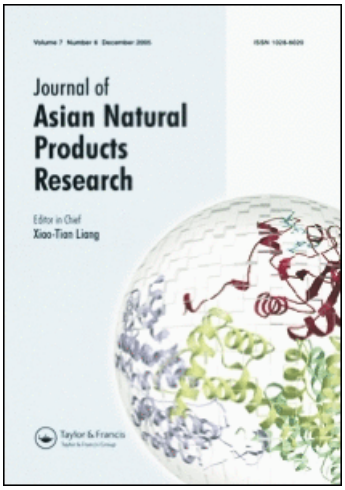

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A new amide from *Asarum forbesii* Maxim.

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A highly unsaturated new amide, (2*E*,4*Z*,8*Z*,10*Z*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide (**1**), was isolated in very small quantities from the whole plant of *Asarum forbesii* Maxim. together with four known compounds, (2*E*,4*E*,8*Z*,10*E*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide (**2**), (−)-sesamin (**3**), (−)-asarinin (**4**) and (*E*)-asarone (**5**). The *Z*/*E* isomers, **1** and **2**, were separated successfully by developed silver-ion medium-pressure liquid chromatography (SIMPLC). Compound **2** and the two diastereoisomers, **3** and **4**, were isolated from this plant for the first time. The characterization of these compounds was achieved by various spectroscopic methods.

Keywords: *Asarum forbesii* Maxim; (2*E*,4*Z*,8*Z*,10*Z*)-*N*-Isobutyl-2,4,8,10-dodecatetraenamide; (2*E*,4*E*,8*Z*,10*E*)-*N*-Isobutyl-2,4,8,10-dodecatetraenamide; (−)-Sesamin; (−)-Asarinin; (*E*)-Asarone

1. Introduction

Asarum forbesii Maxim belongs to the Aristolochiaceae, a family consisting of about 93 species [1], many of which are widely used in traditional medicine. For example, an important Chinese traditional drug, "Xi-xin", was prepared from *A. heterotropoides* Fr. var. *mandshuricum* (Maxim.) Kitag. or *A. sieboldii* Miq. and has been used as an analgesic, antitussive, or anti-allergic remedy [2]. Some detailed phytochemical studies on the above plants have been carried out [3–7]. *Asarum forbesii* Maxim, is a herbal medicine that has many effects such as "analgesic, antitussive, anti-allergic, diuretic and diaphoretic effects" [8]. However, there are very few phytochemical reports on this plant, except for the isolation of four asarumins [8]. Here we report the isolation and characterization of a new amide named (2*E*,4*Z*,8*Z*,10*Z*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide (**1**) from the hexane extract by developed silver-ion medium-pressure liquid chromatography. In addition, (2*E*,4*E*,8*Z*,10*E*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide (**2**), (−)-sesamin (**3**), (−)-asarinin (**4**), and (*E*)-asarone (**5**) (figure 1) were also isolated and identified. Compounds **2**–**4** were isolated from *Asarum forbesii* Maxim. for the first time.

