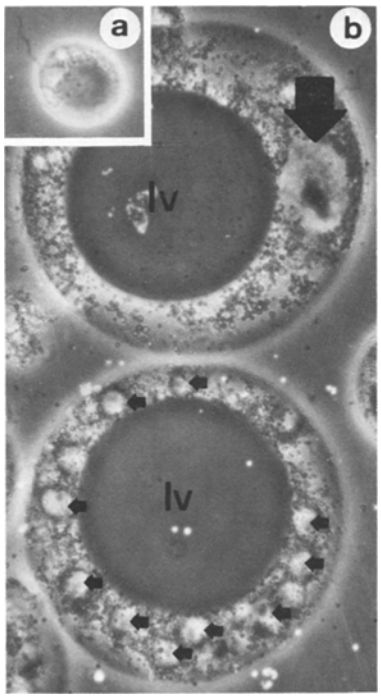

Fig. 1. Light micrographs of *Poterioochromonas*. The cells are unfixed. (a) Control cell, (b) Giant, TEL-treated cells (0.25 m*M* TEL, 7 days). One cell with only one giant nucleus (large arrow) and one cell with many nuclei of normal size (small arrows). *lv* leucosine vacuole, ×640

Table 1. The influence of 0.1 m*M* lead nitrate (Pb^{2+}) and 0.1 m*M* tetraethyl lead (TEL) on the cytokinesis and the nuclear index of *Poterioochromonas*. Nuclei per cell expressed in %. Cultivation time 5 days. Nuclear index: median number of nuclei per cell of a culture

Nuclei per cell	Control	Pb^{2+}	TEL
1	99.1	98.6	26.5
2	0.9	1.2	28.4
3	–	0.2	4.3
4	–	–	18.1
5	–	–	2.3
6	–	–	6.7
7	–	–	2.3
8	–	–	6.6
9	–	–	0.6
10	–	–	1.3
11	–	–	0.2
12	–	–	1.4
13	–	–	0.2
14	–	–	0.6
15	–	–	–
16	–	–	0.3
Nuclear index	1.009	1.015	3.438

aeration of the cultures of very low cell titers – there are further signs supporting the view of an inhibition of cytokinesis:

(1) The giant, normally non-dividing multinucleate cells divide frequently after the agent has been washed out.

(2) In 2- and 4-nucleate cells the nuclei often are arranged symmetrically; presumably this arrangement results from karyokinesis. 2-Nucleate cells from cultures with organolead cannot be distinguished from regular telophase stages of the controls.

(3) The idea of an inhibition of cytokinesis by organolead is further supported by the fact that the leukosine vacuole (LV) was never multiplied like the other organelles. In the normal cytokinesis of *Poterioochromonas* the LV is not – like the other organelles – replicated prior to the cell division but in the last phase of the cell division, when the cytoplasmic bridge connecting the two daughter cells breaks up.

However, the numbers of nuclei in the giant cells often deviate from 2^n (1-2-4-8 · · ·) (Table 1, TEL) which should be expected as a result of the inhibition of cytokinesis and a strictly synchronous karyokinesis.

It is probable that this is caused by disturbances of mitoses of single nuclei in multinucleate cells. As a result cells with many nuclei of different size occur whose number may deviate from 2^n. In addition complete small cells separate from giant, multinucleate ones.

In conclusion we have strong evidence that the two organic lead compounds are potent inhibitors of cytokinesis in *Poterioochromonas malhamensis*.

Work carried out under contract nr. 019-74-1ENV-D of the E.C. Environmental Research Programme. With support of the Commission of the European Communities.

Received July 26, 1977

1. Röderer, G.: Naturwissenschaften *63*, 248 (1976)
2. Schnepf, E., et al.: Protoplasma *92*, 87 (1977)

Recruitment Gland of *Leptogenys chinensis*

A New Type of Pheromone Gland in Ants

U. Maschwitz and P. Schönegge

Fachbereich Biologie der Universität, D-6000 Frankfurt a.M.

