

## GENERAL NOTES

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### DEFENSIVE FLOCCULENT EMISSIONS IN A TIGER MOTH, *HOMOEOCERA STICTOSOMA* (ARCTIIDAE:ARCTIINAE)

Additional key words: antipredator, Euchromiini, Panama, subabdominal pouch, moth, chemical defense.

Tiger moths (Arctiidae) exhibit a wide range of antipredator adaptations, including ultrasound reception and production, reflex immobilization, wasp mimicry, and chemical secretions in the form of foams and liquids (see Beebe & Kenedy 1957, Blest 1964, Rothschild et al. 1979, Fullard 1990, Conner 1999, Weller et al. 1999). While performing field studies in Panama over the course of several years, we observed what appears to be a novel mode of defense for the Lepidoptera- the expulsion of abdominal 'flocculent' material in a neotropical tiger moth, *Homoeocera stictosoma* Druce (Arctiidae, Arctiinae, Euchromiini) (Jacobson & Weller 2002). In this note we summarize our observations on the behavior associated with flocculent release, and describe some structural and chemical characteristics of the flocculent fibers, and the subabdominal pouches where they are produced and stored.

Certain species of the arctiid tribe Euchromiini possess conspicuous pouches on the ventral anterior abdomen. These pouches have been described only in males to date, and are variously referred to as 'ventral valves' or 'subabdominal pouches' (see Weller et al. 2000). Depending on the species, there may be one or two pouches, which contain either non-deciduous hair pencils or deciduous scales (Weller et al. 2000). The latter, when discharged from the pouch, appear as fluffy white or yellowish cotton, called 'flocculent' by Blest (1964). Despite the widespread occurrence of

subabdominal flocculent throughout the Euchromiini (R. Simmons, pers. com.), little is known of its function. Conner et al. (2000) provided experimental evidence for its role in courtship in one species, *Cosmosoma myrodora* Dyar, whereby males increase their probability of mating by enveloping the female with flocculent. Females covered with this material, in turn, are thought to be chemically protected against spiders, due to the high pyrrolizidine alkaloid content of the flocculent filaments. Flocculent has also been documented to play a role in courtship in two other species, *Syntomeida melanthus*, which actively releases flocculent (Sanderford 1992), and *S. ipomoeae*, which flashes its flocculent briefly, but does not release it during courtship (Johnson 2002). In *C. myrodora* and other flocculent-bearing species examined, there is no direct evidence that flocculent functions as a mechanism of defense for the possessor, because its production has not been observed to be evoked by handling or other mechanical disturbances of the individual (Blest 1964, Conner et al. 2000, R. Simmons, pers. com., Yack, pers. obs.). *Homoeocera stictosoma* (Fig. 1) is exceptional in this respect. Blest (1964) commented that this species emits flocculent upon 'light restraint of an individual'. Our observations, outlined below, corroborate Blest's earlier hypothesis.

Moths were collected at ultraviolet and mercury vapor lights in neotropical lowland rainforest on Barro Colorado Island by A. Aiello and R. Silberglied in

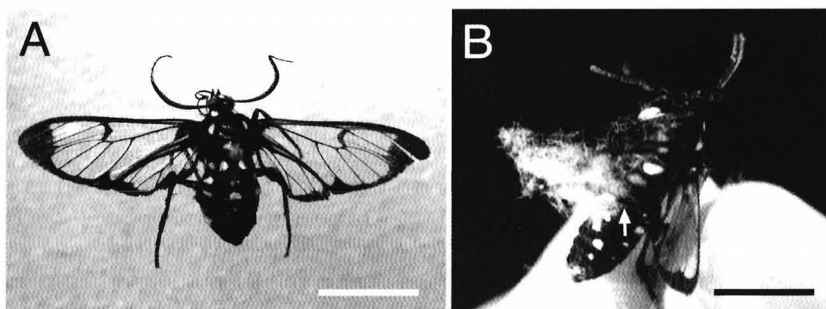


FIG. 1. A. A male *Homoeocera stictosoma*, collected on Barro Colorado Island, Panama. B. A male *H. stictosoma* discharging flocculent while being restrained. An arrow points to the subabdominal pouch, where flocculent is rapidly scooped out by the thoracic legs.

