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Autodetection and chemistry of female and male pheromone in both sexes of the tiger moth *Panaxia quadripunctaria*

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Abstract Female moths of *Panaxia quadripunctaria* PODA (Lepidoptera, Arctiidae), produce (*Z*,*Z*)-6,9-heneicosadiene (I) as the major and (*Z*,*Z*)-6,9-eicosadiene (II) as the minor component of a putative pheromone. Related compounds occur in trace amounts. The abdominal scent glands contain 5–10 μg of (I) and 50–100 ng of (II). Recordings of electroantennogram (EAG) responses to (I), (II), and to female glands are of equal amplitude in both sexes. Females are thus capable of pheromone autodetection in contrast to the majority of moths where females are considered to be anosmic for their own attractant. The EAG threshold to (I) was below 1 ng at the odour source. The odour of the male scent gland (corema) elicited significant EAGs in both sexes. The chemical contents of coremata varied with the provenience of the moths. A variety of ethyl esters was always found, yet hydroxydanaidal (up to 20 μg/corema) and traces of danaidal, only in some samples. All these scents might be components of a male pheromone. Peculiar scent scales on the coremata are exposed during the extrusion. Antennae of both sexes have similar inventories of trichoid sensilla.

Key words Arctiidae · Pheromones · Autodetection · Hydroxydanaidal · Danaidal

Abbreviations *EAG* electroantennogram · *SEM* scanning electron microscope · (*I*) (*Z*,*Z*)-6,9-heneicosadiene · (*II*) (*Z*,*Z*)-6,9-eicosadiene. (*III*) (*Z*,*Z*,*Z*)-3,6,9-heneicosatriene · *PA* pyrrolizidine alkaloid · *HOdal* hydroxydanaidal

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Introduction

Chemical communication in the sex life of moths usually begins with the emission of an odorous attractant by the female. Her male partner is the receiver with his stimulus-specific olfactory receptor neurons on the antenna (Schneider 1992). In the first species in which these relations were studied electrophysiologically, the silkmoth *Bombyx mori*, it appeared that the females were unable to smell their own odour signal (Schneider 1957). This and further observations led to the assumption of a general anosmia of female moths to their own attractant. However, exceptions to this rule were later observed (see Ljungberg et al. 1993 for references) and recently named "autodetection" (Ochieng et al. 1995).

Here we report on the first case of autodetection in a female arctiid moth. Although there is little doubt that the volatiles which the female produces in her abdominal glands act as pheromones, we consider this term to be still tentative before behaviour tests have been carried out. Both sexes possess tubular abdominal scent glands. Female glands are never everted, but dissipate their volatiles by abdominal pumping (see Lenau-Jürgens 1971 for *Panaxia*), in contrast to the large male glands – the coremata (Birch 1979). As such organs in other Lepidoptera emit volatiles with pheromonal functions, we attempted to identify the expected compounds and recorded olfactory responses from the antennae of both sexes.

Materials and methods

Experimental animals

Panaxia (or *Euplagia* or *Callimorpha*) *quadripunctaria* PODA is one of the larger species of the Arctiidae with a wingspan of more than 40 mm and bright, probably aposematic, coloration. It ranges from the south of England over large parts of Europe to western Asia

and is known for its habit of assembling in great numbers (up to 700/m^2) in moist places during summer months on two Greek islands, Rhodes (Walker 1966; Elger 1969; Lenau-Jürgens 1971) and Paros (D. Schneider, unpublished observations).

In 1993 we observed *Panaxia* at Paros and raised specimens from their eggs. In 1994 we bought larvae from France (Provence), observed the moths again at Paros in 1997 (preserved their glands and bodies) and later tested a few males from SW Germany (henceforth called "local"). Courtship has never been observed although captive moths successfully mated (Walker 1966; Elger 1969; Lenau-Jürgens 1971). In the laboratory we found that the larvae accepted a number of different foodplants. Given a choice, they preferred cultivated rape, *Brassica napus* (which is poor in glucosinolates), over wild rape, followed by *Taraxacum officinale*. The larvae also accepted (without special preference) some plants containing pyrrolizidine alkaloids such as *Senecio vulgaris* and *S. fuchsii*.

Morphology

Isolated abdomina of both sexes of *Panaxia* were ligated proximally with surgical clamps. The tubular glands were evaginated (extrused) by pressing air into the abdomen with a syringe. This simulates the natural evagination in males but not in females, since they never expose their glands when calling. The abdomina were then ligated again close to the respective glands and the proximal tissue discarded. The female glands were filled with haemolymph; the male glands contained air. Glands could now be handled easily for morphological inspection or use as an odour source. After drying, mounting and gold sputtering we studied the coremata and the antennae of both sexes with scanning electron microscopes (SEM: Zeiss Novoscan 30, 15 kV and Jeol 6300 F).

Chemistry

Extruded glands of both sexes were cut from the abdomina and kept in pentane at −20 °C until chemically analysed. Bodies of both sexes were preserved in ethanol or pentane and studied for pyrrolizidine alkaloid (PA) content. Mass spectra (70 eV) were obtained with a VG 70/250S mass spectrometer coupled to a Hewlett-Packard HP 5890A gas chromatograph and with a Fisons MD 800 mass spectrometer coupled to a Fisons GC 8000. Gas chromatographic analyses were performed with a Carlo-Erba Fractovap 2101 gas chromatograph, equipped with a flame ionization detector and on-column injection. Separations were carried out using 30 m Rt_x-5 (i.d. = 0.32 mm, $d_f = 0.25\,\mu m$) and 30 m DB-FFAP (i.d. = 0.32 mm, $d_f = 0.25\,\mu m$) fused silica columns with hydrogen as the carrier gas. Identifications are based on comparison of mass spectra and gas chromatographic retention times with those of authentic samples. (*Z*,*Z*)-6,9-heneicosadiene (I) and (*Z*,*Z*)-6,9-eicosadiene (II) (Fig. 3) were synthetized by standard procedures (Yu et al. 1989) as follows: ethyl linoleate was reduced with $LiAlH_4$ to linolenyl alcohol. After conversion to the paratoluol sulfonate, the product was coupled with an appropriate lithium diakyl cuprate to yield (I) or (II), respectively. The triene (III) was prepared using the same transformation starting from ethyl linoleate. The purities of these synthetic polyenes were higher than 98%.

