IDENTIFICATION OF ANTIINFLAMMATORY AGENTS FROM *SIDERITIS* SPECIES GROWING IN SPAIN

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Infusions and decoctions of the aerial parts of *Sideritis* species (Lamiaceae) have been traditionally used in Spain for their digestive and antirheumatic properties (1). The antiinflammatory and antiarthritic activities of borjatriol (7*S*, 14*R*, 15-trihydroxy-8α-13-epoxy-labdane) and of hypolaetin-8-*O*-β-D-glucoside (5,7,8,3′,4′-pentahydroxyflavone-8-*O*-β-D-glucoside) isolated from *Sideritis mugronenis* Borja have been demonstrated (2-4) as well as the inhibitory effect on lipoxygenase (5) of sideritoflavone (5,3′,4′-trihyd-

TABLE 1. Antiinflammatory Compounds in Spanish *Sideritis* Species

Species	Places of Collection[a]	B	SF	HG[b]
S. mugronensis Borja	Bonete (Albacete)	+	+++	++
S. funkiana Willk.	Pozo Lorente (Albacete)	+	++	++
S. angustifolia Lag.	Ayora (Valencia)	−	+++	+++
S. tragoriganum Lag.	Torreblanca (Castellón)	−	+++	++
S. saetabensis Rouy	Xàtiva (Valencia)	−	+++	++
S. reverchonii Willk.	Ronda (Málaga)	−	+++	++
S. leucantha Cav. *leucantha*	Santomera (Murcia)	−	+++	+
S. leucantha Cav. var. *bourgeana* (Boiss & Reuter) F.Q.	Pozocañada (Albacete)	−	+++	+
S. leucantha Cav. var. *incana* (Willk.) F.Q.	Caravaca (Murcia)	−	+++	+
S. pusilla (Lange) Pau	Adra (Almería)	−	+++	+
S. flavovirens Rouy	Puerto Lumbreras (Murcia)	−	+++	+
S. linearifolia Lam.	Zaida (Zaragoza)	−	+++	+
S. javalambrensis Pau	Sierra Javalambre (Teruel)	−	+++	+
S. foetens Clemente ex Lag.	Sierra de Gádor (Almería)	−	+++	+
S. osteoxyla (Pau) Rivas Goday & Gómez	Gata (Almería)	−	++	+
S. granatensis (Pau) Rivas Goday & Gómez	Nerja (Málaga)	−	++	+
S. spinulosa Barnadés ex Asso	Zaida (Zaragoza)	−	+	t
S. arborescens Salzm. ex Bentham subsp. *paulii* (Pau) P.W. Ball ex Heywood	Relumbrar (Albacete)	−	+	t
S. scordioides L. subsp. *cavanillesii* (Lag.) P.W. Ball ex Heywood	Enguera (Valencia)	−	+	t
S. serrata Cav. ex. Lag.	Tobarra (Albacete)	−	+	t
S. hirsuta L. *hirsuta*	Gallocanta (Teruel)	−	t	−
S. hirsuta L. subsp. *iberica* Socorro	Serranía de Cuenca (Cuenca)	−	+	t
S. hirsuta L. var *laxespicata* (Degen & Debeaux) F.Q.	Sierra de Segura (Jaén)	−	+	t
S. glacialis Boiss.	Sierra Nevada (Granada)	−	t	t
S. incana L. *incana*	Alto del Hornillo (Albacete)	−	−	t
S. incana L. subsp. *sericea* (Pers.) P.W. Ball ex Heywood	Bicorp (Valencia)	−	−	t
S. incana L. var. *intermedia* F.Q.	Sierra de Almansa (Albacete)	−	−	t
S. glauca Cav.	Sierra de Orihuela (Alicante)	−	−	t
S. hyssopifolia L.	Valle de Arán (Lérida)	−	−	t

[a]Town or Place (Province).

[b]B=borjatriol (+=present, −=absent); SF=sideritoflavone and HG=hypolaetin-8-*O*-β-D-glucoside (Concentration %: +++=over 30, ++=10-30, +=up to 10, t=trace. Percentage of SF is related to the total flavonoid content in $CHCl_3$ extract.)

roxy-6,7,8-trimethoxyflavone) isolated from several *Sideritis* species (6). In the present study, 29 *Sideritis* taxa growing wild in Spain were surveyed by means of hplc and tlc techniques for the presence of these compounds.

