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Galangoisoflavonoid Isolated from Rhizomes of *Alpinia Galanga*

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ABSTRACT

Galangoisoflavonoside was isolated from the rhizomes of *Alpinia galanga*. Methanolic extract of Alpinia galanga was subjected to column chromatography and eluted with ethyl acetate-methanol (9:1) to yield compound (AG 12) Galangoisoflavonoside. The structure of newer compound was elucidated by various spectral techniques (UV, IR, ^{1}HNMR, ^{13}CNMR, and MS). Chemical investigation of the methanolic extract of the rhizomes of *Alpinia galanga* furnished a new flavonoid (AG 12) Galangoisoflavonoside. Isolation of compound (AG 12) Galangoisoflavonoside in *Alpinia galanga* rhizomes is being reported for the first time.

Keywords: *Alpinia galanga,* Galangoisoflavonoside, methanolic extract

INTRODUCTION

Alpinia galanga is a perennial herb with rhizomatous root stocks and tall leafy stems belonging Zingiberaceae family, commonly known as greater galangal (1–2). This plant is well known for its richness in essential oils such as cineole, methyl cinnamate, myrecene, and methyl eugenol. This plant is also reported to contain various flavones like galangin, alpinin, kampferide and 3-dioxy-4-methoxy flavone (3–4). Alpinia galanga is known to possess antimicrobial activity, antioxidant activity, antifungal activity, anti-cancer activity, and gastroprotective activity (5–7). Present paper reports the isolation of flavone glycoside and structural determination evidences by means of various spectroscopic methods like UV, IR, NMR and Mass.

MATERIALS AND METHODS

General

Melting point was determined in open capillary and is uncorrected. IR spectra were recorded using KBR pellets, recorded on Jasco FTIR-550 spectrophotometer. IH NMR and ^{13}C NMR spectra were recorded on Bruker DPX 300 Hz and Mass spectra on FAB-JEOL-MS 303 system. Purity of isolated compound was checked by TLC aluminium sheets —Silica gel 60 F254 (0.2 mm).

Plant

The dried rhizomes of Alpinia galanga (Zingiberaceae), collected in Pusad province of India 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 Center of this Institute.

Extraction and isolation

Dried, ground 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 yield a dark brown solid (35g), which was subjected to Si-gel column chromatography

(100—120mesh) eluted with, EtOAc—MeOH (9:1) to give compound AG 12 (360mg).

RESULTS

The methanolic extract was column chromatographed over silica gel using EtOAc—MeOH (9:1) as eluants to yield Compound AG 12 as galangoflavonoside, was obtained as a pale yellow crystalline mass. It responded positively to the tests of flavonoid glycosides. Compound AG 12 showed Rf value of 0.57 in EtOAc—MeOH (99:1) solvent system. Its Melting point was determined by open capillary method and it was recorded as mp. 188 °C–190°C, is uncorrected. IR bands 3416, 3355, 3260, 2924, 1725, 1651, 1559, 1516 and 1040 $cm^{-1.}$ Positive FAB-MS m/z 1742 $(M)^+$ ($C_{17}H_{114}O_{14}$), 648 (12.3), 486 (10.4), 397 (31.6), 324 (11.5), 281 (25.9), 265 (20.6), 184 (16.3), 163 (32.2), 133 (65.1), 118 (62.7), 93 (67.8). ^{1}H-NMR and ^{13} C-NMR data: table-1.

DISCUSSION

Compound AG 12 designated as galangoflavonoside was obtained as pale yellow crystals. On the basis of the mass spectral fragmentation pattern, its molecular formula was established as $C_{59}H_{114}O_{14}$. The characteristic UV spectrum UV λ_{max}^{MeOH}: 257 nm (log ε 5.3), $\lambda_{max}^{MeOH + NaOAc}$: 256 nm, $\lambda_{max}^{MeOH1 + NaOme}$: 250 nm, $\lambda_{max}^{MeOH + AlCl_3}$: 269 nm suggesting flavone nature of the molecule. Its IR spectrum showed characteristic absorption bands for hydroxyl groups (3416, 3355, 3260 cm^{-1}) and ester group (1725 cm^{-1}).

