Seasonal Variation of Urushiol Content in Poison Oak Leaves

Barbara L. Gartner, Christian Wasser, Eloy Rodriguez, and William L. Epstein

Urushiol is the causative agent in poison oak and poison ivy dermatitis. Urushiol concentration in poison oak (*Toxicodendron diversilobum*) leaves was quantified weekly between mid-August and late November for several individual genotypes of plants that had been grown in either sun, moderate shade, or deep shade. These individual plants were grown in the same garden to reduce variation caused by environmental factors. Plants cloned from a coastal genotype had higher levels of urushiol than those from an inland site, suggesting that people who live in coastal areas may be at a greater risk for contracting poison oak dermatitis than people who live farther inland. The concentration of urushiol remained relatively constant in the inland clones until early November, at which point it increased by three to six times. This increase in urushiol may be responsible for the greater number of cases that some dermatologists have noticed in the autumn. The level of urushiol in leaf litter was very low, and so it can be considered nonallergenic.

PLANTS of the genus *Toxicodendron* (formerly known as *Rhus* Anacardiaceae) are notorious as causes of contact dermatitis.[1,2] The dermatitis produced by these plants is caused by the urushiol fraction, which is primarily composed of 3-pentadecylcatechol and three heptadecylcatechol analogs in poison oak.[3,4] Several methods have been developed to lessen the severity of the rashes produced by urushiol[5] but the best precaution is to avoid contact with the plant and the chemicals in it.

There are four species in this genus in North America, but *Toxicodendron diversilobum* and *T radicans* are the most common, and thus cause the most cases of dermatitis. The only unambiguous names for members of this genus are the scientific names because "poison oak" and "poison ivy" are used to indicate the same species in some regions and different species between regions. *T radicans* (often called poison ivy) is the predominant species in the eastern half of the United States, and it is divided into a number of subspecies[6]; *T rydbergii* has a wide range in the middle of the United States, and *T toxicarium* is found in the Southeast (Fig 1).

The subject of this paper, *T diversilobum* (most often referred to as poison oak), is the only species in this genus to live west of the Sierra Nevada (Fig 1). It will be referred to hereafter as poison oak. Poison oak can assume a continuum of growth forms from a short shrub to a 30-meter–tall vine. It can be found in full sun or deep shade, in creek beds, on dry hillsides, on a variety of soils, on windswept coastal bluffs, and in a variety of other habitats: coniferous forest, chaparral, and meadows.[7] The differences between the vine and shrub growth forms have recently been shown to be caused by environmental factors (the presence or absence of physical support) and not heredity.[8]

Many studies have been performed on the variability of the chemical constituents of plants. A cursory examination of recent literature indicates that chemical constituents such as monoterpenes and flavonoids, vary markedly in pine trees from different geographic regions.[9-11] Different plants also have been studied for seasonal and environmental variability in the content of monoterpenes,[12] alkaloids,[13] and furanocoumarins.[14] Studies have established that chemical constituents such as furanocoumarins, can increase after the stress of fungal infections or cold night temperatures.[15]

The comparative urushiol composition and content in different species of *Toxicodendron* have been extensively studied, for example, references 16 and 17, and Baer et al[18] established that the urushiol content of leaves can vary with the position of the leaf on the stem. However, no studies have been reported on the differences in concentration and composition of urushiol of plants with different growth habits. Moreover, no data are available on the seasonal variation in urushiol content of poison oak.

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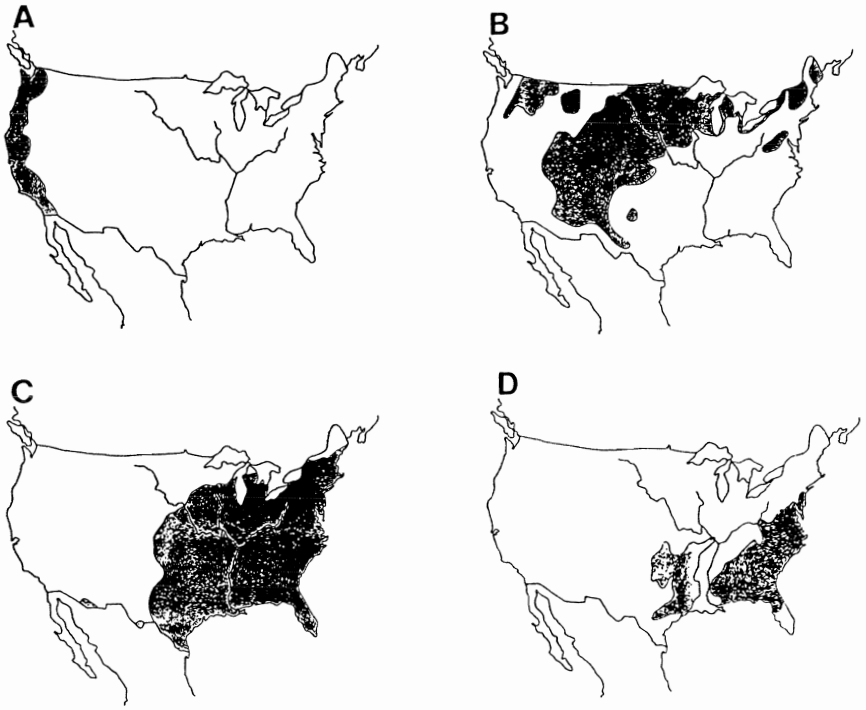

