

COMPOSITION OF THE ESSENTIAL OIL IN *THYMUS OROSPEDANUS*

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Aromatic plants, particularly the various species of the genus *Thymus*, are frequently used in folk remedies throughout southern Spain. Some of the pharmacological effects attributed to *Thymus* species have been demonstrated in controlled laboratory studies (1-5). Several such studies have been carried out in our laboratory (6-9).

No other studies have appeared to date on *Thymus orospedanus* H. del Villar, a species endemic in southern Spain (10). The authors have investigated the pharmacodynamic properties of its essential oil. Findings included potent antimicrobial, spasmolytic, and central nervous system stimulating activities (11), which led us to believe it would be worthwhile to carry out a more detailed analysis of the composition of the essential oil.

The essential oil portion in *T. orospedanus* reached 1.75% of dry weight, a relatively high figure in comparison to most species of the genus *Thymus*, where essential oil content generally varies from 0.58% to 2.5% (12). The oil exhibited the following physical properties:  $d_{4}^{15}=0.8951$ ;  $n_{D}^{15}=1.4864$ ;  $[\alpha]_{D}^{20}=-7.3^{\circ}$ . One gram of oil was soluble in 0.5 ml 90% EtOH.

TABLE 1. Composition of the Essential Oil of *Thymus orospedanus*

Component	Mode of Identification		Relative Concentration (%)
<b>Hydrocarbons</b>			
$\alpha$ -pinene . . . . .	2.45 <sup>a</sup>	ms	3.8
$\alpha$ -thujene . . . . .		ms	
camphene . . . . .	3.02	ms	3.3
$\beta$ -pinene . . . . .	4.17		tr <sup>b</sup>
$\Delta$ -3-carene <sup>c</sup> . . . . .	5.48	ms	
myrcene <sup>c</sup> . . . . .	5.89	ms	4.9
phellandrene <sup>c</sup> . . . . .	5.89	ms	
$\beta$ -ocimene . . . . .		ms	
limonene . . . . .	6.68		0.8
$\gamma$ -terpinene . . . . .	7.01	ms	22.4
<i>p</i> -cymene . . . . .	7.69	ms	22.5
terpinolene . . . . .		ms	
caryophyllene <sup>d</sup> . . . . .	12.51	ms	4.1
$\delta$ -cadinene . . . . .		ms	
$\alpha$ -humulene . . . . .		ms	
<b>Alcohols</b>			
linalool . . . . .	12.06		5.4
terpinen-4-ol . . . . .	13.15		0.8
borneol . . . . .	14.12	ms	5.0
<b>Acetates</b>			
bornyl acetate . . . . .	12.92		0.9
carvacryl acetate . . . . .		ms	
<b>Ketones</b>			
thujone . . . . .	10.04		tr
camphor <sup>d</sup> . . . . .	12.51	ms	4.1
<b>Aromatic alcohols</b>			
thymol . . . . .	24.60	ms	7.3
carvacrol . . . . .	25.96	ms	15.6

<sup>a</sup>Retention time (min);<sup>b</sup>trace—when below 0.5%;<sup>c,d</sup>Compounds not resolved for individual quantitation.

The qualitative and quantitative data obtained in the analysis of the essential oil are reported in Table 1. The analysis revealed that the hydrocarbons were the most prevalent chemical group of all components identified. The predominant components of the *T. orospedanus* essential oil were *p*-cymene,  $\gamma$ -terpinene, carvacrol, thymol, linalool, and borneol. The aromatic alcohols and their biosynthetic precursors. ( $\gamma$ -terpinene and *p*-cymene) represent practically 70% of the total content in essential oil.

