A Sex Attractant for the European Fir Budworm Moth, *Choristoneura murinana*

Ernst Priesner

Max-Planck-Institut für Verhaltensphysiologie, D-8131 Seewiesen

Hermann Bogenschütz

Forstliche Forschungs- und Versuchsanstalt Baden-Württemberg, D-7801 Stegen-Wittental bei Freiburg

and

Heinrich Arn

Eidgenössische Forschungsanstalt für Obst-, Wein- und Gartenbau, CH-8820 Wädenswil

Z. Naturforsch. **35 c**, 390–398 (1980); received February 12, 1980

Pheromones, Sex Attractant, (*Z*)-9-dodecenyl Acetate, Olfactory Receptors, *Choristoneura murinana*, Tortricidae

Traps baited with 100 µg (*Z*)-9-dodecenyl acetate and 10 µg (*Z*)-11-tetradecenyl acetate are highly effective in attracting *C. murinana* males at varying population densities. The attractive mixture was established by electrophysiological and field screening studies combined with chemical analysis of the female pheromone secretion.

(*Z*)-9-dodecenyl acetate is a major constituent in washes of pheromone glands of calling *C. murinana* females. This compound, the first dodecenyl ester pheromone known for a species of the Archipini tribe, is moderately attractive on its own at low doses. Attraction is strongly synergized by 10% (*Z*)-11-tetradecenyl acetate but is inhibited by small amounts of (*Z*)-9-tetradecenyl acetate. Each of the three compounds acts upon a separate type of receptor cell in hair sensilla of the male antenna.

Choristoneura murinana Hb. (Tortricidae: Tortricinae, Archipini), the "Tannentriebwickler" of the silver fir (*Abies alba*) forests of Central and Eastern Europe [1–3], is considered a close taxonomic relative to a group of North American, coniferophagous *Choristoneura* spp. known as the "spruce budworms" [4–9]. In one of these, *C. fumiferana* (eastern spruce budworm), traps baited with the synthetic sex pheromone have proved capable of catching significant numbers of males at population densities well below one larva per branch, when conventional sampling techniques become impractical in detecting incipient infestations [10–12]. Also in *C. murinana*, sex pheromone trapping has great promise in monitoring low population fluctuations in potential outbreak areas ranging from Eastern France and Switzerland to Southern Germany, Austria, Czechoslovakia, and Poland [1–3]. As a first contribution towards a system of forecasting *C. murinana* infestations by sex attractant traps, we report the identification of the major component of the female sex pheromone of the species and the development of a potent sex attractant mixture.

Results (including methodological notes) and conclusions will be presented in a chronological sequence, starting in 1975.

Reprint requests to Dr. E. Priesner.

0341-0382/80/0500-0390 $ 01.00/0

An early field trial

In the North American spruce budworms, two tetradecenyl compounds, (*E*)-11-tetradecenal (*E*11-14:Ald) and (*E*)-11-tetradecenyl acetate (*E*11-14:Ac), have been implicated as the major component of the female sex pheromones of the different species [8, 13–16]. Considering the close affinity of *C. murinana* to its New World relatives, in morphological as well as physiological and ecological characters [9], a pilot field study was undertaken in 1975 [17] in order to test these and a few structurally-related compounds for possible attractancy to male *C. murinana*. Tests were carried out, from July 10 to August 8, in the Black Forest at Biberach, Kinzigtal, in a mature stand of *A. alba*. The stand had suffered some tree mortality due to the last *C. murinana* in-

festation (which collapsed in 1968) but showed no detectable defoliation in 1975. The compounds tested were *E*11-14:Ald, *E*11-14:Ac, (*E*)-11-tetradecen-1-ol (*E*11-14:OH), the corresponding (*Z*) isomers (*Z*11-14:Ald, *Z*11-14:Ac, *Z*11-14:OH), and (*E*)-9-tetradecenyl acetate (*E*9-14:Ac), all ≧ 99% pure. They were made up in polyvinyl chloride formulations [18] containing 3% of the chemical per weight. Pellets of the formulations, each containing 4.5 milligram of a particular test compound, were pinned singly or in various 1:1 combinations in Sectar 1 traps.

Trap catches are listed for three lepidopteran spp. (Table I). Pure *Z*11-14:Ac (treatment No. 2) and the *Z*11-14:Ac + *E*9-14:Ac combination (No. 8) attracted large numbers of *Parasyndemis histrionana* Fröl., another fir-inhabiting leafroller species which closely resembles *C. murinana* and has thus repeatedly caused confusion in early literature (as discussed in [1–3]). Smaller numbers of this species were captured by the other lures containing *Z*11-14:Ac except for the combination with the (*E*)-11 isomer (No. 10).

