

## Essential Oil of *Dyssodia acerosa* DC.

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Samples of the suffrutescent *Dyssodia acerosa* DC. were collected from the Jornada Experimental Range in south-central New Mexico, and GC/MS and retention indices were used to identify 62 components from the steam-distilled oil. Chrysanthenone (34.8%), limonene (14.5%), camphor (12.3%),  $\alpha$ -pinene (6.8%), sabinene (4.3%), camphene (3.9%), (*E*)- $\beta$ -ocimene (2.5%), 1,8-cineole (2.2%), myrcene (2.0%), and *cis*-sabinene hydrate (1.6%) were the major components of the steam-distilled oil.

**Keywords:** *Dyssodia acerosa*; Asteraceae; essential oil composition; chrysanthenone; limonene; camphor

### INTRODUCTION

The study of diet selection by livestock in arid rangelands has been a long-term research focus at the U.S. Department of Agriculture, Agricultural Research Service's Jornada Experimental Range (JER). Recently, we have begun to examine plant–herbivore interactions at the biochemical level (Estell et al., 1994; Fredrickson et al., 1994; King et al., 1996). In an effort to further understand these interactions, we are investigating the essential oil (which likely contains volatiles present at the plant–animal interface) composition of a number of unpalatable shrubs. *Dyssodia acerosa* (prickleleaf dogweed, Hierba de San Nicolas and Contrayerba; typical height of 0.1–0.25 m) is a suffrutescent found mostly on loose limestone soils of the Mojave Desert and northern regions of the Chihuahua and Sonora Deserts and in adjacent grasslands and woodlands at 1000–1800 m elevation in the United States (Benson and Darrow, 1981; Allred, 1988) and extending south to Hidalgo and Zacatecas, Mexico (Powell, 1988). *D. acerosa* has also been placed in the genus *Thymophylla* (K. W. Allred, personal communication). This half-shrub is considered a range pest (Kearney and Peebles, 1951) and is quite unpalatable to livestock (E. H. Bomberger, JER, unpublished data).

Information on the chemical makeup of *D. acerosa* and related species is very limited. Thiophenes and acetylenic and polyacetylenic thiophenes have been isolated from and identified in several species of *Dyssodia* (Bohlmann and Zdero, 1976; Downum and Towers, 1983; Downum et al., 1985) including *D. acerosa* (Bohlmann et al., 1976). Monoterpene ketones have been reported in *Dyssodia decipiens* (Bohlmann and Zdero, 1979). To our knowledge, the oil composition of *D. acerosa* has not been previously reported. In addition to potential involvement in diet selection, these natural products may have alternative uses.

### EXPERIMENTAL PROCEDURES

Samples of *D. acerosa* DC. [identified using Allred (1988)] were collected on the JER from a bajada slope of the San Andres Mountains at an elevation of 1608 m on a ridgetop with loamy-skeletal, carbonatic, thermic, shallow, Typic Pa-

leorthis soils (Tencee series) formed on an old alluvial fan (Neher and Bailey, 1976; Gibbens et al., 1993). Long-term annual precipitation (1922–1990) at the nearest rain gauge (1.7 km from the collection site; elevation of 1585 m) is 257 mm, with 55% of the precipitation occurring in July, August, and September (Gibbens et al., 1993). Total precipitation between October 1995 and September 1996 was 215 mm (73 mm in July, 20 mm in August, and 60 mm in September). Whole plants at the flowering stage were collected in late August, placed in labeled plastic bags, and immediately frozen using dry ice. The plant material was stored at  $-20^{\circ}\text{C}$  until steam distillation was performed. A voucher specimen of *D. acerosa* DC. was placed in the JER herbarium located in Las Cruces, NM.

Steam distillation was conducted in a Nickerson–Likens type apparatus (Nickerson and Likens, 1966; Maarse and Kepner, 1970). A 300 mL three-neck flask, 150 mL of water, and 11.2 g of whole plant material (a composite of three plants) were used. The distillate was continuously extracted during an 8 h distillation with 12 mL of pentane (bp  $35\text{--}36^{\circ}\text{C}$ ) into a 10 mL pear-shaped flask heated with a water bath maintained at  $\sim 62^{\circ}\text{C}$ . The two pentane fractions, and an additional 8 mL of pentane used to rinse the apparatus, were combined and dried over anhydrous magnesium sulfate overnight and filtered. Analyses were performed directly on this solution (before addition of standards) to determine if naturally occurring plant components coeluted with the octane and eicosane standards. Octane and eicosane standards were used only for determination of retention times and not for quantitation purposes: a 100  $\mu\text{L}$  aliquot was diluted with 100  $\mu\text{L}$  of pentane, 100  $\mu\text{L}$  of octane in pentane (1000  $\mu\text{g}/\text{mL}$ ) and 100  $\mu\text{L}$  of eicosane in pentane (1000  $\mu\text{g}/\text{mL}$ ) at a final concentration of 250  $\mu\text{g}/\text{mL}$  of each alkane before injection. Subsequently, the solvent was removed from the remaining filtrate under reduced pressure at  $1^{\circ}\text{C}$  using a rotary evaporator. A clear yellow oil was obtained in a yield of 113 mg (1.01% of fresh weight).

