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„Mouthparts of adult Plodia interpunctella“

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1. Introduction

The Lepidoptera belong to one of the species-richest insect orders with 160,000 described species in 124 families (Kristensen et al., 2007). The majority of the larvae have biting-chewing mouthparts and feed on plant material. Most of the adult Lepidoptera possess a proboscis to imbibe liquid substances (Krenn, 2010). This form of nutrition led to special modifications of the mouthparts, which are modified into a suctorial proboscis, except for the most basal taxa of Lepidoptera (Kristensen, 2003). The morphology of lepidopteran mouthparts has been studied in numerous species of butterflies and moth (e.g. Eastham & Eassa, 1955; Kristensen & Nielsen, 1981; Büttiker et al., 1996; Krenn et al., 2001).

The mouthparts of Glossata consist of a small labrum, greatly elongated galeae of the maxillae, three-segmented maxillary palpi and the labium bearing prominent labial palpi (Kristensen, 2003). The labrum has lateral lobes, the piliferes, bearing bundles of bristles on their lateral edges (Krenn & Kristensen, 2000). In adult Glossata, the mandibles are nonfunctional. The basal parts of the maxilla, the stipes and the cardo, are fused together and form a hemolymph pump on each side of the labium. They bear the enlongated galeae, which compose the coillable proboscis (Kristensen, 2003; Krenn, 2010). The median walls of each galea constitute the food canal. On the dorsal and ventral sides, the galeae are linked by rows of cuticular processes, the legulae (Krenn & Kristensen, 2000). Each galea contains a trachea, a nerve and several muscles inside the lumen. At the base of the proboscis the basal galeal muscle extends from the proximal end of the galea to the dorsal proboscis wall (Krenn, 1990). Two series of intrinsic galeal muscles occur in the galeal lumen. The oblique lateral intrinsic galeal muscles are arranged one upon the other along the lateral proboscis wall. The median intrinsic galeal muscles are arranged longitudinally along the ventral wall (Krenn & Kristensen, 2004). In many taxa, the outer surface of the proboscis is covered with spine-like cuticular processes, referred to as microtrichia (Krenn & Kristensen, 2000).

Several different types of sensilla occur on the lepidopteran proboscis. They are usually classified into bristle-shaped sensilla trichodea, small cone-shaped sensilla
basiconica and the conspicuous sensilla styloconica (Städler et al., 1974; Faucheux, 1991; Paulus & Krenn 1996).

The sensilla on the cephalic appendages of Lepidoptera have been studied in several families (e.g. Faucheux, 2008; Krenn, 1998), including some species of the Pyralidae (Faucheux, 1991; 1995; Honda & Hanyu, 1989). The presence of the labial palp pit organ or vom Rath organ on the third segment of the labial palpi was described by Faucheux (1991, 1995) in two species of Pyralidae.

The species-rich family Pyralidae occurs on all continents except the antarctica. As characteristically concealed feeders, the larvae of many species of Pyralidae cause major damage to crops worldwide (Kristensen, 2003). The pyralid moth *Plodia interpunctella* (Hübner, 1813) is commonly known as the Indian Meal Moth and is distributed worldwide especially in households, as the larvae feed on stored food products, such as grain products, nuts and dried fruit. Up to 400 eggs are oviposited directly on the larval food source. The larvae hatch in three to eight days depending on the temperature. After five to seven larval instars, the development is completed in six to eight weeks. The pupal stage can last from seven to 20 days. Adults do not feed (Fasulo & Knox 1999), however they have been observed to take up water using their proboscis (Krenn, personal communication).

*Plodia interpunctella* has been the subject of many studies concerning development and reproduction (i. a., Savov, 1973; Hoppe, 1981; Johnson et al., 1992) or the effects and the production of ultrasound (Trematerra & Pavan, 1995; Huang & Subramanyam, 2004). No detailed investigations have been made on the morphology of the mouthparts and their sensilla in this species.

