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ATTRACTIVE AND INHIBITORY PHEROMONES PRODUCED IN THE BARK BEETLE, *Dendroctonus brevicomis*,[1] DURING HOST COLONIZATION: Regulation of Inter- and Intraspecific Competition

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Abstract—Quantities of attractive (*exo*-brevicomin and frontalin) and inhibitory (*trans*-verbenol, verbenone, and ipsdienol) pheromones were monitored in both sexes of *Dendroctonus brevicomis* during their colonization of a ponderosa pine. Verbenone was found in males in the greatest amounts at the time of landing, and it declined more rapidly than the other pheromones in either sex. The amounts of frontalin and *exo*-brevicomin in males and females, respectively, increased after initial boring within the host but began to decline after mating. The quantity of *trans*-verbenol in both sexes (females had significantly more) declined more gradually than *exo*-brevicomin, frontalin, and verbenone. Ipsdienol was found only in males during the initial stages of attack when encountering the resin. It is suggested that along with a general decline in all pheromonal components, a sufficient change in the ratio of the attractive pheromones to an inhibitory pheromone, *trans*-verbenol, may play a role in termination of aggregation. *trans*-Verbenol may also function along with verbenone and ipsdienol in limiting the density of attack and thus intraspecific competition. These inhibitory pheromones also appear to cause several competing species of bark beetle to avoid landing in areas infested with *D. brevicomis*, even when their own pheromone is present.

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INTRODUCTION

Intraspecific competition is assumed to occur in most species which utilize a limited food resource (Pianka, 1976), such as a "weakened" and beetle-infested host tree. Furthermore, it is always advantageous for an individual to avoid competition (both intra- and interspecific) whenever possible (Pianka, 1976). As expected from ecological theory, reports of intraspecific competition in several bark beetle species in the genera *Ips, Dendroctonus, Scolytus,* and *Tomicus* have shown that brood output per female decreases at higher densities on the bark (Miller and Keen, 1960; Cole, 1962; Reid, 1963; Eidmann and Nuorteva, 1968; Ogibin, 1972; Svihra, 1972; Berryman, 1974; Mayyasi et al., 1976).

The western pine beetle, *Dendroctonus brevicomis* LeConte, begins the attack of its host, ponderosa pine (*Pinus ponderosa* Doug. ex. Laws.), when a female penetrates the bark and excavates a gallery in the phloem. The attractive pheromone component, *exo*-brevicomin, is produced in females and released with the frass, a mixture of fecal pellets and host material (Silverstein et al., 1968). A male attracted to the entrance tunnel soon releases frontalin (Kinzer et al., 1969; Pitman et al., 1969; Browne et al., 1979) which, together with *exo*-brevicomin and a major component of host resin, myrcene, significantly enhances the attraction of both sexes (Wood et al., 1976; Bedard et al., 1980b) to initiate the mass attack (concentration phase). Several compounds in the beetles that inhibit the attraction response have been suggested to play a role in regulating attack density and intraspecific competition (Byers and Wood, 1980; Byers, 1982, 1983a). Verbenone alone (Renwick and Vité, 1970; Bedard et al., 1980a), verbenone plus *trans*-verbenol (Bedard et al., 1980a), and *trans*-verbenol alone (Bedard et al., 1980a; Byers, 1983a) have been shown to inhibit the response of *D. brevicomis* to the attractive components in the field. In a recent report, Byers (1982) found that only males could synthesize (+)-ipsdienol from myrcene vapors and that ipsdienol inhibited the attraction of both sexes to their pheromone components in the field. Emergent females contain *trans*-verbenol and emergent males *trans*-verbenol and verbenone (Renwick, 1967; Vité and Renwick, 1970; Byers, 1983c), but the amounts in beetles during the period of host colonization have not been determined.

D. brevicomis appears to limit intraspecific competition for food and space by regulating the density of attack. Miller and Keen (1960) have

summarized several early reports on attack densities and found them to range from 5.9 to 23.2 per 0.1 m^2 or "always within certain limits." Production and release of attractants by an individual must be synchronized at the population level with a mechanism of regulating denisty (possibly olfactory/pheromonal) to prevent overcrowding and to terminate aggregation. One theory proposed by Renwick and Vité (1970) and McNew (1970) suggested that males may release verbenone during the latter stages of attack which would reduce response to attractive components in order to terminate the aggregation of beetles. However, Byers and Wood (1980) questioned this theory because they found that males contained the largest quantities of verbenone at the beginning of colonization, and as the attractiveness of the infested log decreased over a 5-day period, the amount of verbenone declined along with *exo*-brevicomin and frontalin. They suggested that verbenone does not terminate the mass attack but may regulate the density of attack at close range, while a reduction in *exo*-brevicomin and frontalin caused termination of long-range attraction (Byers and Wood, 1981).