The traps with pure *E*9-14:Ac, a known sexual attractant for *Gelechia* spp. of the subgenus *Bryotropha* [19–21], caught numbers of *Gelechia* (subg. *Neofaculta*) *betulae* Hw. (Gelechiidae). The attraction response to this chemical does not seem to be markedly affected by the presence of the alcohols or aldehydes (see treatments Nos. 11, 12, 17, 18 *vs.* No. 1) whereas it is abolished by addition of *Z*11-14:Ac or *E*11-14:Ac (Nos. 8, 9). Male *Evergestis for-*

Table 1. Numbers of males of 3 lepidopteran spp. captured by 7 test compounds and their binary 1 : 1 combinations in Sectar-1 traps at Biberach, July 10 to August 6, 1975 [17].

Treatment No. *	Test chemicals *							Total catch of		
	*E*9-14 : Ac	*Z*11-14 : Ac	*E*11-14 : Ac	*Z*11-14 : OH	*E*11-14 : OH	*Z*11-14 : Ald	*E*11-14 : Ald	*Parasyndemis histrionana*	*Gelechia betulae*	*Evergestis forficalis*
1	x							–	58	–
2		x						243	–	–
3			x					–	–	11
4				x				–	–	–
5					x			–	–	–
6						x		–	–	–
7							x	–	–	–
8	x	x						190	–	–
9	x		x					–	–	–
10		x	x					–	–	–
11	x			x				–	21	–
12	x				x			–	30	–
13		x		x				73	1	–
14		x			x			26	–	–
15			x	x				–	–	–
16			x		x			–	–	–
17	x					x		–	19	–
18	x						x	–	48	–
19		x				x		21	–	–
20		x					x	8	–	–
21			x			x		–	–	–
22			x				x	–	–	–
23				x	x			–	–	–
24				x		x		–	–	–
25				x			x	–	–	–
26					x	x		–	–	–
27					x		x	–	–	–
28						x	x	–	–	–

* Each chemical at dose of 4.5 mg, incorporated in polyvinyl chloride; 3 traps per treatment.

ficalis L. (Phycitidae) were selectively attracted to pure *E*11-14:Ac (No. 3), confirming other reports [22, 23].

During the 4 week test period, no male *C. murinana* were captured in any of the traps.

Antennographic and preliminary chemical analysis, 1976/77

Analysis of the *C. murinana* female pheromone was started in 1976/77 by evaluating the "profile" of male electroantennogram (EAG) responses to standard test compounds, using techniques of stimulation and recording as in earlier EAG studies with other tortricid species [24–28]. In the series of mono-unsaturated C_{10} to C_{16} acetates, alcohols, and aldehydes, varying in position and configuration of the olefinic double bond, (*Z*)-9-dodecenyl acetate (*Z*9-12:Ac) evoked the maximum response. Dienic acetates included in these assays were significantly less stimulatory than the corresponding (*Z*) or (*E*) monoenes.

A dodecenyl compound eliciting the highest EAG response in a species of *Choristoneura* was an unexpected discovery, as unsaturated C_{12} pheromones were unknown from the Tortricinae: Archipini tribe (see also below). However, subsequent chemical analysis established the presence of *Z*9-12:Ac as the major component of the *C. murinana* female pheromone secretion [29]. Abdominal tips of 70 virgin females, collected as pupae in the Polish outbreak area near Kielce in June 1977, were extracted in ethyl ether and the extracts analyzed by capillary gas chromatography using a Silar 10C glass capillary column with Ucon 50 HB 5100 as a liquid phase, and mass spectrometry [30]. In these preliminary chemical investigations, a compound with the retention time and mass spectrum of *Z*9-12:Ac was discovered in the extracts in quantities of less than 1 nanogram per female [29]. In some extracts it was accompanied by a smaller amount of another C_{12} acetate which due to its shorter retention time was tentatively assigned as the (*E*)-9 or (*Z*)-8 isomer (*E*9-12:Ac or *Z*8-12:Ac) [29] (for further chemical analysis see below).

Field attraction test, 1978

The electrophysiological and analytical results prompted a series of field experiments to investigate the possible attractiveness of *Z*9-12:Ac, or combinations of this compound with the (*E*)-9 or (*Z*)-8 isomer, to *C. murinana* males. The test area chosen was a mixed-aged stand of *A. alba* in the sub-alpine mountains of Central Poland near Suchedniów, heavily attacked by *C. murinana*. Comparisons were made with 100 µg *Z*9-12:Ac (< 0.1% *E*) by adding *E*9-12:Ac and/or *Z*8-12:Ac in ratios ranging from 100/5 to 100/100. The synthetic formulations were applied to rubber caps (Tellergummikappen No. 90142, Auer Bittmann Soulié AG Zürich) as solutions in hexane. Pherotraps 1C (Zoecon) and tetratraps with flaps [31] were used. They were attached at a heigth of 2 m to the trunks of fir trees at distances of 3–5 m. The traps were set up July 7, 1978, when field observations indicated intense mating flights.