On the basis of the results obtained from analyses carried out in our laboratories as well as from studies published by others on the essential oils of various species of *Thymus*, these can be classified in general terms into two main groups. The first group contains those in which aromatic alcohols and/or their biosynthetic precursors are clearly the predominant component, as in *T. zygis* (13), *T. capitatus* (14-16), *T. serpyllum* (17), *T. serpylloides* (18), *T. fominii* (19), *T. transcasicus* (19), *T. migricus* (20), and *T. rariflorus* (21). In the essential oils of other members of this genus, however, aromatic ring-containing components are scarce (less than 10%) or altogether lacking, whereas other components traditionally considered to be scarce in thyme species dominate. Examples of this second group include *T. granatensis*, whose major components are myrcene (19%) and carophyllene (14%) (7), *T. eriphorus* (22) and *T. praecox* (23, 24), in which linalool is the major component (12% and 27%, respectively), and *T. trautvetteri* whose major component is geraniol (11%) (25). 1,8-Cineole is the most commonly detected compound in the essential oil of *T. membranaceus* (31%), *T. funkii* (51%), and *T. antoninae* (41%) (18).

These two groups notwithstanding, it should be pointed out that certain species in the genus *Thymus* show a chemical polymorphism controlled by genetic factors (12). Hence, Granger (26) has demonstrated that the diversity in the composition of the *T. vulgaris* essential oil reflects six distinct chemotypes, each characterized by one of the following different major components: geraniol, linalool,  $\alpha$ -terpineol, thujan-4-ol and terpin-4-ene, thymol, and carvacrol. Subsequently, various chemotypes were shown to exist within several other species, including *T. nitens* (27), *T. berba-barona* (28), *T. hymalis* (29), and *T. mastichina* (30).

In regards to the essential oil of *T. orospedanus*, the total amount of aromatic alcohols detected was not particularly great (23%), and carvacrol was found to be clearly more abundant than thymol (16% and 17%, respectively). Hence, we consider its essential oil to belong to the first group, representing aromatic alcohol rich thymes. Although neither thymol nor carvacrol were exceptionally abundant, considerable levels of both were found along with the other aromatic alcohols mentioned above or their proposed precursors, i.e.,  $\gamma$ -terpinene and *p*-cymene (31,32)

#### EXPERIMENTAL

The sample material consisted of flowering apices of *T. orospedanus* collected in the Sierra Elvira mountains, in the province of Granada, Spain, during the month of July 1984. Plant identity was confirmed in the Department of Botany, School of Pharmacy (Granada, Spain). The specimens are currently deposited in the Department of Pharmacognosy, School of Pharmacy, Granada, Spain. The essential oil was steam distilled with a Clevenger device during a total time of 5 h.

Gc analyses were performed using a Perkin-Elmer gas chromatograph (model F-11), equipped with a flame ionization detector and a Hewlett-Packard computing integrator (model 3380A). The column packing was 3% DEGS on Chromosorb W (Perkin-Elmer), 5m $\times$ 2mm i.d. Operating conditions were as follows: column temperature, 50 $^{\circ}$  (5 min), 110 $^{\circ}$  (3 min), 150 $^{\circ}$  (5 min), and 170 $^{\circ}$  (rest), with a gradient of 30 $^{\circ}$ /min; injection block temperature, 250 $^{\circ}$ ; detector temperature, 250 $^{\circ}$ . The carrier gas was N<sub>2</sub> at a flow rate of 18 ml/min.

Gc-ms analyses were done on a Hewlett-Packard mass spectrometer (model 5970-B), combined with a Hewlett-Packard gas chromatograph (model 5890), with a Hewlett-Packard methyl-silicone coated capillary column, 12m $\times$ 0.25 mm i.d. Split injection was used throughout the analyses; the split ratio was 100:1. Other conditions of the instrument were as follows: initial column temperature, 100 $^{\circ}$ ; final column temperature, 250 $^{\circ}$  (initial time 2 min, rate 7 $^{\circ}$ /min, and final time, 10 min). Injection block temperature, 250 $^{\circ}$ ; detector temperature, 250 $^{\circ}$ . The carrier gas was He at a flow rate of 2 ml/min.

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