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Isolation of β-Sitosterol Diarabinoside from Rhizomes of *Alpinia Galanga*

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Abstract*—Alpinia galanga* is rhizome, generally known as Greater galangal and is selected for isolation of newer constituents accountable for various therapeutic activities. Present study is intended to isolate glycoside from *Alpinia galanga* rhizomes. *Alpinia galanga* methanolic extract was column chromatograph and eluted with ethyl acetate-methanol (99:1) to isolate compound β-Sitosterol Diarabinoside. Herein, the isolation and structural elucidation of new compound is described. Chemical investigation of methanolic extract of rhizomes of *Alpinia galanga* furnished a new compound β-Sitosterol Diarabinoside. The IR, NMR and MASS investigations of isolated compound confirmed its structure as β-Sitosterol Diarabinoside, which is isolated for the first time from a medicinal plant or any synthetic source.

Keywords*—Alpinia galanga*, methanolic extract, β-Sitosterol Diarabinoside.

I. Introduction

ALPINIA galanga is a rhizomatous root stocks belongs to family Zingiberaceae and commonly known as Greater galangal, Kulingen [1]. Traditionally this plant is used as stomachic, rheumatic pain, antiemetic, antiulcerative, anti-dementia [2-7]. *Alpinia galanga* is known to possess antimicrobial, antioxidant, antifungal, anti-inflammatory, immuno stimulant, anti-cancer, and gastro protective activities [8-11]. It is also reported to use in treatment of AIDS [12]. This plant reported to contain various constituents such as 1, 1, 8-acetoxycineoles, 1'-Acetoxychavicol acetate, Galango galloside, Galango flavonoid β-Sitosterol, diglucoside, β-Sitsteryl Arabinoside [13-15]. The present study contributes to the ongoing investigations on *Alpinia galanga* plant for novel constituents with potent bioactivities. Herein, the isolation and structural elucidation of new compound is described.

II. Material and Methods

A. General

Melting point was determined in open capillary and is uncorrected. IR spectrum was recorded using KBR pellets, on Jasco FTIR-550 spectrophotometer. ^{1}H NMR and ^{13}C NMR spectra were recorded on Bruker DPX 300 Hz NMR spectrometers in $CDCl_3$ or DMSO-*d6* with TMS as internal standard. The Mass spectrum was generated on FAB-JEOL-MS 303 system. For column chromatography silica gel(100-200 mesh; Hi-Media) was used. The purity of isolated compound was determined by TLC aluminium sheets –Silica gel 60 F254 (0.2 mm).

B. Plant

The dried rhizomes of *Alpinia galanga* (Zingiberaceae) were collected form the province of Pusad, Maharashtra and were identified by Prof. Anjula Pandey, Taxonomist, National bureau of plant genetic resources, PUSA, New Delhi. A voucher specimen No. EP-542 is deposited in the Natural Medicine Research Centre, PUSA, New Delhi.

C. Extraction and Isolation

In the continuation of the work done on isolation of constituents from *Alpinia galanga* [16], the air-dried and powdered rhizome of *Alpinia galanga* (3000 g) was defatted with petroleum ether, and successively extracted with methanol using Soxhlet apparatus. The methanolic extract was evaporated to give a dark brown solid (35 g), which was further subjected to Si-gel column chromatography (100–120 mesh) and gradient elution EtOAc–MeOH (99:1) to give compound AG 6, β-Sitosterol Diarabinoside (346 mg).

III. Results

Compound AG 6, β-Sitosterol Diarabinoside is a pale yellow crystalline powder; mp. 182°C-185°C, is uncorrected. IR (KBr) spectrum of compound AG 6, exhibited bands at

TABLE I
^{1}H AND ^{13}C NMR SPECTROSCOPIC DATA FOR COMPOUND AG-6

	^{1}H NMR		^{13}CNMR
Position	**Alpha**	**Beta**	
1	1.37 m	2.39 m	36.83
2	1.94 m	1.82 m	29.16
3	3.63 brm (w 1/218.5)	---	73.47
4	2.89 d	2.92 d (7.9)	41.05
5	---	---	140.45
6	5.33	---	121.20
7	1.59	2.34 dd (13.8,5.5)	29.25
8	---	1.78 m	31.41
9	1.59 m	---	49.61