This diploma thesis investigates the morphology of the mouthparts with emphasis on the sensory equipment in adult *Plodia interpunctella* moths. The purpose of this study is to provide a detailed basic description of the mouthparts of *P. interpunctella*. Emphasis is laid on the types and distribution of the sensilla in order to contribute to establish this moth-species as laboratory animal for further investigations. Due to its relatively short generation time and since this moth is easy to rear in the laboratory, this species would be well suited as a future model organism for scientific research, especially for developmental biology.
2. Material and Methods

2.1 Biometry and light microscopic measurements

The individuals of *Plodia interpunctella* (Hübner, 1813), which have been used for this study, were reared and kept at room temperature and 35% relative humidity at the Department of Integrative Zoology at the University of Vienna, Austria. The body size was measured using a caliper rule in dead adult moth, *n* = 26.

To measure the length of the proboscis, the thorax, abdomen and labial palpi of dried animals were removed under a Nikon SMZ-U stereomicroscope. The heads were put into lactic acid for several days to uncoil the proboscis. Afterwards they were rinsed with alcohol (30%) and put onto a hollow object slide with a drop of glycerine and a cover glass for the analysis in light microscopy.

Photos were taken using a light microscope (Olympus CX41) with attached Olympus E330 digital camera. The photos were transferred to a computer, where the galea length was measured using ImageJ.

In order to study the external morphology of the proboscis in light microscopy, sagittal sections were made. As described above, dried animals were put into lactic acid for several days, then rinsed with alcohol. Afterwards the proboscises were separated into the galeae. They were put onto glass slides and fixed with glycerine and a cover slide.

2.2 Scanning Electron Microscopy

The samples for scanning electron microscopy were prepared and observed at the Core Facility of Cell Imaging and Ultrastructure Research (University of Vienna, Austria).

The specimen used for scanning electron microscopy were fixed in picric acid for several days and afterwards stored in 70% ethanol. The abdomen, wings and legs were removed using a stereomicroscope (Nikon SMZ-U), the head and thorax were dehydrated in 95% ethanol two times for 30 minutes and subsequently in 100% ethanol two times for 15 minutes and two times for 20 minutes. They were
submerged into Hexamethyldisilazane for 30 Minutes and left to air-dry over night under the fume hood.

The protocol followed a standard procedure which was already proven to be useful in studies of lepidopteran mouthparts (e.g. Krenn et al., 2001).

From some specimen the scales of the labial palpi, the maxillary palpi and the proboscis were removed under the binocular using adhesive tape. From other samples the proboscis and/or the labial palpi were completely removed in order to view the basal structures of the mouthparts.

The prepared heads, several dissected proboscides and labial palpi were mounted on aluminium stubs using carbon foils and conductive silver, left to dry for a few hours and were then covered with a thin layer of gold within the Sputter Coater (Agar sputter coater, for 200 seconds).

SEM - Images were taken using a Philips XL 20 SEM and Philips XL 30 ESEM, with an acceleration voltage of 15 kV. The photos were processed using Adobe Photoshop CS6. Contrast enhancement and brightness adjustments were performed in several photos. ImageJ was used for length measurements.

2.3 Semi-thin sections

The specimen were fixed in alcoholic bouin’s solution (Duboscq-Brazil) for 48 hours, afterwards dehydrated with ethanol and acetone. They where embedded in an ERL-4206 epoxy-resin (procedure according to Pernstich et al., 2003). Semi-thin-sections were made on a Reichert ultramicrotome using a Diatome diamond knife. The sections where put on slides and stained with a mixture of 1% azure II and 1% methylene blue in a 1% borax solution. The slides where then placed on a heating plate at 80°C for 30 seconds. The sections were studied using an Olympus CX41 microscope. For taking pictures a drop of glycerin and a cover glass was added on top of the selected section. An Olympus E330 camera was used to take the photos.
3. Results

The mean body size of *Plodia interpunctella* is 6.1 mm (SD ± 0.8; n = 26). In resting position the wings are folded above the body. They proceed beyond the end of the abdomen. The length from the anterior side of the head to the tip of the wings measures 7.8 mm (SD ± 0.9; n = 26).

The mouthparts of *Plodia interpunctella* consist of the labrum bearing the lateral piliferes, small mandibles, the maxillae consisting of proboscis and maxillary palpi and the labium bearing the labial palpi (Fig. 1).

The majority of the mouthparts are reduced in size, except for the galeae, which compose the proboscis and the labial palpi, which cover the proboscis on the lateral sides. No differences in the morphology and anatomy of the mouthparts could be found in the two sexes.