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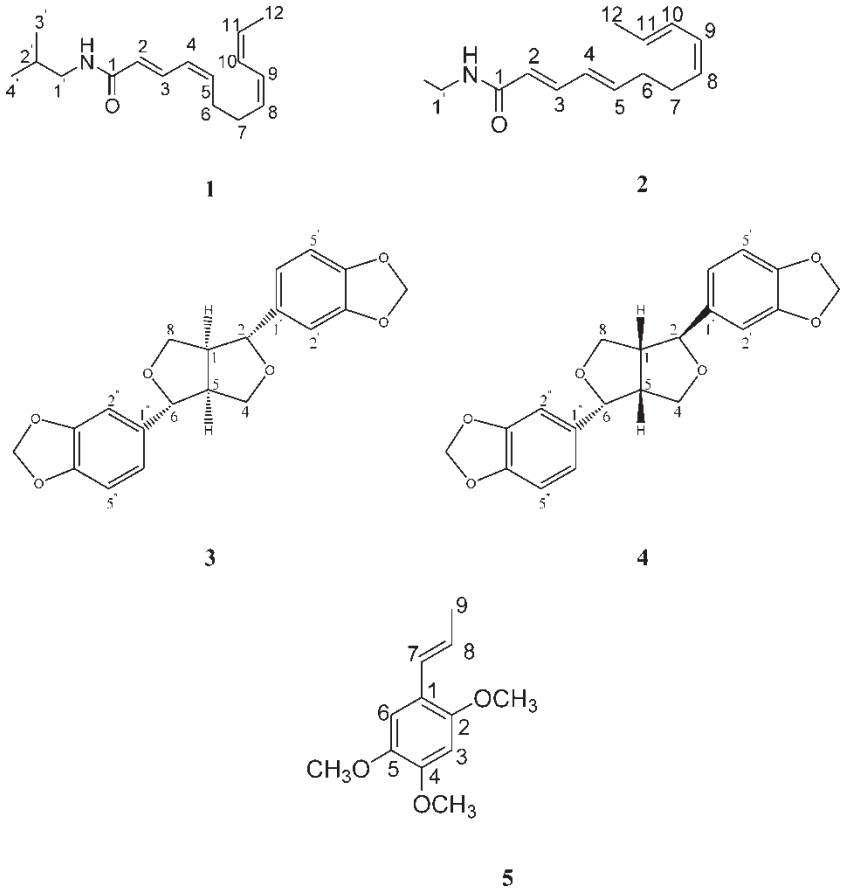

Figure 1. Structures of compounds **1**–**5**.

2. Results and discussion

The n-hexane extract of *Asarum forbesii* Maxim. was subjected to column chromatography on silica gel using n-hexane–diethyl ether (1:1) as eluent to afford 4 fractions. Gas chromatography-mass spectrometry (GC-MS) analysis of fraction 4 indicated that it contained at least two *cis*/*trans* isomers with olefinic bonds. In addition, the very small difference in GC retention times of the two isomers ($t_{R(1)} = 20.06$, $t_{R(2)} = 19.86$ min) indicated that their separation by silica gel column chromatography would be very difficult—as was confirmed in practice. Since silver-ion chromatography is very useful for the separation of such double bond isomers, it was applied to the fraction using silica impregnated with silver nitrate (9%) prepared according to the literature [9] and afforded compounds **1** and **2**. However, the purity of **1** measured by GC was less than 85%. To improve on this, ultrasound was applied for the first time. The impregnated silica gel was exposed to ultrasound to distribute homogeneously the Ag^+ in the silica gel. Under these conditions, the purity of **1** was increased from 84% to 93%, showing that a very efficient, convenient and practical method has been developed for the preparation of silica gel impregnated with silver nitrate.

Compound **1** has a molecular formula of $C_{16}H_{25}NO$, determined by high-resolution TOF-MS (*m/z* 247.1929 $[M]^+$). Cleavage of the allylic C_6—C_7 bond led to the base peak at *m/z* 81 and another major ion peak at *m/z* 167. The presence of the NH group was deduced from the IR band at 3300 cm^{-1} and a broad resonance signal at δ 5.0 in the ^1H NMR spectrum. The UV absorption at 261.7 nm and the IR peaks at 1550 and 1650 cm^{-1} were attributed to the double bond conjugated amide group. The 4-*cis* geometry was deduced from the fact that the chemical shifts for H-4 and H-5 were at higher fields than those recorded for the corresponding protons in (2*E*,4*E*,8*Z*,10*E*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide [10] and the $J_{4,5}$ was at 11.5 instead of 15 Hz, as expected for a *trans*-double bond. The 10-*cis* geometry was also apparent from the higher field shifts for H-10 and H-11 compared to those recorded for (2*E*,4*E*,8*Z*,10*E*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide and also from the $J_{10,11}$ of 10 Hz. Furthermore, chemical shifts (δ) and coupling constants (*J*) for H-2, 3, 4, 5 were similar to those reported for the corresponding protons in (2*E*,4*Z*,8*Z*,10*E*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide [11]; in addition, δ and *J* for H-8-11 were similar to those of the corresponding protons for (2*E*,4*E*,8*Z*,10*Z*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide [10]. Inspection of the ^1H NMR data published for (2*E*,4*E*,8*Z*,10*E*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide, (2*E*,4*E*,8*Z*,10*Z*)-*N*-isobutyl-2,4,8,10-dodeca-tetraenamide, (2*E*,4*Z*,8*Z*,10*E*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide, (2*E*,4*E*,8*E*,10*Z*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide [12] and (2*E*,4*E*,8*E*,10*E*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide [13] revealed that the ^1H NMR data for **1** did not correspond to any of these compounds. Hence this is the first time this isomer has been isolated.

