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Phytochemical Screening of Roots of *Achyranchus Aspera*

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Abstract

Phytochemical screening of the roots of *Achyranthus aspera* yield new xanthone glycosides and its structure characterized as 1, 8-dihydroxy-3, 7-dimethoxy xanthone-5-*O*-*β*-L-rhamnopyranoside by chromatographic, chemical and various spectral studies.

Keywords: Latjeera, *Achyranthus aspera*, medicinal properties, phytochemical.

Introduction

Achyranthus aspera belong to the family Amaranthaceae. Commonly known as Latjeera. Almost all of its parts are useful in treatment of vomiting, asthma, obesity, heart disease, piles, itching, dysentery, leprosy, blood disease, anti-allergic. Preliminary phytochemical revealed the presence of terpenoids, quinine, flavonoids, alkaloids, long chain compounds. The roots of *Achyranthus aspera* were extract with successive different organic solvents in increasing order of polarity resulted glycosides of xanthone **1**.

Result and Discussion

The air dried and crushed roots was extracted successively with hexane, benzene, chloroform and ethyl acetate in a soxhlet extractor. From benzene fraction on flash column chromatography compound **1** was isolated.

Compound **1** m.p. 275^0C, $C_{21}H_{22}O_{11}$ (M^+.450) was glycoside of xanthone which on hydrolysis gave aglycone **1a** and one sugar which was confirmed by comparison with authentic sample and co-paper chromatography with sample as L-rhamnose. Aglycone **1a** was shown to be xanthone by its characteristic colour reaction. It gave violet colour with magnesium and hydrochloric acid (Shinoda test) which was also confirmed by special analysis such as UV, IR, ^{1}HNMR and 13C-NMR.

The compound on acetylation with acetic anhydride and pyridine yielded diacetyl derivative showing the presence of two hydroxyl group and two quartet in ^{13}C-NMR at δ 55.3 (C-3) and 56.7 (C-7) confirms two methoxy group.

The ^{1}HNMR spectrum of compound **1** showed three peaks for aromatic proton at δ 6.20, 6.27, 6.58 which corresponded to 1, 3, 5, 7, 8- pentoxygenated xanthone. This compound was also

insoluble in aqueous sodium carbonate and with 10% alcoholic KOH solution and formamide, a dark red colour was developed, indicating the presence of free hydroxyl group at C-1 and C-8. It was further confirmed by strong peak at 1650 and 1670 cm^{-1} for doubly chelated and non-chelated carbonyl group respectively.

Aglycone **1a** $C_{15}H_{12}O_7$ (M^+ 304) m.p. 210 ^OC on acetylation gave triacetate. Hence one of the hydroxyl group out of three was involved in glycoside linkage with rhamnose sugar. Compound **1** showed red shift of 35nm in band 1 with NaOAc and 97 nm on treatment with HI. Confirming the presence of 1, 4 dihydroxy nature of xanthone, it also confirms that the sugar moiety must be attached at position 5 of xanthone.

The ^{1}HNMR spectrum of xanthone display signal for (i) one proton at δ 6.27(1H, s), 6.58(1H, s), 6.65 (1H, s) (ii) two methoxyl group at δ 3.81 (3H, s) and 3.85 (3H, s). Thus compound **1** is 1, 8-dihydroxy -3, 7-dimethoxy-5-*O*-*β*-L-rhamnopyranoside.

Experimental Section

The plant was collected from Kanpur Dehat, U.P. India, in 2017. TLC, soxhlet and flash column chromatography were carried out on silica gel G. IR spectra were run in form of KBr pellets. ^{1}HNMR spectra was recorded at 300 MHz in $CDCl_3$ using TMS as internal standard. Mass spectra was recorded at 70 eV.

The air-dried and finely crushed root (4.0 kg) was extracted with ethanol. The above concentrated dark brown ethanolic extract was poured into ice-cold water, whereby a coloured residue and water soluble portion was extracted with soxhlet increasing polarity of solvents.

