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ISOLATION AND STUDY OF XANTHIAZONE.7-HYDRXYMETHYL-8,8-DIMETHYL-4,8-DIHYDROXYBENZO(1,4)THIAZINE-3,5-DIONE-(2-O-CAFFEOYL)-B-D-GLUCOPYRANOSIDE FROM THE ROOTS OF XANTHIUM TRUMARIUM.(COMPOSITAE)

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ABSTRACT

The ethyl acetate fraction and methanol extract on repeated column chromatography over silica gel afforded the compound . The identification of the compound was made by concerted use of 1D and 2D-NMR, Mass, UV and IR spectroscopy.

PLANT OF XANTHIUM STRUMARIUM

INTRODUCTION :

Common name is "cocklebur"and chota gokhru in Hindi.It is an annual herb with 2 strong hooked weeks[1.] It is common weed found in India in Himalaya,it is also found in M.P. and Mharastra[2,3.] The genus Xanthium includes 25 species.The whole plants, especially roots and fruits, are used as medicine. According to Ayurveda, X.strumarium is cooling, laxative, fattening. anthelmintic, alexiteric. Tonic, digestive, antipyretic and improves appetite, voice, complexion and memory. It cures leucoderma, biliousness, epilepsy, salivation, fever and poisonous bites of insects.The plant Xanthium yields Xanthinin which acts as a plant growth regulator. Antibacterial activity of Xanthium has also been reported .Seeds yields semi-drying edible oil, which resembles sunflower oil and used in bladder infection, herpes and erysipelas[4,5,6]. Xanthium species have been reported to possess an anti-inflammatory and analgesic[7], anti-ulcerogenic activity[8] Anti-oxidant[9] activity.Its metabolite, 8-epi-xanthatin and its epoxide shows strong evidences of being anti-tumorous, they significantly inhibit the protiferatious of cultured human tumor cell lines. 8-epi-xanthatine acts by tarnesyltransferase inhibitory effect and also inhibit microtubule interfering agents. These inhibitious contribute to the anti- cancer activity of 8-epi-xanthatin (Kim et al. 2003).

Two sesquiterpene lactone glycosides and three kaurene glycosides closely related to carboxyatractyloside and atractyloside, together with the 3′, 4′ didesulphatedcarboxyatracty loside and 3′, 4′-didesulphatedatractyloside, have been isolated from the aerial parts of *Xanthium spinosum* [14] along with xanthanol, isoxanthanol and their C-4 epimers[16] Polyphenol 1,3,5-tri-*O*-caffeoylquinic acid, accompanied by 3,5- di-*O*-caffeoylquinic acid, has been isolated from the fruit of *Xanthium strumarium* [10]. Xanthanolide and bis-norxanthanolide has been isolated from *Xanthium cavanillesii* [15].The present investigation deals with the isolation and identification of Xanthiazone, Xanthiazone-(2-O-caffeoyl)-b-D-glucopyranoside from the ethyl acetate fraction (8.03 g) and methanol extract of whole plants of *Xanthium strumarium* on repeated column chromatography over Si-gel .

PLANT MATERIAL:

Roots of Xanthium strumarium were procured from pachmadi (M.P.) and authenticated by the Botany Department of this college.

EXTRACTION AND ISOLATION:

The air-dried and powderd roots of xanthium strumarium (2 kg) were exhaustively defatted with light petroleum ether (60-800). The petroleum free mass extracted with 90% ethanol. The ethanol extract was concentrated under reduced pressure and a suspension
of the residue was made with water, which was washed with diethyl ether for several times and then partitioned with CHCl3: H2O: MeOH (6:4:4) in a separatory funnel. The aqueous layer was concentrated under reduced pressure and then partitioned with ethyl acetate and 50% aqueous methanol. The ethyl acetate fraction was dried under reduced pressure to give
EtOAc extract (8.03 g). The aqueous methanol extract was concentrated under reduced pressure to give methanol extract (12.5 g). The EtOAc extract (5.0 g) was column chromatographed over Si-gel using gradient elution using CHCl3: MeOH with increasing
MeOH content afforded two fractions. Fraction was subjected to column chromatography on Si-gel using CH2Cl2-MeOH solvent with increasing MeOH contents afforded a
fraction. This fraction was subjects to preparative TLC on Si-gel 60 HPTLC (Merck) using developing system CHCl3: MeOH: NH3 (80:20:3) afforded compound (15 mg).
Compound was yellow crystalline solid (MeOH),m.p..187-189˚C. Elemental Analysis: C=55.59%, H=5.34%,1625, 1605, etc. 1H-NMR: (300 MHz, DMSOd6): d 3.51 (2H, *s*, H-2), 6.61 (1H, *s*, H-6), 1.39 (6H, *s*, H-9, H-10, 2CH3), 4.50 (1H, *d*, *J*=15.0 Hz, H-11), 4.68 (1H, *d*, *J*=15.0 Hz, H-11), 9.30 (1H, *s*, NH), 4.36 (1H, *d*, *J*=7.0 Hz, H-1'), 4.67 (1H, *m*, H-2'), 3.45 (1H, *m*, H-3'), 3.15 (1H, *m*, H-4'), 3.45 (1H, *m*, H-5'), 3.35 (1H, *m*, H-6'), 7.05 (1H, *brs*, H-2"), 6.78 (1H, *d*, *J*=8.0 Hz, H-5"), 7.01 (1H, *d*, *J*=8.0 Hz, H-6"), 7.49 (1H, *d*, *J*=15.0 Hz, H-7"), 6.26 (1H, *d*, *J*=15.0 Hz, H-8"). 13CNMR (75 MHz, DMSO-d6): d 28.85 (C-2), 162.27 (C-3; -C=O), 175.01 (C-5, -C=O), 121.79 (C-6), 164.09 (C-7), 41.47 (C-8), 26.75 (C-9), 26.34 (C-10), 65.72 (C-11), 141.01 (C-8a), 130.03 (C-4a), 101.92 (C-1'), 71.22 (C-2'), 73.55 (C-3'), 73.92 (C-4'), 74.22 (C-5'), 60.01 (C-6'), 125.34 (C-1"), 114.89 (C-2", 145.38 (C-3"), 148.76 (C-4"), 115.80 (C-5"), 121.22 (C-6"), 145.36 (C-7"), 113.73 (C-8''), 165.97(C-9'').

