Assembling and Alerting Scents Produced by the Bedbug *Cimex lectularius* L.

MARX[1] reported in 1955 that the gregarious habit of *C. lectularius* is promoted by the presence of a specific nest odour, which could be provided also by crushed bedbugs. However respective results obtained by other investigators[2-4] seemed to contradict the above findings. SCHILDKNECHT[5] found recently that the scent gland secretion of bedbugs consists approximately of 70% hex-2-en-1-al and 30% oct-2-en-1-al but the function of the secretion is as yet unclear. We have attempted to clarify this somewhat confusing situation by a study of the olfactory behaviour of bedbugs towards their body scent as well as the above aldehydes.

Filter paper discs (thickness 0.38 mm, diam. 25 mm) were scented by impregnation with different levels of hex-2-en-1-al [b.p.$_{20}$ = 51–53 °C, n_D^{18} = 1.4438; 2,4-DNPH: mp = 144°, λ_{max}^{EtoH} = 374 nm], oct-2-en-1-al [b.p.$_{14}$ = 76–78 °C, n_D^{20} = 1.4500; 2,4-DNPH: mp = 126°, λ_{max}^{EtoH} = 372 nm][6] or by the body odour of male and female *C. lectularius*. The latter was achieved by keeping graded numbers of bedbugs in close contact with single paper discs for 48 h in glass-stoppered vials or by adsorbing methanol extracts of the above mentioned discs on test discs. A total of 1 scented and 7 untreated paper discs were placed at a distance of 27° from disc to disc near the circumference of a filter paper arena (diam. 200 mm) sticked onto the floor of a glass dish (Figure 1). Series of 10 adults of either sex (fed 2 days ago on mice blood) were exposed per choice arena maintained at 27 ± 1 °C, 55 ± 5% relative humidity and semidarkness (> 0.1 lumen/sq. ft.). The number of bedbugs aggregating under each of the 8 paper discs was then compared at successive hours (counted at red light). The random distribution of bedbugs among 8 unscented paper discs at the above conditions served as basic control.

Paper discs scented with either hex-2-en-1-al or oct-2-en-1-al or with a mixture of both aldehydes in various amounts and proportions did not produce any aggregation of male or female bedbugs, suggesting that the main constituents of the scent gland secretion have no intraspecific attractant function. They appear to have a protective rôle, since like other *Hemiptera*[7,8], *C. lectularius* release them when irritated (e.g. by abrasive powder), and become excited as well as repelled by them. Application of both aldehydes in proportion of their occurrence in the glandular secretion[5] (approx. 47 µg/per ml of air) caused aggregated bedbugs to disperse rapidly. Their responses to this scent resemble the behaviour induced by alerting pheromones, as defined by BUTLER[9].

On the other hand, bedbugs of both sexes assembled at similar or equal rates under papers which had been scented by the odour of 25 female or male bedbugs. Under experimental conditions, the number of aggregating insects increased gradually within 4 h after exposing them to the choice arena, and remained then relatively constant during the following 16 h (Figure 2). As tactile responses are known to be dominant reactions in the behaviour of *C. lectularius*[10], the above aggregations – once established – are most likely to be preserved by thigmotaxis (Figure 3). It is noteworthy that mated as well as unmated bedbugs are attracted to the assembling scent and that on approaching the latter, the insects erect their previously limp antennae. However after removal of both antennae, male and female bedbugs failed to respond to the above odour.

Number and sex of bedbugs aggregating seem to depend on the source and concentration of bedbug scent (Table I). Females were consistently more responsive than males towards the scent of either males, females or both sexes (1:1), while mixed male and female bedbugs (1:1) responded to the above scents in a slightly different manner than either sex alone. The scents of all 3 sources displayed considerably more attractancy at 25 than at 12 bedbug equivalents, whereas male or female scents

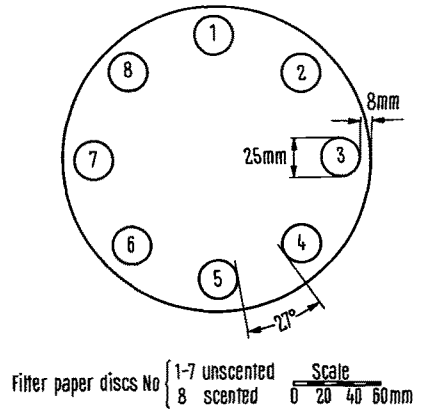

Fig. 1. Choice arena used for testing aggregation of bedbugs (schematic view from above).