March 1980, and by J. Yack in June and October 1998 and September 1999. Moths were collected from sheets and placed in plastic vials where they remained for up to 2 days until being tested. Behavioural observations were made on a total of 9 (3 in 1980; 3 in 1998; 3 in 1999) individuals at the study site, and subsequent anatomical investigations of the subabdominal pouches were carried out on 3 specimens fixed in alcoholic Bouin's (Pantin 1946) or C&C fixative (Chauthani & Callahan 1966), and one dried specimen cleared in warm 10% KOH for 10 minutes. Flocculent material was collected from two live specimens, glued to aluminum stubs and dried overnight. The specimens were coated with gold-palladium for 80 seconds using a Pelco SC-4 Plasma Coater, and examined with an Amray 1810 Scanning Electron Microscope. For chemical analysis, the flocculent was extracted with methanol (2 ml) overnight at room temperature and subsequently analyzed by positive-ion electrospray ionization mass spectroscopy, using a Micromass (Manchester, U.K.) Quattro I mass spectrometer. Moths were identified as male *H. stictosoma* according to their distinctive wing and body markings using Rubio & Pesántez (1997) and Draudt (1915), and through comparison with specimens at the United States National Museum with the assistance of R. Simmons.

Independent behavioral observations were made by one of the authors [AA] in 1980, and another author [JEY] during 1998 and 1999. In 1980, observations on *H. stictosoma* were made while doing palatability experiments with moths on small orb weaver spiders. During these trials, various tiger moth species were tossed from their holding vials into spider webs, and observations were made on the spider's response. To the author's surprise, when a *H. stictosoma* struck the web, it immediately used its legs to pull out abdominal flocculent, and thereby escaped the spider's web, leaving it covered in flocculent. Moths were recaptured and the trials were repeated until the moths ran out of flocculent material. Similar observations were made by J.E.Y. in 1999, whereby a male tossed into a spider web immediately released flocculent and escaped, leaving the web covered with flocculent fibers. In 1998 and 1999, all 6 moths tested discharged flocculent upon being restrained. In no cases was flocculent released while moths were collected or stored in plastic vials, but immediately upon being restrained, all individuals instantaneously responded by releasing clouds of flocculent that surrounded the moth, adhering to the forceps and/or fingers being used to hold the moth (Fig. 1). Flocculent emission discontinued when the moth was placed back in the holding container, but could be provoked repeatedly upon further restraint

until the subabdominal pouches were empty.

In *H. stictosoma*, the flocculent fibers are tightly packed inside two subabdominal pouches formed by abdominal sternites I, II, and III (Fig. 2). The largest of the two, the anterior pouch, is covered ventrally by an enlarged sternal plate (SII) that partially overlays the anterior edge of SIII. The opening to the anterior pouch is directed posteriorly. The outermost edge of SII is lined with clear, rounded, non-deciduous scales that may be interpreted as scent scales (cf. Weller et al. 2000). The opening to the smaller posterior pouch occurs between SIII and SIV, with shallow pockets extending both anteriorly and posteriorly. The posterior edge of SIII is also lined with transparent, rectangular scales that are somewhat smaller than those associated with SII. Corresponding to the pattern found in other euchromiines, female *H. stictosoma* lack pouches (R. Simmons, pers. com.). In a detailed examination of the subabdominal pouches in the euchromiine-ctenuchine clade, Weller et al. (2000) described two main types: a single pouch, and a double pouch that corresponds to that of *H. stictosoma*. Species that possess a constricted abdomen (i.e. mimicry of the hymenopteran petiole - wasp mimics) consistently have either a single pouch or no pouch at all, suggesting that there is a trade off between wasp mimicry and the development of subabdominal pouches. *Homoeocera stictosoma*, accordingly, lacks a restricted abdomen.

