

**STRUCTURE OF STEROIDS**  
**IN *Stelechocarpus burahol* Hook f. & Thomson STEM BARK**  
**Struktur steroid dalam kulit batang**  
***Stelechocarpus burahol* Hook f. & Thomson**

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**ABSTRAK**

Steroids alam digunakan dalam berbagai produk farmasi, terutama sebagai material awal untuk sintesis parsial hormon reproduksi termasuk oral kontrasepsi. Dalam penelitian ini steroid ditemukan sebagai produk sampingan dari proses isolasi dan identifikasi senyawa sitotoksik kulit batang *Stelechocarpus burahol* Hook F & Thomson. Campuran steroid berupa serbuk putih yang terdiri dari 3 komponen : $\Delta$ 5-ergostenol, stigmasterol, dan  $\beta$ -sitosterol. Elusidasi struktur menggunakan GCMS dan <sup>1</sup>H-NMR. Hasilnya dibandingkan dengan literatur standar.

**Kata kunci:** steroid, *Stelechocarpus burahol*

**ABSTRACT**

Natural steroids can be used for pharmaceutical production. Especially served as starting material for their partial synthesis of sex hormones, including the oral contraceptives. In this research, steroid was obtained as by-products found during isolation and identification of cytotoxic compounds from *Stelechocarpus burahol* Hook f. & Thomson stem bark. The steroid mixture appears as white powder, consist of 3 steroid components:  $\Delta$ 5-ergostenol, stigmasterol, and  $\beta$ -sitosterol. Structure elucidation has been done using GC-MS and <sup>1</sup>H-NMR spectroscopic. The result was compared with literature standard.

**Key words:** steroid, *Stelechocarpus burahol*

**INTRODUCTION**

Steroid extracted from plants has important roles on cortison hormon synthesis or sex hormon. For example, diosgenin from dioscorea plant can be synthesized to become progesterone through Marker Degradation. Progesteron hormon is highly needed for family planning. In this research, streoid compound was found during isolation and identification of

cytotoxic compound from burahol stem bark, *Stelechocarpus burahol* Hook f. & Thomson (Clara, 2003). The steroid was obtained in non toxic fraction against *Artemia salina* Leach (Brine shrimp lethality bioassay), the E2-5 fraction. In dilution process of E2-5 fraction using methanol, there was white crystall that gave positif reaction to vanilin-H<sub>2</sub>SO<sub>4</sub> reagent.

## METHODS

### Material

EI and CIMS (direct) and HRMS were determined on JMS AX500 mass spectrometer at 70 eV.  $^1\text{H-NMR}$  spectrum was recorded at 400 MHz and 100 MHz on a JEOL Lambda 400 instruments using DMSO- $d_6$ . TLC chromatographic analysis was out on precoated Silica gel 60 F254 plates (E. Merck). Whereas visualization of the TLC plates was performed using Vanillin - H $_2$ SO $_4$  spray reagent, and UV Lamp 254 nm. Fraction E $_{2-5}$

### Method

Fraction E $_{2-5}$  was diluted in methanol, resulted in white crystal. This crystal was recrystallized with acetone, obtaining SB $_4$  as white needle crystal, and was monitored by Thin Layer Chromatography, using reference standard stigmasterol and  $\beta$ -sitosterol. Afterwards SB $_4$  was identified by using  $^1\text{H-NMR}$  and GC-Mass Spectrometry.

## RESULT AND DISCUSSION

Compound SB $_4$  was obtained as white needle crystal, the TLC chromatogram showed one spot which give positive reaction to the steroid by using vanillin-H $_2$ SO $_4$  spray reagent, giving violet color. The  $^1\text{H-NMR}$  spectrum performed a specific characteristic for the existence of steroid component. (Fig. 1).

The GC chromatogram showed 3 peaks with comparative of the peak width 1:4.1:6.1 (Fig.2). The first peak has 33.68 minutes retention time, that in the EI-MS spectrum has similarity type with ergost-5-en-3-ol ( $3\beta$ ) or  $\Delta^5$ -ergostenol, MW 400, with 99% equality (Fig.

3). The second peak has 33.98 minutes retention time, that in the EI - MS spectrum has similarity type with 5,2-dien-3-ol ( $3\beta$ -22E) or stigmasterol, MW 412, with 96% equality (Fig. 4). The third peak has 34.56 minutes retention time, that in the EI - MS spectrum has similarity type with (23S)-ethylcholest-5-en-3- $\beta$ -ol or  $\beta$ -sitosterol, MW 414, with 99% equality (Fig. 5).

It can be concluded that SB $_4$  (white needle crystal) is a mixture of 3 steroid compounds that consist of  $\Delta^5$ -ergostenol, stigmasterol and  $\beta$ -sitosterol. According to The Merck Index, 2001, the structures of the 3 steroids are in Fig.6, 7, and 8.