Some of the behavioral chemicals produced by *D. brevicomis* may be used to avoid competition by other bark beetle species that also inhabit ponderosa pine phloem and frequently occur together in the same pine tree (Miller and Keen, 1960). For instance, the attraction of *Ips paraconfusus* Lanier to natural pheromone was shown by Byers and Wood (1980, 1981) to be inhibited by volatiles from *D. brevicomis*-infested logs and also by verbenone. (+)-Ipsdienol produced by males of *D. brevicomis* (Byers, 1982) was shown to inhibit the response of *I. pini* (Say) to natural pheromone (Birch et al., 1980). Therefore, in order to better understand the mechanisms that may regulate attack density, terminate aggregation, and reduce interspecific competition, we wanted to determine the quantitative relationships between the above behavioral chemicals found in both sexes of *D. brevicomis* during their colonization of a pine tree and compare them to landing rates of the beetle on the tree.

METHODS AND MATERIALS

A mass attack of *D. brevicomis* was induced on an apparently healthy ponderosa pine to determine when certain behavioral chemicals were present within each sex during host colonization (August 25–September 27, 1978). The ponderosa pine (50.9 cm diam at 1.5-m height) was located in the Sierra National Forest near Oakhurst, California, at 1000 m elevation in a nearly pure stand of this species. A pulley with climbing rope was attached to a tree limb 13 m above ground so that by means of counterbalancing weights and a sling one could hoist oneself up the trunk to obtain ready access to any sampling area of the tree without damaging it. Flat sticky-traps (15.25 × 15.25

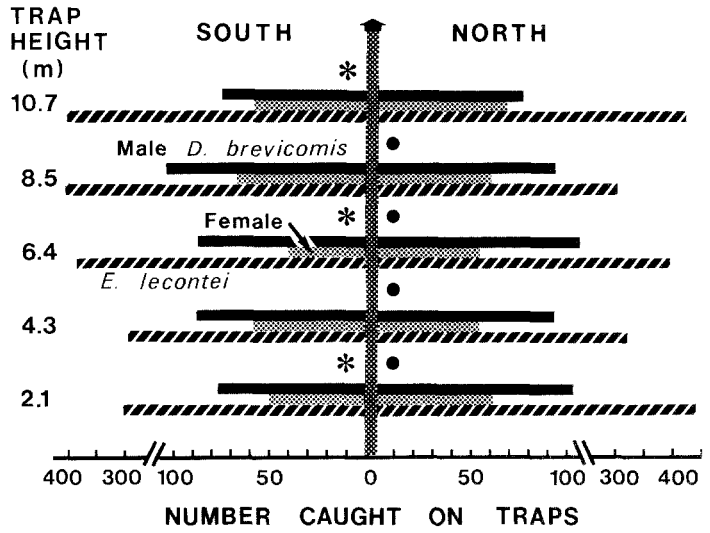

FIG. 1. Number of male and female *Dendroctonus brevicomis* and *Encolerus lecontei* caught on sticky traps at several heights and directions on a ponderosa pine tree from August 25 to September 27, 1978. The tree was baited with synthetic pheromone of *D. brevicomis* (*exo*-brevicomin, frontalin, and myrcene) for one day (August 25) at three heights indicated with an asterisk. *D. brevicomis* were obtained from bark sampled between the traps at heights indicated with darkened circles to determine pheromone content in hindguts.

cm) made from 6.3-mm wire mesh and coated with Stickem Special® were placed on nails 2 cm away from the bark of the tree in north- and south-facing directions at five heights on August 24, 1978 (Figure 1). On the next day three glass salt-shakers were placed on the tree; each one contained three glass tubes filled with neat *exo*-brevicomin, frontalin, (both racemic) and myrcene (all Chemical Samples Co., Cleveland, Ohio), each one releasing about 2 mg/day (Byers and Wood, 1980). The following morning these baits were removed. *D. brevicomis* and *Enoclerus lecontei* Wolc. (Coleoptera:Cleridae) were collected daily for 11 days after the initial attack induced by the baiting (Figure 1).

Chi-square tests were used to compare sex ratios of catch between various trap levels and between north- and south-facing traps. The extent and duration of male stridulation, length of gallery construction, egg maturation within females, and host resin exudation were noted during the first part of the attack sequence from August 25 to September 5 (Table 1). On September 20 the attack density was recorded in several areas between the 2.1- and 10.7-m heights.

