Do Red-Cockaded Woodpeckers Select Cavity Trees Based on Chemical Composition of Pine Resin?

Richard N. Conner

Follow this and additional works at: http://scholarworks.sfasu.edu/forestry

Part of the Forest Sciences Commons

Tell us how this article helped you.

Recommended Citation


http://scholarworks.sfasu.edu/forestry/280

This Article is brought to you for free and open access by the Forestry at SFA ScholarWorks. It has been accepted for inclusion in Faculty Publications by an authorized administrator of SFA ScholarWorks. For more information, please contact cdsscholarworks@sfasu.edu.
DO RED-COCKADED WOODPECKERS SELECT CAVITY TREES BASED ON CHEMICAL COMPOSITION OF PINE RESIN?

Author(s): RICHARD N. CONNER, ROBERT H. JOHNSON, D. CRAIG RUDOLPH, and DANIEL SAENZ


Published By: The Wilson Ornithological Society

DOI: http://dx.doi.org/10.1676/03-032

URL: http://www.bioone.org/doi/full/10.1676/03-032
DO RED-COCKADE WOODPECKERS SELECT CAVITY TREES BASED ON CHEMICAL COMPOSITION OF PINE RESIN?

RICHARD N. CONNER,¹,³ ROBERT H. JOHNSON,² D. CRAIG RUDOLPH,¹ AND DANIEL SAENZ³

ABSTRACT.—We examined resin chemistry of loblolly (Pinus taeda) and shortleaf (P. echinata) pines selected as cavity trees by Red-cockaded Woodpeckers (Picoides borealis) in eastern Texas. We sampled resin from (1) pines selected by Red-cockaded Woodpeckers that contained naturally excavated active cavities, (2) pines selected by forest biologists that contained artificially installed cavity inserts and were actively being used by Red-cockaded Woodpeckers, and (3) control pines of similar age and appearance to the active cavity trees. We hypothesized that if woodpeckers are inducing a change in resin chemistry by excavating resin wells, this change should appear in active cavity trees selected by woodpeckers and trees selected by biologists, but not in control pines. If woodpeckers are selecting pines that have specific resin chemistry, concentrations of some resin components in active cavity trees selected by the woodpeckers for natural cavity excavation should be different from both control pines and pines selected by biologists. A large diterpene acid peak containing an isopimaric-levopimaric-palustric methyl-ester mix in active natural cavity trees was approximately 20% greater than controls and 22% greater than trees with artificial cavities. None of the other eight resin chemicals differed among treatments. The activity of Red-cockaded Woodpeckers at resin wells did not appear to affect the composition of cavity tree resin. Woodpeckers, however, may select pines with specific resin chemistries for cavity trees. Received 17 March 2003, accepted 1 August 2003.

The Red-cockaded Woodpecker (Picoides borealis) nests and roosts nearly exclusively in living pines (Pinus spp.). Red-cockaded Woodpeckers flake off loose bark resulting in a smoother surface on the cavity tree’s bole. The woodpeckers also make daily excavations at small wounds, termed resin wells, around their cavity entrance and on the bole of their cavity tree from which resin flows down the tree (Ligon 1970). These behaviors result in a resin barrier that serves as an effective defense against rat snakes (Elaphe spp.) if the cavity tree can produce adequate resin (Jackson 1974, Rudolph et al. 1990). Rat snakes regularly attempt to climb Red-cockaded Woodpecker nest trees and are known to prey on Red-cockaded Woodpeckers when the resin barrier is inadequate (Jackson 1978a, Neal et al. 1993).

Red-cockaded Woodpeckers make a substantial investment in time and energy when excavating a cavity (Conner and Rudolph 1995), and characteristics used to select cavity trees affect the rate of cavity excavation. Red-cockaded Woodpeckers select pines that have thin layers of sapwood and large diameters of heartwood (Conner et al. 1994), as well as pines that have had their heartwood softened by fungal decay from Phellinus pini infections (Jackson 1977, Conner and Locke 1982, Hooper et al. 1991) because cavities can be more quickly excavated in such pines (Conner et al. 2001).

