Antiinflammatory Constituents from *Heterotheca inuloides*

Guillermo Delgado,*,† María del Socorro Olivares,† María Isabel Chávez,† Teresa Ramírez-Apan,† Edelmira Linares,‡ Robert Bye,‡ and Francisco J. Espinosa-García§

Instituto de Química e Instituto de Biología (Jardín Botánico) de la Universidad Nacional Autónoma de México, Ciudad Universitaria, Circuito Exterior, Coyoacán 04510, México, D. F., and Instituto de Ecología (Campus Morelia) de la Universidad Nacional Autónoma de México, Xangari 58089, Morelia, Michoacán, México

Received October 27, 2000

Three new compounds, cadalen-15-oic acid (**1**), 3,7-dihydroxy-3(4*H*)-isocadalen-4-one (**2**), and dicadalenol (**3**), were isolated from the aerial parts of *Heterotheca inuloides* (Mexican arnica), together with the known compounds 7-hydroxycadalene (**4**), 7-hydroxy-4α*H*-3,4-dihydrocadalene (**5**), 1α-hydroxy-1(4*H*)-isocadalen-4-one (**6**), 1α-hydroxy-4α*H*-1,2,3,4-tetrahydrocadalen-15-oic acid (**7**), 7-(3,3-dimethylallyloxy)coumarin, caryolan-1,9*β*-diol, and quercetin. The structures of the new compounds were elucidated by spectroscopic methods. The antiinflammatory activities of the extracts and the isolated compounds were evaluated by determining the inhibition of TPA-induced mouse ear edema. The natural products **3**, caryolan-1,9*β*-diol, and quercetin were the most active substances tested and displayed dose-dependent activities.

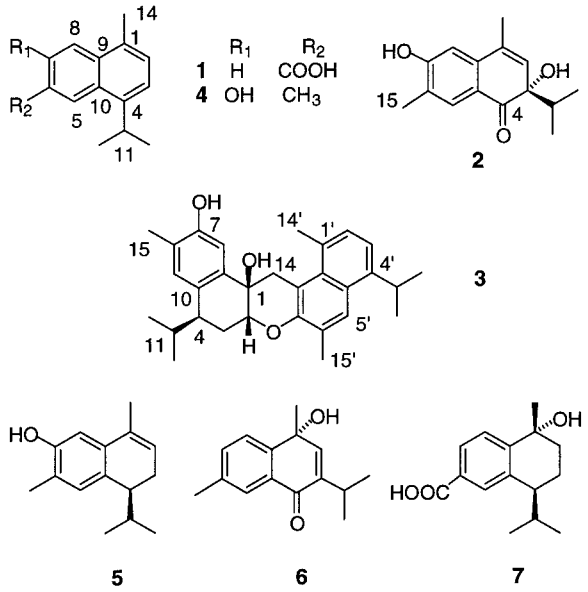
The complex of Mexican medicinal plants known as "árnica" is comprised of several plant species: *Heterotheca inuloides*, *Mentezlia conzattii*, *Zexmenia pringlei*, *and Haplopappus* spp. among others. The label plant of this complex, *Heterotheca inuloides* (Asteraceae), commonly named as Mexican arnica, acahual, cuauteteco, and xochihuepal, is widely used in folk medicine for the topical treatment of contusions and bruises and for the treatment of skin wounds and injuries.[1,2] This species is highly valued and frequently used in different parts of Mexico,[3,4] and some of its uses are similar to those of *Arnica montana*.[5−7]

Previous chemical investigations of this species have yielded polyacetylenes,[8] cadinanes,[6,8,9] triterpenes, sterols,[6] and flavonoids.[5,10,11] The plant has been reported to exhibit antimicrobial activity[9] and plant growth inhibitory activity;[12] cytotoxic and antioxidative properties have been found for some sesquiterpenes isolated from this plant.[13] The antiinflammatory and analgesic effects of extracts of *H. inuloides* have also been evaluated.[14]

In our continuing study of species included in medicinal plant complexes of Mexico,[15−17] it was found that an acetonic extract of the aerial parts of *H. inuloides* exhibited in vivo antiinflammatory activity when evaluated for inhibition of TPA-induced mouse ear edema. Purification of this extract by conventional methods afforded three new constituents (**1**−**3**), along with several known compounds. In this paper we report the isolation and characterization of these natural products together with their antiinflammatory activities.

