Herbicide effects of essential oils

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Laboratory and greenhouse experiments were conducted to determine the herbicidal effect of plant-derived oils and to identify the active ingredient in an oil with herbicide activity. Twenty-five different oils were applied to detached leaves of dandelion in the laboratory. Essential oils (1%, v/v) from red thyme, summer savory, cinnamon, and clove were the most phytotoxic and caused electrolyte leakage resulting in cell death. Each of these essential oils in aqueous concentrations from 5 to 10% (v/v) plus two adjuvants (nonionic surfactant and parafﬁnic oil blend at 0.2% [v/v]) were applied to shoots of common lambsquarters, common ragweed, and johnsongrass in the greenhouse; shoot death occurred within 1 h to 1 d after application. Essential oil of cinnamon had high herbicidal activity, and eugenol (2-methoxy-4-[2-(propenyl)]phenol) was determined to be this oil's major component (84%, v/v). Dandelion leaf disk and whole-plant assays verified that eugenol was the active ingredient in the essential oil of cinnamon. Essential oils are extracted from plants and thus may be useful as “natural product herbicides” for organic farming systems.

Nomenclature: Cinnamon, Cinnamomum zeylanicum; clove, Syzygium aromaticum; red thyme, Thymus vulgaris; summer savory, Satureja hortensis; common lambsquarters, Chenopodium album L. CHEAL; common ragweed, Ambrosia artemisiifolia L. AMBEL; dandelion, Taraxacum ofﬁcinale Weber in Wiggers TAROF; johnsongrass, Sorghum halepense (L.) Pers. SORHA.

Key words: Organic weed control, generally regarded as safe (GRAS).

Weeds, a major problem in U.S. agriculture, cause up to 12% loss in crop yield each year and cost nearly $33 billion (Pimentel et al. 2001). Overall, farmers spend $6.1 billion each year to control weeds, and in 1998 the herbicides accounted for 68% of the total U.S. pesticide sales. In the organic systems of agriculture, growers cannot use synthetic herbicides, and weed infestations result in costly crop losses (Gianessi 1990). Weeds were responsible for up to 5% reduction in yield in organically managed corn (Zea mays L.) in California (Clark et al. 1998). In organic cereal fields, yield losses to weeds may be 20% or higher (Rasmussen and Ascard 1995). Mechanical tillage, mulch, and planted ground cover are major approaches for weed control by organic farmers, but these techniques can be costly and may fail to control weeds adequately. Herbicides for organic systems are limited. Research has demonstrated herbicidal effects of some vegetable oils, and formulations of fatty acids are commercially available as herbicides (Cornish et al. 1993; Gauvrit and Cabanne 1993; Harzios 1998).

Preliminary work in our laboratory demonstrated that essential oils have herbicidal activity; complete weed shoot control was achieved with dilute concentrations of red thyme essential oil (data not shown). Essential oils are natural plant products that contain natural flavors and fragrances that provide characteristic odors (Mukhopadhyay 2000). They are likely to break down quickly in the environment and should be acceptable for weed control by organic farmers. Essential oils are classified as “generally regarded as safe” (GRAS) and can inhibit the growth of microorganisms in food (Beuchat 2001; Ismaiel and Pierson 1950b; Wilson et al. 1997). Essential oils also contain allelochemicals that inhibit seed germination and may be used to inhibit weed seed germination (Dudai et al. 1999). The inhibitory activity against seed varied with the species from which the essential oil was extracted (Dudai et al. 1999). Cinnamon and red thyme are sources of essential oils, and aromatic compounds from these plants have inhibited potato (Solanum tuberosum L.) sprout growth, apparently by killing meristematic cells (Vaughn 1991). However, phytotoxicity caused by direct applications of essential oils to higher plants is unknown.