Electrophysiology

Electroantennograms (EAG) were recorded from isolated antennae using standard methods (Boeckh et al. 1965). Amplitudes of recorded calibration pulses were unchanged over the experiments of ca. 1 h, indicating a stable ohmic resistance of the mounted antennae. Odour sources were pheromone glands of both sexes or filter papers impregnated with given amounts of the respective female pheromone components (solved in pentane). The odour sources, or blank papers for control, were mounted in short glass tubes through which air puffs were blown onto the antennae. In 1993, we tested only freshly expanded glands and in 1994 in addition the synthetic components (I), (II), (III). In 1997, we EAG-tested a few local males with HOdal and substance (I).

Behaviour

In 1997 we field-tested *Panaxia* in Paros with the synthetic female and male components (not with volatile PA derivatives) and later tested caged local males in addition with HOdal.

Results

Morphology

Female gland

The two separate tubular gland systems belong to the dorsolateral VIII/IX intersegmental membrane of the abdomen; their size and shape vary. Some have two, others three or even four branches of ca. 3 mm in length and 0.3 mm in diameter (see similar glands in Conner et al. 1980, Wunderer et al. 1986, Schneider et al. 1992a).

Male gland

The coremata are bifid tubular glands and part of the ventral VII/VIII intersegmental membrane of the abdomen (Fig. 1a, b). They carry a central field of dark, longitudinal scales (Fig. 1a, c) and short, yellow, triangular scales, arranged on the outer edge of folds viewed as fine ribs in the everted organ (Fig. 1b). In the retracted state it appears that this arrangement of the short scales allows the close packing of the coremata (see tip zones in Fig. 1c). The wall of the fully expanded, air-filled organ is transparent, like parchment (Fig. 1b). During the process of eversion of the corema, each side of the organ extends from a resting length of 1.0–1.4 mm through a medium length of 3.5 mm (Fig. 1c) to its full length of 5–6 mm (Fig. 1b).

The ca. 200 short scales have a distal width of 100–160 μm, an equal height (Fig. 2a, b) and very different outer and inner sides. The former (facing toward the outer ends of the organ) appear rather smooth, while the latter have several prominent keels. High-power SEM of the outer sides reveals a complex surface structure, yet no visible pores of over 10 nm diameter (Fig. 2c). The inner sides have many holes of variable shape and width (Fig. 2d). Interestingly, it is on the inner sides of chemically untreated scales that we observed a "contamination", probably a residue of organic substances (upper right corner of Fig. 2d). The 80–100 dark, bilaterally symmetrical central scales are 0.7–1.0 mm in length and 0.12–0.20 mm in width (Fig. 1c).

Antennae

The filiform antennae showed no macroscopic sexual difference in shape and dimension of the antennal flagellum. The density and form of the curved, presumably attractant-sensitive sensilla (trichodea), does not differ between the sexes. These sensilla are 30–40 μm long.

Chemistry

Volatile components of the female glands

The GC-MS analysis of Paros females showed the presence of long-chain polyenes (Fig. 3). These were identified to be (*Z*,*Z*)-6,9-heneicosadiene (I) (about 5 μg/female) as the main component, accompanied by small amounts (less than 1%) of (*Z*,*Z*)-6,9-eicosadiene (II). Trace constituents were (*Z*,*Z*,*Z*)-3,6,9-heneicosatriene (III) (*Z*,*Z*)-6,9-docosadiene (IV) (*Z*,*Z*)-6,9-tricosadiene (V) and another triene, tentatively identified to be (*Z*,*Z*)-6,9,20-heneicosatriene (VI). Females from France contained almost pure (I) at a concentration of 5–10 μg/female. The only trace constituent present in these latter females was (III) (less than 0.1%).

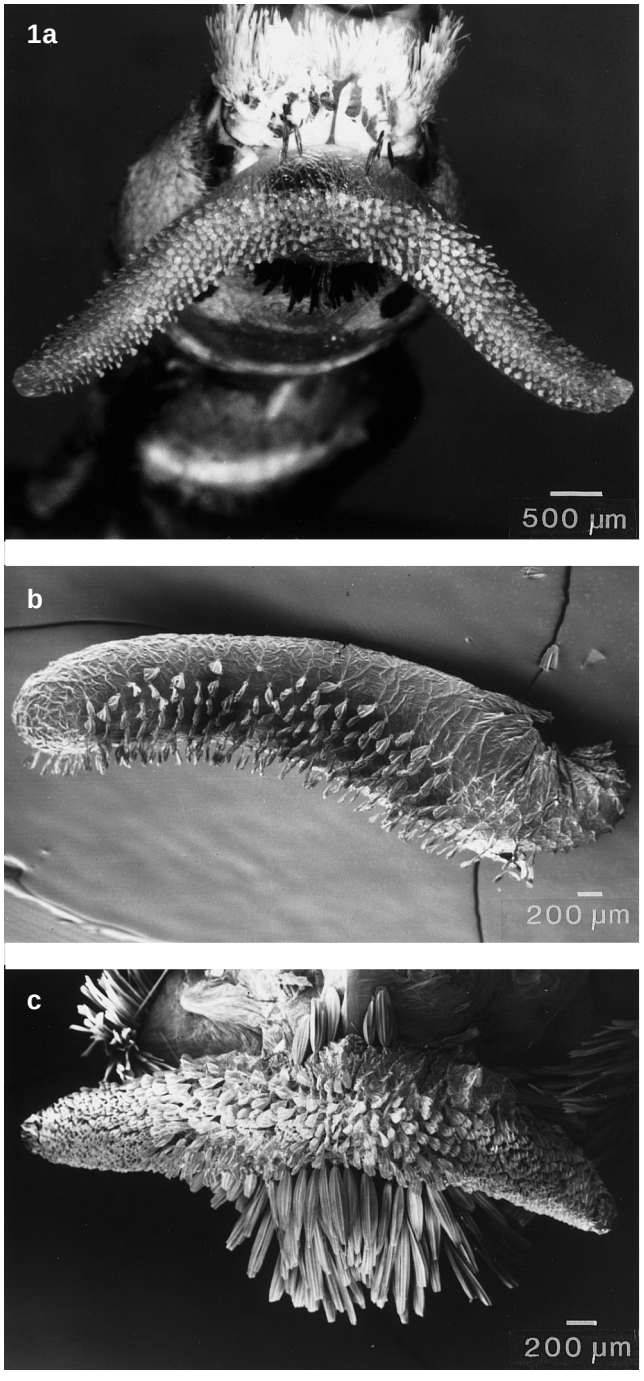

Fig. 1a–c *Panaxia* male odour glands, coremata: **a** fully everted organ; **b** right half of the organ, fully everted (SEM); **c** partially everted organ (SEM)

