Effect of *Newbouldia Leavis* Aqueous Extract on Cardiac Troponin, Creatinine Kinase, Myoglobin and Lactate Dehydrogenase in Albino Rats Treated with Diclofenac

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Abstract

Diclofenac is a commonly used non-steroidal anti-inflammatory drug (NSAID) associated with adverse cardiovascular effects, including myocardial injury marked by elevated levels of cardiac biomarkers such as troponin, creatine kinase-MB (CK-MB), myoglobin and Lactate Dehydrogenase (LDH) *Newbouldia laevis*, a medicinal plant known for its antioxidant and anti-inflammatory properties offer protective effects against such drug-induced cardiotoxicity. This study is aimed to evaluate the serum levels of cardiac troponin, CK-MB, myoglobin and Lactate Dehydrogenase (LDH) in albino rats treated with diclofenac and varying doses of *Newbouldia laevis* extract. Ethical approval was obtained from the ethical committee of Madonna University, Elele Campus, Rivers State. A total of 20 albino rats were randomly divided into five groups of four rats each. Group 1 served as the negative control which received normal rat food and water, Group 2 received normal rat food, water and diclofenac. The mean cardiac myoglobin and LDH level in group one (positive control) were 3.8 ± 2.0 and 137.7 ± 3.2, group two (negative control) were 61.6 ± 3.7 and 195.2 ± 7.3, group 3 were 48.3 ± 13.2 and 180.0 ± 9.1, group four were 21.8 ± 6.9 and 161.2 ± 19.3 while group five were 4.1 ± 2.1 and 147.5 ± 8.8. However, the mean level of myoglobin and LDH shows a notable significant difference ($p < 0.05$). (5 mg/kg), while Groups 3, 4, and 5 were treated with diclofenac and 200 mg, 400 mg, and 800 mg of *Newbouldia laevis* extract, respectively. Blood samples were collected and analyzed using the Finecare fluorescence immunoassay kit for quantitative measurement of Troponin, CK-MB, and myoglobin levels (CTN I/CK- MB/MYO), while LDH was analysed using fortress diagnostic. Data obtained were analysed using SPSS version 27. The diclofenac-only group showed significantly elevated troponin (19.87 ± 4.34 ng/mL) and CK-MB (8.08 ± 2.04 U/L) compared to the control group (0.05 ± 0.04 ng/mL and 1.98 ± 0.44 U/L, respectively). Co-administration with *Newbouldia laevis* extract, particularly at 400 mg, significantly reduced biomarker levels (0.52 ± 0.56 ng/mL for troponin and 3.53 ± 0.55 U/L for CK-MB). Post hoc analysis showed statistically significant differences between the treatment and control groups ($p < 0.05$). This study shows that *Newbouldia laevis* extract has a cardioprotective effect against diclofenac-induced myocardial injury.

Keywords

Newbouldia Leavis Aqueous, Cardiac Troponin, Creatinine Kinase, Myoglobin and Lactate Dehydrogenase, Albino Rats

1. Introduction

Maintaining life depends on the circulatory system, and any conditions that impair cardiac function can have serious negative effects on health, such as heart disease and myocardial damage. It is commonly known that cardiac biomarkers, like cardiac troponin and creatine kinase-MB (CK-MB), can be used to identify myocardial damage [1]. Skeletal and cardiac muscle fibres include a regulatory protein complex called troponin, which aids in contraction. Heart damage, especially myocardial infarction, is indicated by elevated cardiac troponin levels. Another important indicator for identifying myocardial injury is CK-MB, an isoenzyme of creatine kinase that is frequently utilised in combination with troponin assays. [2]. Because of its effectiveness in lowering inflammation and easing pain, the non-steroidal anti-inflammatory medicine (NSAID) diclofenac is commonly recommended. Nevertheless, research has indicated that long-term diclofenac usage may increase cardiac toxicity by raising cardiac enzyme levels, including troponin, CK-MB, and lactate dehydrogenase (LDH), which are indicators of myocardial stress. One well-known indicator of tissue damage, particularly cardiac infarction, is lactate dehydrogenase (LDH). Increased blood levels of LDH signify tissue deterioration and cellular damage, especially in the heart [3]. Another biomarker that is commonly evaluated to determine the degree of myocardial injury is cardiac myoglobin, which acts as an early sign of heart muscle damage. Monitoring these indicators is essential for determining the possible hazards of drug-induced cardiotoxicity, especially in light of the growing usage of NSAIDs like diclofenac.

