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Triterpenoid saponins from *Mitragyna rotundifolia*

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1. Subject and source

The genus *Mitragyna* belongs to the family Rubiaceae and is found in swampy territory in the tropical and subtropical regions of Africa and Asia including four species in Africa and six species in India and Asia (Shellard et al., 1978). *Mitragyna rotundifolia* (Roxb.) Kuntze is a shrub growing in the south of Yunnan province, China. The bark of *M. rotundifolia* was collected in Xishuang-banna and a voucher was deposited in the Herbarium of the Kunming Institute of Botany, Chinese Academy of Science (KUN No 0334819).

2. Previous work

Previous phytochemical investigations on *Mitragyna* led to isolation of indole alkaloids (Shellard et al., 1967, 1978; Houghton and Shellard, 1974) and triterpenoid saponins (Cheng et al., 2002a,b; Fatima et al., 2002; Takayama et al., 2004).

3. Present study

Dried powder of the bark of *M. rotundifolia* (6.4 kg) was extracted three times with ethanol at room temperature. After evaporation of the solvent in vacuo, the concentrated extract was suspended in water and extracted with EtOAC and *n*-BuOH. The *n*-BuOH extract (44 g) was subjected to CC over silica gel (200–300 mesh), developing with $CHCl_3$/MeOH (95:5 to 6:4). The $CHCl_3$/MeOH (8:2) fraction was chromatographed repeatedly by CC over silica gel, and the fraction eluted with $CHCl_3$/MeOH/H_2O (8:2:0.2) was submitted to Sephadex LH-20 CC using MeOH as eluent yielding quinovic acid-3-*O*-β-D-6-deoxy-glucopyranoside 28-*O*-β-D-glucopyranosyl ester (**1**, 57 mg), quinovic acid-27-*O*-α-L-rhamnopyranosyl ester (**2**, 34 mg) and quinovic acid-3-*O*-α-L-rhamnopyranoside (**3**, 54 mg). The $CHCl_3$/MeOH (10:1) fraction was chromatographed on Sephadex LH-20 developing with $CHCl_3$/MeOH (1:1), and the fraction was isolated

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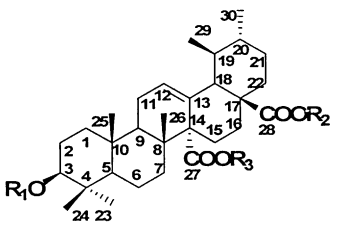
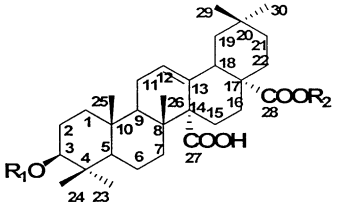
by RP_{18} CC eluted with MeOH/H_2O (7:3) to yield qinovic acid-27-*O*-β-D-glucopyranosyl ester (**4**, 56 mg), quinovic acid-3-*O*-6-deoxy-glucopyranoside (**5**, 107 mg), qinovic acid-27-*O*-β-D-glucopyranosyl ester (**6**, 28 mg), cincholic acid-3-*O*-β-D-6-deoxy-glucopyranoside (**7**, 12 mg), cincholic acid-3-*O*-β-D-6-deoxyglucopyranoside (**8**, 20 mg) and cincholic acid-28-*O*-β-D-glucopyranosyl ester (**9**, 31 mg). The structures of compounds **1**–**9** were determined on the basis of NMR data, including 2D experiments, by comparison with date reported in the literature (Kang et al., 2003, 2004a,b).



1 R_1=6-deoxy-β-D-glc R_2=β-D-glc R_3=H
2 R_1=R_2=H R_3=α-L-rha
3 R_1=α-L-rha R_2=R_3=H
4 R_1=R_2=H R_3=β-D-glc
5 R_1=6-deoxy-β-D-glc R_2=R_3=H
6 R_1=R_2=H R_3=6-deoxy-β-D-glc



7 R_1=6-deoxy-β-D-glc R_2=H
8 R_1=β-D-glc R_2=H
9 R_1=H R_2=β-D-glc

4. Chemotaxonomic significance

Quinovic acid glycosides are mainly found in the family Rubiaceae, and about 34 have been isolated from the genera, *Anthocephalus* (Sahau et al., 1999), *Adina* (Fan et al., 1995; Fan and He, 1997), *Cinchona* (Hani et al., 1995), *Guettarda* (Capasso et al., 1998), *Luculia* (Kang et al., 2003), *Mussaenda* (Zhao et al., 1995), *Nauclea* (Lamidi et al., 1995), *Neonauclea* (Kang et al., 2004a,b) and *Uncaria* (Aquino et al., 1997; Mary et al., 2005). But so far, nine quinovic acid glycosides were isolated from *Mitragyna inermis* (Cheng et al., 2002a,b), *Mitragyna stipulosa* (Fatima et al., 2002), and *Mitragyna africanus* (Takayama et al., 2004). Compounds **2**–**6** isolated from *M. rotundifolia* are reported for the first time from the genus of Mitragyna. Only 10 cincholic acid glycosides have been mainly isolated from the genera *Adina* (Fan et al., 1995; Fan and He, 1997), *Cinchona* (Hani et al., 1995), *Luculia* (Kang et al., 2003), *Mussaenda* (Zhao et al., 1995), *Neonauclea* (Kang et al., 2004a,b), and it is the first time that cincholic acid glycosides have been isolated from the genus of *Mitragyna*.

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