Results and Discussion

The dried aerial parts of *H. inuloides* were extracted with acetone and then with methanol. These extracts showed inhibition on TPA-induced mouse ear edema (61.7 and 4.5%, respectively, at doses of 0.5 mg/ear). The acetonic extract was then subjected to chromatographic procedures to afford three new natural substances (**1**−**3**), together with 7-hydroxycadalene (**4**),[8] 7-hydroxy-4α*H*-3,4-dihydrocadalene (**5**),[9,18] 1α-hydroxy-1(4*H*)-isocadalen-4-one (**6**),[8] 1α-hydroxy-4αH-1,2,3,4-tetrahydrocadalen-15-oic acid (**7**),[19] 7-(3,3-dimethylallyloxy)coumarin,[20,21] caryolan-1,9*β*-diol,[22] quercetin,[23] stigmasterol, and *β*-sitosterol. The known compounds were identified by comparison of their physical and spectral data with those of authentic samples.



Compound **1**, obtained as a solid, gave the molecular ion peak at *m*/*z* 228.2909, indicating the molecular formula $C_{15}H_{16}O_2$. The IR spectrum indicated the presence of a carboxylic acid (3622, 1692 cm^{-1}), and the ^1H NMR spectrum of **1** (Table 1) displayed signals corresponding to ABX (δ_A 8.10, d, J_{AB} 8.5 Hz; δ_B 8.16, dd, J_{AB} 8.5, J_{BX} 1.5 Hz; δ_X 9.05, d, J_{BX} 1.5 Hz) and AB (δ_A 7.43, d, J_{AB} 7.0 Hz; δ_B 7.40, d, J_{AB} 7.0 Hz) systems of a trisubstituted naphthalene nucleus. The presence of methyl, isopropyl, and carboxyl moieties was evident by ^1H and ^{13}C NMR signals (Tables 1 and 2), establishing the cadinane skeleton. The HMBC correlation of the carbonyl (δ_C 172.03) with two benzenoid hydrogens fixed the position of the carboxylic acid at C-15. Thus, compound **1** was determined to be 4-isopropyl-1-methylnaphthalene-15-oic acid and named as cadalen-15-oic acid. This compound has been previously prepared, but not isolated as a natural product.[24] Spectro-

* To whom correspondence should be addressed. Tel: (52)-5622-4446. Fax: (52)-5616-2217. E-mail: delgado@servidor.unam.mx.
† Instituto de Química de la UNAM.
‡ Jardín Botánico, Instituto de Biología de la UNAM.
§ Instituto de Ecología (Campus Morelia) de la UNAM.

10.1021/np0005107 CCC: $20.00
Published on Web 06/12/2001

Table 1. ^{1}H NMR Spectral Data of **1**−**3** and **7** (500 MHz, $CDCl_3$, δ)[a]

H (H′)	1	2	3	3[b]	7
2	7.43 dd (7, 1)	6.05 d (1)	4.57 dd (11,4)	7.13 d (7.5)	1.84 m 2.05 m
3	7.40 d (7)		2.10 ddd (14, 11, 4) α 2.20 ddd (14, 4, 4) *β*	7.10 d (7.5)	1.86 m
4			2.94 ddd (8, 4, 4)		2.69 ddd (6, 6, 6)
5	9.05 d (1.5)	7.73 d (0.5)	7.00 s	7.77 s	7.99 d (2.0)
7	8.16 dd (8.5, 1.5)				7.92 dd (8.5, 2)
8	8.10 d (8.5)	6.71 s	7.05 s		7.70 d (8.5)
11	3.86 hept (6.5)	1.90 m	2.27 m	3.63 hept (7)	2.35 m
12	1.43 d (6.5)	0.89 d (7)	1.10 d (7)	1.34 d (7)	1.07 d (6.5)
13	1.43 d (6.5)	0.85 d (7)	0.86 d (7)	1.34 d (7)	0.79 d (6.5)
14	2.70 d (1)	2.09 d (1)	3.68 d (12) α 3.65 d (12) *β*	2.78 s	1.56 s
15		2.25 d (0.5)	2.20 s	2.38 s	
O*H*			4.86, s; 1.57 s		