Concerns have been raised about the long-term use of NSAIDs due to their possible cardiotoxic effects, particularly in patients who already have cardiovascular risks. Therefore, it is crucial to identify supplemental treatments that can lessen the cardiac damage caused by diclofenac [4].

Known as "Ogirisi" in Igbo, "Aduruku" in Hausa, and "Akoko" in Yoruba, Newbouldia laevis, also called the "African Border Tree" or "Tree of Life," is a medicinal plant that is used extensively in West Africa for its anti-inflammatory, antioxidant, and wound-healing qualities [5]. With a long history in African ethnomedicine, this plant has garnered special attention as a traditional medicinal herb. Newbouldia leaves are well-known for their anti-inflammatory, antibacterial, and antioxidant qualities, which may offer defence against harm brought on by oxidative stress, including cardiotoxicity. Consequently, it is thought that the leaf extract of Newbouldia laevis may operate as a cardioprotective agent to lessen the rise of cardiac enzymes brought on by diclofenac [6].

In albino rats given diclofenac, this study will examine the protective effect of Newbouldia laevis leaves on the levels of cardiac biomarkers, specifically troponin, CK-MB, myoglobin, and lactate dehydrogenase (LDH). This study intends to contribute to safer therapeutic options for regulating inflammation while minimising cardiac risks by investigating Newbouldia's potential function in lowering cardiac enzyme increase [7].

2. Materials and Methods

2.1 Experimental Animals

Twenty (20) albino rats of eight weeks old, weighing between 100 - 200g were obtained from the livestock breeding unit of animal friendly farm, Royce Road Owerri, and were used as the experimental animals. The rats were weighed and kept in cages for two weeks at the animal house, Madonna University, Nigeria Elele Campus Rivers State and allowed to acclimatize with their new environment. Within these periods the animal whose life span is 2.5-3.5 years were feed with vital grower food and water. The protocol was in line with the guideline of the National Institute of Health (NIH) (NIH Publication 86-23, 1985) for laboratory animal's care and use.

2.2 Ethical Approval.

This study was conducted according to the rules and regulations of Madonna University, Elele Campus Ethical committee on the use of experimental animals.

2.3 Reagent

The chemicals and reagent that were used in this work were of analytical grade. They were purchased commercially and the manufacturer’s Standard Operating Procedures (SOP) was strictly followed**.**

2.4 Diclofenac Treatment and Dosage

Diclofenac tablets were purchased from MUTH pharmacy. Each diclofenac tablet had a weight of 100mg.

LD50 =5mg/kg

5mg was dissolved in **10**ml of distilled water**.**

Formula: volume of water used to dissolve drugs / mg of drug dissolved × mg to administer / standard dose of drugs × average body weight of the animals

10ml/5mg× 1.25mg/100g× 100g= 0.3ml

0.3ml of the dissolved drugs was administered per rat daily.

2.5 *Newbouldia laevis* Treatment

Newbouldia leaves were gotten from pharmacognosy garden in Madonna University, Elele Nigeria. The leaves were sorted and dried for a week at room temperature. The leaves were grinded and weighed (529.2g). The grinded mixture was put in a conical flask and 3.2litre of alcohol was added to the grinded mixture. The mixture was thoroughly mixed and closed with foil paper (to avoid contamination). The mixture was stood undisturbed for 72 hours at the Pharmacognosy Laboratory in Madonna University, Elele. After 72 hours, it was filtered with filter paper. It was further separated with Rotary Evaporator into yield, methanol and gas. The yield gotten was then refrigerated till the time for treatment at a temperature of 2 to 8ºc.

DOSAGE:

The *Newbouldia* extract (42200mg/42.2g) was dissolved in 70ml of distilled water.

FORMULA: volume of water (ml) used to dissolve the drugs/total weight of drug(extract) × mg to administer/standard dose of drug ×average body weight of animal used.

Total weight of drug(extract)- 42200mg

Volume of water used to dissolve drugs - 70ml

Average body weight - 100g

Standard dose of drug - 1000mg

2.6 Study Design

The study was designed as a controlled experimental trial aimed at investigating the effect of *Newbouldia* leaves on cardiac troponin and CK-MB enzyme levels in albino rats treated with diclofenac. The study adopted a randomized, controlled, and parallel-group design. Rats were randomly assigned to different treatment groups, which were carefully controlled to assess the effects of *Newbouldia* leaf extract when combined with or administered independently of diclofenac.

