Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

J P P Journal of Pharmacognosy and Phytochemistry

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2015; 4(2): 05-07
Received: 11-05-2015
Accepted: 12-06-2015

N.M. Mofiz Uddin Khan
Lecturer,
Department of Chemistry
Dhaka University of Engineering and Technology (DUET),
Gazipur-1700, Bangladesh.

Md. Sagar Hossain
Department of Applied Chemistry and Chemical Engineering, Faculty of Engineering, University of Dhaka, Dhaka-1000,
Bangladesh.

Correspondence:
N. M. Mofiz Uddin Khan
Lecturer,
Department of Chemistry
Dhaka University of Engineering and Technology (DUET),
Gazipur-1700, Bangladesh.

Scopoletin and β-sitosterol glucoside from roots of *Ipomoea digitata*

N.M. Mofiz Uddin Khan, Md. Sagar Hossain

Abstract
Plants are ancient source of medicine due to presence of bioactive molecules of various compounds in its different parts. Two compounds were isolated from ethanol extract of the roots of *Ipomoea digitata*. The compounds are Scopoletin (7-hydroxy-6-methoxycoumarin) and β-sitosterol glucoside. The structures were elucidated by spectroscopic analysis (^{1}H NMR & ^{13}C NMR). Though the compounds are known natural products, scopoletin is the first report of its occurrence from the plant *Ipomoea digitata*. The compound scopoletin is very much important for its medicinal value.

Keywords: Bioactive, Scopoletin, β-sitosterol glucoside, *Ipomoea digitata*, medicinal, natural product.

1. Introduction
Ipomoea digitata (local name: Bhui Chapa) is a member of family Convolvulaceae and called as Bilai-kand, Bhui-khola, Ksheervidari, Payasvinee, Bhumi-kumra, Bhumi-kushmanda in various languages [1]. From website of Wikipedia it was found that of about 60 genera and 1650 species are distributed in both tropical and temperate regions of the world.
Medicinal value of the plant is enormous. Its tuber powder (administration of 3 g) significantly showed antihypertensive potential which also increased fibrinolytic activity and total antioxidant status with a significant reduction in serum total cholesterol, LDL cholesterol and atherogenic index [1]. The tuberous root is used for the treatment of hypoglycemic, hypolipidemic, for debility, to increase secretion of milk, poor digestion, tuberculosis, enlarged liver etc. It was also found to have alterative, aphrodisiac, cholagogue, demulcent, diuretic, rejuvenative actions [2].
From the web search it was found that an Ayurvedic drug company Himalaya reported that Aligator Yam (*Ipomoea digitata*) has been traditionally used as a medicine in India and parts of Southeast Asia. In Ethno pharmacology of medicinal plants: Asia and the Pacific, author Christophe Wiart writes that in Cambodia, Laos and Vietnam, the tubers are used to prevent obesity and regularize menstruation. The author further emphasizes that the plant should be studied for its potential as a nerve stimulator. In India, Aligator Yam is used as a general tonic, to treat diseases of the spleen and liver and prevent fat accumulation in the body. They also reported the therapeutic constituents that are: the beta-sitosterol which is an antioxidant and ergonovine, an alkaloid, used to stop menstrual bleeding [3].
The compound scopoletin that was found in our study has multidimensional medicinal activity. It showed a significant result to reduce blood glucose level and lipid level [4, 5]. Scopoletin induced a marked time and concentration dependent inhibition of PC_3 cell proliferation. It reduced the protein content and decreased the ACP level in PC_3 cell in a concentration dependent manner and showed typical morphologic changes of apoptosis [6]. Both compounds beta-sitosterol and scopoletin have antioxidant activity [3, 7]. Literature review showed that the chemical constituents obtained from tuberous root of the plant are trierpenid, coumarin, octadecyl(E)-p-coumarate, beta-sitosterol, taraxerol, t-cinnamic acid [undecyl(E)-3-(4-hydroxyphenyl)-2-propenoate], an unknown coumarin (5-hydroxy-7-methoxy coumarin) and a lignan type resin glycoside [8-10].

