

from those observed in field cases. The details concerning the comparative pathology of poisoning by cocklebur seedlings and authentic carboxyatractyloside in swine will be reported elsewhere (B. P. Stuart, R. J. Cole, and S. H. Gosser, unpublished data).

Carboxyatractyloside appeared to be located primarily in the cotyledon of the plant, and the concentration of carboxyatractyloside diminished rapidly after germination of the seed and the disappearance of the cotyledons. Metabolic activity in the cotyledon was directly associated with a concomitant disappearance of carboxyatractyloside and loss of toxicity. This rapid decrease in toxicity as the plant grows older no doubt has been the source of confusion by early investigators about the toxicity or lack of toxicity of the cocklebur plant.

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## Chemistry of Toxic Range Plants. Volatile Constituents of Broomweed (*Gutierrezia sarothrae*)

The major mono- and sesquiterpenes in the essential oil of *Gutierrezia sarothrae* have been identified by gas chromatography-mass spectrometry as  $\alpha$ -pinene, myrcene, linalool, *cis*-verbenol, *trans*-verbenol, verbenone, geraniol, caryophyllene, and  $\gamma$ -humulene.

Broomweed, turpentineweed, or snakeweed (*Gutierrezia sarothrae*), a perennial resinous shrub widespread on dry rangeland from Texas to California and northward to Idaho, has been responsible for major economic losses due to abortion and death among range animals (Kingsbury, 1964). Cattle appear to be particularly susceptible to the abortifacient effects of broomweed, although sheep and goats are also affected to a lesser extent. Common features of broomweed-induced abortion in cattle are production of weak and underweight calves which frequently fail to survive, retained placenta, and hemorrhage. The plants appear to be most toxic in the early growth stages and are more toxic when growing on impoverished, sandy soils than on richer soils.

Intravenous injection of a saponin isolated from broomweed into pregnant rabbits, goats, and cows induced abortion and produced symptoms similar to those observed in cows poisoned by consuming the plant on the range. The saponin was shown to be abortifacient when administered orally to pregnant rabbits. However, both a saponin from lechuguilla (*Agave lecheguilla*), known to be toxic, and a supposedly nontoxic, commercial pharmaceutical-grade saponin produced abortion and/or death when administered intravenously to pregnant rabbits (Dollahite et al., 1962; Shaver et al., 1964).

Since certain terpenes have been reported to stimulate the central nervous system and may also cause menorrhagic abortion (von Oettingen, 1963), knowledge of the composition of the volatile fraction of broomweed could provide information for comparison of the volatile terpene

constituents with those of other abortifacient range plants.

The composition of the steam-volatile essential oil obtained from broomweed is reported herein.

#### EXPERIMENTAL PROCEDURES

Leaves and small stems of *Gutierrezia sarothrae* (Pursh.) Britton et Rusby were collected from young plants in the preflower stage on April 5, 1979, near Eunice, NM. The material was air-dried and ground to pass through a 1-mm screen.

A sample of the ground plant material (100 g) in deionized, odor-free water (500 mL) was extracted with heptane (50 mL) containing Ionox 330 in a Likens-Nickerson apparatus held at 100-mm pressure (~50 °C water temperature) for 8 h. The heptane extract was concentrated by distillation of the solvent through a Vigreux column, the concentrate weighed, and the percentage of heptane remaining determined by gas chromatography. The concentrated extract was then transferred to a small pear-shaped flask and the residual heptane distilled off through an air condenser.

Gas chromatography (GC) was carried out by using a Hewlett-Packard 5830 equipped with a 30 m  $\times$  0.25 mm i.d. glass SP2100 methyl silicone column and flame ionization detector. The column was programmed from 60 to 250 °C at 4 °C/min. Samples were injected with a split ratio of 1:50 at a linear velocity of 23.5 cm/s. Identification of components was based upon GC-MS data obtained with a quadrupole-type mass spectrometer (Electronic Associates Quad 300 mass filter; Finnigan Corp. 3000-1B