Analyses were performed by gas chromatography coupled to mass spectrometry (GC/MS), using a Finnigan ion trap mass spectrometer (EI, 70 eV) in conjunction with a Varian Model 3400 gas chromatograph equipped with a DB-5 column (30 m  $\times$  0.25 mm fused silica capillary column, film thickness 0.25  $\mu\text{m}$ ) using helium as carrier gas (1 mL/min), 1  $\mu\text{L}$  injection size, and a programmed (injector temperature,  $220^{\circ}\text{C}$ ; transfer line temperature,  $240^{\circ}\text{C}$ ; initial column temperature,  $60^{\circ}\text{C}$ ; final column temperature,  $240^{\circ}\text{C}$ ,  $3^{\circ}\text{C}/\text{min}$ ) temperature run (Adams, 1995). Identifications of oil components were performed by a comparison of mass spectra with literature data (NIST, 1990; Adams, 1995) and by a comparison of their relative retention times with those of authentic compounds or by comparison of their retention indices with those in the literature (Adams, 1995). The relative amounts (RA) of

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**Table 1. Constituents of the Oil of *D. acerosa***

compound	RI <sup>a</sup>	RT <sup>b</sup>	% RA <sup>c</sup>
hexanal	798	167	t <sup>d</sup>
( <i>E</i> )-2-hexenal	854	211	t
( <i>Z</i> )-3-hexenol	857	216	t
1-hexanol	869	226	t
tricyclene	926	298	0.2
$\alpha$ -thujene	930	304	1.2
$\alpha$ -pinene	938	315	6.8
camphene	953	338	3.9
sabinene	976	377	4.3
$\beta$ -pinene	979	383	1.2
myrcene	991	405	2.1
$\alpha$ -phellandrene	1004	431	1.1
$\delta$ -3-carene	1011	443	t
$\alpha$ -terpinene	1017	454	0.2
<i>p</i> -cymene	1025	469	0.3
limonene	1030	481	14.5
1,8-cineole	1033	484	2.2
( <i>Z</i> )- $\beta$ -ocimene	1040	496	0.1
phenylacetaldehyde	1043	504	t
( <i>E</i> )- $\beta$ -ocimene	1051	518	2.5
$\gamma$ -terpinene	1061	541	0.7
<i>cis</i> -sabinene hydrate	1069	559	1.6
terpinolene	1088	608	0.2
<i>trans</i> -sabinene hydrate	1096	628	0.3
linalool	1097	633	0.1
isophorone	1118	678	0.1
chrysanthenone	1123	696	34.8
unknown <sup>e</sup>	1140	729	0.1
camphor	1144	740	12.3
unknown <sup>f</sup>	1157	771	t
<i>cis</i> -chrysanthenol	1163	785	0.7
borneol	1165	792	0.4
terpinen-4-ol	1177	822	1.0
<i>p</i> -cymen-8-ol	1184	840	t
$\alpha$ -terpineol	1189	855	0.5
<i>cis</i> -piperitol	1194	868	t
verbenone	1205	899	0.1
<i>trans</i> -carveol	1217	927	0.1
<i>cis</i> -carveol	1230	958	t
carvone	1243	990	t
piperitone	1253	1016	t
carvacrol	1299	1146	t
eugenol	1356	1286	0.3
$\alpha$ -copaene	1376	1339	t
$\beta$ -cubebene	1389	1376	t
methylugenol	1402	1409	0.1
$\beta$ -caryophyllene	1419	1448	0.1
$\alpha$ -humulene	1454	1533	t
germacrene D	1481	1603	0.8
bicyclogermacrene	1495	1641	0.3
$\gamma$ -cadinene	1513	1684	t
$\delta$ -cadinene	1523	1707	t
eugenyl acetate	1525	1712	0.1
spathulenol	1576	1835	t
caryophyllene oxide	1582	1848	t
<i>epi</i> - $\alpha$ -cadinol	1640	1984	0.1
$\alpha$ -muurolol	1646	1996	t
$\alpha$ -cadinol	1653	2014	0.1
eicosane	1999	2744	t
heneicosane	2099	2934	t
1-docosane	2194	3106	0.2
<i>n</i> -docosane	2201	3118	t
tricosane	2302	3294	t
tetracosane	2403	3462	t

