

Phytochemical Screening, Acute Toxicity and Analgesic Activity of Extracts From *Newbouldia laevis* in Laboratory Animals

Odo Peter¹, Odo Ekene¹, Ihemadu Chiaguguom¹, Felix Grace¹, Nwaubani Daniel^{1,2}, Amako Ngozi¹, Omekara Israel¹

¹ Michael Okpara University of Agriculture, Umudike

PMB 7267, Umuahia Umudike, Abia State, Nigeria

² Morgan State University

1700 East Cold Spring Lane, Baltimore, Maryland, 21251, USA

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Corresponding Author:

Odo Ekene

odo.samuel@mouau.edu.ng

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Abstract. Extraction was by cold maceration using methanol. The extract was concentrated in vacuo to yield a brown solid of 120.191 g. The crude methanol extract was partitioned into n-hexane 0.1 g, dichloromethane 2.5 g, ethylacetate 4.6 g, and methanol 10.0 g fractions via coarse chromatography. Secondary metabolites identified; 21.73±0.36% alkaloids, 40.78±0.27% flavonoids, 15.99±0.044% saponins, 6.088±0.06% tannins, 3.086±0.03% terpenoids and 12.13±0.01% Cardiac Glycosides. Acute toxicity test showed no death in the rats administered with low dose (1000 mg/kg) and high dose (6000 mg/kg). Analgesic activities of crude methanol extract and fractions showed that at a low dose of 4000 mg/kg of the extract and the fractions have percentage inhibition of pains as methanol crude (63.48±4.62), methanol fraction (79.14±7.39), dichloromethane fraction (60.79±6.69), ethylacetate fraction (23.26±9.75) and n-hexane fraction (64.82±9.75). At a high dose of 8000 mg/kg, the percentage inhibition of pain was 5200±2.00, 71.87±7.04, 80.31±6.20, 45.61±12.60 and 43.87±8.13. Statistical analysis; P>0.05 confidence level, methanol fraction recorded highest analgesic activities while ethylacetate fraction had least.

Keywords: Alkaloids; Cardiac glycosides; Flavonoids; Pains; Rats.

INTRODUCTION

Africa, particularly Nigeria, has an excellent history of using herbal medicine to treat diverse health problems with indisputable success. This superb traditional practice is transferred from generation to generation. The efficacy of herbal medicine cannot be currently argued locally and internationally. Nature has provided vegetation to humanity, which comprises abundant medicinal plants of different diversities located in different habitats like tropical Rain Forest, Savanna Grass Land, Sahel Savanna and desert regions of the world. The various medicinal plant parts (root, bark, stem, flower, leaves, fruits and seeds) could be used in folklore and allopathic medicine to fight against numerous health challenges [1]. This is done to improve the healthy living of the populace and facilitate the growth of the improved productive economy.

Humanity faces different challenges due to the proliferation of diseases caused by organisms found everywhere in the environment. The author [2] opined that using plants to manage and treat diseases is an age-long practice. According to him, it has been found that many plants possess medicinal value, and different parts of these plants have helped synthesise medicines used in hospitals, clinics, and other health care centres. Traditional medicine is leading the front-line research for finding a solution to many health challenges which are so common in society. In Nigeria, many diseases were treated and are still being treated with the use of folk medicine, most of which have shown positive results. These diseases include pains, muscle inflammation, ulcers, convulsions, diarrhoea, bacterial and fungal infections, asthma, malaria, diabetes, and typhoid fever. Some of the numerous medicinal plants already in use in Nigeria are: *Garcinia kola*, which is used for

the treatment of asthma; *Carica Papaya* used for hypertension; *Ocimum basilicum* for typhoid fever; *Senna occidentalis* for skin diseases and *Newbouldia laevis* for pains [3].

These medicinal plants' efficacy is based primarily on their diverse phytochemical constituents. Plants contain an array of phytochemicals certified to be pharmacologically active and have been mostly utilised in treating many diseases wrecking both man and animals.

