

Brief Communication

**Use of the dart apparatus by the hermaphroditic land snail
Polymita muscarum (Lea, 1834)**

BERNARDO REYES-TUR^{1,*}, JORIS M. KOENE²

¹ *Departamento de Biología, Facultad de Ciencias Naturales, Universidad de Oriente, Ave. Patricio Lumumba, Santiago de Cuba 90500, Cuba*

² *Institute of Ecological Science, Faculty of Earth and Life Sciences, Vrije Universiteit, Amsterdam, The Netherlands*

Abstract—Many species of pulmonate land snails are equipped with one or more so-called “love darts”. Even though the number and shape of these calcareous darts vary considerably between species, dart use has only been investigated in very few species. Here, we redescribe the mating behaviour of *Polymita muscarum* because previous reports did not include the use of the dart apparatus. Mating in this hermaphroditic land snail can be divided into three stages: courtship, copulation and post-copulatory activity. During courtship, full eversion of the genital atrium is reached, thus exposing the sensitive zone, genital lobes and dart apparatus. We observed that *P. muscarum* pushes the everted dart apparatus repeatedly onto different parts of the partner’s body and does not lose its dart after stabbing. Dissected specimens had a single, slender dart with a round base, a broad corona and a circular cross-section. We propose that the morphology of *P. muscarum*’s dart is consistent with the idea of simple darts needing to be stabbed more often in order to increase the transfer of mucus, which contains a biologically active substance (i.e. allohormone) that enhances the chances of paternity. Besides adding to the growing diversity in the use of love darts in land snails, these findings contribute to the understanding of the evolution of this peculiar reproductive act.

Keywords: Gastropod; hermaphrodite; love dart; sexual selection; stylommatophora.

INTRODUCTION

Pulmonate land snails are simultaneous hermaphrodites, and many species are equipped with one or more so-called “love darts” in their reproductive tract (Koene and Schulenburg, 2005; Schilthuizen, 2005). Despite the diversity in number and shape of these calcareous darts, their use has only been investigated in very few species. The best-studied species, the garden snail *Cantareus aspersus* (Müller,

*Corresponding author, e-mail: breyes@cnt.uo.edu.cu and/or bernardo635547@yahoo.es

1774), uses one forceful stab to pierce the dart through the skin of its partner during courtship (Adamo and Chase, 1988). Successful shooting results in a transfer of mucus from the associated glands into the partner's blood (Adamo and Chase, 1990) and increases fertilisation success of the sperm that are transferred after shooting (Rogers and Chase, 2002; Chase and Blanchard, 2006). Although the helicid *C. aspersus* loses its dart in the process, some xanthonychid (syn. helminthoglyptid) and bradybaenid species seem to stab their partner repeatedly with the same dart (Webb, 1942, 1952; Koene, 2006; Koene and Chiba, 2006). Detailed information about such behavioural differences is essential to understand the evolution of this peculiar reproductive act.

Tree snails of the genus *Polymita* Beck, 1837 are endemic to Cuba. The species we investigated, *Polymita muscarum* (Lea, 1834), has a distribution along the north coast of eastern Cuba from Sagua de Tánamo to Nuevitas (Torre, 1950). This hermaphrodite exchanges sperm simultaneously reciprocally in a mating that consists of a courtship, copulation and post-copulatory activity phase (Reyes-Tur and Fernández, 1998). Moreno (1950) described the love dart of *P. muscarum* as a calcareous stiletto with a very sharp, closed tip. Interestingly, a previous description of the mating behaviour of *P. muscarum* made no mention of the dart apparatus (Bidart et al., 1998), most likely because this organ was mistaken for the penis. The aim of the present communication is, therefore, to redescribe the mating behaviour for this species by including the use of the dart apparatus.

MATERIALS AND METHODS

We studied the reproductive biology of *P. muscarum* at the Malacology Laboratory of the Universidad de Oriente (Cuba). Adult specimens with a reflected lip at the shell aperture were collected from Holguín, Cuba. Test snails were individually housed in small plastic containers (7 × 7 × 12 cm) at least 3 days before mating trials. They were fed a fungi complex that lives on leaves of several plant species (Reyes-Tur et al., 2000). The temperature was maintained between 22 and 28°C with a light : dark cycle of 12 h : 12 h. At the beginning of the mating trials, eight to ten snails were placed together in an experimental plastic container (50 × 25 × 20 cm). The courting pairs were carefully separated from the group for detailed observation of the mating behaviour.

Four specimens were dissected after mating to remove the darts and check whether spermatophores were exchanged. To avoid damaging the dart, each dart apparatus was carefully cut out of the reproductive tract and placed for 24 h in 2N NaOH, which dissolved all the tissue and mucus but left the dart intact. The darts were subsequently placed under a stereo microscope (Stemi 2000-C, Zeiss) and digital photographs were taken.