D. brevicomis were collected live as they walked on the bark surface at two times in the afternoon on the first day of baiting, August 25, and three

TABLE 1. RELATIONSHIPS BETWEEN AGGREGATION OF *Dendroctonus brevicomis* and *Enoclerus lecontei* ON PONDEROSA PINE AND MALE STRIDULATION, EGG MATURATION, AND GALLERY LENGTH OF *D. brevicomis*, AND HOST RESIN EXUDATION (AUGUST–SEPTEMBER 1978)

	Dendroctonus brevicomis					
Days after initial attack	Trap catch ♂:♀	Male stridulation	Gravid ♀♀	Range of gallery lengths (cm)	*E. lecontei*	Resin exudation
Aug. 24	0	0	0	0	0	0
Aug. 25[a]	57:34	++[b]	0	0	5	0
Day 1	97:53	+++	0	*[d]	15	0
Day 2	151:120	+++	0	*	15	+[e]
Day 3	255:153	+++	+[c]	*	455	+++
Day 4	172:111	++	+++	0.6–2.5	614	+++
Day 5	54:27	++	+++	1.2–3.8	519	+
Day 6	25:15	+	+++	2.0–5.1	302	0
Day 7	29:18	+	+++	2.5–6.2	483	0
Day 11	rain	0	+++	< 9.0	rain	0

[a] The tree was baited with *exo*-brevicomin, frontalin, and myrcene for one day only (August 25) which elicited a mass attack of *D. brevicomis*.

[b] Relative levels (from none = 0 to maximum = +++ as judged by J.A. Byers) of male stridulation, number of eggs in dissected females, and rate of resin flow.

[c] Little or no feeding occurred in *D. brevicomis* prior to August 28 (day 3) when phloem was first observed in hindguts of most males and females. Approximate percentage levels of dissected females with eggs, from none = 0 to >95% = +++.

[d] Not measured as beetles were taken from resin tubes, outer bark area, or after just penetrating the phloem.

[e] Relative rates of resin flow from none = 0 to maximum = +++ (judged by J.A. Byers).

times one day later (results pooled and shown for August 26 in Figure 2) and from galleries (lengths in Table 1) at 1200 and 1800 hr of each day (2–6, 8, 11, 14, 20, and 27 days) after the initial attack. The beetles were obtained in about equal portions from 2-m sections of the tree (Figure 1). Within 1 hr of collections, the beetles from each date and time were separated by sex, and the hindguts (about 20–25) of each group were excised and extracted with 300 μl diethyl ether (Byers and Wood, 1980). The amounts of pheromones present in these gut extracts were analyzed by GLC (3.6 m × 2 mm ID glass column of Ultrabond II on 100/120 mesh at 60° and 110° C and N_2 flow of 30 ml/min; 1.8 × 2 mm ID glass column of 3% Apiezon L on 100/120 Gas Chrom Q at 100° C and N_2 flow of 12 ml/min). Authentic samples of verbenone (Chemical Samples Co.), frontalin, *exo*-brevicomin, myrcene, and *trans*-verbenol (Glidden Organics, Jacksonville, Florida) were each GLC purified >99% and used for comparison to the gut extracts. Exponential regressions of the amounts of

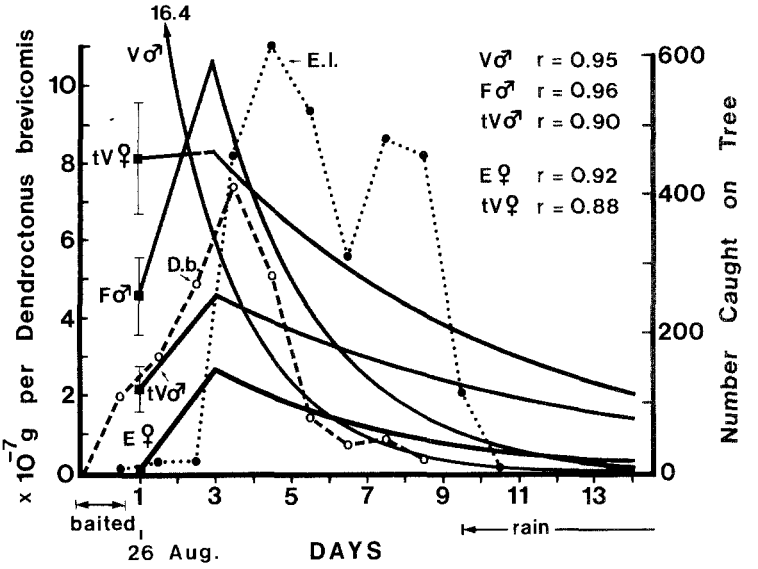

FIG. 2. Average content of the pheromones, verbenone (V), frontalin (F), *trans*-verbenol (tV), and *exo*-brevicomin (E), in guts of male and female *Dendroctonus brevicomis* feeding in a ponderosa pine tree. Daily catch of *D. brevicomis* (D.b.) and *E. lecontei* (E.l.) indicated by broken and dotted lines, respectively. Exponential regression curves are from data collected on days 3–20 except for V (1–20), V: $Y = 26.4e^{-0.48X}$, F: $Y = 31.4e^{-0.36X}$, tV♂: $Y = 6.4e^{-0.11X}$, E: $Y = 5.0e^{-0.20X}$, tV♀: $Y = 12.3e^{-0.13X}$. Brackets represent ± SEX for $N = 5$ on day 1 (August 26) for nonfeeding, recently landed *D. brevicomis*. Tree baited with E + F + myrcene for time shown to attract beetles to begin colonization.

pheromones in each sex through time were calculated from the GLC analyses (as indicated in Figure 2) since this type of function might be expected for release of volatile compounds from a substrate.