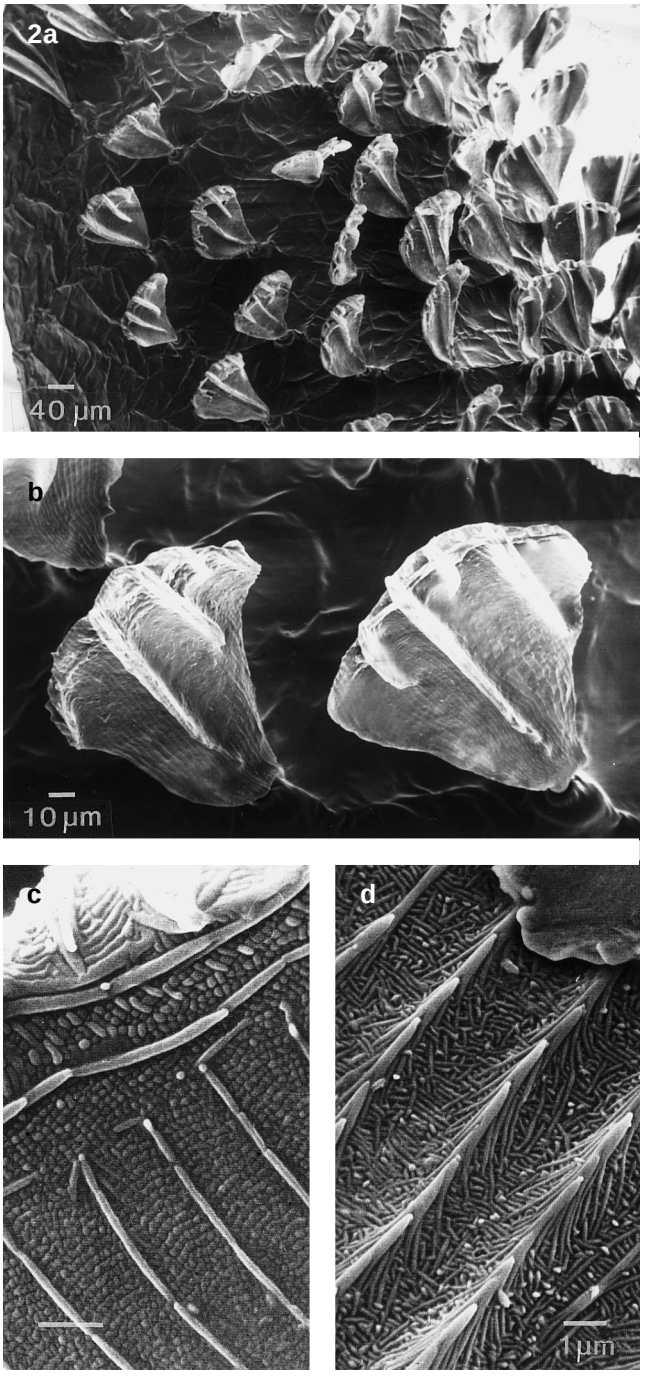

Fig. 2a–d *Panaxia* male odour gland; SEMs of scent scales: **a, b** triangular scales, inner sides with ribs; **c** outer side; **d** Inner side with precipitate on its top-right area

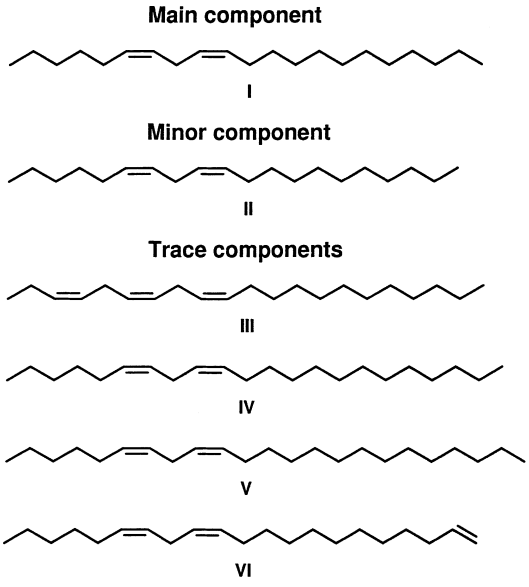

Fig. 3 Components of the female gland odour: I (*Z*,*Z*)-6,9-heneicosadiene; II (*Z*,*Z*)-6,9-eicosadiene; III (*Z*,*Z*,*Z*)-3,6,9-heneicosatriene; IV (*Z*,*Z*)-6,9-docosadiene; V (*Z*,*Z*)-6,9-tricosadiene; VI (*Z*,*Z*)-6,9,20-heneicosatriene

Volatile components of the male glands

Freshly everted coremata of the 1997 Paros males and of the local males surprised us by a (quickly fading) "green" smell which could still be perceived from the extracts of the glands. In all Paros samples (1993 and 1997) a range of ethyl esters was present. Main constituents were ethyl esters of ubiquitous acids, namely ethyl oleate, ethyl linoleate, and ethyl palmitate. Minor constituents were ethyl stearate, ethyl hexadecenoate, some usual hydrocarbons like tricosane or pentacosane, and the steroid cholesterol. Samples from France were similar, but contained much fewer ethyl esters. The main constituent of these glands was cholesterol. In 1997, different from all our analyses before, the coremata from Paros and of the local males contained ca. 20 µg and 1.5 µg, respectively, of HOdal/male and traces of danaidal. The bodies of both sexes of these specimens contained a variety of PAs.

