EUROPEAN JOURNAL OF BIOMEDICAL AND PHARMACEUTICAL SCIENCES
http://www.ejbps.com

ISSN 2349-8870
Volume: 12
Issue: 9
Year: 2025

COMPARATIVE PHYTOCHEMICAL AND ANTI-INFLAMMATORY ASSESSMENT OF *PERSEA AMERICANA* SEED AND *NAUCLEA LATIFOLIA* ROOT EXTRACTS

Nourou-dine Balogoun[1], Amoussatou Sakirigui*[2], Franck Yovo[3], Jean-Robert Klotoé[4], Victorien Dougnon[5], Gado Dramane[6]

[1]Ph.D. Student, Department of Chemistry, Kaba Research Laboratory in Chemistry and Applications (LaKReCA), University of Technical Sciences, Engineering and Mathematics (UNSTIM), Republic of Benin.
[2,3]Lecturer, Department of Chemistry, Kaba Research Laboratory in Chemistry and Applications (LaKReCA), University of Technical Sciences, Engineering and Mathematics (UNSTIM), Republic of Benin.
[4]Professor, Department of Animal Physiology, Research Unit in Applied Microbiology and Pharmacology of Natural Substances (URMAPha), University of Abomey-Calavi, Cotonou, Republic of Benin.
[5]Lecturer, Department of Animal Physiology, Research Unit in Applied Microbiology and Pharmacology of Natural Substances (URMAPha), University of Abomey-Calavi, Cotonou, Republic of Benin.
[6]Lecturer, Department of Animal Physiology, Laboratory of Natural Sciences and Applications (LSNA), University of Technical Sciences, Engineering and Mathematics (UNSTIM), Republic of Benin.

***Corresponding Author: Amoussatou Sakirigui**
Lecturer, Department of Chemistry, Kaba Research Laboratory in Chemistry and Applications (LaKReCA), University of Technical Sciences, Engineering and Mathematics (UNSTIM), Republic of Benin.

Article Received on 26/07/2025 **Article Revised on 16/08/2025** **Article Accepted on 06/09/2025**

ABSTRACT
Dysmenorrhea, a common menstrual pain condition, is primarily linked to an inflammatory process triggered by excessive prostaglandin production. These mediators cause pelvic pain, digestive, and musculoskeletal disorders, impairing the quality of life of women who suffer from them. Due to the inaccessibility of certain allopathic treatments, the interest in natural, effective, and affordable alternatives must be considered. In this context, the present study aimed to evaluate and compare the anti-inflammatory activity of *Persea americana* (PA) seeds and *Nauclea latifolia* (NL) roots, a plant traditionally used for dysmenorrhea treatment. Hydro-ethanolic extracts of both substrates were obtained by maceration and then subjected to qualitative (coloration and precipitation reactions) and quantitative (spectrophotometry) phytochemical analyses. Anti-inflammatory activity was evaluated *in vivo* in Wistar rats using the carrageenan-induced paw edema model. Both extracts exhibited similar phytochemical profiles, with the presence of polyphenols, flavonoids, tannins, saponins, and reducing compounds. The PA seeds extract was distinguished by its high polyphenol (989.12 mg/g) and flavonoid (26.82 mg/g of extract) content. Biological tests showed significant edema inhibition at doses of 200 and 400 mg/kg, reaching up to 46.15% for PA seeds and 65.38% for NL. PA activity was particularly pronounced during the first three hours, the critical phase of acute inflammation. These results suggest that *Persea americana* seeds, considered a waste product, could be a promising natural alternative for the treatment of dysmenorrhea.

KEYWORDS: Anti-inflammatory activity, *Persea americana* seeds, *Nauclea latifolia* roots, Hydro-ethanolic extracts, Phytochemical analyses.

INTRODUCTION
Perimenstrual inflammation is one of the manifestations of primary dysmenorrhea, characterized by recurrent menstrual pain without identifiable pelvic lesions.[1-3] Indeed, dysmenorrhea is one of the most common gynecological disorders in women of reproductive age, affecting between 50% and 90% of women worldwide.[4,5] In Africa, the prevalence ranges from 56% to 83%, particularly among adolescent girls and university students.[6,7] These data highlight the urgent need for targeted interventions to reduce the impact of this largely underestimated disorder. Despite its major impact on quality of life and productivity, it remains neglected in reproductive health policies.[8] Conventional treatments, such as nonsteroidal anti-inflammatory drugs, remain effective but sometimes have side effects.[9] In addition, many women resort to self-medication due to a lack of access to appropriate care.[10] In this context, the exploration of natural alternatives is gaining interest. Furthermore, the literature reports that extracts from the roots of *Nauclea latifolia* and *Persea americana* seeds are recognized for their anti-inflammatory and antioxidant properties.[11-16] On the one hand, the roots of *Nauclea latifolia* are widely used by the Beninese

population for the management of pain related to perimenstrual inflammation. On the other hand, in Benin, *Persea americana* seeds are considered as waste contributing to environmental pollution. The comparative study of the two substances will contribute to the valorization of the seeds and slow down the destruction of *Nauclea latifolia* plants. This work therefore aims to compare the phytochemical composition and anti-inflammatory activity of hydro-ethanolic extracts of the roots of *Nauclea latifolia* and the seeds of *Persea americana* with a view to enhancing them.