One bark beetle extract for each sex collected at 1800 hr on days 2, 8, 11, 14, and 20, and the above authentic standards as well as ipsdienol (Chemical Samples Co., GLC purified >99.5%) were further analyzed by GC-MS to confirm the identifications and relative amounts obtained with GLC. A Finnigan 4023 gas chromatograph–mass spectrometer–computer system with a 60-m Carbowax 60 N column (J & W) was used isothermally at 180° C with a helium carrier gas flow rate of 28 cm/sec using a Grob injection technique. A compound was considered identified if the mass spectrum and retention time of the unknown and the standard were similar (retention time ±5 sec). For quantitative analyses, ion characteristics of the spectra of the pheromonal standard were measured in comparison to the unknown at the appropriate retention times (min:sec):frontalin (*m/e* 142; 4:48); *exo*-

brevicomin (*m/e* 114; 5:02); ipsdienol (*m/e* 85; 6:51); *trans*-verbenol (*m/e* 109; 7:10); and verbenone (*m/e* 107; 7:59).

RESULTS

There were no significant differences ($P > 0.1$) in sex ratios of total catch in all comparisons between the five heights or between the north and south directions on the tree (Figure 1). Also, no significant differences were observed in the proportion of males to females caught each day from the initial attraction induced by the pheromone bait, day 0, to day 7 ($P > 0.1$). A total of 866 males and 557 females were caught during the entire period yielding a sex ratio of 1.55 (1.35–1.79, 99% binomial confidence limits). This ratio was significantly different from 1 ($P < 0.001$), the sex ratio of broods as they emerge from infested trees (Miller and Keen, 1960; Stephen and Dahlsten, 1976). The catch of *E. lecontei,* a predator, was substantially higher than *D. brevicomis.* The catch of *E. lecontei* also appears to be uniformly distributed in areas of the tree where traps were placed (Figure 1).

No beetles were caught until placing the pheromone baits on the tree. The aggregation was initiated, and it continued after the baits were removed for about eight days during a period of warm sunny weather (daily highs of 30–34° C). This concentration phase was essentially finished on day 8 after the initial attack, at which time only 4.7% as many beetles were caught as on the peak catch on day 3. Flight ceased the next day when rain and cooler weather intervened for over a week. The sex ratio in galleries was usually 1 : 1 (from day 4), and no males were found alone. The concentration phase was synchronized over a relatively short period of time as indicated by the ranges of gallery lengths and the large proportion of the total catch (66%) during the first four days (Table 1). Females began to produce mature eggs at about the time phloem was first observed in hindguts of both males and females. Male stridulation was heard almost as soon as females penetrated the bark, reaching a maximum during the peak aggregation three days after baiting and then diminishing as the catch decreased (Table 1). Host resin exudation began when females reached the phloem–xylem interface and apparently caused significant mortality of *D. brevicomis* since many "pitch tubes" contained from one to three or more dead beetles of either sex. However, the resin flow appeared to cease by day 6, and the death of the tree was confirmed the following year. Evidence of successful brood production and emergence holes were also observed. The final density of attack was about 15.1/0.1 m^2 (range 11.8–17.2, $N = 8$) and appeared relatively uniform in all areas sampled.

The peak aggregation of *D. brevicomis* was correlated with higher quantities of *exo*-brevicomin, frontalin, and other pheromone components in their guts (Figure 2). Males contained large quantities of verbenone (1.6

μg/male) when they landed on the tree, and the levels appeared to decline the most rapidly of all pheromone components measured (Figure 2). Frontalin levels in males that had just landed may increase upon exposure to resin or feeding, but its depletion was also rapid after 2–3 days. Females contained only trace or undetectable amounts of *exo*-brevicomin upon landing on the tree but apparently synthesize the component during feeding (Figure 2). *trans*-Verbenol was found in both sexes upon landing, although females had significantly more (Figure 2). The decline in *trans*-verbenol content in hindguts appeared to be the most gradual of all the components measured.

The identifications and relative amounts of pheromone components in gut extracts as determined by GLC (Figure 2) were confirmed by GC-MS. Females apparently did not have ipsdienol (<0.1 ng/female) during the attack, while males contained the largest amount (80 ng/male) when in the resin (day 2) but less on days 8 and 11 (5 and 7 ng, respectively) and none (<0.1 ng) was detected in the male extract on day 14.