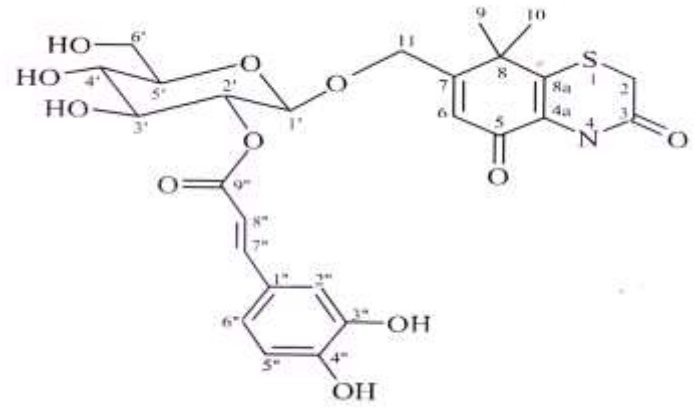
ACID HYDROLYSIS OF COMPOUND:

(10mg) was dissolved in 5% H2SO4 and refluxed on water bath for 3 hrs. The reaction mixture was cooled and poured on crushed ice and stand for 30 min. The precipitate was purified by re-crystallization from MeOH. The aglycone was identified as Xanthiazone by
comparison with authentic sample and the sugar was identified as b-D-glucose by paper chromatography.

RESULT AND DISCUSSION

The molecular weight of the compound wasfound to be 563 by positive-ion FAB mass spectrum which exhibited a molecular ion peak [M+H]+ at m/z 564 (calc. for C26H29O11NS) and [M+Na]+ peak at m/z 586 (calc. for C26H29O11NSNa). The IR spectrum displayed presence of an a,b-unsaturated carbonyl
Group at 1665 cm-1, another carbonyl group at 1625 and an oleiifnic absorption band at 1605 cm-1. The UV spectrum displayed an absorption maxima at 250,280 335 nm indicated presence of a carbonyl group, double bond and an aromatic ring in the molecule. The 1H-NMR spectrum of exhibited sixn signals in aromatic region, ten signals in aliphatic region and a singlet in the aromatic heterocyclic region. The 13C-NMR spectrum displayed twelve
Signals in aliphatic region and fourteen signals in thearomatic region. The DEPT spectrum showed presenceof eleven methine, three methylene, two methyl and ten quaternary

carbon atoms which accounts for twenty three protons. The one proton singlet at d 9.30
was assigned for a proton attached to N-atom ofheterocyclic ring. The remaining protons must present as hydroxyl groups. In the aliphatic region of 1H-NMR spectrum a six
protons singlet at d 1.39, and two protons singlet at d3.51, were assigned for two methyl groups attached at C-8 and a methylene group adjacent to carbonyl group of Xanthiazone moiety [10]. In addition to these signals, two doublets (J = 16.5 Hz), each for 1H at d
4.50 and 4.68 was assigned for gem protons of oxymethyl group attached C-7 of Xanthiazone moiety. The 1H-NMR spectrum also displayed presence of a
trans-disubstituted [d 7.49 (1H, *d*, *J*=15.0 Hz, H-7"), 6.26 (1H, *d*, *J*=15.0 Hz, H-8") and a tri-substituted [d 6.61 (1H, *s*, H-6)] double bond in the molecule. The 1H-NMR spectrum also displayed a broad singlet at d 7.05 and two A2B2 type *ortho*-coupled doublets (J =
8.0 Hz) indicated the presence of caffeoyl group in the molecule [11, 12]. A 1H doublet (J = 7.0) d 4.36 for anomeric proton along with other proton signals assignable to sugar moiety indicated presence of a bsugar in the molecule. On acid hydrolysis compound afforded a sugar which was confirmed as b-D-glucose by paper chromatography. In agreement with the
Above discussed 1H-NMR spectral data the ^{13}C-NMR spectrum displayed presence of a caffeoyl group [δ 125.34 (C-1"), 114.89 (C-2"), 145.38 (C-3"), 148.76 (C-4"), 115.80 (C-5"), 121.22 (C-6"), d 145.36 (C-7"), 113.73 (C-8") 165.97 (C-9")]. [7], ana,b-unsaturated carbonyl group at d 175.01 (C-5, -C=O), and the olefinic carbons at d 121.79 (C-6), 164.09 (C-7) of
Xanthiazone moiety. In addition to these the 13C-NMR spectrum displayed chemical shift of anomeric carbon of b-D-glucose at d 101.92 and other carbon resonances of sugar moiety. The downfield chemical shift of methylene carbon at d 65.72 (C-11) and that of (C-1') of glucose at d 101.92 suggested that the C- 1' of glucose is attached at methylene carbon of
Xanthiazone moiety. Furthermore the downfield chemical shift of C-2' carbon at d 71.22 suggested that the caffeoyl group is attached at the C-2' of glucose. On the basis of above discussed spectral data compound was characterized as 7-hydroxymethyl- 8,8-dimethyl-4,8-dihydroxybenzo[1,4] thiazine-3,5- dione-(2-O-caffeoyl)-b-D-glucopyranoside, which was
further confirmed by comparing data with those of reported data [13.]