Fig 1. Geographic ranges of the four species of *Toxicodendron* in the United States: (A) *T diversilobum* (Western poison oak); (B) *T rydbergii* (Rydberg's poison ivy); (C) *T radicans* (poison ivy); and (D) *T toxicarium* (Eastern poison oak). Data from Gillis.[6]

MATERIALS AND METHODS

Collection of Samples

Poison oak plants were grown from cuttings taken from three different source plants, a vine (plant A) and a shrub (plant B) from Jasper Ridge Biological Preserve, Woodside, CA, in the coastal range near Stanford University, and a shrub (plant C) from a bluff overlooking Pomponio Beach, CA, 20 km southwest of Jasper Ridge. Three replicate cuttings ("clones") from each source plant were planted into each of three light environments: full sun, 45% sun, and 15% sun.[19] Clones were sampled for this study 1½ years later, to assure that all buds and leaves had developed in the light environment in which the clone, not the source plant, had grown.

On May 14, 1989, samples were collected from all plants to learn the variance among source plants and light treatments for total urushiol and the proportion of the different catechols in the urushiol. Each sample consisted of nine leaves (three from each of the three clones of a given plant). Samples were stored in plastic bags and analyzed within 4 days of collection.

From mid-August 1989 until late November when the majority of leaves had dropped, clones A and B in 15% sun were sampled weekly to determine the seasonal change in total urushiol content of the leaves. Three weeks later, their abscised (dead or fallen) leaves were sampled. We refer to these as "leaf litter."

Analysis of Samples

Poison oak leaves (2.0 g, fresh weight) were extracted with 30 mL of methanol. The extracts were then centrifuged for 5 minutes at 3,000 rpm. Five milliliters of the supernatant was evaporated to dryness, redissolved in 5 mL of dichloromethane, filtered, and evaporated. The extract was then redissolved in 5 mL of high-pressure liquid chromatography (HPLCl)-grade methanol. Each sample was then filtered through a Miniclean Cartridge (C18, Alltech, Deerfield, IL) before injection on the HPLC column. HPLC analysis was performed using a C18 reversed phase cartridge (Alltech, Econosphere C18, 250 × 4.6 mm, 5 mm particle size) and methanol-water (95:5) mixture at 1.2 mL/min with UV detection at 275 nm.

Four different catechol derivatives (Fig 2) were detected, but heptadecylcatechol was present in extremely low concentrations and could not be analyzed accurately. Heptadecenylcate-

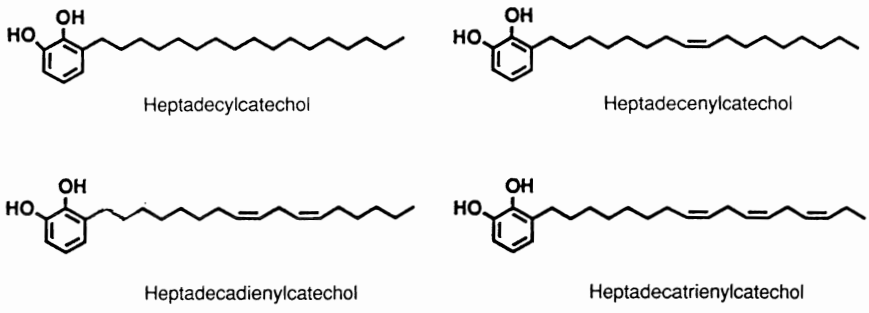

Fig 2. Catechol derivatives of poison oak: heptadecylcatechol, heptadecenylcatechol (HDEC), heptadecadienylcatechol (HDDC), and heptadecatrienylcatechol (HDTC).

chol (HDEC), heptadecadienylcatechol (HDDC), and heptadecatrienylcatechol (HDTC) were present in larger concentrations and were analyzed throughout the experiment. The concentration of catechols in each sample was determined by comparison with a set of external standards of purified urushiol (0.1, 0.2, 0.3, 0.4, and 0.5 mg/mL). The detector response was linear over the range of these standards. Urushiol used for standards was purified according to a procedure described elsewhere.[20]

RESULTS AND DISCUSSION

On May 14, 1989, after leaves had fully expanded but before they had begun to senesce, the foliar concentration of urushiol was greater in clones of plants from the coastal area (plant C) than in the clones from Jasper Ridge (plants A and B, Fig 3; significant at $P < .01$, one-factor analysis of variance [ANOVA]). The individual plants that grew in full sun had slightly more urushiol than the individual plants that grew in shaded areas for all clones. A lower proportion of the urushiol was composed of HDTC in individual plants grown in the full sun rather than in individual plants grown in the shade.

