



Central nervous system depressant activity of fractions of *Globimetula braunii* Engl. (Loranthaceae) growing on *Terminalia catappa* L. (Combretaceae) and isolation of lupeol

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Abstract

Globimetula braunii is a parasitic plant belonging to the family Loranthaceae. Traditionally, the plant has been reported to be used in the treatment of insomnia. The study was carried out to investigate the central nervous system (CNS) depressant activity of the fractions of *Globimetula braunii* growing on *Terminalia catappa* and to isolate the phytochemical compound(s) present in the most active fraction. The CNS depressant activity of all the fractions was investigated using diazepam induced sleep. The most active fraction was further subjected to the hole board test and beam walk assay. The chromatographic technique was used for the isolation of phytochemical compound. Hexane fraction significantly ($p < 0.05$) reduced latency to sleep and prolonged the sleeping time. Both chloroform and ethyl acetate fractions at highest and median doses showed significant increase in the duration of sleep compared to normal saline. The n-butanol fraction at all doses tested do not have any effect on time of onset and duration of sleep when compared with normal saline treated group. Hexane fraction significantly ($p < 0.05$) decreased the number of head dip in a dose dependent manner and delayed the time to reach the goal box compared to normal saline treated group. Lupeol was isolated from n-hexane fraction.

Keywords: *Globimetula braunii*; Lupeol; Central nervous system; Depressant activity

INTRODUCTION

The practice of traditional medicine is as old as man himself. In Nigeria, traditional medicine is well established in socio cultural system of the people and it varies from one community to another. Moreover, traditional medical practitioners played a vital role and remain the major source of new finding in natural product. The use of herbal medicinal

plants is one of the oldest known forms of therapy [1-3].

In developing countries, people living in rural areas used herbal medicinal plants for their primary health care needs. Similarly, in Nigeria the major consumers of medicinal plants have been until recently, the local population [1]. These may be due to its easy accessibility, availability, low cost and few

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side effects. The part of plant that could be used in herbal medicine is either the barks, roots, leaves or whole plant.

Globimetula braunii (Loranthaceae) known as Mistletoe (English) and Kauchi (Hausa) is a parasitic plant growing on many plants. The plant is widely distributed in tropical countries including Nigeria. It is used in the treatment of insomnia, nervousness and pain [4].

Our earlier study on *Globimetula braunii* revealed that the crude ethanol leaves extract possesses central nervous system depressant activity [5]. The present study therefore aimed to assess the central nervous system depressant activity of the fractions of extract of *Globimetula braunii* leaves using Diazepam-induced sleep, Hole-board test and Mouse beam-walk assay and to isolate the possible phytochemical constituent(s) present in the most active fraction.

EXPERIMENTAL METHODS

Collection and identification of plant material. The plant *Globimetula braunii* growing on *Terminalia catappa* was collected in December 2017, from its natural habitat around Aminu Kano Teaching Hospital, Zaria road, Tarauni Local Government Area, Kano State. The plant was identified and authenticated by Mallam Namadi Sunusi a plant taxonomist at the Herbarium Unit of the Department of Botany Sciences, Faculty of Life Sciences, Ahmadu Bello University, Zaria by comparing with voucher specimen number (2839) already deposited in the Herbarium Unit.

Extraction and fractionation. The leaves of *Globimetula braunii* were washed with water, dried and pulverized. The pulverized sample (2500 g) was extracted by maceration with 20 L 90% v/v ethanol. The filtrate collected was concentrated in rotatory evaporator at 40°C under reduced pressure. The ethanol crude extract was dissolved in water; the aqueous solution was successively partitioned with n-

hexane, chloroform, ethyl acetate and n-butanol. The fractions (n-hexane, chloroform, ethyl acetate and n-butanol) were concentrated and dried.

Preliminary phytochemical screening. The qualitative evaluation for the presence of phytochemical groups in the fractions of *Globimetula braunii* leaves extract was carried out according to established procedures described by Evans 2002 [6].

