#### AN ABSTRACT OF THE THESIS OF

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Three coleopterous predators associated with the Douglasfir beetle (Dendroctonus pseudotsugae Hopkins in a second growth
forest of Douglas-fir (Pseudotsugae menziesii (Mirb.) Franco)
in western Oregon are Enoclerus sphegeus Fabricius, E. lecontei
Wolcott, and Thanasimus undatulus Say. A study was undertaken
with the following objectives: (1) to determine the diurnal and seasonal flight patterns of these predators in relation to the flight of
the Douglas-fir beetle, and (2) to determine the mechanism by which
these predators locate their prey. Additional studies were conducted
to detect the possible production of an olfactory sex attractant by E.
sphegeus.

Information on the flight of the three species was obtained with six electrically driven rotary insect nets and six sheet metal olfactometers. The attractiveness of various materials to the predators

was also tested with the olfactometers.

The diurnal flight patterns of <u>E</u>. sphegeus and <u>T</u>. undatulus and apparently <u>E</u>. lecontei are similar to that of the Douglas-fir beetle except that flight of the predators is restricted mainly to a six or seven hour portion of each day.

The seasonal flight of  $\underline{E}$ . sphegeus and  $\underline{T}$ . undatulus adults is initiated each spring with the first temperatures of 55° to 60°F. as it is with the Douglas-fir beetle, but  $\underline{E}$ . sphegeus adults apparently fly little after they locate a log infested with Douglas-fir beetles. Adults of  $\underline{E}$ . lecontei appear to fly after the main flight period of the Douglas-fir beetle and is thus not closely associated with this scolytid.

Adults of <u>E</u>. <u>sphegeus</u> and <u>T</u>. <u>undatulus</u> are attracted to oleoresin from Douglas-fir, grand fir, and ponderosa pine, and also to alpha-pinene, beta-pinene, limonene, and camphene which are major constituents of these oleoresins. In view of this information it appears that the predators <u>E</u>. <u>sphegeus</u> and <u>T</u>. <u>undatulus</u> locate concentrations of prey insects by being attracted directly to volatile materials escaping from the tree host of the prey insects. No attraction was found with adults of E. lecontei.

# THE FLIGHT AND OLFACTORY BEHAVIOR OF CHECKERED BEETLES (COLEOPTERA:CLERIDAE) PREDATORY ON THE DOUGLAS-FIR BEETLE

by

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# THE FLIGHT AND OLFACTORY BEHAVIOR OF CHECKERED BEETLES (COLEOPTERA:CLERIDAE) PREDATORY ON THE DOUGLAS-FIR BEETLE

#### INTRODUCTION

The Douglas-fir beetle, <u>Dendroctonus pseudotsugae</u> Hopkins, is probably the most destructive insect of Douglas-fir, <u>Pseudotsuga</u> <u>menziesii</u> (Mirb.) Franco, throughout the natural range of this economically important tree. The volume of green timber killed by this insect during the 1951-1954 epidemic amounted to over three billion board feet in Oregon and Washington alone. A lesser epidemic started in Oregon in 1964 causing losses for that year estimated at 250 to 500 million board feet of green timber.

Although this beetle normally attacks unhealthy or recently downed trees, epidemic populations and primary tree killing usually occur about every ten to 15 years following extensive windthrows and fires which provide abundant suitable food material for good survival of developing broods. These broods mature in the functional phloem and cambium of the tree host throughout the summer and overwinter there, mainly as callow adults, while their obligatory low-temperature requirement is being met. The adults emerge in the spring with the first sufficiently warm temperatures and attack new suitable host material. During the periods of high population levels, when there is an insufficient quantity of fresh fallen

timber to absorb the beetles, healthy trees may be overcome and killed by large numbers of invading beetles. Such mass attacks are accomplished with the help of a pheromone produced by feeding virgin female beetles. This pheromone attracts both males and additional females to the area of initial attack. This account of the life history, flight, and olfactory behavior has been summarized from the reported investigations of Hopkins (1909), Bedard (1933, 1950), Walters (1956), Furniss (1957), Vité and Rudinsky (1957), Mc Cowan and Rudinsky (1958), Rudinsky (1963, 1965), Hendrickson (1965), and Jantz (1965).

Although the main regulatory factor of the Douglas-fir beetle population is apparently the availability of suitable brood material, other factors such as weather, competition, host resistance, and biological agents certainly play a more or less important role from time to time. It is through a knowledge of these individual factors that a safe, effective, and economical method of integrated control may be achieved. Entomophagous insects have often been considered important factors in reducing or preventing the development of high populations of destructive forest insects.

Three species of checkered beetles (Coleoptera:Cleridae)
which may exert regulative pressures on the Douglas-fir beetle population in western Oregon are Enoclerus sphegeus Fabricius,
Thanasimus undatulus Say, and E. lecontei Wolcott. The close

association of these clerids with the Douglas-fir beetle has prompted past investigations into their biologies and estimates of their effectiveness, but the flight periods and the means by which these predators locate their prey has not been investigated.

The lack of knowledge in these areas regarding these clerids has prompted the study reported here with the following objectives: (1) to determine the flight patterns of these insects in relation to the flight of the Douglas-fir beetle, and (2) to determine the mechanism by which these predators locate their prey. Once the study was underway and it was determined that the predators were attracted to volatile fractions of the oleoresin produced by the tree hosts, further study was undertaken on the mechanism of sex recognition and/or attraction of  $\underline{E}$ .  $\underline{sphegeus}$ . The reason for this side study was that such an olfactory response may aid the predators in finding a site for mating and ovipositioning in addition to feeding.

Knowledge of this directed flight of these clerids may be of value to workers attempting to control the Douglas-fir beetle and other scolytids by: (1) providing a means of estimating population levels of the clerids in a given area through trapping, (2) determining the associations between uncertain predators and bark beetles through comparisons of their flight schedules and tree hosts, (3) furnishing a method of trapping large numbers of Cleridae for release in other areas where scolytids are epidemic, and (4) aiding in the

prediction of the success of introduced predator species into new areas by comparing the oleoresin composition in the tree hosts of the new and original prey species.

#### LITERATURE REVIEW

The family Cleridae is composed of approximately 3300 species of which about 317 species are represented in North America north of Mexico (Papp, 1960). With very few exceptions, the members of this family are closely associated with bark- and wood-boring insects, mainly Scolytidae, as predators in both the larval and adult stages (Böving and Champlain, 1921; Balduf, 1935; Knull, 1951).

Balduf (1935) considers the clerids in general to be more specialized as predators than most Carabidae and Cicindelidae because they have limited their prey to a group of insects with more or less uniform habits and development. This limitation is said to be indicated by the predator and prey life cycles which appear to be correlated seasonally and numerically. He also considers the development of these predators to be parallel to that of their prey which is evidenced by the fact that the clerid larvae feed on the immature stages of the prey and the clerid adults upon the prey adults. These points agree at least in part with the information available on <u>E</u>. <u>sphegeus</u>, <u>E</u>. <u>lecontei</u>, and <u>T</u>. <u>undatulus</u>.

Evaluations of the effectiveness of these three species as predators have ranged from little or no value to highly beneficial depending on the location and/or the prey insect (Keen, 1928; De Leon, 1934; Struble, 1942; Bedard, 1950; Kline and Rudinsky, 1964; Cowan

and Nagel, 1965).

It is at least of historical interest that the first insect to be introduced into the United States to combat a forest insect pest was a clerid, Thanasimus formicarius Lec. (Chamberlin, 1939; Dixon and Osgood, 1961). This introduction was made by Hopkins in 1892 against the southern pine beetle, Dendroctomus frontalis Zimm., in West Virginia (Hopkins, 1897). The species apparently did not become established due at least in part to a collapse of the host population shortly after the release (Dixon and Osgood, 1961).