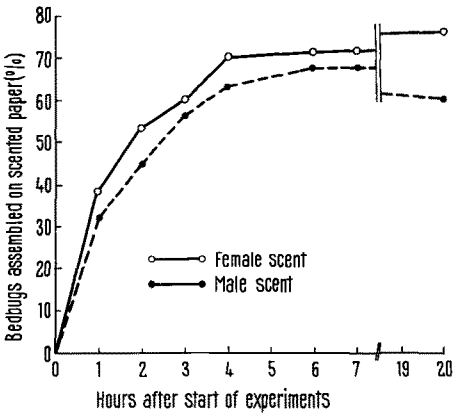

Fig. 2. Velocity of bedbug attraction to the scent of 25 female or male *C. lectularius*. Female and male bedbugs accumulated under the scented papers in almost equal proportion.

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Fig. 3. Aggregation of bedbugs (females) on filter paper scented by the odour of female or male bedbugs. The paper disc was inverted for photographing (approximately 2 × natural size).

Table I. Attraction of male and female bedbugs to the scents of male and female *C. lectularius*

Scent source			Aggregation of		
Males		Females	Males (%)[a]	Females (%)[a]	Both sexes 1:1 (%)[a]
12		–	30	68	33
25		–	68	92	63
100		–	73	93	
–		12	12	57	33
–		25	68	86	70
–		100	86	84	
6	+	6	15	50	48[b]
12	+	12	25	75	56[b]
25	+	25	60	80	82[b]
50	+	50			100[b]

[a] 60 adults of either sex or mixed sexes were tested per scent level; their aggregation under the scented papers was recorded 4 h after start of experiments. [b] Females and males were attracted in equal proportion.

Table II. Attraction of *C. lectularius* to graded levels of mixed female and male scents

Bedbug equivalents[a]	0.1	1	2.5	5	10	20	50
Females assembled under scented paper (%)	0	14	28	42	58	62	90

[a] Methanol extract of female and male scent 1:1.

were similarly attractive at 100 and 25 bedbug equivalents. It is interesting that fifth instar nymphs respond only slightly to those scents, despite the fact that in natural harborages the former are usually found in clusters with adults. The latter may be mainly due to tactile reactions among bedbugs of different ages.

Methanol extraction of papers impregnated by the odour of male and female *C. lectularius* (1:1) yields a relatively effective scent solution, of which an aliquot of 1 bedbug equivalent attracts 14% of the exposed females (Table II). However the above scent mixture was found to be insoluble in diethylether. The scent can be efficiently volatilized from living bedbugs at 32 °C and atmospheric pressure, and can then be adsorbed on filter paper suspended in the air above the insects.

Since the bedbug scent causes hetero-as well as homosexual attraction, we have classified it among the assembling scents[11]. It should be recalled however, that scents to which this term has been applied, may differ widely in their modes of action[9,12]. We assume that the above mentioned odour induces bedbugs of both sexes to move into harborages which provide the tactile stimuli necessary to maintain the aggregation of these insects. The mutually opposed effects of assembling and alerting scents may be of ecological significance[13].

Zusammenfassung. Es wird gezeigt, dass die Aggregation der Bettwanzen (*Cimex lectularius*) durch einen von beiden Geschlechtern abgegebenen Versammlungsduft eingeleitet wird. Letzterer ist in Methylalkohol, aber nicht in Diäthyläther löslich, verdampft bei 32 °C und lockt unbegattete sowie begattete Wanzen beider Geschlechter an. Die Lockreaktion ist von der Duftmenge abhängig und bleibt bei antennenlosen Wanzen aus. Die Stinkdrüsen gereizter, respektive verletzter Bettwanzen sondern Hex-2-en-1-al und Oct-2-en-1-al als Alarmduft ab, der die Zerstreuung der Wanzengruppen hervorruft.

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L-Isoleucine: An Inducer of the Feeding Response in Decapod Crustaceans

Pachygrapsus crassipes, the lined shore crab, exhibits the following feeding behavior when dead squid is presented: the animals scrape the bottom of the tank with their claws and lift their claws to their mouths. This behavior is followed by searching movements during which the crabs walk around the tank until they contact the squid. The crabs take the squid in their claws, tear it into small pieces, and place it in their mouths. Experiments were performed to determine what elicits this response.

The nature of the feeding response was determined in a preliminary study using filtered squid juice (homogenate of 10 g of frozen squid in 30 ml of seawater). A piece of Whatman No. 1 filter paper (1–3 mm²), pre-