



STIGMASTEROL ISOLATION AND ANTICONVULSANT ACTIVITY OF METHANOL EXTRACT FROM THE AERIAL PARTS OF *INDIGOFERA STENOPHYLLA* (FABACEAE)

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Introduction: *Indigofera stenophylla*, a member of the Fabaceae family, has been traditionally used in folk medicine for various ailments, including epilepsy among others. This study aimed to isolate and characterize phytochemical(s) from the methanol aerial parts extract of *Indigofera stenophylla* (MAEIS) and evaluate its anticonvulsant potential. **Methods:** The MAEIS affords the isolation of compound AR2 through chromatography. The anticonvulsant effect of MAEIS was determined using pentylenetetrazol (PTZ) and strychnine (SCN) induced seizure models. **Results:** The spectroscopic analysis of the compound AR2 affords 3 β , 22-stigmasta-5, 22-dien-3-ol (stigmasterol). The MAEIS demonstrated significant anticonvulsant activity at graded doses of 150, 300, and 600 mg/kg, reducing both seizure onset and duration in PTZ and SCN models at $p < 0.05$ and $p < 0.001$. The 600 mg/kg dosage provided the most substantial protection. **Conclusion:** The column chromatographic and spectroscopic analysis of MAEIS led to the isolation of stigmasterol which is a potential bioactive compound with anticonvulsant activity. This is the first isolation of stigmasterol from the plant.

Keywords: *Indigofera stenophylla*; Stigmasterol; Isolation; Characterization; Anticonvulsant

INTRODUCTION

Epilepsy is a chronic neurological condition characterized by unpredictable seizures, which can be either provoked or unprovoked. This disorder arises from abnormal electrical activity in the brain, often due to recurrent and spontaneous discharges of a group of neurons, leading to seizure episodes in patients¹. Epilepsy affects approximately 50 million people worldwide, making it one of the most prevalent neurological disorders². The global annual incidence ranges from 20 to 70

new cases per 100,000 people³. In Africa, around 14 million individuals are estimated to have epilepsy, with incidence rates varying between 50 and 100 new cases per 100,000 annually^{4,5}. Nigeria, in particular, has one of the highest prevalence rates in Africa, ranging from 4 to 10 per 1,000 people, and an incidence rate estimated between 50 and 100 new cases per 100,000 each year⁶. These statistics underscore the significant public health challenge posed by epilepsy, particularly in sub-Saharan Africa.

Treatment options for epilepsy primarily include antiepileptic drugs (AEDs), targeting different mechanisms in the pathophysiology of the diseases. For patients with drug-resistant epilepsy, surgical interventions like resective surgery and corpus callosotomy may be considered^{7,8}. Additional treatments include vagus nerve stimulation⁹ and responsive neurostimulation¹⁰. Alternative therapies, such as the ketogenic diet, have shown effectiveness, particularly in children¹¹, while cannabidiol (CBD) has demonstrated promise in reducing seizures¹². Techniques like biofeedback and neurofeedback may also help to manage seizures¹³, alongside mindfulness practices, including yoga, that can help reduce stress-related seizure triggers¹⁴. Natural sources offer promising alternatives for managing the condition, particularly medicinal plants that remain largely underutilized despite numerous ethnomedicinal claims^{15,16}.

Indigofera stenophylla has been utilized in folk medicine to address a variety of health concerns, such as bacterial infections, pain relief, inflammation, epilepsy, and even cancer¹⁷. Its medicinal applications also extend to treating bronchitis and skin wounds, reflecting its ethnobotanical significance. Several species within the *Indigofera* genus have been documented for their pharmacological effects, which includes anti-inflammatory, anti-cancer, and neuroprotective properties^{17,18,19}. There is a noticeable lack of comprehensive phytochemical and pharmacological studies specific to *Indigofera stenophylla*¹⁸. This gap presents a valuable opportunity for this research aimed at uncovering the therapeutic potential and bioactive compounds within this species, which could lead to the development of novel molecules. This study aimed to isolate, and characterize some phytochemical(s) and to validate its folkloric claims of the plant in the management of convulsions.

