

A Novel Antimicrobial Substance in Etiolated Seedlings of Adlay

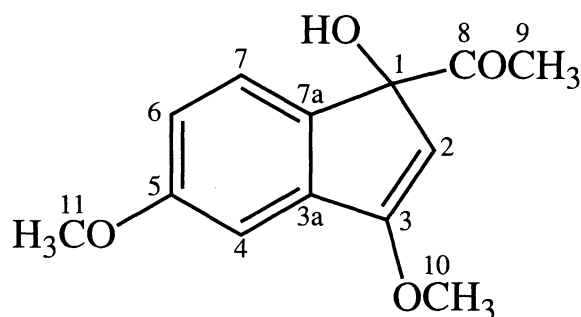
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A novel antimicrobial substance was isolated from the extract of etiolated seedlings of Adlay (*Coix lachryma-jobi* L. var. *ma-yuen* Stapf). This substance showed antimicrobial activity against bacteria, but no activity against yeast and fungi.

Recently, many antimicrobial substances have been isolated from plants.¹⁻⁸ We have found a novel antimicrobial substance from the methanol extract in etiolated seedlings of Adlay (*Coix lachryma-jobi* L. var. *ma-yuen* Stapf). In this report, we describe the isolation and structure elucidation procedures of this substance (1).



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The seeds of Adlay were germinated at 25 °C for 5 days in the dark. The etiolated seedlings (150 g) which were lyophilized, were extracted twice with methanol at room temperature for 2 days and filtered. The filtrate was concentrated *in vacuo*. The residue was dissolved in chloroform and methanol (2:1) by Folch partition method.⁹ The chloroform fraction was sequentially partitioned into acetone, ether, and pyridine fractions. The acetone fraction, which showed the highest antimicrobial activity, was chromatographed on GPC, was purified by reversed phase HPLC. Finally, the antimicrobial substance (1.5 mg) was isolated. The molecular formula C₁₃H₁₄O₄ of this substance was assigned by CI-MS of ion peaks [m/z 219 ([M-CH₃)⁺], 235 (M+H)⁺, by EI-MS of ion peaks [m/z 190 ([M-H-CH₃CO]⁺), 191 ([M-CH₃CO]⁺), 219 ([M-CH₃)⁺], 233 ([M-H]⁺), and by HR-EIMS of molecular ion peak [m/z 233.0723 (calcd 233.0814) ([M-H]⁺)]. From ¹H-NMR ((CD₃)₂CO) [δ 2.01 (3H, s, 1-COCH₃), 3.84 (3H, s, 5-OCH₃), 4.00 (3H, s, 3-OCH₃), 5.68 (s, 2-H), 7.08 (1H, dd, J=8.55, 2.67Hz, 6-H), 7.37 (d, J=2.67Hz, 4-H), 7.57 (d, J=8.55Hz, 7-H)], ¹³C-NMR ((CD₃)₂CO) [δ 31.6 (q, 8-CH₃), 55.9 (q, 5-OCH₃), 57.1 (q, 3-OCH₃), 76.0 (s, 1-C), 99.1 (d, 2-C), 109.5 (d, 4-C), 117.2 (d, 6-C), 128.7 (d, 7-C), 129.0 (s, 3a-C), 137.0 (s, 7a-C), 160.3 (s, 5-C), 168.3 (s, 3-C), 205.0 (s, 8-C)], HMBC (2-H / 1, 3, 3a-C, 4-H / 3, 6, 7a-C, 6-H / 4, 7a-C, 7-H / 1, 5, 3a-C, 9-H / 8-C, 10-H / 3-C, 11-H / 5-C), and FT-IR (1569,

1712, 3401 cm^{-1}) spectra of this substance, we proposed the structure as 1-acetyl-1-hydroxy-3,5-dimethoxy-1*H*-inden **1**. The ^{13}C -NMR spectrum of this substance which has an inden skeleton, agreed with that of 1*H*-inden.¹⁰⁾ This substance is a new type antimicrobial substance of an inden skeleton. The antimicrobial activity of this substance against several microorganisms was measured by the paper disc method.¹¹⁾ This substance showed antimicrobial activity against bacteria such as *Bacillus subtilis* IFO 3009, but no activity against yeast and fungi such as *Saccharomyces cerevisiae* IFO 0304 and *Aspergillus niger* JCM 5697.

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