Electroantennograms

Recordings were taken from 12 antennae in 1993. Eight of these experiments are shown in Table 1, the remaining four had to be excluded for technical reasons. Male and female antennae, when stimulated with female gland odour, exhibited similar responses with an amplitude more than five times higher than in response to the control stimulus. The amplitudes of the EAG responses to the odour of the male glands were about one-half of the amplitude evoked by female glands but in most experiments still well above control. The stimulus strength of the male glands varied more than that of the female

Table 1 Overview of eight representative EAG recordings from *Panaxia M* Male, *F* female (listed sequence of use), *C* control stimulus. Male glands 1–8 (sequence of dissection), were either fresh or kept deep frozen for a few days between experiments; coremata with the same number were first measured intact and then (marked[a]) after squeezing with forceps. Glands were expanded in the stimulatory apparatus. The figures shown are EAG amplitudes to single stimuli or the means of several stimuli $n(x)$. EAG amplitudes in mV. Gland odour was tested at random

No. Ant	Odour stimuli															
	Male glands (coremata)														Female glands	
	C	1[a]	2	2[a]	3	3[a]	4	4[a]	5	5[a]	6	7	8	8[a]	1	2
M1	0.18 (10)	0.40 (3)	0.43 (5)	0.43 (2)	0.19 (6)	0.68 (3)									0.70 (2)	
M8	0.05 (8)	0.12 (1)		0.17 (2)		0.28 (3)	0.27 (1)	0.23 (3)	0.08 (3)		0.12 (1)				0.64 (3)	0.75 (1)
M9	0.03 (2)			0.06 (2)		0.34 (1)		0.09 (2)	0.06 (1)						1.36 (1)	
M12	0.05 (2)	0.12 (1)		0.46 (2)		0.60 (1)		0.08 (1)		0.16 (1)	0.24 (1)	0.16 (2)	0.16 (1)	0.26 (4)	0.58 (2)	0.56 (3)
F6	0 (3)	0.39 (2)		0.59 (3)		0.55 (2)									0.88 (1)	0.77 (1)
F7	0.12 (2)	0.38 (2)		0.31 (2)		0.50 (1)									0.75 (1)	0.75 (1)
F10	0.11 (2)	0.22 (1)		0.33 (1)		0.47 (3)		0.11 (1)	0.37 (1)		0.30 (12)				0.67 (1)	0.73 (1)
F11	0.14 (3)	0.40 (2)		0.38 (2)		0.50 (2)		0.38 (1)	0.21 (2)	0.42 (1)	0.17 (1)		0.50 (1)		0.54 (1)	0.71 (4)

[a] Coremata after squeezing with forceps

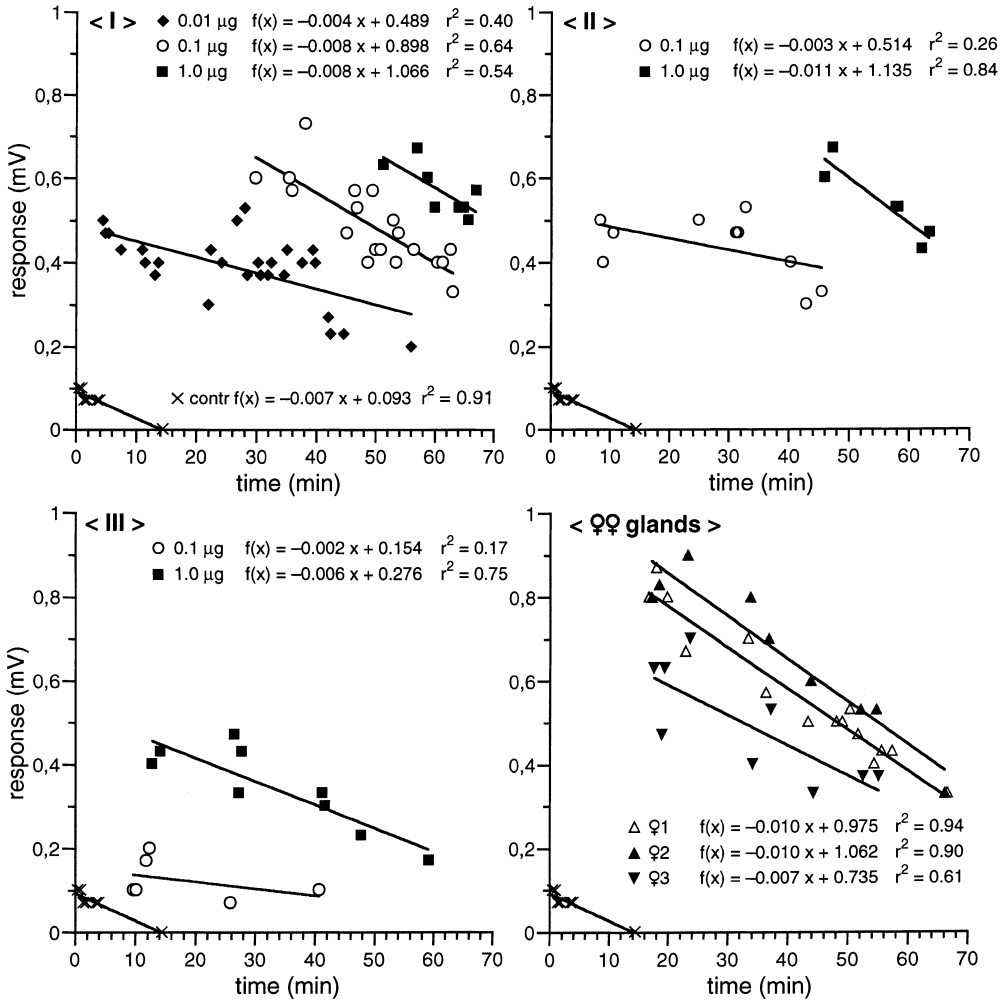

Fig. 4 EAG responses of a male antenna to female gland components (I), (II), (III) in different amounts (in µg on the odour source) and to female gland odour. *x*: control air-puff. Ordinates: EAG amplitudes in mV. The responses to the four types of odour stimuli are shown as realtime events during the 70-min experiment. Weak stimuli were applied in the early phase of the experiment, stronger ones later. The *symbols* show the moment when the measurement was taken. The abscissa shows the common progress of the experiment. Stimulus intervals were ca. 1 min

glands. In five cases we squeezed the expanded corema before or after the initial EAG. In three cases (no. 3/3[a], 5/5[a], 8/8[a]), the EAG amplitude was higher after squeezing.

