PROC. ENTOMOL. SOC. WASH.
100(2), 1998, pp. 353–360

ON THE STRUCTURE AND FUNCTION OF THE FEMALE GENITALIA IN FLEA BEETLES (COLEOPTERA: CHRYSOMELIDAE: ALTICINAE)

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Abstract.—Structure, terminology and possible homology of female genitalia in flea beetles (Alticinae) are discussed. The relative position of the male and female genitalia was examined in a pair of *Aphthona formosana* Chen preserved in copulation. It is speculated that the vaginal palpi of the female genitalia and the median lobe of the male genitalia function together during copulation as an "internal courtship device" to regulate sperm transfer and egg fertilization.

Key Words: Female genitalia, homology, function, Alticinae

The earliest documented observations on beetle genitalic structures are by Foudras (1859, 1860). That predates the claim of Nichols (1986) for Thomson (1875) by nearly 15 years. In the introduction to his famous "Alticides" Foudras (1859) admitted the usefulness of the "aedeagus" for species differentiation and further (Foudras 1860) provided diagnostic characters for many European species. Baly (1879) and Weise (1886, 1889a, b) discussed the taxonomic importance of the median lobe in several genera of Cryptocephalinae, Chrysomelinae, Donaciinae and Clytrinae. Early in this century, male genitalia (median lobe of the aedeagus in particular) were employed for species differentiation in flea beetles by Heikertinger (1911, 1913, 1921, 1925) and the median lobe has been illustrated in many descriptions of Palearctic species published subsequently, especially for such large genera as *Altica* Geoffroy, *Aphthona* Chevrolat, and *Longitarsus* Latreille.

The first attempt to describe female genitalia (mostly the spermatheca) was by Spett and Lewitt (1925). Later Spett and Lewitt (1926) described the spermathecae of 63 species of Alticinae from 15 Palearctic genera. However, only since the 1960s has the spermatheca become a popular source of species level characters in flea beetles (Samuelson 1966, Berti and Rapilly 1973, 1977, Leonardi 1970, 1975). A comprehensive review of research on alticine spermathecae was published by Döberl (1986). The other structures of the female genitalia received much less attention (Suzuki 1982, 1983, 1988). To my knowledge Lyubishchev (1959, 1969) and Kevan (1962) were the first to use vaginal palpi for discrimination of species in the genus *Altica*. Several new characters in the shape, proportions and degree of sclerotization of the vaginal palpi (= styli in Konstantinov 1987, 1994) as well as intraspecific variability have been found more recently in this genus (Konstantinov 1987). Kangas and Rutanen (1993) confirmed these characters for Finnish species of *Altica*. Since these publications, illustrations of the vaginal palpi usually have accompanied redes-

criptions of *Altica* species (Doguet 1994, LeSage 1995). There was a single attempt to review the female genitalia of the Palearctic genera (Konstantinov 1994). Aside from *Altica* (Lyubishchev 1959, 1969, Kevan 1962, Konstantinov 1987, Kangas and Rutanen 1993, Doguet 1994, LeSage 1995), *Crepidodera* Chevrolat (Konstantinov 1996), *Aphthona* (Konstantinov and Vandenberg 1996), *Bellaltica* Reid (Reid 1988), *Pedilia* Clark (Duckett 1995) and *Homichloda* Weise (Cox 1997) almost nothing has been published about these structures in other flea beetle genera.

Despite a demonstrated usefulness of female genitalia for species identification and derivation of relationships of flea beetles, they are much less known than male genitalia. Homology, terminology and functions of their structures remain obscure.

Methods

Techniques for dissecting and studying female genitalia are simple and have been well documented (Tanner 1927, Döberl 1986, Duckett 1995). In these and the present study, beetle abdomens were treated with hot 10 % KOH, then washed in water and dissected. The genitalia were transferred to a mixture of water and glycerin (which prevents collapse of the spermatheca and vaginal palpi). A second method involves everting the entire genital apparatus without dissecting the abdomen, however this method is not recommended as the process puts structures in artificial conditions changing their relative position, and preservation of the natural position of the structures is essential for their recognition, naming and comparison.