Essential oils may be used as a viable weed control technology in organic farming systems, but basic information on phytotoxicity is required before field experiments are conducted. The objectives of this research were to (1) screen commercially available oils from plants, especially essential oils, for herbicidal activity, (2) determine whole-plant herbicidal efficacy of candidate oils that were identified in the initial screening, and (3) identify the active ingredient in an oil with strong herbicidal activity.

Materials and Methods

Dandelion Leaf Disk Assay

Essential oils from red thyme, summer savory, cinnamon, clove, basil (Ocimum basilicum L.), fennel (Foeniculum vulgare Miller), and sweet birch (Betula nigra L.) were purchased. Other plant oils were provided or purchased from commercial sources, including corn oil; sesame (Sesamum indicum L.) seed, jojoba (Simmondsia chinensis (Link) C. Schneider), flax (Linum usitatissimum L.) seed, and hazelnut ( Corylus avellana L.) oils; olive (Olea europaea L.) oil; canola (Brassica napus L.) oil; soybean [Glycine max (L.) Merr.] and sunflower (Helianthus annuus L.) oils; peanut (Arachis hypogaea L.) oil; grape (Vitis amurensis Rupr.) seed, caraway (Carum carvi L.), cranberry (Vaccinium macrocarpon Ait.), plum (Prunus sp.) seed, and...
meadowfoam (*Limnanthes alba* Hartweg ex Benth.) oils, cotton (*Gossypium hirsutum* L.) seed oil, and safflower (*Carthamus tinctorius* L.) oil.

The concentration range for studying oil activity was determined in a preliminary experiment by placing leaf disks of dandelion in a series of concentrations of cinnamon oil ranging from 0.001 to 10.0% (v/v). Electolyte leakage, as measured by conductivity, increased at a cinnamon oil concentration of 1.0% (8.6 ± 1.6% leakage). All oils were tested for activity from 0.5 to 2.0% oil concentration. In subsequent experiments, four concentrations of each oil (0.5, 1.0, and 2.0%, v/v) were prepared in mineral oil. An untreated water control was also used. In July and August, uniform leaves of dandelion were collected from plants in the field near Kearneysville, WV, placed on ice in a cooler, and brought into the lab for use within 1 h. Leaves were dipped in the oil treatment and placed in a glass jar containing water to maintain high humidity for 1 h. Leaf disks (1-cm diameter) were cut from each leaf, placed in vials containing 5 ml deionized water, and allowed to equilibrate for 18 h with shaking (300 rpm). A visual estimate of injury to leaf disks was taken at this time. Injury ratings included three categories: 0 indicated little or no injury (necrosis and water-soaked appearance); + indicated injury to nearly half the leaf disk; and ++ indicated severe injury to most of the leaf disk. Electrical conductivity of the solution containing the leaf disk was measured, after which leaf disks were autoclaved for 1 h, and leaf disk solutions were equilibrated with shaking for 4 h before the conductivity was measured again. Leaf injury caused by treatments was calculated for each leaf disk with conductivity before autoclaving (electrolyte leakage caused by treatment) divided by the conductivity after autoclaving (maximum electrolyte leakage of a killed leaf disk). Leaf injury caused by treatment was expressed as a percentage of maximum conductivity. Each treatment was replicated five times in a completely randomized design, and the experiment was repeated.

### Whole-Plant Assay with Common Lambsquarters, Common Ragweed, and Johnsongrass

Plants were grown from seed during September through March in 0.6-L pots with soilless media in a greenhouse near Kearneysville, WV. Plants were grown with supplemental light to maintain a 12-h photoperiod (average midday light intensity, 1.045 μmol m⁻² s⁻¹; photon flux density; 22 ± 3°C). One common ragweed, two common lambquarters, and three johnsongrass plants per pot were grown for 3 mo, and plants were treated when they were approximately 25 to 30 cm tall. These weeds were chosen to represent grass and broadleaved weeds found in fruit orchards.

