Variation in Monoterpene Composition of Loblolly Pine as Related to Geographic Source of Seed'

A. R. Gilmore
University of Illinois at Urbana-Champaign2

A number of studies have shown local and/or regional variation in the monoterpenes of some coniferous species. One of the most comprehensive studies was conducted by Smith et al. (1969) on ponderosa pine. Based on terpene analyses, they found evidence to support the establishment of at least four major and four minor regional types of ponderosa pine. 'No study of this nature has been reported for the southern pines. There are many reasons why this is true, one being the scarcity of suitable plant material growing in an area where differences in environment are relegated to a minimum.

A 21-year-old plantation in southern Illinois provided an opportunity to study the relationship between monoterpenes and geographic races of loblolly pine (Pinus taeda L.) growing in a uniform area but outside its natural range. The most northerly range of loblolly pine in the Mississippi River Valley is in northern Mississippi and western Tennessee, which is about 200 miles south of the plantation used in this study.

Methods

Seed from six geographic sources of loblolly pine were obtained from trees growing in the following areas and planted during the 1948 season in the Union State Nursery near Jonesboro, Illinois: (1) southwestern Arkansas (Ark.); (2) throughout Mississippi (Miss.); (3) throughout South Carolina (S.Car.); (4) Pender County, North Carolina (N.Car.); (5) Matthews County, Virginia (Va.); and (6) Worcester County, Maryland (Md.).

Planting stock was graded to eliminate culls, according to the quality standards used in the Jonesboro nursery at that time, and 1-0 seedlings were planted at 6- x 6-foot spacing in two blocks of a randomized block design. The plantation is located in the "claypan" area of southern Illinois about 4 miles west of West Frankfort. The soil type is a Wynoose silt loam (Typic Albaqual) and is relatively uniform over the planted area. Growth measurements were taken by staff members of the Carbondale Research Center, U. S. Forest Service, at the end of the tenth growing season. At this time mean tree height ranged from 19.3 feet to 22.3 feet for the six seed sources, and survival percentages ranged from 81.0 percent to 95.3 percent.

'Plantation established by L. S. Minckler (retired) while he was a member of the staff at Carbondale Research Center. The plantation is a part of the "Plan for Pine Seed Source and Pine Hybrid Studies in Southern Illinois," dated September 15, 1948. The author expresses his gratitude to the North Central Forest Experiment Station, U.S.D.A., for permitting him to use ma- from the plantation in this study.

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At the end of 21 growing seasons, one tree in each of the 8 interior rows in a plot was randomly selected for oleoresin analysis. Thus oleoresin samples were collected from 16 trees in each seed source. A V-notch was cut at breast height on the north side of each tree that extended through the bark to the xylem. Oleoresin oozing from each cut was placed in a small vial, and the air in the vial was replaced with nitrogen. The vials were placed on dry ice shortly after collection, and the oleoresin was kept frozen until it was used for terpene analysis.

Terpene analysis was determined with a Hewlett-Packard Model 5750 Gas Chromatograph. Oleoresin was dissolved in pentane, and the analysis was performed using a stainless steel column packed with 20 percent Carbowax 20M liquid phase on 60-80 mesh, acid-washed Chromosorb W solid support. In addition, peak separation was checked on another Column using 5 percent B, B - oxidipropionitrile in place of the Carbowax 20M. This check revealed that no peaks were produced that were not observed when the original column was used.

Concentration of each terpene is expressed as a percentage of the total terpene concentration.

Results

Five terpenes were found in the oleoresin of all seed sources. The order in which they eluted from the Gas Chromatograph column were alpha-pinene, camphene, beta-pinene, myrcene, and limonene. The average composition according to seed source is shown in Table 1.

Table 1. Average monoterpene composition according to geographic source of seed.

Monoterpene	Miss.	Ark.	S.Car.	N.Car.	Va.	Md.
			Percent			
alpha-pinene [a/]	71.59	73.22	76.48	79.94	81.55	83.54
Camphene [b/]	1.04	1.27	1.04	1.46	1.37	0.80
Beta-pinene [c/]	20.29	17.45	16.29	10.24	11.00	10.84
Myrcene	3.77	4.59	4.55	6.13	4.33	3.57
Limonene [e]	3.31	3.47	1.64	2.23	1.73	1.25

[a/] Miss. different from N.Car., Va., and Md. at 1 percent level. Ark. different from Va. and Md. at 1 percent level and from N.Car. at 5 percent level.
[b/] Md. different from Va. and N.Car. at 1 percent level and from Ark. at 5 percent level. N.Car. different from S.Car. and Miss. at 5 percent level.
[c/] Miss. different from N.Car., Va., and Md. at 1 percent level. N.Car. different from S.Car. and Ark., and Ark. different from Va. and Md. at 5 percent level.
[d/] N.Car. different from all other sources at 1 percent level.
[e/] Miss. different from Va. and Md. at 1 percent level and from S.Car. and N.Car. at 5 percent level. Ark. different from S.Car., Va., and Md. at 1 percent level and from N.Car. at 5 percent level.

