to investigate the sperm state in the spermatophore before transfer to the female.

Seven spermatophores were artificially triggered by bending down the lamina with a pair of tweezers and thus sperm was ejected. The sperm gained that way was analyzed under three different conditions: 1) no addition of fluid to the sperm, just air contact to observe the desiccation process \((n = 2)\). 2) Addition of insect Ringer’s saline solution \((154 \text{ mmol l}^{-1} \text{ NaCl}, 2.68 \text{ mmol l}^{-1} \text{ KCl}, 2.25 \text{ mmol l}^{-1} \text{ CaCl}_2, \text{pH 6.4})\) \((n = 3)\) to imitate the fluid conditions inside the female’s genital atrium. 3) Addition of human Ringer’s saline solution \((\text{MERCK-Ringer tablets, Merck KGaA, 64293 Darmstadt, Germany, pH 7.4})\) to control for the specificity of the sperm’s reaction to the female genital condition \((n = 2)\).

Remating Trials
Ten females were offered the opportunity to remate one (seven females) or two times with different males (three females). Three of them had a mating plug aged 3 d; the other females were previously anesthetized and their mating plugs removed. For details see Table 1. It was recorded whether the female rejected the male actively (R), accepted a new mating, or whether there was no reaction from both sides (nr).

RESULTS
Presence and Visibility of the Mating Plug
Seventeen of 20 mated females had a mating plug (85%). In all mating plugs, sperm was found. In all but one sample (7 d; 94%), the mating plugs consisted mainly of sperm. Three mating plugs aged 12 h, 3 d, and 14 d were smaller and shorter than the others (18%). In one case, it was too small to fill the female gonopore. Those mating plugs also lacked or contained only very few sperm packages, but, nevertheless contained free spermatozoa. If a mating plug was present, it was visible from the outside of the female gonopore in 88% of the cases \((n = 15; \text{Fig. 1})\). The visibility from outside does not seem to be related to age or size of the mating plug. The two plugs that could not be seen from outside were a 6 h and a 3 d mating plug of median and large size. With the exception of the 7 d mating plug that did not mainly consist of sperm, the mating plugs were not anchored firmly in the female and could easily be removed.

Composition and Form of the Mating Plug
The following description is based on the 13 mating plugs that were not aberrant in size or sperm content. Aged, mating plugs were about 1 mm in maximum length and width; in a lateral view, they appear conical in shape (Figs. 1B and 2). As the substance for the mating plug is transmitted in a soft-gelatinous form, the mating plug fits perfectly into the female gonopore. The broader end of the mating plug faces toward the outside of the female (Fig. 2A). The outer part of the mating plug has a granular appearance and the inner part is smoother (Fig. 2).

A mating plug consists of sperm packages, free, uncoiled spermatozoa, and secretion. The sperm packages are found in the outer part of the mating plug (Fig. 2A–C). They appear increasingly dense packed toward the inside of the female (Figs. 2B,C and 3A,B). One package contains roughly 500-900...
spermatozoa. The sperm heads lie in parallel, are highly regularly arranged, and densely packed (Figs. 4A,D). The flagella are twisted in a screw-like manner and the bundle of flagella lies in parallel to or next to the sperm heads. The flagella as well are densely and regularly packed (Fig. 4C). The spermatozoa in the packages are embedded in a secretion that also encloses the sperm packages as a whole with a thin layer. The sperm packages are surrounded by another secretion (Fig. 4A,B).

The outer part is followed by the median part, in which sperm packages become looser and start to uncoil to single, free spermatozoa (Figs. 2A–C and 3C,D). In the inner part of the mating plug, only free spermatozoa in varying densities are found (Fig. 3E,F). The spermatozoa have a relatively long and slim head that is not broader than the middle piece (Fig. 5B). The globular structures (Fig. 5D) are probably by-products of sperm differentiation (M. Stoffel, unpublished data).

Secretion is found filling the space between the sperm packages especially in the outermost part of the mating plug (Fig. 3A). The free sperm is embedded in secretions of different densities as well (Fig. 5). Furthermore, fibrillose (Fig. 5B) components are visible in the secretions of the mating plugs aged 3 h and 3d.

