Supporting Information

Cytotoxic New Nortriterpene from Roots of *Potentilla atrosanguinea* var. *argyrophylla* and its UPLC Quantification

Mayanka Walia¹, ², Dharmesh Kumar³, Pawan Kumar², Bikram Singh¹, ², Yogendra S. Padwad³, Vijai K. Agnihotri¹, ²

Affiliations

¹Academy of Scientific and Innovative Research, CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh, India
²Natural Product Chemistry and Process Development Division, CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh, India
³Biotechnology Division, CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh, India

Correspondence

**V. K. Agnihotri**

Natural Product Chemistry and Process Development Division, CSIR-Institute of Himalayan Bioresource Technology
Palampur, Himachal Pradesh 176061
India
Phone: +91 1894 233 339
Fax: +91 1894 230 433
kantvijai@yahoo.com
vijai@ihbt.res.in
Table of Content

UPLC-DAD method validation

Mass spectrometry analysis conditions

**Fig. S1** ESI-QTOF-MS chromatogram for compound 1.

**Fig. S2** IR spectrum for compound 1.

**Fig. S3** $^1$H NMR spectrum for compound 1 (Chloroform $d$).

**Fig. S4** $^{13}$C NMR spectrum for compound 1 (Chloroform $d$).

**Fig. S5** DEPT spectrum for compound 1 (Chloroform $d$).

**Fig. S6** HMQC spectrum for compound 1.

**Fig. S7** HMBC spectrum for compound 1.

**Fig. S8** NOSEY spectrum for compound 1.

**Table S1** Percentage growth inhibition of extract, fractions and compound 1.

Method validation

By plotting nominal standard concentration \((x)\) against the peak area \((y)\) of analytes, a calibration curve was obtained. The linearity of the method was obtained by analyzing a series of a standard solution of compound 1. The intra-day and inter-day variability were determined by analyzing the sample for three times per day and three consecutive days. LOD and LOQ were determined by injecting dilute standard solutions until the signal to noise ratios \((S/N)\) were 3:1 and 10:1, respectively. Recovery was performed for ascertaining the accuracy of the method by adding three different concentrations \((0.01, 0.02 \text{ and } 0.03 \text{ mg/mL})\) of standard to the extract.

Mass spectrometry analysis conditions

ESI-MS analysis was performed by direct infusion with syringe pump (flow rate 0.28 mL/min). Different parameters were set as: cone voltage 30 V; cone gas flow 50 L/h; capillary voltage 3.2 kV; desolvation gas flow 400 L/h; desolvation temperature 220°C; source temperature 80°C; scan time 1.0 s and interscan delay 0.1 s.

Fig S1 ESI-QTOF-MS chromatogram of compound 1.
Fig S2 IR spectrum of compound 1.

Fig. S3 $^1$H NMR spectrum of compound 1 (Chloroform $d$).
**Fig. S4** $^{13}$C NMR spectrum of compound 1 (Chloroform $d$).
Fig. S5 DEPT spectrum of compound 1 (Chloroform d).
Fig. S6 HMQC spectrum of compound 1.
Fig. S7 HMBC spectrum of compound 1.
Fig. S8 COSY spectrum of compound 1.
Fig. S9 NOESY spectrum of compound 1.
Table 1S Percentage growth inhibition of standard and compound (1).

<table>
<thead>
<tr>
<th>Sample</th>
<th>Conc. in μg/mL</th>
<th>SiHa</th>
<th>KB</th>
<th>Colo-205</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>10</td>
<td>18.3 ± 0.5</td>
<td>15.1 ± 2.1</td>
<td>1.2 ± 1.0</td>
</tr>
<tr>
<td></td>
<td>25</td>
<td>44.3 ± 0.6</td>
<td>56.6 ± 2.7</td>
<td>84.2 ± 2.4</td>
</tr>
<tr>
<td></td>
<td>50</td>
<td>70.0 ± 0.5</td>
<td>65.8 ± 3.2</td>
<td>57.4 ± 0.5</td>
</tr>
<tr>
<td></td>
<td>100</td>
<td>84.6 ± 1.3</td>
<td>89.5 ± 1.8</td>
<td>68.8 ± 1.9</td>
</tr>
<tr>
<td>Vinblastin</td>
<td>1 μM</td>
<td>63.5 ± 1.6</td>
<td>53.0 ± 0.6</td>
<td>72.5 ± 2.6</td>
</tr>
</tbody>
</table>