In the 1994 EAG recordings, we tested female glands and components (I), (II) and (III). The two experiments shown here (Figs. 4, 5) are representatives out of a total of seven. The remaining five were in agreement with the results shown in Figs. 4 and 5, and all experiments thus corroborate the 1993 observation of equal sensitivity of both male and female antennae to the female gland odour (Table 1). Component (I) is chemically and physiologically prominent, followed by (II) and (III) with ca. 1/2 and 1/4 of the EAG amplitudes, or approximately 1/10 and 1/100 of the stimulus strength. The decline of the EAG amplitudes in the course of the experiment is independent of the type of stimulus and is also seen in the control EAGs (note the real time abscissa).

In another experiment (not shown) which lasted 30 min, we tested the EAG threshold for substance (I). The responses indicate that the level of the EAG threshold is below 1 ng on the odour source. The mean amplitude of 32 control stimuli was 0.015 mV (SD 0.019); mean amplitude of 26 test stimuli 1.0 ng of (I): 0.088 mV (SD 0.038 mV).

In 1997 we tested three antennae of three local males with 100 µg of HOdal at the evaporation site (comparable to the content of 5 coremata) and elicited EAGs of two times the control amplitude. Female components were as effective as before.

Behaviour

In June 1997 at Paros, we offered the just aggregating moths the components (I, II, III) and corema-esters (not yet the volatile pyrrolizidine derivatives) in different concentrations but observed no reaction at daytime or early evening (before sunset). Later, we tested local, caged males with female components and HOdal, again without clear response. At the assembling place at Paros, *Panaxia* is quite active during the daytime (avoiding direct sunlight, drinking, nectaring). Our local caged males, tended vigorously to escape towards the lighted side at dusk, independent of the air current and the gland odours.

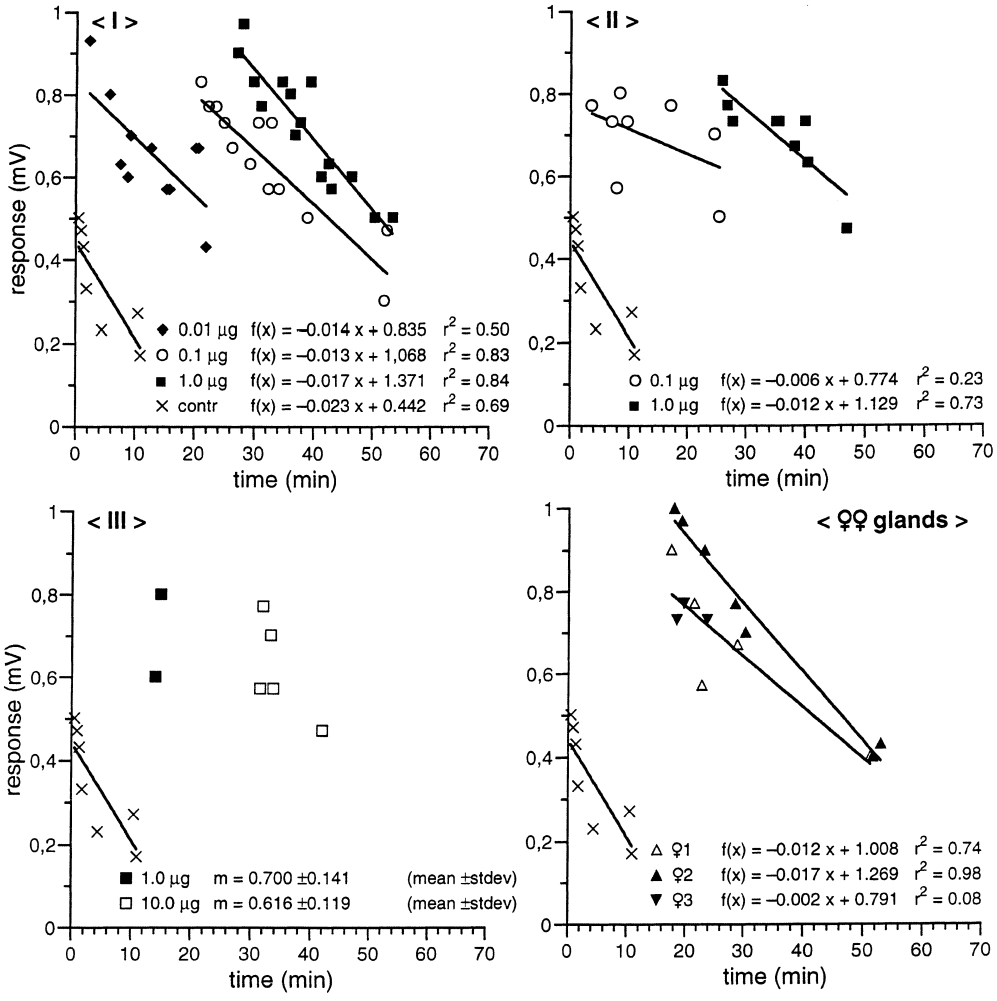

Fig. 5 EAG responses of a female antenna as described in Fig. 4

Discussion

Panaxia

The male scent gland of *Panaxia* was first mentioned and depicted by Birch (1979) and Birch et al. (1990). Its peculiar scales differ from the delicately structured, hairlike odour-emitting scales found on the scent organs of other male Lepidoptera (Wunderer et al. 1986; Boppré and Schneider 1989; Boppré and Vane-Wright 1989; Schneider et al. 1992b). The position of the *Panaxia* scales on the outer edges of the folds (cf. *Creatonotos*; Egelhaaf et al. 1992) allows optimal packing when the organ is deflated and withdrawn into its intersegmental pocket.