Red-cockaded Woodpeckers also select cavity trees based on the ability of pines to produce resin (Conner et al. 1998a), which affects the defense of the cavity. The breeding male woodpecker selects the cavity tree that produces the most resin for his roost tree, which usually becomes the woodpecker group’s nest tree during spring. Cavity trees producing higher yields of resin at resin wells likely provide better barriers against rat snakes, enhancing the probability of survival of the breeding male and his offspring (Conner et al. 1998a). Presumably, the woodpeckers either taste the resin or sense the viscosity of the resin early in the process of excavating a cavity and abandon the tree if they do not detect the proper resin composition. If the cavity is human made (artificial), the tasting-sensing stage may be circumvented, and the woodpeckers use this convenient resource be-
cause it requires little or no time-energy investment in excavation, and the availability of suitable pines for cavity excavation is often low.

Pine resin (oleoresin) is a complex mixture of resin acids (diterpenes) and other chemicals dissolved in monoterpene solvents (Zinkel et al. 1971, Hodges et al. 1979). Pine resin, which the woodpecker uses to create a barrier against rat snakes, serves as the pine’s primary defense against bark beetle infestation (Wahlenberg 1946, Hodges et al. 1977, Conner et al. 1998a). Red-cockaded Woodpecker cavity trees in eastern Texas, especially active cavity trees, are regularly attacked and killed by southern pine beetles (*Dendroctonus frontalis*) and occasionally by various species of engraver beetles (*Ips* spp.; Conner et al. 1991, Conner and Rudolph 1995, Rudolph and Conner 1995). High resin flow rate and total production (yield), along with a fast crystallization rate if flow rate is high, are known to favorably influence the pine’s ability to physically repel a bark beetle attack (Wahlenberg 1946, Hodges et al. 1979). In addition, Hayes et al. (1994a) suggest that 4-allylanisole, a component of pine oleoresin, may serve to repel bark beetles.

Although the adhesive properties of pine resin alone may serve as the deterrent to rat snakes attempting to climb Red-cockaded Woodpecker cavity trees (Rudolph et al. 1990), repellent properties of the chemical components of pine resin also may serve this purpose (Jackson 1974). The monoterpene limonene is known to be toxic to bark beetles (Tomlin et al. 2000) and may have a similar effect on rat snakes. Thus, similar to their selection of pines for cavity excavation that have high resin yields, thin sapwood, large diameters of heartwood, and decayed heartwood, Red-cockaded Woodpeckers also may use specific resin chemistries as a selection criterion. An additional possibility is that daily excavation at resin wells on active cavity trees induces a wound response in the pine that alters resin chemistry by affecting the concentrations of various resin acid components. Wound response in pines usually involves increased resin flow volume (Ruel et al. 1998), whereas resin chemical composition normally is considered a function of the pine’s genotype (Trapp and Croteau 2001). However, short-term wound response in grand firs (*Abies grandis*) can involve a moderate increase in monoterpene concentration and a slight increase in diterpene concentration (Steele et al. 1998, Trapp and Croteau 2001).

We examined resin chemistry in active Red-cockaded Woodpecker cavity trees in loblolly (*Pinus taeda*) and shortleaf (*P. echinata*) pines in eastern Texas. We sampled resin from (1) pines selected by Red-cockaded Woodpeckers that contained naturally excavated cavities in active cavity trees, (2) pines selected by forest biologists that contained artificially installed cavity inserts and were actively being used by Red-cockaded Woodpeckers, and (3) control pines of similar age and appearance to the active cavity trees. If Red-cockaded Woodpeckers are inducing a change in pine resin chemistry by daily excavation at resin wells, this change should appear in active cavity trees which have been selected by woodpeckers for naturally excavated cavities, as well as in active cavity trees that have been selected by biologists for artificial insert installation, but not in control pines. If Red-cockaded Woodpeckers are selecting pines for cavity trees that have specific resin chemistry, concentrations of some resin components in active cavity trees selected by the woodpeckers for natural cavity excavation should be different from both control pines and pines selected by biologists that contain active cavity inserts.

**METHODS**

We used increment borers to collect xylem wood tissue and the resin it contained from Red-cockaded Woodpecker cavity trees in loblolly and shortleaf pines on shrink-swell soils on the northern portion of the Angelina National Forest (62,423 ha; 31° 15′ N, 94° 15′ W) in eastern Texas. A small subpopulation of Red-cockaded Woodpeckers occurs on this portion of the national forest (Conner and Rudolph 1989). We used woodpecker activity at resin wells, amount of bark scaling, and condition of the cavity entrance as indicators of tree status (see Jackson 1977, 1978b).