During the 1 week test period, pure *Z*9-12:Ac and most of the mixture formulations attracted low numbers of *C. murinana* males. However, up to the 100/100 ratio the (*E*)-9 and (*Z*)-8 isomer failed to show any pronounced synergistic or inhibitory effect. Compared to traps baited with 1 live virgin female or the hexane extract from 10 virgin females, catches by the synthetic formulations were generally low. This suggested that either the 100 microgram dose was not at the optimum, or that an essential minor component(s) in the synthetic attractant was missing.

Single receptor analysis

In several cases of incomplete chemical identification of a multi-component moth pheromone, single cell analysis of male receptor types has provided the information for the development of an effective attractant mixture (see refs. [26, 35]). For example, the synthetic formulation presently used in monitoring pine beauty moth (*Panolis flammea*) populations by sex attractant traps in different parts of Europe [32, 33], a 100/5 mixture of *Z*9-14:Ac and *Z*11-14:Ac, is based on combining two "receptor key compounds" [34]. This same procedure was applied to *C. murinana* in a search for receptor types responsive to further potential pheromone components. As in earlier studies on other tortricid species [25–28, 35], nerve impulse responses were monitored via the cut end of a male hair sensillum (*S. trichodeum*). This procedure maintains the responsiveness of individual receptor cells for several hours, thus allowing the

effects of a broad set of test chemicals to be evaluated on the same sensory cell.

Results of these investigations indicate four different types of receptor cells in *C. murinana*, which in the order of nerve impulse amplitude were named cells A to D. The type A cells (largest amplitude) responded maximally to *Z*9-12:Ac; the B cells, to (*Z*)-9-tetradecenyl acetate (*Z*9-14:Ac); and the C cells, to (*Z*)-11-tetradecenyl acetate (*Z*11-14:Ac). The D cells showed moderate responses to several monoenic and dienic C_{12} acetates, including *E*9-12:Ac, but the key structure has not been definitely determined. Thus, in addition to receptor cells specific to the major component of the pheromone blend, *Z*9-12:Ac, already identified, other types of cell responded maximally each to a particular C_{14} acetate. These results strongly suggested that a C_{14} acetate, not searched for in the chemical analysis and not apparent from the EAG response spectrum, could be involved in the *C. murinana* pheromone blend.

Field tests in Poland and France, 1979

The field program set up for the 1979 flight season was accordingly directed at the effects of (i) various concentrations of pure *Z*9-12:Ac, and (ii) the admixture of different amounts of *Z*9-14:Ac and *Z*11-14:Ac to *Z*9-12:Ac. Dispensers and trap types were the same as in 1978 (see above). Each test series consisted of a set of 5 to 7 treatments in at least 3 replicates. Unbaited check traps (without caps) were included in each series. Trap distances were 3–5 m within sets and 50 m or more between replicates. To account for possible interactions between neighbouring traps, the sequence in treatment positions was changed for every replicate. Unless indicated otherwise, traps were attached at a heigth of 2 m to the trunks of fir trees. The same test program was conducted in parallel at Suchedniów (the heavily-infested test area of 1978, see above) and at Guebwiller in the Vosges mountains of Eastern France, a local area of moderate *C. murinana* infestation (for additional field tests, carried out in an area of very low population density in the Black Forest near Freiburg, see further below).

The effect of different loads of pure *Z*9-12:Ac (< 0.1% *E*) is apparent from Tables II and III. At Suchedniów, due to considerable variations in the local population the total catch strongly varies between the different replicates. However, in the 5 replicates with the highest catches (No. 5–9) by far the most males responded to the lowest dose of 1 µg (Table II). The 10 µg dose was about 4 fold less attractive whereas catches with 100 µg and 1000 µg were not significantly different from blank traps. An inhibitory effect of these higher doses, suggested by replicate 9, is supported by the results of a further replicate (not included in the Table) in which an unusually high number of 81 to 117 males were captured in the blank trap and the 1 µg, 10 µg, and 100 µg traps, but only 22 males in the 1000 µg trap (see also discussion below).

In the corresponding experiment at Guebwiller, traps were placed out on July 2, checked and repositioned on July 12 and 17, and collected up July 24. No caps were replaced during the course of the experiment. Results are accordingly presented for the three successive periods, July 2–12, July 13–17, and July 18–24 (Table III). As shown by these data, in period I in each replicate the 1 µg or 10 µg trap were the most attractive; 100 µg was only slightly attractive; and 1000 µg again did not catch significant numbers of males. However, in period II and III

Table II. Numbers of *C. murinana* males captured in Pherocon 1 C traps at a heigth of 2 m, baited with 4 doses of *Z*9-12:Ac (< 0.1% *E*), in 9 replicates at Suchedniów, June 21 to July 7, 1979.