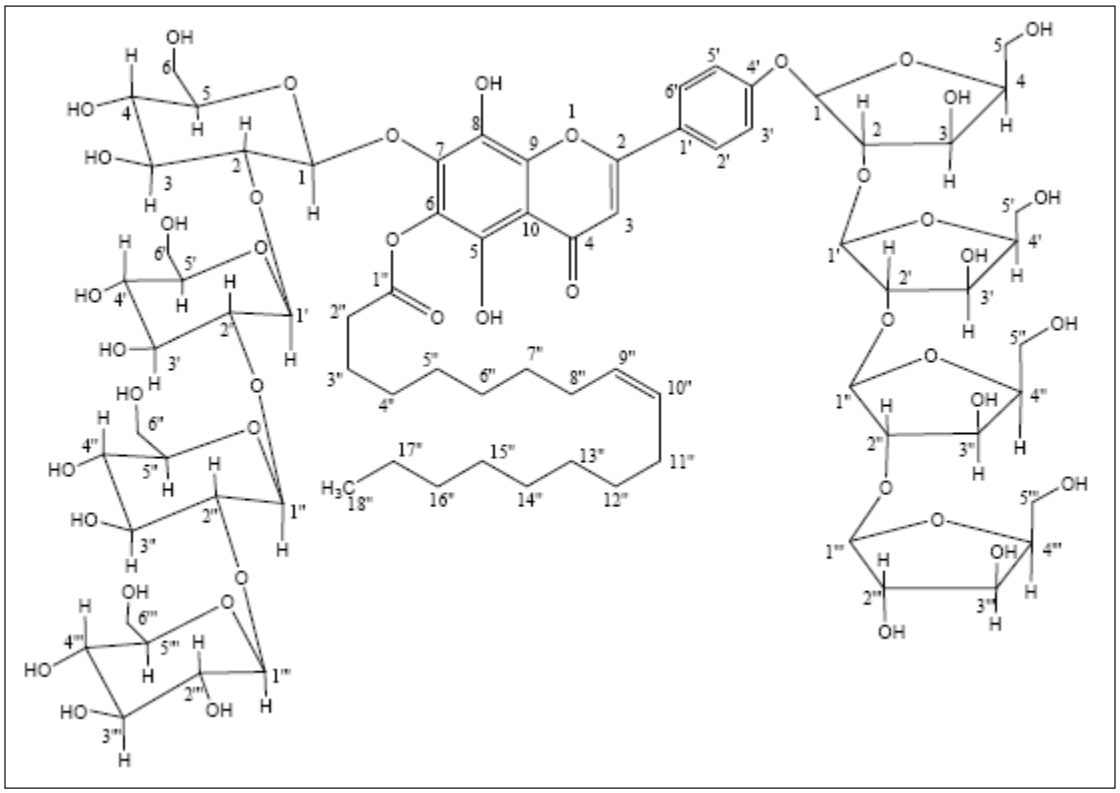
The ^{1}HNMR spectrum of compound AG 12, showed the signals at δ 7.13 (br, H-2), δ 7.03 (m, H-2') and δ 6.70 (m, H-6'), 6.50 (m, H-3'), 6.50(m, H-5'), 5.31,(Vinylic

Table 1: ^{1}H and ^{13}C NMR values of Galangoisoflavonoside (compound 1)

