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NATURE OF RESISTANCE OF PINES TO BARK BEETLES†

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PATTERNS of susceptibility of pines to attack by certain species of *Dendroctonus* bark beetles suggest that a resistance mechanism exists. This situation was first called to my attention in 1949 by John M. Miller, entomologist at the Berkeley Forest Insect Laboratory. He was studying the resistance of pines to insects, at the Institute of Forest Genetics near Placerville, California. Miller worked closely with N. T. Mirov, who was studying differences in the volatile constituents of wood oleoresins of pine species. Miller, as others had suggested before, felt that differences in oleoresin constituents might cause the specificity of attacks by *Dendroctonus brevicomis* Lec. on *Pinus ponderosa* Laws.

Following his lead, I explored possible resistance mechanisms involved in this host specificity. Volatile constituents of oleoresin exuding from wounds in the xylem of pines emerged as possible insect toxicants. In a report prepared in 1952* I expanded and developed the hypothesis that differential tolerance for turpentine compounds might account for host specificity in *Dendroctonus* and be one key to resistance. Subsequent research has supported this hypothesis. Further, it suggests that turpentine chemicals could be one of the most important resistance factors that any insect encounters in attacking living pines. This paper is a restatement and updating of the key points in my earlier report.

HOST SPECIFICITY

Host specificity is one manifestation of resistance. Allee *et al.* (1949, p. 258) define it as usually referring "to the tendency of many parasites to

* Callaham, R. Z. 1952. Host specificity in the *Dendroctonus Pinus* complex. (Unpublished report on file at the Pacific Southwest Forest and Range Experiment Station, U.S. Forest Serv., Berkeley, Calif.)

† This paper was not presented during the Advanced Study Institute, but was given in summary form by the author during a discussion period.

attack a single species or a limited number of taxonomically related species". For *Dendroctonus* and *Pinus* the term refers to the restriction of successful (i.e. tree-killing) attacks of one beetle species to one or more pine species—its natural hosts. Host specificity and its causes have been discussed to only a limited extent in the literature, and it is difficult to find a definite statement on the subject in general texts.

Information on host specificity is particularly limited for the genus *Dendroctonus*. Hopkins (1909, 1916) merely pointed to its occurrence and to its taxonomic value. Perhaps the closest approach to the subject of causality was made by Gordon.† He stood on the threshold when he recommended investigations of the quantities and physical state of resinous substances in oleoresin of resistant and susceptible trees. He also recommended testing responses of a given insect to various tissues of its host and of other species. Gordon suggested that "these investigations might lead to information as to the cause of specificity which conditions the exclusive attack by a given insect on a given species of tree in certain cases".

Experiments have demonstrated that host specificity of *Dendroctonus* species disappears if the host pines are not living. *Dendroctonus* bark beetles made successful (brood-producing) attacks on trap logs of nonhost pine species, but even when forced they cannot successfully attack and kill living trees of nonhost species. Some feature of the living tree enables it to resist attacks by

† Gordon, A. 1932. Tree injection experiments in white fir and ecological studies of insects attacking yellow pine. Season of 1931. (Unpublished progress report on file at the Pacific Southwest Forest and Range Experiment Station, U.S. Forest Serv., Berkeley, Calif.)

certain species of *Dendroctonus*. What then might be the resistance mechanism of the living pine?

Painter (1951) breaks down the causes of resistance into three categories—host preference, tolerance, and antibiosis. These three resistance mechanisms can be examined in reference to specificity of *Dendroctonus* bark beetle attacks on pine species.

According to Painter (1951), "Preference or non-preference is used to denote the group of plant characters and insect responses that lead to or away from the use of a particular plant or variety, for oviposition, for food, or for shelter, or for combinations of the three." After nearly half a century forest entomologists still know little about how *Dendroctonus* bark beetles select trees to attack (Miller and Keen, 1960). Significantly, no one has resolved whether initial attacks occur at random or in response to some stimuli. We cannot yet establish the role or importance of preference in host specificity.

Tolerance refers to the ability of a tree to repair insect injury or to grow despite insect attacks. Tolerance does not play an important role in this host specificity problem. A concentrated, successful attack—one producing brood around the entire circumference of the tree—always kills the host. Patch kills are not unknown, but neither are they common. When they occur, it is a matter of pseudo-resistance, not of host specificity. Recovery from a successful concentrated attack is unknown, and seems unlikely. The adult beetles and their broods mine through and destroy the vital phloem tissues and cambial layer. They also desiccate the host and inoculate it with blue-stain organisms (Caird, 1934).

