FURTHER NEW ALKALOIDS FROM CASSIA SIAMEA LAM. LEAVES

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ABSTRACT

Two new isoquinolone alkaloids were isolated from the leaves of Cassia siamea Lam. and their structures were designated as 8-acetyl siamine and 3,8-dimethyl-6,7-methylene dioxy isoquinolone.

INTRODUCTION

In a previous publication, the authors isolated three isoquinolone alkaloids, siamine, siaminine A and siaminine B from Cassia siamea Lam. Leaves.

Siamine, an isoquinolone alkaloid, was obtained by Ahn and Zymalkowski from the seeds of the same plant. However, piperidenol and monoterpenic alkaloids have been isolated from several Cassia species.

The leaves of C. siamea were subjected to further phytochemical investigation with the aim of isolating and characterizing other new alkaloids.

EXPERIMENTAL

General Experimental Procedure:

Melting points were determined with a Kofler hot stage apparatus and uncorrected. UV spectra were recorded in MeOH using Unicam SP1750 Spectrometer. IR spectra were determined in KBr discs using Unicam SP1025 Spectrometer. $^1$H- and $^13$C-NMR were recorded on Jeol FI models FX-60 and FX-90.
Spectrometers in DMSO-d₆ using TMS as internal standard. Mass spectra were determined on a Finnegan Quadrupol 4023 instrument. TLC on silica gel G (Merck) plates, developers: CHCl₃-MeOH (45:5) (syst. I) and CHCl₃-MeOH (4:1) (syst. II). Spots were sprayed with modified Dragendorff's reagent.

**Plant Material:**

The plant was collected during flowering in April 1986, from the Experimental Station of Ornamental Plants (Zohria Gardens), Cairo. It was identified by the late Prof. F.Y. Amin, Prof. of Floriculture and Horticulture, Faculty of Agriculture, Assiut University.

**Extraction and Fractionation:**

The powdered defatted leaves (2 kg) were extracted with ethyl alcohol (70%) at room temperature. The concentrated extract was fractionated with chloroform. The chloroform residue (7 g) was chromatographed on silica gel column with chloroform and chloroform-methanol gradient. The concentrated fractions giving positive response with Dragendorff's reagent were purified by preparative TLC and the eluted compounds were recrystallized from MeOH.

**Compound I:** Colourless fine needles (MeOH), m.p. 216-18°C, [α]²²_D -0.93 (C, 0.75, EtOH); UV (MeOH) max nm (log ε): 226(3.3), 242(3.36), 250(3.8), 292(3.24)

+AlCl₃: -; +NaOH: 224 (3.24), 256(3.42), 330(3.16); IR(KBr) ν cm⁻¹: 3300-3100, 1720, 1645(lactam), 1570, 1500, 1440, 1320, 1160, 890; ¹H-NMR (DMSO-d₆)

δ ppm: 2.19(3H, S, C₃-Me), 2.28(3H, S, CH₃-C), 5.98(1H, S, H₄), 6.58(1H, S, J=2.4Hz, H₅), 6.72(1H, d, J=2.4Hz, H₇), 8.35(1H, br. S, NH); ¹³C-NMR (DMSO-d₆)

δ ppm: C₁ (177.6), C₃ (138.1), C₄ (101.2), C₅ (110.2), C₆ (164.2), C₇ (113.2), C₈ (161.1), C₉ (158.9), C₁₀ (118.1), C₁₁ (19.3), C₁₂ (204.2), C₁₃ (29.6), MS m/z (rel.int.): 234(1.4) (M⁺+1), 233(8.9) (M⁺), 232(14.7) (M⁺-H), 215 (7.2) (M⁺-18), 191(12.3) (M⁺-42), 190(71.4) (M⁺-43), 189(25.4) (M⁺-44), 163 (2.8), 162(21), 161 (20.4), 105(5.6), 91(5.3), 77(13.3), 43(100).
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Compound II: Light yellow crystals (MeOH), m.p. 222-225°C; UVMeOH
max
nm(log $\varepsilon$): 228(3.65), 254(3.7), 315 (3.16), 330(3.1), 366(3.15). IR
(KBr) $\nu$ cm$^{-1}$: 3100-3000, 1660(lactam), 1580, 1440, 950($\nu$-CH$_2$-0); $^1$H-NMR
(DMSO-d$_6$) $\delta$ ppm: 2.18(3H, S, C$_3$-Me), 2.32(3H, S, C$_8$-Me), 6.06(1H, S, H$_4$),
6.46(2H, br. S, 0-CH$_2$-0), 6.83(1H, S, H$_5$), 8.32(1H, br. S, NH); $^{13}$C-NMR
(DMSO-d$_6$) $\delta$ ppm: C$_1$(160.4), C$_3$(137.9), C$_4$(100.4), C$_5$(105.6), C$_6$(158.5),
C$_7$(154.8), C$_8$(112.2), C$_9$(152.9), C$_{10}$(120.5), C$_{11}$(19.2), C$_{12}$(98.6),
C$_{13}$(23.7); MS. m/z(rel.int.) 215(3)(M$^+$-2), 213(100), 185(7.1), 170(4.3),
156(5.4), 141(3.8), 128(5.4), 115(7.5), 106(11.4), 77(11.2), 63(14.7),
51(14.4), 43(28.4).