Amount of *Z*9-12:Ac per cap [µg]	Replicate No.									Total *	(%)
	1	2	3	4	5	6	7	8	9		
0	0	2	4	1	1	3	8	8	28	55 b	(9.7)
1	8	6	6	5	20	94	34	89	96	358 a	(63.4)
10	3	2	9	2	3	20	32	11	12	94 ab	(16.6)
100	4	1	2	1	6	4	7	2	7	34 b	(6.0)
1000	3	4	1	2	1	2	4	5	2	24 b	(4.3)

* Capture totals followed by the same letter are not significantly different at the 95% probability level (P = 0.05) as indicated by analysis of variance followed by Duncan's multiple range test.

Table III. Numbers of *C. murinana* males captured in Pherocon 1 C traps at a heigth of 2 m, baited with 4 doses of *Z*9-12:Ac (< 0.1% *E*), in 5 replicates at Guebwiller, July 2 to 24, 1979.

Amount of *Z*9-12:Ac per cap [µg]	Period I, July 2 – 12						Period II, July 13 – 17						Period III, July 18 – 24					
	replicate No.					Σ *	replicate No.					Σ *	replicate No.					Σ *
	1	2	3	4	5		1	2	3	4	5		1	2	3	4	5	
0	3	5	3	1	1	13 c	1	1	0	0	0	2 b	0	0	0	0	0	0 c
1	60	34	8	36	42	180 a	0	1	0	0	0	1 b	0	1	0	0	0	1 bc
10	81	5	19	8	9	122 ab	11	1	2	0	4	18 ab	5	2	0	1	6	14 ab
100	20	6	7	0	14	47 bc	52	2	0	1	15	70 a	11	2	0	0	16	29 a
1000	3	2	0	0	6	11 c	12	1	1	0	2	16 ab	2	0	0	0	2	4 abc

* Values in the same column followed by the same letter are not significantly different at P = 0.05.

Table IV. Numbers of *C. murinana* males captured in Pherocon 1 C traps at a heigth of 2 m, baited with varying combinations of *Z*9-12:Ac, *Z*9-14:Ac, and *Z*11-14:Ac, in 6 replicates at Suchedniów, June 22 to July 10, 1979.

Amount of chemical per cap [µg]			Replicate no.						Total *	(%)
*Z*9-12:Ac	*Z*9-14:Ac	*Z*11-14:Ac	1	2	3	4	5	6		
0	0	0	18	74	8	0	11	2	113 b	(4.3)
100	0	0	9	4	5	4	7	0	29 b	(1.1)
100	3	0	7	2	2	1	6	4	22 b	(0.8)
100	10	0	4	7	5	1	1	6	24 b	(0.9)
100	0	3	131	146	125	108	112	149	771 a	(29.7)
100	0	10	208	176	159	146	167	194	1050 a	(40.4)
100	5	5	128	116	79	10	104	157	594 a	(22.8)

* Capture totals followed by the same letter are not significantly different at P = 0.05.

the lower doses had apparently lost their attractiveness as most males now responded to the 100 µg traps (Table III). Thus, at least in a higher population density as present at Guebwiller in test period I, pure *Z*9-12:Ac seems to be far more attractive at the lower doses of 1 – 10 µg (see also discussion below). This finding could in part explain the poor results obtained with the standard dose of 100 µg in 1978.

The second series of field experiments, conducted in the same test areas during the same time, investigated the possible synergistic effects of *Z*9-14:Ac and *Z*11-14:Ac (the two C_{14} compounds mentioned above as candidate structures for secondary pheromone components). As results with the lower *Z*9-12:Ac doses were not yet available, comparisons were made to 100 µg *Z*9-12:Ac by adding one or both chemicals (purity ≧ 99%) at ratios of 100/3, 100/10, and 100/5/5. Trapping results are listed in Tables IV and V.

At Suchedniów, agin considerable numbers of males were recorded in the blank traps (Table IV). Compared to these, pure *Z*9-12:Ac and the *Z*9-12:Ac + *Z*9-14:Ac mixture do not show any increase but rather a decrease of male catches (*e. g.*, in replicates 1 and 2), comparable to the results with the 100 µg and 1000 µg doses of pure *Z*9-12:Ac, mentioned above. However, in all 6 replicates the addition of *Z*11-14:Ac strongly increased catches, the maximum occurring with the 100 µg + 10 µg blend of the two compounds, in all replicates (Table IV). The ternary mixture (ratio 100/5/5) caught significantly less males, again suggesting an inhibitory effect of the *Z*9-14:Ac.

At Guebwiller too, in all 6 replicates the blend of 100 µg *Z*9-12:Ac with 10 µg *Z*11-14:Ac gave the highest catches (Table V). The 100/3 mixture of these two compounds, and the 100/5/5 combination of all three compounds, were significantly less attractive. Pure *Z*9-12:Ac and the *Z*9-12:Ac + *Z*9-14:Ac mixture again were essentially unattractive (Table V).