MATERIALS AND METHODS

Experimental Animals

Adult Swiss albino mice of 25–30 g of both sexes were sorted and used for the experiments. The animals were housed under standard laboratory conditions with a 12-hour light/12-hours dark cycle and had free access to

food and water. All procedures adhered to the ethical guidelines to ensure humane treatment in accordance with committee on Animal care and use Ahmadu Bello University, Zaria with an approval number of ABUCAUC/2023/162, which is in conformity with United States National Institute of Health Guideline for care and use of laboratory animal in biomedical research²⁰.

Chemicals, Reagents, Apparatus, and Equipment

The apparatus used included a TLC tank, a 3.5×75 cm column, a burette, beakers, pipettes, and a pre-coated aluminum thin-layer chromatographic plate (60 GF257, 20×20 cm, Sigma Aldrich, Germany). Freshly prepared reagents comprised Salkowski's reagent, Lieberman-Burchard reagent, and ferric chloride. The solvents were methanol, acetone, ethyl acetate, dichloromethane, n-hexane, and acetic acid (Qualikems). PTZ and SCN were sourced from Sigma-Aldrich, while silica gel with a mesh size of 60-120 µm was obtained from Qualikems. All chemicals and reagents were of analytical grade. Essential equipment included an analytical balance, an oven, a rotary evaporator, and an Electro-thermal melting point apparatus (Department of Pharmaceutical and Medicinal Chemistry, Ahmadu Bello University, Zaria). Additional instruments featured the ABB-MB 3000 UV spectrophotometer, Bruker AVANCE III (400 MHz) NMR spectrometer (Multi-user Laboratory, Department of Chemistry, Faculty of Physical Sciences, Ahmadu Bello University, Zaria), and an IR spectrophotometer from the National Research Institute for Chemical Technology (NARICT), Zaria.

Collection, identification, and preparation of *Indigofera stenophylla* plant material

Indigofera stenophylla was collected in September, 2023, identified, and authenticated by Namadi Sanusi at the Habermium Unit, Department of Botany, Faculty of Life Sciences, Ahmadu Bello University, Zaria by comparing with herbarium reference voucher specimen (No. 0763). The aerial parts were sorted, air-dried, and ground into a fine powder, labelled and stored at room temperature²¹.

Extraction procedures

The powdered aerial parts of *Indigofera stenophylla* (1500 g) was extracted with 70% v/v methanol (10 L) using maceration method for 3 days. The extract was concentrated by removing the solvent *in-vacuo* using rotary evaporator at 45°C to afford a crude extract subsequently referred to as methanol aerial parts extract of *Indigofera stenophylla* (MAEIS). The stored MAEIS at -20°C 150 g was fractionated successively with 5 L of n-hexane, and 2.5 L of n-butanol to obtain n-hexane fraction (HF), and n-butanol fraction (BF) respectively²¹.

Isolation, characterization, and purification of compounds AR2

The column (3.5x75 cm) was packed with silica gel (60-120 µm) by wet slurry method. The HF weighing 12.7 g was subjected to extensive column chromatography by adsorbing the sample on silica gel. The elution was carried out using gradient elution starting with n-hexane (100 %), then n-hexane: ethyl acetate (95:5) and the polarity was gradually increased by 10unit, until ethyl acetate (100 %). The characterization of the isolated compounds was determined using solubility test, chemical test, melting point, thin layer chromatography (TLC), ultraviolet (UV), infrared (IR), and proton and carbon nuclear magnetic resonance (¹H-NMR and ¹³C-NMR) spectroscopic experiments alongside with reported literature data²².

Pentylenetetrazole-Induced Seizures

A total of 30 mice were randomly divided into five groups, each containing six mice. The groups were as follows: Group 1 (control) received saline per oral, Group 2, 3 and 4 received MAEIS at the doses of 150, 300 and 600 mg/kg per oral, and Group 5 received standard anticonvulsant, diazepam at 5 mg/kg per oral,. After a 30-minute period following extract administration, all groups received an *intraperitoneal* injection of PTZ at a dose of 60 mg/kg body weight. Seizure onset, duration, and severity were recorded using a standardized scale^{1,23,24}.