[a] The assignments are based on 1D and 2D NMR experiments, including COSY, HMQC, and HMBC. [b] The values in the column correspond to the second unit of cadalene of the dimer (**3**), numbered with primes (′).

Table 2. ^{13}C NMR Spectral Data of **1**−**3** and **7** ($CDCl_3$, 125 MHz, δ)[a]

C (C′)	1	2	3	3[b]	7
1	132.16 s	130.09 s	68.75 s	131.03 s	70.46 s
2	129.34 d	131.96 d	75.75 d	129.77 d	37.17 t
3	122.46 d	79.39 s	25.95 t	119.24 d	19.78 t
4	144.78 s	203.99 s	40.69 d	142.67 s	43.26 d
5	128.01 d	130.04 d	130.98 d	124.06 d	130.36 d.
6	125.72 s	123.57 s	123.76 s	126.83 s	139.84 s
7	124.69 d	159.71 s	152.46 s	149.20 s	127.73 d
8	125.42 d	110.74 d	111.21 d	112.10 s	126.58 d
9	135.72 s	139.71 s	139.40 s	133.14 s	148.85 s
10	130.68 s	126.76 s	130.15 s	127.98 s	145.80 s
11	28.42 d	39.73 d	33.51 d	28.51 d	31.69 d
12	23.78 q	17.10 q	21.50 q	23.64 q	21.36 q
13	23.78 q	16.81 q	18.04 q	23.64 q	17.79 q
14	19.40 q	19.11 q	39.92 t	26.54 q	31.00 q
15	172.03 s	15.30 q	15.67 q	17.69 q	170.09 s

[a] Multiplicity bt DEPT, assignments by HMBC and HMQC experiments. [b] The values in the column correspond to the second unit of cadalene of the dimer (**3**), numbered with primes (′).

scopic assignments of **1** were made by HMQC, HMBC, and NOESY experiments.

Compound **2** was obtained as an optically active solid ($[\alpha]_D$ + 5.0), and its molecular formula was determined to be $C_{15}H_{18}O_3$ by HREIMS. The IR spectrum of **2** shows bands at 3618 and 1696 cm^{-1} corresponding to hydroxyl and carbonyl group absorptions, respectively. The ^{1}H NMR spectrum of **2** (Table 1) displays two 1H singlets corresponding to two benzenoid hydrogens (δ_H 7.73 and 6.71), suggesting they are *para* to each other. This was confirmed by the HMBC spectrum, which showed the low-field hydrogen (H-5) correlated with a methyl carbon (δ_C 15.30, C-15) and the upfield hydrogen (H-8) correlated with the carbon *ipso* to a phenol (δ_C 159.71, C-7). The presence of a vinylic methyl group at a trisubstituted double bond was established from ^{1}H NMR signals for the methyl (δ_H 2.09, 3H, d, J 1.0 Hz) and vinylic (δ_H 6.05, 1H, q, J 1.0 Hz) hydrogens. The HMBC experiment showed correlations of the carbonyl group (δ_C 203.99, C-4) with the vinylic hydrogen (H-2), with the aliphatic methine (of an isopropyl group, H-11), and with the low-field benzenoid hydrogen (H-5), indicating that this substance is 3,7-dihydroxy-3(4*H*)-isocadalen-4-one (**2**), where the isopropyl group is located at C-3 (isocadinane skeleton). HMQC and NOESY experiments confirmed the structure of **2**. The *β*-orientation of the isopropyl group was proposed on biogenetic grounds, but has not been established.

