(*10*), and probably indicates the limitations of the sensitivity of this assay. Detection of MuLV antigen at 15 months of age in 13 percent of the grossly normal C57BL mice treated with methylcholanthrene suggests that the chemical may activate the latent leukemogenic virus prior to the development of overt lymphoma. The complement-fixing MuLV antigens have been demonstrated in nonlymphomatous spleens of other strains of mice, with the incidence approximating 100 percent in strains with high incidences of leukemia (*11*). To determine the temporal relation between the activation of latent leukemia virus and the development of overt lymphoma, mice in the groups inoculated with 10 μg of methylcholanthrene, 100 μg of methylcholanthrene, and trioctanoin alone were bled from the orbital sinus at 6, 12, 20, 37, 52, and 62 weeks after inoculation. The serums were tested for complement-fixing antibody to murine leukemia virus against a complement-fixing antigen prepared from subcutaneously transplanted lymphomas originally induced by radiation in C57BL/6 mice (*8*). This antigen reacts with rat antiserum (group reactive) to MuLV and is a sensitive and specific complement-fixing antigen for detection of antibody to indigenous leukemia virus in C57BL mice. Six to 33 percent of the serums obtained at week 12 and week 20 from mice treated with methylcholanthrene had antibody to murine leukemia virus (Table 2). Most of the serums had titers of 1 : 10 to 1 : 20. Since the lymphomas induced by methylcholanthrene did not occur until after week 20, the early and repeated detection of complement-fixing antibody to murine leukemia virus indicated the activation of leukemia virus prior to the development of overt lymphoma. Two of the 50 mice inoculated with trioctanoin showed antibody at 6 weeks, but antibody could not be detected again until 52 weeks.

In the mice inoculated with methylcholanthrene, 21 extracts contained MuLV antigen—11 lymphoma extracts and 10 extracts of normal spleen-thymus (Table 1). Of the 21 mice which yielded the antigen-positive extracts, 19 had previously demonstrable complement-fixing antibody. Of the 11 mice with antigen-positive lymphomas, all had antibody to murine leukemia virus in two or more of their serum samples.

Of 37 lymphomas induced by methylcholanthrene, urethan, or diethylnitrosamine, 21 contained mouse leukemia viral antigen, and lymphomas induced by methylcholanthrene were preceded by the development of antibody to murine leukemia virus in the serum. The finding of MuLV antigen and antibody in a few control mice over 1 year old suggests that the latent leukemia virus may occasionally become spontaneously activated. Treatment with methylcholanthrene accelerated and increased the magnitude of viral activation and the subsequent development of lymphoma.

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Synthetic Juvenile Hormone: Induction of Sex Pheromone Production in Ips confusus

Abstract. *Topical application of 25, 50, or 100 micrograms of 10,11-epoxyfarnesenic acid methyl ester in peanut oil induced male* Ips confusus *to produce sex pheromone in the hindgut Malpighian tubule region. Twenty-four hours after treatment of male beetles with 100 micrograms of hormone, their hindgut Malpighian tubule extract was more attractive to female beetles in a laboratory bioassay than was extract from males producing pheromone naturally in ponderosa pine logs.*

Within 4 to 6 hours after entering host logs (*1*), male *Ips confusus* (Coleoptera: Scolytidae) begin to produce three sex pheromone compounds (*2*) in the hindgut Malpighian tubule region (*3*). There is then a rapid degeneration of the flight muscles, and the insects approach reproductive maturity (*4*). Degeneration of the flight muscles can be induced in both sexes by topical application of a synthetic juvenile hormone (*5*), a fact which suggests that other events associated with reproduction, such as sex pheromone production, might be similarly induced. In cockroaches, sex pheromone production is dependent on the presence of the corpora allata (*6*), and, in male locusts, the corpora allata control the production of a maturation pheromone (*7*). We report that synthetic juvenile hormone (*8*) (10,11-epoxyfarnesenic acid methyl ester) (EFA) induces male *I. confusus* to produce sex pheromone.

We used beetles which had emerged from a colony maintained in ponderosa pine logs in cages. Groups of 12 to 20 male beetles were treated (i) with EFA in peanut oil applied topically to the abdominal venter; (ii) with peanut oil applied in the same manner; or (iii) with introduction into fresh pine logs to induce normal pheromone production (*1*). Topical applications were made with capillary pipettes, and insects so treated were held on moist lichen in closed glass jars at room temperature. After a specified time, usually 24 hours, all insects were removed from the jars or excised from the host logs. Their hindguts with Malpighian tubules were dissected out and extracted in benzene (one gut to 0.025 ml); the extract was bioassayed with groups of 10 to 16 female beetles in an open stage, multiple airstream olfactometer (*1*). Each extract attracting more than 20 percent of the test females was serially diluted and bioassayed until a nonattracting stimulus was attained.