It is likely that only the corema scales carry and emit the scent. This is supported by the fact that the inner sides of the short scales not only have the expected complex perforations of scent-emitting scales but seem to be smeared by material which is not yet characterized with respect to its chemical nature. It may be a residue of the chemically identified ethyl esters and their "cosmetic formulation". So far, all our chemical findings have been based on extractions of whole coremata.

The new finding of PAs in the bodies of both sexes and of HOdal and danaidal in the coremata, corresponds to related observations in other arctiids which use HOdal as a male pheromone. To produce HOdal, larvae need to feed on plants which contain the PAs, store them also for their own protection and eventually use them for the pheromone biosynthesis (see Eisner and Meinwald 1987, Boppré 1990, Schneider 1992 for ref.). The lack of PAs and HOdal in our earlier samples must be due to a PA-free food of the polyphagous larvae. An interpretation of the complex and varying composition of the male corema-odourants has to await behaviour studies.

The female glands of *Panaxia* show structural and chemical features similar to those known from other arctiids (Conner et al. 1980; Wunderer et al. 1986; Percy-Cunningham and MacDonald 1987; Wheatherston and Percy 1977; Krasnoff and Roelofs 1990; Yin et al. 1991; Arn et al. 1992). There is little doubt that the components (I) and (II) are the crucial elements of the female attractant scent. Arguments in favour of this claim are:

1. They display great similarity to related long-chain C20–23 polyenes shown to act as pheromones in females of other arctiids (Arn et al. 1992).

2. Large EAGs and low EAG threshold indicate that they are biologically important compounds.

3. Behaviour observations with late season males and females in separate neighbouring cages showed female abdominal pumping and male attempts to reach them (Lenau-Jürgens 1971).

Specimens from France contained mainly substance (I) (5–10 μg/moth). This high absolute value puts *Panaxia* females among the moths producing the largest amounts of pheromone [see Steinbrecht (1964) for *Bombyx* (1.5 μg) and Schal et al. (1987) for the arctiid *Holomelina* (5–10 μg)].

Interestingly, we found that the females in both Paros samples (August 1993 and June 1977) produced the tentative pheromone components (I) and (II), but only the late-season 1993 females carried ripe and inseminated eggs. If reproduction is a late summer affair in the Paros populations (cf. Walker 1966, Elger 1969, Lenau-Jürgens 1971 for Rhodes), we wonder about the seasonally early production of pheromonal odourants in both sexes.

Autodetection

Female antennae of most moth species are apparently anosmic to their own odour while autodetection of female pheromones is a less frequently observed phenomenon. *Panaxia* is an additional exception to the rule. In contrast to the females, males of Lepidoptera were never found to be anosmic for their own pheromones (Schneider and Seibt 1969; Grant 1970, 1971; Grant et al. 1972). Pheromone anosmia of female moths, such as the Saturniidae, is often paralleled by a striking sexual dimorphism in which female antennae are smaller and less complex than those of the males and/or lack the long pheromone-sensitive hair sensilla (Boeckh et al. 1960; Schneider et al. 1964).

Autodetection of female pheromone was earlier found in the Noctuidae, Tortricidae and Yponomeutidae (Nesbitt et al. 1973; Birch 1977; Den Otter et al. 1978; Palaniswamy and Seabrook 1978a,b; van der Pers and den Otter 1978; Ross et al. 1979; Saad and Scott 1981; Seabrock et al. 1987; Ljungberg et al. 1993; Den Otter et al. 1996). Receptor cells of females with the capability of autodetection are often less sensitive to the odour than the respective cells of the males. In *Spodoptera littoralis*, both sexes share one type of receptor cells with the same high and specific sensitivity to one of the female pheromone components while the other type, which responds to another component, is only present on male antennae. Interestingly, most moth species with autodetection in the female do not show a substantial sexual dimorphism of their antennae (Ljungberg et al. 1993; Ochieng et al. 1994; Anton and Hansson 1994). To date, there are too few cases of female autodetection to generally relate these physiological facts to the absence, presence or extent of antennal dimorphism. The complexity of the situation is shown by *Bombyx*: females are pheromone anosmic, but dimorphism of the big antennae is limited (Steinbrecht 1970).

Anosmia, in the context, is understood as the lack of a behavioural response and the lack of an EAG. EAG reactions are indicative of a large number of similarly reacting receptor cells. Pheromone "anosmic" female antennae could thus carry small numbers of "autodetective" cells which would not show in an EAG. In females of *Bombyx*, the immunocytochemical detection of pheromone-binding protein in some medium-sized sensilla trichodea and s. basiconica points in this direction, but test recordings from individual receptor cells are needed to confirm this view (Steinbrecht et al. 1995). If the *Bombyx* female does possess such cells, pheromone anosmia in female moths would not be a qualitative but rather a quantitative sexual distinction (R. A. Steinbrecht, personal communication).

Pheromone anosmic female moths do not have the male-specific macroglomerular complex in the olfactory brain where the incoming signals of the female odour receptors are processed (*Antheraea*, Boeckh and Boeckh 1979; *Bombyx* and *Lymantria*, Koontz and Schneider 1987; *Manduca*, Hildebrand 1996). The brain of the female *Spodoptera*, a species with female autodetection, has a special glomerulus of this kind (Anton and Hansson 1994).

Central nervous processing of messages of male pheromones has never been studied and little is known of the respective morphological substrate. In an indicative histological note on the olfactory brain of *Creatonotos* (with its huge coremata and corresponding pheromone production) and the wax moth *Galleria* (with an attractant produced by the wing glands of the male), large glomeruli, potentially specialized to pheromones, were found in both sexes (Schneider and Wunderer 1990).

At present, we can only speculate about the biological meaning of a (total or only partial) anosmia or autodetection. For autodetection, the following functions have been discussed: (1) establishment of "social contacts" among females such as lek formation (sensu Alexander 1975), joint calling, or spacing on food plants (den Otter et al. 1978, 1996), (2) control of timing of pheromone release (Palaniswamy and Seabrook 1985), (3) spacing to avoid interference of pheromone plumes (Anonymous 1997). We now consider that *Panaxia* (which lives several months in aggregation on Rhodes and Paros) might profit from autodetection during gregarious activities. Finally, sensitivity of the autodetective systems of female moths is not necessarily stable since it can be modulated by juvenile hormone, supposedly in relation to the mating and egglaying, behaviour (Palaniswamy et al. 1979).