STRUCTURE OF XANTHIAZONE.7-HYDRXYMETHYL-8, 8-DIMETHYL-4,8-DIHYDROXYBENZO(1,4),THIAZINE-3, 5-DIONE-(2-O-CAFFEOYL)-β-D-GLUCOPYRANOSIDE

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REFERENCES

[1] Agharkar S.P. Medicinal plants of Bombaypresidency. Scientific Publishers, Jodhpur (India).230 (1991).
[2] Oudhia P. Phyto-sociological studies of rainy season wasteland weeds with special reference to Parthenium hysterophorus L. in Raipur (India) district. Asian J. Microbiol. Biotech. Environ. Sci.3 (1-2) 89-92 (2001).
[3] Oudhia P. and Dixit A. Weeds in Ambimkapur region (Madhya Pradesh) and their traditional use.Weed News. 1(2) 19-21 (1994).
[4] Caius J.F. Medicinal and poisonous plants of India. Scientific Publishers, Jodhpur (India). 375-376 (1986).
[5] Oudhia P. and R.S. Tripathi. Possibilities ofutilisation of medicinal weeds to increase the Income of the farmers. In: Abstract. NationalSeminar on Medicinal Plant Resources Development, Gandhi Labour Institute,Ahmedabad (India), 4-5 Oct p. 3 (1998).
[6] Sastry T.C.S. and Kavathekar K. Y. Plants forreclamation of wastelands. Publications and Information Directorate, CSIR, New Delhi (India):421-422 (1990).
[7] Han T., Li H.-L., Zhang Q.-Y., Han P., Zheng H.-C., Rahman K. and Qin L.-P. Bioactivity-guided fractionation for anti-inflammatory and analgesic

Properties and constituents of Xanthium strumarium. Phytomedicine 14(12) 825-829 (2007).

[8] Laura S. F., Alejandra O.M. M., Graciela H. W.,Eduardo J. B., Oscar S. G., Lilian P. and Carlos E. T. Anti-ulcerogenic activity of xanthanolide sesquiterpenes from Xanthium cavanillesii. Journal of Ethnopharmacology. 100(3) 260-267(2005).

[9] Scherer R. and Godoy H. T. Antioxidant activity index (AAI) by DPPH method. Food Chemistry, 112(3) 654-658 (2009).

[10] Ageta I., Goto, S., Hatano T., Nishibe S., and Okuda T. 1,3,5-tri-O-caffeoylquinic acid, from of Xanthium strumarium. Phytochemistry, 33 508 (1993).

[11] Merfort I. Caffeoylquinic acids from flowersof Arnica montana and A. chamissonis. Phytochemistry 31 2111 (1992).

[12] Qin L., Han T., Li H., Zhang Q. and Zheng H. Anew thiazinedione from Xanthium strumarium.Fitoterapia 77 245 (2006).

[13] Paulo A., Gomes E.T., Steele J., Warhurst D.C. and Houghton P.J., Planta Medica 66 30-31(2000).

[14] Piacente S., Pizza C., Tommasi N. D., and Simone F. D. Sesquiterpene and diterpene glycosides from Xanthium spinosum. Phytochemistry 41(5), 1357-1360 (1996).

[15] Riscala E. C., Fortuna M. A., Catalán C. A.N.,Díaz J. G., and Herz W. Xanthanolide and bis **D.P.Pandey et al** /Int.J. ChemTech Res.2012,4(1) 271norxanthanolide from Xanthium cavanillesii.Phytochemistry 35(6) 1588-1589 (1994).

[16] Marco J.A., Sanz-Cervera J. F., Corral J., Carda M., and Jakupovic J. Xanthanolides from Xanthium. Absolute configuration of xanthanol, isoxanthanol and their C-4 epimers. Phytochemistry 34(6) 1569-1576 (1993).