The author [4] stated that a significantly greater area of Nigeria's ecological zones is made up of numerous plant species, which have found their usefulness in the healthcare of the populace. According to [5], the medicinal uses of most of these plants are numerous. They cannot be exhausted in respect of oral traditions and folklores from time immemorial that have continued to increase the medicinal potencies of these plants and their crude extracts. He further stated that there are different bioactive properties embedded in medicinal plants, such as flavonoids, alkaloids, tannins, saponins, glycosides, etc. These bioactive properties obtainable from a wide range of pharmaceutically derived medications consist of components obtained from plants' phytochemicals. The bioactive properties in medicinal plants possess the healing property attributed to such plants [6].

The medicinal plant's secondary metabolites are believed to be intermediates in metabolic processes found in nature and are usually small molecules. Primary metabolites are involved directly in average growth, development and reproduction, for example, (ethanol, acetic acid, citric acid, lactic acid) and cell components (lipids, vitamins and polysaccharides). Secondary metabolites on their own are not directly involved in those processes and usually have a duty not crucial for the organisms, e.g. antibiotics, proteins and carotenoids.

One of such medicinal plants abundant in nature is *Newbouldia laevis* P.Beauv (*Bignoniaceae*). The plant is known to possess great potency in treating *elephantiasis*, *syphilis*, rheumatic swellings, and as a vermifuge to roundworms. The plant has also been identified to help treat sore feet, ear ache, chest pain, children's convulsion and epilepsy [1]. Many ethnic medicinal activities have been reported to be associated with *Newbouldia laevis* leaves extract, such as uterine stimulants, treatment of

arthritis and rheumatism, gastro-intestinal treatment and all kinds of body pain [7].

Investigations from [8] revealed that some phytochemicals are embedded in *Newbouldia laevis*. The leaves of *Newbouldia laevis* P.Beauv are used in folk medicine to treat various ailments like body pain, rheumatism, arthritis, inflammations and others. The crude extract has been found potent in some diseases. This is why the decoction of the leaves is consumed locally as a remedy to many health challenges. There are essential drugs isolated from *Newbouldia laevis* leaves, such as digoxin and digitoxin, which help treat congestive heart failure [9]. Taxol is another drug extracted from the plant. Taxol is used as a cancer chemotherapeutic drug [10]. Newbouldine is another drug for pain relief and antimalarial. Also, lysergic is a drug produced from ursolic acid obtained from *Newbouldia laevis* [11]. Similar research by [12] states that phytochemicals are chemical substances found naturally in plants. Some of these phytochemicals are responsible for colour and organoleptic properties, such as the deep purple of blueberries and the smell of garlic. In contrast, many others are responsible for protecting fruits, plants, vegetables, cereals, beans, and plant-based beverages like tea and wine. Phytochemical screening of the methanolic extract showed the presence of flavonoids, tannins, glycosides, alkaloids, terpenes and steroids [8, 13].

Research done recently by [14] revealed that the leaf crude methanol extract using white whisker albino rats showed no toxic effect at $LD_{50} > 6000$ mg/kg. The researcher conducted similar research on the same laboratory animals. The plant was found to be a very active analgesic at all concentrations and doses. Despite a lot of research in human medicine, challenges are still encountered. One of these leading challenges is body pain, which arises from other ailments and symptoms of a particular sickness. The effect of such medicinal disease conditions is prevalent in Nigeria due to the relative unavailability of medicines with promising efficacy and the proliferation of adulterated analgesic drugs [1].

This research mainly focused on the extraction of the bioactive properties of *Newbouldia laevis*, the study of its structures and the evaluation of their analgesic efficacy in some laboratory animals.

MATERIALS AND METHODS

Extraction. A solvent distillation machine (PS/1598) was used to distil the solvents, and big glass containers were used for cold maceration.

Toxicity Test. Feeder (gavage), cage (Hamster), plates/spoons, weighing balance, string and experimental animals (white albino rats) were used.

Analgesic Test. Weighing balance, string and white albino rats (experimental animals) were used.

Collection and preparation of plant material.

The leaves of the plant *Newbouldia laevis* were collected at Amede, Eha-Amufu, Enugu State, on 8th November 2019. Foresters confirmed the leaves at the College of Natural Resources and Environmental Management, Michael Okpara University of Agriculture, Umudike, Abia State. The Herbarium Number was identified as Dar-amola FHI 35500.

