m, H-17b), 172.67 (C-18), 2.79 (2 H, t, J = 6.3 Hz, H-19)/28.97, 2.87 (2 H, t, J = 6.3 Hz, H-20)/29.17, 174.26 (C-21), 168.61 (C-22); for selected HMBC correlations, see Fig.

5. Chiral amino acid analysis

Trichoderide A (**1**, 2.0 mg) was hydrolyzed in 1 mL of 6 N HCl at 110 °C for 20 h, and then dried under vacuum. The hydrolysate was eluted from a C18 column (Dikma) using MeOH/H_2O (10 : 90). The elute was dried under vacuum and reconstituted with 100 μL of H_2O prior to analysis [CHIRAL PAK CR(+), 4.61 × 50 mm; detection: UV 200 nm; injected amount: 5 nmol; mobile phase: pH 1.5 $HClO_4$ in H_2O, flow rate 0.4 ml/min]. The hydrolysate was chromatographed alone and co-injected with standards to confirm assignments. Retention times (min) of the four amino acids of **1** were as follows: L-Ala (4.68), L-Val (7.06), L-Pro (3.82), and L-Orn (5.25), which were identical with the authentic amino acids. The standard retention times (min) of the corresponding D-amino acids were D-Ala (3.54), D-Val (5.85), D-Pro (3.08), and D-Orn (4.30).

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Research Department of Natural Medicine[1], China-Japan Research Institute of Medical and Pharmaceutical Sciences[2], Shenyang Pharmaceutical University, Shenyang, P.R. China

Two new triterpenoids from the carpophore of *Xanthoceras sorbifolia* Bunge

Wei Li[1], Xian Li[1], Jia Yang[2], Lin-Hao Li[2], Ning Li[1], Da-Li Meng[1]

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Prof. Xian Li, Research Department of Natural Medicine, Shenyang Pharmaceutical University, Box 49, Shenyang, 110016, P.R. China
proflixian@163.com

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Two new triterpenoids 3-*O*-β-d-glucopyranosyl (1→6)-β-d-glucopyranosyl, 28-*O*-β-d-glucopyranosyl (1→6) [α-l-rhamnopyranosyl (1→2)]-β-d-glucopyranosyl, 16-deoxybarringtogenol C (**1**) and 22-*O*-acetyl-21-*O*-(4′-*O*-angeloyl)-β-d-fucopyranosyl theasapogenol B (**2**), were isolated from the dried carpophore of *Xanthoceras sorbifolia* Bunge (Sapindaceae). **1** and **2** were found to have activity of inhibiting the proliferation of two human tumor cell lines.

Xanthoceras sorbifolia Bunge (Sapindaceae) is a shrub mainly growing in Inner Mongolia, China. Its bark and fruits are used to treat rheumatism and enuresis of children as a folk medicine. Previous phytochemical studies on this plant revealed the presence of saponins (Chen et al. 1985a, 1985b), flavonoids (Ma et al. 2000). We investigated the chemical constituents of the carpophore of *X. sorbifolia* Bunge and report here the isolation and identification of two new triterpenoids (Fig.), as well as their inhibiting activities against human tumor cell lines.

Compound **1**, white powder from MeOH, was deduced to have the molecular formula $C_{60}H_{100}O_{28}$, on the basis of its ESIMS (m/z 1291 $[M + Na]^+$) and NMR data and was considered to be a triterpen glycoside due to a positive Liebermann-Burchard and Molish reactions. Furthermore, the fragmentation patterns indicated the loss of sugar moieties (m/z 1291 $[M + Na]^+$, 1145 $[M + Na\text{-rha}]^+$, 1129 $[M + Na\text{-}162]^+$, 983 $[M + Na\text{-}146\text{-}162]^+$, 967 $[M + Na\text{-}146\text{-}2 \times 162]^+$, 821 $[M + Na\text{-}146\text{-}2 \times 162]^+$, 659 $[M + Na\text{-}146\text{-}3 \times 162]^+$). On hydrolysis, two kinds of monosaccharide units were obtained and identified as glucose and