MATERIAL AND METHODS

MATERIAL

Plant Material

Nauclea latifolia root : The *N. latifolia* roots used in this study were harvested in Aoro, in the commune of Bassila, Donga department northern Benin.

Persea americana seeds: The avocado seeds used were collected in Natitingou, in the Atacora department, northern Benin.

Animal Material

For all tests, we used adult female and male albino rats of the Wistar strain, weighing between 80 g and 150 g. The rats were raised in the animal facility of the Applied Microbiology and Pharmacology of Natural Substances Research Unit of the University of Abomey-Calavi (URMAPHA/UAC) at a temperature between 23 and 30°C. They were handled in accordance with the rules of good practice for the use of experimental animals.

METHODS

Powder Preparation

Nauclea latifolia root : The carefully harvested roots were cleaned with clean water to remove debris. They were then crumbled using a cutter and dried in the shade at room temperature in the laboratory for one week. They were then ground into powder using an electric grinder and stored in a glass jar away from light and heat.

Persea americana seeds: The seeds were cleaned and crumbled using a mechanical mortar and then dried in the shade at room temperature in the laboratory for one week. They were then ground into powder using an electric grinder and stored in a glass jar away from light and heat.[15]

Extraction Preparation

The extracts were prepared by maceration using the method of Sakirigui et al., 2020. To 60 g of powder, we added 600 ml of the v/v mixture of distilled water and 96° ethanol. After 72 hours of maceration away from light, the mixture was filtered and then evaporated at 35°C under low pressure until a brown powder was obtained: this is the hydro-ethanolic extract.[15] The yield was calculated according to the following formula:

R = 100 X Mass of dry powder obtained / Mass of plant material

Phytochemical Screening

The phytochemical screening test enabled the identification of the major phytochemical families present in our extract through solubility tests, color reactions, precipitation, and ultraviolet light examinations using the Hughton and Ramann method, revised and adapted to our laboratory conditions.[17] Polyphenol and flavonoid contents were calculated by reading absorbances on a spectrophotometer based on the method of Fagbohoun et al. in 2015.[18,19]

Assessment of Anti-inflammatory Activity

Acute inflammation was induced by injecting carrageenan (1%, 0.5 ml) under the plantar fascia of the rat's right hind paw. Five groups of five rats were formed. Group 1 was treated with normal saline (10 ml/kg), group 2 with diclofenac (10 mg/kg), and groups 3 and 4 received the extract at doses of 200 and 400 mg/kg orally, respectively. These administrations were carried out 30 minutes before the carrageenan injection.[20] Paw volume measurements were taken before and after carrageenan injection for five hours. The percentage inhibition of anti-edema activity was assessed using the following formula : % inhibition = A- B/A x 100

where A represents the edema volume of the control group and B represents the paw edema of the test groups.

Statistical Analysis

Means and standard deviations were calculated using an Excel spreadsheet. Differences between mean values of normally distributed data were assessed using the ANOVA (Dunnett's Test) and tstudent's test. The significance level was set at 5%.

RESULTS

Yield

Following the extractions, the yield was 11.2 ± 3% for *Nauclea latifolia* roots and 11.5 ± 5% for *Persea americana* seeds.

Phytochemical Screening

The qualitative and quantitative analysis of the hydroethanolic extracts of *Persea americana* seeds and *Nauclea latifolia* roots indicated the results shown in Table 1. This table shows that the extracts of *Persea americana* seeds (PA) and *Nauclea latifolia* roots (NL) contain polyphenols, flavonoids, alkaloids, and tannins. However, the terpenes and mucilages present in NL are absent in PA [Table 1].

Table 1: Qualitative and Quantitative Analysis of Hydroethanolic Extracts of *Persea americana* seeds and *Nauclea latifolia* Roots.

Compounds	QL PA	QN PA (mg/g d'extrait)	QL NL	QN NL (mg/g d'extrait)
Reducing compound	+		+	
Alkaloids	+		+	
Flavonoids	+	26.82 ±0.01	+	35.39 ± 0.01
Tanins catechic	+		+	
Tanins gallic	+		+	
Anthocyanins	-		-	
Leuco-anthocyanins	+		+	
Quinonics compound	-		-	
Saponin	+		+	
Coumarin	+		+	
Terpenoids	-		+	
Mucilages	-		+	
Cartenoids	-		-	
Free Anthracenics	-		-	
O-heterosides	-		+	
Polyphénol	+	989.12±0.02	+	48.0±0.02

+ : Presence of metabolite. - : Absence of metabolite

QL PA : Qualitative analysis of *Persea americana* seeds; **QN PA :** Quantitative analysis of *Persea americana* seeds; **QL NL:** Quantitative analysis of *Nauclea latifolia* roots and **QN NL:** Quantitative analysis of *Nauclea latifolia* roots.