A male and female of *Aphthona formosana* Chen preserved *in copula* with the internal sack inflated were studied to determine the approximate position of the median lobe of the male and sclerotized structures of the female genitalia during the last phases of copulation and permit some speculation on the functions of some parts of the female genitalia.

Structure of the Female Genitalia in Flea Beetles

The chitinized structures of the female genitalia (Fig. 1) are situated in the abdomen and covered by the strongly sclerotized 7th tergite and sternite. As in all other leaf beetles, the ventral side of the abdomen consists of 5 visible sternites. The first true sternite is reduced and the first visible sternite is composed of the second and third true sternites. The dorsal side of the abdomen consists of 7 visible tergites. The vagina is formed by invagination of the body surface and its opening (vulva) is situated between the 9th tergite and 8th sternite. The sclerotized structures of the female genitalia are derivatives of the 8th and 9th abdominal segments, but their homology and terminology is poorly known (Lindroth 1957, Teotia 1958, Konstantinov 1994). The 8th and 9th sclerites lie telescoped within the 7th segment. The 8th tergite is situated under the 7th, and the 9th tergite is situated under the 8th. They have preserved the plate shape, and are easily recognizable. The 8th and 9th tergites are connected with each other by membranes which join the distal margin of the upper sclerite with the proximal margin of the lower sclerite.

The sternites are highly modified. The 8th sternite is represented by a weakly sclerotized plate lying above the 7th sternite, usually with a well developed, strongly sclerotized, narrow projection called a tignum (Kevan 1962, Konstantinov 1994, Konstantinov and Vandenberg 1996), spiculum gastrale (Kasap and Crowson 1985, Crowson and Crowson 1996), spiculum (Reid 1988) or long apodeme (Teotia 1958). All these terms are synonyms and their usage does not create contradictory hypotheses of homology between the structures.

The paired vaginal palpi are elongate, partly sclerotized structures with several long setae on the apex and a cavity inside, contained within the vagina and attached to the inner wall of its dorsal surface. They extend posteriorly along the dorsal side of

