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New Compound Isolated from Stem Bark of the *Alstonia Scholaris* R. Br.

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ABSTRACT:

Two new compounds have been isolated from stem bark of plant *Alstonia Scholaris* using column chromatography from the chloroform fraction is namely Alscholaridine and its Structure was elucidated on the basis of spectral data.

Key Words: Alstonia Scholaris, Alscholide, Alscholaridine, Column chromatoraphy.

INTRODUCTION

Alstonia scholaris R. Br. Belong to family Apocynaceae and is known as "saptparni" in India is a medicinal, distributed throughout the tropical and subtropical region of the world. In Jaipur, Rajsthan (India) several species of *Alstonia* are being used by the Ayurvedic medicinal system[1,2]. α-amyrin acetate isolated from bark of *Alstonia scholaris* have been reported for suppression of fertility in male albino rats[3]. Lupeol acetate isolated from *Alstonia scholaris* has shown antifertility effects in male albino rats[4]. α-amyrin linoleate and a-amyrin palmitate isolated from this *Alstonia sps.* have been reported for non-competitive inhibitor of trypsin and chymotrypsin[5]. Root extract of this plant is reported for cytotoxic activity against human lung cancer cells adenocarcinoma and large cell carcinoma[6] and for the treatment of malaria[7,8]. The plant extract is also reported to exhibit anti-inflammatory activity[9], anti-plasmodial activity[10].

The medicinal importance of *Alstonia* sps. leads us to chemical screening and identification of compounds in search of active constituents. During identification process two new compounds have been obtained.

ISOLATION OF COMPOUNDS

Stem bark of plant *Alstonia scholaris* was collected, shade dried and grinded to powder. This powder was extracted with methyl alcohol for approximately 50 hrs. Methanol was removed by distillation leaving behind a brown sticky mass. This brown mass was treated with acetonitrile for the removal of fats, waxes etc. This brown fat removed mass was re-extracted with chloroform. Solvent chloroform was removed and dry 20 gm of extract was subjected to column chromatography for the isolation of chemical components. For this purpose a column of 1.5m in height with 2.5cm diameter filled with 500 g silica gel G (60-120 mesh) was used. This column was eluted with various solvents and solvent mixtures in their increasing polarity. When column was eluted with solvent mixture of petroleum ether and benzene in ratio 3:1 compound 1 was obtained and when eluted with solvent mixture chloroform and ethyl acetate in ration 1:1 compound 2 was obtained.

EXPERIMENTAL:

Spectral Observations of Compound 2

Compound 2 has shown R_f value 0.32 in pet ether acetone (4+1) system has been calculated for $C_{24}H_{32}O_4$. Compound 2 has shown its melting point at 212°C.

MS (M/Z) : 384, 383 (M^+), 381, 368, 367, 353, 337, 279, 253, 236 etc.

^{1}H NMR (δ, ppm) : ($CDCL_3$) : 0.76 (6H, brs), 1.18 (1H, brs), 2.37 (3H. s), 3.2 (remaining 19H, s), 4.15 (2H, s), 11.8 (1H, brs).

^{13}C NMR (δ, ppm) : ($CDCl_3$) : 20.5 (C-1), 31.5 (C-2), 28.1 (C-3), 54.9 (C-4), 55 (C-5), 69.8 (C-6), 61.8 (C-7), 39.8 (C-8), 49.9 (C-9), 48.5 (C-10), 29.1 (C-11), 25.1 (C-12), 28.6 (C-13), 30.1 (C-14), 109 (C-15), 150 (C-16), 130 (C-17), 126 (C-18), 177 (C-19), 76.8 (C-20), 27.1 (C-21), 15 (C-22), 18 (C-23) etc.

RESULT AND DISCUSSION :

Compound Alscholaridine

The compound showed molecular peak at 384 on the basis of ^{1}HNMR spectrum. The number of protons was calculated to be 32 and ^{13}C NMR spectrum showed 24 signals for the carbon atoms on this basis the molecular formula for compound has been calculated as $C_{24}H_{32}O_4$.

A characteristic broad singlet observed at δ12 in the ^{1}HNMR, is assigned to the proton of a hydroxyl group attached to C-16.