The aggregation of *E. lecontei* on the tree appeared to immediately follow the peak aggregation of *D. brevicomis,* but the catch of this predator remained high for several days after attraction of the bark beetle had essentially ended. The attractive components released on the tree on day 0 (August 25) appeared to have resulted in very little attraction of *E. lecontei.*

DISCUSSION

Intraspecific Competition. The final attack density of *D. brevicomis* on the ponderosa pine was within normal limits (Miller and Keen, 1960), and the range of densities in various sections indicated a rather uniform distribution throughout the bole. The duration of landing and mass attack on the tree also was within the ranges observed in earlier studies in California (Miller and Keen, 1960; Byers and Wood, 1980; Bedard et al., 1983). The catch was highest when galleries were under 2.5 cm which agrees with Bedard et al. (1983), who found logs most attractive when galleries ranged from 2 to 5 cm with both sexes present.

Landing places with respect to height and direction on the tree appeared similar for *E. lecontei* and both sexes of *D. brevisomis.* However, more males were caught than females (1.55:1) and this ratio was significantly different from the brood emergence ratio (1:1). This may have been due to (1) the absolute sex ratio in the area, (2) a sexual difference in attraction as noted recently for *Ips paraconfusus* (Byers, 1983b), and (3) differential landing activity by the male when "searching" for female entrance holes—as suggested by Stephen and Dahlsten (1976) to explain the higher proportions of male *D. brevicomis* caught on baited trees. The third possibility appears more likely in view of the high numbers of clerids also caught on traps which appeared due to

their high activity with multiple landings in search of prey on the bark. Furthermore, sexual differences in response to attractive components have not been observed in the laboratory olfactometer or in the field, or to infested logs (Byers and Wood, 1980, 1981).

Based on the diameters near the base of the tree (50.9 cm) and at the 10.7 m height (41.2 cm), the ten traps covered at most 1.5% of the bark surface. If extrapolations are made from this figure and the catch, then over 27,000 beetle landings occurred in this area of the tree on day 3 and over 91,000 during the mass attack period. However, only about 4700 beetles would need to land in this part of the tree over the entire period (15.1 attacks/0.1 m^2) if every pair were successful. The discrepancy between the number of galleries and the apparently much higher number of landings, as reflected in trap catch, was probably caused by some combination of the following: (1) multiple landings by beetles, (2) mortlity caused by predators and tree resin, and (3) beetles that visited the tree and did not stay. The amounts of *exo*-brevicomin and frontalin (each 6 mg/day) released from the baits on the first day were similar to what might be expected to be released per day from the 4700 beetles as calculated from the results of Browne et al. (1979) during the early stage of attack (however, the amount of myrcene from baits was about two orders of magnitude less).

Frontalin and verbenone from males, and *trans*-verbenol from both sexes, appear to be released soon after "contact" with the host (cf. Vité et al., 1972) since they are found in large amounts upon landing. However, the production of *exo*-brevicomin and possibly frontalin may be influenced by feeding and/or hormones. *exo*-Brevicomin in females increased as feeding commenced (also found by Pitman et al., 1969; Hughes and Renwick, 1977), while its decline appeared to begin immediately after mating (eggs in females). Earlier, Hughes (1973) found that mating caused a significant decline in the content of *exo*-brevicomin in females feeding in logs of ponderosa pine. Hughes and Renwick (1977) discovered that *D. brevicomis* females produced large quantities of *exo*-brevicomin when treated with juvenile hormone (JH III). The amounts of ipsdienol found only in males during the early stages of colonization are apparently produced after exposure to myrcene precursor in the host resin (Byers, 1982). The long-term decline in *trans*-verbenol levels of beetles during the colonization appears due to its continued but declining production from the α-pinene precursor in the resin and phloem (Hughes, 1973; Byers, 1983a).

The highest quantities of *exo*-brevicomin and frontalin were found in *D. brevicomis* guts when the maximum catch occurred (Figure 2). This indicates that maximal flight attraction resulted from the release of the highest relative amounts of these pheromone components. The relative amounts of various pheromones within the beetles during the colonization presumably reflect the release rates of these compounds in nature. This assumption is probably

correct since feeding and defecation are known to occur at the beginning of colonization (Silverstein et al., 1968), and females must continue to feed to sustain egg production (e.g., 56 eggs contain over 3.4 times the beetles' weight, from Figure 2 in Miller and Keen, 1960). Furthermore, the evaporation rates of compounds of similar volatility from fecal pellets would be expected to be nearly porportional to their mole percent (Raoult's Law, cf. Byers, 1981b). However, absolute release rates are dependent on the number and density of beetles attacking the tree. Therefore, comparisons between dates of relative release rates from the infested tree (Figure 2) cannot be done accurately until the population density has stabilized, a few days after baiting (gallery lengths in Table 1 indicate maximum density was reached after only a few days). Some loss of pheromone components may have occurred during the <1-hr transport of beetles from the tree to the laboratory, although amounts are similar to those found in *D. brevicomis* feeding in logs that had been immediately dissected and extracted (Byers and Wood, 1980; Byers, 1983c).