Compound **3** was obtained as an optically active solid, and its HREIMS showed a molecular ion at 444.6121, consistent with the molecular formula $C_{30}H_{36}O_3$. UV absorptions at 243, 288, and 323 nm suggested the presence of aromatic rings, and the hydroxyls groups were evident by the strong IR band at 3621 cm^{-1}. The ^{1}H NMR spectrum of **3** shows the presence of three sets of aromatic hydrogens: two *ortho*-coupled aromatic protons (δ 7.13, d, 7.5 Hz; δ 7.10, d, J 7.5 Hz), two *para*-oriented aromatic protons (δ 7.00, brs; δ 7.05, s), and one aromatic proton of a pentasubstituted benzene ring (δ 7.77, s). The presence of three methyl groups on the benzenoid rings was evident from the HMQC experiment (δ_H 2.78 with δ_C 26.54; δ_H 2.38 with δ_C 17.69, and δ_H 2.20 with δ_C 15.67). Further analysis of the HMQC spectrum indicated the presence of a phenol (δ_H 4.86, δ_C 152.46), a tertiary alcohol (δ_H 1.57, δ_C 68.75), an oxymethine (δ_H 4.57, δ_C 75.75), two methylenes (δ_H 2.07, 2.20, δ_C 25.95; δ_H 3.68, 3.65, δ_C 39.92), a benzylic methine (δ_H 2.94, δ_C 40.69), and two isopropyl groups. The COSY spectrum indicated that one methylene is isolated (C-14) and the second (C-3) is flanked by two methines (C-2 and C-4). These fragments could be accommodated considering two cadinane units, one with five unsaturations (including a 1,2,4,5-tetrasubstituted benzenoid ring) and the second with seven unsaturations (a pentasubstituted naphthalene), connected by an additional heterocycle that includes a tertiary alcohol (to complete 13 degrees of unsaturation). In the HMBC experiment significant correlations were observed between C-7 and H-15/H-5; C-9 and H-14a,b/H-8; C-4 and H-2/H-5/H-11/H-12/H-13; C-2′ and H-14′/H-3′; and C-5′ and H-15′. Additionally, HMBC correlations between C-7′ and H-2/H-14a,b, and C-9′ and H-14a,b established the C(2)−O−C(7′) and C(14)−C(8′) connectivities of a dihydropyran ring joining the dimeric cadinane. Compound **3** was named dicadalenol.

The relative stereochemistry of **3** was determined from the coupling constants of relevant protons and from the observed NOESY cross-peaks. The coupling constants of H-2 (11 and 4 Hz) indicated an axial orientation of this proton, and the couplings for H-4 (8, 4, and 4 Hz) suggested a pseudo-axial orientation of the C(4)−C(11) *σ*-bond. These couplings established a pseudo-boat for the cyclohexene ring, since in the chair form, the equatorial orientation of the isopropyl at C-4 would result in strong interaction with H-5, which lies on the same plane. Consistently with these observations, H-2 shows NOE cross-peaks with the hydroxyl proton of the alcohol at C-1, with H-11, and with H-12, indicating that the C-4 isopropyl, H-2, and the C-1 hydroxyl group are all oriented on the same side. Additional NOE interactions were observed between H-4 and H-5, H-2′ and H-14′, H-14′ and H-11a,b, and H-8 and H-14a,b,

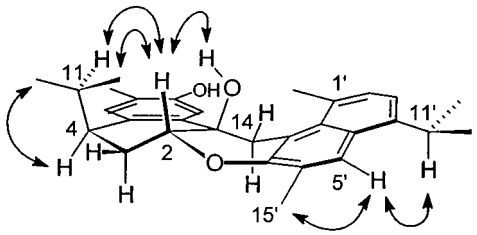


Figure 1. Selected NOE correlations of **3**.