One of the most remarkable phenomena in the behavior of a number of ponerine ants is their highly organized group-predation [1]. Up to now, little is known about the signals that trigger and coordinate this behavior. The object of our investigations was a ponerine ant from Sri Lanka, *Leptogenys chinensis*, which feeds mainly on termites, preferably the genus *Odontotermes*. Single foraging ants search the surrounding area of the nest for feeding places of termites. The prey is transported to the nest along a previously laid exploration trail. There the returning ants excitedly run about among their nestmates dragging their gaster on the ground. This stimulates a varying number of ants, a group of which gathers at the entrance. They are guided to the feeding place by the successful scout which, for this purpose, lays a new trail out of the nest with its extruded sting.

At first we supposed the secretion of the venom gland of *L. chinensis* to be the recruiting signal, since it has orientating and recruiting effects in other *Leptogenys* species [2, 3]. The venom of *L. chinensis*, however, releases only orientation reactions and has no recruiting effects. While searching for the signal that triggers the characteristic group formation and group running, we found an intersegmental gland which opens between the last and the second to last gaster tergum [4]. Its secretion is highly stimulating. In test situations trails from the secretions of the dorsal gland and the venom gland were followed with equal frequency. When the content of the dorsal gland was offered in the nest, the forming of the characteristic group at the nest entrance was observed. Only a few animals of this group followed a fresh-laid venom trail leading out of the nest, more following a trail laid with the secretion of the dorsal gland. The greatest effect by far was achieved by using a mixture of both secretions (Fig. 1).

A feeding place of termites to which such a trail was laid was exploited as under normal conditions. This led to the conclusion that the secretion of the dorsal gland is the relevant signal for group recruitment. It must be brought to the ground together with the venom as the ant bends its gaster. When the effect of the venom gland was eliminated by closing the tip of the gaster with wax, these ants were still capable of recruiting groups of nestmates. But, when

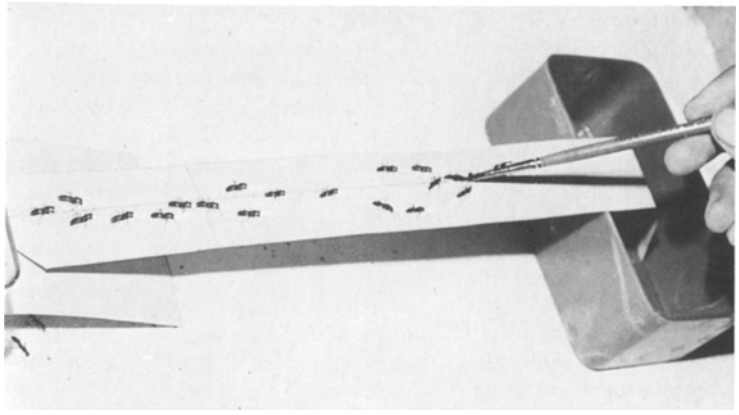

Fig. 1. Release of group recruitment in *L. chinensis* by laying an artificial trail with a mixture of venom and dorsal gland secretion.

the dorsal gland was inhibited with wax, these animals were not able to recruit more than one or two nestmates, although they used their intact sting for laying trails. These experiments indicated that our hypothesis on the function of the dorsal gland was correct.

Moreover, the pattern of group recruitment is an eminent clue to understanding the nest moving of *L. chinensis*. The emigration of the entire colony can be initiated by a single scout who has found a new nest site. We are able to induce large groups of ants to leave the nest by laying a trail with a mixture of the secretion of the venom and dorsal glands through the nest entrance and at the same time offering large quantities of it in the nest. In a few experiments even single pupae were transported, which is characteristic for the beginning of nest moving. This did not happen when only one of the gland secretions was offered. We observed that in the course of each normal colony emigration, without exception, one or more animals chirped with their gastral stridulation organ. Scouts with blocked dorsal glands who had found a new nest site were not able to form an emigration group in the old nest, although they stridulated and laid trails with their stings. When, on the other hand, the stridulation organs were blocked, but the dorsal glands left intact, the scouts did not loose their group recruiting ability in nests ready for emigration. Their ability to initiate colony emigration, however, was strongly weakened. In only a few experiments was a regular colony emigration initiated by these workers. In these cases we observed other emigrating workers chirping.