Isolation and purification. The n-hexane which was found to have better CNS depressant activity was subjected to column chromatography. The n-hexane fraction (4 g), was chromatographed on silica gel column (2.9 cm by 70 cm) packed with 160 g of silica gel of 60-120 mesh size (Loba Chemie) to 45 cm length of the column. Gradient elution was carried out starting with n-hexane 100% followed by n-hexane: ethyl acetate 95:5 and continue with constant increase of 5% ethyl acetate up to 60:40 n-hexane: ethyl acetate giving a total of 228 fractions which were pooled together based on their TLC profiles and coded A-I. The fraction C was re-purified by crystallization and afforded a white crystalline compound weighing 7 mg coded GBH₁. The TLC profile of GBH₁ using Hexane:Ethyl acetate (8:2) revealed a single spot with R_f value 0.63.

Spectroscopic characterization of GBH₁. The isolated compound (GBH₁) was characterized using chemical and spectroscopic techniques (UV, IR and NMR). The UV spectrum was measured on Jenway 7315 Spectrophotometer. The IR spectrum was measured on a Cary630 agilent technologies fourier transform infrared spectrophotometer. Nuclear magnetic resonance (NMR)-spectra were carried out at Institute of Pharmacy and Biomedical Sciences, University of Strathclyde, Glasgow, UK, recorded on a Bruker Avance spectrometer at a frequency of 600 MHz for ¹H-and 100 MHz for ¹³C-NMR.

Animals. Swiss albino mice of either sex (weighing between 18-30g) were used for the determination of LD₅₀ as well as central nervous system depressant activity evaluation. The mice were obtained from the Animal House of the Department of Pharmacology and Therapeutics, Faculty of Pharmaceutical Sciences, Ahmadu Bello University, Zaria. The ethical approval (ABUCAUC/2021/011) for the experimental procedure was sought from the Ahmadu Bello University Animal Ethics Committee and the mice were maintained in a well-ventilated room in cages with stainless steel wire mesh covers. The experiment was conducted according to guidelines of Ahmadu Bello University Animal Ethics Committee.

Acute toxicity study. The intraperitoneal median lethal doses (LD₅₀) of partitioned fractions of *Globimetula braunii* were determined in mice according to the method described by Lorke, 1983 [7]. The study was carried out in two phases; in the first phase, the mice were grouped in to three groups of three mice, each group received partitioned fractions of *Globimetula braunii* at doses of 10, 100 and 1000 mg/kg body weight intraperitoneally and then observed for signs of toxicity including death within 24 hrs. In the second phase, three mice were treated with the partitioned fractions at doses of 1600, 2900 and 5000 mg/kg body weight. The mice were observed for signs of toxicity and death within 24 hours. The LD₅₀ value was calculated as the geometric mean of the lowest lethal dose and the highest non-lethal dose.

Diazepam-induced sleep in mice. Sleep potentiating effect of n-hexane, chloroform, ethyl acetate and n-butanol fractions of *Globimetula braunii* at different doses were determined according to the method described by Rakotonirina *et al.*, 2001 [8]. The mice were grouped in to four groups of six animals each; groups 1, 2, and 3 received n-hexane fraction at doses of 250, 500 and 1000 mg/kg body weight intraperitoneally whereas, chloroform,

ethyl acetate and n-butanol were administered at doses of 150, 300 and 600 mg/kg body weight intraperitoneally while mice in the fourth group received normal saline 10 ml/kg to serve as negative control. Thirty minutes later, diazepam at a dose of 20 mg/kg was administered intraperitoneally to mice in all the groups to induce sleep. The loss of righting reflex was considered as time of onset of sleep [9], and sleeping time was measured as the time between disappearance and recovery of righting reflex [10]. Based on the results obtained from this test the most active fraction was subjected to the hole board test and beam walk assay.

Hole board test. The influence of the n-hexane fraction of ethanol leaves extract of *Globimetula braunii* on the exploratory behaviour of mice was determined according to the method described by Files and Wardill 1975 [11]. Mice were randomly divided into five groups of six mice each. Mice in group 1, 2 and 3 received the n-hexane fraction at doses of 250, 500 and 1000 mg/kg body weight intraperitoneally respectively. The animal in the fourth group received diazepam 0.25 mg/kg intraperitoneally while group 5 received normal saline 10 ml/kg intraperitoneally. Thirty minutes post treatment each mouse was placed at one corner of the board and then allowed to move about and dipped its head into the holes to indicate exploratory behaviour. The number of head dips in 5 minutes was recorded [12].