2. Materials and methods
2.1. General experimental procedure
The ^{1}H and ^{13}CNMR spectra were recorded by a Bruker AMX-400 (400 MHz) instrument using $CDCl_3$ and the chemical shift was reported in ppm with respect to TMS or residual non deuterated solvent signals.

2.2. Plant material processing
The plant sample was collected from Natore district, northern part of Bangladesh and identified by taxonomist. 3.30 kg of fresh tuberous roots of the plant (*Ipomoea digitata*) were cut into small pieces and sun-dried and then, dried in an oven at reduced temperature (≤ 50 ^{0}C).

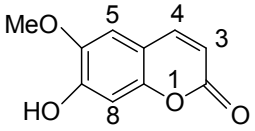
2.3. Extraction and isolation
The plant sample was soaked into four (4L) liters aqueous ethanol (80%) for three (03) days at room temperature for cold extraction and repeated the extraction four times. The crude extract was dried by rotavapor and desiccators. Dried crude aqueous ethanol extract was fractionated individually by n-hexane and water with the help of partition method solvent extraction. Then dried water soluble part was fractionated by dichloromethane (DCM) in the same process. DCM extract (5 g) was subjected to column chromatography. Single compounds were found from fraction F_9 (Compound 1), eluted with mixture of DCM and methanol (93:07) and F_{14} (Compound 2), eluted with mixture of DCM and methanol (85:15). The compound 1 was yellowish crystalline solid that exhibited blue fluorescence under UV light at 365nm. The compound 2 was white crystalline solid with boiling point 288-289 ^{0}C which turned pink on treatment with ceric sulfate indicating that it might be steroidal type compound.

2.4. Characterize Properties of isolated compounds spectrophotometrically

Scopoletin (1): Yellowish crystalline solid, exhibit blue fluorescence under UV light; ^{1}H NMR (400 MHz $CDCl_3$): δ 3.88 (3H, s, OCH_3-6), 6.20 (1H, d, *J* = 9.2 Hz, H-3), 6.80 (1H, s), 6.90 (1H, s), and 7.70 (1H, d, *J* = 9.2 Hz, H-4); ^{13}C NMR δ 162.60 (C-2), 108.10 (C-3), 144.20 (C-4), 111.60 (C-5), 145.30 (C-6), 151.00 (C-7), 102.90 (C-8), 149.80 (C-9), 110.90 (C-10) and 55.80 (O-CH_3).

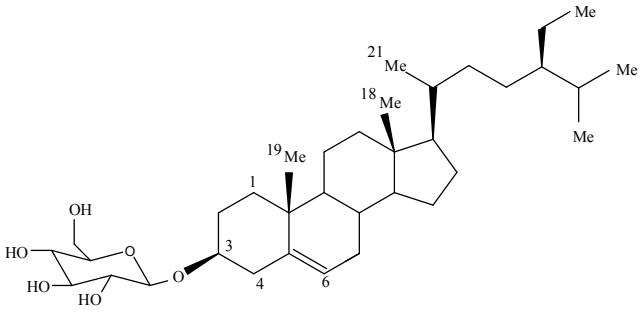
β-sitosterol glucoside (2): White crystalline solid, turned into pink color with ceric sulfate; ^{1}H NMR (400 MHz $CDCl_3$): δ 0.58 (3H, s, H_3-18), 0.68 (3H, d, *J* = 8, H_3-29), 0.71 (3H, d, *J* = 8, H_3-27), 0.75 (3H, d, *J* = 7.2, H_3-26), 0.83 (3H, d, *J* = 5.6, H_3-21), 0.90 (3H, s, H_3-19), 3.66 (1H, m, H-3) and 5.26 (1H, bd-s, H-6); ^{13}C NMR δ 37.50 (C-1), 29.44 (C-2), 78.96 (C-3), 42.13 (C-4), 55.80 (C-5), 122.1 (C-6), 39.20 (C-7), 31.70 (C-8), 50.01 (C-9), 35.93 (C-10), 20.85 (C-11), 39.56 (C-12), 45.68 (C-13), 55.87 (C-14), 25.9 (C-15), 28.00 (C-16), 56.56 (C-17), 11.66 (C-18), 18.73 (C-19), 36.51 (C-20), 19.03 (C-21), 38.49 (C-22), 28.99 (C-23), 49.30 (C-24), 24.05 (C-25), 18.50 (C-26), 19.48 (C-27), 22.86 (C-28), 11.58 (C-29), 100.92, 77.03, 76.70, 75.60, 70.0, 61.66 ppm (for six carbons of glucose unit).