Table 3. Antiinflammatory Activities of Dicadalenol (**3**), Caryolan-1,9β-diol, and Quercetin[a]

compound	doses (mg/ear)	% inhibition of edema	ED_{50} (mg/ear)
	0.01	17.08* ± 4.05	
3	0.05	29.45** ± 6.10	0.11
	0.10	57.06** ± 4.73	(0.03, 0.46)
	0.50	70.15** ± 3.71	
	0.05	20.74* ± 3.22	
caryolan-1,9β-diol	0.10	28.96** ± 4.87	0.34
	0.50	56.17** ± 3.01	(0.27, 0.41)
	1.00	67.76** ± 6.57	
	0.05	29.93** ± 7.38	
quercetin	0.10	41.86** ± 5.47	0.16
	0.50	67.42** ± 3.92	(0.12, 0.21)
	1.00	83.60** ± 3.05	
	0.05	26.35*	
indomethacin	0.10	34.77**	0.18
	0.50	60.53**	(0.07, 0.52)
	1.00	93.48**	

[a] *$p < 0.05$, **$p < 0.01$.

confirming the assigned structure and the relative stereochemistry of *rel*-1β,7-dihydroxy-2βH-1,2,3,4-tetrahydro-14.8′,2(*O*)·7′-dicadalenol (**3**). Figure 1 shows selected NOE correlations of **3**, considering the β-orientation of the C(4)−C(11) bond as is usually seen in sesquiterpenoids from Asteraceae.

The effects of topically administered natural products on TPA-induced mouse ear edema are summarized in the Experimental Section. All the tested compounds displayed activity, with dicadalenol (**3**), caryolan-1,9β-diol, and quercetin as the most active compounds. The dose−response curves for these compounds were determined and, together with the calculated ED_{50}, are shown in Table 3. Because the amount available was limited, dicadalenol (**3**) was tested at lower doses; nevertheless it was found to be the most active compound. Considering the cadinanes and isocadinanes (**1**−**7**), it can be observed that the presence of phenolic or carboxylic groups does not influence the antiinflammatory activities. The different structural pattern of caryolan-1,9β-diol with respect to the cadinanes suggests that it could act at a different step in the inflammation process. It is interesting to note the similarities between the structures and the bioactivities of dicadalenol (**3**) and quercetin. Although they are formed by different biosynthetic pathways, they could have a similar mode of biological action.

In conclusion, the aerial parts of *H. inuloides* contain a series of secondary metabolites that display topic antiinflammatory activities, and these results correlate with some of the popular uses of the Mexican arnica. Further investigations are in progress on the chemical constituents of different populations of this species at various stages of development.

Experimental Section

General Experimental Procedures. The ^1H and ^{13}C NMR spectra were recorded on a Varian Unity Plus-500 instrument, and the chemical shifts are expressed in ppm (δ) relative to tetramethylsilane. Samples for NOE experiments were degassed and sealed under argon. Standard pulse sequences were used for COSY, NOEdiff, DEPT, HMQC, and HMBC experiments. Infrared spectra were recorded with Nicolet Magna IR TM 750 and Perkin-Elmer 283B instruments. MS data were recorded with a JEOL JMS-AX 505 HA mass spectrometer. Electron impact mass spectra were obtained at 70 eV ionization energy. TLC: Merck silica gel 60 F254 plates (0.25 mm). TLC visualization was accomplished with either a UV lamp (254 and 365 nm) or a charring solution (12 g of ceric ammonium sulfate dihydrate, 22.2 mL of concentrated H_2SO_4, and 350 g of ice).

Plant Material. Aerial parts (leaves, flowers, and stems) of *Heterotheca inuloides* Cass. were obtained from Atlixco, Puebla, México. The plant material was identified by E. Linares and R. Bye, and a voucher specimen (R. Bye + E. Linares 19401) is kept in the Ethnobotanical Collection of the National Herbarium (MEXU), Instituto de Biología de la Universidad Nacional Autónoma de México.