After 24 hours, guts from males treated with 100 μg of EFA were highly attractive to female beetles (Table 1). When bioassayed at a stimulus concentration of 2 male equivalents (two guts in approximately 0.05 ml of benzene), these guts were more attractive than guts from males that were allowed to produce pheromone naturally in logs. However, at a stimulus concentration of 0.02 male equivalents, extract from males which had produced pheromone in logs was significantly attractive (20.3 percent), whereas extract from males treated with EFA elicited only a trace response (Table 1). Evidently, males boring in logs produced greater amounts of the pheromone compounds than males treated with the hormone. There was a progressively weaker response as the EFA dose decreased, and after treatments with 10 μg no pheromone activity could be detected in the hindguts.

Only a trace response by females occurred to guts from control males treated with pure peanut oil (Table 1). In certain species of bumblebees and wasps, farnesol acts as an attractive pheromone (*9*). Therefore, we tested the attractiveness of 100 μg of EFA in 0.05 ml of benzene, but only 1 of 71 females responded positively to this stimulus.

Eighteen hours after treatment of male beetles with 100 μg of EFA, their gut extract elicited a significant response of 26.6 percent (25 of 94 females) when bioassayed at 2 male equivalents. However, similar treatments failed to induce pheromone production after 3, 6, and 12 hours in 13, 13, and 12 male beetles, respectively. Synthetic hormone applied topically apparently required more than 12 hours to elicit a response, whereas male beetles excised from host logs 3 hours after introduction possessed highly attractive hindguts. Evidently, boring into and feeding on the bark of host logs provide a stimulus which promotes rapid synthesis or release of the insect's juvenile hormone, or both, thereby inducing a more immediate physiological response. One suggestion is that topically applied EFA is slowly absorbed and converted to a hormonal terpene identical or closely related to "paper factor" (juvabione) (*10*) or cecropia juvenile hormone (*11*), while beetles in the bark receive a "paper factor" type stimulus directly from the host tree.

In nature, *I. confusus* males produce pheromone while feeding and release it by discharge of fecal pellets (*3*). Moreover, the terpene alcohol pheromone compounds are closely related to naturally occurring constituents in the host tree. Therefore, one hypothesis explaining the biogenesis of scolytid pheromones is that host tree precursor compounds are ingested and metabolized to produce pheromone in the hindgut (*12*). The fact that males treated with EFA were not allowed to ingest fresh host material supports the alternative hypothesis (*12*) that the pheromone compounds of *I. confusus* are true secretions. A further possibility is that absorbed EFA is metabolically converted by the beetle to form one or more of the known sex pheromone compounds (*2*).

Table 1. Response of *I. confusus* females to benzene extracts of hindguts from males 24 hours after topical application of 10,11-epoxyfarnesenic acid methyl ester (EFA) in peanut oil compared with response to hindgut extracts from males treated with peanut oil, or excised from host logs.

Stimulus (male equivalents)	Tested (No.)	Responders (No.)	Response (%)	Probability (χ^2)
1 μl of peanut oil				
2.0	103	8	7.8	Control
In log				
2.0	141	95	67.3	<.001
0.2	81	29	35.8	<.001
0.02	79	16	20.3	<.05
0.002	79	3	3.8	
100 μg of EFA in 1 μl of peanut oil				
2.0	101	82	81.2	<.001
0.2	119	57	47.9	<.001
0.02	102	10	9.8	<.70
0.002	102	1	1.0	
0.5 μl of peanut oil				
2.0	160	7	4.7	Control
50 μg of EFA in 0.5 μl of peanut oil				
2.0	80	41	51.3	<.001
0.2	78	22	28.2	<.001
0.02	80	4	5.0	<.90
25 μg of EFA in 0.5 μl of peanut oil				
2.0	80	12	15.0	<.01
10 μg of EFA in 0.5 μl of peanut oil				
2.0	83	2	2.4	

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Satellite-like Particle of Tobacco Ringspot Virus That Resembles Tobacco Ringspot Virus

Abstract. *A satellite-like nucleoprotein serologically indistinguishable from multicomponent tobacco ringspot virus, also resembles it in size, shape, and electrophoretic mobility. Although the protein shell of the nucleoprotein is similar to, if not identical to that of tobacco ringspot virus, each shell of the nucleoprotein contains many small strands of nucleic acid, which replicate only in mixed infections with the virus.*

The satellite virus (SV) of tobacco necrosis virus (TNV) was originally named "satellite" by Kassanis (*1*) because "it is always in association with, and dependent upon, the large virus particles" (that is, TNV). Similarly, the nucleoprotein, which is the subject of this report, appears to be dependent upon the multiplication of tobacco ringspot virus (TRSV), but I refer to it as "satellite-like" (SL) because its functional relation to TRSV has not yet been determined. The new particle, SL-TRSV, has the same shape and size as TRSV and is serologically related to TRSV, whereas SV-TNV is smaller than TNV and does not appear to be serologically related to TNV (*2*).

The nucleoprotein SL-TRSV was discovered in bean plants previously inoculated with a laboratory source of TRSV. The new component appears to be noninfectious in both cowpea *Vigna sinensis* (Torner) Savi, cv. Blackeye and bean *Phaseolus vulgaris* L. cv. Black Valentine plants, which are hosts