3.1 Labrum

The labrum lies beneath the clypeus (Fig. 2) and above the basal joint between the proboscis and the head capsule. The labrum is reduced to a tiny triangle. It bears several microtrichia and is entirely covered with bristles (Fig. 3).

The two piliferes, i.e., lateral lobes of the labrum, are located at both sides of the labrum and have several small microtrichia at the base. They bear numerous very long sensory bristles, which are oriented towards the proboscis and are in contact with the proximal end of the galea. Their mean length is 68.3 µm (SD ± 4.9; n = 7).

They are striated and are shorter on the edge of the piliferes, they become longer and thicker to the medium side of the pilifer (Fig. 4).

3.2 Mandibles

The mandibles are rudimentary, quite small and triangular. They are located below the piliferes. Their cuticle is deeply striated. They bear pointed, prickle-shaped thorns of unknown function (Fig. 5).
3.3 Maxillae and Proboscis

The basal parts of the maxillae, the stipes and cardo, are located on each side of the labium (Fig. 2). The galea originates from the anterior end of the stipes. The two galeae are prolonged and interlocked, they are forming a sucking tube with a single central food canal. The mean length of this proboscis is 3.3 mm (SD ± 0.32; n = 16).

In its resting position, the proboscis is curled up between the labial palpi beneath the head in 4 – 4.5 coils (Fig. 6). The outermost coil is covered in scales, it touches the ventral side of the head at the labium.
The cuticula of the proboscis is formed by cuticular ribs from the base to the tip. They are broader at the base and become narrower towards the tip (Tab. 1).

**Table 1:** Width of the ribs on the proboscis surface; SD standard deviation; n sample size

<table>
<thead>
<tr>
<th>Region</th>
<th>Mean (µm)</th>
<th>SD (µm)</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>Base</td>
<td>7,03</td>
<td>0,55</td>
<td>10</td>
</tr>
<tr>
<td>Near base</td>
<td>6,54</td>
<td>0,8</td>
<td>13</td>
</tr>
<tr>
<td>Middle</td>
<td>5,97</td>
<td>0,53</td>
<td>11</td>
</tr>
<tr>
<td>Near tip</td>
<td>5,44</td>
<td>0,43</td>
<td>13</td>
</tr>
<tr>
<td>Tip</td>
<td>5,08</td>
<td>0,59</td>
<td>10</td>
</tr>
</tbody>
</table>

The surface of the proboscis is completely covered with microtrichia. These cuticular processes are long and thin at the base of the galea (Fig. 7). At the base, they contact the sensilla of the piliferes. They are short and pointed on the other regions of the proboscis (Fig. 8). The two galeae are held together by the legulae. These cuticular structures form the dorsal and ventral linking construction of the galeae. There are two rows of legulae per galea on the dorsal and ventral side, which intertwine with their counterparts on the other galea at the opposite side (Fig. 9). The ventral legulae are tightly connected from the base to the tip of the proboscis. The dorsal legulae are less tightly interlocked with each other, since they often opened up during SEM-preparation of the specimen (Fig. 10). In the distal part of the proboscis, the two rows of dorsal legulae melt to a single row. This so called tip region is about 0.2 mm long. It corresponds to about 6.5% of the total proboscis length (Fig. 10).

There are 5 types of sensilla on the proboscis, i.e., bristle shaped sensilla chaetica and sensilla trichodea, cone shaped sensilla basiconica types 1 and 2 and sensilla styloconica. Sensilla basiconica type 2 occur only inside the food canal.

Sensilla chaetica were found only at the base of the proboscis. They are distributed between and beneath the scales. They have a very small socket and a long, striated sensory bristle with a mean length of 37.2 µm (SD ± 1.87; n = 6). The sockets resemble those of scales, but they are tighter around the base of the sensilla (Fig. 7). Sensilla trichodea have a mean length of 10.9 µm (SD ± 1.52; n = 21). They are composed of a small socket and a long sensory bristle (Fig. 11). They are scattered...
all over the surface of the proboscis. The bristles are longer in the basal region of the proboscis and become shorter towards the tip (Tab. 2). The bristles do not show any pores (Fig. 11).