Changes Over Time

Mating plugs are retained for at least 14 d after mating. The mating plugs aged 0.5 h till 6 h are of soft consistence. Especially, the inner part is almost gel like to fluid during the first 3 h (Fig. 6A). Therefore, mating plugs aged 0.5–1 h were deformed by removal. From 3 h, a hardening processing from the outside was observable that continues toward the inner part of the mating plug. After 3 to 5 days, the hardening process was completed (Fig. 6B). During hardening, the appearance of the mating plug changes. When the mating plug is still soft, the inner part is whitish and opaque. As the hardening is complete, it appears vitreous. While hardening, the mating plug has a more or less smooth surface and no sharp edges can be seen. When hardening is completed, the edges are sharp and the surface of the mating plug is no longer even, especially in the inner part of the mating plug. The inner end of the mating plug, while first being rounded (Figs. 2A and 6A) becomes jagged during hardening (Fig. 6B–D). The inner part consisting of free sperm and secretion becomes shorter even after it has hardened from 5 d till 14 d (Fig. 6B–D). The amount of sperm packages (i.e., the outer and median parts of the mating plug) decreases after 3 h, but a certain amount of sperm packages can still be seen in hardened mating plugs (lines indicated on Fig. 6).

Preinsemination vs. Postinsemination Spermatophore

In the preinsemination spermatophore, the sperm is located in the sperm reservoir beneath the capsular region of the spermatophore (Fig. 7A). The sperm is exclusively found in pack-
ages (Fig. 7B), no uncoiled sperm is detectable. Between the loosely packed sperm packages, secretion can be seen. Separated from the sperm packages, a gelatinous fluid fills the lower spermatophore cavity, which is expelled after the sperm when the spermatophore is triggered. (Fig. 8)

Tests on the Mating Plug Formation Process

In Treatment 1 (air contact), the contents of the artificially triggered spermatophores hardened almost completely within an hour ($n = 2$; Fig. 9A). The sperm packages do not change. They are loosely packed and secretion is found between the packages (Fig. 9B).

When sperm and fluid are transferred into insect Ringer's saline solution (Treatment 2), the sperm packages start to uncoil partially and sperm can be seen moving actively in the solution ($n = 3$; Fig. 9C). The spermatozoa were moving for up to 6 h.

When sperm and fluid are transferred into human Ringer's saline solution (Treatment 3), the sperm packages do not change either ($n = 2$; Fig. 9D). Neither the fluid nor the sperm becomes hard but they stay soft and whitish in color.

Remating Trials

None of the 10 females remated (Tab.1). All but one showed clear rejection behavior toward the
male as soon as they attempted to grasp the females’ pedipalps. In one case, no contact between male and female occurred.

**DISCUSSION**

**Sperm as the Main Component of the Mating Plug in *E. italicus***

Mating plugs originating from products that are transmitted by the male with the ejaculate are a frequent mode of mating plug formation (Kingan et al., 2003; Moreira and Birkhead, 2003; Polak et al., 1998). However, it has rarely been reported that the mating plug mainly consists of sperm. In primates, for example, it is known that a protein mediates coagulation of the ejaculate and so a plug is formed (Kingan et al., 2003). Nevertheless, this mating plug is not as permanent and as hard as the one of *E. italicus*. In arachnids, a mating plug consisting of sperm was hitherto unknown. For

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**TABLE I. Remating trials with the details for the 10 females used**

<table>
<thead>
<tr>
<th>Female No.</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
<th>10</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plug removed (h)</td>
<td>0.5</td>
<td>1</td>
<td>3</td>
<td>10</td>
<td>15</td>
<td>24</td>
<td>72</td>
<td>p</td>
<td>p</td>
<td>p</td>
</tr>
<tr>
<td>Male offered (d)</td>
<td>3</td>
<td>1</td>
<td>5</td>
<td>1</td>
<td>3</td>
<td>3</td>
<td>4</td>
<td>3</td>
<td>3</td>
<td>3</td>
</tr>
<tr>
<td>Reaction of female</td>
<td>R</td>
<td>nr</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>R</td>
<td>R</td>
</tr>
</tbody>
</table>

Hours the mating plug was removed after mating; time after mating in days when one or two further males were offered to the female; reaction of the female.

nr, no reaction; p, mating plug not removed; R, rejection of the male by the female.