The quantities of frontalin and *exo*-brevicomin in beetles during the first 3–4 days of colonization (Figure 2) agree with the relative release rates of these components observed by Browne et al. (1979) from beetles for only 1 day and for 3 days in the field. If the average amounts of *exo*-brevicomin and frontalin in beetles are compared to their estimates of release (Browne et al., 1979) for the same period of time (frontalin = 8.6×10^{-7} g/day and *exo*-brevicomin = 4.1×10^{-6} g/day), then males would have a turnover rate of about one gut content per day and females about 20 gut contents per day. It appears that further work is needed before we can have confidence in this type of comparison. However, it does indicate that at least *exo*-brevicomin and *trans*-verbenol were produced over an extended period, otherwise they would have been exhausted after just a few days assuming the above release rates.

Renwick (1967) and Pitman et al. (1969) reported that emergent males contain large amounts of verbenone. Byers (1983a) further showed that the appearance of verbenone in emergent beetles occurred in the absence of host material and that (+)- and (−)-α-pinene did not appear to serve as a precursor, at least at this time. We found verbenone in the largest amounts in males as they landed on the tree, and its content appears to immediately decline and more rapidly than that of any other pheromone component in the hindgut (although ipsdienol was not compared). These results indicate the verbenone is not synthesized ore released in the latter stages of the concentration phase when termination is occurring. This compound was earlier hypothesized to cause termination (Renwick and Vité, 1970; NcNew, 1970). Instead, verbenone is produced before landing and possibly shortly thereafter and may operate as a "close-range" inhibitor (Byers and Wood, 1981) to regulate density of attack, since a "long-range" inhibition at this time would be nonadaptive. Browne et al. (1979) indicated that verbenone is associated with females because they collected the compound from air

passed over females in logs inside steel barrels. Addition of males did not significantly increase the release of verbenone. However, the ratios of verbenone–*exo*-brevicomin and verbenone–frontalin were much less (1 : 106 and 1 : 11.2) than that observed in our study (days 1–4, Figure 2). This discrepancy may be due to the artificial conditions inside the barrels affecting behavior or due to problems of differential entrainment of the volatiles.

Both the increase and decrease in the landing rate, and probably the attraction, of *D. brevicomis* were positively correlated with the presence of *exo*-brevicomin and frontalin in the gut. However, the decline in landing may have been made more precipitous because of a change in the ratio of attractive to inhibitory pheromones. There is evidence that *trans*-verbenol, in combination with verbenone (Bedard et al., 1980a) or by itself (Hughes and Pitman, 1970; Bedard et al, 1980a; Byers, 1983a) inhibits the attraction of *D. brevicomis* to their attractive components. Feeding males contained about half as much *trans*-verbenol as females (Figure 2), which is in contrast to earlier reports that males contained little or none of this compound (Renwick, 1967; Renwick and Vité, 1970; Vité and Renwick, 1970; Pitman et al., 1969). *trans*-Verbenol may function with verbenone and ipsdienol during the aggregation to regulate density of attack. In addition, *trans*-verbenol may inhibit new attacks during the termination phase since it decreased in both sexes more gradually than other components and was the only component still present in significant amounts when the catch decreased to low levels. Therefore, the ratio of *trans*-verbenol to *exo*-brevicomin and frontalin within certain absolute release rates may function to terminate attack as well as regulate attack density and intraspecific competition. In Figure 3 we propose a revised version of the mechansim of attack (Renwick and Vité, 1970) based on the above discussion. However, additional work is needed to establish and further delimit the functions of the inhibitory pheromones, (+)-ipsdienol, verbenone, and (−)-*trans*-verbenol and their interactions with the attractive pheromones, (+)-*exo*-brevicomin and (−)-frontalin.

Several other bark beetles appear to utilize inhibitory pheromones to regulate density and/or termination of attack. *D. pseudotsugae* males release methylcyclohexenone that inhibits response to female-released pheromone (Rudinsky and Michael, 1972; Pitman and Vité, 1974). Male *Trypodendron lineatum* release volatiles that inhibit response to female pheromone (Nijholt, 1973). In *D. frontalis,* males release verbenone which at high release rates inhibits both sexes or males more (Renwick and Vité, 1970; Payne et al., 1978), and may be used in a similar way as in *D. brevicomis.* Another inhibitor, *endo*-brevicomin, from males reduces attraction of flying beetles (Payne et al., 1978). In *I. paraconfusus,* however, attractive pheromene components from males at high release rates appear to have a sex-specific inhibitory effect on males which may function to regulate their attack density (Byers, 1983b). In contrast, the attractive response of *D. brevicomis* does not