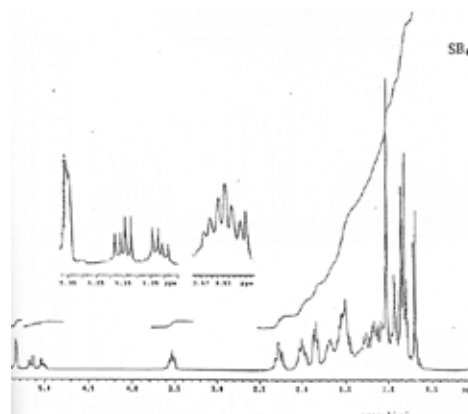


Figure 1. Spectrum  $^1\text{H-NMR}$  SB $_4$

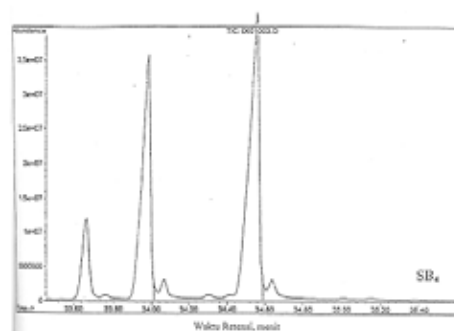


Figure 2. GC chromatogram SB $_4$

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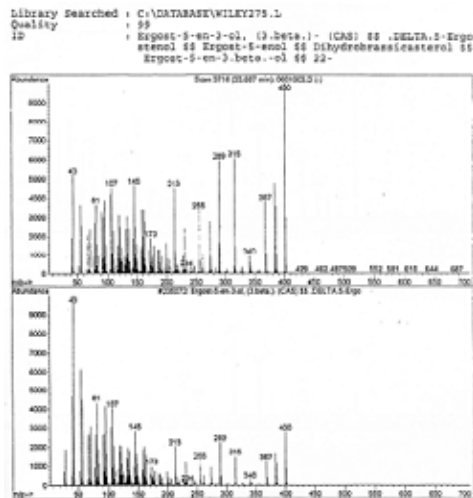


Figure 3. MS spectrum Peak 1 (RT=33.68 minutes) identical with  $\Delta^5$ ergostenol

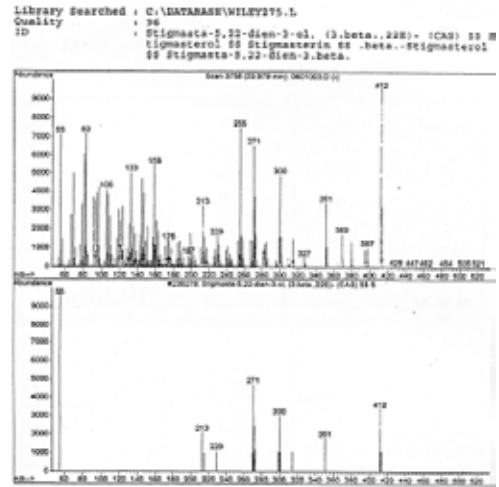


Figure 5. MS spectrum Peak 3 (RT=34.56 minutes) identical with  $\beta$ -sitosterol

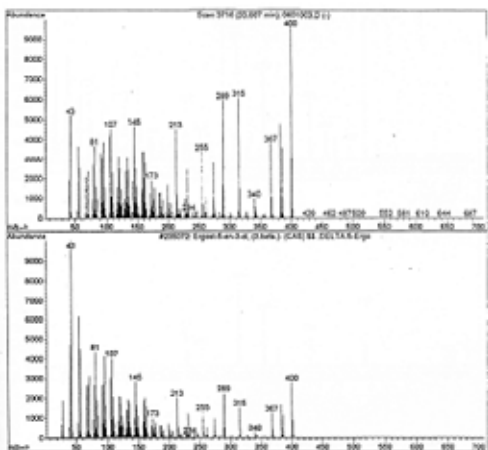


Figure 4. MS spectrum Peak 2 (RT=33.98 minutes) identical with stigmasterol

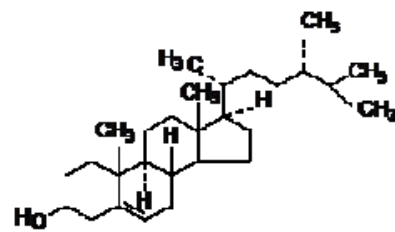


Fig. 6. Structure of  $\Delta^5$ ergostenol (The Merck Index, 2001)

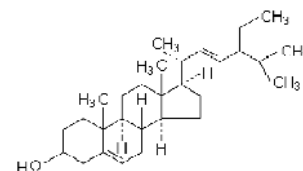


Fig. 7. Structure of Stigmasterol (The Merck Index, 2001)

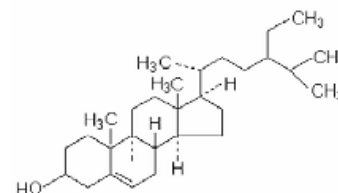


Fig. 8. Structure of  $\beta$ -sitosterol (The Merck Index, 2001)

## LITERATURES

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The Merck Index. 2001. *An Encyclopedia of Chemicals, Drugs, and Biologicals*, Thirteenth Edition, Merck & CO., Inc., Whitehouse Station, NJ., 8627, 8893