Figures 6-9: Proboscis of *Plodia interpunctella*; **Fig. 6** Oblique lateral view of coiled proboscis (*p*); *mp* maxillary palp, *sc* scales; **Fig. 7** Dorsal view on the base of the proboscis showing the elongated microtrichia (*mi*) and the sensilla chaetica (*se*); *lp* labial palp; **Fig. 8** Ventral view of the proboscis covered with microtrichia (*mi*), showing the tight ventral legulae (*vl*) and some scattered sensilla trichodea (*st*); **Fig. 9** Dorsal view on proboscis. The dorsal legulae (*dl*) opened up due to SEM-preparation; **Fig. 10** Tip of proboscis; two rows of legulae melt to a single row (region marked by arrows); both dorsal and ventral legulae were separated as an artefact of SEM preparation; *ss* sensilla styloconica
Sensilla basiconica possess a small socket and a short sensory cone with a pore on the apex. They are only 4.4 \( \mu \text{m} \) long (Fig. 12). They are much less frequent and only present on the dorsal surface of the proboscis, where they are situated in a row near the dorsal legulæ. In the tip region a few sensilla basiconica are located on the lateral side of the proboscis, that have a much longer sensory cone than more proximally on the galea. Their mean length is 7.3 \( \mu \text{m} \) (SD ± 0.86; \( n = 6 \)).

### Table 2: Length of sensilla trichodea in the different regions of the proboscis; SD standard deviation

<table>
<thead>
<tr>
<th>Region</th>
<th>Mean (µm)</th>
<th>SD (µm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Base</td>
<td>12,16</td>
<td>1,11</td>
</tr>
<tr>
<td>Middle</td>
<td>10,82</td>
<td>1,00</td>
</tr>
<tr>
<td>Tip</td>
<td>9,57</td>
<td>1,24</td>
</tr>
</tbody>
</table>

Figures 11-14: Sensilla and microtrichia of the proboscis; Fig. 11 Bristle shaped sensillum trichodeum (st); mi microtrichia: Fig. 12 Cone shaped sensillum basiconicum (sb) showing a terminal pore (arrow); le legulæ Fig. 13 Sensillum styloconicum (ss) with the massive socket and the 4 pikes; Fig. 14 Sensillum basiconicum inside the food canal with a terminal pore (arrow)
Sensilla basiconica were also found inside the food canal, where they are shorter than on the outside (3.9 µm; SD ± 0.37; n = 4). They are scarce, and count only 11-13 per galea (n = 6). These sensilla are widely spaced in the proximal half of the proboscis and closer towards the tip. They have no socket and are composed of just the sensory cone, which bears a pore on the apex (Fig. 14).

Sensilla styloconica are restricted to the tip region. They are only found on the most distal 5% of the proboscis (Fig. 13). Their mean length is 14.2 µm (SD ± 1.7; n = 17). Compared to the other sensilla, the socket is long (7.9 µm; SD ± 0.93; n = 17) and massive, with four pikes surrounding it. The sensory cone is similarly long with 6.2 µm (SD ± 0.87; n = 17) and has a pore on the apex. There are 9-10 sensilla styloconica per galea; they occur in the same pattern on each galea. A group of 7-8 sensilla are found close together at the apex. Another two sensilla are situated in a row besides the dorsal legulae (Fig. 15).

Fig. 15 Arrangement of sensilla styloconica (ss) at the tip region of the proboscis; most of the sensilla are located near the apex, two sensilla are situated further along the proboscis besides the dorsal legulae (dl); the proboscis is covered in microtrichia till the tip; sb sensillum basiconicum, st sensillum trichodeum
The two galeae enclose the central food canal. In cross-section each galea contains a nerve, a trachea and several intrinsic galeal muscles (Fig. 16). The intrinsic galeal muscles can be divided into lateral intrinsic galeal muscles, which are arranged along the lateral proboscis wall, and median intrinsic galeal muscles, which are arranged longitudinally along the ventral wall. The nerve and the trachea are suspended at a longitudinal septum. Whereas the nerve proceeds to the tip, no trachea could be found in the distal region of the proboscis (Fig. 17).