Extraction and Isolation. Dried and powdered plant material (3.5 kg) was extracted with Me_2CO at room temperature (3 times/24 h) and then with MeOH (3 times/24 h), to afford, after evaporation of the solvent, 195 and 180 g of residue, respectively. The acetonic extract was purified by vacuum liquid chromatography (VLC)[25,26] using hexane and mixtures of hexanes−EtOAc. According to the TLC profiles, 14 crude fractions were obtained (A−N). Fraction E (7 g, eluted with hexanes−EtOAc, 19:1) was rechromatographed by VLC, and some fractions were further chromatographed on Si gel to afford 33 mg of 7-hydroxy-3,4-dihydrocadalene (**5**) [mp 103−104 °C [lit. 103.5 °C][8] R_f 0.33 (*n*-hexanes−EtOAc, 9:1)] and 35 mg of 7-hydroxycadalene (**4**) [mp 116−117 °C [lit. 118−119 °C],[9,18] R_f 0.35 (hexanes−EtOAc, 9:1)]. Fraction G (9 g, eluted with *n*-hexanes−EtOAc, 9:1) was purified by successive VLC on silica gel (hexanes−EtOAc gradient) to afford stigmasterol (350 mg), β-sitosterol (225 mg), and 1α-hydroxy-1(4*H*)-isocadalen-4-one (**6**) (75 mg) as an oil[8] [R_f 0.62 (*n*-hexanes−EtOAc, 85:15), $[\alpha]^{25}_D$ +5 (*c* 0.3, MeOH)] and 45 mg of 7-(3,3-dimethylallyloxy)coumarin[20,21] [R_f 0.55 (hexanes−EtOAc, 85:15)]. Fraction I was chromatographed using VLC with mixtures of hexanes−EtOAc to afford 8.5 mg of cadalen-15-oic acid (**1**) and 15 mg of 3,7-dihydroxy-3(4*H*)-isocadalen-4-one (**2**). Fraction L (eluted with *n*-hexanes−EtOAc, 7:3) was purified by successive VLC to afford 10 mg of 1α-hydroxy-4α*H*-1,2,3,4-tetrahydrocadalen-15-oic acid (**7**).[19] The data reported in ref 19 (for the methyl ester of **7**) are inverted with those of its epimer at C-1. The data for the natural product **7** (not previously reported) are the following: mp 203−205 °C; $[\alpha]^{25}_D$ +21 (*c* 0.2, MeOH); R_f 0.36 (*n*-hexanes−EtOAc, 7:3); UV (MeOH) λ_{max} (log ϵ) 206 (3.86), 237 (3.42), 277 (2.6) nm; IR ν_{max} ($CHCl_3$) 3294, 2957, 2872, 1680, 1433, 1273 cm^{-1}; ^1H and ^{13}C NMR (see Tables 1 and 2); EIMS *m*/*z* 248 (M^+, 3), 233 (54), 187 (73), 143 (100), 128 (28), 115 (14). From fraction M (eluted with *n*-hexanes−EtOAc, 3:2) were isolated, by successive VLC, caryolan-1,9β-diol (32 mg)[22] [oil, R_f 0.43 (*n*-hexanes−EtOAc, 4:1)] and dicadalenol (**3**, 15 mg). Rechromatography of fraction N (eluted with *n*-hexanes−EtOAc, 1:1) afforded 55 mg of quercetin. [23]

Cadalen-15-oic acid (1): mp 168−170 °C; R_f 0.44 (*n*-hexanes−EtOAc, 2:1); UV (MeOH) λ_{max} (log ϵ) 213 (3.30), 242 (3.52), 286 (2.19), 331 (2.22) nm; IR ($CHCl_3$) ν_{max} 3618, 1696, 1607 cm^{-1}; ^1H and ^{13}C NMR (see Tables 1 and 2); EIMS *m*/*z* 228 [M^+] (72), 213 (100), 169 (12), 153 (17), 154 (26); HREIMS *m*/*z* 228.2909, calcd for $C_{15}H_{16}O_2$ 228.2902