The study consisted of the following groups:

Group 1 (Negative Control Group): This group received rat feed and water only.

Group 2 (Positive control group): Rats in this group were treated with rat feed and water and 5mg of diclofenac only.

Group 3: Rats in this group was administered with Diclofenac and 200mg of *Newbouldia* leaf extract.

Group 4: Rats in this group was administered with Diclofenac and 400mg of *Newbouldia* leaf extract.

Group 5: Rats in this group was administered with Diclofenac and 800mg of Newbouldia leaf extract.

2.7 Animal Sacrifice and Sample Collection

Blood samples for the analysis of cardiac troponin, CK-MB, myoglobin and LDH enzyme levels were collected from all rats at pre-determined time points. The blood collection was performed by cardiac puncture, to ensure minimal stress to the animals while obtaining the required samples. The blood was drawn under sterile conditions to prevent contamination, and the animals were anesthetized with chloroform before sampling to minimize pain and distress. Samples were collected in plain tubes and allowed to clot at room temperature. After clotting, the samples were centrifuged to separate the serum from the cellular components. The resulting serum was then stored at 2-8^{O}c until analysis for cardiac troponin and CK-MB levels.

2.8 Laboratory Assay

Troponin, Creatinine kinase, lactate dehydrogenase and myoglobin were determined by standard method.

3. Statistical Analysis

Data obtained from this study were analysed using the Statistical Package for Social Sciences (SPSS) version 27 for windows 10. The results were presented in tables as Mean ± Standard Deviation, and analysis of variance (ANOVA) followed by a post-hoc test if significant differences are found. Values were considered significant at P >0.05 and non-significant at P < 0.05.

4. Results

Table 1. Showing the Cardiac Troponin and Creatinine kinase Enzyme levels among different groups induced with Diclofenac and Newbouldia leaf extract

GROUPS	TROPONIN (ng/ml)	CK-MB(U/L)
GROUP 1	0.05 ± 0.04	1.98± 0.44
GROUP 2	19.87 ± 4.34	8.08± 2.04
GROUP 3	15.44 ± 2.77	5.51 ±0.23
GROUP 4	0.52 ± 0.56	3.53± 0.55
GROUP 5	1.00 ± 1.35	2.15± 0.85
F -Value	68.644	24.134
P - Value	0.000	0.000

Significant at p< 0.05;

Non-Significant at p> 0.05

Table 1 Shows the serum level of Troponin and CK-MB among different groups induced with Diclofenac and Newbouldia extract. Group 1 exhibited mean Troponin serum levels of 0.05± 0.04ng/ml and CK-MB was 1.98± 0.44

U/L. In Group 2, Troponin was 19.87± 4.34ng/ml and CK-MB at 8.08 ± 2.04U/L. Group 3 had Troponin at 15.44± 2.77ng/ml and CK-MB at 5.51±0.23U/L. For group 4, Troponin was 0.52± 0.56ng/ml and CK-MB was 3.53± 0.55 U/L. Group 5 exhibited Troponin level at 1.00 ± 1.35ng/ml and CK-MB at 2.15± 0.85U/L. The F-value of Troponin was 68.644 with a P-value less than 0.05, indicating significant difference. The F-value of CK-MB was 24.134 with a P-value less than 0.05, indicating significant difference.

Table 2. Showing the multiple comparisons of the mean values of Cardiac Troponin and Creatinine Kinase among different groups induced with Diclofenac and Newbouldia extract

GROUPS	TROPONIN (NG/ML)	CK-MB (U/L)
Group 1 VS Group 2	0.000*	0.000*
Group 1 VS Group 3	0.000*	0.002*
Group 1 VS Group 4	0.989	0.273
Group 1 VS Group 5	1.000	0.999
Group 2 VS Group 3	0.100	0.023*
Group 2 VS Group 4	0.000*	0.000*
Group 2 VS Group 5	0.000*	0.000*
Group 3 VS Group 4	0.000*	0.107
Group 3 VS Group 5	0.000*	0.003*
Group 4 VS Group 5	0.997	0.371

Table 3. Showing the Cardiac myoglobin and Lactate Dehydrogenase (LDH) Enzyme levels among different groups induced with Diclofenac and Newbouldia leaf extract

Test (g/l)	GROUP 1	GROUP2	GROUP3	GROUP4	GROUP5	P value	F Value
Myoglobin	3.8 ± 2.0	61.6 ± 3.7	48.3 ±13.2	21.8 ±6.9	4.1 ± 2.1	0.00	55.66
LDH	137.7± 3.2	195.2 ± 7.3	180.0 ± 9.1	161.2 ±19.3	147.5 ± 8.8	0.00	18.17

Table 3 shows that cardiac myoglobin yielded a statistically significant result, with an F-value of 55.66 and a p-value of 0.000, while serum LDH also shows a statistically significant result, with an F-value of 18.17 and a p-value of 0.000. This highly significant p-value ($p < 0.001$) indicates that there are notable differences in cardiac myoglobin and serum LDH levels among the experimental groups: control, diclofenac-only, and combined treatment (diclofenac + extract).