<sup>a</sup> RI, retention index as determined on a DB-5 column using the homologous series of *n*-hydrocarbons (Kovats index). <sup>b</sup> RT, retention time on a DB-5 column in seconds. <sup>c</sup> RA, relative area (peak area relative to total peak area). <sup>d</sup> t, trace ( $\leq 0.05\%$ ). <sup>e</sup> Either *trans*-*p*-menth-2-en-1-ol or *trans*-pinene hydrate. <sup>f</sup> Either  $\beta$ -pinene oxide or isopulegol.

individual components of the oil are expressed as percent peak area relative to total peak area.

## RESULTS AND DISCUSSION

Table 1 shows the identity, retention index, retention time, and percent composition of the oil of *D. acerosa*. Sixty-two compounds were identified in the oil of prickleleaf dogweed, accounting for >95% of the com-

position of the oil, and only 1 unidentified compound (RT = 641) accounted for >0.2% of the total area. Among the identified compounds were 15 monoterpene hydrocarbons (39.3%), 8 sesquiterpene hydrocarbons (1.3%), 18 oxygenated monoterpenes (54.2%), 5 oxygenated sesquiterpenes (0.2%), and 6 long-chain hydrocarbons (0.2%). The unknown at RT = 641 accounted for 1.3% of the total area and, on the basis of its mass spectral pattern [150 (1, M<sup>+</sup>), 135 (2), 121 (40), 107 (82), 105 (20), 91 (53), 80 (98), 79 (100)], appears to be a monooxygenated monoterpene, possibly structurally related to verbenone or chrysanthenone. The absence of thiophene derivatives (Bohlmann et al., 1976) in our distillate would indicate that this method is not suited for the isolation of such compounds from *D. acerosa*.

The oil of *D. acerosa* has a very pleasant smell strongly reminiscent of the *Artemisia*- and *Chrysothamnus*-dominated plant communities common to western North America. The dominant compound, chrysanthenone, has been reported in a patent to be an effective component in toothpaste for the removal of tobacco stains (Inoue et al., 1987). Chrysanthenone is also known to be a photoinduced rearrangement isomer of the allelochemical verbenone (Hurst and Whitman, 1960; Kostyk et al., 1993), which was present in the oil in low concentrations. Whether verbenone, a well-known beetle antiaggregant (Gijzen et al., 1993; Kostyk et al., 1993), is the precursor for chrysanthenone in *D. acerosa* is not clear. Although chrysanthenone has been shown to be biologically inactive as an antiaggregant for one species of pine-beetle (Kostyk et al., 1993), several of the main oil constituents of *D. acerosa*, including limonene,  $\alpha$ -pinene, borneol, terpinolene (Elliott and Loudon, 1987), camphor (Sinclair et al., 1988), and myrcene (Bucyanayandi et al., 1990), which account for >36% of the total oil content of *D. acerosa*, have been negatively correlated with herbivory.

In conclusion, *D. acerosa* has a moderate oil content and a high proportion of monoterpenes in its profile. The dominance of chrysanthenone and the presence of verbenone in the profile make *D. acerosa* a good candidate for further study in terms of its allelochemical properties, and the agreeable fragrance of the oil makes it of possible interest to the perfume and flavor industry.

## LITERATURE CITED

- Adams, R. P. *Identification of Essential Oil Components by Gas Chromatography/Mass Spectroscopy*; Allured Publishing: Carol Stream, IL, 1995.
- Allred, K. W. *A Field Guide to the Flora of the Jornada Plain*; New Mexico Agricultural Experiment Station Bulletin 739; New Mexico Agricultural Experiment Station: University Park, NM, 1988; p 44.
- Benson, L.; Darrow, R. A. *Trees and Shrubs of the Southwestern Deserts*, 3rd ed.; The University of Arizona: Tucson, AZ, 1981; pp 310–311.
- Bohlmann, F.; Zdero, C. Über Inhaltsstoffe der Gattung *Dyssodia*. *Chem. Ber.* **1976**, *109*, 901–905.
- Bohlmann, F.; Zdero, C. Über Dimere Terpenketone aus *Tagetes gracilis*. *Phytochemistry* **1979**, *18*, 341–343.
- Bohlmann, F.; Zdero, C.; Grenz, M. Inhaltsstoffe Einiger Gattungen der Tribus Helenieae und Senecioneae. *Phytochemistry* **1976**, *15*, 1309–1310.
- Bucyanayandi, J.-D.; Bergeron, J.-M.; Menard, H. Preference of Meadow Voles (*Microtus pennsylvanicus*) for Conifer Seedlings: Chemical Components and Nutritional Quality of Bark of Damaged and Undamaged Trees. *J. Chem. Ecol.* **1990**, *16*, 2569–2579.
- Downum, K. R.; Towers, G. H. N. Analysis of Thiophenes in the Tageteae (Asteraceae) by HPLC. *J. Nat. Prod.* **1983**, *46*, 98–103.

