A new glycoside from *Alpinia officinarum*

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Abstract: Aim To investigate the glycosidic constituents in the rhizomes of *Alpinia officinarum* Hance. **Methods** The isolation and purification of glycosides were done with column chromatography on macro porous resin, polyamides and Sephadex LH-20, whilst the structure elucidation was done by HRCI-MS and NMR (1D and 2D) methods. **Results** A glycosidic ester identified as 4′-hydroxy-2′-methoxyphenol-β-*D*-{6-*O*-[4″-hydroxy-3″, 5″-dimethoxy(benzoate)]}-glucopyranoside (**I**), along with a known compound *n*-butyl-β-*D*-fructopyranoside (**II**), were isolated and characterized. **Conclusion** **I** was found to be a new compound, named as alpinoside A, whilst **II** was isolated from the genus *Alpinia* for the first time.

Key words: *Alpinia officinarum*; glycosides; alpinoside A

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高良姜根茎中的一个新糖苷

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摘要: 目的 对高良姜(*Alpinia officinarum* Hance)根中的糖苷类成分进行分离和结构鉴定。方法 通过大孔树脂、聚酰胺和凝胶反复柱色谱分离得到糖苷类化合物, 利用多种波谱技术鉴定其化学结构。结果 分离鉴定了2个糖苷类化合物, 其结构分别为4′羟基-2′甲氧基苯酚-β-*D*-{6-*O*-[(4″羟基-3″, 5″二甲氧基)苯甲酸]}-吡喃葡糖苷(**I**)和正丁基-β-*D*-吡喃果糖苷(**II**)。结论 化合物 **I**为新化合物, 命名为高良姜苷 A, 化合物 **II**为首次从该属植物中分离得到。

关键词: 高良姜; 糖苷; 高良姜苷 A

Introduction

The rhizomes of *Alpinia officinarum* Hance are used as traditional herbs in China for relieving stomachache, treating colds, invigorating the circulatory system, and reducing swellings[1]. Previous studies have demonstrated that the extracts of *A. officinarum* possessed various biological activities, such as anti-inflammation, as a result of inhibitory effect against prostaglandin biosynthesis, antiemetic[2] and antioxidant effects[3]. Some aromatic glycosides recently isolated from the methanol extract of fresh rhizomes of *A. officinarum* were found to show certain antioxidant activities[4,5]. Here we report a new glycoside, 4′-hydroxy-2′-methoxyphenol-β-*D*-{6-*O*-[4″-hydroxy-3″, 5″-dimethoxy(benzoate)]}-glucopyranoside, named as alpinoside A (**I**) (Figure 1) along with a known compound *n*-butyl-β-*D*-fructopyranoside (**II**) isolated from the dried rhizomes of *A. officinarum*.

Results and discussion

Compound **I** was isolated as white crystals, with an $[\alpha]_D$ value of -4.6°(MeOH) and a mp of 154-156 ℃. The molecular formula was established to be $C_{22}H_{26}O_{12}$ based on molecular ion peak $[M^+]$ at *m/z*

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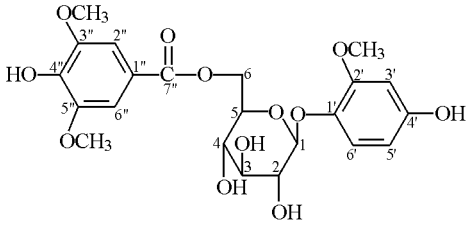