One broad doublet has been observed at δ4.15 in the ^{1}HNMR, has been calculated for two protons attached to C-15 & C-18 which are bonded with double bond with C-16, C-17 respectively. It has been further confirmed by ^{13}C NMR spectra

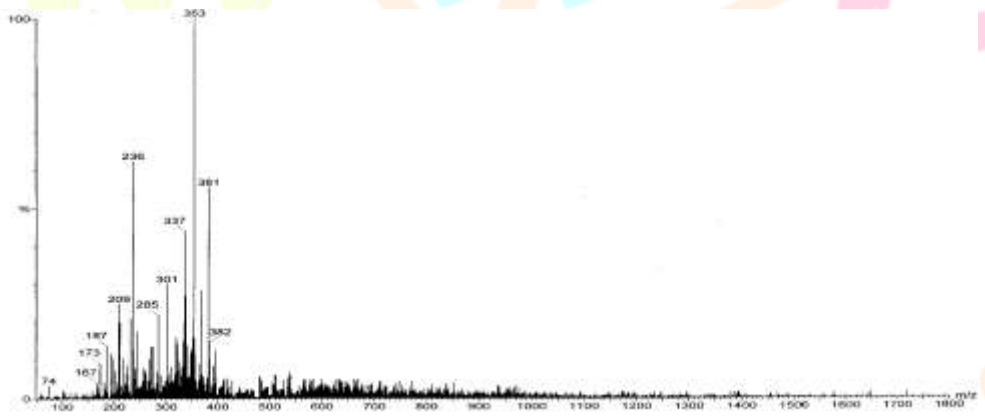
In ^{13}C NMR spectra, absorbance has been observed at δ109 & δ150 and these values are assigned to C-15 & C-16 respectively. C-16 carbon atom has shown absorbed at δ150 due to the attachment of –OH group with it. Proton of hydroxyl group attached to olefinic carbon atom showed absorption at δ11.8 as broad singlet out of two protons, one proton is assigned to be attached at C-15 carbon atom. Similarly absorption at δ130 and δ126 has been assigned to C-17 & C-18 respectively. Remaining second proton observed at δ4.15 is assigned to be attached to C-18[11]. Absorption at δ130 is assigned to C-17 & this shift is due to presence of side chain attached to it. Presence of side chain has been evident by proton NMR. ^{1}H NMR spectra shows absorbance for two methyl groups at δ0.76 & one proton attached to the tertiary carbon atom of side chain which has been observed at δ1.18 in proton spectra and tertiary carbon at δ27.1 in ^{13}CNMR spectra i.e. proton attached to C-21 atom has shown its absorbance at δ1.18 in ^{1}H NMR spectrum.

C-12, C-22 & C-23 carbon atoms of side chain have shown absorption at δ27.1, δ15 & δ18 respectively[12]. Absorption at δ54.9 and δ55 is assigned to C-4 and C-5 respectively. Both carbon atoms are attached with C-19 & C-20 carbon atoms. C-19 carbon atom is linked with oxygen atom by double bond. Carbon atoms C-19 and C-20 are bonded with oxygen atom forming a cyclic structure.

Absorption at δ69.8 and δ61.8 have been assigned to C-6 and C-7 atoms respectively. Both carbon atoms are bonded with oxygen atom forming exocyclic ring structure. Absorbance of C-6 and C-7 carbon atoms have been shifted in down field due to the attachment of this oxygen atom p[13]. C-24 atom of methyl group attached to C-6 showed absorption at δ2.37.

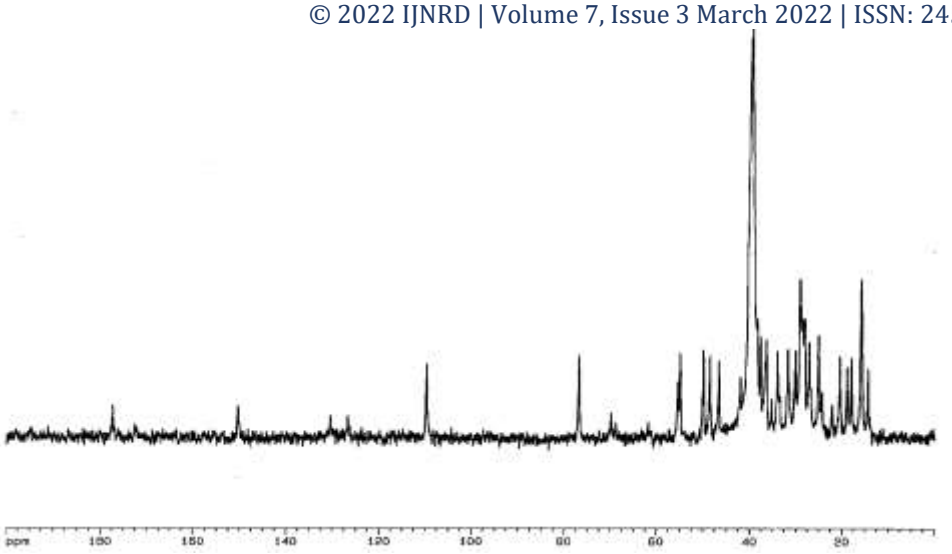
Absorbance of C-19 is shifted to δ177 due to the presence of oxygen atom bonded with it through double bond and one more oxygen is attached to it. Therefore C-19 carbon atom is attached to two oxygen atoms.

