



Enantiomeric ratio of sesamin in the bark of *Pentaspadon motleyi* (*Anacardiaceae*)

(Rasio Senyawa Enansiomer sesamin di dalam kulit batang *Pentaspadon motleyi* (*Anacardiaceae*))

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Abstract: The barks of *Pentaspadon motleyi* Hook. f. (*Anacardiaceae*) were collected from the Poso district in Central Sulawesi, Indonesia, in 1992 which it has been used traditionally as a remedy for malaria and tuberculosis in the local area. It was found that a mixture of (+)-sesamin (**1**) and (-)-sesamin (**2**) in a ratio of 100:86 were present together with lupeol in the bark of *Pentaspadon motleyi*. This was the first evidence of sesamin being isolated in almost equivalent amounts of (+)- and (-)-forms from a single plant species, and indicates that the biosynthetic pathway of sesamin in the plant is not enantioselective or fuzzy.

Keywords: (+)-sesamin, (-)-sesamin, enantiomeric ratio, *Pentaspadon motleyi*, *Anacardiaceae*.

Abstrak: Kulit batang *Pentaspadon motleyi* Hook. F (*Anacardiaceae*) yang dikoleksi dari daerah Poso (Sulawesi Tengah), Indonesia tahun 1992 dan secara tradisional digunakan untuk pengobatan malaria dan tuberkolosis. Senyawa kimia dalam bentuk campuran (+)-sesamin (**1**) dan (-)-sesamin (**2**) dengan perbandingan 100:86 bersama dengan senyawa lupeol telah ditemukan dari kulit batang *Pentaspadon motleyi*. Ini adalah laporan yang pertama kali bahwa sesamin yang diisolasi yang mempunyai jumlah hampir sama antara (+)-sesamin dan (-)-sesamin dari satu spesies tanaman ini, dan ini menunjukkan bahwa jalan biosintesis sesamin dalam tanaman tersebut tidak enansioselektif atau tidak begitu jelas.

Kata kunci: (+)-sesamin, (-)-sesamin, rasio enansiomer, *Pentaspadon motleyi*, *Anacardiaceae*.

INTRODUCTION

(+)-SESAMIN (**1**), a furofuran-type lignan, was first reported as a chemical constituent of sesame oil at the end of the 19th century^(1,2). Since then, several pharmacological activities attributed to the anti-oxidative effect⁽³⁾ of (+)-sesamin have been reported and (+)-sesamin has received much attention from the viewpoint of alternative medicines.

Biosynthetic research of lignans⁽⁴⁾ has revealed

that (+)-sesamin (**1**) is biosynthesized through two steps, **a•b-i** and **c-i** (Figure 1). The first step is the formation of a free radical at the β -position of the coniferyl alcohol (**3**) and the subsequent stereoselective coupling of the radicals^(5,6) via an intermediate **i** to afford (+)-pinosresinol (**4**). The second step is the formation of a methylenedioxy ring⁽⁷⁾ that enables a reaction from **4** to (+)-sesamin (**1**).

On the other hand, isolation of the antipode (-)-sesamin (**2**) from a plant has rarely been reported⁽⁸⁾, and detailed studies on the biosynthetic pathway of (-)-sesamin have never been reported. However, it could be considered that the pathway of **2** is similar to

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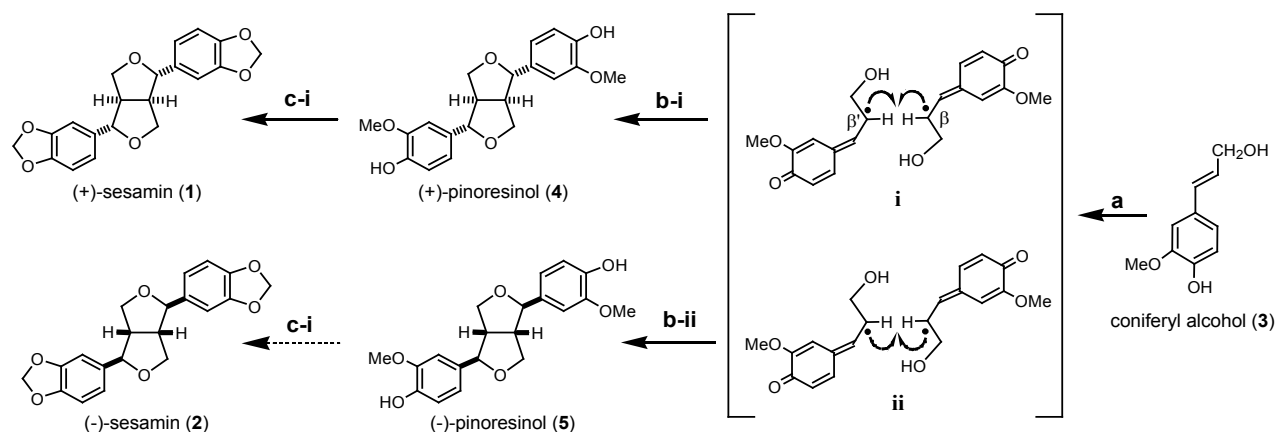