To compare the two most effective attractants found in these two test series, the 1 – 10 µg dose of

Table V. Numbers of *C. murinana* males captured in tetratraps at a heigth of 2 m, baited with varying combinations of *Z*9-12:Ac, *Z*9-14:Ac, and *Z*11-14:Ac, in 6 replicates at Guebwiller, July 2 to 24, 1979.

Amount of chemical per cap [µg]			Replicate no.						Total *	(%)
*Z*9-12:Ac	*Z*9-14:Ac	*Z*11-14:Ac	1	2	3	4	5	6		
0	0	0	0	0	1	1	0	0	2 c	(0.2)
100	0	0	0	0	1	0	1	0	2 c	(0.2)
100	3	0	0	0	0	0	0	1	1 c	(0.1)
100	10	0	2	1	0	0	0	1	4 c	(0.3)
100	0	3	70	3	69	10	101	30	281 b	(22.1)
100	0	10	102	79	109	64	173	187	714 a	(56.2)
100	5	5	44	17	57	16	83	39	266 b	(20.9)

* Capture totals followed by the same letter are not significantly different at P = 0.05.

Table VI. Numbers of *C. murinana* males captured in tetratraps *vs.* Pherocon 1 C traps, baited with combinations of *Z*9-12:Ac and *Z*11-14:Ac, at Guebwiller, July 17 to 24, 1979; trap heigth 2 m; 3 replicates each.

Amount of chemical per cap [µg]		Tetratrap				Pherotrap			
*Z*9-12:Ac	*Z*11-14:Ac	replicate No.			Σ *	replicate No.			Σ *
		1	2	3		1	2	3	
0	0	1	0	0	1 i	1	2	0	3 ef ***
1	0.1	8	56	4	68 fg	39	5	**	
10	0	1	17	4	22 h	6	4	18	28 gh
10	1	34	90	16	140 de	186	91	171	448 ab
10	3	38	157	115	310 bc	71	84	103	258 bc
100	10	43	89	21	153 cd	102	262	354	718 a

* Capture totals followed by the same letter are not significantly different at P = 0.05.
** Trap missing.
*** The statistical treatment assumes a calculated value of 35 for the missing trap.

pure *Z*9-12:Ac and the 100 µg + 10 µg combination of *Z*9-12:Ac with *Z*11-14:Ac, a further field experiment was set up at Guebwiller in mid July. Also included were mixtures of the two compounds at the lower doses of 1 + 0.1 µg, 10 + 1 µg, and 10 + 3 µg. Pherotraps and tetratraps were used, in 3 replicates each, to evaluate possible effects of trap design (Table VI).

Evidently, the mixtures were far more attractive than the 10 µg dose of pure *Z*9-12:Ac (Table VI). The strong synergistic effect of a 10% admixture of *Z*11-14:Ac was now shown also at the lower dose levels, the 10 + 1 µg combination being almost equivalent to 100 + 10 µg. However, a remarkable difference between the two trap types was noted with respect to the preferred mixture ratio: whereas in the tetratraps the 10 + 3 µg mixture was the most attractive formulation, in the pherotraps the 10 + 1 µg and 100 + 10 µg mixtures caught more males. This unexpected result suggests a possible role of the synergist (*Z*11-14:Ac) in close-range male behaviour in which features in trap design would come into play. The effects of synthetic formulations in relation to trap type could not be followed up further during the 1979 flight period but will be a major subject of the *C. murinana* field program in 1980.

Search for secondary pheromone components

Results of the field attraction and receptor studies strongly suggest involvement of *Z*11-14:Ac as a minor component in the *C. murinana* pheromone blend. In a search for this compound, further chemical analysis was carried out in 1979 [29] on washes of virgin females obtained from pupae collected in the Kielce area. Females (mostly 3–4 days old), in calling position after the beginning of the dark period, were extracted by washing the extruded glands in methylene chloride for 20–30 sec [27].

Aliquots of a wash of 10 females were analyzed in a Finnigan 4000 GC-MS system equipped with a Silar 10C glass capillary column and using chemical ionization with isobutane [30, 36].

Spectra search after data aquisition showed the presence of approx. 4 nanogram per female of a dodecenyl acetate as one of the three major components in the chromatogram. Co-injection of the wash with standards, using multiple ion monitoring (*m/e* 167 + 227), showed that this component eluted together with *Z*9-12:Ac (which under the given chromatographic conditions is separated from all positional isomers except 11-12:Ac), thus confirming the identity of *Z*9-12:Ac as a major component of the *C. murinana* female pheromone [29].

No other dodecenyl acetate was present in these washes. Furthermore, contrasting the results of the receptor and field studies, mass chromatograms at *m/e* 255 showed no evidence for the presence of a tetradecenyl acetate; if present, its amount would be less than 0.2% of that of *Z*9-12:Ac [29]. Also antennographic detection [37], using antennae of different moth species known to be sensitive to various acetates [36–38], showed a response only at the retention time of *Z*9-12:Ac. The nature of possible secondary pheromone components, if present, thus remains for future studies.

