SUPPLEMENTARY MATERIAL

A New Piperidine Alkaloid from the Leaves of Microcos paniculata L.

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Abstract

A new piperidine alkaloid, microcosamine C (1), and one known compound, microcosamine A (2) were isolated from the leaves of *Microcos paniculata*. Structure elucidation was carried out using HR-ESI-MS, 1D and 2D NMR spectroscopic methods and by comparison to data reported in the literature. The absolute configuration at the C-3 hydroxy group of 1 was established by a Mosher esterification procedure. Both of the isolates (1–2) were evaluated for cytotoxicity against four selected tumor cell lines and showed only weak activity against RAW 264.7 cell line.

**Keywords:** piperidine alkaloids; *Microcos paniculata*; cytotoxicity
List of Supplementary Material

Figure S1. $^1$H-$^1$H COSY, $^1$H-$^{13}$C HMBC and NOESY correlations for compound 1.
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(pyridine-$d_5$, 600 MHz).
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determination of the absolute configuration of the 3-hydroxy position by
a Mosher ester procedure.

Table S1. $^1$H and $^{13}$C NMR data for compounds 1 and 2.

Table S2. Cytotoxicity data for compounds 1 and 2.
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Figure S2. HR-ESI-MS of compound 1.
Figure S3. $^1$H NMR spectrum (600 MHz, CD$_3$OD) of compound 1.
Figure S4. $^{13}$C NMR spectrum (150 MHz, CD$_3$OD) of compound 1.
Figure S5. DEPT spectrum (150 MHz, CD3OD) of compound 1.
Figure S6. $^1$H-$^1$H COSY spectrum (600 MHz, CD$_3$OD) of compound 1.
Figure S7. $^1$H-$^{13}$C HMOC spectrum (600 MHz, CD$_3$OD) of compound 1.
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Figure S9. $^1$H NMR spectrum (600 MHz, CDCl$_3$) of compound 1.
Figure S10. $^1$H-$^1$H COSY spectrum (600 MHz, CDCl$_3$) of compound 1.
Figure S11. NOESY spectrum (600 MHz, CDCl₃) of compound 1.
Figure S12. Overlaid proton spectra of the (R)- and (S)-MTPA esters of 1 (pyridine-$d_5$, 600 MHz).
Figure S13. $^1$H–$^1$H COSY spectrum of (R)-MTPA ester of 1 (pyridine-$d_5$, 600 MHz).
(A) $^1$H NMR data of S-MTPA ester of 1
(B) $^1$H NMR data of R-MTPA ester of 1
(C) $\Delta \delta_H (\delta_S - \delta_R)$ values of derivatized products
(D) S-configuration of C-3 in 1

**Figure S14.** (R)- and (S)-MTPA derivatives of 1 with chemical shift values used in the determination of the absolute configuration of the 3-hydroxy position by a Mosher ester procedure.
Table S1 $^1$H (600 MHz) and $^{13}$C NMR (150 MHz) data (J value in Hz, δ in ppm) of compound 1 in CD$_3$OD and 2 in CDCl$_3$.

| position | 1 | 2 |
|----------|---|---|
| 2        | 1.99 (m) | 67.4 | 2.60 (m) | 58.3 |
| 3        | 3.19 (m) | 72.3 | 3.23 (m) | 73.4 |
| 4α       | 2.01 (m) | 67.4 | 2.05 (m) | 34.0 |
| 4β       | 1.40 (m) | 33.9 | 1.47 (m) | 34.0 |
| 5α       | 1.51 (m) | 31.7 | 1.62 (m) | 32.5 |
| 5β       | 1.68 (m) | 1.75 (m) | 32.5 |
| 6        | 2.69 (m) | 69.1 | 3.26 (m) | 58.6 |
| 1'       | 5.51 (dd, 15.1, 8.9) | 135.2 | 5.52 (dd, 15.6, 7.2) | 136.0 |
| 2'       | 6.26 (m) | 134.5 | 6.21 (m) | 129.7-131.2 $^a$ |
| 3'       | 6.15-6.23 $^a$ | 131.7-134.9 $^a$ | 6.06-6.22 $^a$ | 129.7-131.2 $^a$ |
| 4'       | 6.15-6.23 $^a$ | 131.7-134.9 $^a$ | 6.06-6.22 $^a$ | 129.7-131.2 $^a$ |
| 5'       | 6.15-6.23 $^a$ | 131.7-134.9 $^a$ | 6.04 (m) | 129.7-131.2 $^a$ |
| 6'       | 6.15-6.23 $^a$ | 131.7-134.9 $^a$ | 5.70 (dt, 15.1, 7.2) | 135.9 |
| 7'       | 6.09 (m) | 131.0 | 2.10 (q, 7.0) | 32.9 |
| 8'       | 5.76 (dt, 15.1, 6.7) | 138.2 | 1.38 (m) | 31.4 |
| 9'       | 2.12 (q, 7.0) | 26.9 | 1.36 (m) | 22.2 |
| 10'      | 1.01 (t, 7.4) | 14.0 | 0.86 (t, 7.2) | 13.9 |
| 2-CH$_3$ | 1.28 (d, 6.2) | 15.9 | 1.24 (d, 7.0) | 18.5 |
| N-CH$_3$ | 2.30 (s) | 40.3 |

$^a$ Multiplicity patterns unclear due to signal overlapping.
Table S16. Cytotoxicity data against HeLa, HepG2, A-549 and RAW 264.7 Cell Lines of Compounds 1–2.

| compound | IC$_{50}$ ($\mu$M) |
|----------|---------------------|
|          | HeLa    | HepG2   | A-549   | RAW 264.7 |
| 1        | Inactive$^a$ | Inactive | Inactive | 31.5     |
| 2        | Inactive  | Inactive | Inactive | 39.8     |
| Adriamycin | 0.7    | 0.6     | 0.3     | 0.8      |

$^a$ Inactive: indicates <50% inhibition of cell proliferation at 100 $\mu$M.
