A NOVEL TANNIN FROM *PROSOPIS JULIFLORA* ROOTS

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ABSTRACT

From the ethanolic extract of roots of *Prosopis juliflora* DC a novel tannin has been isolated and characterised as 1,3-glucose diester of 4,4′-dimethoxy 3,3′,5,5′-tetrahydroxy diphenic acid.

PROSOPIS juliflora DC (N.O. Caesalpinaceae) is well known for its economical and medicinal values[1-4]. The presence of polyphenols and tannins in this plant has been reported earlier[5]. Recently, the chemical constituents of its stem bark[6], pods[7-9] and roots[9-11] have been reported. In the present paper the chemistry of a novel tannin has been discussed.

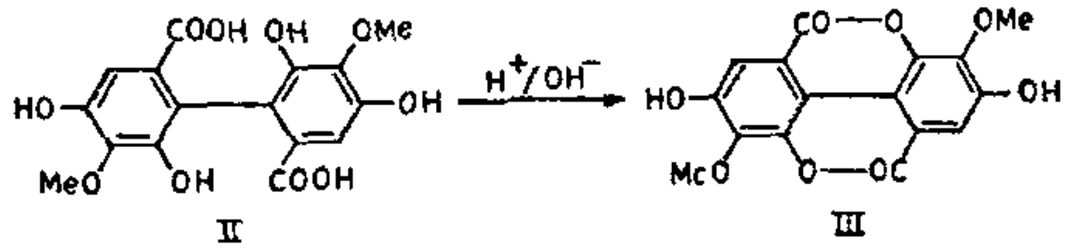
Air-dried roots of the plant were extracted with acetone and dried. The roots were then extracted with boiling ethanol. An amorphous deposit was obtained by keeping the concentrated ethanolic extract at 5° C for 7–8 days which could be separated by centrifugation at -10° C at 10000 rpm. It was crystallised to a chromatographically homogeneous buff-coloured semimicro crystals, m.p. 139° (d), $(\alpha)_D^{25}$ (pyridine) –29° and analysed for $C_{22}H_{22}O_{14}$.

It was found to be a non-reducing glycoside, very hygroscopic and sensitive to aerial oxidation. Dark green colour with alcoholic ferric chloride and yellow colour with alkali indicated its phenolic nature. The above tests combined with spectral data *i.e.*, λ_{max}^{EtOH} at 271 nm and definite ester band at 1740 cm^{-1} in IR spectrum[12] suggested that the compound may be a glycoside or a sugar ester of a phenolic acid. Strong peaks at 1180 and 3440 cm^{-1} in IR spectrum indicated the presence of methoxyl and hydroxyl groups in the glycoside. Alkoxyl estimation indicated the presence of two methoxyls per mole of the glycoside[13].

On acid hydrolysis the glycoside yielded an aglycone along with a sugar identified as D-glucose by direct comparison with an authentic sample and osazone preparation. The glycoside could also be hydrolysed with alkali, which further supported the presence of ester links in the compound[12].

The aglycone, $C_{16}H_{10}O_8$, m.p. 240° C was identified as 3,3′-di-O-methyl ellagic acid(II) by its absorption maximum at 274 nm; and superimposable IR spectrum which showed strong peaks at 3430, 1735, 1720 and 1180 cm^{-1}. A bathochromic shift of 28 nm with sodium ethylate and no shift with sodium acetate clearly indicated the presence of two methoxyl groups at 3,3′-position of ellagic acid[10,12]

The presence of ester linkages in the glycoside and subsequent formation of 3,3′-di-O-methyl ellagic acid (III) by alkali hydrolysis suggested that this aglycone is not present as such in the glycoside but rather has been formed as a result of cyclisation of the corresponding methyl ether of hexahydroxydiphenic acid, under the conditions of hydrolysis. It is well known that under these conditions hexahydroxydiphenic acid cyclises to give the corresponding lactone *i.e.*, ellagic acid[12]. Thus, it could be concluded that 4,4′-dimethoxy 3,3′,5, 5′-tetrahydroxy diphenic acid (III) is esterified through the bidental linkages with two hydroxyls in glucose moiety; one of these must be at the anomeric carbon atom, since the glycoside is non-reducing. (chart 1).