- Downum, K. R.; Keil, D. J.; Rodriguez, E. Distribution of Acetylenic Thiophenes in the Pectidinae. *Biochem. Syst. Ecol.* **1985**, *13*, 109–113.
- Elliott, S.; Loudon, A. Effects of Monoterpene Odors on Food Selections by Red Deer Calves (*Cervus elaphus*). *J. Chem. Ecol.* **1987**, *13*, 1343–1349.
- Estell, R. E.; Fredrickson, E. L.; Anderson, D. M.; Mueller, W. F.; Remmenga, M. D. Relationship of Tarbush Leaf Surface Secondary Chemistry to Livestock Herbivory. *J. Range Manage.* **1994**, *47*, 424–428.
- Fredrickson, E. J.; Thilsted, J.; Estell, R.; Havstad, K. Effect of Chronic Ingestion of Tarbush (*Flourensia cernua*) on Ewe Lambs. *Vet. Hum. Toxicol.* **1994**, *36*, 409–415.
- Gibbens, R. P.; Havstad, K. M.; Billheimer, D. D.; Herbel, C. H. Creosote Vegetation After 50 Years of Lagomorph Exclusion. *Oecologia* **1993**, *94*, 210–217.
- Gijzen, M.; Lewinsohn, E.; Savage, T. J.; Croteau, R. B. Conifer Monoterpenes, Biochemistry and Bark Beetle Ecology. In *Bioactive Volatile Compounds from Plants*; Teranishi, R., Buttery, R. G., Sugisawa, H., Eds.; ACS Symposium Series 525; American Chemical Society: Washington, DC, 1993; pp 8–22.
- Hurst, J. J.; Whitman, G. H. The Photochemistry of Verbenone. *J. Chem. Soc.* **1960**, *21*, 2864–2869.
- Inoue, A.; Naganuma, T.; Hozumi, S. Dentifrices Containing Monoterpenes for Removing Tobacco Stains from Teeth. *Jpn. Kokai Tokkyo Koho JP 62,181,212* [87,181,212] (Cl. A61k7/16), Aug 8, 1987; *Chem. Abstr.* **1987**, *107*, 204969k.
- Kearney, T. H.; Peebles, R. H. *Arizona Flora*; University of California: Berkeley and Los Angeles, CA, 1951; pp 932–933.
- King, D. W.; Estell, R. E.; Fredrickson, E. L.; Havstad, K. M.; Wallace, J. D.; Murray, L. W. Effects of *Flourensia cernua* Ingestion on Intake, Digesta Kinetics, and Ruminal Fermentation of Sheep Consuming Tobosa. *J. Range Manage.* **1996**, *49*, 325–330.
- Kostyk, B. C.; Borden, J. H.; Gries, G. Photoisomerization of Antiaggregation Pheromone Verbenone: Biological and Practical Implications with Respect to the Mountain Pine Beetle, *Dendroctonus ponderosae* Hopkins (Coleoptera: Scolytidae). *J. Chem. Ecol.* **1993**, *19*, 1749–1759.
- Maarse, H.; Kepner, R. E. Changes in Composition of Volatile Terpenes in Douglas Fir Needles During Maturation. *J. Agric. Food Chem.* **1970**, *18* (6), 1095–1101.
- Neher, R. E.; Bailey, O. F. *Soil Survey of White Sands Missile Range, New Mexico*; USDA Soil Conservation Service, U.S. Department of the Army, and New Mexico Agricultural Experiment Station: University Park, NM, 1976.
- Nickerson, G. B.; Likens, S. T. Gas Chromatographic Evidence for the Occurrence of Hop Oil Components in Beer. *J. Chromatogr.* **1966**, *21*, 1–5.
- NIST MS Library, Finnigan, Standard Format. National Institute of Science and Technology, Gaithersburg, MD, 1990.
- Powell, A. M. *Trees and Shrubs of Trans-Pecos Texas*; Big Bend Natural History Association: Big Bend National Park, TX, 1988; pp 472–473.
- Sinclair, A. R. E.; Jogia, M. K.; Andersen, R. J. Camphor from Juvenile White Spruce as an Antifeedant for Snowshoe Hares. *J. Chem. Ecol.* **1988**, *14*, 1505–1514.

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