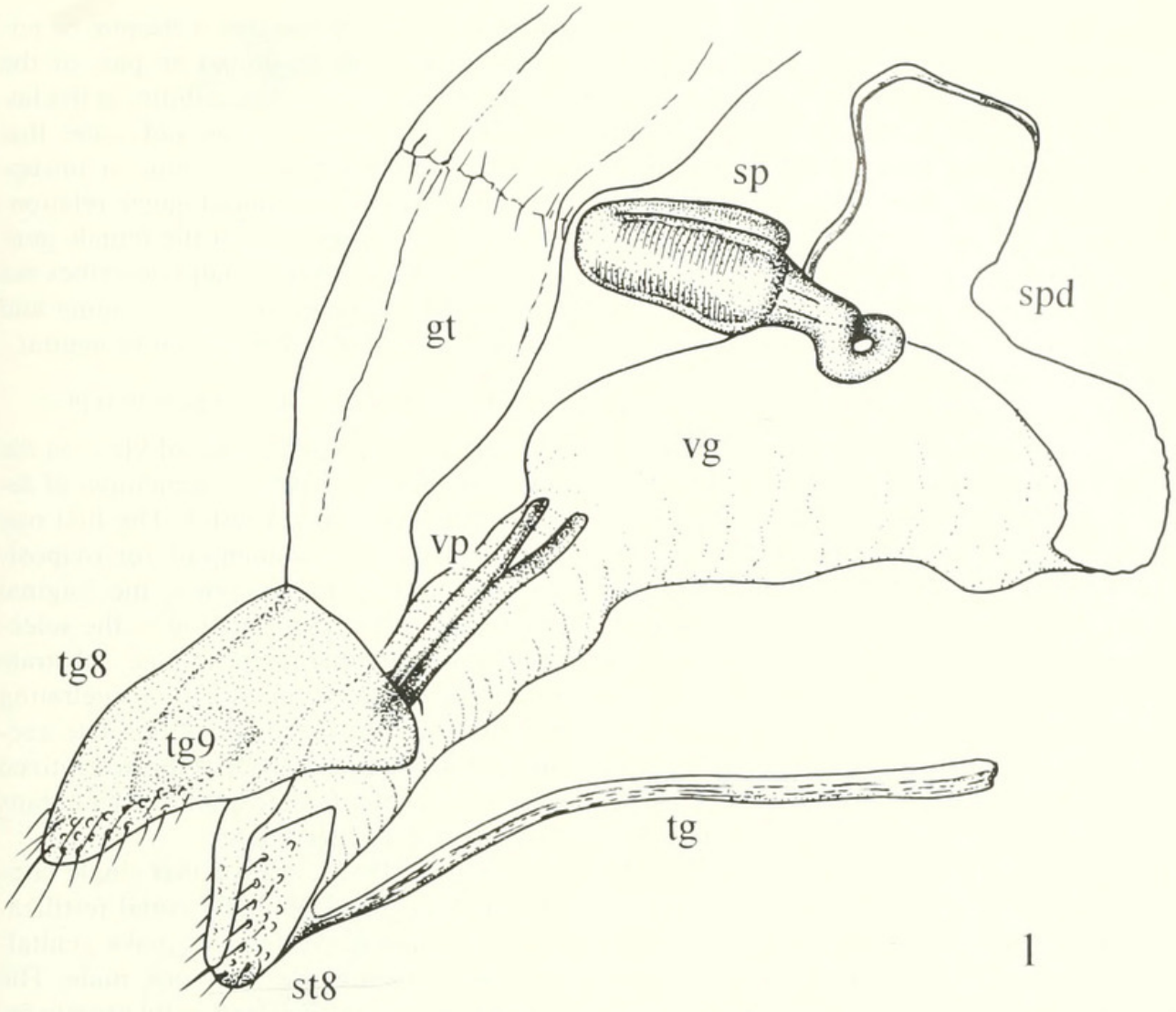


Fig. 1. Female genitalia of *Altica tamaricis* (lateral view). Abbreviations: gt = gut; sp = spermatheca; spd = spermathecal duct; st8 = 8th sternite; tg = tignum; tg8 = 8th tergite; tg9 = 9th tergite; vg = vagina; vp = vaginal palpi.

the vagina, terminating below the basal part of the 9th tergite. The apical part of the palpus is circularly sclerotized; basally the sclerotization continues on the dorsal side as a narrow stripe along the medial side. The ventral side of the palpus is usually membranous near the middle and forms a sclerotized plate basally. Unfortunately nothing is known about the origin of the vaginal palpi. It is unlikely that they represent a modified 9th sternite, as I proposed previously (Konstantinov 1994) because of the position of the vaginal palpi on the dorsal wall of the vagina.

Many terms have been applied to vaginal palpi: gonocoxa (Duckett 1995), hemisternite (Reid 1988), ovipositor (Crowson and Crowson 1996), styli (Konstantinov 1987, 1994), vaginal palps (Kasap and Crowson 1985, Konstantinov and Vandenberg 1996), and unjointed coxites (Teotia 1958). The term gonocoxa is used in various meanings, but more preferably as a coxite of gonopods (Tuxen 1956). Hemisternites are primarily defined as 2 sclerites of the 9th sternite (Tuxen 1956, Lindroth 1957) surrounding the gonopore. The term ovipositor does not imply any homology but determines the function of the structure. Styli are the distal part of the gonopods, movably attached to basal segment of the gonopods (coxites) or to abdominal sterna (Tuxen 1956), usually

divided into 2 articulated joints (Lindroth 1957). Coxites are the basal parts of the appendages of the 9th (rarely 8th) genital segment carrying styli (Tuxen 1956). The application of these terms to the vaginal palpi would consider them as: 1) sclerites attached to the 9th sternite; 2) basal part of the appendages of the 9th genital segment (either tergite or sternite) carrying styli; 3) styli themselves, which are the apical part of the aforementioned appendage; 4) the whole appendage attached to the 9th tergite.