Figure 1. Biosynthetic pathway of (+)-sesamin (**1**) and the plausible pathway of (-)-sesamin (**2**). [a: free radical formation at the β -position; b-i and b-ii: stereoselective coupling of the radicals; c-i and c-ii: formation of methylenedioxy ring].

that of (+)-sesamin (**1**); in particular, **2** is biosynthesized through conversion of coniferyl alcohol (**3**) via an intermediate **ii** to (-)-pinoresinol (**5**), and then from **5** to **2**, as shown in steps a•b-ii and c-ii in Figure 1.

Isolation of (+)-sesamin (**1**) and (-)-sesamin (**2**) from a single plant species has been reported in two papers^(9,10). In both cases, it was reported that (+)-sesamin (**1**) was predominantly obtained, together with small amounts of (-)-sesamin (**2**), but the enantiomeric ratio were not reported.

In this paper, we report the isolation of (+)- and (-)-sesamins (**1** and **2**, respectively) in a mixture of almost equivalent amounts from the bark of the *Anacardiaceae* species, *Pentaspadon motleyi* Hook. f.

MATERIALS AND METHODS

MATERIALS. Plant. The barks of *Pentaspadon motleyi* Hook. f. were collected from the Poso district in Central Sulawesi, Indonesia, in 1992 and were identified at the Herbarium Bogoriense, Research Center for Biology, Indonesian Institute of Sciences, Indonesia. *P. motleyi* has been used traditionally as a remedy for malaria and tuberculosis in the local area.

Instruments. Optical rotations were measured with a JASCO DIP-360 digital polarimeter. FAB-MS was obtained on a JMS 102 A mass spectrometer. IR spectra were run on a Shimadzu FT-IR 8500 spectrometer, and UV spectra on a Hitachi U-3500 spectrometer. ¹H and ¹³C-NMR spectra were recorded on a JEOL JNM-Lambda 500 spectrometer (500 MHz and 125.65 MHz, respectively). HPLC was performed on a Hitachi D-7000 system. Column chromatography was carried out on Silica gel 60 (230-400 mesh, Merck).

METHODS. Extraction and Isolation. The barks of *P. motleyi* (350 g) were extracted three times with 700 mL of hot methanol. The combined extract solution was evaporated under reduced pressure to give a methanol

extract (42 g, 12% from the bark), and the methanol extract (2.0 g) was partitioned into ethyl acetate-water (1:1) to produce an ethyl acetate-soluble portion (1.02 g, 6.1%) and a water-soluble portion (0.98 g, 5.9%). The ethyl acetate-soluble portion was subjected to silica gel column chromatography (SiO₂ 100 g) elution with *n*-hexane-ethyl acetate (20:1) to (2:1) and then further purified by semi-preparative HPLC (YMC-Pack SIL) using *n*-hexane-ethyl acetate (19:1) to produce sesamin fraction (90 mg, 0.54%) and with lupeol (110 mg, 0.66%). A portion of the sesamin fraction (25 mg) was separated on HPLC system (Chiral Pak IC) with mobile-phase of *n*-hexane-ethanol (4:6) with a flow-rate of 0.5 mL/min and UV detector at 220 nm to produce (+)-sesamin (**1**, 13.4 mg, 0.29% of the bark) and (-)-sesamin (**2**, 11.6 mg, 0.25%). The isolated sesamins (**1** and **2**) were directly identified by comparison of physicochemical data of authentic (+)-sesamin and (-)-sesamin, respectively, such as melting point (**1**, 121-122 °C; **2**, 121-122 °C) and optical rotation [α]_D (**1**, +63.7°, c 1.03 in CHCl₃; **2**, -67.5°, c 1.01 in CHCl₃) as well as ¹H & ¹³C-NMR spectra^(1,2).