Beam walk assay. Mice were trained to walk from a start platform along a ruler (80 cm long, 3 cm wide) elevated 30 cm above the bench by a wooden support to a goal box. Each mouse was tried three times, and the test was designed in such a way that the mice tested would be aware that there was a goal box that could be reached. A ruler was used for the training so that the mouse will find it easier to cross; moreover, it will induce minimum anxiety. The mice that successfully walked along the ruler were grouped into five groups of six mice

each. The first group received normal saline at the dose of 10 ml/kg i.p. Hexane fraction at doses of 250, 500 and 1000 mg/kg was administered intraperitoneally to the second, third and fourth groups respectively, while the fifth group received diazepam (0.25 mg/kg, i.p.). Thirty minutes post-treatment, each mouse was placed on the beam at one end and allowed to walk to the goal box. Mice that fell were returned to the position they fell from, with a maximum time of 60 seconds allowed on the beam. The time to complete the task; the number of foot slips (one or both fore and hind limbs slipped from the beam) and the number of falls were recorded [13].

Statistical analysis. Data was expressed as mean \pm standard error of mean (Mean \pm SEM.). The means difference was analyzed by one-way ANOVA followed by Dunnett's post hoc test. The values were considered to be significantly different at $p < 0.05$. SPSS version 20 was used for statistical analysis.

RESULTS AND DISCUSSION

The percentage yield of n-hexane, chloroform, ethyl acetate and n-butanol was calculated to be 28.14%, 5.00%, 6.67% and 5.67 respectively (Table 1).

Phytochemical constituents. The Preliminary phytochemical screening of fractions of *Globimetula braunii* showed that, all fractions contained glycoside, steroids and saponins; whereas alkaloid was only present in aqueous fraction. Flavonoids were found to be present in all fractions except n-hexane. Tannis were found to be present in only ethyl acetate and aqueous fraction. Triterpenes were found to be absent in only aqueous fraction (Table 2). As in our earlier study it was reported that ethanol leaf extract of *Globimetula braunii* contained saponins, flavonoids, tannins, steroids, triterpenes and glycosides [5]. Similarly in the present study ethyl acetate and n-butanol to contain all secondary metabolites that are present in the crude extract.

Table 1: Percentage yield of partitioned fractions

Solvent	Weight (g)	Yield (%)	Appearance
Hexane	42.21	28.14	Green
Chloroform	7.50	5.00	Bright green
Ethyl acetate	10.00	6.67	Light brownish
Butanol	8.50	5.67	Dark brownish
Aqueous	15.23	10.15	Brownish
Total	83.44	55.63	

Weight of Ethanol leaf extract = 150 g

Table 2: Preliminary phytochemical constituents of partitioned fractions of *Globimetula braunii* growing on *Terminalia catappa*

Constituents	Test	Observation	Inference			
			HF	CF	EAF	BF
Alkaloids	Mayer's	Cream precipitate	-	-	-	-
	Dragendoff's	Rose red precipitate	-	-	-	-
	Wagner's	Whitish precipitate	-	-	-	-
Tannins	Lead sub-acetate	Cream precipitate	-	-	+	+
Saponins	Frothing	Froth persists for 10 min	-	+	+	+
Steroids	Lieberman-Bucchard's	Reddish ring at interface	+	+	+	+
Triterpenes	Salkowski's	Red color	+	+	+	+
Antbraquinones	Bontrager's	Bright pink colour	-	-	-	-
Flavonoids	Ferric chloride	Green precipitate	-	+	+	+
	Sodium hydroxide	Yellow colour	-	+	+	+
	Shinoda'	Red color	-	-	+	+

- = absent, + = present

HF = n-hexane fraction; CF = Chloroform fraction; EAF = Ethyl acetate fraction; BF = n-butanol fraction