Compound **2** was characterized as (2*E*,4*E*,8*Z*,10*E*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide by comparison of the ^1H NMR, IR, UV and MS data with those reported in the literature [9]. The structures of the other three known compounds were identified as (−)-sesamin (**3**), (−)-asarinin (**4**) and (*E*)-asarone (**5**) by ^1H and ^{13}C NMR, IR, UV and MS spectral data. These structures were confirmed by comparison with literature data [14–16].

3. Experimental

3.1 General experimental procedures

Melting points were determined on an X-4 micromelting apparatus and are uncorrected; Optical rotations were obtained on a JASCO P-1010 digital polarimeter, whereas UV and IR spectra were recorded on a Waters 996 spectrometer and a Perkin-Elmer 683 infrared spectrophotometer in KBr disks, respectively. ^1H and ^{13}C NMR spectra were recorded on a Bruker-DRX 400 spectrometer; chemical shifts (δ ppm) are from TMS as an internal standard. EI (EIMS) and high-resolution EI mass spectra (HR-EIMS) were recorded on a Micromass GC-TOF mass spectrometer fitted with an electron impact (EI+) source, EI 70 eV, source temperature 180°C, column DB-5MS (J & W, 20 m × 0.18 mm, 0.18 μm film thickness), injection temperature 250°C, carrier gas He, flow rate 1.0 ml min^{-1}, split ratio 500:1, column temperature program 50°C for 2 min, then raised to 250°C at a rate of 10°C min^{-1} and held at this temperature for 7 min.

3.2 Plant material

The plant material was collected in Jiangsu province, China, in June 2002, and identified by C. Chen, Professor of Botany, Liaoning Normal University, where a voucher specimen has been deposited.

3.3 Extraction and isolation

The air-dried whole plants of *Asarum forbesii* Maxim. (1 kg) were ground and extracted twice with refluxing n-hexane (2 × 2 l) for 24 h. The extract was subjected to column chromatography on silica gel using n-hexane-diethyl ether (1:1) as eluent to afford 4 fractions. Frs. 1–4 were respectively recrystallized from n-hexane to yield compound **3** (51 mg), compound **4** (28 mg), compound **5** (528 mg) and a mixture of amides. The mixture containing **1** and **2** was further separated by medium-pressure liquid chromatography on silica gel impregnated with silver nitrate (9%), using cyclohexane-ethyl acetate (10:1) as eluant and monitoring with gas chromatography to afford **1** (6 mg) and **2** (16 mg).

New method of preparing silica gel impregnated with silver nitrate (9%) for medium pressure liquid chromatography: an aqueous solution of silver nitrate (3.6 g) in distilled water (80 ml) was mixed with 200–300 mesh silica gel (40 g). The mixture was then treated with an ultrasonic wave generator for 1 h and then dried in an oven at 150°C for a further hour. The almost white resulting powder was stored in a beaker wrapped with dark paper and dried over phosphorus pentoxide in a vacuum desiccator. Columns were packed in the same way as ordinary silica columns and wrapping with dark paper was not necessary for the medium-pressure liquid chromatography.