Although some workers have found these predators to be of little apparent value in several areas of the western United States, possible future benefits from these insects should not be overlooked. In light of the increasing use of silvicultural operations as our forests are converted from virgin stands to intensively managed ones, the ability to manipulate the environmental conditions in favor of the predators becomes more feasible. Bőving and Champlain (1921) and Person (1940) foresaw this possibility and suggested that cutting of bark beetle infested trees be timed to coincide with periods when the immature stages of the predators were not beneath the bark. The latter author felt that many earlier cut and burn operations of beetle infested trees were unsuccessful because their poor timing reduced the predator populations to levels below those needed for adequate subsequent predation.

Although the life histories of these three species have been investigated by various workers, attention in this section is given mainly to information on the adults. Particular emphasis is placed on the flight and olfactory behavior of each species and the volatile materials produced by the associated tree species which may be chiefly responsible for the olfactory responses exhibited by the predators.

## Enoclerus sphegeus

The adult Enoclerus sphegeus was first described by Fabricius in 1787 (Leng, 1920; Papp, 1960) and was more thoroughly described recently by Brown (Reid, 1957). Larval instars have been described by Struble (1942), Reid (1957), Kline and Rudinsky (1964), Cowan and Nagel (1965). E. sphegeus is distributed from Alberta, Canada southward along the Pacific coast states into Mexico and eastward into Colorado, Arizona, and New Mexico (Reid, 1957; Vaurie, 1952; Papp, 1960).

The period of adult flight activity appears to occur at various times of the year in different parts of the species' range. In the central Sierra Nevada Mountains of California at elevations of 4,500 to 6,500 feet <u>E</u>. sphegeus adults appeared in the greatest numbers from September to November (Struble, 1942). These adults apparently overwintered in bark crevices and reappeared the following

spring. Blackman (1931) found adult E. sphegeus to be most abundant from late May through early July with a rapid decrease in numbers thereafter. Furniss (1957) reported adults were most numerous in Idaho between mid May and early July. E. sphegeus adults in western Oregon were reported to emerge with the Douglas-fir beetle in April and May by Kline and Rudinsky (1964), but Cowan and Nagel (1965) concluded that this species on Marys Peak in western Oregon had a two year life span with the adults emerging in late summer, overwintering in bark crevices and litter, and reappearing the following spring with the emergence of the Douglas-fir beetle. Although these adults could be found from the last part of April through July, they were seldom seen in flight after June 1, in 1964. After a release of 25 marked beetles on a log June 12, 1964, the relative numbers of marked to unmarked beetles changed little during the following two weeks. It was concluded that few additional beetles had flown to the log.

The forests within the area occupied by <u>E. sphegeus</u> are composed chiefly of conifers in the family Pinaceae, and this predator has been reported to occur with most of the species. Champlain summarized the observations on file with the United States National Museum and wrote, "It is found in most of the western pines, spruce, and fir; also <u>Pseudotsuga taxifolia</u> and <u>Larix occidentalis</u>." (Böving and Champlain, 1921). The observations were made by Hopkins,

Burke, Webb, Edmonston, Fiske, Brunner and Champlain. It has also apparently been found on western hemlock, <u>Tsuga heterophylla</u> (Raf.) Sarg., by McGhehey (1965).

Although E. sphegeus has been studied in nuermous areas by several workers, information is lacking on the mechanism by which it finds concentrations of prey insects. Without any explanation, Furniss (1957) reported that the greatest number of adults were found on the stumps of felled trees, and Boving and Champlain (1921) stated that E. sphegeus were attracted to trees containing Dendroctonus ponderosae Hopkins. Reid (1957) was able to find E. sphegeus on a log only after it had been attacked the previous day by Ips sp.

The predators were not on the log approximately one week after the Ips had entered the bark. Vite and Gara (1962) were able to attract E. sphegeus to ponderosa pine logs infested with species of Dendroctonus and Ips and also to uninfested logs. They concluded that attraction was greater to infested logs, but did not elaborate on the causes of this attraction.

Other behaviors which may involve olfactory responses are the finding, excitation, and recognition of sexes among members of the species. Hitchcock (1963) suggested an external sexual dimorphism as the means used by the clerid Necrobia rufipes De Greer for sex recognition. He reported no evidence was found of an olfactory stimulus from Y-tube olfactory tests and observations. He then

interchanged elytra between the sexes and attached elytra of both sexes to wooden blocks and exposed these beetles and wooden models to males. He concluded that locating of females by males was by random searching, but that tactile stimuli were important in recognition of the females and that probably the elytral setae were the source of the stimuli. Simmons and Ellington (1925) first noted that the sexes of N. rufipes differed by the elytral setae which project forward only on the females.

Cowan and Nagel (1965) tested <u>E</u>. <u>sphegeus</u> females for possible production of an attractant or excitant by placing <u>E</u>. <u>sphegeus</u> males and <u>T</u>. <u>undatulus</u> females together in containers which held <u>E</u>. <u>sphegeus</u> females just prior to the test. No interspecific matings were observed in these tests, but they stated that the larger <u>E</u>. <u>sphegeus</u> males were unable to catch the smaller, more rapid <u>T</u>. undatulus females.

In summation for <u>E</u>. <u>sphegeus</u> adults, information is available on the general seasonal appearance of the beetle in several areas, but no mention is made of diurnal activities nor the factors affecting flight. Concerning oriented flight, concentrations of these predators have been noted on scolytid infested and on uninfested fresh logs, and attraction has been found to infested and uninfested ponderosa pine logs, but in neither case have explanations been given.

### Enoclerus lecontei

The nomenclature of Enoclerus lecontei has been confused by changes of both the generic and specific names. The adult E. lecontei was first described by Le Conte in 1861 as Clerus nigriventris. Wolcott renamed the species Clerus lecontei in 1910, but this name was later changed to Enoclerus lecontei Wolc. (Leng, 1920; Böving and Champlain, 1921; Wolcott, 1947). Since this last revision, the name Thanasimus nigriventris has been used by Balduf (1935), Blackwelder (Kline and Rudinsky, 1964) and Townes (1960), and Thanasimus lecontei (Wolc.) by Person (1940). Descriptions of the larvae have been made by Böving (1928) and Kline and Rudinsky (1964). The range of E. lecontei extends from British Columbia east to Michigan and south to Guatemala (Papp, 1960; Wolcott, 1947), but it is apparently most abundant in Washington, Oregon, and California (Person, 1940).

The time of flight activity for <u>E</u>. <u>lecontei</u> appears to differ only slightly throughout its range. The beetles were reported to be present from April to October by Böving and Champlain (1921) while Bedard (1933) found them to be most numerous from the latter part of July through August in Idaho. Person (1940) concluded from his studies in California that newly emerged adults appeared twice each year, from May 15 to June 20 and also late each summer. This

second emergence group was composed of 80 to 95 percent of the brood from the spring emerged adults. The remaining five to 20 percent overwintered as mature larvae and made up the spring emergence group. Studies on Marys Peak by Cowan and Nagel (1965) revealed <u>E. lecontei</u> adults to be present from about June 15 through September.

This insect is best known as a predator of <u>D. brevicomis</u>

Leconte attacking ponderosa pine (Keen, 1928; Doane et al., 1936;

Person, 1940), but it has also been found associated with <u>Ips confusus</u>

(Leconte) on ponderosa pine (Struble and Hall, 1955), <u>D. ponderosae</u>

on lodgepole pine and western white pine (De Leon, 1934), <u>D. obesus</u>

(Mannerheim) on engelmann spruce (Massey and Wygant, 1954),

and <u>D. pseudotsugae</u>, <u>Scolytus unispinosus</u> Leconte, and <u>Pseudohy-lesinus sp.</u> attacking Douglas-fir (Bedard, 1933 and 1950; Kline and Rudinsky, 1964).

Vite and Gara (1962) concluded from field olfactory tests that

E. lecontei were attracted to sections of ponderosa pine logs which contained various species of <u>Ips</u> and <u>Dendroctonus</u> in early and advanced stages of infestation as well as to uninfested sections.