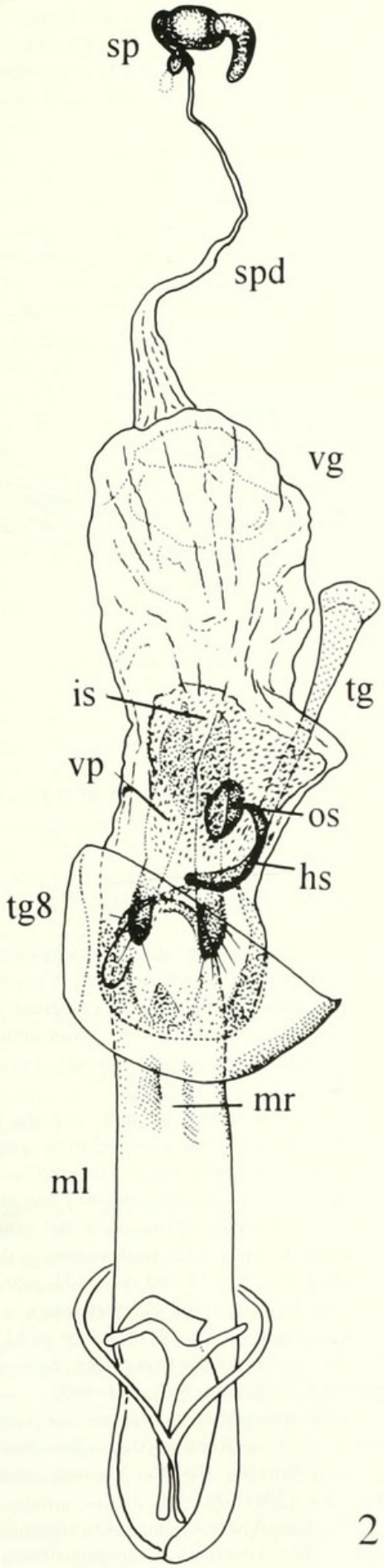
The structure of the female genitalia in Alticinae is extremely different from that in other families of beetles (Bils 1976, Burmeister 1976, 1980, Jablokoff-Khnzorian 1974, Mickoleit 1973). It is also different from that in other leaf beetle subfamilies. Donaciinae have well developed paraprocts with attached setose structures that can be interpreted as coxites (Askevold 1988). In Sagrinae the paraprocts contain coxites with attached styli (Mann and Crowson 1989). The female genitalia of Chrysomelinae (Konstantinov and Rusakov 1993) lack the 9th tergite and are extremely diverse: ranging from *Timarcha tenebricosa* F. with well developed paired structures situated inside the vagina, tignum and spermatheca to *Gonioctena viminalis* L. without 8th sternite, vaginal palpi and sclerotized spermatheca. No visible paraprocts and no structures associated with paraprocts have been found in Alticinae, and therefore no structures can be easily homologized with the gonocoxite and gonostylus. Therefore the names hemisternite, styli, coxites and gonocoxa cannot be applied to vaginal palpi in flea beetles until a connection between vaginal palpi and the 9th segment is found. There are 3 possibilities to homologize the vaginal palpi: 1) vaginal palpi are paraprocts (halves of the 9th tergite) which moved inside the vagina; 2) vaginal palpi are hemisternites (appendages of the 9th sternite) and by definition they should be at the gonopore; 3) vaginal palpi are inverted and sclerotized parts of the vaginal wall; in this case they do not represent any segment. Since it is improbable that a sternite or tergite can become integrated as part of the vagina, the most likely possibility is the latter. That alternative is the only one that does not imply either erroneous or unsupported ideas on the homological relationships between structures of the female genitalia. The name vaginal palpi describes the location of the palpi inside the vagina and because of that this name is more neutral.