Strychnine-Induced Seizures

A total of 30 mice were randomly divided into five groups, each containing six mice. The groups were as follows: Group 1 (control) received saline per oral, Group 2, 3 and 4 received MAEIS at the doses of 150, 300 and 600 mg/kg per oral, and Group 5 received standard anticonvulsant, phenobarbital at 30 mg/kg per oral. After a 30-minute period following extract administration, all groups received an *intraperitoneal* injection of SCN at a dose of 1 mg/kg body weight. Seizure onset, duration, and severity were recorded using a standardized scale^{25,24}.

Statistical analysis

The data is expressed as mean ± Standard Error of Mean (SEM) analyzed using one-way analysis of variance (ANOVA) followed by Dunnett's post-hoc test, with significance set at $p < 0.05$ and $p < 0.01$.

RESULTS AND DISCUSSION

Table 1: Thin layer chromatographic Profiles of Isolated Compound AR2.

Solvent System	Number of spots	Colour of spot after heating	R _f Value
N-hexane: ethyl acetate (8: 2)	1	Yellow	0.79
N-hexane: ethyl acetate (7: 3)	1	Yellow	0.63

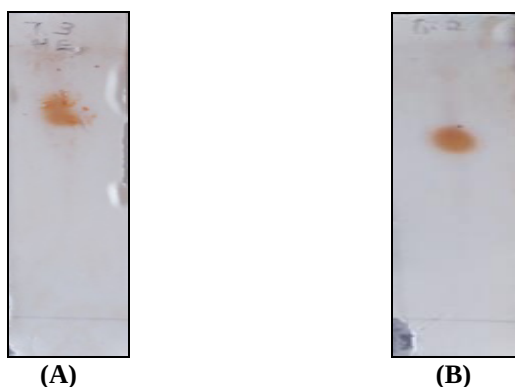


Plate I: TLC Profiles of Isolated Compound AR2 .

(A) N-hexane: Ethyl acetate (8:2).

(B) N-hexane: Ethyl acetate (7:3).

Table 2: Physico-Chemical Properties of Compound AR2.

Physico-chemical properties	Compound AR2
Physical appearance	White
Melting point	163 - 166°C
In Ethyl acetate	Insoluble
In Chloroform	Completely soluble
(a). Liebermann-Burchard test	Green
(b). Salkowski test	Reddish brown ring at the interphase
(c). Ferric chloride test	Green

UV Spectrum of compound AR2

The UV spectrum of compound AR2 displays an absorption maximum at 254 nm. as seen in **Fig. 1**.

IR Spectrum of compound AR2

In the IR spectrum, the following signals are observed: 3354.0 cm⁻¹, 2959.7 cm⁻¹, 2855.0 cm⁻¹, 1736.9 cm⁻¹, 1453.0 cm⁻¹, and 1379.0 cm⁻¹. as observed in **Fig. 2**.

¹H-NMR spectrum of compound AR2

The proton NMR spectrum reveals the following signals: δ 5.12 (dd, J = 15.0, 8.5 Hz,

1H), δ 5.04 (dd, J = 15.0, 7.0 Hz, 1H), δ 3.62 (tt, J = 11.0, 4.5 Hz, 1H), δ 0.69 (s, 1H), δ 0.81 (s, 1H), δ 0.88 (s, 1H), δ 0.94 (s, 1H), δ 1.00 (s, 1H), and δ 1.02 (s, 1H) as shown in **Fig. 3**.

¹³C-NMR Spectrum of compound AR2

The ¹³C-NMR spectrum displays the following signals: δ 37.26, δ 31.67, δ 71.81, δ 42.31, δ 140.76, δ 121.71, δ 31.90, δ 31.90, δ 50.16, δ 36.51, δ 21.21, δ 39.68, δ 42.22, δ 56.87, δ 24.36, δ 28.92, δ 55.96, δ 12.05, δ 21.08, δ 40.49, δ 23.07, δ 138.31, δ 129.28, δ 51.24, δ 29.15, δ 18.98, δ 19.62, δ 25.23, and δ 12.27. as observed in **Fig. 4**.