The leaves were properly washed and air dried. It was further grounded into powder, weighed and was found to be 800 g. It was soaked with distilled methanol, and after two weeks, it was filtered, and the filtrate was refluxed. Thus the solvent was recovered. The crude methanol extract was then kept to air dry. After one week, it was weighed and found to be 120.191 g. It was thus labelled MCNL.

After the fresh leaves of *Newbouldia laevis* were collected and identified, it was washed, chopped and dried under shade. The dried leaves were thus pulverised to give 800 g. The 800 g was put in a big glass container, and 100 % of methanol was poured into the container to the brim. The container was thus covered and kept. After two weeks, the sample was filtered, the filtrate was refluxed, and the crude extract was kept to dry.

The residue was resoaked again in 100 % methanol, and one week later, it was filtered, and the filtrate was refluxed. Thus, the crude extract was left to air dry. The dried crude extract was weighed to give 120.191 g. It was therefore labelled *Newbouldia laevis* crude methanol (NLCM).

This method of extraction is called cold maceration. The NLCM obtained was used for bioassay, phytochemical screening and fractionation.

The NLCM 120.191 g was thus partitioned via coarse chromatography to give different fractions as NLH – 0.1 g, NLD – 2.5 g, NLE – 4.6 g, and NLM – 10.0 g.

Phytochemical screening. The phytochemical screening of the NLCM was done according to standard methods. Thus, the phytochemical analysis is as follows:

Test for Alkaloids. This was done according to [15]. About 5 g of the sample was weighed into a 250 ml beaker, and 200 ml of 10 % acetic in ethanol was added. It was covered and allowed to stand for 2 hours. This was thus filtered, and the extract was concentrated to $\frac{1}{4}$ of the original volume in a water bath. Concentration $\text{NH}_{3(\text{aq})}$ was then added dropwise till a precipitate was formed. The deposit was collected and washed with dil. $\text{NH}_{3(\text{aq})}$ and filtered. The residue was dried and weighed, and calculated as:

$$\% \text{ Alkaloid} = \frac{W_1 - W_2}{W_t \text{ of sample}} \times 100. \quad (1)$$

Test for Flavonoids. The test was by [15], 5 g of the sample was boiled in 50 ml of 2mHCL solution for 30 min under reflux. It was allowed to cool and then filtered. A measured volume of NLCM extract was treated with equal ethylacetate starting with a drop. The solution was filtered into a weighed crucible. The filtrate was heated to dryness in an oven at 60 °C. The dried crucible was weighed again, and the difference in the weight gave the quantity of flavonoid and calculated as:

$$\% \text{ Flavonoid} = \frac{W_t \text{ of dried extract}}{W_t \text{ of sample}} \times 100. \quad (2)$$

Test for Saponins. The method adopted for this test is according to [16]. 100 cm³ of 20 % aqueous methanol was added to a conical flask. The mixture was heated over a water bath for 4 hours. It was stirred at 55 °C. The residue of the mixture was re-extracted with another 100 cm³ of 20 % aqueous methanol after filtration and heated for 4 hours at a temperature of 55 °C with constant stirring. The combined extract was evaporated to 40 cm³ over a water bath at 90 °C. 20 cm³ of diethylether was added to the concentrated solution in a 250 cm³ separator funnel and vigorously agitated from

which the aqueous layer was recovered while the ether layer was discarded. This was repeated twice. 60 cm³ of n-butanol was added and extracted twice with 10 cm³ of 5 % NaCl. After discarding the NaCl layer, the remaining solution was heated in a water bath for 30 minutes. The answer was transferred into a crucible and was dried in an oven to a constant weight. It was calculated as:

$$\% \text{ Saponin} = \frac{W_i \text{ of Saponin}}{W_i \text{ of sample}} \times 100. \quad (3)$$

Test for Tannins. Tannins were determined via a method by [17]. 1 ml of the sample extract of concentration 1 mg/ml was taken in a test tube. The volume was made up to 1 ml with distilled water, and 1 ml served as the blank to 0.5 ml of Folin's phenol reagent (1:2), followed by 5 ml of 35 % Na₂CO₃ added and kept at 25 °C for 5 minutes. A blue colour was formed, and the colour intensity was read at 640 nm. The tannin was determined and calculated as:

$$\% \text{ Tannin} = \frac{AT \times VF \times DFX}{AS \times VE \times WS} \times 100, \quad (4)$$

where *AT* – Absorbance of tannin solution; *AS* – Absorbance of sample solution; *VF* – Total volume of filtrate; *VE* – Volume of extract analysed; *DF* – Dilution factor; *WS* – Sample weight.