Essential oils of red thyme, summer savory, cinnamon, and clove were diluted in water to concentrations of 0, 1.0, 5.0, and 10.0% (v/v), with 0.1% concentrations of combinations of nonionic surfactant and paraffinic oil blend, and were applied to each weed species with a handheld sprayer to the point of drip. Adjuvants used in the treatments were selected on the basis of results of a preliminary field experiment in which the essential oil of red thyme was diluted in water to produce concentrations of 0, 1.0, 5.0, and 10% (v/v), with 0.2% concentrations of different adjuvants, and was applied (100 ml m⁻²) with a handheld sprayer to a variety of weeds. A nonionic surfactant and a paraffinic oil blend appeared to improve weed control and were used for greenhouse experiments. Weed injury in the greenhouse was evaluated as the average visual estimates taken by two people 7 and 30 d after treatment. Injury ratings were on a scale from 0 (no injury) to 100 (dead plant). Each treatment was replicated five times in a factorial design, and the experiment was repeated. Effects of essential oil and rate were statistically analyzed with the general linear model procedure (SAS 1988).

### Active Ingredient Determination

Cinnamon oil had strong herbicidal activity in the dandelion leaf disk and whole-plant assays; therefore, the active ingredient is...
ingredient in cinnamon oil was determined. Cinnamon oil (1 μl) was injected in a gas chromatograph equipped with a 30-m nonpolar column, and individual components were determined with a mass selective detector. Helium was the carrier gas, and run conditions included 250 C (injector); 315 C (detector); 50 C for 5 min, thermal gradient of 3 C per minute to 250 C, then 250 C for 10 min before returning to the initial conditions (column). Only four components occurred at concentrations greater than 2%, and their identities were verified with standards. These four components were further analyzed for herbicidal activity with the dandelion leaf disk and whole-plant assays described previously.

Results and Discussion

Dandelion Leaf Disk Assay

Essential oils of red thyme, summer savory, cinnamon, and clove injured dandelion leaf disks at concentrations of 1 and 2% (Table 1). Dandelion leaf injury was less for essential oils of basil and sweet birch than for those of red thyme, summer savory, cinnamon, and clove. No other plant oil caused leaf injury. On the basis of these results, the most active essential oils (red thyme, summer savory, cinnamon, and clove) were applied in greenhouse experiments.

Essential oils have reduced sprout growth in potatoes, possibly by killing meristematic cells (Vaughn 1991). Essential oils have also controlled harmful insects and harmful microbes (Farag et al. 1989). Yet, essential oils contain antioxidants that suppress lipid oxidation (Farag et al. 1989). Ismaiel and Pierson (1990a) investigated the mechanism of the inhibition of Clostridium botulinum by essential oils. They found no effect on DNA, RNA, or protein synthesis and suggested that essential oil altered membrane permeability. Based on the electrical conductivity measurements of the electrolyte leakage, the current work indicates that essential oils increase membrane permeability in leaf disks of dandelion. However, the mechanism of injury caused by essential oils remains to be determined.

Whole-Plant Assay with Common Lambsquarters, Common Ragweed, and Johnsongrass

Weeds showed signs of injury within 1 h of treatment, and maximum injury occurred within 24 h of application of the essential oil (data not shown). Rapid plant injury occurs with pelargonic acid (nonanoic acid) (Hatzius 1998), and active components in essential oils apparently caused rapid ion leakage and similar plant injury in this experiment. Treatment effects of the essential oils, the concentrations, and the interaction of oil and concentration were significant (P > F = 0.01).