Antibiosis refers to the injury or destruction of insects by the host. It has only recently been investigated as a factor in the host specificity problem. Gordon made a few tests of the contact and fumigant toxicity of oleoresin compounds but left no data. He concluded the compounds were toxic.

In 1951 and 1952, Miller and I forced adults of *Dendroctonus brevicomis* Lec., *D. monticolae* Hopk., and *D. jeffreyi* Hopk. to attack nonhost species of pines. We concluded from our observations that wood oleoresin of certain pine species is very toxic to individuals of certain *Dendroctonus* species. In some of our experiments, the first pitch flow

from the wounded nonhost trees killed the attacking insect (Fig. 1). The point of attack was not marked by a copious flow of pitch. For example, ponderosa pine (*Pinus ponderosa* Laws.) and Jeffrey pine (*P. jeffreyi* Grev. and Balf.) were tested for resistance to the western pine beetle and the mountain pine beetle. Attacks of these beetles on their host—ponderosa pine—resulted in production of very large pitch masses at the entrances to galleries. On the other hand, attacks of these beetles on the nonhost—Jeffrey pine—resulted in no pitch flow. Apparently the oleoresin was extremely toxic to the beetles. Adults usually succumbed before any pitch was visible on the bark surface. Their attacks never penetrated farther than the first contact with the phloem. First contact of beetle and oleoresin decided resistance of the host. Obviously oleoresin had an antibiotic action. It deserved further study as an antibiotic causing host specificity.

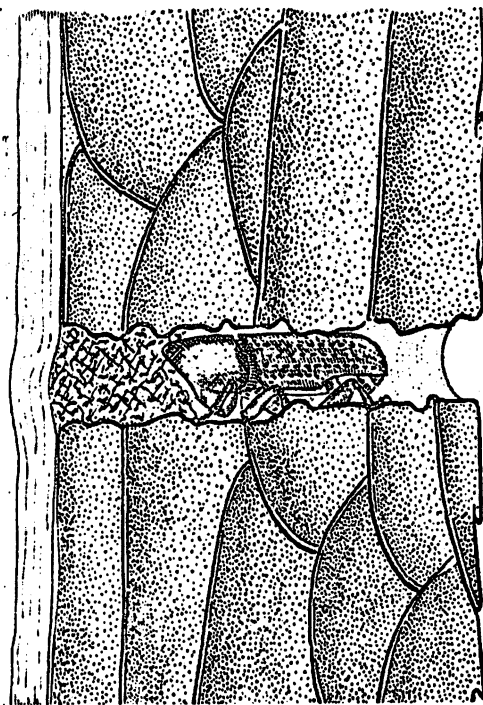


FIG. 1. In the entrance gallery of *Dendroctonus brevicomis* attacking the nonhost—*Pinus jeffreyi*—the first resin flow was lethal.

OLEORESIN AS AN ANTIBIOTIC

Oleoresin exuding from severed pine resin ducts consists of two major fractions. Resin acids comprise the rosin fraction, about 70 to 90 per cent by volume. The remainder is the volatile turpentine or essential oil. Rosin is of unknown but doubtful importance as a toxic agent to insects because of its chemistry. Turpentine, on the other hand, is composed of various aliphatic hydrocarbons, terpenes, and sesquiterpenes; its toxicity has been demonstrated in the laboratory (Blackman, 1915, p. 65; Smith, 1963).

We might propose that turpentine constituents are the antibiotics controlling host specificity. Accordingly, we would expect to find major differences between species in turpentine constituents, and relatively minor differences between trees of a species. The vast amount of literature on turpentine constituents supports this expectation. Species have highly heritable and specific characteristic turpentine constituents (Mirov, 1961; Williams and Bannister, 1962). Individuals of some species show great similarity in kinds and amounts of turpentine constituents (Bannister *et al.*, 1962; Smith, 1964a). Strong evidence for intraspecific variation in constituents in other species (Smith, 1964b) is emerging. Individuals, races, varieties, or subspecies of widely distributed species may vary considerably in turpentine constitution (Mirov and Iloff, 1958; Peloquin, 1964*).

To evaluate insecticidal properties of turpentine compounds, we must be familiar with properties of toxic chemicals. Brown (1951, p. 65) summarizes trends in insecticidal activity in relation to chemical structure. When the chemical structure of turpentine compounds is evaluated in terms of toxicity, several generalizations can be made:

1. None of the compounds is of optimum molecular weight—300 to 400—for use as a contact insecticide. Sesquiterpenes have the largest molecules, but they are present only in limited amounts. They are the only compounds exhibiting molecular weights over 200.

2. Except for tricyclic sesquiterpenes, most of the compounds have only one or two rings of

carbon atoms. This enhances their value as contact insecticides.