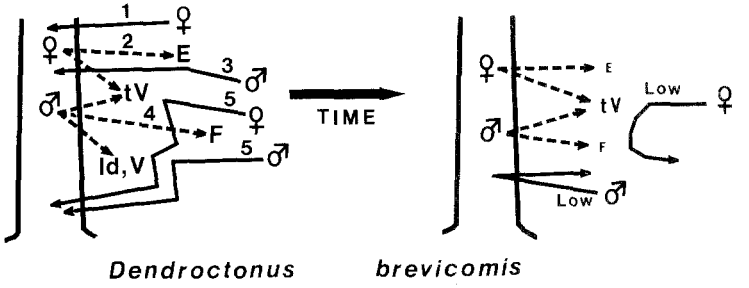

FIG. 3. Theoretical mechanism for regulation of attack density (intraspecific competition) and termination of aggregation in *Dendroctonus brevicomis* during colonization of a ponderosa pine. The female beetle arrives first (1) and bores into the trunk and after feeding produces *exo*-brevicomin, E (2), which primarily attracts males (3). Males, upon locating a female gallery, soon release frontalin, F (4), which synergizes with E to elicit a mass aggregation (5). However, at the same time females and males produce *trans*-verbenol (tV) and males produce verbenone (V) and (+)-ipsdienol (Id). At close range these compounds apparently inhibit the attraction of beetles to E and F (tV appears to primarily affect females while Id and V affect both sexes), which would regulate the attack density. After several days the production and release of E and F diminishes to unattractive levels (at long range). The few females attracted during this latter period may be inhibited from attacking by the still significant, although reduced, levels of tV. The few males would not find any unpaired females and so would continue searching elsewhere.

seem to be inhibited by similar levels of its attractive components released in the same olfactometer in which *I. paraconfusus* was inhibited (Byers and Wood, 1981; Byers, 1983b). Mating appears to cause a reduction in pheromone production in *D. brevicomis* (Hughes, 1973; and our study), *D. frontalis* (Coster and Vité, 1972), *Scolytus multistriatus* (Peacock et al., 1971; Elliott et al., 1975; Gore et al., 1977), and *I. paraconfusus* (Byers, 1981a). Thus, the reduction in the quantity of attractive pheromones is as important as the release of inhibitory compounds during termination of the concentration phase. In fact, a reduction in attractive pheromones is apparently the only olfactory mechanism of termination in some bark beetles, *S. multistriatus* and *I. paraconfusus* (Gore et al., 1977; Byers, 1981a), although these species may have as yet undiscovered short-range olfactory mechanisms.

Interspecific Competition. The inhibitory pheromone components may not only play a role in intraspecific communication but also may serve as interspecific messages (allomones) for reducing possible competition between cohabiting species (Figure 4). *D. brevicomis* and *I. paraconfusus* are sympatric in our study area near Yosemite National Park but compete for the same host tissue with a third beetle, *I. pini,* about 100 km northward where

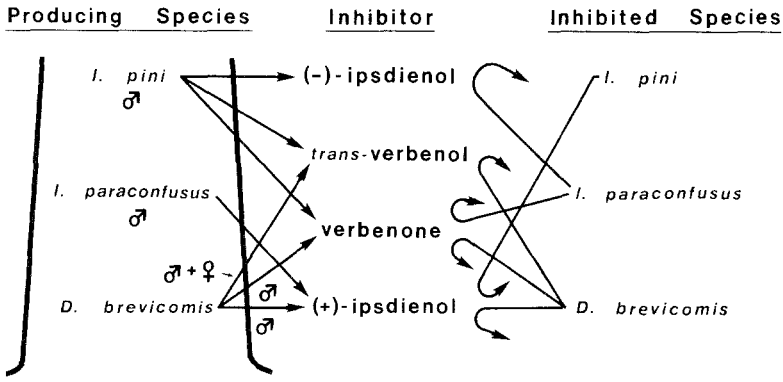

FIG. 4. Inhibition of the attraction response to conspecific pheromone by pheromones/kairo-allomones produced by three sympatric bark beetles, *Dendroctonus brevicomis, Ips paraconfusus,* and *I. pini* which may function to reduce interspecific competition for their host ponderosa pine in California. Response inhibition by *trans*-verbenol, verbenone, and (+)-ipsdienol may also reduce intraspecific competition in *D. brevicomis.*