In brief, the information available on the flight and olfactory behavior of <u>E</u>. <u>lecontei</u> is limited to statements of the seasonal appearance of adults in several areas, and attraction to scolytid infested and uninfested log sections of ponderosa pine. No explanation

was given for this attraction.

## Thanasimus undatulus

The adult Thanasimus undatulus was first described by Say (1835) under the name Clerus undatulus. Because of revisions of the generic nomenclature, it was placed in the genus Thanasimus (Leng, 1920), but through a typographical error the specific name was listed as undulatus (Kline and Rudinsky, 1964; Wolcott, 1947). This error has been carried forward by Chamberlin (1939), and Papp (1960). Kline and Rudinsky (1964) state that Bedard (1933) mistakenly identified T. undatulus as T. dubius (Fab.). This identification is also used by Bedard in 1950.

At least three subspecies or forms have been recognized by Chamberlin (1939) and Papp (1960) while five varieties have been listed by Wolcott (1947). Barr preferred to call all the forms T. undatulus (Kline and Rudinsky, 1964). If all the varieties or forms are considered as a single species, this insect is distributed from Alaska to eastern Canada and south into New Mexico (Chamberlin, 1939; Papp, 1960; Wolcott, 1947).

Few studies have been conducted on <u>T. undatulus</u>, but the adults appear to be active during most of the summer months throughout the species' range. Beetles were reported to fly from May to September by Böving and Champlain (1921). Bedard (1933) found them to be

most abundant in Idaho in late May and June while Kline and Rudinsky (1964) found them in April and May in western Oregon.

On Marys Peak in western Oregon, most adults were found to emerge in September, overwinter, reappear in the spring, and remain active until mid August (Cowan and Nagel, 1965).

Although several hardwood species occur within the range of T. undatulus, it has been reported associated only with bark beetles attacking coniferous species. Referring to observations made by Hopkins, Burke, Fiske, and Champlain, Böving and Champlain (1921) state, "It is a predator of Dendroctonus and other bark beetles in coniferous trees, Pinus, Picea, Pseudotsuga, Larix, Abies, and cedar..."

Although no information is available on the mechanism by which this species finds concentrations of prey insects, Cowan and Nagel (1965) found evidence of the production of an olfactory sex stimulus by the females. They were able to produce interspecific matings between <u>T. undatulus</u> males and <u>E. sphegeus</u> females when the two insects were placed in a container occupied by a <u>T. undatulus</u> female just prior to the test. The test was repeated with <u>T. undatulus</u> males and Douglas-fir beetles and it was found that the clerid males would also attempt to mate with them.

In conclusion, no information is available concerning the factors affecting the seasonal flight of  $\underline{T}$ . undatulus, no mention is

made of the diurnal patterns of flight, and no mechanism has been found by which this species finds its insect prey.

## Flight Behavior of Dendroctonus pseudotsugae

The diurnal and seasonal flight patterns of the Douglas-fir beetle on Marys Peak have been investigated during the last four years by Rudinsky (1965) and the diurnal and seasonal patterns of response have been reported by Rudinsky (1963), Hendrickson (1965), and Jantz (1965). The most important physical factors affecting flight of this beetle were considered to be temperature, light, and wind.

The diurnal flight was found to occur essentially from 10:00 AM until 7:00 to 8:00 PM standard time, or until darkness, with the greatest number of individuals flying about 2:00 PM. Exceptions to this pattern were found when temperatures exceeded approximately 75°F. Two-peaked flight curves were obtained on these days, due to a mid-day depression of flight activity. The beetles were also found to rarely fly below 58°F. although flight activity was noted to occur occasionally at lower temperatures.

The seasonal flight of the Douglas-fir beetle was found to occur from April through July with the main period of primary flight activity occurring during May. Secondary peaks of flight were found to occur in July which were considered to be made up

predominantly of reemerged beetles. A gradual decline in flight activity was reported to occur after July.

#### Compositions of Oleoresins

Because oleoresins and their fractions were found to be important to the finding of bark beetle prey by the checkered beetles in this study, this section is included to provide information concerning the physical and chemical natures of these substances.

Many investigations have been made on the physical and chemical properties of the oleoresins of the pines, but information is lacking for most other species. Oleoresin is defined by the American Society for Testing Materials as,

the nonaquous secretion of resin acids dissolved in a hydrocarbon oil which is (1) produced in or exuded from the intercellular resin ducts of a living tree, (2) accumulated, together with oxidation products, in the dead wood of weathered limbs and stumps (Mirov 1961, p. 157).

Mirov adds to this statement that the resin acids may also be dissolved in paraffin hydrocarbons or even benezene derivations.

Oleoresin may be separated physically into rosin, a mixture of various non-volatile materials, and turpentine which is the composite of completely volatile substances. These volatile substances are predominantly cyclic hydrocarbons, terpenes with the formula  ${}^{\rm C}_{10}{}^{\rm H}_{16}$ , most often in mixtures with sesquiterpenes,  ${}^{\rm C}_{15}{}^{\rm H}_{24}$ , but occasionally with nonterpene substances (Mirov, 1961). Kurth (1952)

states that the volatile materials from wood include essential oils and volatile acids, but that the distinction between the two groups is arbitrary since the essential oils may contain acids. He says that the chief substances in essential oils are terpenes, sesquiterpenes, and the oxygenated derivatives of alcohols, aldehydes, or ketones and that phenols, phenolic ethers, esters, oxides, and acids may also be present.

The composition of terpentines is a genetically fixed character of each tree and changes but little during the growing season (Mirov, 1961). Composition differences may be great between genera of the same family and even species of the same genus (Kurth, 1952; Mirov, 1961) and considerable differences may be found among different parts of the same tree (Kurth, 1952). This variation is shown by Kurth (page 565) who summarized the findings of Johnson and Cain (1937), Schorger (1917), and Benson and Mc Carthy (1925) on the various oleoresins of Douglas-fir.

Volatile oils from Douglas-fir

Source	Known constituents				
Wood	l-a-Pinene, 30%; l-camphene, 6%; l-limonene, 14%; l-a-terpineol, 32%				
Bark	l-a-Pinene, 27%; l-B-pinene, 24%; l-camphene, 7%; dipentene, 8%; geraniol, 6% and azulenogenic sesquiterpenes.				
Leaves and twigs	1-B-Pinene, 33%; dipentene, 18%; 1-a-pinene, 12%; 1-camphene, 7%; geraniol (caprate or acetate), 12%; phenols (salicyclic acid) 0.07%; and capric acid.				

Volatile oils from Douglas-fir

Known constituents

Source

Oleoresin caused by 1-a-and B-pinene and small amounts of

injuries l-limonene and l-terpineol.

Such differences among various tree parts most likely occur also in other species and are important determinants of the quality of vapors released from fallen trees. Unfortunately, such complete information is unavailable for most tree species. The fact that most of the bark beetles preyed upon by these clerids penetrate the bark of the stem and larger limbs, causing volatilization of resin from only these parts, contributes some uniformity to the oleoresin quality.

Table 1 presents the compositions, when they are known, of the oleoresins produced by the more common species of conifers in western United States and also by those species with which E. sphegeus, E. lecontei, and T. undatulus have been associated. Larix decidua is listed in place of L. occidentalis because no information is available on the latter species. It should be noted that the fractions alphapinene, beta-pinene, and limonene are present in most of the pines (Pinus), true firs (Abies), Douglas-fir (Pseudotsuga), the larch (Larix), and at least one spruce (Picea). Another fraction, beta-phallandrene, is present in lodgepole pine, and some of the true firs. Also to be noted in this table is the absence of the fractions geraniol, alpha-terpineol, and myrcene in most of the species. The chemical nature of these fractions will be presented in the following section.

Table 1. The compositions of the oleoresins of some coniferous species.