Figure 1 Structure of alpinoside A

482.144 6 (calcd. 482.142 4) in the HRCI-MS spectrum. The UV spectrum of **I** exhibited absorption bands at 279, 219, and 206 nm. The ^{1}H NMR spectrum showed four aromatic signals at δ 6.05 (1H, dd, J = 8.5, 2.5 Hz), 6.41 (1H, d, J = 2.5 Hz), 6.89 (1H, d, J = 8.5 Hz) and δ 7.31 (2H, s). The former three were attributed to a 1, 2, 4 trisubstituted benzene ring whilst the latter one to a 1, 3, 4, 5 tetrasubstituted benzene ring under highly symmetrical circumstance. In addition, there also existed signals at δ 3.76 (3H, s) and δ 3.85 (6H, s), corresponding to three methoxyl groups, and resonances around δ 3.35 - 4.70 to a sugar moiety in the ^{1}H NMR spectrum (Table 1) of **I**. The presence of two substituted benzene ring, three methoxyl groups and a sugar moiety were confirmed by the ^{13}C NMR data (Table 1) of **I**. In addition, the ^{13}C NMR spectrum suggested the presence of a carbonyl group (C-7″, δ 168.34). The location of aromatic hydroxyl and methoxyl groups in the aromatic units were determined mainly on the basis of NMR data and the HMBC spectrum (Figure 2). EIMS showed fragment ions of 198 and 140. These data indicated the presence of 3, 5-dimethoxy-4-hydroxybenzoate and 1, 4-dihydroxy-3-methoxylbenzene.

The determination of a glucopyranoside moiety was mainly by comparing the six signals around δ 65.75 - 104.75 in the ^{13}C NMR spectrum with known compounds[4], and was further confirmed by hydrolysis of **I**. In addition, the coupling constant of 7.5 Hz for the anomeric proton at δ 4.70 indicated the β-configuration of the glucosidic linkage.

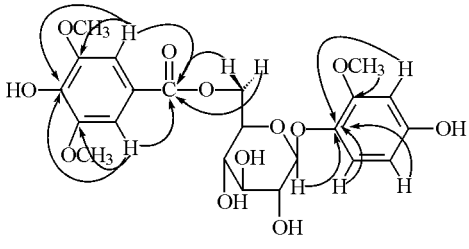

Figure 2 Key correlations in the HMBC spectrum of alpinoside A

Table 1 NMR data of alpinoside A [500 MHz for ^{1}H NMR and 125 MHz for ^{13}C NMR (CD_3OD)]

Position	^{1}H NMR (Hz)	^{13}C NMR
1	4.70 (d, 7.5)	104.75
2	3.45 (m)	75.52
3	3.45 (m)	78.23
4	3.35 (m)	72.56
5	3.64 (m)	76.12
6	4.37 (dd, 7.5, 12.0), 4.65 (dd, 2.0, 12.0)	65.75
1′	-	140.70
2′	-	152.70
3′	6.41 (d, 2.0)	102.25
4′	-	155.52
5′	6.05 (dd, 2.0, 8.5)	107.88
6′	6.89 (d, 8.5)	121.09
1″	-	121.79
2″	7.31 (s)	108.79
3″	-	149.43
4″	-	141.33
5″	-	149.43
6″	7.31 (s)	108.79
7″	-	168.34
2′-OCH_3	3.85 (6H, s)	57.37
3″, 5″-OCH_3	3.76 (3H, s)	56.98

The connections of the glucose unit and two benzyl moieties were established based on the corrections between the anomeric proton signal (δ 4.70) and C-1′ signal (δ 140.70) and between the carbonyl signal (δ 168.34) and H-6 (δ 6.37 and 6.65) in the HMBC spectrum (Figure 2). Thus this compound was determined to be 4′-hydroxy-2′-methoxyphenol-β-*D*-{6-*O*-[4″-hydroxy-3″, 5″-dimethoxy (benzoate)]}-glucopyranoside, named as alpinoside A.

Experimental

Melting points were determined using a Fisher Johns apparatus and was uncorrected. UV spectra were measured on a Philips PYE Unican Pu8800 spectrophotometer. One- and two-dimensional NMR spectra were recorded on a Bruker ARX 400 spectrometer. The EIMS were obtained on a VG ZAB-2f mass spectrometer. Precoated Silica gel plates (Qingdao Haiyang Chem. Co.) were employed for TLC and HPTLC. For column chromatography, macro porous resin (Tianjing Nankai Chem. Co.), polyamids (Zhejiang Taizhou Chem. Co.) and Sephadex LH 20 (Pharmacia) were used.