Test for Terpenoids. The method adopted here is from [18]. Dried plant leaves extract 100 mg (*W_i*) was taken and soaked in 9 ml of ethanol for 24 hours. The extract after filtration was extracted by 10 ml of petroleum ether using a separating funnel. The quote was thus separated in pre-weighed glass vials and waited for its complete drying (*W_f*). Ether was evaporated, and the formula measured the yield (%) of total terpenoids content:

$$\% \text{ Terpenoids} = \frac{W_i - W_f}{W_i} \times 100. \quad (5)$$

Test for Cardiac Glycosides. The method recommended by [16]. 22.5 g of the dried sample was weighed into a 250 cm³ flask, and about 200 cm³ of distilled water was added to 1g of each dry wood powder sample and allowed to

stand for 2 hours for autolysis to occur. Full distillation was carried out in a 250 cm³ conical flask containing 20 cm³ of 2.5 % NaOH in the sample after adding an antifoaming agent (tannic acid), cardiac glycosides (100 cm³), 8 cm³ of 6 M NH₄OH and two cm³ of 5 % KI were added to the distillate(s), mixed, and titrated with 0.02 M AgNO₃ using a micro burette against a black background. Continuous turbidity indicates the endpoint. Cardiac glycosides were calculated as:

$\% \text{ Cardiac glycosides} =$

$$= \frac{\text{Titre value (cm}^3\text{)} \times 1.08 \times \text{Extract value}}{\text{Aliquot volume (cm}^3\text{)} \times \text{Sample } W_i \text{ (g)}} \times 100 \quad (6)$$

Toxicity Test. Animal stock: The experimental animal used was 18 adult white whisker albino rats of mixed sex. The rats were obtained from an animal house in the College of Veterinary Medicine, Michael Okpara University of Agriculture Umudike, Abia State. The rats were starved of food and water for 24 hours inside a rate cage. The method used for the acute toxicity test (LD₅₀) was according to [19].

Method for Acute Toxicity (LD₅₀) Study of The Extract. About 1.6 g of the crude methanol extract was used for the acute toxicity test. Eighteen adult white whisker albino rats were used. The rats were divided into six groups of 3 rats each, with each group used in a separate cage.

Group 1 was administered 1000 mg/kg body weight of the extract, while groups 2, 3, 4, 5 and 6 were administered 2000, 3000, 4000, 5000 and 600 mg/kg body weight of the animals, respectively. All treatments were done via the oral route using oral gavage, and animals used for the experiment were handled by the OECD rules for care and use of laboratory animals [20]. After the administration, the rats were returned to top cages. They were allowed free access to feed and water and, at the same time, monitored for the development of toxicity signs and mortalities within 24 hours of treatment and an additional seven days. At the end of the period, LD₅₀ values for the extract were determined according to Kaber's formula:

$$L_{D50} = L_{D100} - \frac{\sum DDXMD}{N}; \quad (7)$$

LD₅₀ > 600 mg/kg

Analgesic Test. The method employed for the analgesic test of the crude methanol extract and fractions was according to [21].

Fourteen adult white whisker albino rats were used. The rats were divided into seven groups of 2 rats each, with each group housed in a separate cage. The body weight of each group was taken. Group 1, the control group, were thus given a dose of (1000 mg/kg) of a standard drug (aspirin). The analgesic activity was evaluated by observing the number of writhes they made in 30 minutes. Lower doses of (400 mg/kg) of crude methanol and methanol n-hexane extracts were given in Vivo to groups 2, 3, 4, 5, 6 and 7, respectively, and the number of writhes the rats made in 30 minutes were observed for the analgesic activity and calculated through mean \pm SEM. Also, higher doses of (800 mg/kg) of the crude methanol, methanol, dichloromethane, ethylacetate and n-hexane extracts were given in Vivo to groups 2, 3, and 4, 5, 6 and 7, respectively. The number of writhes the rats made in 30 minutes was observed for the analgesic activities of each extract, which was calculated via mean \pm SEM.