3,7-Dihydroxy-3(4*H*)-isocadalen-4-one (2): mp 198−200 °C; $[\alpha]^{25}_D$ +5.0 (MeOH, *c* 0.2); R_f 0.56 (*n*-hexane−AcOEt, 7:3); UV (MeOH) λ_{max} (log ϵ) 205 (3.18), 256 (3.47) nm; IR ($CHCl_3$) ν_{max} 3618, 2975, 2930, 1696, 1607 cm^{-1}; ^1H and ^{13}C NMR (see Tables 1 and 2); EIMS *m*/*z* 246 $[M]^+$ (26), 204 (98), 175 (100), 176 (90), 161 (19), 71 (16), 43 (52), 41 (18); HREIMS *m*/*z* 246.3050, calcd for $C_{15}H_{18}O_3$ 246.3054.

Dicadalenol (3): mp 258−260 °C; R_f 0.35 (*n*-hexane−AcOEt, 4:1); UV (MeOH) λ_{max} (log ϵ) 205 (4.04), 243 (4.05), 288 (3.32), 323 (3.59), 338 (2.65); IR ($CHCl_3$) ν_{max} 3621, 2973,

2928. 2875, 1437 cm^{-1}; ^1H and ^{13}C NMR (see Tables 1 and 2); EMIE *m/z* (int rel) 444 (48), 426 (29), 383 (12), 218 (29), 175 (100), 83 (4), 69 (6), 43 (9); HREIMS *m/z* 444.6121, calcd for $C_{30}H_{36}O_3$ 444.6126.

Antiinflammatory Activities. The bioassays were performed as described previously.[27,28] The major antiinflammatory activity was found in the acetonic extract using the inhibition of edema in mouse ear induced by TPA (61.7% of inhibition in the acetonic extract and 4.5% in the methanolic extract, 0.5 mg/ear). The isolated natural compounds displayed the following antiinflammatory activities (% inhibition of edema, 0.5 mg/ear). **1**: 60.24 (± 10.0), **2**: 57.24 (± 11.6), **3**: 70.15 (± 10.3). **4**: 46.31 (± 11.8), **5**: 31.58 (± 2.9), **6**: 41.71 (± 10.2), **7**: 24.60 (± 15.3), 7-(3,3-dimethylallyloxy)coumarin: 14.17 (± 9.8), caryolan-1,9β-diol: 66.71 (± 13.5), quercetin: 66.05 (± 16.2). ED_{50} for **3**, caryolan-1,9β-diol, and quercetin are shown in Table 3.

Acknowledgment. We thank Rocío Patiño, Luis Velasco, Javier Pérez-Flores, Beatriz Quiroz, and Héctor Rios (Instituto de Química de la UNAM) for technical assistance. M.S.O. thanks the Consejo Nacional de Ciencia y Tecnología (México) for providing a graduate scholarship. This work was supported in part by Dirección General de Asuntos del Personal Académico de la UNAM (Project IN 221198).