The total foliar urushiol content was also analyzed 17 times between late spring and late fall in the clones from plants A and B (Fig 4). The amount of urushiol remained relatively constant until early November, at which point it increased from the baseline level (about 0.02% to 0.4% by fresh weight) to 0.9% to 2.6% of fresh weight. High content was maintained for 2 weeks, after which the leaves showed low levels of urushiol again. Two

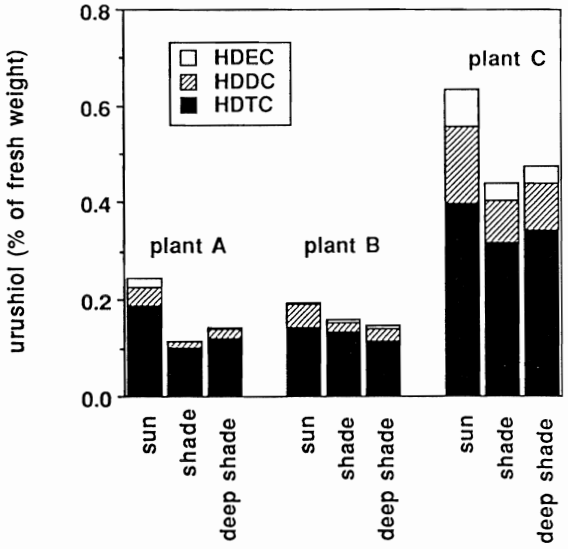

Fig 3. Foliar urushiol content and composition in three clones (from an inland vine [A], and inland shrub [B], and a coastal shrub [C]), in three different light environments.

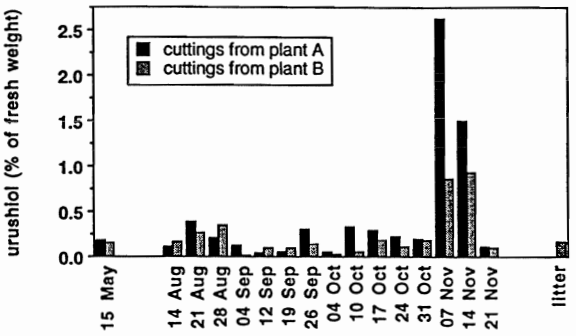

Fig 4. Seasonal variation in foliar urushiol content in clones of two plants growing in deep shade.

weeks after the increase in urushiol, the leaves abscised (dropped). The leaf litter contained a very small quantity of urushiol. These data were collected on a fresh weight basis, and there are many more dead than live leaves per gram; therefore, the low level of urushiol in dead leaves underemphasizes the actual decrease in urushiol concentration that occurs between live and dead leaves. Thus, if urushiol content were expressed per leaf surface area rather than per fresh weight, the contrast between urushiol content of litter and attached leaves would be greater.

These results led us to conclude that urushiol concentration and composition are affected by the light environment of the plant. The greater concentration of urushiols in the individual plants cloned from plant C (the coastal shrub) suggested that a plant's "genotypic origin" or "race" can affect urushiol content. The increase in catechol concentration observed during November occurred as the leaves senesced, shortly before they fell off the plant. This could be a byproduct of the stresses of cold nights, but more likely, the increase is caused by an apparent enrichment in urushiols as other components (carbon, nutrients, and water) are exported from the leaf. Up to 30% of a leaf's carbon and 70% of its nitrogen and phosphorus can be exported from a leaf before senescence,[21] and the leaf can lose water as well; therefore, its fresh weight per unit leaf area would be expected to be much lower in litter than living tissue.

CONCLUSIONS

This study suggests that the environment and the plant's genetic composition (genotype) influence the quantity and composition of urushiols produced in *T diversilobum*. Urushiol is a threat to the health of more than 50% of the North Ameri-

can population,[22] so it is essential to identify when and where the risk of contacting these allergens is highest. As long as the leaves are attached to the plant, their urushiol content is sufficient to cause an allergic reaction in sensitized individuals. However, the leaf litter can be considered benign. Our study suggests that the chances of contracting poison oak dermatitis are higher in coastal areas and in late fall as the leaves begin to senesce. Many dermatologists have noted an increase in poison oak cases in the fall, but its basis had not been investigated previously.

ACKNOWLEDGMENT

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