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WHOLE-PLANT OILS FROM TWO *EUPHORBIA* SPECIES GROWING IN SARDINIA

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Key Word Index—*Euphorbia dendroides*; *E. cupanii*; whole plant oils; fatty acids; hydrocarbons; sterols.

Abstract—Chemical compositions of acetone extracts from *Euphorbia dendroides* and *E. cupanii* are reported. Both plants grow wild in Sardinia and represent a balance between territorial distribution and size. The extracts are interesting as possible future sources of chemicals.

INTRODUCTION

Whole-plant oils extracted with organic solvents from several plant species and in particular from *Euphorbiae* have been considered in the past as possible alternative fuel sources to crude-oil products [1] and also as sources of a wide variety of chemicals [2, 3].

Previously we have reported the results obtained with organic solvent extracts from *E. lathyris* and *E. characias* [4, 5]. We now wish to report the results of an analogous investigation on *E. dendroides* and *E. cupanii*. The four aforementioned species of *Euphorbia* represent a convenient balance between territorial distribution and plant size among the 27 species growing wild on the island of Sardinia [6]; furthermore, it should be noted that *E. cupanii* is endemic to this island.

RESULTS AND DISCUSSION

The extraction of vegetable material was carried out following the experimental scheme proposed in ref. [7] and successfully employed by us in previous work [5]. The yield of raw organic materials (based on dry wt) obtained by extraction with acetone from *E. dendroides* and *E. cupanii* are reported in Table 1; the amount of extract was high in the case of *E. cupanii* reaching 14.3%, comparable with that found for *E. lathyris* and *E. characias* [4]. A solid phase separated during the extraction process; it was filtered off and the IR spectra showed characteristic peaks of polyphenolic compounds.

Table 1. Extractives and residues from two *Euphorbia* species

	Acetone extract (wt%)	Cyclohexane extract (wt%)	Residue (wt%)
E. dendroides	11.8 (1.9*)	0.2	88.0
E. cupanii	14.3 (1.2*)	0.1	85.6

*Material which separated from acetone solution during extraction process.

Only very small amounts of hydrocarbons (0.1–0.2%) were obtained from the residues by extraction with cyclohexane after the acetone treatment of both plants (Table 1).

The primary products of the acetone extraction were partitioned between hexane and 95% ethanol in order to separate the oil from the polyphenol fraction; Table 2 reports the results obtained in this separation process and in the elementary analyses of the oil fractions. *E. dendroides* produced only a small amount of oil (4.1%), this value representing one of the lowest oil contents found in extraction processes of *Euphorbia* species [8]. In this species the polyphenol fraction is more abundant than the oil fraction and the composition of the acetone extracts is atypical in comparison with the other *Euphorbia* species investigated. This polyphenolic material, which is a very complex mixture of aromatic compounds, was not further analysed.

Saponification of the oil fractions from *E. dendroides* and *E. cupanii* afforded a considerable amount of unsaponifiable matter (58 and 54%, respectively). This is the most interesting material in the oil as a source of fine chemicals, in addition to the fatty acids (40 and 44%, respectively). In Table 3 the distribution of the fatty acids determined by GC analysis in the saponifiable matter of both oils is shown.

Both oils showed a similar qualitative and quantitative composition of the fatty acid mixture. Myristic acid is four

Table 2. Hexane–ethanol (95%) partitioning of *Euphorbia* extractives.

	E. dendroides	*E. cupanii*
Oil fraction (wt%)	4.1	8.5
Polyphenol fraction (wt %)	5.8	4.6
Elemental analyses of oil fraction		
C (wt %)	79.39	73.81
H (wt %)	11.53	10.90
O (wt %)	9.08	15.29

Table 3. Relative proportions of fatty acids contained in the oils extracted from *Euphorbia* species

Fatty acid	*E. dendroides*		*E. cupanii*		Standard
	(R_f)	(wt %)	(R_f)	(wt %)	(R_f)
12:0	3.5	1.9	3.4	0.4	—
14:0	5.6	12.3	5.6	3.4	5.7
15:0	6.8	0.5	6.6	2.8	—
16:0	8.0	32.0	8.0	28.5	8.0
16:1	—	—	8.7	0.8	—
18:2	9.7	37.5	9.8	45.3	9.8
18:1	10.1	4.2	10.1	4.5	—
18:0	—	—	—	—	10.2
20:0	11.7	1.5	—	—	—
18:3	12.1	2.5	12.1	1.6	—
22:0	13.9	3.5	13.9	2.8	—
24:0	15.6	1.7	15.6	5.9	15.8
Others	—	2.5	—	4.0	—

times more abundant in the oil of *E. dendroides* than in that of *E. cupanii*. The unsaponifiable matter of the two plants after purification by CC was qualitatively analysed by TLC and compared with an unsaponifiable fraction from *E. lathyris* previously analysed. The following four classes of components were revealed: sterols, triterpene alcohols, saturated and unsaturated hydrocarbons and traces of tocopherols. Qualitative separation and determination of these fractions, accomplished by standard methods [9], gave the results reported in Table 4. It is noteworthy that the overall amount of the sterol fraction (12% of oil extracted) found for *E. cupanii* is one of highest reported for oils derived from *Euphorbia* speices [10].

