SPINASTEROL : STEROIDS FROM *Filicium decipiens* STEM BARK

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ABSTRACT

The aim of the study was to isolate and structure elucidate compound from extract of Filicium decipiens stem bark, Sapindaceae family. A stigmasta-7,22-dien-3-ol (spinasterol) was isolated from the Filicium decipiens, Sapindaceae family. The fractionation of ethyl acetate stem bark extracts by colomn chromatography led to the isolation of Compound 1 (32 mg). The white powder of Compound 1 had melting point 168.1 – 168.3°C. The structure was established on the basis of UV, IR, NMR spectral data and by comparison with literature data. The isolated compound was concluded as stigmasta-7,22-dien-3-ol (spinasterol).

Keywords – *Filicium decipiens, Sapindaceae, steroid, NMR*

1. INTRODUCTION

Filicium decipiens (Wight et Arn.) Thwaites (Familiy : Sapindaceae) also called fern leaf tree is a tree which growing in tropic zones like Asia and Africa (Figure 1). This plant is commonly found in areas with altitudes up to 1000 meters. *Filicium decipiens* has 25 meters gray brown stem with a lot of branches and cracked bark. The leaves are compound with 10 – 24 leaflet and is 15-20 cm each leaflet in lenght. Dark green leaves has a smooth upper surface, the bottom shiny and wavy leaf edge.

Fig 1. *Filicium decipiens* (Wight et Arn.) Thwaites

In Indonesia, *Filicium decipiens* commonly found in roadside and green open spaces. This plant has a high reduction of the lead which is emissions from motor vehicles, making it great to use as a pollution-absorbing trees. This is called ki sabun in Indonesia because all

parts of the plant contain saponin compounds[1]. The high polar secondary metabolites of *Filicium decipiens* make this plant has high antioxidant activity and triterpenoidal saponin/steroid have good anti-inflammantory effect[2]. *Filicium decipiens* is traditionally used for antidiabetic in India and Srilanka[3].

Four triterpenoidal saponins[4] and norneohopane caffeate[5] have been reported from stem bark methanolic extract of *Filicium decipiens*. Sitosterol and flavonol glycosides were identified from leaves and fruit of *Filicium decipiens*[2,6]. In this paper, we report the isolation and structure elucidation of a new compound of *Filicium decipiens* species, stigmasta-7,22-dien-3-ol or spinasterol (Figure 2) from ethyl acetate extract of the stem bark.

2. MATERIALS AND METHODS

2.1 General experimental procedures

UV spectra were obtained using a Shimadzu® UV1700 spectrophotometer. 1D dan 2D NMR spectra were recorded in $CDCl_3$ on a JEOL spectrometer (500 MHz) using TMS as internal reference. IR spectra were recorded on a Thermoscientific FTIR spectrometer using KBr pellets. Melting point were determined on Gallenkamp apparatus. Colomn chromatography was performed on silica gel 60, 70-230 mesh, ASTM (Merck). TLC was performed on precoated aluminium sheets, silica gel 60 $PF_{254 +365}$ (Merck), detected under UV light 254 and 365 nm.

2.2 Plant material

The stem bark of *Filicium decipiens* was collected in Andalas University, Padang, West Sumatera, Indonesia, November 2014. The plant has been authentified by Dr. Nurainas in Herbarium Laboratory of Andalas University (ANDA).

2.3 Extraction and Isolation

The air-dried powdered stem bark of *Filicium decipiens* (5 kg) were extracted by maceration with n-hexane, ethyl acetate and methanol at room temperature. The combined of each extracts were concentrated under reduce pressure to afford n-hexane (15 g), ethyl acetate (31 g) and methanol (280 g) extracts. The ethyl acetate extract (15 g) was chromatographed on a flash colomn of silica gel (200 g), eluting with n-hexane and ethyl acetate (5:5). Twenty fractions collected were group into twelve according to their TLC profile. Fraction 3 was rechromatographed on silica gel (15 g), eluting with step polarity n-hexane and ethyl acetat and got ten group fractions. Fraction C_4 dissolved with n-hexane and the solid was dissolved with aceton to get Compound 1 (32 mg).

