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Morphology and development of the accessory glands in various female cricket species

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ABSTRACT

The study presents new results with regard to the morphometric and ultrastructural development of the accessory glands in females of the three cricket species Gryllus bimaculatus, Gryllus assimilis, and Acheta domesticus. Furthermore, possible age-dependence of secretory productivity of single organs was analyzed by application of the ligature technique introduced in a previous contribution. Within the first 12 days of the adult phase, the accessory glands of all investigated cricket species exhibit a significant increase in length and width which assumes values between 50 and 100%. This gland growth is rather the result of a continuous increase in cellular volume and less that of mitotic cell propagation. In all species height and width of single gland cells increase by 60-80% within the studied time interval. These changes in morphometry are commonly accompanied by ultrastructural modifications. Total glandular secretion is subject to an increase from the 5th to the 12th day of adult age. This development corresponds well with the number of eggs contemporaneously oviposited into the substrate and thus underlines the hypothesis, according to which the main function of the secretion consists in acting as a lubricant for the facilitated transport of the oocytes through the ovipositor.

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1. Introduction

As found by numerous previous studies (e.g., Brunet, 1952; Gerber et al., 1971; Happ et al., 1977; Happ, 1984; Locke, 1985; Quennedey, 1998; Sturm and Pohlhammer, 2000; Sturm, 2002a), insect glands belonging to the reproductive system are subject to an extensive developmental process, before the adult animal attains its full reproductive functionality. In many cases, this development, starting immediately after the imaginal molt, includes both a significant morphogenesis and a related cytological differentiation. Whilst morphogenetic processes mainly result in an increase of gland size and volume (Sturm and Pohlhammer, 2000; Sturm, 2002a, 2008), characteristic features of cellular differentiation among other comprise a significant enhancement of cell volume as well as diverse ultrastructural changes such as growth of the nucleus (nucleolus) and extensive propagation or synthesis of those cell compartments being involved in metabolic processes (Berry, 1968; David, 1977; Locke, 1980a,b; 1985; Happ, 1984; Sturm, 2002b, 2008).

Among numerous orthopteran insects male and female accessory glands are characterized by essential morphometric and structural changes during early adulthood (Gillott, 1988; Kaulenas, 1992; Sturm, 2002a). Detailed investigation of the accessory glands in females of the black field cricket Teleogryllus commodus furnished proof that within a time period of 12 days organ size may be subject to a 100%-increase. This phenomenon, however, is exclusively caused by gland cell hypertrophy, with single cellular units experiencing volume gains between 400% and 700%. Ultrastructural changes of single cells include a simultaneous growth of the nucleus and nucleolus, a significant increase of the number of mitochondria, and a remarkable multiplication of rough and smooth endoplasmatic reticulum (Sturm and Pohlhammer, 2000; Sturm, 2002a). In order to maximize the productive capacity of single gland cells, basal cell surfaces undergo a successive increase by forming deep invaginations which enable an enhanced uptake of basic substances from the hemolymph (Sturm, 2000, 2008).

Although morphology, histology, and function of female accessory glands in diverse cricket species have been largely clarified in the meantime (e.g., Lococo and Huebner, 1980; Sturm and Pohlhammer, 2000; Sturm, 2002b), development of these organs during the early adult phase still raises lots of open questions. In order to overcome this scientific deficiency, the female accessory glands of three cricket species (Gryllus bimaculatus, Gryllus assimilis, and Acheta domesticus) have been investigated to developmental

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aspects (Fig. 1). For this study the methodical canon formerly introduced by Sturm (2002a) has been applied to a large extent. The present contribution has been conducted on the basis of the hypotheses that (a) gland development passes very similar stages among the examined species and (b) intensity of organogenetic and cytologic changes may differ significantly from one species to the other.

2. Materials and methods

2.1. Animals

Rearing of the Mediterranean field cricket (*Gryllus bimaculatus*), Jamaican field cricket (*G. assimilis*), and house cricket (*A. domesticus*) was conducted in a climate chamber at the Department of Organismic Biology, University of Salzburg. Nymphal and adult animals of all species were fed on green lettuce, standard diet for laboratory rats (Altromin 1222), and water. Climatic conditions were set to constant values, using a photoperiod of 12 h, a mean temperature of 25 °C as well as a relative humidity of 60%. Nymphal stages of the crickets were kept in special plastic boxes ($40 \times 30 \times 20$ cm) filled with a 3 cm thick layer of peat soil and diverse accessories for shelter (e.g., egg cartons). Adult animals, on the other hand, were separated by gender and transferred into glass vessels (volume: 5 L) filled with wrinkled sheets of paper (Sturm and Pohlhammer, 2000; Sturm, 2002a, 2002b, 2011, 2014).

2.2. Microscopic investigations

For light- and electron-microscopic studies of the accessory glands selected females were anesthetized in a stream of carbon dioxide (CO_2) and decapitated. After that, the ventral abdomen was opened at the 7th and 8th segment. Before removal of the accessory glands could take place, all overlying and covering fatty tissue had to be macerated with the help of fine tweezers and preparation needles. The organs were taken out by a precise section at the orifice and transferred into a small glass vessel filled with insect Ringer's solution (Sturm and Pohlhammer, 2000).

Light-microscopic studies were carried out after transferring the glands together with Ringer's solution on a glass slide ($55 \times 20 \text{ mm}$) and covering the preparation with a cover slip ($20 \times 20 \text{ mm}$) placed on small petioles of plasticine. Description of the gland structure and morphometry was conducted on a Reichert Polyvar II microscope which enabled usage of the interference-contrast mode. For a more detailed depiction of single cellular



Fig. 1. General appearance of the accessory glands in females of *Teleogryllus commodus* (a), *Gryllus bimaculatus* (b), *Gryllus assimilis* (c), and *Acheta domesticus* (d) after their isolation from the genital tract, maceration from fatty tissue and unwrapping.

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