References and Notes

(1) (a) Bye, R.; Linares, E.; Estrada, E. *Rec. Adv. Phytol*. **1995**, *29*, 65−82. (b) Linares, M. E., Bye, R.; Flores, B. *Tes Curativos de México*, 2nd ed.; Cuadernos del Instituto de Biología 7, Universidad Nacional Autónoma de México, 1990.
(2) Martínez, M. *Las Plantas Medicinales de México*; Editorial Botas; México, D. F., 1969.
(3) Nicholson, M. S.; Arzeni, Ch. B. *Econ. Bot*. **1993**, *47*, 184−192.
(4) Lozoya, X.; Aguilar, A.; Camacho, J. R. *Rev. Méd. IMSS (Méx.)* **1987**, 283−291.
(5) Willuhn, G.; Röttger, P. M. *Dtsch. Apoth. Zeit.* **1980**, *120*, 1039−1042.
(6) Willuhn, G.; Schneider, R. *Arch. Pharm. (Weinheim)* **1987**, *320*, 393−396.
(7) Willuhn, G.; Schneider, R.; Matthiesen, U. *Dtsch. Apoth. Zeit*. **1985**, *125*, 1941−1944.
(8) Bohlmann, F.; Zdero, Ch. *Chem. Ber*. **1976**, *109*, 2021−2025.
(9) Kubo, I.; Muroi, H.; Kubo, A.; Chaudhuri, S. K.; Sánchez, Y.; Ogura, T. *Planta Med*. **1994**, *60*, 218−221.
(10) Jerga, Ch.; Merfort, I.; Willuhn, G. *Planta Med*. **1990**, *56*, 122−123.
(11) Jerga, Ch.; Merfort, I.; Willuhn, G. *Planta Med*. **1990**, *56*, 413−415.
(12) Kubo, I.; Ishiguro, K.; Chaudhuri, S. K.; Kubo, Y.; Sánchez, Y.; Ogura, T. *Phytochemistry* **1995**, *38*, 553−554.
(13) Kubo, I.; Chaudhuri, S. K.; Kubo, Y.; Sánchez, Y.; Ogura, T.; Saito, T.; Ishikawa, H.; Haraguchi, H. *Planta Med*. **1996**, *62*, 427−430.
(14) Gené, R. M.; Segura, L.; Adzet, T.; Marin, E.; Iglesias, J. *J. Ethnopharm*. **1998**, *60*, 157−162.
(15) Delgado, G.; Reza-Garduño, R. G.; Toscano, R. A.; Bye, R. A.; Linares, E. *Heterocycles* **1988**, *27*, 1305−1312.
(16) Delgado, G.; García, P. E.; Roldán, R. I.; Bye, R. A.; Linares, E. *Nat. Prod. Lett*. **1996**, *8*, 145−150.
(17) Aguilar, M. I.; Delgado, G.; Bye, R. A.; Linares, E. *Phytochemistry* **1993**, *33*, 1161−1163.
(18) Gallagher, M. J.; Sutherland, M. D. *Aust. J. Chem*. **1965**, *18*, 1111−1113.
(19) Bohlmann, F.; Rajindek, K. G.; King, R. M.; Robinson, H. *Phytochemistry* **1982**, *21*, 2982−2984.
(20) Chiang, M. T.; Bittner, M.; Silva, M.; Mondaca, A.; Zemelman, R.; Sammes, P. G. *Phytochemistry* **1982**, *21*, 2753−2755.
(21) Lassak, E. V.; Southwel, I. A. *Aust. J. Chem*. **1972**, *125*, 2491−2596.
(22) Hegmann, H.; Tezuka, Y.; Kikuri, T.; Supriyatna, S. *Chem. Pharm. Bull*. **1994**, *42*, 138−146.
(23) Wadsworth, T. L.; Koop, D. R. *Biochem. Pharmacol*. **1999**, *57*, 941−949.
(24) Reddy, M. P.; Rao, G. S. K. *Indian J. Chem., Sect. B* **1982**, *21B*, 885−886.
(25) Pelletier, S. W.; Chokshi, H. P.; Desai, H. K. *J. Nat. Prod*. **1986**, *49*, 892−900.
(26) Coll, J. C.; Bowden, B. F. *J. Nat. Prod*. **1986**, *49*, 934−936.
(27) Della Logia, R.; Tubaro, A.; Sosa, S.; Becker, H.; Saar, St.; Isaac, O. *Planta Med*. **1994**, *60*, 516−529.
(28) Tubaro, A.; Dri, P.; Delbello, G.; Zilli, C.; Della Logia, R. *Agents Actions* **1985**, *17*, 347−349.

NP0005107