Alpha-pinene averaged 71.59 percent in the most southerly seed source and increased almost linearly to the Maryland source, which averaged 83.54 percent. The concentration of camphene appeared to be greatest near the midpoint of the species natural range, decreasing to a low level at the extreme of its range. Beta-pinene was greatest in the southerly sources and the least concentrated in the northerly sources. Myrcene was similar to camphene in that the concentration was greatest near the midpoint of the species range and least at the extremes of the range. Limonene followed the general pattern of beta-pinene in that its concentration was greatest in trees grown from seed obtained from southern sources and least in the northern sources.

There was considerable tree-to-tree variation in the composition of the terpene fractions not only for all trees (Table 2) but also for trees within a seed source.

Table 2. Ranges in terpene concentration for all trees.

Terpene	Range (percent)
Alpha-pinene	56.56 - 95.51
Camphene	0.03 - 2.21
Beta-pinene	1.13 - 37.30
Myrcene	1.53 - 9.29
Limonene	0.26 - 6.84

There was no difference in terpene composition by blocks. The total height of the plantation was relatively uniform and averaged about 50 feet. The diameter at breast height (dbh) of the study trees ranged from 4.1 to 10.1 inches with the mean 6.5 inches. No correlation was found between either terpene concentrations and dbh or total height.

Discussion

When compared with other studies, terpenes in this study show similarities as well as differences. For example, Smith et al. (1969) found five major terpenes in ponderosa pine oleoresin, four of them the same as those found in loblolly pine. They reported only a trace of camphene in ponderosa but found large amounts of 3-carene. Lotan and Joye (1970) reported that lodgepole pine contained the five terpenes found in loblolly, plus four others. In slash pine, Roberts (1970) found two additional terpenes beside the five reported for loblolly. These differences demonstrate that terpenes differ widely in Pinus species, even in the closely related species such as yellow pines.

Environment had little effect on terpene concentration in the study, as the trees are growing on a relatively uniform area. Therefore, differences in terpene concentrations must be related to genetics, even though tree-to-tree variations in terpene levels are large.

An interesting aspect of the study is to speculate on the relationship of monoterpenes to plant-insect interactions. Since beetles react differently to terpene variation, a paramount consideration is the behavior of beetles to resin vapor prior to tissue invasion. The volatile fraction of oleoresin, the terpenes, apparently plays a dual role in pine-beetle relationships. Vapors of individual terpenes have the ability either to attract beetles to the source tree (Chapman, 1963), or to repel or kill the insect. Smith (1965) has demonstrated the variable toxicity of pine resin vapor to several species of Dendroctonus bark beetles. He noted that vapor concentration seems to be the factor determining whether the terpenes will be attractants or deterrents. Also, Chararas (1958) found that high concentrations the terpenes, alpha-pinene, beta-pinene, and terpineol were all repellent to bark beetles, but attractants in low concentration. Concentration may not be the only factor that determines the toxicity or attractiveness of the terpenes. Olfactometer tests with Douglas-fir beetles (Heikkenen and Hrufiord, 1965) indicated that alpha-pinene is an attractant, whereas beta-pinene may repel the beetles. They hypothesized that when the ratio of alpha-pinene to beta-pinene is low, the beetles are repelled; this is the normal situation, but, according to their hypothesis, the ratio can be upset when low soil moisture produces a reduction in the cooling properties of evapotranspiration; then the heating effect of radiant energy might cause volatilization of alpha-pinene, resulting in beetle attraction.

The author is unaware of any study pertaining to the susceptibility of these geographic races of loblolly pine to insect attack. Nevertheless, terpene composition has been shown to be an inherited factor in a number of coniferous species and that terpenes might play a part in attracting or repelling insects to a particular host. For example, Smith et al. (1969) found distinct patterns of monoterpene composition in geographic races of ponderosa pine. They noted that the western pine beetle is not found in the region occupied by ponderosa pine of var. scopulorum and that trees in this region have a high concentration of 3-carene. Also, the beetle is not found in regions occupied by var. arizonica, where alpha-pinene is high. In general, they noted that this beetle was not normally found in areas of ponderosa pine where a monoterpene occurred as a large percentage of the terpene composition.

Many clues of host-insect attractants need to be investigated, and hopefully this mystery will become clear in the not too distant future.

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