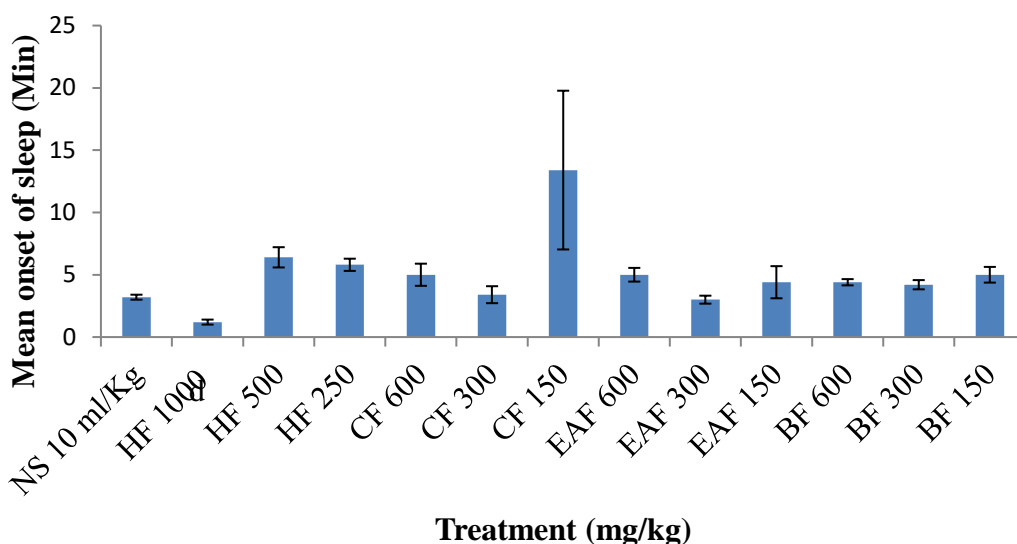
Table 3: Intraperitoneal Median Lethal dose Value of the fractions

Fraction	LD ₅₀ (mg/kg body weight)
n-hexane	3800
Chloroform	2100
Ethyl acetate	2100
n-butanol	2100

Table 4: Effect n-hexane fraction of *Globimetula braunii* in Hole board test

Treatment	Mean number of head dip \pm Std. Error
Diazepam 0.25 mg/kg	2.50 \pm 0.56 ^a
Hexane fraction 1000 mg/kg	1.00 \pm 0.26 ^a
Hexane fraction 500 mg/kg	2.50 \pm 0.72 ^a
Hexane fraction 250 mg/kg	4.17 \pm 1.86 ^a
Normal Saline 10 ml/kg	20.33 \pm 3.39

Data are expressed as mean \pm SEM (n = 6); means were compared by Bonferroni test ($p < 0.05$). a= $p < 0.05$

**Figure 1:** Effect of partitioned fractions of *Globimetula braunii* on diazepam-induced sleep in mice.

Values are presented as mean \pm SEM; the data were analysed using one-way ANOVA followed by Dunnett's post hoc test, n = 6, ^d $P < 0.05$ significant difference from normal saline group. Key: NS = Normal saline, HF = n-hexane fraction, CF = Chloroform fraction, EAF = Ethyl acetate fraction, BF = n-butanol fraction.

As in our earlier study it was reported that ethanol leaf extract of *Globimetula braunii* contained saponins, flavonoids, tannins, steroids, triterpenes and glycosides [5]. Similarly in the present study ethyl acetate and n-butanol to contain all secondary metabolites that are present in the crude extract.

Median lethal dose. The intraperitoneal median lethal dose of n-hexane fraction was estimated to be 3800 mg/kg, whereas that of chloroform, ethyl acetate and n-butanol

fraction was estimated to be 2100 mg/kg body weight in mice. This showed that, chloroform, ethyl acetate, and n-butanol fractions have similar median lethal dose. However, n-hexane has the highest median lethal dose compare to all other fractions (Table 3). From the result obtained n-hexane fraction is relatively safer than the chloroform, ethyl acetate and n-butanol. Moreover, n-hexane fraction has a higher median lethal dose compared to ethanol extract of *Globimetula braunii* (2800 mg/kg) as reported in previous study [5].