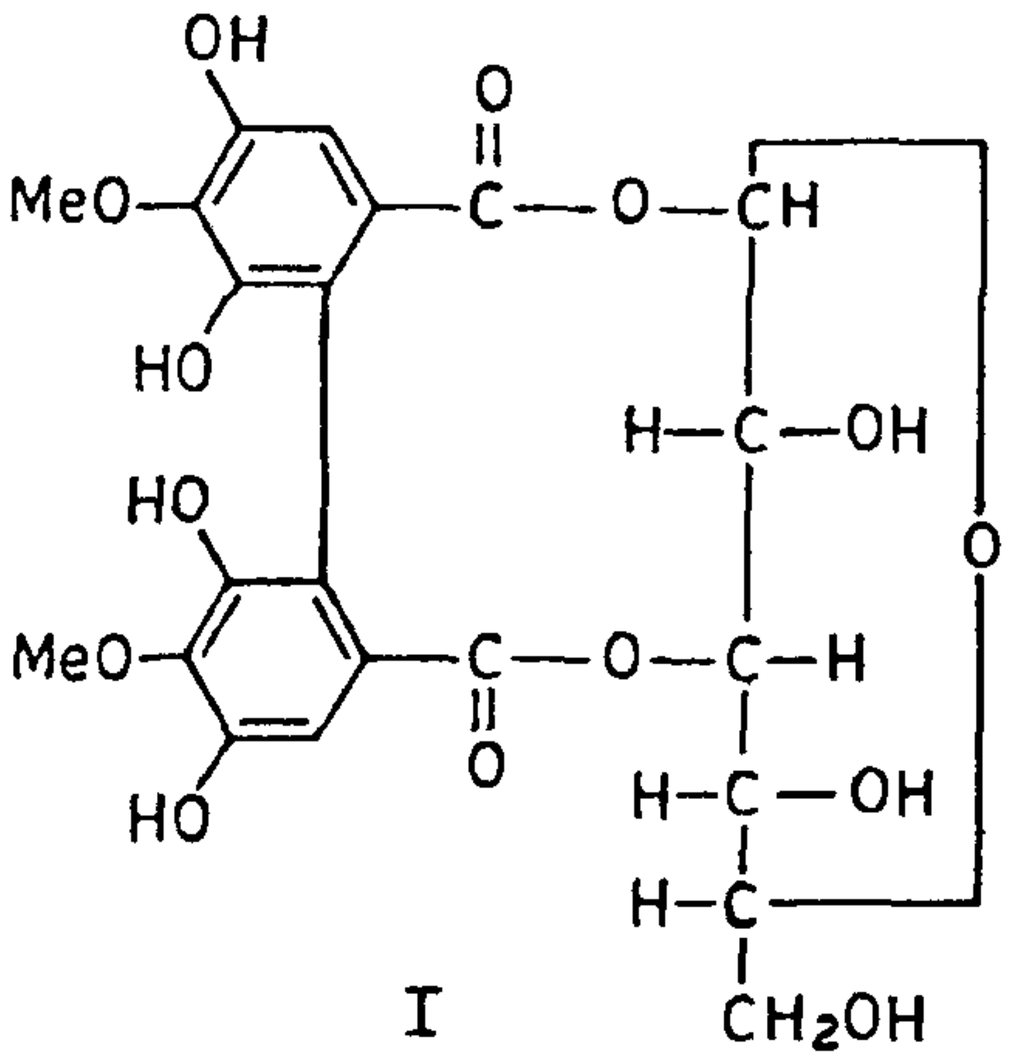
II III

Methylation of the glycoside with diazomethane followed by acid hydrolysis, yielded a pale yellow product, mp 238° (d). It was identified as hexamethoxydiphenic[10,12] acid by mmp 239° C (lit. 238–40° C) and superimposable IR spectrum, λ_{max}^{KBr} at 3000, 1680, 1570, 1460, 1410, 1320, 1180, 1128 and 1000 cm^{-1}.

Quantitative hydrolysis and subsequent sugar estimation[15] showed the presence of only one mole of glucose per mole of the glycoside. Out of the two ester links, one was decidedly at C-1 of glucose, as shown by its non-reducing nature. The position of the second ester link was decided by permethylating[16] the glycoside followed by acid hydrolysis when 2,4,6-tri-O-methyl glucose was identified by its R_G value[17]. This showed that hydroxyls at position C-1 and C-3 of glucose moiety are involved in esterification.

Hence a probable structure could be assigned to this sugar ester as 1,3-glucose diester of 4,4′-dimethoxy, 3,3′-5, 5′-tetrahydroxy diphenic acid I (chart 2).

This sugar ester can be classified as an ellagitannin as on hydrolysis it yielded an ellagic acid derivative.



I

This is the first case reported so far in nature of an ellagitannin in which hexahydroxydiphenyl unit is partially methylated.

EXPERIMENTAL

Plant material: Plant material was locally collected and identified by the Botanical Survey of India, Allahabad. m.ps. are uncorrected and recorded on electrically heated plate. Ascending type of chromatography was done except in sugars, solvents used are: (a) *n*-BuOH: HOAc: H_2O (4:1:5, v/v), (b) 10% HOAc and (c) *n*-BuOH : HOAc : H_2O (5:1:4 v/v).

EXTRACTION

Air-dried powdered and acetone extracted roots (3 kg) were extracted with EtOH (5×5 l) at reflux temperature. EtOH extract was concentrated under reduced pressure (150 ml) and kept at 5° for 7-8 days when a buff-coloured amorphous solid separated out. The solution was centrifuged at −10° C and 10,000 rpm for 50 min. The buff-coloured residue was washed with dry EtOH and crystallised from containing traces of pyridine as semi-micro crystals, m.p. 139°(*d*), α_D^{25}(pyridine) -29°, R_f 0.21 and 0.46 (PC, solvent; a and b respectively; spray: alcoholic $FeCl_3$).