3.3.1 (2*E*,4*Z*,8*Z*,10*Z*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide (1). Colourless needles (n-hexane). mp 63 °C, UV_{max} (MeOH) (nm): 261.7, 235.8; IR ν_{max} (KBr) (cm^{-1}): 3300 (NH), 2910 (CH), 1720 (C═O), 1650 (CONH), 1550, 990 (*trans*, C═C), 690 (*cis*, C═C); Micromass Q-Tof MS *m/z* 247.1929 calcd. for $C_{16}H_{25}NO$ $[M]^+$ 247.1936. EIMS 70 eV, *m/z* 247.2 $[M]^+$ (0.02), 246.2 $[M-H]^+$ (0.04), 167.1 $[M-C_6H_8]^+$ (5.6), 81.0 $[M-C_{10}H_{16}NO]^+$ (100), 67.0 $[M-C_{11}H_{18}NO]^+$ (3.2), 41.0 $[M-C_{13}H_{20}NO]^+$ (5.5); ^1H NMR (400 MHz, $CDCl_3$) δ (ppm): 5.82 (1H, d, $J = 15$ Hz, H-2), 7.54 (1H, dd, $J = 15$, 11.5 Hz, H-3), 6.11 (1H, dd, $J = 11.5$, 11.5 Hz, H-4), 5.78 (1H, dt, $J = 11.5$, 7 Hz, H-5), 2.33 (2H, br dt, $J = 7$, 7 Hz, H-6), 2.27 (2H, br dt, $J = 7$, 7 Hz, H-7), 5.42 (1H, dt, $J = 10$, 7 Hz, H-8), 6.32 (1H, brt, $J = 10$ Hz, H-9), 6.25 (1H, tq, $J = 10$, 7 Hz, H-10), 5.55 (1H, dq, $J = 10$, 7 Hz, H-11), 1.75 (3H, dd, $J = 7$, 2 Hz, H-12), 3.16 (2H, t, $J = 7$ Hz, H-1′), 1.80 (1H, m, H-2′), 0.93 (6H, d, H-3′, 4′).

3.3.2 (2*E*,4*E*,8*Z*,10*E*)-*N*-isobutyl-2,4,8,10-dodecatetraenamide (2). Colourless needles (n-hexane), mp 63 °C; UV_{max} (MeOH) (nm): 261.7, 235.8. Micromass Q-Tof MS *m/z* 247.1929 calcd. for $C_{16}H_{25}NO$ $[M]^+$ 247.1936. EIMS 70 eV, *m/z* 247.2 $[M]^+$ (1), 167.1 $[M-C_6H_8]^+$ (10), 81.0 $[M-C_{10}H_{16}NO]^+$ (100), 67.0 $[M-C_{11}H_{18}NO]^+$ (6.5), 41.0 $[M-C_{13}H_{20}NO]^+$ (11); ^1H NMR (400 MHz, $CDCl_3$) δ (ppm): 5.80 (1H, d, $J = 15$ Hz, H-2), 7.20 (1H, dd, $J = 15$, 10 Hz, H-3), 6.18 (1H, dd, $J = 15$, 10 Hz, H-4), 6.10 (1H, dt, $J = 15$, 7 Hz, H-5), 2.28 (2H, m, H-6), 2.28 (2H, m, H-7), 5.25 (1H, dt, $J = 10$, 7 Hz, H-8), 5.97 (1H, brt, $J = 10$ Hz, H-9), 6.30 (1H, br dd, $J = 15$, 10 Hz, H-10), 5.70 (1H, dq, $J = 15$, 7 Hz,

H-11), 1.80 (3H, br d, $J = 7$ Hz, H-12), 3.16 (2H, t, $J = 7$ Hz, H-1′), 1.80 (1H, m, H-2′), 0.93 (6H, d, $J = 7$ Hz, H-3′,4′).

3.3.3 (−)-Sesamin (3). Light green needles (n-hexane), mp 118°C; $[\alpha]_D^{28}$ −59.2 ($CHCl_3$, *c* 0.05); UV_{max} (MeOH) (nm): 203.0, 238.2, 287.7; IR ν_{max} (KBr) (cm^{-1}): 2820, 1610, 1480, 1435, 1250, 1080, 1020, 780; ^{1}H NMR (400 MHz, $CDCL_3$) δ (ppm): 3.05 (2H, m, H-1α, H-5α), 3.86 (2H, dd, $J = 8$, 4 Hz, H-4β, H-8β), 4.23 (2H, dd, $J = 8$, 4 Hz, H-4α, H-8α), 4.71 (2H, d, $J = 4$ Hz, H-2β, H-6β), 5.95 (4H, s, 2 × OCH_2O), 6.78 (2H, d, $J = 8$ Hz, H-5′, H-5″), 6.80 (2H, dd, $J = 8$, 1.2 Hz, H-6′, H-6″), 6.85 (2H, d, $J = 1.2$ Hz, H-2′, H-2″); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm): 54.3 (C-1, 5), 71.7 (C-4, 8), 85.8 (C-2, 6), 101.1 (2 × OCH$_2$O), 106.5 (C-2′, 2″), 108.2 (C-5′, 5″), 119.3 (C-6′, 6″), 134.9 (C-1′, 1″), 147.1 (C-4′, 4″), 147.9 (C-3′, 3″).