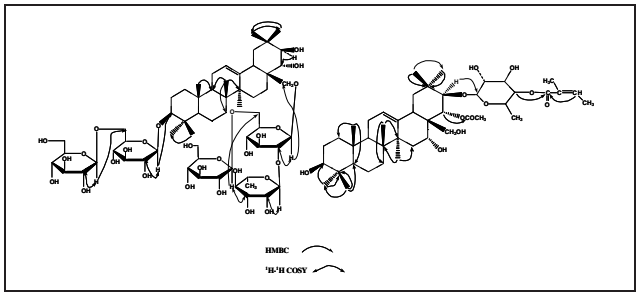

Fig.: Important correlations of compound **1** and **2** = 1

rhamnose by co-TLC with authentic sample. In the ^{1}H NMR spectrum revealed seven tertiary methyl signals at δ 0.94, 0.98, 1.06, 1.23, 1.24, 1.24, 1.27, one trisubstituted olefinic proton of δ 5.34 and five anomeric protons at δ 4.73 (1 H, d, J = 9.5 Hz), 4.88 (1 H, d, J = 7.7 Hz), 5.02 (1 H, d, J = 7.6 Hz), 5.14 (1 H, d, J = 7.7 Hz), 6.53 (1 H, s) which indicated the configuration of the glucose were of β orentation. By comparision of the ^{13}C NMR spectral data of **1** with those of 16-deoxybarringtogenol C (Doddrell et al. 1974), the distinguished downfield shifted of C-3 and C-28 indicated that the two sugar moieties of **1** were linked at C-3 and C-28. In the HMBC experiment, the long-range correlations were observed between H-1‴′ (δ 4.73) and C-28 (δ 74.3), H-1‴′ (δ 5.02) and C-6‴ (δ 70.2), H-1″ (δ 5.14) and C-6′ (δ 70.5), H-1′ (δ 4.88) and C-3 (δ 89.1). Combined with the ^{1}H-^{1}H COSY, HMQC and TOCSY experiments, the spin systems and linkages for sugar were assigned. Accordingly, the structure of **1** was established as 3-*O*-β-D-glucopyranosyl (1→6)-β-D-glucopyranosyl, 28-*O*-β-D-glucopyranosyl (1→6) [α-L-rhamnopyranosyl (1→2)]-β-D-glucopyranosyl 16-deoxybarringtogenol C.

Compound **2** was obtained as white needles ($CHCl_3$-MeOH) which responded positively to Liebermann-Burchard and Molish reactions. The molecular formula of **2** was deduced as $C_{43}H_{68}O_{11}$ from ESI-MS (m/z 783.3 $[M + Na]^+$) and NMR data. The sugar was identified as fucose by acid hydrolysis, and co-TLC with an authentic sample. In the ^{1}H NMR spectrum of **2**, the signals at δ 1.94 (3 H, brs), 2.08 (3 H, d, J = 7.2 Hz), 5.92 (1 H, d, J = 7.2 Hz) could be assigned to an angeloyl function. Additionally, an acetoxyl group could be observed at δ_H 2.34 (3 H, s) and δ_C 171.8, 22.1. Besides that, the ^{1}H NMR spectrum also gave seven singlet signals of methyls at δ 0.90, 0.93, 1.05, 1.25, 1.40, 1.48, 1.84 and the signal of the anomeric proton of fucose at δ 4.87 (1 H, d, J = 7.3 Hz) which indicated that the anomer of the fucose was of β orientation. The ^{13}C NMR data of compound **2** were very similar to those of 22-*O*-acetyl-21-*O*-(4′-*O*-acetyl-3′-*O*-angeloyl)-β-D-fucopyranosyl protoaescigenin (Chen et al. 1985a) except for an upfield shift of the C-24 signal by 48 ppm (from 64.6 in compound D to 16.6 in **1**) which suggested the absence of the OH group at C-24 and inexistence of an acetoxyl attached to the fucose. Furthermore, the downfield shift of C-4′ signal by 1.5 ppm and the upfield shift of C-2′, C-3′ signals as comparision with fucose suggested that the angeloyl group was linked at C-4′ of fucose. In the HMBC spectrum, the long-range correlations were observed between δ 5.05 (H-21) and δ 105.7 (C-1′), δ 5.67 (H-4′) and δ 168.0 (C-1″), δ 6.16 (H-22) and δ 171.8 (COMe). With the data above, the structure of **2** was established as 22-*O*-acetyl-21-*O*-(4′-*O*-angeloyl)-β-D-fucopyranosyl theasa-pogenol B (**2**).

Compounds **1** and **2** were tested for their anti-tumour activtity *in vitro* against two cell lines (Hela and A375-S2). **1** was found to have weak activity against the Hela with an IC_{50} value of 499.33 μmol/L and to be inactive against A375-S2. **2** showed mild activity against both the cell lines with IC_{50} values at 101.33 and 174.25 μmol/L.

Experimental

1. Apparatus

Melting points were measured on a Yanaco micro-hot-stage apparatus and are uncorrected. 1D and 2D NMR spectra were recorded on a Bruker-ARX-300 or an AV-600 spectrometer, using TMS as an internal standard. The ESI-MS was determined by Finnigan LCQ spectrometer.

2. Plant material

The carpophores of *Xanthoceras sorbifolia* Bunge were collected at Shenhe district, Shenyang, China. A voucher specimen (No.0187512) is deposited in Herbarium Department of Institute of Applied Ecology, Chinese Academy of Sciences.

3. Extraction and isolation

Dried carpophores of *Xanthoceras sorbifolia* Bunge (10 kg) were extracted with 70% ethanol. The extract (800 g) was concentrated *in vacuo*, then half of the extract was eluted with H_2O, 70% ethanol and 95% ethanol on macroporous resin. The 70% ethanol fraction (110 g) was first subjected to column chromatography on silica gel yielding fraction 15–19 [CH_3Cl_3 : MeOH (100 : 16–100 : 24), 28.0 g] which was then rechromatographed on ODS eluted with H_2O : MeOH (40 : 60) to give compound **1** (15.0 mg), and yielding fraction 7–9 [CH3Cl3 : MeOH (100 : 7–100 : 9), 9.3 g] which was then rechromatographed on silica gel eluted with petroleum ether : acetone (2 : 1) to give compound **2** (6.7 mg).