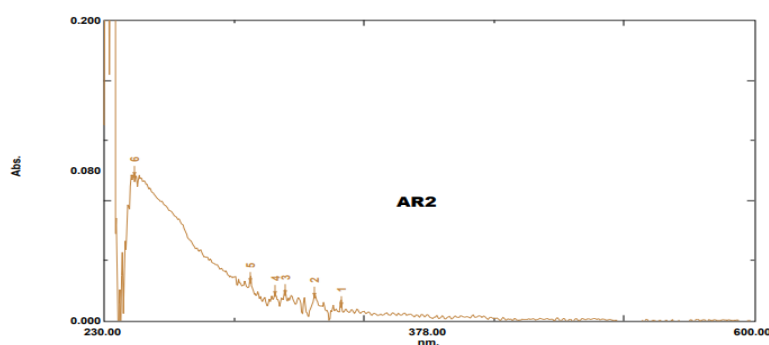


Fig. 1: UV Spectrum of Compound AR2.

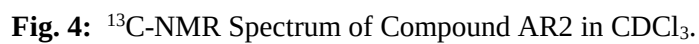
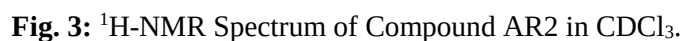
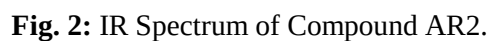


Table 3: ^1H and ^{13}C -NMR Spectral Data of Compound AR2 Compared with Literature.

Position	^{13}C -NMR Compound	^1H -NMR AR2	^{13}C -NMR	^1H -NMR ²²
1	37.26	1.86, 1.07		37.3 1.85
2	31.67	1.46, 1.85		31.6 1.46
3	71.81	3.62, 1H, tdd, J= 4.5, 4.0 and 3.9Hz	71.8	3.52 m
4	42.31	2.28, m, 2.25,	42.3	2.27
5	140.76	-----		140.8 -----
6	121.71	5.36, 1H, d, J= 5.0Hz	121.7	5.35 br s
7	31.90	1.97, 1.48		31.9 1.96
8	31.85	1.49		31.9 1.48
9	50.16	0.94		51.2 0.93
10	36.51	-----		36.5 -----
11	21.21	1.51, 1.47		21.1 1.49
12	39.68	1.17, 1.98		39.7 1.16
13	42.22	-----		42.3 -----
14	56.87	1.06		56.9 1.05
15	24.36	1.57, 1.16		24.4 1.56
16	28.92	1.81, 1.26		28.4 1.70
17	55.96	1.13		56.1 1.13
18	12.04	0.69, 3H, s		11.0 0.69 s
19	21.07	1.00, 3H, s		21.2 1.01 s
20	40.49	2.02, s		40.5 2.02
21	23.07	1.02, 3H, d, J= 7.0Hz		21.2 1.02(d, 7.5)
22	138.31	5.12, 1H, dd, J= 9.0 and 15.0Hz	138.3	5.10
23	129.28	5.04, 1H, dd, J= 9.0 and 15.0Hz,	129.3	5.03
24	51.24	1.53		51.2 1.53
25	29.15	1.65		31.9 1.65
26	18.98	0.77, 3H, d, J= 7.0Hz		21.2 0.79 (d, 6.5)
27	19.62	0.88, 3H, d, J= 7.0Hz		19.0 0.84 (d, 6.5)
28	25.23	1.16, 1.44		25.5 1.15
29	12.27	0.81, 3H, t, J= 7.0Hz		12.1 0.80 (t, 7.5)

Effect of Methanol Aerial Parts Extract of *Indigofera stenophylla* using Pentylentetrazole Induced Seizure in Mice

In the evaluation of the anticonvulsant effects of the methanol aerial parts extract of *Indigofera stenophylla* using the PTZ seizure model, the normal control group exhibited a mean seizure onset of 394.00 ± 0.88 seconds, mean seizure duration of 41.25 ± 1.02 and zero percentage protection. In contrast, the groups receiving the methanol extract at doses of 300 mg/kg and 600 mg/kg demonstrated significant ($p < 0.05$) prolongation in mean onset seizure, while reduction in the mean seizure duration was only observed at 600 mg/kg of the extract when compared to normal saline control group. Subsequently, the extract offered 50 and 75% protection at 300 and 600 mg/kg respectively. Diazepam (a positive control anti-seizure drug) significantly ($p < 0.05$) elongated the mean onset of seizure and significantly ($p < 0.05$) reduced the mean seizure duration compared to the normal saline control group. In addition,

diazepam also offered 100% protection in seizure model induced by PTZ in mice **Table 4**.