Function of the Vaginal Palpi

There are two main points of view on the function of the sclerotized structures of female genitalia in flea beetles. The first one considers them as adaptations for oviposition. According to this view the vaginal palpi serve a sensory function in the selection of a favorable ovipositional substrate and/or mechanical function in penetrating the substrate during oviposition. The second point of view considers the sclerotized structures of the female genitalia as organs participating in copulation.

Eberhard (1985) showed that single copulation does not necessarily entail fertilization in groups where females make genitalic contact with more than one male. His hypothesis of sexual selection by cryptic female choice suggests that "male genitalia function as an "internal courtship" device to increase the likehood that females will actually use a given male's sperm to fertilize her eggs rather than those of another male." Therefore the female is choosing males based on the structure of their genitalia.

This suggests that females should have a sense organ which would be in mechanical contact with the male genitalia and recieve stimuli provided by male genitalia during intromission. In the case of flea beetles there are only 2 structures that can function as this sensory organ: the vagina and the vaginal palpi. In most flea beetle genera the diameter of the vagina is much larger than the diameter of the median lobe of the aedeagus, it also has no indication of any sensory structures. The vaginal palpi are at-



tached to the dorsal wall of the vagina, slightly bent ventrally (in lateral view) so their apices are located slightly above the middle of the vagina. The wholly sclerotized apices with a cavity inside and several long setae, strong medial sclerotization and sclerotized ventral side with membranous medial part make this organ quite sensitive to mechanical influence. In this example of copulation (Figs. 2, 3), in which the internal sack of the median lobe is inflated, there is contact between the apices of the vaginal palpi and the apical part of the ventral side of the median lobe. This part of the median lobe is the most species specific in flea beetles. In *Altica* it has many longitudinal impressions and transverse ridges. In *Aphthona,* especially in the *hammarstroemi* group of species it has a moderately developed median ridge and two lateral impressions. In the *A. formosana* pair in copulation the median ridge of the median lobe of the male is situated between the apicies of the vaginal palpi of the female. The chitinized structures of the internal sac are represented by an oval structure and a long hook to the left of the oval structure. The membrane of the sac, especially apically, is covered with numerous short setae (Fig. 3). All these structures are situated just below the vaginal palpi where they might contribute to their stimulation.

Based on the structure, position and orientation of the vaginal palpi and the median lobe of the aedeagus I suggest that the vaginal palpi function as sense organs, the female component of a so called "internal courtship device" which receives stimuli produced by movement of the median lobe of the aedeagus that induce sperm transfer and egg fertilization.

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Fig. 2. Male genitalia inside vagina (dorsal view) of *Aphthona formosana.* Abbreviations: hs = hook structure of the internal sack; is = internal sack of the aedeagus; ml = median lobe of the aedeagus; mr = median ridge of the median lobe; os = oval structure of the internal sack. Other abbreviations as in Fig. 1.