Alkaline hydrolysis of I: 10 mg was dissolved in 5 ml MeOH to which 5
ml of 5% methanolic KOH was added. The mixture was stirred at room
temperature for 18 hours. MeOH was distilled and 10 ml water was added.
The solution was extracted with (4 x 10 ml) CHCl$_3$. The extract was dried
over anhydrous Na$_2$SO$_4$ and solvent was distilled under reduced pressure
to give light yellow residue. The latter was chromatographed using system I
and the major zone was scraped and extracted with CHCl$_3$.

**RESULTS AND DISCUSSION**

The chloroform fraction of the defatted alcoholic extract
of *Cassia siamea* Lam. leaves was found to show, at least, six
Dragendorff's-positive and fluorescing spots on silica gel
chromatograms.

Fractionation of this fraction on a silica gel column and
purification of the subsequent fractions by preparative TLC
afforded two pure compounds (I and II). Both compounds gave
orange colour with Dragendorff's reagent, fluoresced under UV
light and failed to give hydrochloride salts.
Compounds I & II showed UV absorption maxima between 230-370 nm indicating a highly conjugated chromophore. Their IR spectra exhibited intense peaks at 3000-3100 cm\(^{-1}\) assignable to N-H stretching vibration of lactam, while peaks at 1660, 1645 cm\(^{-1}\) were consistent with the presence of a \(\beta\)-lactam carbonyl\(^\text{10}\). IR spectrum of I showed another band at 3300 cm\(^{-1}\) for OH which is supported by a bathochromic shift in UV upon addition of NaOH and formation of green colour with FeCl\(_3\).

Mass spectrum of I revealed a molecular ion peak at m/z 233, in addition to a peak at m/z 190 (M-43) for the cleavage of a lactam group. Its \(^1\)H-NMR showed three aromatic protons, two of them appeared as doublets with meta coupling for H\(_5\) and H\(_7\) at \(\delta\) 6.58 and 6.72 ppm respectively, while, the third appeared as a singlet at \(\delta\) 5.98 ppm for C\(_4\)-H. A singlet for 3H at \(\delta\) 2.19 assignable to C\(_3\)-Me which appeared in \(^1\)H-NMR at \(\delta\) 19.3 ppm (characteristic for C\(_3\)-Me)\(^\text{11}\). An acetoxy group could be deduced by the appearance of a singlet for 3H at \(\delta\) 2.28 ppm in \(^1\)H-NMR which is supported by an intense band at 1720 cm\(^{-1}\) in the IR spectrum and mass fragment (100%) at m/z 43 in addition to a signal at \(\delta\) 204 ppm for C\(_1\) (\(^1\)C-NMR spectrum). A broad singlet assignable to one proton of NH at \(\delta\) 8.35 ppm was also observed. Trials to obtain a chelate complex with AlCl\(_3\) were unsuccessful indicating that the acetoxy group must be at C-8 while OH is at C-6. On these bases, compound I might be \(\beta\)-acetyl siamine. This assumption was confirmed by the appearance of 12 signals in \(^1\)C-NMR, comparable with similar compounds\(^\text{11}\). Alkaline hydrolysis produced a compound identical with siamine (m.m.p and CO-TLC).

Compound II: IR was similar to compound I with an additional band for aromatic methylene dioxy at 940 cm\(^{-1}\), supported by a broad singlet for 2-protons at \(\delta\) 6.46 ppm in \(^1\)H-NMR.
The latter revealed a broad singlet at $\delta$ 8.32 ppm for one proton of NH and two singlets each for 3H, one at $\delta$ 2.18 ppm (C$_3$-Me) and the other at $\delta$ 2.32 ppm (C$_5$-or C$_8$-Me). The singlet at $\delta$ 6.06 ppm was ascribed to C$_4$-H. Another singlet was observed at $\delta$ 6.83 ppm assignable either to C$_5$- or C$_8$-H. Similar compounds$^{11}$ showed that C$_5$-H and C$_8$-H resonate at $\delta$ 6.57-6.9 and 7.58-7.87 ppm respectively. Therefore, the singlet at $\delta$ 6.83 ppm corresponds to the resonance of C$_5$-H proton and a methyl group is, accordingly, attached to C-8. Compound II is suggested to be 3,8-dimethyl-6,7-methylene dioxy isoquino-lone.$^{13}$ C-NMR showed 12 signals comparing favourably with related compounds$^{11}$.

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