	Constituents 4									-	
Tree species	camphene	delta 3_carene	geraniol	limonene	myrcene	beta- phallandrene	alpha— pinene	beta- pinene	alpha- terpineol	others <sup>b/</sup>	Source of information
Abies amabilis				+		+	+	+			Kurth 1952
Abies balsamea						+	+	+			Kurth 1952
Abies grandis	19					5	20	15		<b>4</b> 1	Trupp and Fischer 1939
Chamaecyparis lawsoniana				3			46			51	Kurth 1952
Larix decidua							+				Kurth 1952
Picea glauca							50				Kurth 1952
Pinus contorta						P	+				Mirov 1961
Pinus lambertiana						2-3	70-75	5		12-15	Mirov 1961
Pinus monticola				7			32	45		16	Mirov 1961
Pinus ponderosa		25-64		0-15	0-5		1 -45	0-50		0-10	Mirov 1961
Pseudotsuga menziesii wood	6			14			30		32	18	Kurth 1952
Pseudotsuga menziesii bark	7		6				28	24		35	Kurth 1952
Tsuga canadensis	12			6	2		18	3		50	Shaw 1951

<sup>&</sup>lt;sup>a/</sup>Numerals represent the percent of total weight, + represents presence without a determination of percentage, P represents the predominating constituent.

 $<sup>\</sup>frac{b}{}$  Includes other volatile fractions, unknown fractions, and nonvolatile materials.

#### MATERIALS AND METHODS

The investigations reported here were conducted during the spring and summer months of 1963, 1964, and 1965 with the major contributions coming from the latter two years. All the field studies were on the northeast slope of Marys Peak 14 miles west of Corvallis, Oregon. Metal olfactometers and screened cages containing various baits were used to trap insects in flight responding to the attractive substances while electrically driven insect nets were run to collect those insects in undirected or random flight. Several laboratory tests were also conducted to confirm the results obtained in the field and to study close range olfactory behavior.

## Study Area

The main study area (Figure 1) is along a ridge extending west to east at an elevation of 1100 feet in the Marys Peak watershed of the Siuslaw National Forest. The stand density varies from moderate along the ridge crest to high along the flanks of the ridge while several clearcuts occur nearby. The overstory is predominantly 250 to 300 year old second growth Douglas-fir, Pseudotsuga menziesii (Mirb.) Franco, but with scattered trees of western red cedar, Thuja plicata Don.; western hemlock, Tsuga heterophylla (Raf.) Sarg.; and grand fir, Abies grandis Lindl. The common

understory woody plants include vine maple, Acer circinatum Pursh.; salal, Gautheria shallon Pursh.; and Oregon grape, Berberis nervosa Pursh.

#### Nets

Six electrically driven rotary insect nets (Figure 2) were run periodically during the spring and summer of all three years to sample the flying populations of Cleridae and Scolytidae. These nets were spaced approximately 100 feet apart at standard positions in a curved line which extended along the flank and crest of the ridge. These traps were identical to those described by Vite and Gara (1962) which were found the most suitable for sampling bark beetle populations. Each net assembly consisted of a net bag mounted on a shaft which was rotated horizontally by an electric motor equipped with a reduction gear box and 90° coupling. The motors hung by brackets from the top of six foot step ladders. Three portable electric generators driven by gasoline engines supplied the 110 volt current needed for the motors.

Information on the seasonal flight patterns of the insects was obtained by running the nets approximately one day each week of the spring and summer for three years. Deviations from this schedule occurred when climatic conditions were not conducive to flight and in the late summer when insect flight levels were low. The total



Figure 1. A view of the study area showing that portion with the highest stand density.



Figure 2. A rotary net used to trap insects in random flight.

number of days on which nets were run for the three years 1963 through 1965 are respectively 31, 20, and 18.

Diurnal flight patterns were obtained throughout the three flight periods, but only those gathered early each season during the periods of high flight activity contained sufficient numbers of insects to be reliable. On each of the collection days, the nets were started before flight activity began and were emptied each one or one-half hour until the insects being studied ceased to be collected.

#### Olfactometers

The metal olfactometers (Figure 3) were the chief source of data on both flight and olfactory response. They were located 100 feet apart in a straight line along the ridge crest with each location as nearly like the others in respect to exposure as was possible under natural conditions.

These olfactometers were patterned after those used and described by Vité and Gara (1962) to study olfactory behavior of forest insects in California. They are essentially sheet metal tubes standing approximately six feet high. Midway inside each olfactometer is an electric fan which draws air from the base, containing the olfactory materials being tested, and forces it out the opening in the top. Above the fan is a wire screen funnel pointing downwards and ending in a removable glass jar which receives the attracted insects.



Figure 3. A sheet metal olfactometer used for attraction studies.

Transparent plexiglas baffles are centered atop each olfactometer to stop the insects following the scent stream to the source. The insects were collected from the olfactometers at one-half or one hour intervals.

Most of the tests were made using a different olfactory material in each of five olfactometers with the sixth one containing either ethanol, benzene, or nothing as a control. Table 2 lists the materials tested during 1964 and 1965 and the total number of hours each one was tested. All the solutions were tested at known, standardized concentrations in identical bottles to assure that the evaporation rates were comparable exclusive of variations in vapor pressure of the various substances. A  $2\frac{1}{2}$  percent solution of Douglas-fir oleoresin with 95 percent ethanol was exposed during most tests as a standard attractant source for determining the seasonal flight pattern and also for comparing the attractive qualities of materials that were tested on different days. Tests early in the 1964 season indicated this concentration to be the most attractive. All the solutions were diluted in 95 percent ethanol to the concentrations shown in Table 2 except for the fraction myrcene and grand fir oleoresin which were diluted in benzene because they were not completely soluble in ethanol.

The various substances tested were obtained from several sources. The Douglas-fir and ponderosa pine oleoresins were colubected from living trees approximately five miles north of Corvallis,

Table 2. The materials tested and their total hours of exposure in sheet metal olfactometers during 1964 and 1965. 24

Materials tested	Total hours of exposure
½% Douglas-fir oleoresin	$11\frac{1}{2}$
1% Douglas-fir oleoresin	$11\frac{1}{2}$
$2\frac{1}{2}\%$ Douglas-fir oleoresin	166
5% Douglas-fir oleoresin	27
10% Douglas-fir oleoresin	$11\frac{1}{2}$
15% Douglas-fir oleoresin	$2\frac{1}{2}$
20% Douglas-fir oleoresin	$11\frac{1}{2}$
50% Douglas-fir oleoresin	6
100% Douglas fir oleoresin	$8\frac{1}{2}$
$2\frac{1}{2}\%$ Ponderosa pine oleoresin	$71\frac{1}{2}$
$2\frac{1}{2}\%$ Grand fir oleoresin	$24\frac{1}{2}$
l% a -pinene	84
1% β-pinene	84
1% Camphene	$50\frac{1}{2}$
1% Geraniol	54
1% Limonene	73
1% Myrcene	$29\frac{1}{2}$
1% a -terpineol	$32\frac{1}{2}$
Uninfested Douglas-fir log	$9\frac{1}{2}$
Douglas-fir log infested with Scolyti	$.dae   39\frac{1}{2}$
Empty control	87
Ethanol control	30
Benzene control	8

Grand fir oleoresin and myrcene were diluted in benzene.
All other solutions were with 95% ethanol.

Oregon while the grand fir oleoresin was collected from trees in the watershed. The oleoresin of grand fir was collected from small pockets in the outer bark of green trees while the other two oleoresins were collected from the xylem. The trees were tapped by boring a hole through the bark and cambium and driving a short iron tube into it. A rubber balloon was fitted over the end of the tube to collect the oleoresin exuded over a period of three to seven days. The fresh oleoresin was diluted 50 percent with ethanol and stored in glass bottles at 40°F. The dilution was necessary to prevent crystallization of the oleoresin. The oleoresin was further diluted to the desired concentration just prior to testing.

