Isolation of Hex-2-ulofuranosyl hexopyranoside and GC-MS profile of different extracts from *Capparis decidua* (Forssk.)

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**ABSTRACT**

In this phytochemical studies Hex-2-ulofuranosyl hexopyranoside (sucrose) was been isolated from the aerial parts of *Capparis decidua*. The compound has been identified by different spectroscopic techniques including X-ray crystallography. The chloroform and ethylacetate extracts of the plant were further subjected to GC-MS which result in the identification of interesting compounds.

**Key words:** Hex-2-ulofuranosyl hexopyranoside, GC-MS profile, *Capparis decidua*

**INTRODUCTION**

*Capparis decidua* Family (Capparaceae) is widely distributed in typical deserts and semi-deserts areas in northern and central Sudan, especially on sandy soils and in low rainfall savanna on clays ¹. It is also found in Blue Nile, Upper Nile, western and eastern Sudan besides northern areas of the country ². In Sudan *C.decidua* is used traditionally as anti-helmintic, analgesic, aphrodisiac, carminative, diaphoretic, emmenagogue and laxative. The bark extract is used for asthma and cough management. The paste of young leaves and branches are applied as protective coat on boils and swelling for the absorption of serious secretions ³, and for their anti-inflammatory, astringent effects, they are used also as stomachic, laxative, antidote, and used for skin diseases ². The poultices of the twigs are used against head-ache ⁵. Decoction preparation from the roots is used to relieve fever and is also used for jaundice. As fumigation, roots are used to treat fever and rheumatism ⁶. The aerial part is used for rheumatism, gout; externally the infusion is used for boils, eruption and ulcers, while internally as antidote to poisons ⁷.

**EXPERIMENTAL SECTION**

2-1. Plant material

*Capparis decidua* (aerial parts) was collected from Shambat, Khartoum north-Sudan. The plant was authenticated at the Medicinal and Aromatic Plant Research Institute (MAPRI), Sudan and voucher specimens deposited in the Herbarium.
2-2. Extraction and isolation procedure
Collected plant was dried under shade and then powdered. The powdered plant material (200 gram) was extracted by cold maceration method with sufficient quantity of 80% methanol for 48 hrs. The process of extraction was repeated twice for the completion of the extraction. The extract was filtered using Whatman filter paper and the filtrates were concentrated under reduced pressure which afforded 39 g of a concentrated extract. A 30 g of this extract was fractionated on a normal phase silica gel column eluted with petroleum ether-chloroform and chloroform–ethyl acetate mixtures of increasing polarity to give the sub-fractions (1, 2 and 3). Compound MW-1 was obtained as colourless crystalline solid from the fraction of 50% methanol in ethylacetate during elution of column packed with methanol extract of *Capparis decidua*. The crystals were collected and washed several times with methanol.

2-3. X-Ray crystallography
Conducted at X-Ray crystallography apparatus (diffractorometer with Molybdenum source). Bruker SMART APEX-Germany

2-4. Gas chromatography- Mass spectrophotometry (GC-MS) of different sub-fractions
GC-MS were carried out on the Schimadzu GCMS-QP1020 spectrometer operating at 45 to 500 MHz. 1 ml of compound being tested was dissolved in methanol. The solution was filtered through micro filter. Then 1 microliter of the solution was injected using Hamilton microlitr syringe. The runs on the GC was done according to the following method:


2-5. Nuclear magnetic resonance (NMR)
^1^H-NMR- and ^13^C-NMR spectra were carried out on the Bruker AM 500 and 700 spectrometer operating at 500 and 700 MHz (^1^HNMR) in spectroscopic grade solvents D$_2$O, MeOD and CDCl$_3$. The chemical shifts values are expressed in δ (ppm) units using (TMS) as an internal standard and the coupling constants (J) are expressed in Hertz (Hz). Standard pulse sequences were used for generating COSY, HMQC and HMBC spectra (2D experiments).