Acknowledgements Dr. Ernst Priesner went missing whilst conducting pheromone field studies in the Bavarian Alps soon after he

had performed the second series of *Panaxia* EAG recordings for this study. He was never found. This paper will be the last of many in which Ernst Priesner reported on moth pheromones, a field which he mastered like few others in the world. We lost a colleague who shared the interests of one of us (D.S.) for over 30 years. The authors thank Drs. K. E. Kaissling, T. Keil, R. A. Steinbrecht, L. Williams and an anonymous reviewer for critical and constructive advice. J. Berz, C. Bock, G. Brehm G. Laplanche, E. Roth, C. Schmid and H. Söchting-Mayr gave technical assistance. Our experiments comply with the US and German rules for animal care.

References

Alexander RD (1975) Natural selection in specialized chorussing behavior in acoustical insects. In: Pimentel D (ed) Insects, science, and society. Academic Press, New York, pp 35–77

Anton S, Hansson BS (1994) Central processing of sex pheromone, host odour, and oviposition deterrent information by interneurones in the antennal lobe of female *Spodoptera littoralis* (Lepidoptera: Noctuidae). J Comp Neurol 350: 199–214

Arn H, Tóth M, Priesner E (1992) List of sex pheromones of Lepidoptera and related attractants, 2nd edn. Int Org Biol Control. Montfavet, Switzerland

Birch MC (1977) Responses of both sexes of *Trichoplusia ni* (Lepidoptera, Noctuidae) to virgin females and synthetic pheromone. Ecol Entomol 2: 99–104

Birch MC (1979) Eversible structures. In: Heath J, Emmet AM (eds) Moths and butterflies of Great Britain and Ireland. Curven, London, pp 9–18

Birch MC, Poppy GM, Baker TC (1990) Scents and eversible scent structures of male moths. Annu Rev Entomol 35: 25–58

Boeckh J, Boeckh V (1979) Threshold and odor specificity of pheromone sensitive neurons in the deutocerebrum of *Antheraea pernyi* and *A. polyphemus* (Saturnidae). J Comp Physiol 132: 235–242

Boeckh J, Kaissling KE, Schneider D (1960) Sensillen und Bau der Antennengeißel von *Telea polyphemus* (Vergleiche mit weiteren Saturniden: *Antheraea*, *Platysamia*, und *Philosamia*). Zool Jahrb Ant Ontogen 78: 559–584

Boeckh J, Kaissling KE, Schneider D (1965) Insect olfactory receptors. Cold Spring Harbor Symp Quant Biol 30: 265–280

Boppré M (1990) Lepidoptera and pyrrolizidine alkaloids. J Chem Ecol 16: 165–185

Boppré M, Schneider D (1989) The biology of *Creatonotos* (Lepidoptera: Arctiidae) with special reference to the androconial system. Zool J Linn Soc 96: 339–356

Boppré M, Vane-Wright RI (1989) Androconial systems in danainae (Lepidoptera): functional morphology of *Amauris*, *Danaus*, *Tirumala* and *Euploea*. Zool J Linn Soc 97: 101–133

Conner WE, Eisner T, Van der Meer RK, Guerrero A, Meinwald J (1980) Sex attractant of an arctiid moth (*Utetheisa ornatrix*): a pulsed chemical signal. Behav Ecol Sociobiol 7: 55–63

Den Otter CJ, Schuil HA, Sander-van Oosten A (1978) Reception of host plant odor and female sex pheromone on *Adoxophyes orana* (Lepidoptera: Tortricidae): electrophysiology, and morphology. Entomol Exp Appl 34: 370–378

Den Otter CJ, De Cristofaro A, Voskamp KE, Rotundo G (1996) Electrophysiological and behavioural responses of chestnut moths, *Clydia fagiglandana* and *C. plendana* (Lep., Tortricidae) to sex attractants and odours of host plants. J Appl Entomol 120: 413–421

Egelhaaf A, Rick-Wagner S, Schneider D (1992) Development of the male scent organ of *Creatonotos transiens* (Lepidoptera, Arctiidae) during matamorphosis. Zoomorphology 111: 125–139

Eisner T, Meinwald J (1986) Alkaloid-derived pheromones and sexual selection in Lepidoptera. In: Prestwitch GD, Blomquist GJ (eds) Pheromone biochemistry. Academic Press, Orlando FL, pp 251–269

Elger R (1969) Freilandstudien zur Biologie and Ökologie von *Panaxia quadripunctaria* (Lepidoptera, Arctiidae) auf der Insel Rhodos. Oecologia 2: 162–197

Grant GG (1970) Evidence for a male sex pheromone in the noctuid, *Trichoplusia ni*. Nature 227: 1345–1346

Grant GG (1971) Electroantennogram responses to the scent brush secretions of several male moths. Ann Entomol Soc Am 64: 1428–1431

Grant GG, Brady UE, Brand JM (1972) Male armyworm scent brush secretion: identification and electroantennogram study of major compounds. Ann Entomol Soc Am 65: 1224–1227

Hildebrand JG (1996) Olfactory control of behavior in moths: central processing of odor information and functional significance of olfactory glomeruli. J Comp Physiol A 178: 5–19

Koontz MA, Schneider D (1987) Sexual dimorphism in neuronal projections from the antennae of silk moths (*Bombyx mori*, *Antheraea polyphemus*) and the gypsy moth (*Lymantria dispar*). Cell Tissue Res 249: 39–50

Krasnoff SB, Roelofs WL (1990) Evolutionary trends in the male pheromone systems of arctiid moths: evidence from studies of courtship in *Phragmatobia fuliginosa* and *Pyrrharctia isabella* (Lepidoptera: Arctiidae). Zool J Linn Soc 99: 319–338

Lenau-Jürgens H (1971) Untersuchungen zum Migrationsverhalten von *Panaxia quadripunctaria* PODA (Lepidoptera, Arctiidae) auf der Insel Rhodos. Forma Functio 4: 1–45