The volume (ml) of the extracts administered to the rats were calculated:

$$V_{ia} = \frac{\text{Dose} \times \text{Average body weight}}{\text{Concentration of extract}} \times 100. \quad (8)$$

RESULTS AND DISCUSSION

The extracts NLM, NLE, and NLDCM obtained were used to conduct chemical analysis viz: phytochemical screening, column chromatography, thin layer chromatography, and NMR spectroscopy (Table 1).

Also, a toxicity test was carried out on the crude extract to evaluate if it is toxic or edible. The analgesic test was equally done on the natural extract to examine its analgesic activity on laboratory animals.

Table 1 – The extracts NLM, NLE and NLDCM with their yields and appearance

Extract	Yield (%)	Appearance
NLM	10.0	Reddish brown
NLE	4.6	Greenish yellow
NLDCM	2.5	Dark green
NLH	0.1	Yellow

The quantitative phytochemical screening of methanol crude extract in Table 2 shows that NLCM contains 21.73 ± 0.36 % alkaloids, 40.78 ± 0.27 % flavonoids, 15.99 ± 0.04 % saponins, 6.088 ± 0.06 % tannins, 7.086 ± 0.03 % terpenoids, 0.00 ± 0.01 % steroids, 12.13 ± 0.01 % Cardiac Glycosides.

Table 2 – Results of qualitative and quantitative phytochemical screening of methanol crude extract of *Newbouldia laevis* leaves

Phytochemicals Present		Mean \pm SEM (%)
Alkaloids	++	21.73 ± 0.36
Flavonoids	++	40.78 ± 0.27
Saponins	++	15.99 ± 0.04
Tannins	+	6.088 ± 0.06
Terpenoids	+	7.086 ± 0.03
Steroids	-	0.00 ± 0.01
Cardiac Glycosides	+	12.13 ± 0.01

Alkaloids possess a lot of pharmacological and therapeutic potentials such as analgesic, antihyperglycemic, anticancer, anti-inflammatory, anti-diarrhoeal, etc. Many alkaloids are used as bitter supplements to stimulate the digestive system Bruneton [5].

Also, alkaloids in foods and supplements help improve immune activities, nervous systems and physical performance, as reported by [22]. *Newbouldia laevis*, rich in alkaloids, can be used to prevent and treat infections and relieve pains from arthritis, inflammations, rheumatism and others. The use of *Newbouldia laevis* leaves in traditional herbal medicine as an analgesic, uterine stimulant, antimalarial, and anticonvulsant may be attributed to its high amount of alkaloids.

Flavonoids possess antibacterial, anti-inflammatory, anti-allergic and antiviral activity [23]. Many observed effects have been linked to their known functions as potent antioxidants, free radical scavengers and metal chelators [24]. Foods rich in flavonoids help fight against cancer, stroke, and obesity and lower blood pressure in hypertensive patients [25].

The high occurrence of flavonoid [40.78 ± 0.27] in the NLCM implies that *Newbouldia laevis* leaves an excellent pain killer, anti-inflammatory, anticancer, antioxidant, antihyperglycaemic and other pharmacological activities attributed to *Newbouldia laevis* leaves.

Saponins were also present in the NLCM extract of *Newbouldia laevis* leaves. Saponins have broader applications in beverages, food ingredients and cosmetics. Reports reveal that saponins are an excellent antioxidant, anti-inflammatory, anti-allergic, anticancer, antiviral and antifungal [26–32].

Also, Saponins possess anti-hypercholesterolemia activity. Hypercholesterolemia is implicated in cardiovascular diseases [33]. This implies that consumption of *Newbouldia laevis* decoction may help prevent and reduce the risk of cardiovascular diseases.

Tannins are used in traditional herbal medicine to precipitate proteins. It also possesses pharmacological activities such as antidiarrheal, anti-inflammatory and wound healing [34]. Tannins are a class of compounds found in the leaves of *Newbouldia laevis*. These polyphenolic compounds are used to relieve fatigue and sore throat immediately. Tannins equally heal burns and stop bleeding [35]. These therapeutic effects of tannins are attributed to the ability of tannins to act as free radical scavengers and to activate antioxidant enzymes [36]. The presence of tannins in *Newbouldia laevis* leaves may be responsible for using the plant in traditional medicine as a pain-killer and for treating rheumatism and arthritis.