The absorbance value of other carbon atoms were assigned as δ20.5 (C-1), 31.7 (C-2), 28.1 (C-3), 39.8 (C-8), 49.9 (C-9), 48.5 (C-10), 29.1 (C-11), 25.1 (C-12), 28.6 (C-13) and 30.1 (C-14).



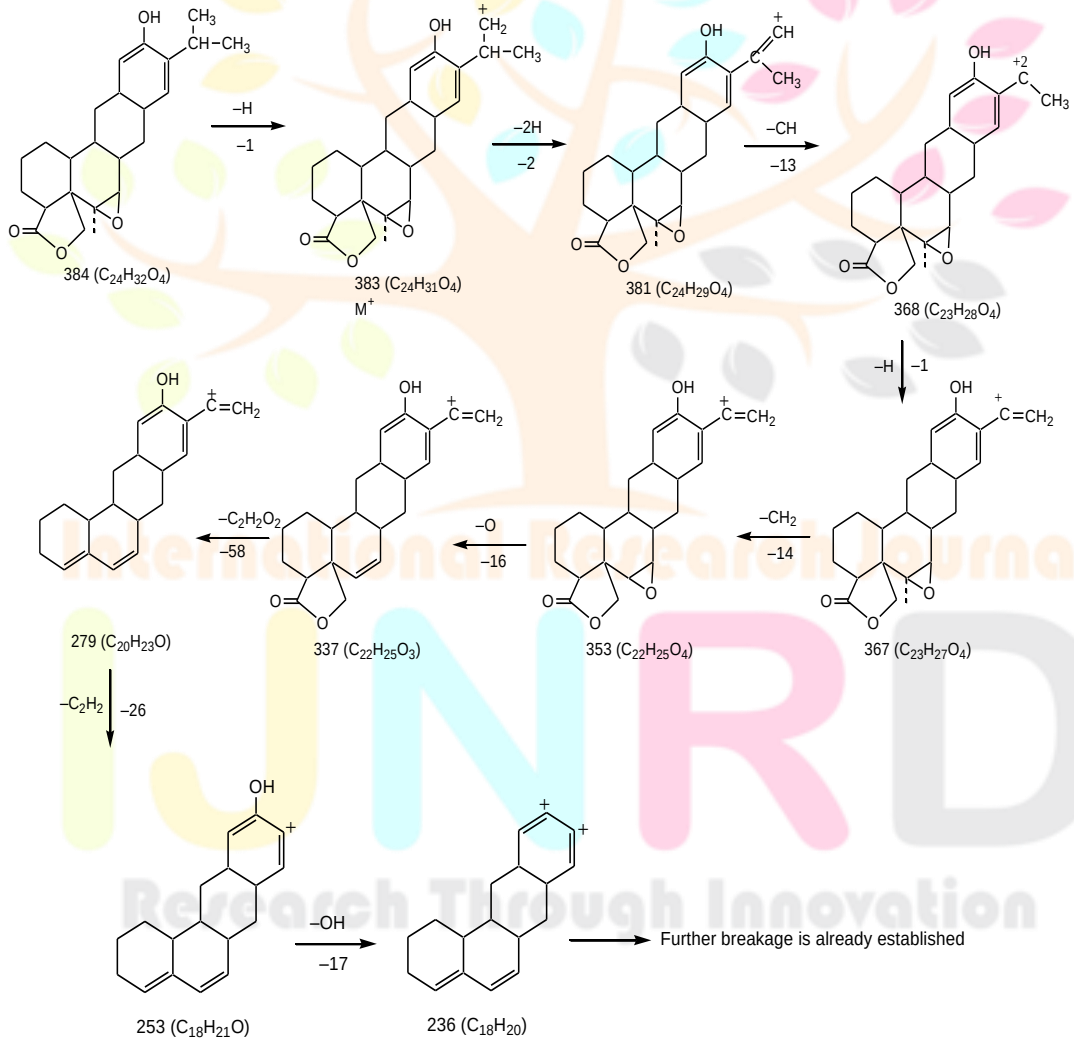
Mass spectra of Compound Alscholaridine

^{1}H NMR Spectra of Compound Alscholaridine

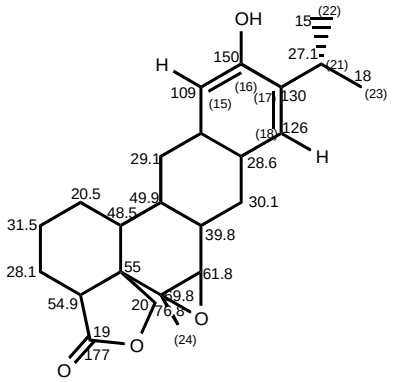


^{13}C NMR Spectra of Compound Alscholaridine

On the basis of molecular ion peaks in mass spectra a splitting pattern for this proposed compound has been established. This splitting pattern also supports the proposed structure.



On the basis of above discussion following structure has been proposed for compound Alscholaridine.



Compound Alscholaridine

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