Effect of Methanol Aerial Parts Extract of *Indigofera stenophylla* Using Strychnine Induced Seizure in Mice

In the SCN seizure model, the normal saline control group showed a mean seizure of 609.75 ± 1.06 seconds and a mean seizure duration of 62.00 ± 0.75 seconds with zero percentage protection. However, the extract at 300 and 600 mg/kg showed significant ($p < 0.05$) extension in the mean seizure onset, while no significant ($p > 0.05$) reduction in mean seizure duration was observed at all the tested doses of the extract compared to normal saline control group. The extract at the highest dose of 600 mg/kg offered 50% protection in this model. Phenobarbitone (a reference anti-seizure drug) significantly ($p < 0.05$) extended the mean seizure onset and significantly ($p < 0.05$) reduced mean seizure duration when compared to normal saline control group. Phenobarbitone offered 100% protection in seizure induced by strychnine in this study

Table 5.

Table 4: Effect of Methanol Aerial Parts Extract of *Indigofera stenophylla* using Pentylene-tetrazole Induced Seizure in Mice.

TREATMENTS (mg/kg)	NUMBER OF ANIMALS	MEAN ONSET SEIZURE (Second)	MEAN DURATION SEIZURE (Second)	% PROTECTION
N/S 10 mL/kg	6	394.00±0.88	41.25±1.02	0
MEIS (150)	6	415.00±0.78	45.75±0.90	0
MEIS (300)	6	483.75±0.74*	26.75±0.88	50
MEIS (600)	6	639.00±0.84*	9.50±0.82*	75
DZP (5)	6	1800.00±0.00*	0.00±0.00*	100

N/S; normal saline, MAEIS; methanol aerial parts extract of *Indigofera stenophylla*, DZP; diazepam. Each value represents Mean ± Standard Error of Mean (SEM), n=5, one-way ANOVA; Dunnett's post hoc test, *p<0.05 statistically significant compared to normal saline control.

Table 5: Effect of Methanol Aerial Parts Extract of *Indigofera stenophylla* Guil.and Perr. (Fabaceae) using Strychnine Induce Seizure in Mice.

TREATMENTS (mg/kg)	NUMBER OF ANIMALS	MEAN ONSET SEIZURE (Second)	MEAN DURATION SEIZURE (Second)	% PROTECTION
N/S 10 mL/kg	6	609.75±1.06	62.00±0.75	0
MEIS (150)	6	624.50±0.98	71.50±1.02	0
MEIS (300)	6	714.25±0.88*	65.00±0.89	25
MEIS (600)	6	781.00±0.78*	52.75±0.85	50
PHENO (30)	6	1800.00±0.00*	0.00±0.00*	100

N/S; normal saline, MAEIS; methanol aerial parts extract of *Indigofera stenophylla*, PHENO; phenobarbitone. Each value represents Mean ± Standard Error of Mean (SEM), n=5, one-way ANOVA; Dunnett's post hoc test, *p<0.05 statistically significant compared to normal saline control.

Discussion

The TLC analysis of the isolated compound AR2 showed retention factor (R_f) value of 0.79 and 0.63 using N-hexane/Ethyl acetate (8:2) and N-hexane/Ethyl acetate (7:3) respectively with a single spot as shown in Plate I confirming the purity of the isolated compound AR2 in **Table 1**. The isolated compound AR2 appeared white amorphous substances and exhibited a melting point range of 163-166°C, insoluble in ethyl acetate but completely soluble in chloroform. The formation of a green colour to Liebermann-Burchard test, a reddish-brown color at the interface of the two layers for Salkowski as

observed in **Table 2** which confirmed the presence of triterpene skeleton and a green coloration to ferric chloride test indicates the presence of a hydroxyl group at the C-3 position of triterpene skeleton²². The UV spectrum of the isolated compound AR2 showed absorption maxima at 254 nm in **Fig. 1**, which are characteristic of the unsaturated triterpene skeleton²². The IR spectrum exhibited absorption bands at 3354.0 cm^{-1} (hydroxyl group), 2959.7 cm^{-1} and 2855.0 cm^{-1} (aliphatic C-H stretching), 1736.9 cm^{-1} (olefinic C=C), and 1453.0 cm^{-1} and 1379.0 cm^{-1} (gem-dimethyl groups) in **Fig. 2**. The IR signals are consistent with the structural