![Figures 16-17](image)

**Figures 16-17**: Cross sections of the proboscis in different regions; **Fig. 16** Section near the proboscis base showing the prominent nerve (^n^), the trachea (^t^), and the intrinsic galeal muscles (^igm^); the ventral legulae (^vl^) are more tightly locked than the dorsal legulae (^dl^); **Fig. 17** Cross sections from middle to tip of the proboscis; the trachea is suspended at a septum (arrow); two series of muscles can be distinguished: the lateral intrinsic galeal muscles (^lim^), and the median intrinsic galeal muscles (^mim^); ^fc^ food canal; the smallest section near the proboscis tip pictured at the bottom shows the nerve (^n^) and distinct muscles (^m^), but no trachea;

### 3.4 Maxillary palpi

The maxillary palpi are 172.1 µm long (SD ± 24.4; n = 10) and consist of 3 segments (Fig. 18).

The first segment is short, tapering in the direction of the proboscis. This segment bears 2-4 sensilla trichodea, which are 48.5 µm long (SD ± 5.2; n = 7). They touch the microtrichia of the proboscis (Fig. 19).

The second segment is longer than the first one and has a triangular form. The third segment is oval-shaped and similarly long as the second. No sensilla were found on those segments.
There are several microtrichia on the first and on the second segment (Fig. 20). No scales were found on the first segment. A few scales occur on the second, whereas the third segment is covered in numerous scales except for the base.

**Figures 18-20:** Maxillary palpus, scales partially removed; **Fig. 18** Dorsal view showing 3-segmented maxillary palp; on the third segment, the sockets of the numerous scales can be seen (arrow); *sc* scales, *mi* microtrichia, *st* sensilla trichodea, *md* mandible; **Fig. 19** Lateral view of the first segment bearing sensilla trichodea (*st*) touching the base of the proboscis (*p*); **Fig. 20** Microtrichia (*mi*) on the second segment

### 3.5 Labium

The labium is reduced except for the prementum and the labial palpi. The prementum constitutes the ventral side of the head (Fig. 2). It forms a groove, which has numerous cuticular processes, where the proboscis is resting in its recoiled position.

### 3.6 Labial palpi

The labial palpi consist of three segments (Fig. 21). They measure 1.1 mm (SD ± 0.1; n = 6) in length. There is a right angle between the first and the second segment. The second and third segment stand in front of the head in a dorso-ventral position. The labial palpi are quite big compared to the head and surmount it (Fig. 1).

The first segment is short (296.8 µm; SD ± 34.7; n = 4), followed by a much longer second segment (575.9 µm; SD ± 22.1; n = 4), the third one is short again (235.5 µm; SD ± 46.7; n = 5) and rounded.
The labial palpi are entirely covered in scales, which extend from a round cuticular socket (Fig. 22). The scales are shorter and broader at the base and long and thin towards the tip. The outside of the labial palpi is more densely covered than the medial side facing the proboscis.

Figures 21-25: Labial palpus, scales have been removed; Fig. 21 Light microscopic picture showing 3-segmented labial palp on lateral side; po pit organ; Fig. 22 Second and third segment bearing sensilla chaetica (se) and sensilla trichodea (st); Fig. 23 Detailed view of third segment showing the location of the pit organ (po); sc scales; Fig. 24 Sensillum campaniformium (sa) on the first segment; Fig. 25 Detailed view of labial palp pit organ showing a sensillum chaeticum (se), several sensilla coeloconica (so) around the edge, leaf-shaped sensilla (sl) and sensilla campaniformia (arrow) deeper in the pit;

Four types of sensilla were found on the labial palpi, i.e., sensilla chaetica, sensilla trichodea, sensilla campaniformia and sensilla coeloconica inside the pit organ.

The sockets of the sensilla chaetica resemble those of the scales, but they are smaller and much tighter around the base of the sensilla (Fig. 22). The sensory bristles are long and striated. There are more sensilla on the second segment, with a
mean length of 68.2 µm (SD ± 11.42; n = 8). On the third segment there are few and shorter sensilla (60.9 µm; SD ± 6.27; n = 10). The standard deviation is pretty high, because of the different length of the sensilla on the inner and outer side of each labial palpus.

Sensilla campaniformia have a small socket with a dome shaped sensory cone barely surmounting it. They were found only on the inner side of the first segment (Fig. 24).

