Note

Isolation and Identification of Three Insecticidal Principles from the Red Alga Laurencia nipponica Yamada

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A number of sesquiterpenoids and nonterpenoid C15 acetogenins have been isolated from red algae of the genus Laurencia (Rhodomelaceae). In the course of our screening program for insecticidal compounds of marine origin, we found that the methanol extract of the red alga Laurencia nipponica Yamada possessed a strong insecticidal activity against mosquito larvae (Culex pipiens pallens). The bioassay-guided isolation afforded three active principles which were identified as deoxyprepacifenol (1), Z-laureatin (2) and Z-isolaureatin (3). Here, we describe the isolation and identification of these compounds as well as their insecticidal activities.

The dried alg specimens (180g) were extracted with methanol, whose extracts were partitioned between water and ethyl acetate. The ethyl acetate portion (2.0g), which showed insecticidal activity against the mosquito larvae (C. pipiens pallens), was chromatographed on a silica gel column with hexane-ethyl acetate systems. The active fraction eluted with hexane-ethyl acetate (9:1, v/v) was further purified by preparative thin-layer chromatography to afford 20mg of compound 1: mp 122~124°C; [α]D25 = +20° (c=0.20, CHCl3); IR vmax (KBr) cm⁻¹: 1660, 1485, 1440, 1140, 1080, 1055, 1010 and 880; MS m/z 416, 414, 412, 410 (M⁺) for C15H21OBr2Cl (HRMS); 1H NMR δ (CDCl3): 1.15 (s, 3H), 1.18 (s, 3H), 1.60 (s, 3H), 1.67 (s, 3H), 2.10~2.50 (m, 6H), 2.90 (d, 1H, J=3 Hz), 4.65 (dd, 1H, J=13, 5 Hz), 6.22 (1H, d, J=3 Hz). Based on the spectral data, compound 1 was identified as deoxyprepacifenol, which had previously been isolated from the sea hare Aplysia californica.

The second active fraction eluted with hexane-ethyl acetate (7:1, v/v) contained a mixture of two active principles (2 and 3), which were separated from each other by preparative silica gel thin-layer chromatography to afford 80mg of pure compound 2 and 120mg of compound 3. Compound 2 showed the following spectral data: mp 80~82°C; [α]D25 = +96° (c=0.25, CCl4); MS m/z 394, 392, 390 (M⁺) for C15H20O2Br2 (HRMS); IR vmax (KBr) cm⁻¹: 3300, 2100, 1140, 1085, 1045 and 975; 1H-NMR δ

Fig. 1. Structures of Compound 1~6.
Table I.  Insecticidal Activity of Compounds 1 ~ 6 against Mosquito Larvae (Culex pipiens pallens)

<table>
<thead>
<tr>
<th>Compound no.</th>
<th>Lethal activity( LC_{50} ) (ppm)</th>
<th>Emergence inhibitory activity( IC_{50} ) (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>6.83</td>
<td>0.48</td>
</tr>
<tr>
<td>2</td>
<td>2.86</td>
<td>0.06</td>
</tr>
<tr>
<td>3</td>
<td>6.14</td>
<td>0.50</td>
</tr>
<tr>
<td>4</td>
<td>&gt;10</td>
<td>&gt;10</td>
</tr>
<tr>
<td>5</td>
<td>&gt;10</td>
<td>&gt;10</td>
</tr>
<tr>
<td>6</td>
<td>&gt;10</td>
<td>&gt;10</td>
</tr>
</tbody>
</table>

* The lethal activity was observed at 24 hr after treatment.

**Experimental**

Melting points (mp) were measured on a Mitamura Riken micro-melting point apparatus and are uncorrected.

Mass spectra were obtained with a Hitachi M-003 spectrometer, and IR spectra were recorded on a Hitachi 260-10 spectrometer in KBr pellets. Specific rotations were measured with a Jasco DIP-181 polarimeter. \(^1\)H and \(^13\)C NMR spectra were recorded on a Hitachi R90H instrument, using TMS as an internal standard.

**Bioassay.** The immersion method was adopted for the bioassay using the mosquito larvae (Culex pipiens pallens), 10 mg of each tested sample being dissolved in a mixture of \( \text{H}_2\text{O} \), xylene and Sorpol \(^8\) (5 ml). A portion of the solution (0.5 ml) was added to 100 ml of deionized water, into which 20 last-instar larvae were pipetted. The mortality was assessed after 24 hr, and the surviving larvae were reared until their emergence. The inhibitory effect was also observed.

**Isolation.** The dried L. nipponica specimens (180 g) collected at Nomo in Nagasaki Prefecture in March 1982 were extracted with methanol. The extract was dried in vacuo to give 9.5 g of an oily residue which was partitioned between \( \text{H}_2\text{O} \) and ethyl acetate. The ethyl acetate layer was concentrated to give 2.0 g another oily residue. This residue was chromatographed on silica gel (100 g) with hexane-ethyl acetate as the eluent. The fraction eluted with 9:1 hexane-ethyl acetate (v/v) was purified by preparative thin-layer chromatography on silica gel 60 (HRMS); IR \( \nu_{\text{max}} \) (KBr) cm\(^{-1} \): 3300, 2150, 1130, 1110 and 1095; \(^1\)H NMR and \(^13\)C NMR spectra were recorded on a Hitachi R90H instrument, using TMS as an internal standard.

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Three Insecticidal Principles from *Laurencia nipponica Y.*

References