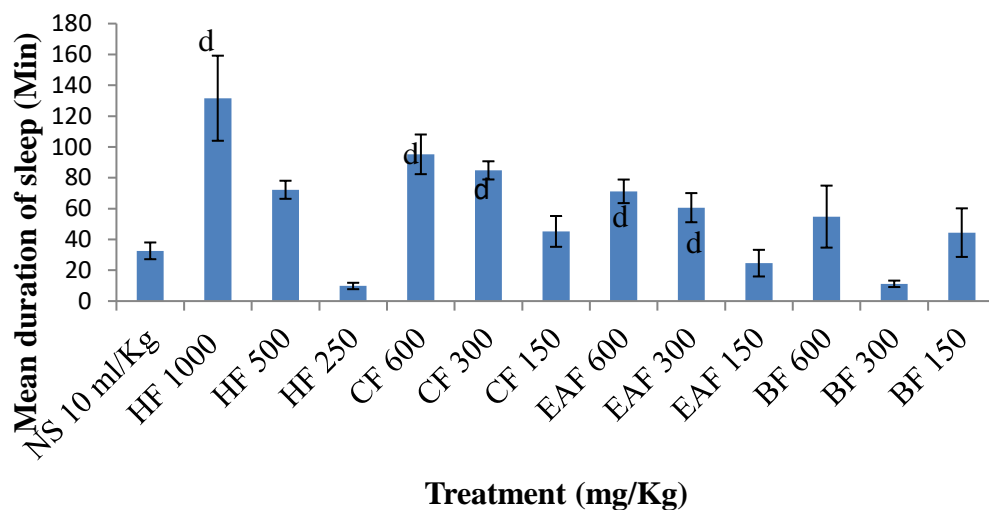


Figure 2: Effect of partitioned fractions of *Globimetula braunii* on diazepam-induced sleep in mice. Values are presented as mean \pm SEM; the data were analysed using one-way ANOVA followed by Dunnett's post hoc test, $n = 6$, ^d $P < 0.05$ significant difference from normal saline group. Key: NS = Normal saline, HF = n-hexane fraction, CF = Chloroform fraction, EAF = Ethyl acetate fraction, BF = n-butanol fraction.

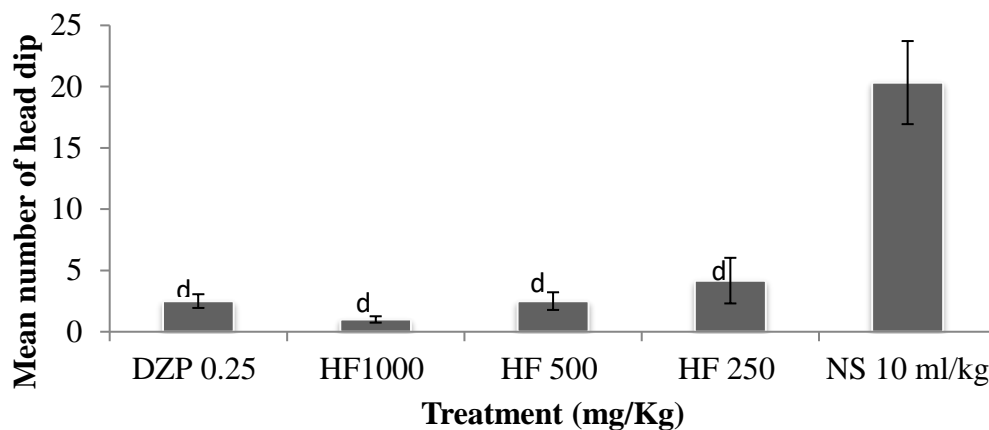


Figure 3: Effect of n-hexane fraction of *Globimetula braunii* on exploratory behavior of mice in hole board test. Values are presented as mean \pm SEM; the data were analysed using one-way ANOVA followed by Dunnett's post hoc test, $n = 6$, ^d $P < 0.05$ significant difference from normal saline group. Key: DZP = Diazepam, HF = n-hexane fraction, NS = Normal saline

Effect of partitioned fractions of *Globimetula braunii* on diazepam-induced sleep in mice. The n-hexane fraction at highest dose (1000 mg/kg) showed significant decrease ($p < 0.05$) in the time of onset of sleep and prolonged the duration of sleep ($p < 0.05$). However, there is no significant difference between the normal saline (10 ml/kg) and the two lower doses of the n-hexane fraction (500

and 250 mg/kg) (Figure 1). The increase in the duration of sleep exhibited by n-hexane fraction was found to be dose dependent. Moreover, both highest and median dose of chloroform fraction; 600 and 300 mg/kg (95.20 ± 12.86 and 84.80 ± 5.90 respectively) showed significant ($p < 0.05$) increase in the duration of sleep compared to normal saline (32.60 ± 5.42) (Figure 1).