We sampled a total of 52 pines (36 loblolly and 16 shortleaf pines) each month during August and September 1997, including 13 active naturally excavated cavity trees, 13 active cavity trees that contained one artificial cavity.
insert, and 26 control pines of similar age and general appearance to the active cavity trees. Two cores (0.5 cm diameter by 3 cm long) of xylem tissue were extracted at breast height from opposite sides of the outermost xylem rings of each pine. Both xylem core samples removed from each pine were immediately placed in foil-sealed scintillation vials in the field that were filled with 10 ml of pentane containing 0.2 mg of the internal standard tetradecane. The lids were securely tightened and the vials placed on ice in insulated coolers for transport back to the laboratory; samples were maintained in explosion-proof freezers until processed.

We analyzed terpenoid components of the resin samples using a Varian 3400 gas chromatograph with flame ionization detector. Components were separated on a J&W Scientific DB-5 column (30 m length × 0.52 mm internal diameter) using a thermal gradient program (initial temperature 70°C maintained for 5 min, increasing to 250°C by 6°C/min and held for 10 min). The injector port was maintained at 260°C and the detector at 280°C; peaks were integrated using Varian ChemStar software on a PC (Varian, Inc. 1995). Samples were prepared for chromatographic analysis using a modified method of Lewinsohn et al. (1993), which allows the simultaneous analysis of mono- and diterpenoids of conifer oleoresin. Approximately 48 h prior to sample preparation, 5 ml of MTBE (methyl tert butyl ether) containing 0.25 mg of tricosanoic acid (internal standard) were added to the pentane-containing sample vials and allowed to further extract at room temperature. The mixture was then evaporated to 2.5 ml under N₂ gas and split into two 1.0-ml aliquots: one processed for monoterpenes and the other for diterpene acids. Diterpenes were esterified using TMS-diazomethane (Aldrich Chemical Co.) because it is commercially available and more stable than lab-generated diazomethane. After processing, the two fractions were recombined, further concentrated to approximately 300 ml, and analyzed. Monoterpene peaks were quantified against tetradecane, and diterpene resin methyl esters were quantified against the methyl ester of tricosanoic acid.

We identified individual peaks by comparison with published peak retention times (Hayes et al. 1994b) under replicated chromatographic conditions and confirmed them by GC-mass spectra produced from a single sample that exhibited all peaks being quantified. Mass spectra were produced using a Varian 3400GC with a 30-m DB-5 column coupled to a Saturn 3 mass detector under chromatographic conditions similar to those used for the quantitative analysis. Total terpenoid, total monoterpene, total diterpene resin acid yields, and the concentration of individual terpenoid components were statistically analyzed using StatView ver. 4.5 (SAS Institute, Inc. 1998). We tested all data for normality and homogeneity of variance. We pooled data across months because no monthly differences were detected in an initial two-way ANOVA design (treatment × month), and tree types (natural cavity, artificial cavity, and control) were compared subsequently with a one-way ANOVA followed by a Tukey’s multiple comparison test if significant differences among means were detected. Under situations where the assumptions of parametric statistical tests were not met, data were log transformed (logₑ+1). A two-way ANOVA design (treatment × tree species) was used to explore possible differences in production of specific resin components between tree species detected a difference only for α-pinene (Table 1).

RESULTS

Seven volatile monoterpene peaks, including α-pinene, β-pinene, limonene, 4-allylani sole, and three unknown smaller peaks were identified and quantified. Five diterpene methyl-ester peaks, including pimaric acid, isopimaric-levopimaric-palustric acids (quantified as a single large unresolved peak due to poor separation on the DB-5 column), dehydroabietic acid, abietic acid, and neoabietic acid also were identified and quantified. We detected no significant differences in total volatile monoterpene or total diterpene acid contents among the three treatments (Table 1). We compared individual peaks across the three treatment levels; likewise, no significant differences were seen for α- or β-pinene, limonene, or 4-allylani sole. However, the large diterpene acid peak containing the isopimaric-levopimaric-palustric methyl-ester mix did show a significant difference among the three treatment levels (Table 1), with levels found within the active natural cavity trees being
TABLE 1. Amounts of monoterpenes and diterpenes (means ± SE, mg/two-core sample) measured from increment cores extracted during August and September 1997 from active naturally excavated and active artificial insert Red-cockaded Woodpecker cavity trees and morphologically similar control pines on the Angelina National Forest in eastern Texas. Red-cockaded Woodpeckers selected pines with higher iso-levo-palustric acid levels. Means for all components except α-pinene were compared with a one-way ANOVA (df = 2, 49).