they all occur continuously along the Sierra to the Cascades and into Oregon. In Oregon and Idaho, *D. brevicomis* and *I. pini* are predominant. Verbenone, in addition to its intraspecific effects, appears to have another function for *D. brevicomis* since its release by males inhibits the response of its competitor, *I. paraconfusus* (Byers and Wood, 1980, 1981). Lanier et al. (1980) found verbenone in *I. pini* males from Idaho that had fed in red pine (*P. resinosa* Ait.) logs, but they could not ascribe any "biological activity" to the compound. Birch and Wood (1975) showed that the responses of *I. pini* and *I. paraconfusus* were mutually inhibited by volatiles from infested logs of the opposite species, and Light and Birch (1979) determined that (−)-ipsdienol from *I. pini* inhibited the attraction of *I. paraconfusus.* However, it may be that the verbenone in *I. pini,* a major component, contributes to this inhibition of *I. paraconfusus* by (−)-ipsdienol, and thus the behavioral effect of verbenone on *I. paraconfusus* has been naturally selected because of pressures to reduce interspecific competition from both *D. brevicomis* and *I. pini* (Figure 4). Furthermore, the inhibitory effects of verbenone on *D. brevicomis* could be not only the result of intraspecific competition but partly the result of selection pressure exerted by *I. pini.* This could also be true of the inhibitory effects of *trans*-verbenol on *D. brevicomis* (Byers, 1983a) which is produced by *I. pini* (Vité et al., 1972; Lanier et al., 1980) (Figure 4). The (+)-ipsdienol produced by male *I. paraconfusus* (Silverstein et al., 1966) and by male *D. brevicomis* in the early stages of colonization may not only function to reduce intraspecific competition in both species (Byers, 1982; Byers, 1983b) but also

reduce interspecific competition from *I. pini* in both species (Birch et al., 1980; Byers, 1982).

The system of density regulation and termination of aggregation in *D. brevicomis* appears to be complex. It apparently involves several attractive and inhibitory pheromones that are produced and released at varying ratios and absolute concentrations, depending on the stage of colonization. The absolute and relative concentrations of various pheromones are influenced by many factors such as attack density and sequence of arrival (compression of mass attack) which in turn are dependent on weather, population density, and host resistance. Physiological factors such as mating and nutrition as well as presence of pheromone precursors in the host (Byers, 1981b) also may effect release rates of pheromones. Male stridulation and other behavioral processes also probably have a role in regulation of attack density. Interspecific effects also could influence the density of colonization as hypothesized by Byers and Wood (1980, 1981) for *I. paraconfusus* and *D. brevicomis*. In Figure 5, a theoretical scheme is presented for the interaction of ecological factors which appear to influence the significance of pheromones, kairomones, and

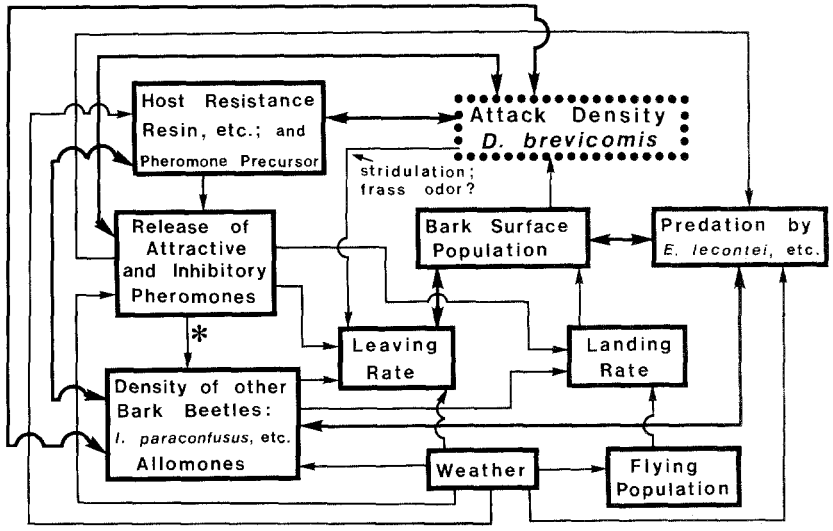

FIG. 5. Schematic of theoretical interactions between ecological components which may influence the attack densities of *Dendroctonus brevicomis* (and competing bark beetle species) on a ponderosa pine. Heavy lines indicate two-way interactions and possible direct feedback or regulatory functions, while thin lines indicate one-way effects which in some cases can have indirect feedback effects. The model assumes that pheromones/allomones are released after beetles have located sites of attack. *Pheromone components of *D. brevicomis* can also act as allomones in reducing the response of competing species to their pheromones.

allomones in regulating inter- and intraspecific competition (attack density) among *D. brevicomis* and other bark beetle species (e.g., *I. paraconfusus*).

Further work on determining the absolute concentrations of pheromones released from a tree under colonization and on the variation of production (cf. Birgersson et al., 1984) and release between individual galleries is needed. In addition, detailed studies of response to combinations of pheromones must be done before a rather complete understanding of aggregation and colonization can be achieved.

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