Isolation of new racemic sugar (D/L galacturonic acid) from leaves of *Paederia foetida* Linn.

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Abstract

New racemic sugars (D/L galacturonic acid) were isolate from the leaves of *Paederia foetida* Linn by the column chromatography. Identification of these isolated compound achieved by TLC, ^{1}H-NMR, ^{13}C-NMR, FTIR and Mass spectral data.

Keywords: Isomer, TLC, NMR, *Paederia foetida* Linn,

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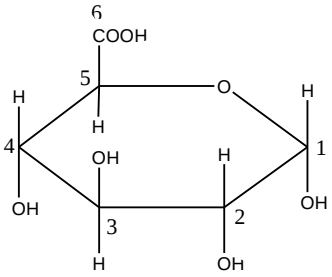
INTRODUCTION:

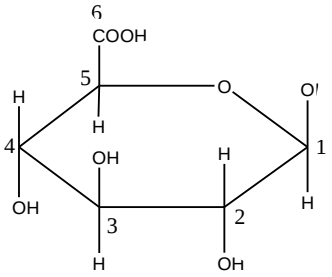
Paederia foetida Linn (Hindi – *Ghandhali*, Assameese – *Bedolilata*) belonging to family Rubiaceae is a fast growing vine found in region of Himalayas. It is also reported to be used in gout, vesical calculi, diarrhoea, dysentery, piles, inflammation of the liver and emetic [1-4]. It possesses various types of biological activities such as anti- inflammatory [5], antidiarrhoeal [6], antinocieptive [7], antitussive [8], antiviral [9], anti-tumor promoters[10].

RESULT AND DISCUSSION:

Methanolic extract of the leaves of *Paederia foetida* Linn was given two compounds which were identified as following types.



D - galacturonic acid



L - galacturonic acid

Figure 1 **Figure 2**

Compound B-I - D – galacturonic acid (Figure 1) was obtained as a white flake; the molecular formula of B-I was determined as $C_6H_{12}O_7$ using IR, ESMS and NMR spectral data. Its IR spectrum showed the absorption bands for hydroxyl group at 3392, 3730 cm^{-1} and carboxylic group at1670 cm^{-1}. The ^1H – NMR spectrum of B – I showed a one proton doublet at δ 4.75 (J = 7.1 Hz), assigned to anomeric H – 1 proton. One proton doublet at δ 3.96 (J = 2.8 Hz, 2.8 Hz) was ascribed to carbinol H – 4 proton. Two-one proton doublet at δ 3.55 (J = 7.2Hz) and 3.42 (J = 2.8 Hz) were attributed to hydroxymethine H – 2 and H – 5, respectively. One proton multiplet at δ 3.17 was accounted to carbinol H – 3. ^{13}C – NMR Spectrum exhibited signal for carboxylic carbon at δ 182.13 (C – 6), anomeric carbon at δ 74.43 (C – 1). The remaining carbonyl carbons resonated between δ 72.49 –71.21. The ESMS of B – I displayed a molecular ion peck at m/z 194 corresponding to the sugar molecular formula $C_6H_{10}O_7$.

Compound B-II - L – galacturonic acid (Figure 2) was obtained as a white flake; the molecular formula of B-II was determined as $C_6H_{12}O_7$ using IR, ESMS, NMR spectral

data. Its IR spectrum showed the absorption bands for hydroxyl group at 3421, 3390 cm^{-1} and carboxylic group at1653 cm^{-1}. The ^{1}H – NMR spectrum of B – II showed a one proton doublet at δ 4.89 (J = 7.1 Hz) assigned to anomeric H – 1 proton. One proton doublet at δ 3.96 (J = 2.8 Hz, 2.8 Hz, H – 4) was ascribed to carbinol H – 4 proton. Two-one proton doublet at δ 3.57 (J = 7.1Hz, H – 2) and 3.47 (J = 2.8 Hz, H – 5) were attributed to hydroxymethine H – 2 and H – 5, respectively. One proton multiplet at δ 3.21 was accounted to carbinol H – 3 and ^{13}C – NMR Spectrum exhibited signals for carboxylic carbon at δ 182.13 (C – 6), anomeric carbon at δ 74.43 (C – 1). The remaining carbonyl carbons resonated between δ 72.49 – 71.21. The ESMS of B – II displayed a molecular ion peck at m/z 194 corresponding to the sugar molecular formula $C_6H_{10}O_7$.