The distribution of components in the isolated and purified linear hydrocarbon fractions for the two *Euphorbia* species, determined by GC analysis, is shown in Table 4: the predominant components for both plants were C_{27}, C_{29} and C_{31}. Substantial differences were found in the alcohol fractions of the two plants. (i) The fraction from *E. dendroides* showed five main components, namely cycloartenol (33%), 24-methylenecycloartanol (27%), lanosterol (12%), α-amyrin and butyrrospermol (8%), besides many other unidentified compounds present in minor amounts, whereas in the same fraction from *E. cupanii* only 24-methylenecycloartanol and cycloartenol were found in significant (13 and 40%, respectively). (ii) Fatty alcohols, such as C_{28}, C_{30} and C_{32} were detected only in the fraction derived from *E. cupanii* (23%). (iii) An unidentified triterpenic alcohol with an R_f lower than that of lanosterol comprised *ca* 8% of the fraction from *E. dendroides*.

Finally, the main components of the sterol fractions from both *Euphorbia* species were identified as β-sitosterol (46% and 25% in *E. dendroides* and *E. cupanii* respectively) and Δ_7-stigmasterol (18 and 39% in *E. dendroides* and *E. cupanii*, respectively). A small quantity (2%) of stigmasterol was found in both fractions.

On the basis of these results it is now possible to obtain some comparative data on the properties of the four most interesting Sardinian *Euphorbia*. In Tables 5 and 6 the results concerning the extraction of these four plants are collected. It is interesting to note that *E. cupanii*, in spite of a rather high content of polyphenolic material, shows the highest content of oil. This species represents also the most favourable quantitative balance between the classes of components in the oil fractions. Particularly significant is the high content of sterols, which accounts for 1.5% of the dried vegetable feedstock (Table 7).

EXPERIMENTAL

Homogeneous samples of starting materials were obtained from wild *E. dendroides* and *E. cupanii* plants collected in north west Sardinia, by drying at 105° for 4 hr, grinding and then sieving to pass a 80 mesh. A portion of this material was extd in a Soxlet apparatus with Me_2CO for 100 hr. The Me_2CO soln was evapd and the residue purified as described elsewhere [5].

The refined oils were saponified by treatment with 2 N KOH in EtOH and the unsaponifiable material extd from the alkaline soln with petrol (40–60°). The fatty acids obtained after acidification were converted into Me esters by treatment with an Et_2O soln of CH_2N_2. GC analysis of the Me ester mixt was carried out

Table 4. Composition of the purified unsaponifiable fraction from two *Euphorbia* species*

	Hydrocarbons (wt %)	Alcohols (wt %)	Sterols (wt %)	Other compounds (wt %)
E. dendroides	6.4	60.0	13.9	19.6
E. cupanii	2.4	55.2	22.4	20.0

*Determined by TLC and GC.

Table 5. Composition of main hydrocarbons found in two *Euphorbia* species

Hydrocarbons*	*E. dendroides*	*E. cupanii*
C_{16}	0.54	3.05
C_{20}	0.72	1.03
C_{21}	1.03	2.53
C_{22}	1.65	0.84
C_{23}	3.22	2.50
C_{24}	3.10	0.87
C_{25}	6.61	4.79
C_{26}	5.26	6.01
C_{27}	7.80	10.25
C_{28}	3.47	4.39
C_{29}	7.64	8.40
C_{30}	1.64	0.64
C_{31}	7.64	11.10

*Wt % of fraction; hydrocarbons occurring in very small amounts (<0.5%) are not listed.

on a 2 m × 2 mm using 6 ft column packed with 3% SE 30 on Chromosorb W under the following conditions: 100° for 1 min and prog. at 8°/min up to 260° for 2 min.

The unsaponifiable material, after purification by CC on Al_2O_3[5], was fractionated by prep. TLC using Merck 20 × 20 cm plates with a 2 mm thick coating of silica gel G. Five fractions were obtained (satd and unsatd hydrocarbons, triterpene alcohols and two different sterol fractions) from the plate by scraping and extraction with hot $CHCl_3$. GC analyses of the sepd fractions were performed, under the following conditions: *Hydrocarbons.* 70° for 1 min and then prog. at 16°/min up to 300° for 15 min. *Triterpene alcohols.* 100° for 1 min and then prog. at 32°/min up to 260° for 16 min. *Sterols* (as TMS ethers). 120° for 1 min and then prog. at 4°/min up to 290° for 4 min.

Preliminary IR analyses of polyphenolic fractions showed groups of bands characteristic of phenolic OH groups (3000–3500 cm^{-1}) and of a 4H-pyran-4-one sytem (1600 cm^{-1} and 1200–1450 cm^{-1}) [9].

Table 6. Extractive from four Sardinian *Euphorbia* species

	Acetone extract*	Oil fraction*	Polyphenol fraction*
E. lathyris†	11.5	8.1	3.4
E. characias†	13.5	7.5	6.0
E. dendroides	11.8	4.1	7.7
E. cupanii	14.3	8.5	5.7

*Wt % on the dried plants.
†From ref. [5].

Table 7. Composition and yield of whole-plant extract oils from four Sardinian *Euphorbia* species

	Euphorbia			
Components	*lathyris*	*characias*	*dendroides*	*cupanii*
Fatty acids	30.0	27.1	40.0	48.0
Hydrocarbons	10.6	10.8	3.7	1.5
Sterols				
Lanosterol	11.0	—	4.4	—
α-Amyrin	8.2	18.3	2.9	—
Cycloartenol	8.5	12.8	11.5	13.2
24-Methylenecycloartanol	10.8	8.9	9.4	4.3
Campesterol	0.2	—	0.2	—
Stigmasterol	0.2	—	0.1	0.3
β-Sitosterol	2.5	3.3	3.7	3.3
Δ_7-Stigmasterol	0.1	—	1.5	5.1
Others*	17.9	18.8	22.6	24.3
Yield (% dry wt)	8.1	7.5	4.1	8.5

*Other compounds are glycerol, fatty alcohols and trace amounts of tocopherols.

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