	^{1}HNMR	^{13}CNMR		^{1}HNMR	^{13}CNMR
1	……..	……..	G-1'	4.98 brs,	104.42
2	7.13 brs	151.81	G-2'	3.87 m	72.53
3	……..	121.26	G-3'	3.59	69.33
4	……..	174.52	G-4'	3.47 m	66.39
5	……..	160.15	G-5'	4.25 m	75.82
6	……..	134.29	G-6'	6.13, 6.15	60.84
7	……..	166.43	G-1''	4.96 brs	104.23
8	……..	124.16	G-2''	3.87 m	72.53
9	……..	157.87	G-3''	3.59 m	69.15
10	……..	115.84	G-4''	3.47 m	66.25
1'	……..	123.93	G-5''	4.25 m	73.52
2'	7.03 m	130.15	G-6''	6.13,6.15	60.03
3'	6.50 m	115.34	G-1'''	4.96 brs	88.31
4'	…….	144.35	G-2'''	3.86 m	72.48
5'	6.50 m	115.84	G-3'''	3.59 m	69.85
6'	6.70 m	130.06	G-4'''	3.47 m	66.44
1''	……..	173.48	G-5'''	4.25 m	73.65
2''	2.51 brs,2.49 brs	55.59	G-6'''	6.13, 6.15	60.01
3''	1.47 m	33.69	A-1	5.01 brs	102.41
4''	1.22 brs	32.41	A-2	4.25 m	77.68
5''	1.22 brs	29.08	A-3	3.63 m	73.52
6''	1.22 brs	26.05	A-4	3.35 m	84.26
7''	…….	25.53	A-5	3.25, 3.19	63.13
8''	1.99 m	51.46	A-1'	5.00 brs	101.35
9''	5.31 m	124.21	A-2'	4.25 m	77.59
10''	5.29 m	115.	A-3'	3.63 m	73.61
11''	1.84 m	38.04	A-4'	3.33 m	82.61
12''	1.22 brs	33.73	A-5'	3.25,3.20	63.13
13''	1.22 brs	31.40	A-1''	5.00 brs	101.19
14''	1.22 brs	29.08	A-2''	4.23 m	76.81
15''	1.22 brs	29.08	A-3''	3.63 m	73.61
16''	1.22 brs	24.54	A-4''	3.32 m	81.35
17''	1.22 brs	22.17	A-5''	3.23, 320	63.19
18''	0.83 t (6.2)	13.99	A-1'''	5.00 brs	99.81
G-1	4.98 brs	106.51	A-2'''	4.23 m	76.75
G-2	3.09 m	72.53	A-3'''	3.63 m	3.52
G-3	3.59 m	69.29	A-4'''	3.30 m	81.94
G-4	3.35 m	66.44	A-5'''	3.23, 3.20	62.15
G-5	4.25 m	76.81			
G-6	6.13, 6.15	61.15			

Where G = Glucose unit and A = Arabinose unit

Coupling constants in Hertz are provided in parenthesis.



Compound AG 12: IUPAC NAME: 5,6,7,8,4'-pentahydroxy isoflavone-6-(9" octadecenoate)-7-[β-D-glucopyranosyl(G-2 →G-1')- β-D-glucopyranosyl(G-2' →G-1")- β-Dglucopyranosyl- (G-2" → G- 1"')- β-D-glucopyranosyl]4'-[β-L-arabinofuranosyl-(A-2 → A-1')- β-L-arabinofuranosyl-A-2 →A-1') β-L-arabinofuranosyl(A-2" →A-1"')- β-L-arabinofuranoside.

2H, m, H-9"), 5.29 (Vinylic 5H, m, H-10") and two (one proton) doublets for C-2" methylene protons adjacent to the ester linkage at δ 2.51 and δ 2.49. A three protons triplet at δ 0.88 (J=11.3 Hz) was accounted to C-18" terminal primary methyl protons. Signals appeared at δ 5.36 (2H, m) and δ 5.33 (2H, m) were accounted to δ C-9" and δ C-10" vinylic protons respectively. The remaining methylene protons appeared between δ 1.99–1.22. The anomeric protons resonated as multiplets between δ 5.00–4.98. The other sugar protons appeared in the range of δ 4.25–3.13. The signals for flavone ring carbons appeared between δ 166.52–103.58. The signals for other sugar carbons appeared in the range of δ 82.81 – 60.03. Acid hydrolysis of compound AG 12 yielded oleic acid, D-glucose, arabinose and the isoflavone aglycone. On the basis of spectral data analysis and chemical reactions structure of compound AG 12 has been elucidated 5, 6, 7, 8, 4'-pentahydroxy isoflavone-6-(9"octadecenoate)-7-[β-D-glucopyranosyl-(G-2→G-1')-β-D-glucopyranosyl-(G-2' → G-1")-β-D-glucopyranosyl-(G-2"→G-1"')-β-D-glucopyranosyl]4'-[β-L-arabinofuranosyl-(A-2→A-1')-β-L-arabinofuranosyl-(A-2'→A-1")β-L-arabinofuranosyl (A-2' → A-1"')- β-L-arabinofuranoside

CONCLUSION

The present study deals with isolation and structural elucidation of a newer flavone compound AG 12 designated galangoflavonoside for the first time. After observing the UV, IR, ^{1}H-NMR, ^{13}CNMR and MASS data, also chemical test results for compound AG 12, it is concluded that the elucidated structure is in full agreement with all analytical data. Further pharmacological investigations are in progress to study biological activity of isolated compounds

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