The oleoresin fractions were obtained as commercial preparations from two companies. The fractions D-alpha-pinene, betapinene, D-camphene, myrcene, DL-limonene, and geraniol were obtained from K & K Laboratories, Inc. Plainview, New York and a sample of alpha-terpineol was obtained from Hercules Power Company Incorporated, Wilmington, Delaware. These materials were stored full strength in air-tight brown glass bottles until just prior to testing when they were diluted to a one percent concentration with 95 percent ethanol.

The fractions of oleoresin tested for attractiveness in this study may be separated into five groups based on their chemical properties and structure. Limonene (Figure 4) is the sole

representative of the monocyclic terpenes while myrcene (Figure 5) is the only olefinic open chain terpene tested. Two terpene alcohols were tested, alpha-terpineol (Figure 6) and geraniol (Figure 7). The three fractions alpha-pinene (Figure 8), beta-pinene (Figure 9) and camphene (Figure 10) are members of the bycyclic terpene group.

## **Attraction Cages**

Screened cages measuring 2 ft. × 2 ft. × 5 ft. were used in the study area all three years. The number of cages used varied from three to 12. These cages usually contained Douglas-fir log bolts infested with 40 virgin Douglas-fir beetle females, but other materials used as baits were logs of ponderosa pine, white pine, and grand fir with and without female beetles, and Douglas-fir logs with freshly punched holes or male Douglas-fir beetles. No solutions were tested with these cages because they were not comparable to the metal olfactometers with forced air systems.

## Observation Logs

After the results of the flight studies suggested that <u>E. sphegeus</u> adults may remain for extended periods on a log once it is found, releases of marked beetles and periodic counts afterwards were undertaken. Three logs were selected at different locations in the watershed. One log was windthrown during the 1964-1965 winter while the

Figure 5. The structural formula of myrcene.

Figure 6. The structural formula of alpha-terpineol.

Figure 7. The structural formula of geraniol.

other two were cut May 18, 1965. The diameter at breast height of these trees were 13, 15, and 26 inches. The number of Douglas-fir beetle attacks on the logs averaged nine per square foot on the windthrown one and  $1\frac{1}{2}$  on the other two.

The insects to be released were marked with different colors of nail polish or paint to distinguish the two sexes and the logs they were released on. The first releases were unsuccessful because the insects immediately flew away. The attempts used beetles collected in the olfactometers and stored at 40°F. until release. A more successful method was to spread the beetles along the log while they were still cold from being kept in cooler chests with salted ice. Fewer beetles flew away as they warmed up than in the first attempts, but the results were still not satisfactory. The most dependable method found was to mark naturally-occurring beetles on the logs where they were found. Any sudden movements would excite the beetles to the point of flying away or falling from the log so extreme care was necessary. The logs were then inspected approximately each week and the number of marked and unmarked beetles were recorded.

# Meteorological Records

Measurements and records were taken during the three seasons of temperature, humidity, wind, and sky overcast. Two

hygrothermographs, were run continuously during the flight seasons. One instrument was permanently located in the study area under a dense tree canopy while the second instrument was located in a clear-cut near the study area. Both hygrothermographs were protected from direct sunlight by open enclosures approximately four feet above the ground. Additional temperature measurements were taken from mercury thermometers at half of the net and olfactometer sites. When air movements were noted, the direction was recorded and velocity was measured with a hand-held wind meter. Whenever clouds were present, their extent was recorded as the percent of sky covered.

## Laboratory Olfactory Tests

Several laboratory tests were conducted to confirm the field results of the attraction to oleoresin and to investigate the possibility of a mechanism for sex determination of <u>E</u>. <u>sphegeus</u>. Two devices tried in this work, an arena olfactometer and an arrestment board, have been used by previous workers for testing olfactory responses of scolytids. Another device was designed specifically for detecting the presence of a sex attractant of these beetles. Observations were also made of paired beetles prior to mating.

<sup>&</sup>lt;sup>1</sup>Made by the Foxboro Company of Foxboro, Massachusetts.

<sup>&</sup>lt;sup>2</sup>Made by the F. W. Dwyer Manufacturing Company of Michigan City, Indiana.

The arena olfactometer, as described by Hendrickson (1965), allows beetles to choose one of five air streams to follow containing different substances. Introduced into these air streams were Douglas-fir and ponderosa pine oleoresins, alpha-pinene, beta-pinene, and air from containers of  $\underline{E}$ . sphegeus of the opposite sex of those being tested. Both sexes were tested for responses and attractant production. Tests were run in both total darkness and in light to determine the best conditions.

The arrestment boards, as described by Jantz (1965) were tried to detect any arrestment of movement by the beetles over holes containing oleoresin or their fractions. Such a response under natural conditions may aid female beetles in ovipositing near bark beetle entry holes.

Equipment was designed to determine the possible presence of a sex attractant or excitant produced by <u>E. sphegeus</u> females as Cowan and Nagel (1965) reported for <u>T. undatulus</u>. The apparatus pumped air from a jar containing 40 <u>E. sphegeus</u> females into a plastic box containing an <u>E. sphegeus</u> male. Females of other clerid species as well as dead <u>E. sphegeus</u> females were put into the box with the male to see if he would attempt to mate. Fifty different males were checked in this manner with 25 beetles exposed to each type of female.

#### RESULTS AND DISCUSSION

Although the flight and olfactory behavior of three species of Cleridae were investigated, the amount of information obtained varied for each species. E. sphegeus was the most abundant of the three species near the study plot on Marys Peak and the information gathered on it was the most complete. T. undatulus was the second most numerous and the amount of information obtained was intermediate to that collected on the other two species. Little information was obtained concerning E. lecontei because specimens were seldom encountered in the area.

### Diurnal Flight Patterns

# Enoclerus sphegeus

Although the nets and olfactometers were run periodically throughout the seasons of flight of the predators, sufficient numbers of  $\underline{E}$ . sphegeus were caught on only one or two days each season to provide reliable information concerning the diurnal flight patterns.

The lowest temperature at which <u>E</u>. <u>sphegeus</u> was found flying was 55°F. Olfactometers containing attractive oleoresins from Douglas-fir and ponderosa pine were run while the ambient air

temperature was near the suspected temperature threshold for the species' flight. E. sphegeus were caught at the olfactometers only after the air temperature within the stand had reached 55°F.

The highest temperature within the stand at which <u>E</u>. <u>sphegeus</u> was found flying was 80°F. on July 23, 1965. It can not be concluded from the data that this temperature is the limit for the species' flight activity because insufficient numbers were caught for reliable evidence from mid-summer on when the temperatures are more often above 80°F. within the stand.

Although some individuals of this species were found to fly at 55°F., flight activity generally did not begin until the temperature was approximately 60°F. In late April and May during the main seasonal flight period, the typical diurnal flight pattern exhibited by E. sphegeus (Figure 11) is such that flight is usually initiated from 10:00 to 11:00 AM standard time and is terminated by 5:00 PM. This restricted period of flight occurred although the air temperature was sufficiently high for flight activity before and after this six or seven hour interval and suggests that light intensity and/or humidity influence the diurnal flight pattern of E. sphegeus.

Consistency of the hour of flight termination is shown by the flight patterns on those days when flight was delayed by morning low temperatures (Figure 12). Although the insects did not begin flying until 12:30 to 1:30 PM standard time, the flight did not extend more

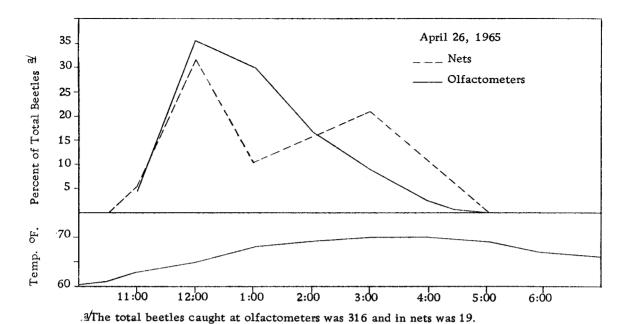


Figure 11. A typical diurnal pattern of flight activity of E. sphegeus.