3. Result and discussion
The ^{1}H NMR spectrum of compound 1 displayed signals characteristics of a 6,7-dioxygenated coumarin. The spectrum revealed two doublets at δ 6.20 ppm (^{1}H, *d, J=9.2 Hz)* and δ 7.70 ppm (^{1}H, *d, J=9.2 Hz)* characteristic of H-3 and H-4 protons respectively of the pyrone ring of a coumarin [11]. The presence of two aromatic proton singlets at δ 6.90 ppm and δ 6.80 ppm were attributable to H-5 and H-8 respectively. In this spectrum a three proton singlet at δ 3.88 ppm was assigned for protons of methoxy group at C-6. The ^{13}C NMR spectrum of compound 1 showed 10 carbon present in the compound and DEPT 135 spectra indicated that 4 out of 10 carbons were attached to single proton. The ^{1}H NMR and DEPT 135 spectra of compound 1 revealed the presence of four methine carbons, one *O*-methyl group and one phenolic hydroxyl group. Finally, the structure of compound 1 was confirmed by comparing its - ^{1}H NMR and ^{13}C NMR data to those reported for the Scopoletin isolated from the plant *Macaranga gigantifolia* Merr [11, 12]. On this basis compound 1 was identified as Scopoletin (7-hydroxy-6-methoxy coumarin) and the structure is-



Scopoletin (7-Hydroxy-6-methoxycoumarin)

The ^{1}H NMR spectrum of compound 2 showed six spectra at δ 0.58, 0.68, 0.71, 0.75, 0.83 and 0.90 ppm for methyl hydrogen (-CH_3) at C-18, C-29, C-27, C-26, C-21, C-19 respectively. One proton at C-3 appeared as multiplet at 3.66 ppm and a doublet at 5.26 ppm was the characteristics of double bond in the ring in between quaternary carbon and methine carbon C-5 and C-6. ^{13}C NMR indicated that the compound 2 consists of 35 carbons in its structure with some non-characterized signals due to presence of slight impurities. From DEPT it is appeared that 29 carbons of beta-sitosterol part contained six methyl carbon (–CH_3), eleven methylene (-CH_2-) carbon, nine methine (=CH-) carbon and three quaternary (=C=) carbon. Methyl carbons are C-18, C-19, C-21, C-26, C-27, C-29 and appeared at 11.6, 18.73, 19.03, 18.50, 19.48, 11.58 ppm; methylene carbons are C-1, C-2, C-4, C-7, C-11, C-12, C-15, C-16, C-22, C-23, C-28 and appeared at 37.5, 29.44, 42.13, 39.2, 20.85, 39.56, 25.9, 28.0, 38.49, 28.99, 22.86 ppm; methine carbons are C-3, C-6, C-8, C-9, C-14, C-17, C-20, C-24, C-25 and appeared at 78.96, 122.1, 31.7, 50.01, 55.87, 56.56, 36.51, 49.3, 24.05 ppm and quaternary carbons are C-5, C-10, C-13 and appeared at 55.8, 35.93, 45.68 ppm respectively. The glucose unit contained six carbons of which oxygenated carbon C-1 appeared at 100.92 ppm and methylene carbon C-6 appeared at 61.66 ppm. The other four carbons of the glucose molecule were appeared at 70.01, 75.60, 76.7 and 77.03 ppm. ^{1}H NMR and ^{13}C NMR revealed the isolated compound as beta-sitosterol glucoside that was confirmed by the reported data [13]. The structure of the compound is shown below:



β-sitosterol glucoside

4. Conclusion
The above investigation elucidates that the plant has a great medicinal importance. Its tuberous root contained scopoletin and β-sitosterol glucoside those have antioxidant property. The plant can be used as medicine to control diabetics (blood glucose level), hypertension (systolic, diastolic and mean blood pressure), lipid profile (serum total cholesterol, LDL cholesterol level). Isolation and characterization of more new bioactive compounds would be our priority in the future investigation.

5. References

1. Jain V, Verma SK, Katewa SS. Therapeutic validation of Ipomoea digitata tuber (Ksheervidari) for its effect on cardio-vascular risk parameters. Indian J. of Tradi. Knowl. 2011; 10(4):617-623.
2. Chandira M, Jayakar B. Formulation and evaluation of herbal tablet containing *Ipomoea digitata* extract. Int. J. of Pharm. Scis. 2010; 3(1):101-110.
3. Aligator Yam, Milky Yam. http://www.himalayawellness.com/herbfinder/ipomoea-digitata.htm
4. Verma A, Dewangan P, Kesharwani D, Kela SP. Hypoglycemic and hypolipidemic activity of scopoletin (Caumarin derivative) in streptozotocin induced diabetic rats. Int. J. Pharm. Sci. 2013; 22(1):79-83.
5. Obasi SC, Njoku OU, Obidoa O. Effect of single oral doses of scopoletin and aflatoxin B1 on the clotting time, serum cholesterol and phospholipid levels of chicks. Indian J. Physiol Pharmacol. 1994; 38(2):89-94.
6. Xue-Li L, Liang Z, Xin-Lu F, Kai C, Bo-Chu Q. Effect of scopoletin on PC_3 cell proliferation and apoptosis. Acta pharmacol sin. 2001; 22(10):929-933.
7. Malik A, Kushnoor A, Sainil V, Singhao S, Kumar S, Yadav YC. *In vitro* antioxidant properties of scopoletin. J. of Chem. and Phar. Res. 2011; 3(3):659-665.
8. Hao-Fu D, Jiang X, Jun Z, Zhong-Tao D. The chemical constituents from roots of Ipomoea digitata. Acta metallurgica sinica 2000; 22(2):1-3.
9. Rao CBS, Suseela K, Subba RPV, Krishna GP, Subba RGV. Some Indian medicinal plants. Indian J. of Chem. 1984; 23(B):787-788.
10. Madhavi D, Rao BR, Sreenivas P, Krupadanam GLD, Rao PM, Reddy KJ *et al.* Isolation of secondary products from *Ipomoea digitata* a medicinally important plant. www.pharminfo.net. 20, September, 2010.
11. Darmawan A, Kosela S, Kardono LBS, Syah YM. Scopoletin, a coumarin derivative compound isolated from *Macaranga gigantifolia* Merr. J. of App. Pharm. Sci. 2012; 2(12):175-177.
12. Kurdekar RR, Hegde GR, Kulkarni MV, Mulgund GS. Isolation and characterization of scopoletin- an anticancerous compound from the bark of *Hymenodictyon obovatum* Wall. Int. J. of Pharm. and Phytopharm. Res. (accepted article).
13. Rahman SMM, Mukta ZA, Hossain MA. Isolation and characterization of β-sitosterol-D-glycoside from petroleum extract of the leaves of *Ocimum sanctum* L. Asian J. Food Ag-Ind. 2009; 2(1):39-43.