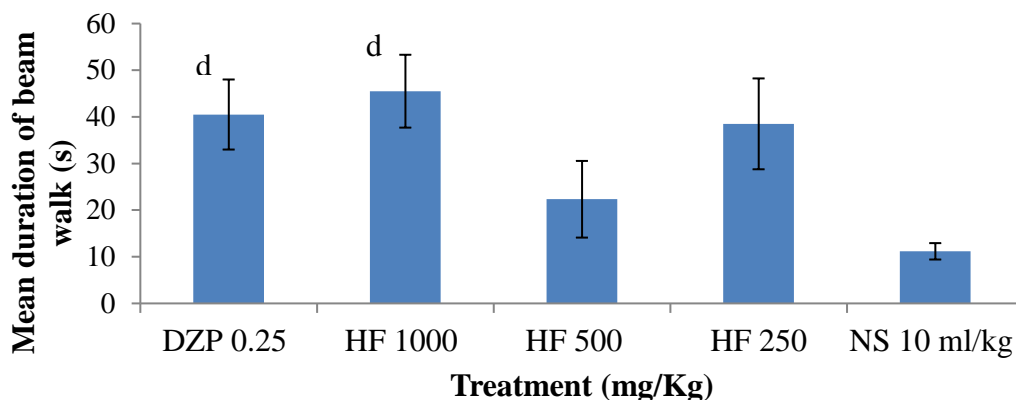


Figure 4: Effect of n-hexane fraction of *Globimetula braunii* on motor coordination in mice. Values are presented as mean \pm SEM; the data were analysed using one-way ANOVA followed by Dunnett's post hoc test, $n = 6$, ^d $P < 0.05$ significant difference from normal saline group. Key: DZP = Diazepam, HF = n-hexane fraction, NS = Normal saline

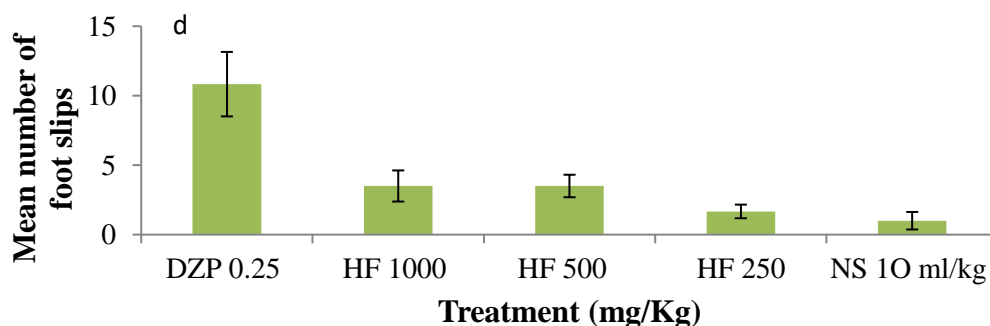


Figure 5: Effect of n-hexane fraction of *Globimetula braunii* on motor coordination in mice. Values are presented as mean \pm SEM; the data were analysed using one-way ANOVA, $n = 6$. Key: DZP = Diazepam, HF = n-hexane fraction, NS = Normal saline

However, there is no significant difference ($p > 0.05$) between normal saline and all doses of chloroform fraction in the time of onset of sleep compared to control (Figure 1).

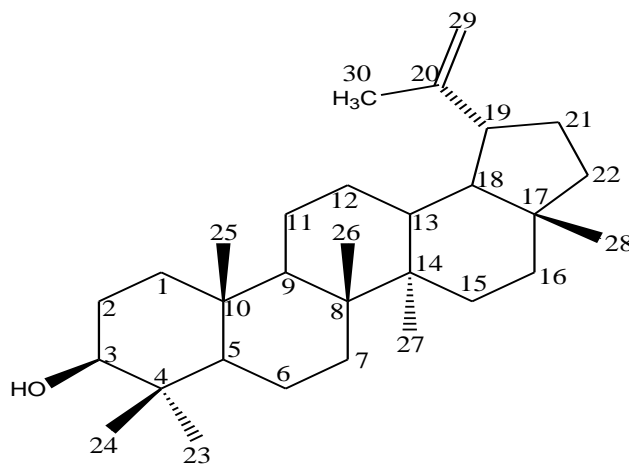
The ethyl acetate fraction produced a significant ($p < 0.05$) increase in the duration of sleep at the highest and median dose (600 and 300 mg/kg respectively) as in chloroform fraction. However, there is no significant increase in the sleep duration demonstrated between normal saline and the chloroform fraction at lowest dose (150 mg/kg). Additionally, the decrease in the time of onset of sleep exhibited by the ethyl acetate fraction is not dose dependent. The n-butanol fraction at all doses tested did not have any effect on time of onset as well as duration of sleep when

compared with normal saline treated group (Figures 1 and 2).