Some sensilla trichodea were found on the third segment near the apical pit organ (Fig. 22).

The labial palp pit organ is located in the middle of the inner side of the third segment (Figs. 21, 23). The pit is ovaly shaped with a length of 17.2 µm (SD ± 0.99; n = 5) and a width of 11.8 µm (SD ± 1.91; n = 5). It contains a single sensillum chaeticum with a length of about 15.6 µm, numerous shorter sensilla coeloconica and several leaf-shaped sensilla around the edges. Some sensilla campaniformia were present at the base of the pit (Fig. 25).
4. Discussion

Insect mouthparts show an amazing diversity of adaptations to their different feeding habits. One of the most formidable of these adaptations is the proboscis of Lepidoptera. With exception of the most basal families like Micropterigidae, in which adults have biting-chewing mouthparts (Kristensen, 1999), Lepidoptera have elongated galeae forming a coilable proboscis specialized to feed liquid nutrients (Krenn & Kristensen, 2000; Krenn, 2010). The lepidopteran proboscis is well-investigated in butterflies (e.g. Eastham & Eassa, 1955; Krenn, 1997; Krenn et al., 2001) and Noctuidae (e.g. Devitt & Smith, 1982; Banziger, 1982), only a few studies have been made about the outer morphology of the mouthparts in Pyralidae (Faucheux, 1991; 1995). The inner morphology of the proboscis was described superficial in the comparative study of proboscis musculature of Krenn and Kristensen (2004).

In this study the mouthparts of adult *P. interpunctella* (Pyralidae) were subjected to a detailed morphological investigation. The proboscis of *P. interpunctella* has the basic equipment of sensilla like other Glossata, it also shows the typical musculature of a lepidopteran proboscis, although the adults do not ingest nectar.

4.1 The basal parts

In basal lepidopteran families, the labrum is quite prominent (Faucheux, 2008), in higher Lepidoptera, where Pyralidae belong as well, it is highly reduced in size. The lateral lobes, the piliferes, bear long sensory bristles (Krenn & Kristensen, 2000).

As typical for the Glossata (Kristensen, 2003), the mandibles of *P. interpunctella* are small and rudimentary. They are non-functional and bear numerous cuticular processes.

The surface of the immovable basal mouthparts, the labrum and the mandibles, is striated, especially around the joints. This might be considered as protection against water and contamination from a functional point of view.
4.2 Maxillae and proboscis

The length of the proboscis of *P. interpunctella* corresponds to half of its body length. It approximates to the proboscis length of small butterflies (Kunte, 2007; Krenn, 2010).

In many taxa, the microtrichia covering the proboscis become less numerous and shorter towards the tip (Krenn & Kristensen, 2000). However, in *P. interpunctella* they keep the same size till the tip and become less numerous only when they are displaced by the sensilla styloconica near the apex. Büttiker et al. (1996) described a hemilachryphagous Pyralidae, *Pionea damastesalis*, which shows a resembling tip region to that of *P. interpunctella* and is covered with cuticular spines to the tip as well. At the base of the proboscis of *P. interpunctella*, the microtrichia are long and thin, they contact the sensilla of the piliferes as well as the sensilla of the first segment of the maxillary palpi. This contact might be important to determine the position of the proboscis relative to the head and to detect proboscis movements (Krenn, 1997; Krenn & Kristensen, 2000).

The dorsal legulae are less tight than the ventral legulae, since the linkages opened after the preparation procedure for SEM-analysis. In the distal part of the proboscis, the two rows of dorsal legulae melt to a single row. This region is specialized for fluid intake, the animals are able to ingest liquid through dorsal drinking slits between the legulae (Krenn, 2010). This so called tip region corresponds to 5-20% of the proboscis length in butterflies (Paulus & Krenn, 1996). In *P. interpunctella*, it amounts to 6.5% of the proboscis length.

The cuticular ribs forming the proboscis walls become narrower towards the tip as the proboscis gets thinner. Likewise the sensilla trichodea are shorter in the distal region of the proboscis. It is concluded that the thinner the proboscis becomes towards the tip, the cuticular structures become smaller and shorter accordingly.