RESULTS AND DISCUSSION

The methanol extract of the bark of *Pentaspadon motleyi* was partitioned into a mixture of ethyl acetate and water to give an ethyl acetate-soluble portion (6.1% from the bark) and a water-soluble portion (5.9%). The ethyl acetate-soluble portion was separated by silica gel column chromatography and subsequent HPLC using a normal-phase adsorbent to afford a sesamin fraction (0.54%) together with lupeol (0.66%). The spectroscopic data (FAB-MS, IR, UV, ¹H-NMR, ¹³C-NMR) of the sesamin fraction was the same as those of the authentic (+)-sesamin. However, the absolute value of the optical rotation, [α]_D +4.6°, was smaller than that of the optically active (+)-sesamin. Therefore, we subjected

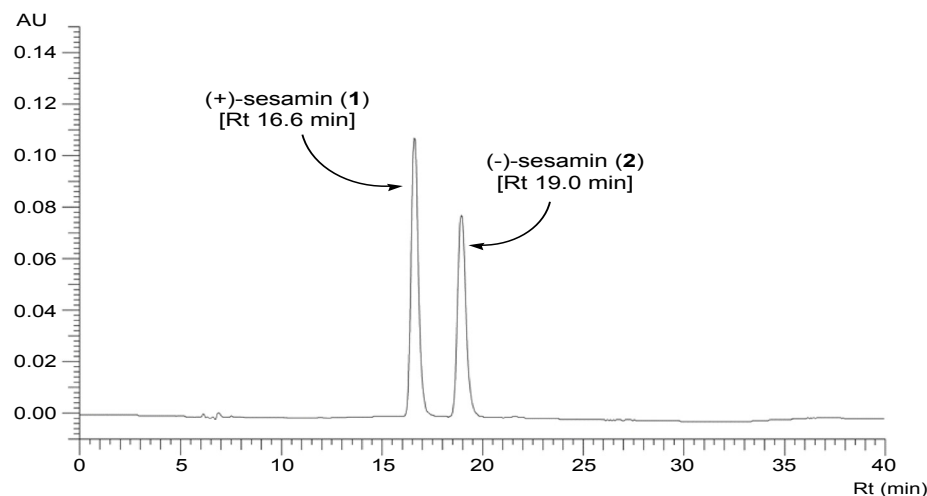


Figure 2. HPLC chromatogram of the sesamin fraction from the bark of *Pentaspadon motleyi*. Conditions: Chiral Pak IC (4.6 mm x 250 mm); flow-rate: 0.5 mL/min; *n*-hexane-ethanol 4:6; UV detector at 220 nm.

the sesamin fraction to HPLC using a chiral adsorbent to find that the enantiomeric ratio of (+)-sesamin (**1**) and (-)-sesamin (**2**) was 100:86, as shown in Figure 2. In fact, the HPLC separation of the sesamin fraction gave (+)-sesamin (**1**, 0.29% from the bark) and (-)-sesamin (**2**, 0.25%). The physicochemical properties were identified as those of the authentic (+)-sesamin and (-)-sesamin, including the optical rotations [**1**, $[\alpha]_D +63.7^\circ$; **2**, $[\alpha]_D -67.5^\circ$]^(1,2).

The present study found that sesamin isolated from a single plant species, *Pentaspadon motleyi* (Anacardiaceae), had a respective enantiomeric ratio of 100:86 for (+)-sesamin (**1**) and (-)-sesamin (**2**). The results also indicated that the key biosynthetic steps of **b-i** and **b-ii** in Figure 1, leading to (+)-sesamin (**1**) and (-)-sesamin (**2**) respectively, are not enantioselective or fuzzy in the plant.

Finally, it should be noted that this study presents the first evidence of the furofuran-type lignin, sesamin, in an Anacardiaceous plant. In addition, the content yield (0.54%) of the sesamin fraction is by no means inferior to that of sesame seeds.

CONCLUSION

Isolation of a mixture of (+)-sesamin and (-)-sesamin from the bark of *Pentaspadon motleyi* Hook. F. (Anacardiaceae) which was collected from Poso district in Central Sulawesi, demonstrated a ratio of 100:86.

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