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![Fig. 4. Transmission electron micrographs of different cross-sections of sperm packages in the outer part of the mating plug. (A) Sperm package. (B) Two adjoining sperm packages. (C) Middle pieces of spermatids in a sperm package. (D) Sperm heads in sperm package. fl, flagellum(s); mpc, middle piece(s); mt, mitochondria; sec 1/2, two different secretions; sh, sperm head(s). Scale bars = 10 µm for A; = 5 µm for B; = 1 µm for C, D.](image-url)
example, in the Araneae, the mating plugs are mostly products of a specialized male palpal gland or parts of the palp that break off during mating (Fromhage and Schneider, 2006; Masumoto, 1993; Nessler et al., 2007). In the Acari, the spermatophore itself, substances transmitted through the spermatophore, or parts of the spermadactylus that break off at mating may function as mating plugs (Alberti and Coons, 1999; Alberti et al., 2000). We found for the first time that spermatozoa in scorpions can have a dual function, meaning that they are not only responsible for fertilization but also in directly blocking the genital opening. Dual functions of spermatozoa are also known from other invertebrates (Swallow and Wilkinson, 2002). Those dual functions and the enhanced production of sperm are believed to have evolved under sperm competition (Birkhead and Möller, 1998). Therefore, the mating plug might have evolved under the pressure of sperm competition.

The mating plug of *E. italicus* is mostly of male origin. The secretion found between the sperm packages in the outer part of the mating plug is most probably also of male origin and should partly be the fluid that follows the sperm package mass when the spermatophore is triggered; the dried sperm mass and secretion of the artificially triggered spermatophore (Fig. 9B) are identical to the outer part of the mating plug (Fig. 3A) when sectioned.

So, males block the genital opening with the sperm and secretion transferred and no other substances added by females seem to be necessary for the mating plug formation. This is in line with the basic sexual conflict theory: males try to block the genital opening of the female to prevent her from remating (Chapman et al., 2003; Parker, 1979). Indeed, females of *Euscorpius* sp. seem to be single mated as shown by our data and by Benton (1992). Single matings are not frequent in scorpions, most of them are multiple mated (Castelvetri and Peretti, 1999; Peretti and Carrera, 2005). In *E. italicus*, the mating plug is occluding the gonopore and is complete after one mating only. This is in contrast to the gel-like mating plugs found in some bothriurids, which increase in size with every remating (Mattoni and Peretti, 2004; Peretti, 2003).

Fig. 5. Transmission electron micrographs of inner parts of mating plugs. A, 0.5 h; B, 3 h; C, 3 d; D, 3 d. ep, sperm end piece; fs, fibrillar substance; gs, globular structure; mpc, sperm middle piece; sec, different secretions; sh, sperm head. Scale bars = 5 µm for A, C; = 2 µm for B, D.
Efficient mating plugs after one mating only are found among the sclerotized plugs in the Vaejovidae (Contreras-Garduño et al., 2006) and in the Bothriurus asper group (Mattoni and Peretti, 2004). Those mating plugs bear conspicuous hooks and distal barbs and get anchored strongly in the female genital atrium. The females of these species

Fig. 6. Photographs of different aged mating plugs, the line indicating the border between sperm packages (outer part and median part) and free sperm (inner part). A, 3 h; B, 5 d; C, 7 d; D, 14 d. ip, inner part with free sperm; mp, median part; op, outer part with sperm packages. Scale bars = 500 µm.

Fig. 7. Preinsemination spermatophore. (A) Longitudinal section of the upper spermatophore region. (B) Sperm packages in sperm reservoir. cap, capsular region; fla, flagella; sec, secretion; shs, sperm heads; sp, sperm packages; wa, wall of sperm reservoir. Scale bars = 500 µm for A; = 20 µm for B.

Bothriurus asper group (Mattoni and Peretti, 2004). Those mating plugs bear conspicuous hooks and distal barbs and get anchored strongly in the female genital atrium. The females of these species
are all single mated. (Contreras-Garduño et al., 2006; Mattoni and Peretti, 2004). Because of their efficiency in preventing further spermatozoa from entering the genital tract of the female, they are interpreted as a male strategy to avoid direct sperm competition (Contreras-Garduño et al., 2006). If mating plugs serve this strategy, females should be prevented from remating by the mating plug. No female mated a second time after one successful mating. Females did not remate even when the mating plug had been removed by us. In contrast, most females from a previous mating that had been interrupted after spermatophore deposition mated a second time (10 of 13; S.A., personal observations). These data are anecdotal only, but, they indicate that females’ behavior changes after sperm transfer.