Table I. Constituents, Composition, and GC Retention Times of *G. sarothrae* Volatiles

compd	composition, %	retention time, min
$\alpha$ -pinene <sup>a</sup>	1.3	8.01
myrcene <sup>a</sup>	1.4	9.75
linalool <sup>a</sup>	1.4	13.75
<i>cis</i> -verbenol <sup>a</sup>	1.7	15.26
<i>trans</i> -verbenol <sup>a,b</sup>	6.0	15.55
C <sub>10</sub> H <sub>16</sub> O	1.7	16.35
C <sub>10</sub> H <sub>18</sub> O	1.5	17.03
verbenone <sup>a</sup>	4.2	17.55
C <sub>10</sub> H <sub>14</sub> O	2.1	18.79
geraniol <sup>a</sup>	53.8	19.77
C <sub>10</sub> H <sub>16</sub> O	1.0	23.52
caryophyllene <sup>a</sup>	1.0	25.22
C <sub>15</sub> H <sub>24</sub>	2.9	26.80
$\gamma$ -humulene <sup>b</sup>	12.2	27.15
C <sub>15</sub> H <sub>24</sub>	2.1	27.67
C <sub>15</sub> H <sub>24</sub>	2.8	28.23
unidentified	1.8	29.87
unidentified	1.2	30.67

<sup>a</sup> Identified by mass spectra and GLC retention time.

<sup>b</sup> Identified by NMR and IR spectra.

electronics) connected to a 500 ft  $\times$  0.03 in. i.d. stainless steel SF96(50)/Igepal column by a silicon rubber membrane type separator. Preparative GC was performed by using a 12 ft  $\times$  0.25 in. i.d. glass column packed with 10% OV-17 on 80–100 mesh Chromosorb W-HP.

Nuclear magnetic resonance (NMR) spectra were obtained by using a Varian EM-390 spectrometer. Infrared (IR) spectra were recorded on a Perkin-Elmer 727B spectrophotometer. Individual constituents were identified by comparison of GC retention times and mass spectra with reference compounds or by preparative GC and determination of the NMR and IR spectra.

## RESULTS

The yield of volatile constituents from dry plant material was 0.32%. The essential oil contained 18 components comprising 1% or more of the total. Nine of these components, comprising 83% of the total, were identified unequivocally. The components, percentage composition, and retention times are presented in Table I.

*trans*-Verbenol was identified by preparative GC and comparison of the NMR spectrum with that reported by Cooper et al. (1967).  $\gamma$ -Humulene was similarly identified by preparative GC and comparison of the NMR and IR spectra with literature data (Bohlmann et al., 1974; Yano and Nishijima, 1974).

## DISCUSSION

A significant similarity among several abortifacient plants is their highly resinous nature. Thus, broomweed, ponderosa pine, Monterey cypress, and juniper all contain considerable quantities of essential oil and have been shown to produce abortions under range conditions, while the essential oils of tansy (*Tanacetum vulgare*) and pennyroyal (*Mentha pulegium*) have historically been used to induce abortion in humans. Moreover, the symptoms observed in abortion produced by consumption of broomweed and ponderosa pine needles are remarkably similar. Analysis and comparison of the essential oil composition of these plants may therefore provide information regarding the nature of the abortifacient agent.

Plants of the genus *Gutierrezia* have not previously been examined with regard to composition of the essential oil. The aerial parts of *Gutierrezia lucida* and *Gutierrezia*

*mandonii* have been shown to contain diterpene acids, and the roots of these species and of *G. sarothrae* to contain triterpenes. The presence of germacrene D in *G. mandonii* was also demonstrated (Bohlmann et al., 1979). A novel chloroditerpene, gutierolide, has been isolated from *Gutierrezia dracunculoides* (Cruse et al., 1971).

The essential oil of *G. sarothrae* is characterized by high levels of monoterpene alcohols (>63%) with geraniol (53.8%) predominating. In contrast with ponderosa pine needle oil, which consists mainly of monoterpene hydrocarbons (Zavarin et al., 1971), the level of the latter in broomweed is less than 3%. In addition, broomweed contains fairly high levels of verbenone (>4%). It is interesting to note that this ketone is synthesized from  $\alpha$ -pinene by female bark beetles, as a male-repellent pheromone, during attack on ponderosa pine (Brand et al., 1976). The role of verbenone as an insect deterrent in *G. sarothrae* is hypothetical.

The sesquiterpene hydrocarbons are dominated by  $\gamma$ -humulene, comprising 12.2% of the total essential oil. The presence of germacrene D, previously detected in *G. mandonii* by Bohlmann et al. (1979), could not be demonstrated in *G. sarothrae* by examination of the mass spectra of the unidentified sesquiterpene hydrocarbons.

In view of the considerable qualitative and quantitative differences in the mono- and sesquiterpene composition of *G. sarothrae* and *Pinus ponderosa*, it appears unlikely that the remarkable similarity in abortifacient symptoms induced by these two plants can be attributed to the essential oil fraction.

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