$CHCl_3$ extracts were analyzed for borjatriol and sideritoflavone. Bojartriol was detected only in *S. mugronensis* and *S. funkiana*, whereas sideritoflavone was the principal flavone in the greater part of species studied (Table 1). Butanolic extracts were analyzed for hypolaetin-8-*O*-β-D-glucoside. This substance was the most abundant in *S. angustifolia* (Table 1). *S. mugronensis, S. tragoriganum, S. funkiana, S. saetabensis,* and *S. reverchonii,* which are taxonomically closely related to *S. angustifolia*, contained this glycoside in rather large amounts (Table 1).

The results suggest that *S. angustifolia* and related species are the most interesting ones as a source of the studied antiinflammatory substances and show that distribution of these compounds can be used for systematic purposes within this genus (7). From the ethnopharmacological point of view, we have found that the most frequently used species are, indeed, those containing the highest levels of active principles.

EXPERIMENTAL

PLANT MATERIAL.—Aerial parts of the *Sideritis* taxa were collected (sites listed in Table 1) at flowering, and voucher specimens were deposited in the herbaria of the Faculty of Biology, University of Murcia, and the Faculty of Pharmacy, University of Valencia. The species listed in Table 1 were authenticated by Dr. J.B. Peris (Department of Botany, Valencia) and Dr. D. Rivera (Department of Botany, Murcia).

ANALYSIS.—Air-dried material was extracted first with $CHCl_3$ in a Soxhlet apparatus and then macerated with EtOH-H_2O (7:3). The $CHCl_3$ extract was retained for analysis, and the alcoholic extract was concentrated under reduced pressure to remove the EtOH; the remaining aqueous fraction was then extracted three times with *n*-BuOH. Borjatriol was identified by comparison against an authentic marker, on Si gel with toluene-HOAc (4:1) and revealed with Munier and Macheboeuf reagent [KI/basic $Bi(NO_3)_3$ in HOAc]. Sideritoflavone was identified by tlc on Si gel (8) and quantified by hplc on a C-18 reverse phase column as described previously (9). Hypolaetin-8-*O*-β-D-glucoside was identified and quantified by hplc on a C-8 (5μm) reverse phase column. Analyses were carried out with a Perkin-Elmer liquid chromatograph with a 25 cm×4.6 mm i.d. column. Runs were made for 20 min. The elution solvents were H_2O-HCOOH (19:1) (pump B) and MeCN (pump A), at a flow rate of 1.5 ml/min with pump A providing 15% MeCN increasing 1%/min; detection at 340 nm.

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LITERATURE CITED

1. P. Font Quer, "Plantas Medicinales," Labor, Barcelona, 1980, pp. 661-663.
2. A. Villar, R. Salom, and M.J. Alcaraz, *Planta Med.*, 90 (1984).
3. A. Villar, M.A. Gascó, M.J. Alcaraz, S. Máñez, and D. Cortes, *Planta Med.*, 70 (1985).
4. A. Villar, M.A. Gascó, and M.J. Alcaraz, *J. Pharm. Pharmacol.*, **36**, 820 (1984).
5. T. Yoshimoto, M. Furukawa, S. Yamamoto, T. Horie, and S. Watanabe-Kohno, *Biochem. Biophys. Res. Commun.*, **116**, 612 (1983).
6. F.A.T. Barberán, J.M. Núñez, and F. Tomás, *Phytochemistry*, **24**, 1285 (1985).
7. V.H. Heywood, in: "Flora Europaea," Ed. by T.G. Tutin, V.H. Heywood, N.A. Burges, D.M. Moore, D.H. Valentine, S.M. Walters, and D.A. Webb, vol. 3, Cambridge University Press, Cambridge, 1972, pp. 138-143.
8. F.A.T. Barberán, F. Tomás, and F. Ferreres, *J. Chromatogr.*, **315**, 101 (1984).
9. F.A.T. Barberán, F. Tomás, L. Hernández, and F. Ferreres, *J. Chromatogr.*, **347**, 443 (1985).

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