features of the triterpene nucleus as reported in the literature. The proton nuclear magnetic resonance (^1H NMR) spectrum showed signals at δ 5.12 (dd, $J = 15.0, 9.0$ Hz, 1H) and δ 5.04 (dd, $J = 15.0, 9.0$ Hz, 1H), characteristic of the olefinic protons at C-22 and C-23. The signal at δ 3.62 (tdd, $J = 4.0, 4.5, 3.9$ Hz, 1H) was assigned to the H-3 proton. The presence of six methyl signals at δ 0.69, 1.00, 1.02, 0.77, 0.88, and 0.81 where two methyl signals are singlets (angular methyl at C-18 and C-19), three correspond to doublets (secondary methyl at C-21 and isopropyl group at C-26 and C-27), and one triplet (primary methyl at C-29), confirmed the phytosterol skeleton of stigmasterol, as reported by²², as observed in the **Fig. 3** The ^{13}C -NMR spectrum displayed 29 carbon signals in **Fig. 4**, consistent with the molecular formula of stigmasterol. The signals at δ 138.31 (C-22) and δ 129.28 (C-23) were attributed to the olefinic carbons. The signal at δ 71.81 was assigned to the C-3 carbon bearing the hydroxyl group²², which confirmed the identification of the isolated compound AR2 as stigmasterol, a pentacyclic triterpene alcohol.

Stigmasterol as shown in **Fig. 5** is a phytosterol found in various plants, known for its multiple pharmacologic properties. It exhibits antivenin activity, anti-inflammatory effects, and has demonstrated antioxidant activity. Stigmasterol also possesses antimicrobial activity against various pathogens. Furthermore, it may support immune function and promote wound healing²². These validated properties suggest its potential therapeutic applications.

The effect of MAEIS on pentylenetetrazole (PTZ)-induced seizures in mice showed that the extract may possess antiseizure potential. PTZ is a well-established chemical agent used to induce seizures in experimental animals^{23,1,24}.

This is simply because PTZ is a **Gamma-Amino butyric Acid (GABA)** antagonist and thus, facilitate excitatory neurotransmission²⁶. In this study, the concomitant prolongation of onset of seizure as well as reduction in seizure duration by MAEIS suggests an antiseizure potential. The antiseizure potential of the extract was further corroborated by the varying degree of protection offered by the extract in this study. Therefore, the probable neural mechanism of antiseizure potential of the extract may be suggested to involve the enhancement of GABAergic pathways, since PTZ acts as a GABA antagonist to elicit seizure in mice²⁷. This finding agrees with earlier medicinal plants suggested to possess antiseizure potential in PTZ-induced convulsion model in mice^{27,24}. The active constituents of MAEIS, Stigmasterol have been identified as a positive modulator of GABA_A receptors, which play a crucial role in inhibitory neurotransmission and are implicated in epilepsy. Stigmasterol enhances GABA-induced currents at various GABA_A receptor subtypes, with a significantly higher potentiation observed at extrasynaptic δ subunit for efficacy. It has been reported that stigmasterol could be considered as a steroidal drug for treating neurological disorders due to its positive modulation of GABA receptors²⁸.

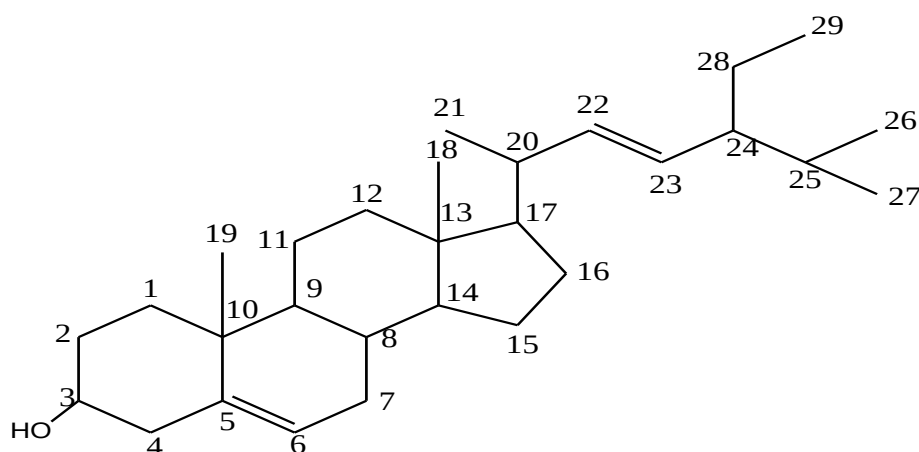


Fig. 5: Stigmasterol.