Terpenoids are another naturally occurring secondary metabolite found in the leaves of *Newbouldia laevis*, and terpenoids serve as cancer chemotherapy. Other important pharmacological and therapeutic uses of terpenoids are antihyperglycemic, anti-inflammatory, anti-arthritis, antioxidant and antimicrobial. Terpenoids like taxol derivatives (paclitaxel and docetaxel) are good cancer drugs. Also, terpenoids like α and β – amyrins are used in herbal folk medicine as analgesics [37]. Terpenoids present in *Newbouldia laevis* are why the decoction of this plant is consumed as a remedy for body pain.

Cardiac glycosides are a class of compounds present in *Newbouldia laevis* leaves. Cardiac glycosides have the potency to increase the heart's output force and decrease its contraction rate by acting on the cellular Sodium-potassium ATPase [38]. Cardiac glycosides are potent therapies for chronic heart failure, oedema, arrhythmias and complete ventricular rate control in persons with atrial fibrillation; this is why the plant is used in traditional medicine as anti-inflammation.

Table 3 – Acute toxicity study of Methanol leave extract of *Newbouldia laevis* LD₅₀ result of the extract

Groups	Dose administered (mg/kg)	No of deaths	Dose difference (DD)	Mean death (MD)	DD × MD
1	1000	0	500	0	0
2	2000	0	1000	0	0
3	3000	0	1000	0	0
4	4000	0	1000	0	0
5	5000	0	1000	0	0
6	6000	0	-	-	-

The LD₅₀ value is determined according to Karbar's formula, which states that:

$$LD_{50} = LD_{100} - \frac{\sum DD \times MD}{N} \quad (9)$$

$$LD_{50} > 6000 \text{ mg/kg.}$$

Results of the acute toxicity study showed that no death was observed in all test groups administered the extract at all dose levels. The animals instead retained their agility and were physically active throughout the period. The rats helped the highest dose (6000 mg/kg), though they appeared calm immediately after administration of the extract and soon regained their physical activities within a short time. These results are presented in Table 3 above. This finding is by [14], which showed no toxic effect when white whisker rats were treated with leaf crude methanol extract of *Newbouldia laevis*.

Treatment with the crude extract and fractions significantly inhibited acetic acid-induced pain in the experimental rats compared with control ($P < 0.05$). While in the control group, the number of writhes made in 30 minutes was 30.00 ± 5.33 and represented 0 % percentage inhibition of pain, the group treated with aspirin made 8.80 ± 1.02 writhes, representing 64.20 ± 9.74 % inhibition of pain. Percentage inhibitions of pain for 400 mg/kg of crude, methanol, dichloromethane, ethyl acetate and N-hexane extracts were 63.48 ± 4.62 , 79.14 ± 7.39 , 60.79 ± 6.69 , 23.26 ± 9.75 and 64.82 ± 10.14 %, respectively. However, at 800 mg/kg extract treated groups, the inhibitions for the same extracts were 52.00 ± 12.00 , 71.87 ± 7.04 , 80.31 ± 6.20 , 45.61 ± 12.60 and 43.87 ± 8.13 %, respectively. Lower doses of the crude, methanol and N-hexane extracts had better analgesic effects than higher doses. The opposite was in groups treated

with dichloromethane and ethyl acetate extracts. The analgesic activities of the sections also did not significantly differ from that of aspirin ($P>0.05$), except for the ethyl acetate low dose group. From the results obtained, the extract with the highest activity was the methanol extract, while ethyl acetate extract had a minor analgesic activity. The results are presented in Table 4 and Figure 1.