4.3 Sensilla on the proboscis

Like in all studied Lepidoptera, sensilla trichodea are the most numerous sensilla on the proboscis of *P. interpunctella*, they occur throughout the entire proboscis-length.
No pores could be found on the bristles of either the sensilla trichodea, or the sensilla chaetica, which occur only at the base of the proboscis. Thus they are considered to be mechanosensitive in butterflies (Städler et al., 1974; Zacharuk, 1980; Krenn, 1998) and so it can be concluded for *P. interpunctella*.

In resting position of the proboscis, the coils are in close contact with each other. Under the assumption that sensilla trichodea function as mechanosensilla, they might provide information on the correct resting position from each coil to the other and to the labium like it was stated for butterflies (Krenn, 1990; 1997).

Sensilla basiconica occur only on the dorsal surface of the proboscis of *P. interpunctella*. They are less frequent than the sensilla trichodea throughout the proboscis. In *Vanessa cardui* (Nymphalidae), the sensilla basiconica on the outside of the proboscis as well as the ones inside the food canal, posses a single terminal pore. They both were considered to function as contact chemoreceptors (Krenn, 1998). Faucheux (1991, 1995) found multiporous sensilla basiconica on the surface of the proboscis of two species of Pyralidae, namely *Homoeosoma electellum* and *H. nebulella*. Faucheux (1991) suggested an olfactive function of these sensilla. On *H. electellum*, a second type of sensilla basiconica with only one apical pore was described. The sensilla in the food canal of both species are uniporous. These uniporous sensilla basiconica are considered to have gustative function (Städler et al., 1974). In *Plodia interpunctella*, both internal and external sensilla basiconica have been observed to possess an apical pore. However, if there were further wall pores, they could not be distinguished with SEM using the standard techniques of preparations.

Contrary to the external sensilla, the sensilla of the food canal of *P. interpunctella* have no socket, they just consist of the sensory cone. Functional reasoning suggests that a socket, which would enable deflection of the cone, is not necessary in the food canal, as these sensilla do not need to detect any mechanical forces. They just need to indicate, if liquid is coming through the food canal. Like in *Homeosoma nebuella* (Faucheux, 1991), abnormal forms with a flattened peg could be found in the food canal of *P. interpunctella*. The higher density of sensilla towards the tip could be explained by the fact, that the tip of the proboscis is the first to touch the liquid when feeding. As those sensilla might provide information on the flow rates in butterflies
(Krenn, 1998), the sensilla on the tip are the first to give this information. Although adult *P. interpunctella* are not nectar-feeding, the sensilla basiconica inside the food canal might be important for the detection of water flow through the proboscis.

Sensilla styloconica appear only in Lepidoptera (Krenn & Kristensen, 2000; Krenn et al., 2005). According to Zacharuk (1980) they are uniporous sensilla. In *P. interpunctella*, the sensilla styloconica seem to show only one terminal pore. The wall of the sensory cone appears to be smooth, no additional pores could be detected in the investigated sensilla. Due to their tubular body at the base of the cone and the apical pore, they might be considered as chemo- and mechanosensilla in butterflies (Krenn, 1998) as well as in Pyralidae. The clear arrangement of sensilla styloconica in the tip region is conspicuous, it is nearly identical in all investigated individuals. In *Pionea damastesalis*, the sensilla look quite similar to those of *P. interpunctella*. Even the arrangement on the proboscis tip shows similarities (Büttiker et al., 1996), despite the fact that this pyralid moth is regularly found on mammal eyes where it takes up lachrymal fluid. Also in *Homoeosoma electellum* the sensilla styloconica look resembling having pikes surrounding the socket (Faucheux, 1995). Another tear feeding Pyralidae, *Filodes mirificales* on the other hand, greatly differs in form and arrangement of the sensilla and the microtrichia on the proboscis (Büttiker et al., 1996). This suggests that the morphology and arrangement of the sensilla styloconica in representatives of Pyralidae rather is not related to the feeding preferences, like it has been shown in nymphaid butterflies (Krenn et al., 2001) and, at least, some tear-feeding or piercing blood-sucking Noctuidae (Büttiker et al., 1996; Zaspel et al., 2007).