*Z*11-14:Ac, here reported as a "receptor key compound" and strong attraction synergist for *C. murinana* males, is the main component of the female pheromone of the oblique-banded leafroller moth, *Choristoneura rosaceana* [39, 40]. The compound has to our knowledge not been isolated from further *Choristoneura* spp. but is a common pheromone constituent in various other members of the Archipini tribe, including the genera *Archips, Archippus, Argyrotaenia, Adoxophyes, Clepsis, Cacoecimorpha, Epichoristodes, Pandemis,* and *Platynota* [41–56]. *Z*9-12:Ac, on the other hand, has not previously been reported from this taxonomic group in which dodecenyl pheromones were unknown. The establishment of *Z*9-12:Ac as the major pheromone component in *C. murinana* thus presents an exception to the "rule" [57] that species of Archipini (and other groups of the subfamily Tortricinae as well) generally produce C_{14} (mono- or di-unsaturated) pheromones. *Z*9-12:Ac is known as a major pheromone constituent from other tortricids belonging to the subfamily Olethreutinae, as the grape berry moth *Paralobesia viteana* [58], the cereal tortrix *Cnephasia pumicana* [59], and the pine shoot borer *Eucosma sonomana* [60]; and species of other lepidopteran families as well, including the European grape moth *Eupoecilia ambiguella* (Cochylidae) [61, 62] and the fall armyworm moth *Spodoptera frugiperda* (Noctuidae) [63, 64]. The synergistic combination of *Z*11-14:Ac with *Z*9-12:Ac has recently been reported [65] from a tea tortrix species, *Homona magnanima* (Olethreutinae) in which females produce the two compounds in a 10:1 ratio. Combinations of the two compounds with *Z*9-12:Ac as the major constituent have to our knowledge not been recorded before.

The possible role of the *Z*9-14:Ac, which is a "key compound" of another type of receptor cell on the male antenna (see above), has not been fully clarified in the present study. At the concentrations and blends used in these tests, the presence of *Z*9-14:Ac causes a reduction in catches, a phenomenon commonly referred to as inhibition. Specialist cells for attraction synergists and inhibitors, combined within the same male hair sensillum, have been well established in some other tortricid species (see refs. [25, 26, 35]).

Field test in the Black Forest, 1979

As in the North American spruce budworms, the mating flight of *C. murinana* takes place mainly in the upper crown region of the host tree as shown by traps baited with live virgin females which gave by far the highest catch at the canopy of the tree [9, 66]. However, any extensive monitoring program must

Table VII. Numbers of *C. murinana* males captured in tetratraps at a height of 9 – 16 m, baited with varying combinations of *Z*9-12:Ac, *Z*9-14:Ac, and *Z*11-14:Ac. Freiburg, June 29 to July 26, 1979.

Amount of chemical per cap [μg]			Catch *
*Z*9-12:Ac	*Z*9-14:Ac	*Z*11-14:Ac	
0	0	0	0
1	0	0	1
10	0	0	1
100	0	0	0
100	3	0	0
100	10	0	0
100	0	3	14
100	0	10	101
100	5	5	19
1000	0	0	0

* One trap per treatment

rely on traps operated at ground level. In *C. fumiferana*, trap catches at a heigth of 2 m are reported about 6 times lower than at crown level [67] but sufficient for monitoring purposes [10–12]. To evaluate for *C. murinana* the effect of trap heigth, in a final field experiment one series of trap replicates was operated in the lower crown region of silver fir trees at a heigth of 9–16 m. A mature stand of *A. alba* in the Black Forest near Freiburg with a very low population of *C. murinana* was chosen as test area.

Results for the crown-trap series are listed in Table VII. They show a total of 101 males captured by the 100/10 mixture; 14 and 19 males by the 100/3 and 100/5/5 mixtures, respectively; and single males by 1 and 10 µg of pure *Z*9-12:Ac. The other four lures did not attract any males (Table VII). Compared to the results obtained with ground-level traps in the test areas of higher population density (Tables IV to VI), the same mixtures were again effective but the selectivity seems to be more pronounced. In the corresponding replicates operated at 2 m, the 100/10 mixture attracted small but significant numbers of males (not specified in Tables). Thus, the synthetic attractant reported here, a combination of 100 µg *Z*9-12:Ac with 10 µg *Z*11-14:Ac, seems to be appropriate for monitoring mating flights of *C. murinana* with traps at ground level even when the local population is at low density.