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10	---	---	36.22
11	2.01 m	1.45 m	20.60
12	1.13 m	1.81 m	37.96
13	---	---	40.03
14	1.16 m	---	56.18
15	1.13 m	1.50 m	23.87
16	1.79 m	1.48 m	27.79
17	1.41 m	---	55.43
18	0.64 brs	---	11.75
19	0.95 brs	---	19.71
20	---	2.12 m	35.71
21	0.91 d (6.0)	---	18.62
22	1.50 m	1.16 m	33.35
23	1.23 brs	1.23 brs	25.46
24	1.20 m	1.23 brs	45.15
25	1.50 m	---	28.72
26	0.82 d (6.0)	---	19.09
27	0.80 d (6.1)	---	18.95
28	1.16 m	1.55	22.62
29	0.78 d (6.3)	---	11.16
1’	5.19 d (7.1)	---	100.78
2’	4.35 m	---	88.23
3’	3.98 m	---	72.42
4’	4.25 m	---	81.29
5’	3.04 d (9.3)	3.01 d (9.3)	61.10
1’’	4.89 d (6.9)	---	100.78
2’’	4.20 m	---	76.75
3’’	3.85 m	---	70.12
4’’	4.21 m	---	67.74
5’’	3.12 d (8.4)	3.09 d (8.4)	60.76
1’’’	---	---	171.03
2’’’	2.54 d (11.7)	2.50 d (11.7)	59.24
3’’’	1.50 brs	1.45 brs	38.13
4’’’	1.23 brs	1.23 brs	29.25
5’’’	1.23 brs	1.23 brs	29.15
6’’’	1.23 brs	1.23 brs	28.72
7’’’	1.23	1.23 brs	28.72
8’’’	0.84 t (6.1)	---	18.62

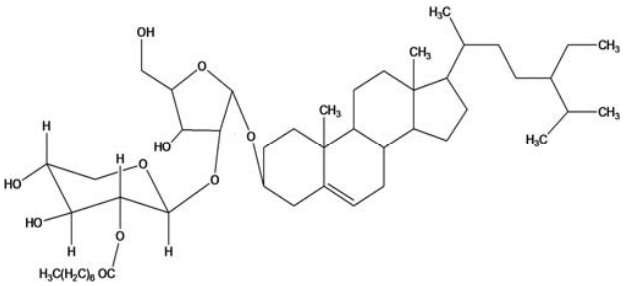
3416, 3355, 3260 cm^{-1}. The positive FAB-MS exhibited various ionic peaks at m/z 391 $[C_{18}H_{31}O_9]^+$, 127 $[CO\ (CH_2)_6\ CH_3]^+$, 143 $[OOC\ (CH_2)_6\ CH_3]^+$, 264 $[391\text{-}CO(CH_2)_6CH_3]^+$, and 413 $[M\text{-}C_{18}H_{31}O_9]^+$. ^1H and ^{13}C NMR spectroscopic data is given in Table I.

IV. Discussion

Compound AG 6, named β-sitosterol diarabinoside, was deduced to have molecular formula from its positive FAB mass spectrum at m/z 804 corresponding to a sterol diglycosyl ester, ($C_{47}H_{80}O_{10}$).

The ^{1}HNMR spectrum of AG 6 displayed signals for vinylic H-6 proton at δ 5.33 (2H,d, J=5.3 Hz), α-oriented carbinol H-3 proton at δ 3.63 (m, ,18.5 Hz,), secondary C-21, C-26, C-27 and primary C-21 methyl protons at δ 0.91 (J=6.0), δ 0.82 (J=6.0 Hz), δ 0.80 (J=6.1 Hz) and δ 0.78 (J=6.3 Hz). The 1H NMR spectrum of AG 6, for tertiary methyl protons at δ 0.64 (3H, m, C-18), δ 0.95 (3H, m,C-19), anomeric protons δ 5.19 (H, d H-1’,) and δ 4.89 (H, d, H-1’’) oxygenated methylene protons of the sugar moieties at δ 3.04 (H, d, J=9.3 Hz), δ 3.01 (H, d, J=9.3 Hz) and δ 3.12 ((H, d, J=8.4 Hz) and δ 3.09 ((H, d, J=8.4 Hz).

The ^{13}C NMR spectrum data of AG 6, exhibited important signals for vinylic carbons at δ 140.45 (C-5) and δ 121.20 (C-6), ester carbons at δ 171.03 (C-1’’’), anomeric carbons at δ 100.78 (C-1’, C-1’’), and methyl carbons at δ 11.75 (C-18), δ 19.71 (C-19), δ 18.62 (C-21), δ 18.95 (C-27), δ 19.09 (C-26), δ 11.26 (C-29) and δ 18.62 (C-8’’’). The appearance of C-2’ carbinol carbon in the deshielded region at δ 88.23 supported the attachment of the second sugar moiety at C-2’. The C-2’’ signal appearing at δ 76.75 indicated the location of the ester linkage at this carbon. The existence of one sugar carbon signed at δ 89.29 indicated the presence of arabinofuranose conformation of one the sugar residue.



β-Sitosterol Diarabinoside

V. Conclusion

The IR, NMR and MASS investigations of isolated compound AG 6, deduced and confirmed the structure as β-Sitosterol Diarabinoside.

This compound is isolated for the first time from the medicinal plant of *Alpinia galanga*.

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