5 mg of the compound being tested was dissolved in 0.6 ml of suitable solvent used and the experiments were sent to NMR instrument at temperature of 296.1 K.

RESULTS AND DISCUSSION

3-1. Structure Elucidation of Compound MW-1
Compound -1 was identified by spectroscopic technique including (1D and 2D NMR) and confirmed by X-ray crystallography.

Compound MW-1, was obtained as colorless crystalline solid from the fraction of 50% methanol in ethyl acetate during column elution. The ^1^H NMR spectrum of MW-1 shown one proton doublet at δ 4.16 (J = 8.5Hz), another proton shown a double doublets at δ 4.00 (J = 4, 9 Hz) ascribed to oxygenated methane is assigned to H$_7$ and H$_8$ respectively. One proton shown a broad singlet at δ 5.36 is assigned to anamoric proton (H$_5$). While a third proton shown a double doublets at δ 3.43 (J = 9. 9 Hz) ascribed to oxygenated methine proton is assigned to H$_4$. Three set of protons (2 each) shown a broad multiplet at δ 3.76, 3.68 and a doublet at δ 3.51 (J = 9.5 Hz) were accounted to the remaining protons. ^13^C-NMR data shown a clear signals attributed to quaternary carbons at δ 103.68 is assigned to C$_5$. One anamoric carbon at δ 92.19 is assigned to C$_6$. Three oxygenated methylene carbons at δ 60.14, 61.38 and 62.38 are assigned to C$_{10}$, C$_{12}$, and C$_{11}$ respectively. Six oxygenated methane carbons at δ 72.42, 69.24, 72.58, 71.08, 76.47, 74.02 and δ 81.37 are assigned to C$_1$, C$_2$, C$_3$, C$_4$, C$_7$, C$_8$ and C$_9$ respectively. The DEPT 90 spectrum of MW-1 showed that the signals at δ 69.2, 71.08, 72.4, 72.58, 74.02, 76.47 and 92.19 were related to the oxygenated methane protons.
The existence of $^1$H NMR signals in the deshielded region at $\delta$ 5.36 and $^{13}$C NMR signals at $\delta$ 92.19 and 103.68 supported the linkages of the two sugar units. The $^1$H-$^1$H Cosy spectrum of MW-1 showed correlations between $H_4$ with $H_3$ and $H_7$ with $H_8$.

The HMBC spectrum of MW-1 exhibited interactions of $H_9$ ($\delta$ 3.62) with $C_7$ ($\delta$ 76.47) by three bonds correlation, $C_6$ ($\delta$ 103.68) by four bonds correlation. In HSQC spectrum of MW-1, $C_2$ at $\delta$ 69.24 interacted with $H_2$ at $\delta$ 3.70; $C_4$ at $\delta$ 71.08 with $H_4$ at $\delta$ 3.43; $C_1$ at $\delta$ 72.42 with $H_1$ at $\delta$ 3.83; $C_3$ at $\delta$ 72.58 with $H_3$ at $\delta$ 3.72; $C_4$ at $\delta$ 74.02 with $H_8$ at $\delta$ 4.00; $C_7$ at $\delta$ 76.47 with $H_7$ at $\delta$ 4.16; and $C_6$ at $\delta$ 81.37 with $H_6$ at $\delta$ 3.62. The X-ray crystallography of compound MW-1 clearly shows the structure of the compound as well as the bond orientation (fig-18). On the bases of these evidences the structure of MW-1 has been established as Hex-2-ulofuranosyl hexopyranoside [Sucrose] ($C_{12}H_{22}O_{11}$).

X-ray crystallography also indicates clearly the actual structure of the compound confirming our prediction.