The concentrated benzene fraction was loaded over a flash column and eluted with different solvents of measuring polarity on dilution with benzene: $CHCl_3$ (8:2 v/v) compound **1** (0.529) was isolated m. p. 275^{0}C, $C_{21}H_{22}O_{11}$ (found: C, 56.05; H, 5.00 Calculated for $C_{21}H_{22}O_{11}$:C, 56.00; H, 4.8;) homogenous on TLC , R_f 0.56, ν^{max} 3350 br (chelated hydroxyl) 1668, 1645, 1615 and 1585 cm^{-1}. (Chelated unsaturated carbonyl group indicated a xanthone skeleton), λ^{max} (MeOH) 237, 266, 310, 345, 400; +$AlCl_3$ 235, 275, 328, 389 nm. ^{1}H NMR ($CDCl_3$, 300MHz**)** δ 6.27(1H, s), 6.58(1H, s), 6.65 (1H, s), 3.81 (3H, s) ,3.85 (3H,s), 5.80 (1H, d, J = 6.6Hz), 4.70 (1H, dd, J=6.6 and 0.98 Hz), 3.72 (1H, dd, J=6.5 Hz), 3.61 (1H, m, J=6.5 Hz), 3.69 (1H, m, J=6.5 Hz), 1.14(3H, d).

Enzymatic Hydrolysis

The glycoside (0.1g) in 50% aqueous ethanol (20ml) and emulsion solution (10ml) prepared from almonds were added. The mixture was left at 40-45^{0}C for two hours and then at room temperature for four days. The solution was extract with ethyl acetate and the remaining hydrolysate was concentrated by co-paper chromatography gave single spot identical to authentic sample of rhamnose. (*Rf* 0.16, solvent system reagent (B: A: W) spray, AHP).

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From the above evidences the structure of compound **1** was identified as 1,8-dihydroxy-3,7-dimethoxy xanthone-5-*O*-*β*-L-rhamnopyranoside.

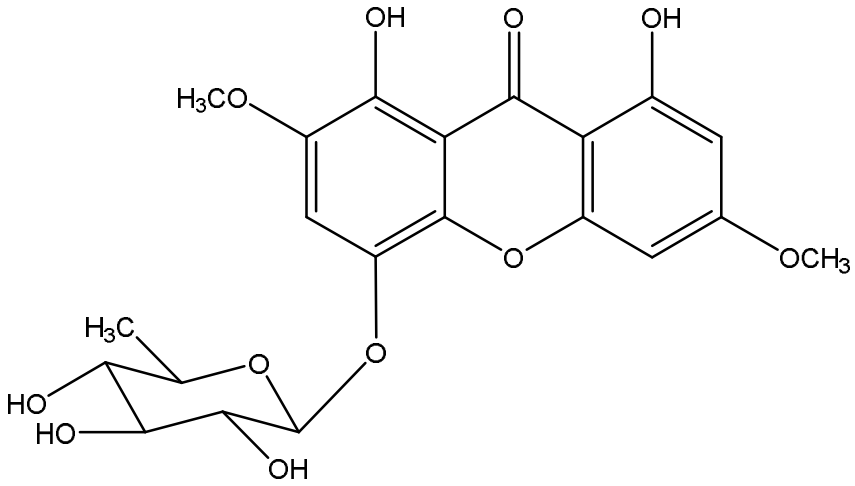

Table 1.^{13}C -NMR(δ) date of compounds 1, 1a

Carbon	1	1a
1	163.9(s)	163.5(s)
2	96.4(d)	95.4(d)
3	166.0(s)	162.0(s)
4	93.2(d)	93.1(d)
5	143.9(s)	133.5(s)
6	108.3(d)	107.3(d)
7	130.0(s)	129.0(s)
8	144.5(s)	143.5(s)
9	185.2(s)	184.5(s)
4a	156.0(s)	155.8(s)
9b	138.2(s)	137.8(s)
8a	115.5(s)	118.5(s)
10b	105.2(s)	104.2(s)
1′	109.4(d)	
2′	73.8(d)	
3′	71.4(d)	
4′	73.7(d)	
5′	74.2(d)	
6′	18.5(q)	
--OCH_3	55.3(q)	
--OCH_3	56.7(q)	

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