Although the flocculent fibers are highly compressed within the pouches, when dispersed, they appear as lightweight 'fluff' that floats around the moth. Upon examination with the scanning electron microscope, the flocculent material of *H. stictosoma* is filamentous, composed of flattened, sculptured scales (Fig. 3). Within the Euchromiini, the morphology of flocculent varies considerably, from ribbon like scales to those that resemble twisted chains or even solid cylinders (Sanderford 1992, Boada 1997, Conner et al. 2000). Those described here for *H. stictosoma* appear to be constructed for lightness and strength, and are described as being thin and ribbon like with a network pattern, and covered with a thin translucent film. They most closely resemble the flocculent of *Gymnelia salvini* Butler, *Sarosa* sp., *Myrmecopsis crabonis* Druce, and *Pseudophex polistes* Hubn. (Boada 1997). In *H. stictosoma*, the flocculent also bears a rather strong phenolic odour when released. Mass spectra obtained showed strong ions of  $m/z$  134 and 222, characteristic for a number of pyrrolizidine alkaloids (PAs), therefore suggesting the presence of PAs in the flocculent. The small amounts of flocculent available for analysis however, did not yield enough material for further mass spectroscopic or NMR spectrometric in-

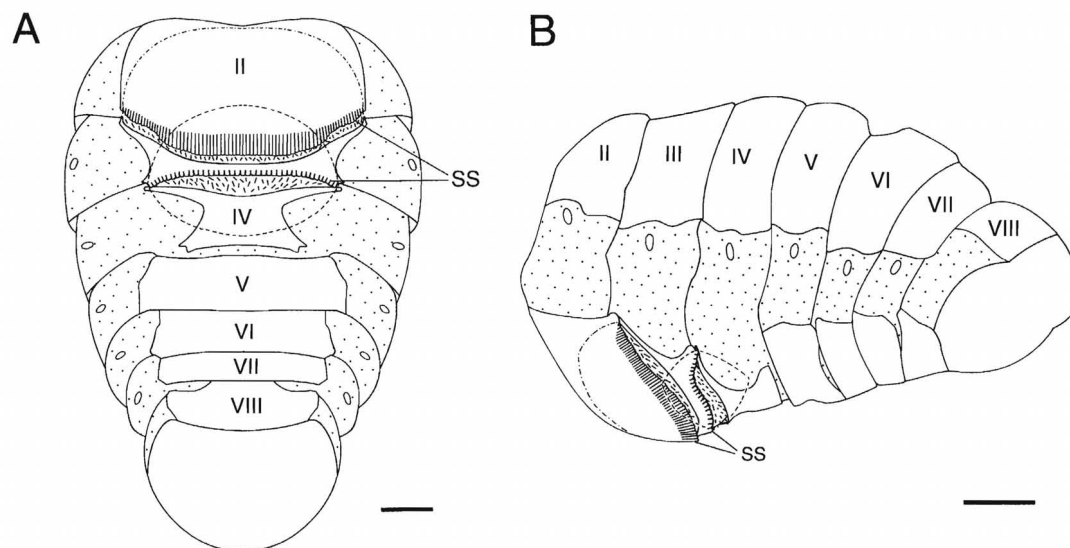


FIG. 2. Schematic representations, based on camera lucida drawings, of the ventral (A) and left lateral (B) views of the abdomen of a male *H. stictosoma*. Flocculent material is densely packed within two subabdominal pouches, the openings of which are depicted by irregularly spaced short lines. Rectangular-shaped scales, interpreted as scent scales (SS) line the edges of the openings to the subabdominal pouches. The larger anterior pouch is defined by sternite II and is directed anteriorly, while the posterior pouch is directed anteriorly and posteriorly. Scale bar: 1 mm.

vestigations to corroborate this hypothesis.

Defensive secretions, including regurgitation, defecation, autohemorrhaging, and the expulsion of foams and sprays are commonly used among insects (Whitman et al. 1990), but to our knowledge, reflexive discharge of 'scales' has not been documented. Our observations of *H. stictosoma* indicate that flocculent is employed as a defense against spiders and vertebrate predators for the following reasons. 1. Flocculent is consistently discharged upon restraint of an individual. 2. The flocculent scales adhere to the 'attacker' and have a distinct phenolic odour, suggesting a chemical defense. 3. Moths discharge the flocculent when colliding with a spider web, and this is followed by their escape, suggesting that the flocculent coats the web so that the moth does not stick to it. The hypothesis that flocculent functions as an 'instant release' from spider webs and/or an 'anti-consumption' device should be validated experimentally by performing feeding trials with various vertebrate and invertebrate predators, and a detailed analysis of the flocculent chemistry. Also, the role that flocculent plays in the courtship behaviour of *H. stictosoma*, and in the courtship and/or defensive behavior of the other 19 listed *Homoeocera* species (Druce 1881-1900; Draudt 1915) should be explored.