![Hex-2-ulofuranosyl hexopyranoside][Sucrose]

Table 1: $^1$H and $^{13}$C NMR Spectral Data of Compound-1 (500 MHz, $D_2$O)

<table>
<thead>
<tr>
<th>Position</th>
<th>$\delta$ $^1$H (Multiplicity, $J$ in Hz)</th>
<th>$\delta$ $^{13}$C</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td>3.83 m</td>
<td>72.42</td>
</tr>
<tr>
<td>2</td>
<td>3.70 m</td>
<td>69.24</td>
</tr>
<tr>
<td>3</td>
<td>3.72 m</td>
<td>72.58</td>
</tr>
<tr>
<td>4</td>
<td>3.43 dd ($J$=9, 9)</td>
<td>71.08</td>
</tr>
<tr>
<td>5</td>
<td>5.36 br</td>
<td>92.19</td>
</tr>
<tr>
<td>6</td>
<td></td>
<td>103.68</td>
</tr>
<tr>
<td>7</td>
<td>4.16 d ($J$=8.5)</td>
<td>76.47</td>
</tr>
<tr>
<td>8</td>
<td>4.00 dd ($J$=4, 9)</td>
<td>74.02</td>
</tr>
<tr>
<td>9</td>
<td>3.62 m</td>
<td>81.37</td>
</tr>
<tr>
<td>10</td>
<td>3.68 m</td>
<td>80.14</td>
</tr>
<tr>
<td>11</td>
<td>3.51 d ($J$=9.5)</td>
<td>62.38</td>
</tr>
<tr>
<td>12</td>
<td>3.76 m</td>
<td>61.38</td>
</tr>
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</table>
3-2. GC-MS analysis of *Capparis decidua* methanolic sub-fractions (1, 2 and 3)
The GC chromatogram of Sub-fraction (1), exhibited four peaks at retention times (Rt) 24.129, 31.549, 32.268 and 42.664 min indicating presence of four compounds, while MS spectrum displayed [M]+ at 177, 185, 150 and 275 m/z respectively, corresponding to different molecular ions. Suggestion of the separated components was accomplished using computer search by matching spectra with reference spectra in the computer library. The suggested compounds are shown in Table-2.

<table>
<thead>
<tr>
<th>Compound number</th>
<th>Retention time in min. (Rt)</th>
<th>Compound name</th>
<th>Chemical formula</th>
<th>Base peak</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td>24.129</td>
<td>Diethyl Phthalate</td>
<td>C₉H₁₀O₄</td>
<td>149</td>
</tr>
<tr>
<td>2</td>
<td>31.549</td>
<td>Tridecanoic acid</td>
<td>C₁₄H₂₈O₂</td>
<td>74</td>
</tr>
<tr>
<td>3</td>
<td>32.268</td>
<td>Dibutyl Phthalate</td>
<td>C₁₆H₂₂O₄</td>
<td>149</td>
</tr>
<tr>
<td>4</td>
<td>42.664</td>
<td>1,2-benzenedicarboxylic acid,diisooctyl ester</td>
<td>C₂₄H₃₈O₄</td>
<td>149</td>
</tr>
</tbody>
</table>
The GC chromatogram of SF-2 exhibited four peaks at retention times (R_t) 28.734, 32.879, 36.562 and 39.945 min indicating presence of four compounds, while the MS spectrum displayed [M]⁺ at 125, 125, 125 and 392 m/z respectively, corresponding to different molecular ions. The suggested compounds are shown in Table-3.
The GC chromatogram of SF-3, exhibited three peaks at retention times (R_t) 29.923, 28.713, and 32.839 min indicating presence of three compounds, MS spectrum displayed [M]^+ at 206, 125 and 125 m/z respectively, corresponding to different molecular ions. The suggested compounds are shown in Table-4.

CONCLUSION

Hex-2-ulofuranosyl hexopyranoside (sucrose) was isolated from Capparis decidua (Forssk.) for the first time, where an advanced spectrosopical techniques were used for its identification. GC-MS technique one of the most powerful methods used for identification of active medicinal plants constituents, in addition it may help in assessing of quality against adulterant and act as a biochemical marker for those medicinally important plants in the pharmaceutical industry.

Acknowledgements

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REFERENCES