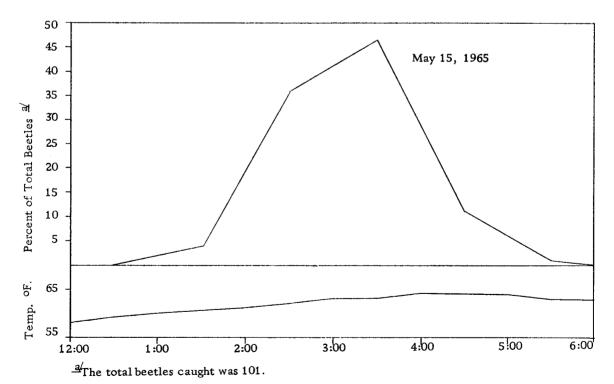


Figure 12. A diurnal pattern of flight activity of E. sphegeus which was delayed because of low temperatures.

than one half hour beyond 5:00. Throughout the 1964 and 1965 seasons, only two specimens of <u>E</u>. <u>sphegeus</u> were collected after 5:00 PM standard time. These collections were made on May 15, 1964 from 4:30 to 5:30 PM and July 23, 1965 between 7:00 and 8:00 PM.

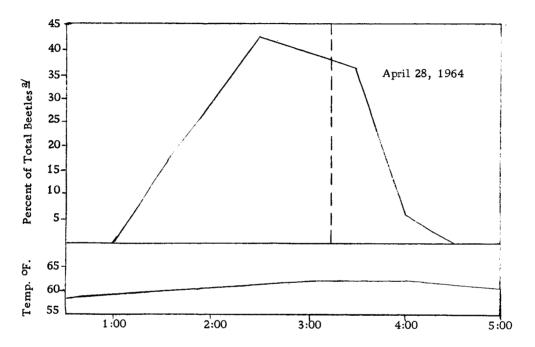
Another environmental factor found to affect the diurnal flight of <u>E</u>. sphegeus was air movements with velocities above approximately five miles per hour within the stand. When continuous winds occurred above this velocity, the numbers of <u>E</u>. sphegeus trapped rapidly diminished to zero (Figure 13). Winds above the forest canopy and short gusts within the stand of velocities greater than about five miles per hour appeared to have little effect on the beetle flight as measured by trapping with the olfactometers and nets.

# Thanasimus undatulus

The lowest air temperature within the stand at which T.

undatulus were caught in the nets and olfactometers was 61°F.

Trapping of this species at this temperature occured on three days in 1964 April 19, May 11, and May 15. The warmest air temperature within the stand when this species was caught in flight was 77°F. on June 22, 1964. Although this species is apparently attracted to oleoresins as is E. sphegeus, sufficient numbers were never trapped to distinguish accurately the periods of low levels of flight activity from those of non-activity.



a The total beetles caught was 83.

Figure 13. The reduction of flight activity of E. sphegeus with the initiation of a five-mile per hour wind at 3:15.

The maximum number of <u>T</u>. undatulus trapped in one day was 12, with the olfactometers, on April 26, 1965 so no reliable diurnal flight pattern can be made. In general, specimens of <u>T</u>. undatulus were trapped during the same time of day as were <u>E</u>. sphegeus, and no specimens were collected before 10:00 AM or after 5:00 PM standard time.

# Enoclerus lecontei

A total of four specimens of <u>E</u>. <u>lecontei</u> were caught by the nets during the three seasons of flight. Two insects were collected on August 2, 1963 between 12:00 and 3:00 PM standard time while the air temperature within the stand increased from 68° to 74°F. A third specimen was collected from 1:00 to 2:00 PM standard time on June 19, 1965 at a temperature of 62° to 64°F. within the stand, and the fourth specimen from 1:00 to 2:00 PM standard time on July 3, 1965 while the temperature was 72° to 73°F.

### Discussion

A comparison of the diurnal flight patterns of E. sphegeus,

E. lecontei, and T. undatulus with that of the Douglas-fir beetle found on Marys Peak by Rudinsky (1965) and Jantz (1965) indicates considerable similarity among the predators and the prey. The most noteworthy difference between the flight patterns of E. sphegeus and

T. undatulus and that of the Douglas-fir beetle is the time of flight cessation. Whereas the predators had discontinued flight by 5:00 PM standard time regardless of the temperature each day except for two occasions, the Douglas-fir beetles were found to fly throughout the evening until darkness when the temperature was permissive. The apparent absence of mid-day flight depressions is most likely due to the early occurrence of E. sphegeus and T. undatulus each season. The highest temperature recorded within the stand during the period of high flight activity in 1964 and 1965 was 72°F. Higher temperatures were not reached until mid-summer when the number of predators in flight was too low to distinguish the presence of such depressions.

### Seasonal Flight Patterns

The information obtained concerning the seasonal flight patterns of the three species varies as it did for the diurnal flight. The numbers of insects of each of the species trapped by the nets were too low for determining differences of flight activity within each season. The information presented here is based on the insects trapped in the olfactometers using a standard attractant source of  $2\frac{1}{2}$  percent Douglas-fir oleoresin unless stated otherwise.

### Enoclerus sphegeus

The variations between the seasonal records of flight for <u>E</u>.

<u>sphegeus</u> in 1964 and 1965 (Figures 14, 15) can be explained by considering the temperature differences of the two years. Flight for each season apparently began with the first occurrence of temperatures sufficiently high for flight activity (55° to 60°F.) and continued at high levels until about June 1 on scattered days whenever the temperatures were not restrictive. Two days of high flight activity occurred in 1964 on April 28 and May 15 while three periods of lower activity occurred in 1965 on April 26, May 10, and June 2.

After May 23 in 1964 and June 2 in 1965, the flight activity, based on olfactory responses, decreased sharply and then continued at low levels through the remainder of each season. In order to explain this pattern, <u>E. sphegeus</u> found on windthrown trees were marked with paint on May 27 and June 1, 1965. Subsequent insect counts yielded insufficient numbers of marked beetles to provide significant evidence for explanation of the rapid decrease in catches to the olfactometers, but the numbers of naturally occurring beetles (Table 3) suggests that this species was most abundant on logs about June 1 and gradually declined in numbers thereafter.

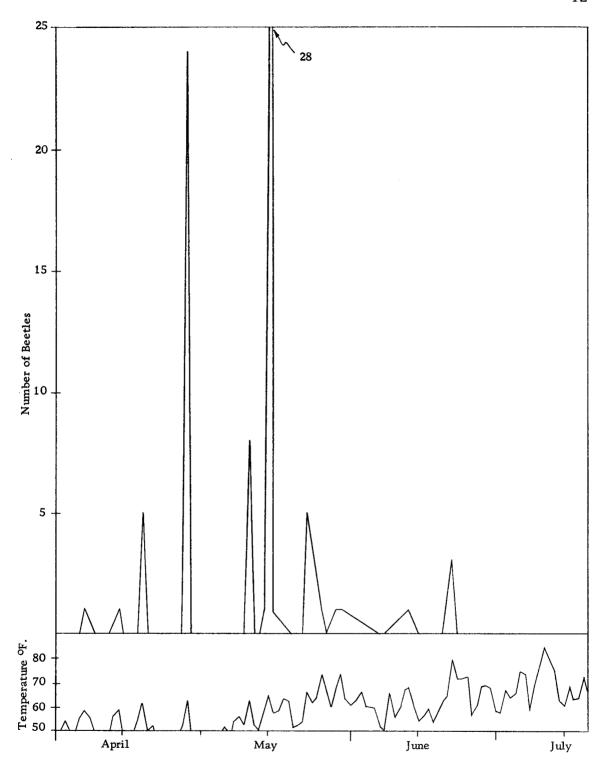


Figure 14. The seasonal pattern of flight activity of <u>E</u>. sphegeus in 1964 based on the highest number of beetles caught per day in a single hour to one olfactometer containing 2 1/2% Douglas-fir oleoresin.