3. Most of the toxic substituent groups like Cl, SCN, NO₂, and CN are lacking in turpentine compounds.

4. None of the compounds possesses unsaturation in an aliphatic side chain to enhance toxicity.

5. Neither the toxophoric C=P—C=O configuration nor ether linkages are present.

6. Most significant is that all compounds rate highly as to lipid solubility. Each cyclic terpene consists of a basic 6-carbon ring and methyl and methyl analogues substituted in the para position. This characteristic adds to its value as a contact insecticide.

7. n-heptane is the most common aliphatic hydrocarbon found in turpentine. It has a greater fumigant toxicity than other saturated paraffin hydrocarbons. Thus, n-heptane may be an important fumigant in the restricted environment of the bark beetle entrance gallery. n-undecane is present in turpentine only occasionally and in small amounts. It would not be important as a fumigant owing to its low volatility.

Turpentine does contain chemically toxic compounds, which theoretically should differ in toxicity. In recent experiments Smith (1965a) demonstrated the variable toxicity of turpentine compounds to *Dendroctonus brevicomis*. Thirty years earlier, Gordon† made crude toxicity tests with bark beetle species. He reported: "The relative toxicity of the essential oils was found to be as follows: Pinene > limonene > carvone > p-cymene." Extensive experiments are warranted now to find the turpentine chemicals most toxic to each species of bark beetle.

TURPENTINE AND RESISTANCE

I would predict that each bark beetle species will have a relatively high tolerance for turpentine compounds found in the pines it normally attacks. Conversely, turpentine compounds not found in its host pines may be relatively more toxic. For example, *Dendroctonus rufipennis* (Kby.) and *D. murrayanae* Hopk. are two very closely related

* Peloquin, R. L. 1964. Geographic variation of the monoterpenes of *Pinus ponderosa*. (Unpublished master's thesis on file at Stanford University, Palo Alto, Calif.)

† Gordon, A. 1933. Apparatus used in a study of the western pine beetle. (Unpublished report on file at the Pacific Southwest Forest and Range Experiment Station, U.S. Forest Serv., Berkeley, Calif.)

species. The former attacks only *Pinus strobus* L. and *P. banksiana* Lamb. In these host species it would encounter only large amounts of α -pinene and smaller amounts of β -pinene, both bicyclic terpenes. *D. murrayanae* attacks only *P. contorta* var. *latifolia* Engelm. In this host it encounters mostly β -phellandrene, a monocyclic terpene. If our hypothesis holds, β -phellandrene should be more toxic to *D. rufipennis*; α - and β -pinene should be more toxic to *D. murrayanae*. Smith (1965b) reports a parallel story involving *D. jeffreyi* and *D. monticolae* versus n-heptane, α -pinene, and Δ_3 -carene.

When relative tolerances for turpentine compounds have been established for each species of *Dendroctonus*, then geneticists will be able to produce resistant pines. The work at Placerville demonstrates that interspecific hybrids resist *Dendroctonus* species which kill the parent species. Resistant interspecific hybrids can be produced in abundance for forest planting. We know that F_1 hybrids inherit the turpentine compounds of their parents (Mirov, 1961; Forde, 1964). In addition, Forde's (1964) work demonstrates that turpentine compounds may be moved from one species to another. By successive selection and crosses, geneticists could produce resistant strains of desired species. Smith's (1964b) current work at Placerville shows that certain ponderosa pines have high concentrations of the more toxic compounds in their turpentine. I expect such trees will be resistant so long as they can produce oleoresin. If this is true, we will soon be able to determine the resistance of ponderosa pines to beetles in only 5 or 6 minutes by using gas chromatography. In the next decade we will develop the ability to produce pines resistant to bark beetles by both hybridization and selection.

Our success in developing bark beetle resistance has three ingredients. First is the timely close association of scientists Miller and Smith—the bark beetle specialists—and Mirov—the biochemist—with the geneticists and their pine hybrids at the Institute of Forest Genetics. Second is our abundant knowledge of turpentine composition in *Pinus*. Third is the advanced technology of gas

chromatography for "fingerprinting" the turpentine of each pine tree or of an individual resin canal* in a few minutes.

Finally, I suggest that turpentine chemistry may be the key to resistance of pines to many other kinds of insects. Any insect encountering oleoresin must have inherent resistance to toxicants in the turpentine. Fumigation tests or topical applications of turpentine and its compounds should expose such resistance or lack of resistance. It should be pointed out that different turpentine compounds are obtained from oleoresin originating in xylem, needles, cortex, buds, or strobili. Hence, care must be taken to expose insects to the appropriate turpentine.