EXPERIMENTAL:

All the reagents were A.R. grade and used without further purification. Melting points were determined on a Veego, Model No. MPI melting point apparatus and are uncorrected. ^{1}H NMR spectra were recorded on Bruker Avance II 400 NMR Spectrophotometer and ^{13}C NMR spectra on Bruker Avance II 100 NMR Spectrophotometer in D_2O using TMS as internal standard. Mass spectra were obtained on VG-AUTOSPEC spectrometer. UV λmax (D_2O) were recorded on Shimadzu UV-1700 and FT-IR (in 2.0cm-1, flat, smooth, abex) were taken on Perkin Elmer – Spectrum RX-I spectrophotometer.

The leaves of *Paederia foetida* Linn. were collected from the Botanical Garden (Department of Life Sciences) Dibrugarh University, authenticated from Botanical Survey of India, Shillong (Ref. No.DU/PSc/HRB-2/08). The authenticated leaves of *Paederia foetida* (Linn.) were dried under shade and coarsely powered and were first extracted with petroleum ether (60 - 80°C) and then methanol in soxhlet apparatus.

In the column methanolic extract eluted successively with different solvents, in order of increasing polarity of the solvents. The column was first eluted with pure petroleum ether (60-80°C) and then with successive series of different solvents in various combinations, such as petroleum ether, petroleum ether:chloroform (75:25, 50:50, 25:75 v/v), chloroform, chloroform : methanol (90:10, 80:20, 70:30, 60:40, 50:50, 40:60, 30:70, 20:80, 10:90 v/v) and methanol.

The fraction collected were subjected to thin layer chromatography (TLC) to check the homogeneity of various fractions chromatographically identical fractions (having same R_f values) were combined together and concentrated.

B-I

Fractions 16-22 were pooled and concentrated and this fraction was taken up for purity using methanol to give final compound.

B – II

Fractions 4-12 were pooled and concentrated. Further washing were done by chloroform, methanol to remove impurities to obtain final compound.

B – I

Elution of the column with methanol: chloroform (80:20) B-I obtain as white flake. It gives positive carbohydrate test.

m. p. : 158-159°C

^{1}H – NMR Spectrum (400 MHz, D_2O): δ 4.75 (1H, d, J = 7.1 Hz, H – 1), 3.96 (1H, dd, J = 2.8, 2.8 Hz, H – 4), 3.55 (1H, d, J = 7.2 Hz, H – 2), 3.42(1H, d, J = 2.8 Hz, H – 5), 3.17(1H, m, H – 3)

^{13}C – NMR Spectrum (400 MHz, D_2O): δ 182.13 (C – 6), 74.43 (C – 1), 72.49 (C – 2, C – 5), 72.26 (C – 4), 71.21 (C – 3)

ES MS (m/z) (rel. int.): 194(11.3) $[M]^+$ ($C_6H_{10}O_7$)

IR Spectrum (in KBr pellet, cm^{-1}): 3392, 3230, 2921, 1670, 1445, 1417, 1370, 1247, 1195, 1147, 1113, 1049, 1003, 894, 732.

B – II

Elution of the column with methanol: chloroform (50:50) furnished white flake, flake successively treated with chloroform, methanol for remove the impurities. It gives positive carbohydrate test.

m. p. : 165 – 167°C

^{1}H – NMR Spectrum (400 MHz, D_2O): δ 4.89(1H, d, J = 7.1 Hz, H – 1), 3.96 (1H, dd, J = 7.1 Hz, H – 2), 3.47 (1H, d, J = 2.8 Hz, H – 2), 3.21(1H, m, H – 3).

ES MS (m/z) (rel. int.): 194(11.3) $[M]^+$ ($C_6H_{10}O_7$).

IR Spectrum (in KBr pellet, cm^{-1}): 3421, 3390, 2924, 1653, 1384, 1050.

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ABBREVATIONS:

TLC = Thin Layer Chromatography

NMR = Nuclear Magnetic Resonance

UV = Ultra Violet Spectroscopy