The result leads to the rejection of the null hypothesis, which posits that there are no significant differences in mean cardiac myoglobin and serum LDH levels across the groups. These findings suggest that diclofenac administration caused a significant elevation in cardiac myoglobin and serum LDH, a marker of cardiac injury, while treatment with *Newbouldia laevis* leaf extract—either alone or in combination with diclofenac—altered this effect.

This result strongly suggests that treatment with *Newbouldia laevis* leaf extract has a modulatory or protective effect on LDH levels, particularly in rats exposed to diclofenac.

Table 4. Showing the Multiple Comparison of the Mean Values of Myoglobin (ml/Kg) and LDH (ml/kg) on albino rats treated with diclofenac using the Post Hoc Test (LSD)

COMPARISON	Myoglobin (ml/kg)	LDH (ml/kg)
Group 1 vs Group 2	.000	.000
Group 1 vs Group 3	.000	.001
Group 1 vs Group 4	.018	.064
Group 1 vs Group 5	1.000	.769
Group 2 vs Group 3	.104	.322
Group 2 vs Group 4	.000	.004
Group 2 vs Group 5	.000	.000
Group 3 vs Group 4	.001	.160
Group 3 vs Group 5	.000	.006
Group 4 vs Group 5	.020	.418

5. Discussion

The purpose of this study was to examine how the aqueous extract of Newbouldia laevis affected the levels of cardiac troponin and creatinine kinase in albino rats given diclofenac. Serum troponin (19.87 ± 4.34 ng/ml) and CK-MB (8.08 ± 2.04 U/L) levels were substantially higher in rats given diclofenac alone (Group 2) than in the control group (Group 1), which had values of 0.05 ± 0.04 ng/ml and 1.98 ± 0.44 U/L, respectively (Table 4.1). This statistically significant rise supports studies of diclofenac-induced cardiotoxicity in animal models and points to considerable cardiac stress or injury. Remarkably, both indicators decreased in a dose-dependent manner when diclofenac and Newbouldia laevis extract were administered together. Troponin and CK-MB levels specifically dropped to 15.44 ± 2.77 ng/ml and 5.51 ± 0.23 U/L at 200 mg (Group 3). Troponin and CK-MB decreased to 0.52 ± 0.56 ng/ml and 3.53 ± 0.55 U/L, respectively, at 400 mg (Group 4), indicating an ideal therapeutic effect. Troponin increased somewhat to 1.00 ± 1.35 ng/ml and CK-MB to 2.15 ± 0.85 U/L at 800 mg (Group 5), which was still less than the positive control group but did not significantly outperform the 400 mg dose.

These results imply that the leaf extract of Newbouldia laevis reduces the increase of cardiac enzymes brought on by diclofenac, most likely as a result of its anti-inflammatory and antioxidant properties. Significant differences between the treatment groups are further supported by the ANOVA's significant F-values (Troponin: 68.644; CK-MB: 24.134) and matching p-values ($p < 0.001$ for both markers).

The results of Table 4.1 were supported by post hoc comparisons of Table 4.2. Both troponin and CK-MB showed significant differences ($p < 0.05$) between the diclofenac-only group and the control group (Group 1 vs. Group 2), suggesting that myocardial damage was caused by diclofenac administration. Interestingly, comparisons between Group 1 and Group 4 and Group 1 and Group 5 revealed no significant difference ($p > 0.05$), indicating that Newbouldia laevis extract was able to restore biomarker levels near normal at both 400 mg and 800 mg doses. Additionally, the extract's capacity to lessen diclofenac-induced cardiotoxicity was demonstrated by extremely significant ($p = 0.000$) comparisons between Group 2 and the treatment groups, particularly Groups 4 and 5. It's interesting to note that there was no statistically significant difference between Groups 4 and 5, suggesting that raising the dosage over 400 mg provides no additional benefits and could instead be a symptom of a plateau effect or possibly biphasic dose-response behaviour.

