The chemical constituents of *S. chinensis* were previously reported to contain nortriterpenoid saponins, flavonoids, and neolignans. To find the biologically active compounds, a detailed phytochemical investigation was carried out on the stem of *S. chinensis*, which resulted in isolation of two new triterpenoid saponins, named as yemuoside YM36 and YM37. Their structures were established on the basis of 2D-NMR experiments and mass spectrometry.

In addition, eight known triterpenoid saponins 3-O-α-L-arabinopyranosyl-30-norhederagenin (3), sinosofoside A (4), yemuoside YM11 (5), 3-O-α-L-arabinopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl-hederagenin (7), 3-O-β-D-xylpyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl-hederagenin (8), hederasapogenin D (9) and 28-O-β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl-hederagenin (10) were isolated from this plant. Compounds 3-4 and 7-10 were isolated from this plant for the first time.

**Figure 1.** Their structures of compounds 1-10.

1. R₁ = H; R² = CH₃OH; R³ = glc(1→6)glc
2. R₁ = xyl(1→3)-rha(1→2)-ara; R² = CH₃OH; R³ = H
3. R₁ = ara; R² = CH₃OH; R³ = H
4. R₁ = ara; R² = CH₃OH; R³ = rha(1→4)-glc(1→6)-glc
5. R₁ = ara; R² = CH₃; R³ = glc(1→6)-glc
6. R₁ = ara(1→3)-rha(1→2)-ara; R² = CH₃; R³ = H
7. R₁ = rha(1→2)-ara; R² = H
8. R₁ = xyl(1→3)-rha(1→2)-ara; R³ = H
9. R₁ = ara; R² = rha(1→4)-glc(1→6)-glc
10. R₁ = H; R² = glc(1→6)-glc

glc: β-D-glucopyranosyl; xyl: β-D-xylpyranosyl;
rha: α-L-rhamnopyranosyl; ara: α-L-arabinopyranosyl

**Key Words:** Triterpenoid saponins, Lardizabalaceae, Stauntonia chinensis
molecule. Compound human hepatocellular liver carcinoma using the MTT assay their cytotoxicities against the cancer cell lines HepG2 fore, the structure of YM determined as 7.0 Hz), the relative configurations of the sugar units were ranosyl-(1,6, the drug concentration that causes 50% of cell growth naro-30-norhederagenin, named as yemuoside YM 28 (81.5; H-1'' at 83.4. According to anomeric proton signals at 8.1 Hz), 6.35 (1H, br s) and 5.35 (1H, d, J = 7.0 Hz), the relative configurations of the sugar units were determined as α-arabinose, α-rhamnose and β-xylene. Therefore, the structure of 2 was identified as 3-O-β-D-xylopyranosyl-(1→3)-α-L-rhamnopyranosyl-(1→2)-α-L-arabinopyranosyl-30-norhederagenin, named as yemuoside YM37.

Triterpenoid saponins 2, 4, 6-8 and 10 were evaluated for their cytotoxicities against the cancer cell lines HepG2 human hepatocellular liver carcinoma using the MTT assay method. MTT assays were used to estimate IC50 values, that is, the drug concentration that causes 50% of cell growth inhibition after 24 h of continuous exposure to the test molecule. Compound 6-8 exhibited moderate cytotoxic activity with an IC50 values of 14.97, 24.46 and 21.32 μM respectively, using cisplatin as a positive control (IC50 31.60 μM). Compound 2, 4 and 10 were inactive against tested cell lines (IC50 > 40 μM).

Experimental

Plant Materials. The stems of Stauntonia chinensis were collected from NanNing, Guangxi Zhuang Autonomous Region, P. R. China and identified by associate chief pharmacist Jin-Wei Huang at Guangxi Institute of Minority Medicine. The voucher specimen (20090801) was deposited with the Herbarium of College of Pharmacy, South Central University for Nationalities.