<table>
<thead>
<tr>
<th>Resin chemicals</th>
<th>Artificial cavity trees (n = 13)</th>
<th>Natural cavity trees (n = 13)</th>
<th>Control pines (n = 26)</th>
<th>F</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Monoterpenes</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>α-pinene*</td>
<td>0.283 ± 0.034</td>
<td>0.373 ± 0.079</td>
<td>0.281 ± 0.026</td>
<td>0.71</td>
<td>0.982*</td>
</tr>
<tr>
<td>β-pinene</td>
<td>0.192 ± 0.021</td>
<td>0.232 ± 0.065</td>
<td>0.204 ± 0.017</td>
<td>0.11</td>
<td>0.900</td>
</tr>
<tr>
<td>Limonene</td>
<td>0.294 ± 0.035</td>
<td>0.244 ± 0.040</td>
<td>0.252 ± 0.028</td>
<td>0.65</td>
<td>0.524</td>
</tr>
<tr>
<td>4-allylanisole</td>
<td>0.501 ± 0.030</td>
<td>0.451 ± 0.052</td>
<td>0.468 ± 0.033</td>
<td>0.54</td>
<td>0.582</td>
</tr>
<tr>
<td>Total monoterpenes</td>
<td>1.390 ± 0.112</td>
<td>1.382 ± 0.230</td>
<td>1.295 ± 0.088</td>
<td>0.27</td>
<td>0.764</td>
</tr>
<tr>
<td><strong>Diterpene resin methyl-esters</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pimaric acid</td>
<td>0.275 ± 0.028</td>
<td>0.305 ± 0.030</td>
<td>0.279 ± 0.021</td>
<td>0.35</td>
<td>0.705</td>
</tr>
<tr>
<td>Iso-levo-palustric acids</td>
<td>1.938 ± 0.114</td>
<td>2.482 ± 0.206</td>
<td>1.992 ± 0.127</td>
<td>3.50</td>
<td>0.034</td>
</tr>
<tr>
<td>Dihydroabietic acid</td>
<td>0.218 ± 0.054</td>
<td>0.227 ± 0.038</td>
<td>0.199 ± 0.020</td>
<td>0.16</td>
<td>0.855</td>
</tr>
<tr>
<td>Abietic acid</td>
<td>0.295 ± 0.036</td>
<td>0.310 ± 0.050</td>
<td>0.388 ± 0.039</td>
<td>2.04</td>
<td>0.136</td>
</tr>
<tr>
<td>Neoabietic acid</td>
<td>0.533 ± 0.046</td>
<td>0.612 ± 0.043</td>
<td>0.615 ± 0.029</td>
<td>1.73</td>
<td>0.183</td>
</tr>
<tr>
<td>Total diterpene methyl-esters</td>
<td>3.259 ± 0.193</td>
<td>3.936 ± 0.278</td>
<td>3.476 ± 0.178</td>
<td>2.14</td>
<td>0.123</td>
</tr>
</tbody>
</table>

* F and P values from two-way ANOVA (treatment × tree species) for α-pinene only; see methods and results.

20% greater than controls (Tukey’s P = 0.043) and 22% greater than trees with artificial cavities (Tukey’s P = 0.066). We detected no difference between trees with artificial cavities and control pines (Tukey’s P > 0.99). There was no significant interaction between treatment and month in the initial two-way ANOVA for the large diterpenic peak. We found no significant differences in any of the terpenoid measures (volatiles, resin acids, or total terpenoid content) between the sampling months (August and September).

Loblolly pines produced significantly more α-pinene than did shortleaf pines (F1,49 = 16.32, P < 0.0001). The two-way ANOVA design (cavity tree type × tree species) detected no significant differences among treatments (cavity types and control) for α-pinene concentrations (Table 1). There was no significant interaction between treatment and tree species (F2,49 = 1.79, P = 0.17). We did not detect a difference between tree species for any of the other resin components.

DISCUSSION

Daily excavation at resin wells coats Red-cockaded Woodpecker cavity trees with fresh pine resin, producing a constant “wick” of resin volatiles that evaporate and diffuse from the trees. Active maintenance of resin wells by Red-cockaded Woodpeckers may induce and maintain elevated chemical defenses in trees. Under this scenario, induction could actually enhance the short term resistance of trees to beetle attack and infestation (defenses are already mobilized and wound response resin is being produced; Ruel et al. 1998), but could lower the long term resistance by depleting carbohydrate stores over time that could otherwise be used to mount an effective defensive response to a future beetle attack. However, in this study, the activity of Red-cockaded Woodpeckers at resin wells did not appear to affect the terpene yield (concentration) or composition of cavity tree resin, and argues against the idea of induced terpene defenses. We found that active cavity trees with artificial cavity inserts had the same terpene yields and composition as the control trees.