Johnsongrass (Figure 1), common lambsquarters (Figure 2), and common ragweed (Figure 3) were injured at 1% concentrations of all oils, and most weeds were killed at 5 and 10% concentrations of all essential oils 7 d after treatment. Injury response of johnsongrass to essential oil concentration followed an exponential pattern, whereas injury response of common lambsquarters and common ragweed followed a sigmoidal pattern, indicating that johnsongrass was sensitive to low concentrations of essential oils. However, small portions of johnsongrass leaves were not killed.
Figure 2. Common lambsquarters (Chenopodium album L.) injury response to essential oils of (A) cinnamon (Cinnamomum zeylanicum), (B) clove (Syzygium aromaticum), (C) summer savory (Satureja hortensis), and (D) red thyme (Thymus vulgaris) 7 d after treatment.

\[ Y = \frac{101.2}{1+e^{-(x-2.4)/0.72}} \]
\[ r^2 = 0.99 \]

\[ Y = \frac{100.5}{1+e^{-(x-2.4)/0.83}} \]
\[ r^2 = 0.99 \]

\[ Y = \frac{99.0}{1+e^{-(x-3.6)/1.4}} \]
\[ r^2 = 0.96 \]

Figure 3. Common ragweed (Ambrosia artemisiifolia L.) injury response to essential oils of (A) cinnamon (Cinnamomum zeylanicum), (B) clove (Syzygium aromaticum), (C) summer savory (Satureja hortensis), and (D) red thyme (Thymus vulgaris) 7 d after treatment.

\[ Y = \frac{100.5}{1+e^{-(x-2.3)/0.73}} \]
\[ r^2 = 1.00 \]

\[ Y = \frac{100.7}{1+e^{-(x-2.30)/1.74}} \]
\[ r^2 = 1.00 \]

\[ Y = \frac{99.4}{1+e^{-(x-2.7)/0.73}} \]
\[ r^2 = 1.00 \]

\[ Y = \frac{100.0}{1+e^{-(x-2.9)/0.69}} \]
\[ r^2 = 1.00 \]
even at the 10% concentration. It is likely that leaf segments without injury were not exposed to essential oils and that more complete coverage would have increased plant injury. Thirty days after treatment, injury ratings had not changed compared with those at 7 d after treatment (data not shown). However, small regrowth from untreated buds was observed in some common ragweed plants. Complete coverage was necessary to injure whole plants, and another application of oil would likely be necessary for continued weed control or plant kill.

Whole-plant injury was related to changes in essential oil rates between 1 and 10%, but the greatest change in injury was between 1 and 5% (Figures 1–3). Greater injury of weeds at low rates may be obtained with adjuvants that increase dispersal and plant coverage with the essential oils. Phytotoxicity at low rates was demonstrated in the dandelion leaf disk experiments where high electrolyte leakage was observed at essential oil concentrations of 2% (Table 1). Cinnamon and clove oils caused the greatest injury to johnsongrass and were among the most active essential oils that injured common lambsquarters and common ragweed. Red thyme and summer savory caused less injury to johnsongrass than did cinnamon and clove oils.

**Active Ingredient Determination**

Twelve components were identified in cinnamon oil, but most of them constituted less than 1% of the oil (data not shown). Only four components occurred in concentrations greater than 2%: eugenol, benzyl benzoate (benzoic acid phenylmethyl ester), humulene (2,6,6,9-tetramethyl-1,4,8-cycloundecatriene), and isoeugenol (2-methoxy-4-propenylphenol) with retention times of 27.5, 43.0, 29.4, and 34.1 min, respectively, and in concentrations of 84.2, 3.9, 3.5, and 2.4%, respectively. In the dandelion leaf disk assay eugenol and isoeugenol had herbicidal activity at 1 and 2% concentrations (Figure 4), but benzyl benzoate and humulene were not phytotoxic at any concentration (data not shown). In the whole-plant assay with common lambsquarters, common ragweed, and johnsongrass, the four major components of cinnamon oil injured or killed plants when applied at concentrations of 5% (Figure 5), but no injury to plants was observed when it was applied at concentrations of 0.2%.

In the whole-plant assay, 10% concentrations of cinnamon oil were phytotoxic (Figure 5). Eugenol concentration would be approximately 8% at this concentration of cinnamon oil, sufficient to cause plant injury. Benzyl benzoate, humulene, and isoeugenol concentrations would be approximately 0.4, 0.4, and 0.2%, respectively, in a 10% concentration of cinnamon oil, too low to cause plant injury. The active ingredient for herbicidal activity in cinnamon oil thus appears to be eugenol.