The ability of the fraction to increase the duration of sleep suggests a possible interaction with GABA (the major inhibitory neurotransmitter in the CNS involved in sedation). Thus potentiating diazepam-induced sleep, n-hexane, chloroform and ethyl acetate seem to possess sedative/sleep inducing properties. The n-hexane fraction has better sedative property compared to other fractions because it decreased the time of onset of sleep and prolonged the duration of sleep. This showed that the n-hexane fraction can be used in the management of insomnia. The sedative property exhibited by n-hexane fraction may be due to phytochemical compounds (steroids and triterpenes) present in the fraction [14].

Table 5: Summary of 1D and 2D Spectral Data for Compound GBH₁

POSITION	$\delta^{13}\text{C}$	$\delta^1\text{H}$, J (Hz)	APT	COSY	HMBC	Abdullahi <i>et al.</i> , 2013 [16] $\delta^{13}\text{C}$
1	39.00		CH ₂			38.7
2	27.42	1.59 (2H, s)	CH ₂	3.12	C-23	27.4
3	79.04	3.12 (1H, dd, 12)	CH	1.59	C-23, C-24	79.0
4	39.00		C			38.9
5	55.32	0.61 (1H, d, 6)	CH		C-3, C-23, C-24	55.5
6	18.34		CH ₂			18.5
7	34.30		CH ₂			34.2
8	41.00		C			40.9
9	50.46		CH			50.5
10	37.19		C			37.2
11	20.95		CH ₂			21.0
12	25.17		CH ₂			25.2
13	38.08		CH			38.1
14	42.50		C			42.9
15	27.42		CH ₂			27.1
16	35.60		CH ₂			35.5
17	43.01		C			43.0
18	48.33		CH			48.3
19	48.00	2.31 (1H, m, 6)	CH			48.0
20	150.97		C			151.0
21	29.87	1.40 (2H, s)	CH ₂			29.9
22	40.02		CH ₂			40.0
23	28.00	0.90 (3H, s)	CH ₃		C-2, C-3, C-5, C-24	28.0
24	15.38	0.69 (3H, s)	CH ₃		C-3, C-5, C-23	15.5
25	16.13	0.72 (3H, s)	CH ₃			16.1
26	16.00	0.93 (3H, s)	CH ₃			16.0
27	14.50	0.88 (3H, s)	CH ₃			14.8
28	18.02	0.72 (3H, s)	CH ₃			18.0
29	109.33	4.5 (s, 1H)	CH ₂	1.6	C-18, C-19, C-30	109.0
		4.62 (s, 1H)	CH ₂	1.6	C-18, C-19, C-30	
30	19.30	1.61 (s, 3H)	CH ₃	4.50 & 4.62	C-18, C-19, C-20, C-29	19.5

**Figure 6:** Proposed structure of GBH₁: LUPEOL

Effect of n-hexane fraction of *Globimetula braunii* on exploratory behavior of mice in hole board test. Hexane fraction at all doses tested produced a significant and dose dependent decrease in number of head dips when compared with control group. Diazepam (0.25 mg/kg) also produced a significant ($p < 0.05$) decrease in number of head dips when compared with normal saline treated group (Figure 3).

The hole board test is a measure used to evaluate the exploratory behaviour in mice after administration of a test substance or agent. A decrease in exploratory behavior in mice as demonstrated by a reduction in the number of head dips is a measure of CNS depressant activity. The n-hexane significantly decreased the number of head dips at all tested doses and this suggests that the n-hexane fraction has neuro sedative activity. Presence of steroids and triterpenes may be responsible for CNS depressant activity of n-hexane fraction as reported by Datta *et al.*, 2004 [15].

Effect of n-hexane fraction on motor coordination in mice. Hexane fraction at highest dose (1000 mg/Kg) significantly ($p < 0.05$) delayed the time to reach the goal box when compared to normal saline treated group (Figure 4). However there is significant difference between n-hexane fraction treated group and normal saline treated group on the number of foot slips (Figure 5). This confirms the result of hole board test. Beam walk assay is a sensitive tool for determining benzodiazepine-induced motor coordination deficits. It also used to predict the clinically sedative doses of new benzodiazepine-like drugs [13]. In this study, the n-hexane fraction did not produce significant increase in the number of foot slips (Figure 5). This suggests that at the doses tested, the fraction may produce its pharmacological effect without marked motor deficits.