Strychnine, a known glycine receptor antagonist which induces seizures by disrupting inhibitory neurotransmission in the central nervous system particularly in the spinal cord, leading to hyperexcitability and convulsions²⁹. In this study, the prolongation of the mean seizure onset as well as the 50% protection offered by the extract at its highest tested dose may suggest antiseizure potential of the extract in mice. This finding is in tune with earlier medicinal plants suggested to possess antiseizure potential in SCN-induced seizure model in mice^{27,24}. It will not be out of place therefore, to suggest that the phytochemicals present in MAEIS may either enhance inhibitory neurotransmission or mitigate excitatory pathways, thereby offering neuroprotective effects. The modulation of neurotransmitters, particularly GABA and glycine, could be crucial mechanisms through which MAEIS exerts its anticonvulsant properties³⁰. The active constituents of *Indigofera stenophylla*, such as flavonoids and alkaloids, have been shown in previous studies to possess neuroprotective effects while improving the overall neurochemical environment in the brain^{17,18,19}. *Indigofera stenophylla* could help stabilize neuronal activity during strychnine-induced seizures. Stigmasterol exhibits anticonvulsant activity through the glycinergic pathway, increasing the inhibitory effect of glycine at glycine receptors³¹. Additionally, stigmasterol has demonstrated neuroprotective effects by restoring antioxidant enzyme activity and reducing oxidative damage in brain tissues, further supporting its potential as a therapeutic agent for epilepsy management³².

Conclusion

The chromatographic separation of HF of the MAEIS afford the isolation of stigmasterol as phytosterol and validation of its anticonvulsant potential which provides the scientific basis for the use of this plant in the management of convulsions.

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نشرة العلوم الصيدلانية جامعة أسيوط



عزل الستغماستيرون والنشاط المضاد للتشنجات لمستخلص الميثانول من الأجزاء الهوائية لنبات *Indigofera stenophylla*

(الفصيلة البقولية - Fabaceae)

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المقدمة

يُعدّ نبات *Indigofera stenophylla* ، وهو من الفصيلة البقولية (Fabaceae) ، من النباتات المستخدمة تقليدياً في الطب الشعبي لعلاج أمراض متعددة، منها الصرع. هدفت هذه الدراسة إلى عزل وتوصيف المركبات الكيميائية النباتية من مستخلص الميثانول للأجزاء الهوائية لنبات *Indigofera stenophylla* (MAEIS)، وتقييم قدرته المحتملة كمضاد للتشنجات.

الطريقة

أُتيح من خلال استخلاص MAEIS عزل المركب AR2 باستخدام تقنيات الكروماتوغرافيا. وقد جرى تقييم التأثير المضاد للتشنجات لمستخلص MAEIS باستخدام نماذج النوبات المستحثة بالـ بيتينيلين تترازول (PTZ) والستريكنين (SCN).

النتائج

أظهر التحليل الطيفي للمركب AR2 أنه β^3 ، ٢٢-ستغماستا-٥، ٢٢-دين-٣-أول (stigmasterol). كما أظهر المستخلص MAEIS نشاطاً ملحوظاً مضاداً للتشنجات عند الجرعات المتدرجة ١٥٠، ٣٠٠،

و ٦٠٠ ملغم/كغم، حيث أدى إلى خفض كل من زمن بدء النوبة ومدة استمرارها في نماذج PTZ و SCN مع دلالة إحصائية $p < 0.05$ و $p < 0.001$ وكانت جرعة ٦٠٠ ملغم/كغم هي الأكثر فعالية في الحماية.

الخلاصة

أدى التحليل الكروماتوغرافي العمودي والتحليل الطيفي لمستخلص MAEIS إلى عزل مركب الستغماستيرون، وهو مركب حيوي فعال ذو نشاط مضاد للتشنجات. ويُعد هذا أول عزل لمركب الستغماستيرون من هذا النبات.