4. Characterization of the compounds

*4.1. Compound **1***

^{1}H NMR (300 MHz, pyridine-d_5): 5.34 (1 H, brs, H-12), 3.74 (1 H, d, J = 9.8 Hz, H-21), 4.30 (1 H, d, J = 9.6 Hz, H-22), 1.24 (3 H, s, H-23), 0.98 (3 H, s, H-24), 0.94 (3 H, s, H-25), 1.06 (3 H, s, H-26), 1.24 (3 H, s, H-27), 1.23 (3 H, s, H-29), 1.27 (3 H, s, H-30), 4.32 (2 H, m, H-28), 4.88 (1 H, d, J = 7.7 Hz, H-1′), 5.15 (1 H d, J = 7.7 Hz, H-1″), 4.73 (1 H, d, J = 9.5 Hz, H-1‴), 5.02 (1 H, d, J = 7.6 Hz, H-1‴′), 6.53 (1 H, s, rha-H-1); ^{13}C NMR (75 MHz, pyridine-d_5): 39.5 (C-1), 25.9 (C-2), 89.1 (C-3), 38.9 (C-4), 55.7 (C-5), 18.5 (C-6), 32.8 (C-7), 40.2 (C-8), 47.9 (C-9), 36.9 (C-10), 23.9 (C-11), 123.8 (C-12), 143.1 (C-13), 41.8 (C-14), 25.8 (C-15), 18.2 (C-16), 43.3 (C-17), 42.1 (C-18), 46.5 (C-19), 36.3 (C-20), 76.9 (C-21), 75.2 (C-22), 28.2 (C-23), 17.1 (C-24), 15.9 (C-25), 16.9 (C-26), 26.3 (C-27), 74.3 (C-28), 30.4 (C-29), 19.7 (C-30), 107.0 (C-1′), 75.6 (C-2, C-2‴′), 78.5 (C-3′, C-3″, C-3‴, C-3‴′), 71.8 (C- 4′, C-4‴), 76.9 (C-5′), 70.5 (C-6′), 105.5 (C-1″), 75.2 (C-2″, rha-C-4), 71.6 (C-4″), 78.4 (C-5″), 62.8 (C-6″), 103.7 (C-1‴;), 80.0 (C-2‴), 76.7 (C-5‴), 70.2 (C-6‴), 105.7 (C-1‴′), 71.7 (C-4‴′), 101.0 (rha-C-1), 72.5 (rha-C-2, rha-C-3), 69.3 (rha-C-5), 18.9 (rha-C-6).

*4.2. Compound **2***

m.p. 239–240 °C, ^{1}H NMR (300 MHz, pyridine-d_5): 1.94 (3 H, brs, 5″-CH_3), 2.08 (3 H, d, J = 7.2 Hz, 4″-CH_3), 5.92 (1 H, d, J = 7.2 Hz, H-3″), 5.05 (1 H, d, J = 9.8 Hz, H-21), 6.16 (1 H, d, J = 9.8 Hz, H-22), 3.63 (1 H, d, J = 10.7 Hz, H-28a), 3.42 (1 H, d, J = 10.7 Hz, H-28b), 4.87 (1 H, d, J = 7.3 Hz, H-1′), 1.20 (3 H, d-like, 6′-CH_3), 0.90 (3 H, s, H-26), 0.93 (3 H, s, H-25), 1.05 (3 H, s, H-24), 1.25 (3 H, s, H-23), 1.40 (3 H, s, H-30), 1.48 (3 H, s, H-29), 1.84 (3 H, s, H-27). ^{13}C NMR (75 MHz, pyridine-d_5): 39.1 (C-1), 28.2 (C-2), 78.0 (C-3), 39.4 (C-4), 55.8 (C-5), 18.8 (C-6), 33.2 (C-7), 40.1 (C-8), 47.1 (C-9), 37.2 (C-10), 23.9 (C-11), 123.8 (C-12), 143.2 (C-13), 41.7 (C-14), 34.8 (C-15), 68.7 (C-16), 47.8 (C-17), 40.1 (C-18), 47.8 (C-19), 37.9 (C-20), 85.5 (C-21), 74.2 (C-22), 28.8 (C-23), 16.6 (C-24), 17.0 (C-25), 15.8 (C-26), 27.5 (C-27), 64.0 (C-28), 30.2 (C-29), 20.2 (C-30), 105.7 (C-1′), 73.0 (C-2′), 73.4 (C-3′), 74.2 (C-4′), 69.4 (C-5′), 17.3 (C-6′), 168.0 (C-1″), 128.6 (C-2″), 138.0 (C-3″), 16.4 (C-4″), 21.1 (C-5″), 171.8 ($COCH_3$), 22.1 ($COCH_3$).

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