Eugenol is a volatile phenol, and this class of compounds may be a valuable source of natural herbicides (Balandrin et al. 1985; Mukhopadhyay 2000). Synthetic phenolic herbicides were used as contact herbicides that caused rapid membrane disruption or uncoupling of oxidative phosphorylation (Anderson 1983). The use of the phenolic herbicide dinesob was cancelled because of the adverse health effects (Anonymous 1998). However, dinesob has a dose lethal to 50% of the test organisms (LD50) of 58 mg kg⁻¹ (rat). In contrast, eugenol is a skin irritant with an oral LD50 > 2,650 mg kg⁻¹ (rat) and is used as a dental antiseptic for analgesic purposes.

Future work should discover the herbicidal activity of other essential oils and their components. Active compounds may be synthesized by cell culture suspensions to produce natural essential oils (Balandrin et al. 1985) or by flavor chemists to produce synthetic natural-identical molecules (Mukhopadhyay 2000). We are currently investigating the activity of several eugenol derivatives. In addition, the practical use of active compounds must be explored. Our preliminary work indicated that eugenol was effective for broadleaved and grass weed control in the field. We are currently investigating formulations that may increase eugenol efficacy and may permit the reduction of the concentration of eugenol in field applications.

Essential oils have herbicidal activity, and nearly complete weed shoot control was achieved with cinnamon and clove essential oils at concentrations of 5 to 10%. Eugenol was the active ingredient in cinnamon oil, and field experiments are being conducted to evaluate eugenol as a contact herbicide beneath fruit trees. Essential oils are extracted from plants and thus may be useful as natural product herbicides for organic farming systems. Additional work is planned to determine the active ingredient(s) and to establish the mechanism of herbicidal action with other essential oils. It is possible that essential oils could be used with other tools to manage weeds. For example, a component of essential oil

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could be used to control weeds that grow through mulch or to injure and predispose weeds to control with biocontrol agents.

**Sources of Materials**

1. Essential oils from red thyme, summer savory, cinnamon, clove, basil, fennel, and sweet birch. Frontier Natural Products, P.O. Box 299, Norway, IA 52318.
5. Canola oil, GFA Brands, Inc., P.O. Box 512, Cresskill, NJ 07626.
6. Soybean and sunflower oils, Beatrice/Hunt-Wesson, Inc., P.O. Box 4800, Fullerton, CA 97634.
7. Peanut oil, C.F. Sauer Co., 200 West Broad Street, Richmond, VA 23220.
8. Grape seed, caraway, cranberry, plum seed, and meadowfoam oils, Badger Oil Co., 1400 South River Street, Spooner, WI 54801.
9. Cotton seed oil, Sigma Chemical Co., P.O. Box 14508, St. Louis, MO 63178.
12. Conductivity meter, Y S I Scientific, 1725 Brannum Lane, Yellow Springs, OH 45387.
13. Seed of common lambsquarters, common ragweed, and johnsongrass, Valley Seed Service, 2172 North Pleasant, Fresno, CA 93705.
14. Metro Mix 510, Scotts-Sierra Horticultural Products Co., 14111 Scottslawn Road, Marysville, OH 43041.
16. Paraffinic oil blend, Superior dormant spray oil, Drexel Chemical Co., 1700 Channel Avenue, Memphis, TN 38113.
18. Hewlett Packard 5890 Series 2, Hewlett Packard, 2850 Cen-
terville Road, Wilmington, DE 19808.
19. DB-5, J & W Scientific, 91 Blue Ravine Road, Folsom, CA 95630.

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Hewlett Packard 5971, Hewlett Packard, 2850 Centerville Road, Wilmington, DE 19808.

21 Eugenol, benzyl benzoate, humulene, isoeugenol, Fluka Chemical Corp., 1001 West St. Paul Avenue, Milwaukee, WI 53233.

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Literature Cited


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