Found: C, 51.06; H, 4.29; -OCH_3, 11.9, $C_{20}H_{16}O_{14}$ (-OCH_3)$_2$ requires 51.76; H, 4.3 and -OCH_3, 12.5%.
UV: υ^{ETOH}_{max}: 222, 275, 295 (sh) nm.
IR: υ^{KBr}_{max}: 3450, 2980, 1730, 1680, 1650, 1630, 1560, 1500, 1470, 1380, 1290, 1180, 1070, 840 and 750 cm^{-1}.

Acid hydrolysis

Compound I (0.01 g) was refluxed with aq H_2SO_4 (7%, 3 ml) for 2.5 hr solution cooled, poured in iced water (20 ml) extracted in Et_2O containing traces of pyridine, crystallised from aglycone:dioxane-light petrol as yellow crystals.

Concentrated hydrolysate was neutrallised with $BaCO_3$, chromatographed on PC, R_f0.18 (solvent:a, spray:aniline hydrogen phthalate).

Aglycone: m.p. 272° (lit. 274°), υ^{EtOH}_{max} nm: 274, +NaOEt: 302 (a bathochromic shift of 28 nm), +NaOAc: 274 nm. υ_{max} (KBr) 3430, 1735, 1720, 1180, 1040 and 940 cm^{-1}.

Alkali hydrolysis

Compound I (0.02 g) dissolved in 5% alc NaOH soln (10 ml) was kept at room temperature for 24 hr. It was acidified with glacial HOAc acid and continuously extracted with Et_2O containing pyridine, concentrated, crystallised from dry Me_2CO–Et_2O as yellow crystals, m.p. 273°, (lit. 275°) identical with the aglycone obtained an acid hydrolysis in all respects.

Quantitative acid hydrolysis

Compound I (0.1 g) was refluxed with aq. H_2SO_4 (70%, 3 ml) for 2.5 hr, cooled, poured onto ice cold water. Aglycone collected by the procedure discussed above and weighed directly. Hydrolysate, neutrallised with $BaCO_3$. made up to 25 ml and sugar estimated by the colorimetric method of Folin and Wu[15].
Found: 3,3′–di–O–methyl ellagic acid, 63.9 and reducing sugar 34%. $C_{22}H_{22}O_{14}$ requires 3,3′–di–O–methyl ellagic acid, 64.75 and reducing sugar 35.27%.

Methylation and hydrolysis

To the compound (I) (0.15 g) in dry Me_2CO (5 ml) excess of CH_2N_2 was added and kept for 24 hr in cold and excess of CH_2N_2 was removed, solvent, distilled off and resulting yellowish syrup crystallised from dry Me_2CO-Et_2O mixture.

This methylated compound was subjected to acid hydrolysis with aq H_2SO_4 (7%, 3 ml) as discussed above to collect pale yellow crystals of methylated aglycone, m.p. 238° (lit. 238–40°), IR υ_{max} (KBr): 3000, 1680, 1570, 1500, 1460, 1410, 1320, 1180, 1120 and 1000 cm^{-1}.

Permethylation

To the diazomethane methylated glycoside (0.1 g) dissolved in dry MeOH (20 ml), Ag_2O (1 g) was added,

CH_3I (3 ml) was added at 40° with stirring, in small lots. It was refluxed for 4 hr, cooled, extracted with $CHCl_3$ and the product obtained was subjected to the above procedure again till the methylation was complete. It was hydrolysed with aq. H_2SO_4 (7%, 3 ml) as discussed earlier. Aglycone:m.p. 237° (lit. 238–40°)[12], sugar: R_f 0.85 (PC, solvent: C, sprary: aniline hydrogen phthalate).

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QUINAZOLONES AND THEIR PSYCHOPHARMACOLOGICAL ACTIVITY

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ABSTRACT

Twenty new substituted quinazolones were synthesized and characterized. They were screened for various central nervous system activities. Some compounds have shown potent tranquillizing, anti-depressant and anti-convulsant activities.

INTRODUCTION

EARLIER studies from this laboratory have shown the potentiality of quinazolone moiety for central nervous system (CNS) activity[1]. The substitution of position three of quinazolone nucleus plays an important role in imparting CNS activity in quinazolones[2]. We have, therefore, incorporated different moieties *via* a phenyl bridge at position three to see the effect of these substituents on CNS activity.

EXPERIMENTAL

Melting points were determined in open capillary tubes and are uncorrected. TLC was carried out by employing silica gel G. Mass spectra were recorded on J. M. S. D. 300 focussing spectrometer with J.M.A. 2000 data. 5-Bromo and 5-iodo anthranilic acids were prepared by known methods[3,4].

2-methyl-3-(3'- acetyl aryl)-6-substituted 4 (3H) quinazolones

These were prepared by the fusion of substituted acetanthranils (0.01 mole) and m-amino acetophenone (0.01 mole). The resultant jelly-like mass was crystallized with ethanol. Quinazolones thus prepared are reported in table 1.