The rhizomes of *A. officinarum* were collected in Guangdong Province of China in 2002 and identified by Prof. Shou-Quan Lin (Institute of Medical Plant Development). A voucher specimen is deposited in the

New Drug Research and Development Center of our Institute.

The dried rhizomes of *Alpinia officinarum* were extracted three times with 95% EtOH at room temperature. The extract was dried under reduced pressure to yield a residue of 2.2 kg, which was subsequently diluted with H_2O and partitioned with petroleum ether, $CHCl_3$, EtOAc and *n*-butanol separatively. The *n*-butanol part (200 g) was dissolved in water and subjected to column chromatography on macro porous resin (40 - 60 mesh) eluting with water first and then with 10%, 30%, 50% and 80% EtOH in order. The fraction eluted with 30% EtOH was chromatographied on polyamides (60 mesh) and eluted with a gradient of ethyl alcohol in water (1∶4 - 1∶1) to give 8 fractions. The resultant fraction 2 was purified with Sephadex LH-20 to give two compounds **I** (5 mg) and **II** (10 mg).

Identification

Compound I $C_{22}H_{26}O_{12}$, white crystals, $[\alpha]_D$ - 4.6°(MeOH), mp 154 - 156 ℃. UVλ_{max}^{MeOH} (log ε) nm: 279(4.33), 219(4.68), 206(4.69). HRCI-MS at *m/z* 482.144 6 (calcd. 482.142 4). ^{1}H NMR and ^{13}C NMR data were listed in Table 1.

Compound II $C_{10}H_{20}O_6$, colorless needles, mp 152 - 154 ℃. ^{1}H NMR(CD_3OD, 500 MHz) δ: 0.94 (3H, t, *J* = 7.5 Hz, 1-CH_3), 1.40(2H, m, 2-CH_2), 1.57(2H, m, 3-CH_2), 3.52(2H, m, 4-CH_2), 3.64(1H, d, *J* = 12.5 Hz, 2′-H), 3.72(5H, m, 1′, 4′, 6′-H), 3.83(1H, m, 5′-H), 3.89(1H, m, 3′-H); ^{13}C NMR (CD_3OD, 125 MHz) δ: 14.84(1-C), 21.01(2-C), 33.82(3-C), 62.13(4-C), 63.96(1′-C), 65.66(6′-C), 71.06(3′-C), 71.60(5′-C), 72.04(4′-C), 102.11(2′-C). These data were in well agreement with those in the literature[6].

References

[1] Li CJ, Chen D, He R. The development of diarylheptanoids in *Alpinia officinarum* [J]. Lishizhen Med Mater Med Res (时珍国医国药), 2000, 11: 367 - 368.

[2] Shin D, Kinoshita K, Koyama K, et al. Antiemetic principles of *Alpinia officinarum* [J]. J Nat Prod, 2002, 65: 1315.

[3] Jian S, Huyi Z, Bo X, et al. The antioxidative constituents of rhizomes of *Alpinia offocinarum* [J]. Nat Prod Res Dev (天然产物研究与开发), 1997, 10: 33 - 36.

[4] Masuda T, Mizuguchi S, Tanaka T, et al. Isolation and structure determination of new antioxidative ferulic acid glucoside easters from the rhizome of *Alpinia spciosa*, a zingiberaceae plant used in okinawan food culture [J]. J Agric Food Chem, 2000, 48: 1479 - 1484.

[5] Ly TN, Yamauchi R, Shimoyamada M, et al. Isolation and structural elucidation of some glycosides from the rhizomes of smaller galanga [J]. J Agric Food Chem, 2002, 50: 4919 - 4924.

[6] Deng YR, He L, Li WQ, et al. Studies on chemical constituents in herbs of *Lamium maculatum* L. var Kansuense [J]. J Chin Mater Med (中国中药杂志), 2003, 28: 730 - 732.