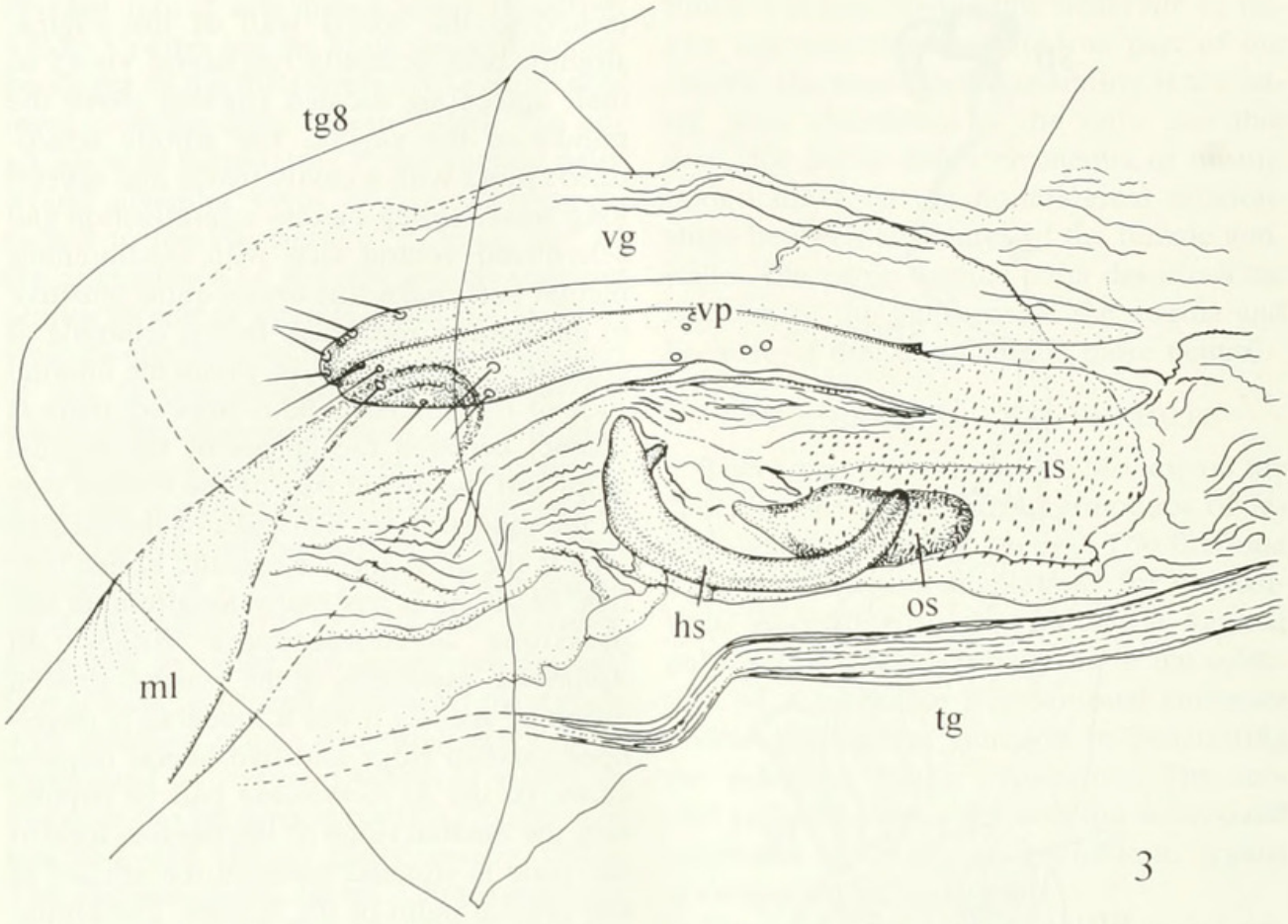

Fig. 3. Male genitalia inside vagina (lateral view) of *Aphthona formosana*. Abbreviations as in Fig. 1, 2.

Acknowledgments

This work was completed during my postdoctoral fellowship at the Systematic Entomology Laboratory, USDA, ARS, funded by the National Biological Control Institute and Systematic Entomology Laboratory. I greatly appreciate support from these organizations, their leaders and staff members. I am also grateful to S. Lingafelter, A. Norrbom, M. Pogue, N. Vandenberg (Systematic Entomology Laboratory, USDA, Washington, DC), M. Coca, and M. Kotrba (Department of Entomology, Smithsonian Institution, Washington, DC) for reviewing earlier versions of this manuscript and their valuable suggestions. D. Furth (Department of Entomology, Smithsonian Institution, Washington, DC) and S. Lingafelter suggested some useful references (Eberhard 1985 and Nichols 1986, respectively).

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