Despite the widespread occurrence of subabdomi-

nal pouches in the Euchromiini, little is known of the taxonomic distribution or functional significance of these interesting structures and their constituent flocculent material. To date, the use of flocculent in the context of courtship has been implicated in only 3 species- *C. myrodora* (Conner et al. 2000), *Syntomeida ipomoeae* (Johnson 2002) and *S. melanthus* (Sanderford 1992). In *C. myrodora*, the flocculent may also function indirectly for defense, by protecting the 'adorned' female from spiders (Conner et al. 2000). *Homoeocera stictosoma* is the only species reported to date that unequivocally discharges flocculent upon being restrained. Two other species, *C. teuthras* and *C. myrodora*, have been suggested to use "fluff from the ventral valve" as part of a defensive behavior (Adams 1990), but no other details were provided, and subsequent attempts to induce flocculent production in *C. myrodora* have proved unsuccessful (Conner et al. 2000). It is not possible at this time to know how these various traits evolved, due to our lack of understanding of the functional distribution of flocculent material within the Euchromiini, and the phylogenetic relationships among Euchromiini species. It is interesting to note that some Arctiidae extrude non-deciduous, brightly colored cervical, genital or abdominal hair tufts upon being restrained, and it is believed that the use of these structures in a defensive context de-

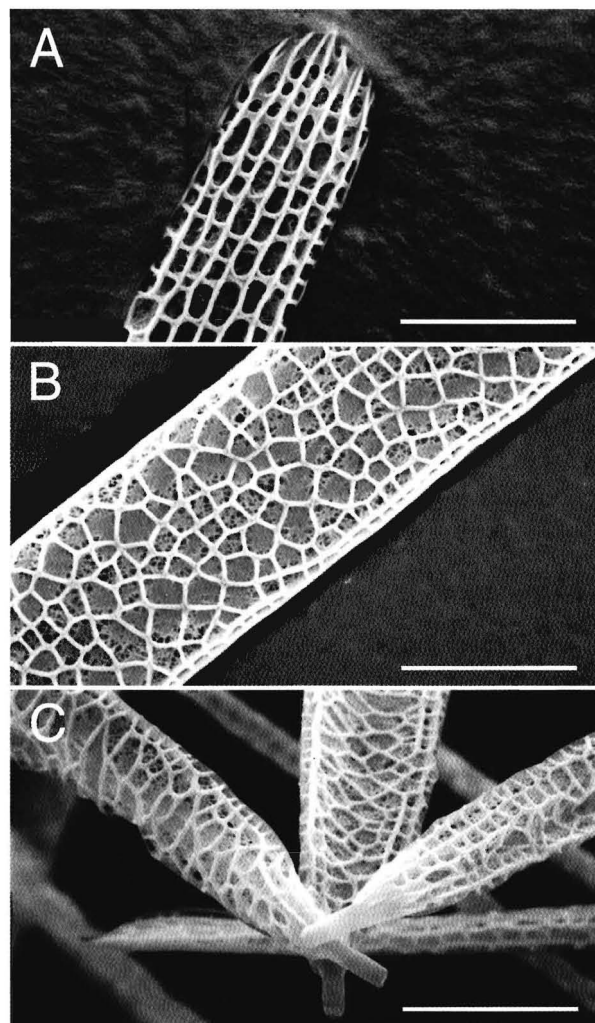


FIG. 3. Filamentous 'scales' from flocculent material in a male *H. stictosoma*. A. Rounded tip of a filament. B. Middle portion of a filament. C. The bases of three filaments where they have broken off of the abdominal pouch. Scale bars: 10  $\mu$ m.

rives secondarily from a mating function (Blest, 1964). Similarly, one can envision that the defensive emission of flocculent, as we have proposed for *H. stictosoma*, evolved secondarily from a courtship function.

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