Acknowledgements

We wish to express our thanks to Drs. Z. Sierpinski and T. Huflert, Forest Research Institute, Warsaw, for their invaluable help in providing the field test facilities in Central Poland and conducting the 1979 field experiments; and Ing. A. Labarriere, Office Nationale des Forêts, Direction Regionale d'Alsace, for providing of field test sites in Eastern France and his helpful advices. We are particularly indebted to Dr. H. R. Buser, Wädenswil, for permitting us to present unpublished results of the chemical pheromone analysis; S. Rauscher, Wädenswil, for his aid in gaining the pheromone extracts and for the statistical analysis of the field data; and Dr. C. J. Sanders, Canadian Forestry Service, Sault Ste. Marie, for generously supplying the synthetic formulations used in the tests of 1975 and for his most valuable comments on a draft of the manuscript. Appreciation is extended to Drs. H. G. Amsel and W. Speidel, Landessammlung für Naturkunde, Karlsruhe, for identification of captured specimens; and Miss J. Schuler, Seewiesen, for skillful technical assistance.

[1] H. Eidmann, Anz. Schädlingskde. **22,** 103 (1949).
[2] J. Patocka, Die Tannenschmetterlinge der Slowakei, Verlag Slowak. Akad. Wiss., Bratislava 1960.
[3] H. Bogenschütz, Tortricinae, Die Forstschädlinge Europas, **Vol. III,** (W. Schwenke, ed.), p. 59, Verlag P. Parey, Hamburg 1978.
[4] T. N. Freeman, Canad. Entomol. **99,** 449 (1967).
[5] G. Stehr, Canad. Entomol. **99,** 456 (1967).
[6] G. T. Harvey and G. Stehr, Canad. Entomol. **99,** 464 (1967).
[7] C. J. Sanders, Science 171, 911 (1971).
[8] C. J. Sanders, G. E. Daterman, and T. J. Ennis, Canad. Entomol. **109,** 1230 (1977).
[9] J. Franz, Z. Pflanzenkrankh. Pflanzenschutz **64,** 578 (1957).
[10] C. J. Sanders, Canad. Entomol. **110,** 43 (1978).
[11] C. J. Sanders, Chemical Ecology: Odour Communication in Animals (F. J. Ritter, ed.), p. 281, Elsevier, Amsterdam 1979.
[12] C. J. Sanders, Mitt. Schweiz. Entomol. Ges. **52,** 227 (1979).
[13] J. Weatherston, W. L. Roelofs, A. Comeau, and C. J. Sanders, Canad. Entomol. **103,** 1741 (1971).
[14] C. J. Sanders, G. E. Daterman, R. F. Shepherd, and H. Cerezke, Canad. Entomol. **106,** 157 (1974).
[15] C. J. Sanders and J. Weatherston, Canad. Entomol. **108,** 1285 (1976).
[16] G. E. Daterman, R. G. Robbins, T. D. Eichlin, and J. Pierce, Canad. Entomol. **109,** 875 (1977).
[17] H. Bogenschütz and C. J. Sanders, unpublished results.
[18] T. D. Fitzgerald, A. D. St. Clair, G. E. Daterman, and R. G. Smith, Envir. Entomol. **2,** 607 (1973).
[19] W. L. Roelofs and A. Comeau, J. Econ. Entomol. **63,** 969 (1970).
[20] H. Arn, C. Schwarz, H. Limacher, and E. Mani, Experientia **30,** 1142 (1974).
[21] A. S. Kovaleva, L. L. Ivanov, I. B. Piatnova, B. G. Kovalev, V. A. Minyailo, and A. K. Minyailo, Proc. 2nd All-Union Symp. on Insect Chemoreception, p. 159, Vilnius 1975.
[22] H. Arn, C. R. 5e Symp. Lutte Integrée en Vergers, OILB, p. 285 (1975).
[23] S. Voerman, Chemical Ecology: Odour Communication in Animals (F. J. Ritter, ed.), p. 353, Elsevier, Amsterdam 1979.
[24] E. Priesner, H. J. Bestmann, O. Vostrowsky, and P. Rösel, Z. Naturforsch. **32 c,** 979 (1977).
[25] E. Priesner, Chemical Ecology: Odour Communication in Animals (F. J. Ritter, ed.), p. 57, Elsevier, Amsterdam 1979.
[26] E. Priesner, Ann. Zool. Ecol. Anim. **11,** (1979, in press).
[27] H. Arn, E. Priesner, H. Bogenschütz, H. R. Buser, D. L. Struble, S. Rauscher, and S. Voerman, Z. Naturforsch. **34 c,** 1281 (1979).
[28] B. Frerot, E. Priesner, and M. Gallois, Z. Naturforsch. **34 c,** 1248 (1979).
[29] H. R. Buser and H. Arn, unpublished results.
[30] H. R. Buser and H. Arn, J. Chromatogr. **106,** 83 (1975).