Physical/Chemical test. Chromatographic separation of n-hexane fraction led to the isolation of GBH₁ compound. GBH₁ was found

to be white crystalline powder partially soluble in hexane, completely soluble in chloroform and insoluble in methanol. It was found to melt between 218- 221°C

The GBH₁ showed a positive result with Liebermann-Burchard test indicating that the compound is either steroid or triterpenoid.

Spectroscopic analysis. UV λ_{\max} was observed at 240 and 295 nm respectively. The IR spectrum of GBH₁ showed weak broad absorption at 3316 cm^{-1} which is O-H vibration, strong absorption at 2919 cm^{-1} and 2851 cm^{-1} is due to the asymmetric and symmetric C-H group respectively. The absorption at 1477 cm^{-1} is due to C-H bend. The ¹H NMR (CDCl₃, 600 MHz) spectrum of GBH₁ showed overlapping signals between δ 0.61 to 2.31 ppm due to CH₃ and CH₂ and, this indicated that GBH₁ contain a steroid or triterpene nucleus. Presence of sextet signal at δ 2.31 (1H, $J = 6$ Hz) indicated that, there is H-19 β which is a characteristic of lupeol [16]. The signal at δ 3.12 (1H, dd, $J = 12$ Hz) is due to a proton attached to hydroxylated C-3. Two broad singlets at δ 4.50 (1H) and δ 4.62 (1H) indicated the presence of an exomethylene group [17]. In addition to that, methyl signal at δ 1.61 ppm (3H, s) is for H-30. These suggested that GBH₁ is a lupane type triterpenoids [16].

The ¹³C-NMR spectrum showed signals at δ 109.33 and δ 150.97, confirming that GBH₁ has olefinic group typical of lupane type triterpenoids. The signal at δ 79.04 indicated the presence of a carbon attached to hydroxyl group assignable to C-3. The proton and carbon assignments were supported by the two-dimensional (2D) NMR data; proton-proton correlations were observed due to the presence of some cross peaks including the correlation between oxygenated methine proton signal at δ_{H} 3.12 (dd, H-3) and methylene signal δ_{H} 1.61 (s, H-2), also between of methine proton signal at δ_{H} 2.30 (m H-19) and methylene proton signal at δ_{H} 1.40 (s, H-21). Another correlation was observed

between methyl proton signal at δ_H 1.61 (s, H-30) and two olefinic protons; H-29a (δ_H 4.5, s, 1H) and 29b (4.62, s, 1H). The long range correlation was observed between oxymethine proton signal at δ_H 3.2 (dd, H-3) and methyl carbon signals at δ_C 28.0 (C-23). The olefinic protons at δ_H 4.50 and 4.62 showed cross peak correlations with a methine carbon signal at δ_C 48.00 (C-19) and methyl carbon signal at δ_C 19.3 (C-30). The methine proton signal at δ_H 2.31 (H- β 19) showed cross peaks with two methylene carbon signals δ_C 29.87 and δ_C 109.33 (C-21 and C-29). Moreover, methine proton signal at δ_H 2.30 (H- β 19) showed cross peak correlations with a methine carbon signal at δ_C 48.33 (C-18), a methyl carbon signal at δ_C 19.30 (C-30) and a quaternary carbon signal at δ_C 151.97 (C-20). The compound gave positive Liebermann-Burchard test indicating the compound is either a steroid or triterpenoid.

The NMR spectral data was compared with reported data from literature (Table 5). Comparison of spectra (UV, IR, 1H -NMR, ^{13}C -NMR, COSY, HSQC, HMBC) with the reported data of lupeol led to the conclusion that, GBH₁ is a pentacyclic tri-terpenoid; lupeol and the structure is presented below (Figure 6).

The isolation of lupeol from *Globimetula braunii* growing on *Terminalia catappa* is being reported for the first time.

Conclusion. In this study, lupeol, was isolated from ethanol leaf extract of *Globimetula braunii*. The isolation is being reported for the first time. The compound were fully characterized using chemical and spectroscopic techniques. The CNS depressant activity of the partitioned fractions of ethanol leaf extract of *Globimetula braunii* was investigated and n-hexane was found to possess significant central nervous system depressant activity. The finding of this study supports the ethno-medicinal claim that mistletoe possessed marked sedative property. *Globimetula braunii* growing on *Terminalia*

catappa should be standardized and used as herbal supplement for the management of insomnia and anxiety.

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