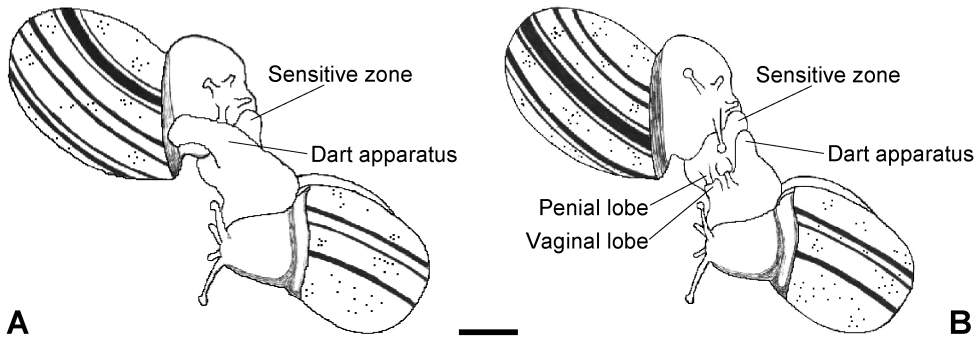


Figure 1. Line drawings of a mating pair of *Polymita muscarum* showing two different positions of the dart apparatus. A: The dart apparatus of one member of the pair is pressed against the edge of the mantle; B: The vaginal and penial lobes of both specimens are now visible and the dart apparatus of the same individual is now contacting the sensitive zone of its partner. (Scale bar = 4 mm.)

RESULTS AND DISCUSSION

In agreement with Bidart et al. (1998), we find that courtship includes reciprocal contacts and circling movements, very similar to circling in *C. aspersus* (Adamo and Chase, 1988), and lasts 28 ± 8 min (mean \pm standard deviation; $N = 21$ pairs). Copulation has an average duration of 85 ± 23 min ($N = 25$ pairs), during which spermatophores are exchanged ($N = 2$ pairs). The copulatory phase is characterised by the inhibition of locomotion and the partial withdrawal of the tentacles. Post-copulatory activity (37 ± 20 min, $N = 21$ pairs) begins when each animal has withdrawn its penis. During this phase both snails frequently exhibit circling movements and extend their tentacles. At the end of the post-copulatory phase the animals separate. The complete mating behaviour lasts 142 ± 29 min ($N = 25$ pairs; see also Reyes-Tur et al., 2000).

In *P. muscarum*, the full eversion of the genital atrium includes the exposure of – in anterior-posterior order – the sensitive zone, the vaginal lobe, the penial lobe and the dart apparatus (fig. 1A, B). Moreno (1950) did not describe the sensitive zone. However, he did report that, in retracted state, one can observe several overlapping leaflet-like structures on the atrium. These structures are abundant in front of the vagina. The position of the sensitive zone during copulation is consistent with the placement of these leaflet-like structures, suggesting that the latter form the sensitive zone when everted.

During mating, the dart apparatus is pressed repeatedly against different parts of the mating partner's skin (e.g., foot, lip, tentacles, mantle edge, penial and vaginal lobes; fig. 1A). The apparatus can then be partially or maximally extended. Such events last between 2 s and 4 min at a frequency of 1-10 times per 5 min. The higher frequencies are observed during copulation. Although both snails can use their dart on their partner simultaneously, they usually seem to do so in alternation. Interestingly, the volume of the sensitive zone increases considerably when contacted by the dart apparatus. Moreover, the action of the apparatus on this



Figure 2. Light microscopy photographs of two darts of *Polymita muscarum* in side view. Cross-sections of the dart are circular, i.e., without blades. (Scale bar = 100 μm .)

zone is perhaps best described as ‘rubbing’. During the rubbing motion the dart apparatus is partially extended (fig. 1B) and seems to make small (semi-)circular movements.

The dart itself is normally obscured from view, but can be clearly seen when the completely everted dart apparatus comes off the skin. As a consequence, we assume that the dart pierces the partner when the dart apparatus is pressed against the skin, with the rubbing motion probably representing repeated stabblings at the same spot. Dissections of four snails immediately after mating showed that the dart is not lost. All four specimens had a single slender dart with a round base, a broad corona and a circular cross-section (fig. 2).

To conclude, our observations show that *P. muscarum* does not lose its dart after use, plus this species uses it repeatedly on the same mating partner. Hence, the way in which the dart is used by *Polymita* clearly differs from that of helicids. The latter shoot their dart once and lose it. The bradybaenid way, also repeated stabbing with a reusable dart (Koene and Chiba, 2006), is different in that the latter always stab into the same area. *Polymita*’s dart apparatus therefore seems unique for its agility. Furthermore, in most dart-possessing families, one type of mucus gland is associated with the dart (Nordsieck, 1987), while all the species of the genus *Polymita* possess two types: the bilobed and peduncular glands. According to Moreno (1950) the peduncular gland secretes calcium, suggesting that this gland may be used to build the dart. For now, it remains unclear whether secretions from either of these glands are transferred to the partner during ‘dart shooting’ in *P. muscarum*, and whether this transfer has a similar effect on sperm storage as in *C. aspersus* (Koene and Chase, 1998a, b; Rogers and Chase, 2002; Chase and Blanchard, 2006). If so, the repeated introduction of gland product with the dart could function to increase the transfer of the biologically active substance (i.e., all hormone: Koene and Ter Maat, 2001, 2002; Koene, 2005) that induces physiological changes in the partner. Finally, the morphology of *Polymita*’s dart is consistent with the idea that simple darts need to be stabbed more often to transfer enough mucus, while darts with a larger surface suffice with one stab (Koene and Schulenburg, 2005; Koene and Chiba, 2006).

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