Compound 1: White powder, mp 168.1-168.3°C. The Liebermann Burchard test indicated the steroid compound. IR v_{max} (KBr) in cm^{-1} : 1370.17, 1457.84 (-CH(CH$_3$)$_2$), 1657.84 (-C=C-), 3422.68 (-OH), ^{1}H NMR (500 MHz, $CDCl_3$) δ (ppm) : 0.55, 0.79, 0.80, 0.82, 0.85, 1.02 (each 3H, s, Me x 6), 3.61-3.58 (1H, m, H-3), 5.02 (1H, dd, H-23) and 5.15, 5.17 (2H, dd, H-7 and H-22). ^{13}C NMR (500 MHz, $CDCl_3$) δ (ppm): Table 1. This compound was identified as stigmasta-7,22-dien-3-ol (spinasterol) according to previously published spectral data[7].

3. RESULTS AND DISCUSSION

Compound 1 was isolated as a white powder with melting point 168.1 – 168.3°C. The ^{1}H NMR spectrum showed three olefinic protons at δ 5.02, 5.15, 5.17 ppm, a hydroxyl methyne at δ 3.58 – 3.61 ppm and six methyl protons at δ 0.55, 0.79, 0.80, 0.82, 0.85, 1.02 ppm. The ^{13}C NMR spectral showed twenty nine carbons with six primary carbons, nine secondary carbons, eleven tertiary carbons and three quartenary carbons by ^{13}C DEPT. The spectral indicated four olefinic carbon at δ 117. 64, 129.60, 138. 37, 139.75 ppm (C-7, C-23, C-22,

C-8), a hydroxyl carbon at δ 71.25 ppm (C-3) and six methyl carbon at δ 12.23, 12.44, 13.23, 19.18, 21.29, 21.56 ppm (C-18, C-29, C-19, C-27, C-26, C-21). The ^{13}C NMR spectral data of Compound 1 was compared with stigmasta-7,22-dien-3-ol (spinasterol) from *Cucurbita maxima* flowers and Compound 1 was assigned as spinasterol compound.

The Heteronuclear Multiple Bond Connectivity (HMBC) spectral showed correlation between H-22 with C-20, C-24, C-23; H-23 with C-20, C-24, C-22; and H-21 with C-20, C-22 (Figure 3). The [1]H-^{1}H Correlated Spectroscopy (COSY) showed correlation between H-3 with H-2,H-5; H-7 with H-6; H-22 with H-21, H-23; and H-23 with H-22, H-24 (Figure 4).

Table 1 : The 500 MHz ^{13}C NMR spectral data of Compound 1 from *Filicium decipiens* stem barks and 100 MHz ^{13}C NMR spectral data of spinasterol from *Cucurbita maxima* flowers.