Table 4 – Analgesic Effects of the Crude Methanol Extract and its fractions of the Leaf extracts of *Newboudia laevis* on Whisker Albino Rat

Groups	Treatment Dose Administered	Number of Writhes in 30 min	% Activity of the Extracts
1	Control	30.00 ± 5.33**	0.00 ± 0.00**
2	Aspirin AS (100mg/kg)	8.80 ± 1.02*	64.20 ± 9.74*
3	Crude methanol extract NLCM (400 mg/kg)	10.00 ± 0.89*	63.48 ± 4.62*
4	Crude methanol extract NLCM (800 mg/kg)	12.00 ± 0.89*	52.00 ± 12.00*
5	Methanol extract NLM (400 mg/kg)	5.60 ± 1.94*	79.14 ± 7.39*
6	Methanol extract NLM (800 mg/kg)	8.40 ± 2.48*	71.87 ± 7.04*
7	Dichloromethane extract NLD (400 mg/kg)	14.00 ± 2.28*	60.79 ± 6.69*
8	Dichloromethane extract NLD (800 mg/kg)	6.00 ± 2.28*	80.31 ± 6.20*
9	Ethyl acetate extract NLE (400 mg/kg)	24.00 ± 4.56**	23.26 ± 9.75**
10	Ethyl acetate extract NLE (800 mg/kg)	17.20 ± 2.73**	45.61 ± 12.60*
11	n-hexane extract NLH (400 mg/kg)	8.60 ± 1.33*	64.82 ± 10.14*
12	n-hexane extract NLH (800 mg/kg)	17.60 ± 2.40**	43.87 ± 8.13*

Results are expressed in Mean±SEM. Means marked * are significantly different from control, while that marked ** are substantially different from aspirin.

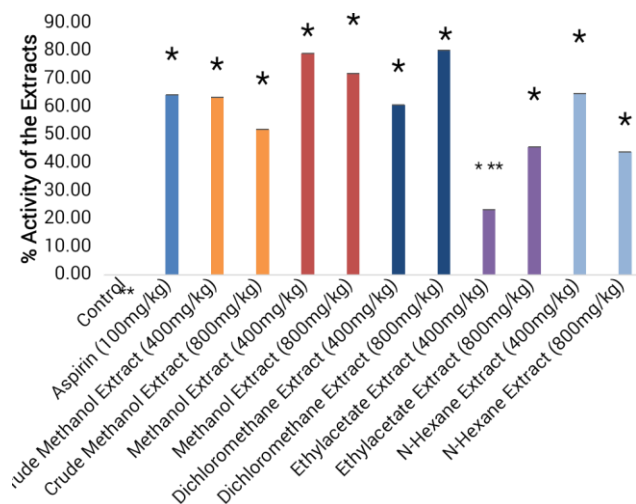


Figure 1 – The percentage analgesic activities of different doses of the extracts

CONCLUSIONS

The results obtained from phytochemical screening, toxicity test and the analgesic test was done on the NLM, NLDCM, and NLHEX extracts from the leaves of *Newboudia laevis* gave the following findings: phytochemical screening revealed the presence of the following phytochemicals; alkaloids, flavonoids, Saponins, Tannins, Terpenoids and cardiac glycosides. The acute toxicity test showed that test groups administered the extract at all doses. The result of the analgesic test showed that methanol extract exhibited the highest inhibition of pain at lower doses of 400 mg/kg body weight of the laboratory animals. In contrast, the ethylacetate extract had minor analgesic activity at the same dose with methanol. From these results and the various analyses done on the leaf extracts of *Newboudia laevis*, it is evident that this plant contains bioactive components that can be used to treat multiple health problems such as pain, inflammation, and oedema, rheumatism and arthritis and heartburn. Thus, it can be said that these research work aims have been achieved.

From the results obtained in this research, people are encouraged to use the leaves of *Newboudia laevis* to treat pains. Pharmaceutical industries should immediately start using the extracts from this plant to produce more potent (pain killer) drugs that will have fewer side effects than Ibumol, Ibuprofen, Diclofenac, et cetera.

The government should enact laws to conserve shrubs, subshrubs and trees like *Newbouldia laevis* to maintain their abundance for various uses. People in different places should embrace the ethnomedicinal services of the plant *Newbouldia laevis* in order to obtain the medicinal benefits embedded in the plant.

This research on the leaf extract of *Newbouldia laevis* shows that people can use it to treat pains. The leaf extract of *Newbouldia laevis* is efficient, non-toxic, potent and reliable for treating body pain. This finding will go a long way in helping further research work on the leaf of *Newbouldia laevis*.

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