SUMMARY

Host specificity of species of *Dendroctonus* attacking pines suggests that living pines have a mechanism for resisting certain types of bark beetles. Painter's three general resistance mechanisms—preference, tolerance, and antibiosis—were considered as they relate to *Dendroctonus* and *Pinus*. Pine oleoresin has antibiotic properties. Turpentine, the volatile fraction of oleoresin, is toxic to *Dendroctonus* adults. Beetle species have inherent resistance to the turpentine of their host species. The compounds in turpentine vary between species, between races of some species, and even between individual pines in populations of certain species. Turpentine compounds differ in their insecticidal properties. Thus, species, races, or individual pines may vary in resistance as their turpentine composition varies. Geneticists have produced interspecific pine hybrids chemically resistant to certain beetle species. When relative tolerances for turpentine compounds have been established for other species of *Dendroctonus*, then resistant pines may be produced by both hybridization and selection. Turpentine chemistry may also be the key to developing pines resistant to other insects that contact oleoresin in their attack or development.

LITERATURE CITED

- ALLEE, W. C., A. E. EMERSON, O. PARK, T. PARK, and K. P. SCHMIDT. 1949. *Principles of animal ecology*. 837 pp. Philadelphia: W. B. Saunders Co.

* Personal communication from Dr. Stanley L. Krugman, Institute of Forest Genetics, Placerville, California, August 1964.

- BANNISTER, M. H., A. L. WILLIAMS, I. R. McDONALD, and M. B. FORDE. 1962. Variation of turpentine composition in five population samples of *Pinus radiata*. *New Zeal. J. Sci.* 5, 486-95, illus.
- BLACKMAN, M. W. 1915. II. Observations on the life history and habits of *Pityogenes hopkinsi* Sw. *N.Y. State Col. Forestry Tech. Pub.* 2, 66 pp.
- BROWN, A. W. 1951. *Insect control by chemicals*. 817 pp. New York: John Wiley and Sons, Inc.
- CAIRD, R. W. 1934. Physiology of pines infested with bark beetles. *Bot. Gaz.* 96, 709-33.
- FORDE, M. B. 1964. Inheritance of turpentine composition in *Pinus attenuata* $\times$ *radiata* hybrids. *New Zeal. J. Bot.* 2, 53-9, illus.
- HOPKINS, A. D. 1909. Contributions toward a monograph of the scolytid beetles. I. The genus *Dendroctonus*. U.S. Dept. Agr., Bur. Ent. Tech. Ser. 17, Pt. 1, 64 pp.
- HOPKINS, A. D. 1916. Economic investigations of the scolytid bark and timber beetles of North America. U.S. Dept. Agr. Program of Work, 1917, p. 353.
- MILLER, J. M., and F. P. KEEN. 1960. Biology and control of the western pine beetle. U.S. Dept. Agr. Misc. Pub. 800, 381 pp., illus.
- MIROV, N. T. 1961. Composition of gum turpentines of pines. *U.S. Dept. Agr. Tech. Bull.* 1239, 158 pp., illus.
- MIROV, N. T., and P. M. ILOFF, JR. 1958. Composition of gum turpentines of pines. XXIV. A report on *Pinus ponderosa* from five localities: central Idaho, central Montana, southeastern Wyoming, northwestern Nebraska, and central eastern Colorado. *J. Amer. Pharm. Assoc., Sci. Ed.* 47, 404-9.
- PAINTER, R. H. 1951. *Insect resistance in crop plants*. 520 pp. New York: Macmillan Co.
- SMITH, R. H. 1963. Toxicity of pine resin vapors to three species of *Dendroctonus* bark beetles. *J. Econ. Ent.* 56, 827-31, illus.
- SMITH, R. H. 1964a. The monoterpenes of lodgepole pine oleoresin. *Phytochem.* 3, 259-62, illus.
- SMITH, R. H. 1964b. Variation in the monoterpenes of *Pinus ponderosa* Laws. *Science* 143, 1337-8, illus.
- SMITH, R. H. 1965a. Effect of monoterpene vapors on the western pine beetle. *J. Econ. Ent.* 58, 509-10, illus.
- SMITH, R. H. 1965b. A physiological difference among beetles of *Dendroctonus ponderosae* ($\neq$ *D. monticolae*) and *D. ponderosae* ($=$ *D. jeffreyi*). *Ann. Ent. Soc. of Amer.* 58, 440-2, illus.
- WILLIAMS, A. L., and M. H. BANNISTER. 1962. Composition of gum turpentines from twenty-two species of pines grown in New Zealand. *J. Pharm. Sci.* 51, 970-5.