[31] H. Arn, S. Rauscher, and A. Schmid, Mitt. Schweiz. Entomol. Ges. **52,** 49 (1979).
[32] H. Bogenschütz *et al.*, in preparation.
[33] J. W. S. Bradshaw *et al.*, in preparation.
[34] E. Priesner, H. Bogenschütz, W. Altenkirch, and H. Arn, Z. Naturforsch. **33 c,** 1000 (1978).
[35] E. Priesner, Proc. Int. Symp. on Insect Neurobiology and Insecticide Action, p. 359, London 1979.
[36] H. Arn, H. R. Buser, and S. Rauscher, Abstract, 3rd Meeting OILB Working Group on Insect Pheromones, Wädenswil 1979.
[37] H. Arn, E. Städler, and S. Rauscher, Z. Naturforsch. **30 c,** 722 (1975).
[38] D. L. Struble, H. R. Buser, H. Arn, and G. E. Swailes, J. Chem. Ecol. **6,** (1980, in press).
[39] W. L. Roelofs and J. Tette, Nature **226,** 1172 (1970).
[40] A. S. Hill and W. L. Roelofs, J. Chem. Ecol. **5,** 3 (1979).
[41] W. L. Roelofs and H. Arn, Nature **219,** 513 (1968).
[42] Y. Tamaki, H. Noguchi, T. Yushima, C. Hirano, K. Honma, and H. Sugawara, Kontyu **39,** 338 (1971).
[43] G. M. Meyer, F. J. Ritter, C. J. Descoins, A. K. Minks, and S. Voerman, Science **175,** 1469 (1972).
[44] A. K. Minks, W. L. Roelofs, F. J. Ritter, and C. J. Persoons, Science **180,** 1073 (1973).
[45] W. L. Roelofs, A. Hill, R. T. Cardé, J. Tette, H. Madsen, and J. Vakenti, Envir. Entomol. **3,** 747 (1974).
[46] C. J. Persoons, A. K. Minks, S. Voerman, W. L. Roelofs, and F. J. Ritter, J. Insect Physiol. **20,** 1181 (1974).
[47] J. R. Miller, T. C. Baker, R. T. Cardé, and W. L. Roelofs, Science **192,** 140 (1976).
[48] W. L. Roelofs, A. Hill, A. M. Cardé, R. T. Cardé, H. Madsen, and J. Vakenti, Envir. Entomol. **5,** 362 (1976).
[49] W. L. Roelofs, A. M. Cardé, A. Hill, and R. T. Cardé, Envir. Entomol. **5,** 649 (1976).
[50] W. L. Roelofs, R. F. Lagier, and S. C. Hoyt, Envir. Entomol. **6,** 353 (1977).
[51] R. T. Cardé, A. M. Cardé, A. S. Hill, and W. L. Roelofs, J. Chem. Ecol. **3,** 71 (1977).
[52] A. S. Hill, R. T. Cardé, W. M. Bode, and W. L. Roelofs, J. Chem. Ecol. **3,** 369 (1977).
[53] C. J. Persoons and F. J. Ritter, Z. angew. Entomol. **77,** 342 (1977).
[54] H. Sugie, K. Yaginuma, and Y. Tamaki, Appl. Entomol. Zool. **12,** 69 (1977).
[55] C. Descoins and B. Frerot, Chemical Ecology: Odour Communication in Animals (F. J. Ritter, ed.), p. 181, Elsevier, Amsterdam 1979.
[56] B. Frerot, M. Gallois, and J. Einhorn, C. R. Acad. Sci. Paris, Sér. D, **288,** 1611 (1979).
[57] A. Comeau and W. L. Roelofs, Entomol. Exp. Appl. **16,** 1991 (1973).
[58] W. L. Roelofs, J. P. Tette, E. F. Taschenberg, and A. Comeau, J. Insect Physiol., **17,** 2235 (1971).
[59] G. Biwer, C. Descoins, M. Gallois, E. Priesner, J. P. Chambon, G. Genestier, and M. Martinez, Ann. Zool. Ecol. Anim. **10,** 129 (1977).
[60] L. L. Sower, G. E. Daterman, C. Sartwell, and H. T. Cory, Envir. Entomol. **8,** 265 (1979).
[61] H. Arn, S. Rauscher, H. R. Buser, and W. L. Roelofs, Z. Naturforsch. **31 c,** 499 (1976).
[62] P. Saglio, C. Descoins, M. Gallios, D. Jaouen, and J. Mercier, C. R. Réunion sur les Phéromones Sexuelles des Lépidoptères, Publ. INRA, p. 188, Bordeaux 1976.
[63] E. R. Mitchell and R. E. Doolittle, J. Econ. Entomol. **69,** 324 (1976).
[64] R. L. Jones and A. N. Sparks, J. Chem. Ecol. **5,** 721 (1979).
[65] H. Noguchi, Y. Tamaki, and T. Yushima, Appl. Entomol. Zool. **14,** 225 (1979).
[66] J. Franz, Z. Angew. Entomol. **27,** 345 (1940).
[67] C. A. Miller and G. A. McDougall, Canad. J. Zool. **51,** 853 (1973).