In contrast, the proportion of isopimaric-levopimaric-palustric resin acids in the oleoresin of Red-cockaded Woodpecker-selected active cavity trees was greater than that found in either the control or active artificial cavity trees. These data suggest that Red-cockaded Woodpeckers may use biochemical information, in addition to other cavity tree selection factors, as selection criteria for determining suitable cavity trees. The chemical components of the resin in pines selected by woodpeckers may have enhanced properties that maintain resin stickiness, which in turn limits...
the ability of rat snakes to gain access to the nest cavity. Alternatively, woodpecker-selected cavity trees may contain higher concentrations of specific diterpene resin acids that are distasteful or toxic to rat snakes and likewise serve to inhibit rat snake climbing and thereby nest predation. Although diterpenes affect resin viscosity, the complete effect of these diterpene resin acids on resin stickiness and toxicity to rat snakes is poorly understood. Limonene, which is one of the most toxic monoterpenes to bark beetles (Coyne and Lott 1976, Raffa and Berryman 1982, Tomlin et al. 2000), was not present in significantly higher levels in cavity trees selected or used by Red-cockaded Woodpeckers. An additional possibility is that Red-cockaded Woodpeckers use resin characteristics to help detect presence of red heart fungus.

Previous research has suggested that the monoterpenes 4-allylanisole in pine resin serves as a repellent to southern pine beetles (Hayes and Strom 1994, Hayes et al. 1994a). Because loblolly and shortleaf pine Red-cockaded Woodpecker cavity trees are regularly infested and killed by southern pine beetles (Conner et al. 1991, 1998b; Conner and Rudolph 1995), we expected to find lower levels of 4-allylanisole in active cavity trees that receive daily wounding from woodpeckers excavating resin wells than in control pines. This was not the case, suggesting that factors other than 4-allylanisole levels, such as the yield of resin from cavity trees (Hodges et al. 1979, Conner et al. 2001), influence cavity tree vulnerability to southern pine beetles.

In present day forest landscapes it may not be possible to obtain a precise determination of Red-cockaded Woodpecker cavity tree preference. Cavity trees currently used by Red-cockaded Woodpeckers and those selected by biologists for cavity insert installation may not provide the resin characteristics fully preferred by Red-cockaded Woodpeckers because they represent a truncated age distribution and an artifact of what was left after the removal of old growth pine forests in the South between 1880 and 1930 (Conner and O’Halloran 1987). However, resin chemistry is controlled primarily by tree genetics (Trapp and Croteau 2001) and likely is not affected by the truncated pine age distribution present throughout the South.

A more precise separation of resin chemicals within the large diterpene peak we detected and a better comprehension of the physical chemistry and toxicity of each resin component also are needed before a more complete understanding of Red-cockaded Woodpecker cavity tree selection can be achieved. Additional research is needed to evaluate resin chemistry in longleaf pines (Pinus palustris). Longleaf pines appear to be the preferred pine species for Red-cockaded Woodpecker cavity trees (Conner et al. 2001), and are known to produce greater yields of resin than either loblolly or shortleaf pines (Hodges et al. 1977, Conner et al. 1998a). Red-cockaded Woodpecker selection of pines with specific resin chemistries may be more pronounced in longleaf pine than in other pine species.

ACKNOWLEDGMENTS

We thank T. Moon for her assistance with laboratory analysis of the resin samples. We thank R. F. Billings, R. Bowman, F. C. James, J. F. Kelly, P. L. Lorio, and J. R. Walters for constructive comments on an early version of the manuscript. Research on the Red-cockaded Woodpecker was conducted under U.S. Fish and Wildlife Service federal permit TE832201-0 to RNC. The use of trade, equipment, or firm names in this publication is for reader information only and does not imply endorsement by the U.S. Dept. of Agriculture of any product or service.

LITERATURE CITED


HAYES, J. L. AND B. L. STROM. 1994. 4-allylanisole as an inhibitor of bark beetle (Coleoptera: Scolytidae) aggregation. J. Econ. Entomol. 87:1586–1594.