Thus, the secretion of the dorsal gland is a crucial signal for recruiting groups for group predation, as well as for colony emigration. For a regular colony emigration, the additional mechanical signal of chirping is necessary.

Received July 13, 1977

1. Wilson, E.O.: Evolution *12*, 24 (1958)
2. Fletcher, D.J.C.: Entomol. (A) *46*, 27 (1971)
3. Maschwitz, U., Mühlenberg, M.: Oecologia *20*, 65 (1975)
4. Jessen, K.: Staatsexamensarbeit, Frankfurt/M. (1976)

2-Ethyl-1,6-dioxaspiro[4.4]nonane, Principal Aggregation Pheromone of *Pityogenes chalcographus* (L.)

W. Francke

Institut für Organische Chemie der Universität, D-2000 Hamburg

V. Heemann, B. Gerken, J.A.A. Renwick, and J.P. Vité

Forstzoologisches Institut der Universität, D-7800 Freiburg i.Br.

Populations of the "Kupferstecher", *Pityogenes chalcographus* (L.), infest Norway spruce, *Picea abies* (L.), recently creating a serious pest problem in European spruce stands. These tiny beetles aggregate like other species of *Pityogenes* [1] on standing trees or slash of freshly cut trees, in response to a pheromone released by the host-selecting male beetles [2].

The isolation of the pheromone followed the principle of differential diagnosis [3]. Only male beetles feeding in the phloem tissue of host material or treated with juvenile hormone analogues (JHA) [4] were highly attractive to male and female beetles in flight [5]. Gas chromatographic analyses of the volatile content of such beetles revealed consistently three major peaks which were not detectable in unfed or untreated males or in female beetles treated in the same manner.

For chemical identification, approximately 100 000 beetles of both sexes were collected from cages as they emerged from naturally infested host material. Subsequently the beetles were treated with JHA ZR 233 (ethyl-3,7,11-trimethyl-2-dodecenoate), employing a novel technique [6]. This treatment was highly efficient in providing a relatively pure source of the candidate compounds.

The three male-specific peaks (Carlo-Erba 2100; 50 m, 0.25 mm i.d., stainless steel column with Marlophen 87, programmed from 50–140 °C at a rate of 3 °C/min) were subjected to mass-spectral analysis. The first substance was identified as 1-hexanol by comparison with an authentic sample on a Varian MAT 111 GC-MS-coupling system.

The two others, which were eluted as a double peak 3 min after 1-hexanol showed parent ions at m/e 156 (2%) and a similar fragmentation pattern on GC-MS, obviously representing two isomeric compounds: 127 (100;100), 87 (64;82), 85 (60;76), 43 (46;79), 56 (42;69), 55=57 (38;60), 41 (31;51), 97 (30;35), 42 (28;42), 69 (29;30), 98 (26;35), 81 (20;22), 39 (16;19), 73 (11;14).

From pentane extracts, the substances were isolated as a 1:1 mixture by preparative GLC (Perkin-Elmer F 21; 9 m, 8 mm i.d., stainless steel column with 5% XE 60 on Chromosorb G, 45–60 mesh). The molecular formula $C_9H_{16}O_2$ was obtained by high-resolution mass spectrometry, and the base peak was determined to be $C_7H_{11}O_2$, indicating the loss of an ethyl group from the molecule (Varian MAT SM 1). The ^{1}H-NMR spectrum showed three characteristic signals at δ=4.07, 3.89, and 3.70 (Bruker WH 270, 30 000 scans), each of which represented one proton in an α-position to oxygen, thus suggesting a bicyclic ketal structure analogous to known bark-beetle pheromones [7–9]. Though the fragmentation patterns of the unknown substances resembled those of the 6,8-dioxabicyclo[3.2.1]octanes and al-