### 4.4 Internal proboscis morphology

The proboscis musculature of *P. interpunctella* shows the regular composition of a Ditrysia. The presence of muscles during the entire proboscis length indicates that the proboscis is movable and is able to take up liquids. Although imagines do not feed on nectar, an intact proboscis including intrinsic galeal muscles is necessary for the intake of water.

In addition to muscles, each galea contains a nerve and a trachea. The nerve extends till the tip of the proboscis, which is very important for movements and sensing. The trachea does not reach to the tip, which is probably because the
proboscis is so thin at the tip, that oxygen transport through haemolymph is sufficient for oxygen supply of the tissue and no trachea is necessary.

4.5 Maxillary palpi

In the most basal groups of Lepidoptera, the maxillary palpi are composed of five segments. In most species of Ditrysia, the palpi are shorter and three-segmented (Kristensen 2003). In another pyralid species, *Homeosoma electellum*, the three-segmented maxillary palpi bear several sensilla on the first and a few on the second segment (Faucheux, 1995). *P. interpunctella* has a maximum of four sensilla on the first and none on the other segments.

4.6 Labial palpi

On the labial palpi of *P. interpunctella*, several types of sensilla were found. Sensilla chaetica and sensilla trichodea probably are machanosensitive (Zacharuk, 1980). Sensilla campaniformia, which were found on the first segment, are supposed to react to mechanical deformations of the cuticule (Keil, 1997). At the third segment of the labial palpi there is an assemblage of sensilla in a pit, the labial palp pit organ or vom Rath organ (Faucheaux, 1991; 2008; Kristensen, 2003). Usually, this pit organ is located on the tip of the third segment (Faucheaux, 1991; 2008; Krenn et al., 2004), in *P. interpunctella* however, it is located in the middle of the inner side. The labial palp pit organ is suggested to have olfactory function in Sphingidae (Kent et al., 1986). However, a functional role in feeding remains dubious in *P. interpunctella* when considering that only water is taken up in imagines.

In general, the proboscides of Lepidoptera, which do not feed as imagines, are more or less reduced in length and complexity (Scoble, 1992). Little is known about the morphology of mouthparts in species, in which imagines do not ingest food. It would be interesting to study non-feeding imagines of other related species to find out, if they have more reductions on their mouthparts than *P. interpunctella*.

Adults of *P. interpunctella* are considered to be non-feeding. However, even though it is not necessary for egg production, adults have been reported to be interested in fruit juice and sugar baits (Fasulo & Knox 1999) and able to take up water from wet
surfaces (Krenn, personal communication). This calls for further investigations on living individuals about the necessity of water supply for reproductive success. In behavioral studies one could observe, if and when they are feeding and if drinking water is necessary maybe at a certain temperature or humidity.
5. References


6. Abstract

The anatomy of the mouthparts and the types and distribution of sensilla on the mouthparts of *Plodia interpunctella* (Pyralidae) have been investigated using light microscopic and scanning electron microscopic techniques. The proboscis of *P. interpunctella* has the basic equipment of sensilla like other Glossata, it also shows the typical musculature of a lepidopteran proboscis, although the adults do not ingest nectar. There are four types of sensilla on the surface of the proboscis, another type was found inside the food canal. Bristle shaped sensilla chaetica occur only at the base of the proboscis. Sensilla trichodea are the most numerous of the sensilla, they are scattered all over the proboscis surface. Both bristle shaped types of sensilla are considered to be mechanosensitive, they appear to provide tactile information on the correct resting position of the proboscis. Cone shaped sensilla basiconica occur in a single row near the dorsal legulae. In the median food canal, there is one longitudinal row of a few sensilla basiconica. Appearing uniporous inside and outside the food canal, they are considered to have chemosensory function. Sensilla styloconica are situated only near the tip of the proboscis, where they occur in clear patterns. Having a tubular socket and an apical pore, they are regarded to be both mechano- and chemosensitive. The comparison to other representatives of Pyralidae having various feeding habits, suggests that the sensilla equipment and arrangement is not related to the feeding preferences. On the labial palpi, the pit organ was found in the middle of the third segment, having numerous different sensilla such as sensilla chaetica, coeloconica and campaniformia, which are considered to have olfactory function, although, probably, not used for the locating of food sources in *P. interpunctella*. 
7. Zusammenfassung

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