Studies by [8] that documented diclofenac-induced cardiac stress and enzyme leakage into circulation, indicating myocardial membrane breakdown, are consistent with the observed increase of cardiac biomarkers in the diclofenac-treated group. Similarly, [9] pointed out that NSAIDs raise troponin and CK-MB levels via oxidative stress and mitochondrial dysfunction.

Results from [10], who documented Newbouldia laevis's antioxidative and anti-inflammatory properties in a variety of animals, corroborate the cardioprotective impact of the plant. These bioactivities are probably in charge of preserving cardiac membranes and lowering myocardial oxidative damage. Previous phytochemical screenings have found flavonoids, tannins, and phenolic compounds in Newbouldia laevis, all of which are known to scavenge free radicals and reduce inflammatory responses, even though the precise phytochemical constituents causing these effects were not isolated in this study.

Furthermore, in drug-induced cardiotoxicity models, research on other plant extracts with comparable profiles, as that conducted by [11] with Vernonia amygdalina, also showed decreases in troponin and CK-MB levels. This lends more credence to the idea that antioxidants produced from plants can be used as supplements to treat drug-induced cardiovascular disease.

Significant cardiac stress or injury was indicated by the significantly higher cardiac myoglobin levels (61.6 ± 3.7 g/L) in the diclofenac-only group when compared to the negative control (3.8 ± 2.0 g/L). This result is consistent with research by [12], who found that long-term exposure to diclofenac raises cardiac biomarker levels because it damages the heart through oxidative stress. LDH showed a similar pattern, increasing from 137.7 ± 3.2 U/L in the control group to 195.2 ± 7.3 U/L in the diclofenac-only group. The findings of [13,14], who connected elevated LDH levels to diclofenac-induced myocardial injury, are supported by these elevated values, which imply that diclofenac causes cardiotoxicity.

A dose-dependent cardioprotective effect was shown after treatment with Newbouldia laevis extract. When compared to the diclofenac-only group, the co-administration of 400 ml/kg and 800 ml/kg of the extract with diclofenac significantly decreased myoglobin and LDH levels. Interestingly, myoglobin and LDH readings decreased to 4.1 ± 2.1 g/L and 147.5 ± 8.8 U/L, respectively, at the maximum dosage (800 ml/kg), approaching levels observed in the control group. These results are consistent with those of [15], who discovered that by dramatically reducing LDH and troponin levels, Newbouldia laevis extract reduced doxorubicin-induced heart injury in rats. The observed dose-dependent impact is similarly in line with [16,17], who showed that in oxidative stress models, greater extract doses provided superior protection against changes in cardiac enzymes.

6. Conclusion

The results of this study unequivocally show that the leaf extract of Newbouldia laevis considerably lowers the levels of cardiac troponin and CK-MB in albino rats given diclofenac, suggesting a preventive effect against the cardiotoxicity caused by diclofenac. Heart biomarkers were reduced in a dose-dependent manner by the extract, with the 400 mg dose

demonstrating the best normalisation of enzyme levels. By demonstrating that Newbouldia laevis extract has a cardioprotective effect and can lessen the myocardial damage usually connected with long-term diclofenac administration, our findings provide an answer to the study issue.

References

[1] Amaechi, A. A., Oli, A. N., Okezie, U. M., Adejumo, S. A., Abba, C. C., Okeke, I. J., & Okoye, F. B. C. (2021). Secondary metabolites of endophytic fungi from Newbouldia laevis and cassia tora leaves: Prospecting for new antimicrobial agents. Recent Advances in Anti-Infective Drug Discovery, 16(1), 50-62.

[2] Osei-Mensah, B., Boakye, Y. D., Anyan, W. K., Agana, T. A., Aboagye, E. A., Bentil, I., Lomotey, E. S., Adu, F., & Agyare, C. (2023). In Vitro Cercaricidal Activity, Acute Toxicity, and GC/MS Analysis of Some Selected Ghanaian Medicinal Plants. Journal of Parasitology Research, 2023, 4589424.

[3] Alabi, O. A., Okorie, B., Simon-Oke, I. A., Atanda, H. C., Olumurewa, J. A. V., & Adebo, T. C. (2024). Cellular toxicity and DNA damage induced by Newbouldia laevis used for male infertility treatment in prokaryotic and eukaryotic models. Protoplasma, 261(1), 53-64.