If an effective mating plug prevents spermatozoa from a second male to enter the female reproductive tract, it could be advantageous for her to repel males before a second mating even starts. As a consequence, selective pressure would be strong on females not to remate after a first mating. Also, even though the mating plug is not anchored in the female, it adjusts perfectly to the shape of the genital opening. It seems improbable that subsequent males would be able to remove a mating plug because of physical limitations and females’ behavior. Possible other causes why females did not remate are: Removal of a mating plug is a purely artificial intervention that possibly changes female’s natural behavior. Female’s receptivity may have changed within the first hours after sperm transfer due to other cues as for example substances that are transmitted with the seminal fluids as has been suggested for a lycosid spider (Estramil and Costa, 2007).

Sperm Activation and Possible Female Influence

Sperm packages uncoil in the median part of the mating plug. This is probably due to the activation of the sperm only in contact with the female genital condition: The spermatozoa were activated only in insect Ringer’s and not in human Ringer’s solution. The possibility that females are able to activate spermatozoa in arachnids was suggested in several studies (Alberti et al., 1999; Burger et al., 2006; Eberhard and Huber, 1998; Peretti and Battán-Horenstein, 2003). The female genital atrium is equipped with glands, which could produce secretions that activate sperm (Peretti, 2003; Peretti and Battán-Horenstein, 2003). The free spermatozoa seen in the inner part of the mating plug and showing a very similar ultrastructure as in other scor-
pions (Alberti, 1983) seem to be embedded into several secretions of different densities. It cannot be ruled out that these different densities correspond to different stages of the hardening process of a single secretion. Some of these secretions might be of female origin: The fibrillosse substance present in the inner part of the mating plug is not found among the substances in the preinsemination spermatophore; thus, it should not be of male origin. In addition, such fibrillosse substances are known to be products of female glands in the genital tract (R. Stockman, unpublished data).

The part of the ejaculate that is not activated and part of the free spermatozoa are subject to a hardening (probably desiccation) process that starts from the outside and continues toward the inside. The hardening of the sperm mass in the female is slower (3-5 d) when compared with the rapid drying of the sperm mass outside the female, i.e., within an hour. This is probably achieved through the mentioned secretions of the female that keep the inner part of the mating plug moist. The outer part that hardens rather quickly forming a hard lid might further reduce loss of humidity. By this, the longevity of the spermatozoa could be prolonged and over a longer period of time sperm could still be activated. Thus, it could be a short-term storage mechanism. Although the presence of a simple spermatheca is reported for Euscorpius (Braunwalder, 2005; Volschenk et al., 2008), no long-term storage of sperm seems to occur (Benton, 1992; Jacob et al., 2004; Pavlovsky, 1925).

The inner part of the mating plug contains large numbers of uncoiled spermatozoa. This could be due to passive enclosure of dead spermatozoa or to female influence on sperm fate and/or hardening of the mating plug. Unfortunately, these problems remain largely unstudied; in addition, very little is known on the sperm longevity and storage in E. italicus and for how long after the mating eggs are fertilized.

The mating plug was still present after 2 wk and no obvious signs of degradation were seen. The inner part becomes slightly shorter from 7 d to 14 d, which could be the first degradation step. In some females that had mated more than a month before, black mating plugs were found by A.J. Similar observations were made in Vaejovis punctatus (Vaejovidae) (Contreras-Garduño et al., 2006) and two bothriurid species of the B. asper group (Mattoni and Peretti, 2004). In the V. punctatus, no changes in mating plug appearance were seen after 15 d, but after 4 mo, the mating plug had turned black in the course of degradation. So, mating plugs may be a safe way for males to prevent remating of a female for several weeks or even months.

Gel-like plugs were described for a few other species. An amorphous substance that envelopes the sperm in the spermatophore seems to form the mating plug in the buthid Leiurus quinquestriatus (Shulov and Amitai, 1958). In some bothriurid species, the gel-like plugs are formed by a granular substance that is produced by accessory glands of the male’s paraxial organs (Peretti, 2003). For Urodacus abruptus (Urodacidae), it was suggested the mating plug was secreted by the female herself (Smith, 1966). Because of the wide distribution of gel-like mating plugs among scorpion families, it seems plausible that they could have different origins.

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LITERATURE CITED

THE MATING PLUG OF E. italicus


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