Ljungberg H, Anderson P, Hansson BS (1993) Physiology and morphology of pheromone-specific sensilla on the antennae of male and female *Spodoptera littoralis* (Lepidoptera: Noctuidae). J Insect Physiol 39: 235–260

Nesbitt BF, Beevor PS, Cole RA, Lester R, Poppi RG (1973) Sex pheromone of two noctuid moths. Nature 244: 208–209

Ochieng' SA, Anderson P, Hansson BS (1995) Antennal lobe projection patterns of olfactory receptor neurons involved in sex pheromone detection in *Spodoptera littoralis* (Lepidoptera: Noctuidae). Tissue Cell 27: 221–232

Palaniswamy P, Seabrook WD (1978a) Behavioral responses of the female eastern spruce budworm *Choristoneura fumiferana* (Lepidoptera, Tortricidae): electrophysiology and morphology. Entomol Exp Appl 24: 570–578

Palaniswamy P, Seabrook WD (1978b) Behavioral response of the eastern spruce budworm, *Choristoneura fumiferana* (Lepidoptera, Tortricidae) to the sex pheromone of her own species. J Chem Ecol 4: 649–655

Palaniswamy P, Seabrook WD (1985) The alteration of calling behavior by female *Chroristoneura fumiferana* when exposed to synthetic sex pheromone. Entomol Exp Appl 37: 13–16

Palaniswamy P, Sibasubramanian P, Seabrook WD (1979) Modulation of sex pheromone perception in female moths of the eastern spruce budworm, *Choristoneura fumiferana* (Lepidoptera/Tortricidae) by altosid. J Insect Physiol 25: 571–574

Percy-Cunningham JE, MacDonald JA (1987) Biology and ultrastructure of sex-pheromone-producing glands. In: Prestwitch GD, Blomquist GJ (eds) Pheromone biochemistry. Academic Press, Orlando, pp 27–75

Pers JNC van der, Den Otter NC (1978) Single cell responses from olfactory receptors of small ermine moths to sex attractants. J Insect Physiol 24: 337–343

Ross RJ, Palaniswami P, Seabrook WD (1979) Electoantennograms from spruce budworm moths (*Choristoneura fumifera*) (Lepidoptera: Tortricidae) of different ages and for various pheromone concentrations. Can Entomol 111: 807–816

Saad AD, Scott DR (1981) Repellency of pheromone released by females of *Heliothis armigera* and *H. zea* to females of both species. Entomol Exp Appl 30: 123–127

Schal C, Charlton RE, Cardé R (1987) Temporal patterns of sex pheromone titers and release rates in *Holomelina lamae* (Lepidoptera: Arctiidae). J Chem Ecol 13: 1115–1129

Schneider D (1957) Elektrophysiologische Untersuchungen an Chemo- und Mechanorezeptoren der Antenne des Seidenspinners *Bombyx mori* L. Z Vergl Physiol 40: 8–41

Schneider D (1992) 100 years of pheromone research – an essay on Lepidoptera. Naturwissenschaften 79: 241–250

Schneider D, Seibt U (1969) Sex pheromone of the queen butterfly: electroantennogram responses. Science 164: 1173–1174

Schneider D, Wunderer H (1990) Olfactory brain structures of two moth species with dual or male pheromone systems: *Creatonotos*, *Galleria*. In: Döving KB (ed) Proc 10th Int Symp Olfaction and Taste. University of Oslo, Norway, pp 159–164

Schneider D, Lacher V, Kaissling KE (1964) Die Reaktionweise und das Reaktionsspektrum von Riechzellen bei *Antheraea pernyi* (Lepidopetra, Saturniidae). Z Vergl Physiol 48: 632–662

Schneider D, Schulz S, Kittmann R, Kanagaratnam P (1992a) Pheromones and glandular structures of both sexes of the weed defoliator moth *Pareuchaetes pseudoinsulata* Rego Barros (Lep., Arctiidae). J Appl Entomol 113: 280–294

Schneider D, Schäfer W, Wunderer H (1992b) Scent organ in male moth: morphology alpha-keto-butyric acid content and evoked antennal responses. Zool Jahb Physiol 96: 365–377

Seabrook WD, Linn CE, Dyer LJ, Shorey HH (1987) Comparison of electroantennograms from female and male cabbage looper moths (*Trichoplusia ni*) of different ages and for various pheromone concentrations. J Chem Ecol 13: 1443–1453

Steinbrecht RA (1964) Die Abhängigkeit der Lockwirkung des Sexualduftorgans weiblicher Seidenspinner (*Bombyx mori*) von Alter und Kopulation. Z Vergl Physiol 48: 341–356

Steinbecht RA (1970) Zur Morphometrie der Antenne des Seidenspinners, *Bombyx mori* L.: Zahl und Verteilung der Riechsensillen (Insecta, Lepidoptera). Z Morphol Tiere 68: 93–126

Steinbecht RA, Laue M, Ziegelberger G (1995) Immunolocalization of pheromone-binding protein and general odorant-binding protein in olfactory sensilla of the silk moths *Antheraea* and *Bombyx*. Cell tissue Res 282: 203–217

Walker MF (1966) Some observations on the behaviour and life history of the Jersey tiger moth, *Euplagia quadripunctaria* Poda (Lep.: Arctiidae), in the ''Valley of the Butterflies'', Rhodes. The Entomologist 99: 1–24

Weatherston J, Percy JE (1977) Pheromones of male Lepidoptera. In: Adiyodi KG, Adiyodi RG (eds) Advances in invertebrate reproduction, vol 1. Peralam-Kenoth, India, pp 295–307

Wunderer H, Hansen K, Bell TW, Schneider D, Meinwald J (1986) Sex pheromones of two Asian moths (*Creatonotos transiens*, *C. gangis*; Lepidoptera-Arctiidae): behavior, morphology, chemistry and electrophysiology. Exp Biol 46: 11–27

Yin LRS, Schal C, Cardé RT (1991) Sex pheromone gland of the female tiger moth *Holomelina lamae* (Lepidoptera: Arctiidae). Can J Zool 69: 1916–1921

Yu HM, Becker H, Mangold HK (1989) Preparation of some pheromone bouquets. Chem Ind Lond 1989: 39–4