[4] Yadav, G., &Ahmaruzzaman, M. (2022). New generation advanced nanomaterials for photocatalytic abatement of phenolic compounds. Chemosphere, 304.

[5] Pham, D. C., Shibu, M. A., Mahalakshmi, B., & Velmurugan, B. K. (2020). Effects of phytochemicals on cellular signaling: reviewing their recent usage approaches. Critical Reviews in Food Science and Nutrition, 60(20), 3522-3546.

[6] Rana, P., Shrama, A., & Mandal, C. C. (2021). Molecular insights into phytochemicals-driven break function in tumor microenvironment. Journal of Food Biochemistry, 45(9).

[7] Lyu, J. Il, Ryu, J., Jin, C. H., Kim, D. G., Kim, J. M., Seo, K. S., Kim, J. B., Kim, S. H., Ahn, J. W., Kang, S. Y., & Kwon, S. J. (2020). Phenolic compounds in extracts of Hibiscus acetosella (Cranberry Hibiscus) and their antioxidant and antibacterial properties. Molecules, 25(18).

[8] Chen, P., Wu, Q., Feng, J., Yan, L., Sun, Y., Liu, S., Xiang, Y., Zhang, M., Pan, T., Chen, X., Duan, T., Zhai, L., Zhai, B., Wang, W., Zhang, R., Chen, B., Han, X., Li, Y., Chen, L., … Sui, X. (2020). Erianin, a novel dibenzyl compound in Dendrobium extract, inhibits lung cancer cell growth and migration via calcium/calmodulin-dependent ferroptosis. Signal Transduction and Targeted Therapy, 5(1).

[9] Mbagwu, I. S., Akah, P. A., &Ajaghaku, D. L. (2020). Newbouldia laevis improved glucose and fat homeostasis in a TYPE-2 diabesity mice model. Journal of Ethnopharmacology, 251.

[10] Alqahtani, F., Mahmood, H. and Alharbi, S. (2022). The impact of diclofenac on oxidative stress and mitochondrial function: A review of its role in hepatotoxicity and cardiotoxicity. Journal of Medical Sciences and Research, 10(2): 134-145.

[11] Ibrahim, M., Oyebanji, E., Fowora, M., Aiyeolemi, A., Orabuchi, C., Akinnawo, B., & Adekunle, A. A. (2021). Extracts of endophytic fungi from leaves of selected Nigerian ethnomedicinal plants exhibited antioxidant activity. BMC Complementary Medicine and Therapies, 21(1).

[12] Zhan, W., Yang, H., Zhang, J., & Chen, Q. (2022). Association between co-exposure to phenols and phthalates mixture and infertility risk in women. Environmental Research, 215.

[13] Amaechi, A. A., Oli, A. N., Okezie, U. M., Adejumo, S. A., Abba, C. C., Okeke, I. J., & Okoye, F. B. C. (2021). Secondary metabolites of endophytic fungi from Newbouldia laevis and cassia tora leaves: Prospecting for new antimicrobial agents. Recent Advances in Anti-Infective Drug Discovery, 16(1), 50-62.

[14] Balakrishnan, R., Azam, S., Cho, D. Y., Su-Kim, I., & Choi, D. K. (2021). Natural Phytochemicals as Novel Therapeutic Strategies to Prevent and Treat Parkinson's Disease: Current Knowledge and Future Perspectives. Oxidative Medicine and Cellular Longevity, 2021.

[15] Murtala, A. A., & Akindele, A. J. (2020). Anxiolytic- and antidepressant-like activities of hydroethanol leaf extract of Newbouldia laevis (P.Beauv.) Seem. (Bignoniaceae) in mice. Journal of Ethnopharmacology, 249.

[16] Bwegne, J. G. B. N., Ayina, C. N. A., wend Pagnangdé, A. H. B., Etaga, N. B., Nzekuie, Q. S., Ouedraogo, M. S., ... & Minkande, J. Z. (2022). Newbouldia's laevis biological effects against metabolic syndrome components: a review. World Journal of Advanced Research and Reviews, 16(2): 121-138.

[17] Ukwubile, C. A., Otalu, O., Abdulrahim, U., Angyu, A. E., Aliyu, Y. K., Njidda, S., & Bingari, M. S. (2020). Ethnomedicinal survey of medicinal plants traditionally used in Ogurugu Community Southeast Nigeria for the treatment of malaria. International Journal of Traditional Complement Medcine; 5(30): 1-15.