Extraction and Isolation. The stems of Stauntonia chinensis (16 kg) were extracted with 60% EtOH three times and then successively partitioned with EtOAc and n-BuOH. The extract of n-BuOH (300 μL) was chromatographed on silicagel with CHCl3-MeOH (100:0, 98:2, 95:5, 9:1, 8:2, 7:3, 1:1, 3:7, 0:100 v/v) to give 15 fractions (fr.1-fr.15). Fr. 10 (9.6 g) was subjected to CC on silica gel with CHCl3-MeOH (1:0, 99:1, 95:5, 9:1, 0:1) and further purified by octadecisilan CC with H2O/MeOH (95:5:→0:1) to obtain 3 (10.3 mg). Fr. 12 (30 g) was subjected to CC on silica gel with EtOAc-EtOH (1:0, 98:2, 95:5, 9:1, 8:2, 7:3, 0:1) to afford compounds 2 (13.0 mg), 6 (8.1 mg), 7 (15 mg), 8 (15.0 mg). Fr. 13 (45 g) was subjected to CC on silica gel with EtOAc-EtOH (1:0, 98:2, 95:5, 9:1, 8:2, 7:3, 0:1) to give five fractions (Frs. 13.1-13.5). Fr. 13.2 (3.6 g) was successively separated by CC (ODS; H2O/MeOH; (9:1→0:1)) to give compound 1 (29 mg). Fr. 13.2.3 (314.8 mg) was successively separated by CC (ODS; H2O/MeOH; (9:1→0:1)) and further purified by semi-prep. HPLC (CH3CN/H2O 36:74, 2.5 mL/min) to afford compound 5 (9.6 mg; tR 15.7 min) and 10 (18 mg; tR 24.4 min). Fr. 13.5 (14.1 g) was subjected to CC on silica gel with CHCl3-MeOH (99:1, 98:2, 95:5, 9:1, 8:2, 7:3, 6:4, 0:1) and further purified by semi-prep. HPLC (CH3CN/H2O 30:70, 2.5 mL/min) to afford compound 4 (14.3 mg; tR 11.8 min) and 9 (14.0 mg; tR 19.2 min).

Yemuoside YM3 (1): white amorphous powder; [α]D20 = +42.5 (c 0.48, MeOH); 1H NMR and 13C NMR (in CDCl3) spectroscopic data, see supporting information; HRESIMS m/z 803.4186 [M+Na]+ (calcd. for C41H39O12Na, 803.4194).

Yemuoside YM37 (2): white amorphous powder; [α]D20 = +40.8 (c 0.50, MeOH); 1H NMR and 13C NMR (in CDCl3) spectroscopic data, see supporting information; HRESIMS m/z 889.4548 [M+Na]+ (calcd. for C42H39O12Na, 889.4562).

Acidic Hydrolysis. Compounds 1-2 (3 mg) were respectively added to trifluoro-acetic acid (TFA, 4 N) solution (6 mL), then heated for 3 h under 90 ºC. After cooled to room temperature, the hydrolytical solution was extracted with chloroform (3 × 6.0 mL). The water layer was concentrated to 0.5 mL. The completely concentrated water layer and reference substances of β-D-glucose, α-L-arabinose and β-D-xylene and α-L-rhamnose were derived by reacting with 1.5 mg hydroxyamine hydrochloride and 0.5 mL pyridine for 1 hour at 90 ºC. After cooling, 0.9 mL Ac2O was added and the
mixture was heated at 90 °C for 1 h. The reaction mixtures were evaporated under reduced pressure, and the resulting aldonitrile peracetates were analyzed by GC-MS. The $t_R$ values of β-D-glucose, β-D-xylose, α-L-arabinose and α-L-rhamnose derivatives were 11.04, 9.08, 8.98 and 8.86 min, respectively.

**Cytotoxic Activity.** Cell viability was measured using the MTT assay. The HepG2 cells were cultured in DMEM medium containing heat inactivated 10% (v/v) fetal bovine serum (FBS) and 100 U/mL penicillin, 100 µg/mL streptomycin. The cells were incubated in a humidified atmosphere of 5% CO$_2$ at 37 °C and pass-aged every 3 days by trypsinization (0.25%). For experiments, HepG2 cells were in-cubated in complete medium with 10% fetal bovine serum in 96-well plates (1000 cells/well). Cells were treated with or without different concentrations of saponins (dissolved in DMSO) when the cells reached 70-80% confluence. Test compounds and blank group had four repetitions. After 24 h, the medium was removed and the serum free DMEM containing 10% MTT (5 mg/mL) were added to each wells. 4 hours later, the culture medium containing MTT was removed and 100 µL DMSO was add to each well to dissolve the formazan and placed on a shaker for 10s. The absorbance values was measured at a wavelength of 492 nm using a microplate reader, and the absorbance values were expressed as a percentage of untreated control cells (control = 100%).

**Acknowledgments.** This work was financially supported by the Natural Science Foundation of Hubei Province of China (2012FFB07409) and National Key Technology R & D Program in the 12th Five Year Plan of China (2012BAI27B06).

**Supporting Information.** NMR spectral data of compounds 1 and 2 are available as Supporting Information.

**References**