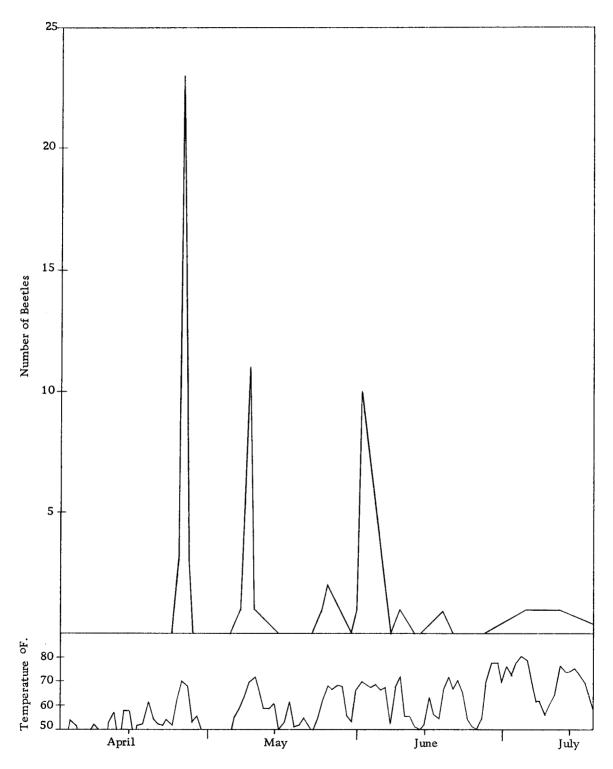


Figure 15. The seasonal pattern of flight activity of <u>E</u>. sphegeus in 1965 based on the highest number of beetles caught per day in a single hour to one olfactometer containing 2 1/2% Douglas-fir oleoresin.

Table 3. The numbers of E. sphegeus adults counted on a log during the last period of major flight activity in 1965.

Date	Number of E. sphegeus
May 25	37
June 1	85
June 10	33
June 16	40
June 28	18
July 6	0
July 13	0

# Thanasimus undatulus

None of the methods used to measure the level of flight activity of these predators trapped sufficient numbers of <u>T</u>. undatulus to establish reliable seasonal flight curves for the species. The first and last days of each season on which <u>T</u>. undatulus were recorded flying (Table 4) are similar to those found for <u>E</u>. sphegeus (Figures 11, 12). The periods of maximum flight activity also appear to coincide in 1964 and 1965 although sufficient evidence was not obtained to fully support it.

Table 4. The first and last day of 1963, 1964, and 1965 on which the flight of <u>T</u>. <u>undatulus</u> was noted by trapping with metal olfactometers, attraction cages, or nets.

Year	First day of flight	Last day of flight
1963	April 28	June 13
1964	March 29	June 22
1965	April 26	June 3

## Enoclerus lecontei

The information obtained concerning the flight activity of

E. lecontei consists of the trapping of two specimens on August 2,
1963, one specimen on June 19, 1965 and another one on July 3,
1965. Each of these four insects were trapped in the nets.

### Discussion

The periods of flight activity of <u>E</u>. <u>sphegeus</u> and <u>T</u>. <u>undatulus</u> found in this study are similar to that found for the Douglas-fir beetle by Rudinsky (1965) and Jantz (1965). While peaks of emergence and flight of the Douglas-fir beetle occur in May, the two species of predators were found to fly during 1964 and 1965 primarily from late April to early June. This synchronization of the predator and prey flight periods tends to favor a higher occurrence of predation by increasing the probability that members of the two insect groups

would occur at the same time on the tree. This simultaneous occurrence of Douglas-fir beetles and <u>E. sphegeus</u> adults on logs was reported by Cowan and Nagel (1965) based on five-minute observation periods made throughout the 1964 season. The trapping of specimens of <u>E. lecontei</u> only from mid June through the first week of August suggests that this predator is not associated with the Douglas-fir beetle as closely as the other two species of predators and that this species may be predatory mainly on another species of Scolytidae.

The periods of flight activity found in this study also conform with the seasonal schedule of adult occurrence reported by Cowan and Nagel (1965) except for the emergence of new adults of <u>E</u>.

sphegeus and <u>T</u>. undatulus during August and September. No increase was found in the number of <u>E</u>. sphegeus or <u>T</u>. undatulus trapped by either the nets or olfactometers during this time of year. Possible reasons for the failure of obtaining evidence of these emergence periods include (1) the newly emerged adults were not attracted to the oleoresins used as bait in the olfactometers, (2) the nets used were not efficient enough for sampling insects with such low population levels, and (3) the newly emerged beetles may remain near the site of emergence and not search for prey on recently downed logs.

## Olfactory Behavior

Evidence of the occurrence of oriented flight of <u>E</u>. <u>sphegeus</u> and <u>E</u>. <u>lecontei</u> was reported by Vité and Gara (1962). They reported the two species of predators to be attracted to ponderosa pine logs infested with two species of Scolytidae as well as to uninfested logs of ponderosa pine. Similar evidence was found by Rudinsky (1965). Small numbers of <u>E</u>. <u>sphegeus</u> and <u>T</u>. <u>undatulus</u> were collected from cages containing Douglas-fir logs infested with <u>D</u>. <u>pseudotsugae</u> and uninfested Douglas-fir logs while no predators were found on empty cages.

Considerable variation in the numbers of predators trapped occurred from one day of testing to another as well as among different hours of the same day. This variation limits the usefulness of the data obtained to the assigning of general qualitative characteristics to the substances tested and has prohibited most attempts to determine the relative attractiveness of the substances. Because of this limitation, it was necessary to obtain evidence that volatile substances escaping from the tree per se were attractive to the predators and that the presence of the prey was not necessary. The results of three separate tests (Tables 5, 6, 7) show that oleoresin collected from living trees of Douglas-fir, ponderosa pine, and grand fir are attractive to E. sphegeus and to a lesser extent, T. undatulus adults.

Table 5. Results of a three-hour olfactometer test on May 23, 1964.

	Number of predators trapped	
Materials tested	E. sphegeus	T. undatulus
2½% Douglas-fir oleoresin	8	1
$2\frac{1}{2}\%$ grand fir oleoresin	10	0
Empty check	0	0

Table 6. Results of a four-hour olfactometer test on May 26, 1964.

	Number of predators trapped	
Materials tested	E. sphegeus	T. undatulus
$2\frac{1}{2}\%$ ponderosa pine oleoresin	4	0
$2\frac{1}{2}\%$ grand fir oleoresin	2	2
$2\frac{1}{2}\%$ Douglas-fir oleoresin	1	3
Empty check	0	0

Table 7. Results of a  $5\frac{1}{2}$ -hour olfactometer test on April 26, 1965.

	Number of predators trapped	
Materials tested	E. sphegeus	T. undatulus
$2\frac{1}{2}\%$ ponderosa pine oleoresin	1 79	2
$2\frac{1}{2}\%$ Douglas-fir oleoresin	59	1

No empty olfactometer was used as a control on April 26, 1965 when the highest numbers of predators were caught to the oleoresins. As can be seen in Table 2, olfactometers containing nothing, 95 percent ethanol, and benzene were run a total of 87, 30, and eight hours respectively during the 1964 and 1965 seasons. One or more of these controls were used on most of the days when high numbers of predators were trapped. After May 9, 1964, at which time the six olfactometers were relocated to provide 100 feet between each one, one <u>E</u>. sphegeus and two <u>T</u>. undatulus adults were the only clerid specimens trapped at an empty, ethanol, or benzene control olfactometer during the two years of study.

The concentration of  $2\frac{1}{2}$  percent used for the oleoresins was selected after preliminary tests appeared to indicate that this concentration was the most attractive (Tables 8, 9).

Table 8. Results of a four-hour olfactometer test on April 28, 1964.

	Number of predators trapped	
Materials tested		T. undatulus
$2\frac{1}{2}\%$ Douglas-fir oleoresin	49	0
50% Douglas-fir oleoresin	14	0

Table 9. Results of a three-hour olfactometer test on May 11, 1964.