| δ_C Compound 1	Types of carbon	δ_C Spinasterol[7]
139.7514	C	139.6 (C-8)
138.3683	CH	138.1 (C-22)
129.6026	CH	129.5 (C-23)
117.6415	CH	117.5 (C-7)
71.2471	CH	71.1 (C-3)
56.0562	CH	55.9 (C-17)
55.2990	CH	55.1 (C-14)
51.4265	CH	51.2 (C-24)
49.6046	CH	49.5 (C-9)
43.4620	C	43.3 (C-13)
41.0392	CH	40.8 (C-20)
40.4288	CH	40.3 (C-5)
39.6276	CH$_2$	39.6 (C-12)
38.1491	CH$_2$	38.0 (C-4)
37.3097	CH$_2$	37.2 (C-1)
34.3910	C	34.2 (C-10)
32.0637	CH	31.9 (C-25)
31.6440	CH$_2$	31.5 (C-2)
29.8126	CH$_2$	29.7 (C-6)
28.7062	CH$_2$	28.5 (C-16)
25.5871	CH$_2$	25.4 (C-28)
23.2026	CH$_2$	23.0 (C-15)
21.7241	CH$_2$	21.6 (C-11)
21.5620	CH$_3$	21.4 (C-21)
21.2949	CH$_3$	21.1 (C-26)
19.1774	CH$_3$	19.0 (C-27)
13.2350	CH$_3$	13.0 (C-19)
12.4433	CH$_3$	12.2 (C-29)
12.2335	CH$_3$	12.0 (C-18)
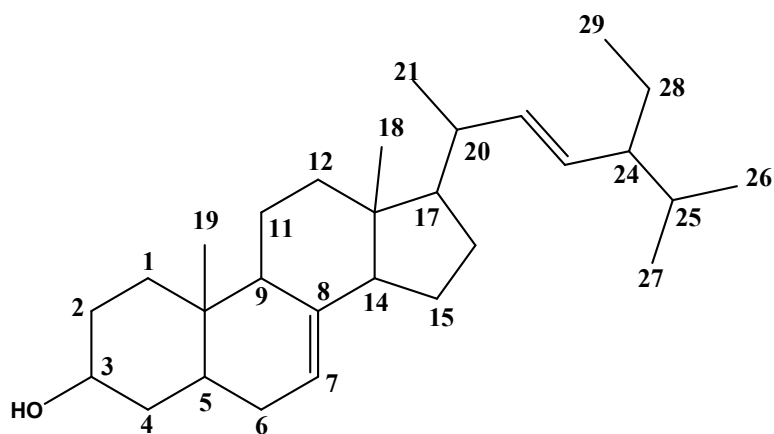
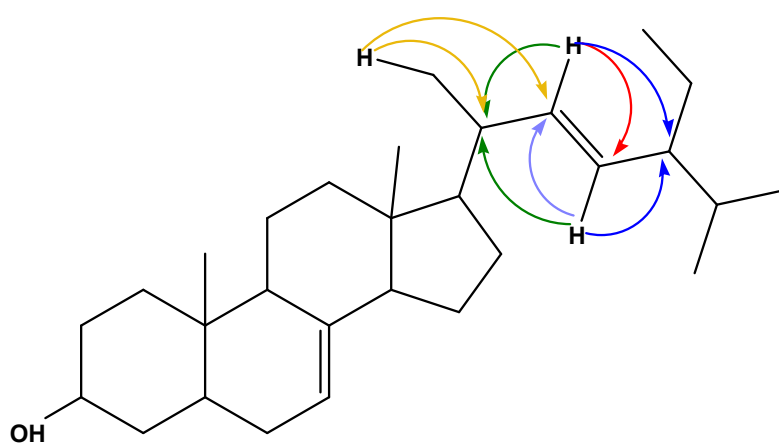Fig 2. Compound 1 (stigmasta-7,22-dien-3-ol or spinasterol) structure
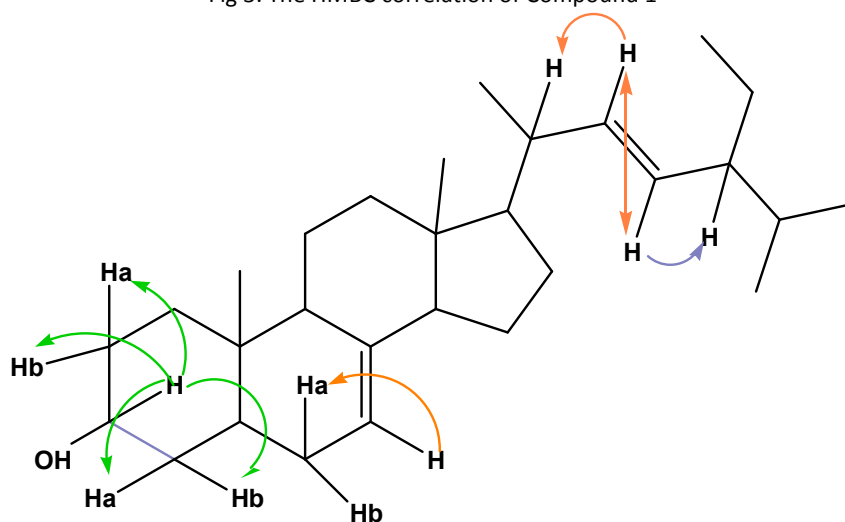

Fig 3. The HMBC correlation of Compound 1

Fig 4. The ^{1}H-^{1}H COSY correlation of Compound 1

4. CONCLUSION

Steroid compound namely stigmasta-7,22-dien-3-ol (spinasterol) has been isolated from ethyl acetate extract of *Filicium decipiens* stem bark. This compound is the first reported from this species.

REFERENCES

1. Eka A., Adji R., Informasi Singkat Benih, 2012, 137: 1 – 2.

2. Sasikala C., Sudhakar Y., Indian Journal of Pharmaceutical Education and Research, 2015, 146 – 151.

3. Lavaud C., Voutquenne L., Massiot G., Serani L., Becchi M., Phytochemistry, 1997, 47(3): hal. 441 – 49.

4. Jayasinghe U.L.B., Bandara A.G.D., Hara N., Fujimoto Y., Fitoterapia, 2001, 72: 737 – 742.

5. Jayasinghe U.L.B., Bandara A.G.D., Fujimoto Y., Natural Product Research, 2004, 18(6): 499 – 502.

6. Jayasinghe U.L.B., Amarasinghe N.R., Arudathie B.G.S., Ayona N.H., Fujimoto Y., Natural Product Research, 2012, 26(8): 717 – 721.

7. Ragasa C.Y., Lim K., Philippine Journal of Sciences, 2005, 134(2): 83 – 87.