3.3.4 (−)-Asarinin (4). White needles (n-hexane). mp 118°C, $[\alpha]_D^{28}$ −116.2 ($CHCl_3$, *c* 0.30); UV_{max} (MeOH) (nm): 203.0, 238.2, 287.7; IR ν_{max} (KBr) (cm^{-1}): 2900, 1490, 1440, 1370, 1260, 1180, 1080, 1040, 930; ^{1}H NMR (400 MHz, $CDCL_3$) δ (ppm): 2.88(1H, m, H-1), 3.31 (2H, m, H-5, H-4β), 3.83 (2H, m, H-8α, H-4α), 4.10 (1H, d, $J = 8$ Hz, H-8β), 4.41 (1H, d, $J = 8$ Hz, H-2), 4.83 (1H, d, $J = 4$ Hz, H-6), 5.95 (4H, s, 2 × OCH_2O), 6.77-6.86 (6H, m, H-2′, H-2″, H-5′, H-5″, H-6′, H-6″); ^{13}C NMR (100 MHz, $CDCl_3$) δ (ppm): 50.1 (C-1), 54.6 (C-5), 69.6 (C-8), 70.9 (C-4), 82.0 (C-2), 87.6 (C-6), 101.0 (2 × OCH$_2$O), 106.4 (C-2′), 106.5 (C-2″), 108.1 (C-5′, 5″), 118.6 (C-6′), 119.6 (C-6″), 132.1 (C-1′), 134.8 (C-1″), 146.5 (C-3′), 147.2 (C-4″), 147.6 (C-4′), 147.9 (C-3″).

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References

[1] Y.Q. Yan. *Zhong. Cihai.*, 155–157 (1996).
[2] P.G. Xiao. *Mod. Chin. Mater. Med.*, **3**, 219–228 (2002).
[3] T. Kosuge, M. Yokota, H. Nukaya, Y. Gotoh, M. Nagasawa. *Chem. Pharm. Bull.*, **26**, 2284–2285 (1978).
[4] I. Yasuda, K. Takeya, H. Itokawa. *Chem. Pharm. Bull.*, **29**, 564–566 (1981).
[5] K. Hashimoto, T. Yanagisawa, Y. Okui, Y. Ikeya, M. Maruno, T. Fujita. *Planta Med.*, **60**, 124–127 (1994).
[6] K. Hashimoto, M. Okada, M. Maruno. *Nat. Med.*, **48**, 39–48 (1994).
[7] M. Takasaki, T. Konoshima, I. Yasuda, T. Hamano, H. Tokuda. *Biol. Pharm. Bull.*, **20**, 776–780 (1997).
[8] M. Kobayashi, M. Asai, T. Ishiguro, T. Kishi, R. Bian, Q.M. Xie. *Shoyakugaku Zasshi*, **43**, 230–234 (1989).
[9] T.S. Li, J.T. Li, H.Z. Li. *J. Chromatogr.*, **715**, 372–375 (1995).
[10] W. Herz, P. Kulanthaivel. *Phytochemistry*, **24**, 173–174 (1985).
[11] R. Martin, H. Becker. *Phytochemisty*, **24**, 2295–2300 (1985).
[12] I.J.O. Jondiko. *Phytochemisty*, **25**, 2289–2290 (1986).
[13] I. Yasuda, K. Takeya, H. Itokawa. *Chem. Pharm. Bull.*, **29**, 564–566 (1981).
[14] N.J. Sun, C.J. Chang, J.M. Cassady. *Phytochemistry*, **26**, 3051–3053 (1987).
[15] S. Shimizu, H. Kawashima, K. Akimoto, Y. Shinmen, H. Sugano, H. Yamada. *Phytochemistry*, **31**, 757–760 (1992).
[16] L. Siergiejczyk, J. Poplawski, B. Lozowicka, A. Dubis, B. Lachowska. *Magn. Reson. Chem.*, **38**, 1037–1038 (2000).