Materials tested	Number of predators trapped	
	E. sphegeus	T. undatulus
½% Douglas-fir oleoresin	0	0
$2\frac{1}{2}\%$ Douglas-fir oleoresin	17	1
5% Douglas-fir oleoresin	2	0
Empty check	0	0

Additional tests were conducted with the olfactometers to determine which of the various fractions of Douglas-fir, ponderosa pine, and grand fir oleoresins were attractive to the two species of predators. The one percent concentration used for all the fractions was selected only because it was less than that used for the oleoresins and not because this concentration was found to be most attractive. The results of some of these tests are presented in Tables 10-14. Although much variability is present among the tests, some of the fractions that consistently attracted E. sphegeus, and to a much lesser extent T. undatulus, were alpha-pinene, beta-pinene, limonene, and camphene while the fractions myrcene, geraniol, and alpha-terpineol usually attracted few or no predators.

Table 10. Results of a four-hour olfactometer test on May 15, 1964.

	Number of predators trapped	
Materials tested	E. sphegeus	T. undatulus
1% beta-pinene	49	1
1% alpha-pinene	36	1
1% Limonene	13	1
l% Myrcene	0	0
Empty check	0	0

Table 11. Results of a  $2\frac{1}{2}$ -hour olfactometer test on April 26, 1965.

	Number of predators trapped	
Materials tested	E. sphegeus	T. undatulus
1% beta-pinene	51	1
1% alpha-pinene	39	4
1% Limonene	38	2
$2\frac{1}{2}\%$ Douglas-fir oleoresin	37	1
1% Camphene	15	1

Table 12. Results of a three-hour olfactometer test on April 26, 1965.

Materials tested	Number of predators trapped	
	E. sphegeus	T. undatulus
$2\frac{1}{2}\%$ Douglas-fir oleoresin	22	0
1% Limonene	14	1
1% Camphene	11	0
l% Geraniol	5	0
l% alpha-terpineol	0	0

Table 13. Results of a three-hour olfactometer test on April 27, 1965.

	Number of predators trapped	
Materials tested	E. sphegeus	T. undatulus
$2\frac{1}{2}\%$ Douglas-fir oleoresin	5	0
1% alpha-Terpineol	4	1
1% Limonene	2	0
1% Geraniol	1	0
1% Camphene	0	2
Empty check	0	1

Table 14. Results of a four-hour olfactometer test on May 10, 1965.

	Number of predators trapped	
Materials tested	E. sphegeus	T. undatulus
$2\frac{1}{2}\%$ Douglas-fir oleoresin	21	1
1% Geraniol	5	1
1% alpha-Terpineol	0	0

It is of interest that alpha-pinene, beta-pinene, and limonene, which are three of the four fractions that appear to be most attractive to <u>E</u>. <u>sphegeus</u> and possibly <u>T</u>. <u>undatulus</u>, are present in the oleoresins of most of the tree species with which these predators have been reported to be associated (Table 1). Also of interest is that the three fractions myrcene, geraniol, and alpha-terpineol which are of questionable attractiveness to the predators, have been found to occur in the oleoresin of only one or two of the tree species the

predators have been reported to be associated with. These two relationships suggest that <u>E</u>. <u>sphegeus</u> and <u>T</u>. <u>undatulus</u> adults locate concentrations of prey insects by being attracted directly to the volatile materials escaping from the tree host of the prey insects. It is also suggested that the predators <u>E</u>. <u>sphegeus</u> and <u>T</u>. <u>undatulus</u> would be effective controlling agents only on those insects that are associated with tree species that produce oleoresins containing specific fractions attractive to these predators.

The complete absence of the third species <u>E</u>. <u>lecontei</u>, at the olfactometers is contradictory to the attraction reported by Vité and Gara (1962). In view of the fact that only four specimens were trapped in the nets during three years of sampling, it appears that either the population level of <u>E</u>. <u>lecontei</u> is very low in the vicinity of the study area, or else the conditions within the stand surrounding the area is unfavorable to the species. In either case, not enough specimens were collected in the nets to conclude that <u>E</u>. <u>lecontei</u> was present but unresponsive in the area.

Attempts were made to find a laboratory procedure which could be used under standardized conditions to test the olfactory behavior of <u>E</u>. sphegeus. Neither the arena olfactometer nor the arrestment board was found to be satisfactory for this purpose because of the excitable nature of this beetle. The handling of the beetles immediately prior to the testing caused them to either feign death or attempt

to escape by running or flying.

Tests designed for determining the possible production of a sex excitant or attractant by <u>E</u>. <u>sphegeus</u> females similar to that reported for <u>T</u>. <u>undatulus</u> females by Cowan and Nagel (1965) provided no indication that such a material was present. No interspecific matings were observed between <u>E</u>. <u>sphegeus</u> males and <u>T</u>. <u>undatulus</u> females from 25 such pairs. Three <u>E</u>. <u>sphegeus</u> males out of 25 attempted to mate with recently killed <u>E</u>. <u>sphegeus</u> females when air from a container with living females was pumped into the chamber. Of a control group of 25 other males tested, without the air being pumped into the chamber, four males attempted to mate with the dead females.

#### **SUMMARY**

The diurnal flight pattern of <u>E</u>. <u>sphegeus</u> was found to be similar to that of the Douglas-fir beetle. Adults of <u>E</u>. <u>sphegeus</u> were found to fly only when ambient air temperatures within the stand were 55°F. or higher. Flight was observed at 80°F., but no maximum temperature limit of flight activity was determined. Flight activity was found to be restricted by continuous winds of velocities greater than approximately five miles per hour within the stand while no effect was detected by winds above the forest canopy or gusts within the stand. Flight occurred almost exclusively between 10:00 AM and 5:00 PM standard time although air temperatures before and after this seven hour interval were high enough for flight activity.

The diurnal flight pattern exhibited by <u>T. undatulus</u> was found to be similar to that of <u>E. sphegeus</u>. The minimum and maximum air temperatures occurring within the stand while <u>T. undatulus</u> adults were trapped in flight were 61° and 77°F. respectively, but no temperature limits of flight activity were determined.

E. lecontei were trapped while in flight during mid-day while the air temperatures within the stand were between 62° and 74°F., but the lower and upper temperature limits of flight activity were not determined.

The seasonal flight patterns of <u>E</u>. <u>sphegeus</u> and <u>T</u>. <u>undatulus</u>

were found to closely resemble that of the Douglas-fir beetle. <u>E</u>.

<u>lecontei</u> were observed in flight only after mid-June which is after the main flight period of the Douglas-fir beetle.

Adults of <u>E</u>. <u>sphegeus</u> and <u>T</u>. <u>undatulus</u> were found to be attracted to vapors of the oleoresins from Douglas-fir, ponderosa pine, and grand fir as well as to alpha-pinene, beta-pinene, camphene, and limonene which are constituents of these oleoresins. No definite attraction was found to the oleoresin fractions geraniol, myrcene, or alpha-terpineol. It is concluded from the attraction studies that <u>E</u>. <u>sphegeus</u> and <u>T</u>. <u>undatulus</u> adults locate concentrations of prey insects by being attracted directly to volatile materials escaping from the tree host of the prey insects. It is also suggested that the predators <u>E</u>. <u>sphegeus</u> and <u>T</u>. <u>undatulus</u> would be effective controlling agents only on those insects that are associated with tree species that produce oleoresins containing specific fractions attractive to these predators.

Attraction of  $\underline{E}$ . lecontei was not found to any of the materials tested.

An arrestment board and arena olfactometer used in the laboratory were found to be unsatisfactory for testing the attraction of

E. sphegeus to volatile materials. The nature of this species is such that handling or other disturbances causes them to either feigh death or attempt escape by running or flying.

No evidence was found of the presence of a volatile sex attractant or excitant produced by E. sphegeus females.

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