Anti-inflammatory activity (anti-edema test)

Evolution of edema

Figure 1 shows a clear induction of edema (inflammation), with a progressive increase in edema up to the 4th hour in the control group (NaCl), reaching 0.30. This evolution confirms the effectiveness of the model. The decrease observed thereafter, particularly under treatment, indicates the anti-inflammatory effect of the tested extracts [Figure 1].

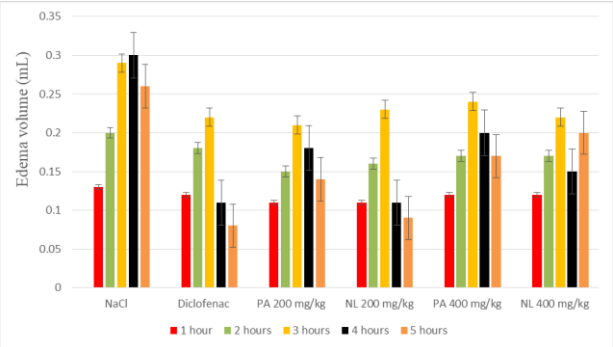

Figure 1: Evolution of edema (inflammation).

Effect of *P. americana* seeds extract on edema

The curves in Figure 2 show the effect of the two doses of hydroethanolic extract of PA seeds on induced edema by determining the inhibition rate. The maximum values after five hours are 46.15%, 34.62%, and 69.23%, respectively, for the extract at doses of 200 mg/kg, 400 mg/kg, and diclofenac.

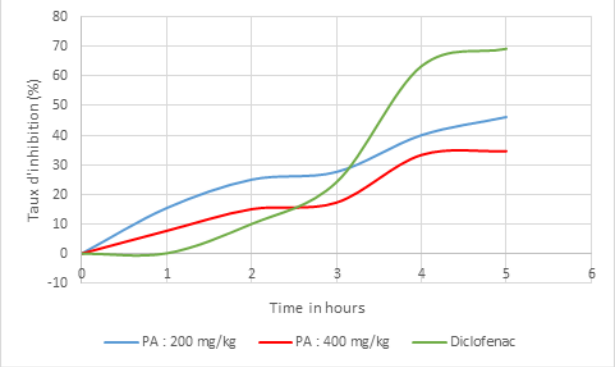

Figure 2: Effect of *P. americana* seeds extract on induced edema in Wistar rats.

Effect of *N. latifolia* extract on edema

The effect of the two doses of the hydro-ethanolic extract of NL roots on induced edema is shown in Figure 3. The maximum levels of 65.38%, 23.08% and 69.23% were respectively determined for the extract at doses of 200 and 400 mg/kg and then at the dose of 10 mg/kg for diclofenac.

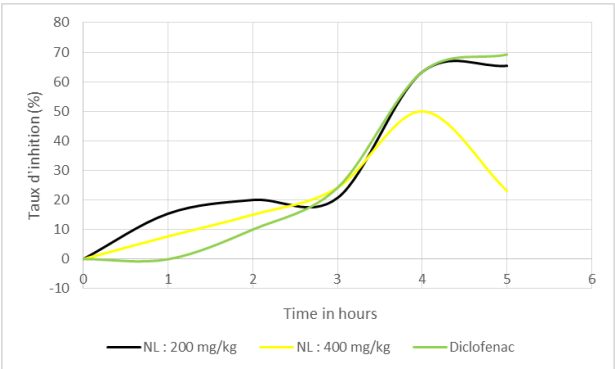

Figure 3: Effect of *N. latifolia* extract on induced edema in Wistar rats.

Comparative study of the activities of the two extracts

The comparative effect of the two doses of the two extracts on induced edema in Wistar rats is shown in Figure 4. These rates increase from 15.38 to 65.38 for the NL extract at 200 mg/kg and then from 7.69 to 23.08 for NL at 400 mg/kg. They increase from 15.38 to 46.15 for the PA extract at 200 mg/kg and then from 7.69 to 34.62 for the extract at 400 mg/kg. The inhibition rate of PA at 200 mg/kg remains highest between 0 and 3 hours. After three hours until the end, the NL rate at 200 mg/kg takes over.

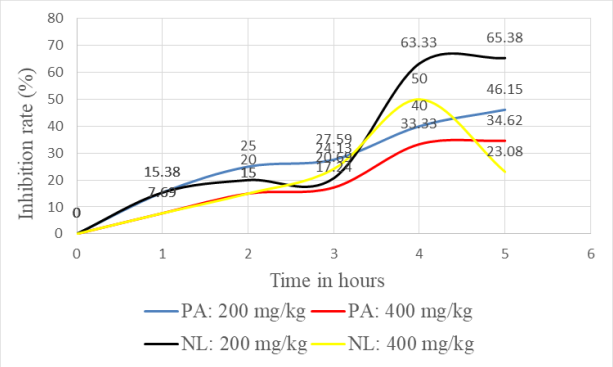

Figure 4: Comparative effects of PA seeds and NL root extracts on induced edema.

DISCUSSION

The comparative evaluation of PA seeds and NL roots extracts in the acute inflammation test (carrageenan-induced edema) reveals an interesting and differentiated temporal dynamic between the two extracts. During the first three hours, the PA seeds extract, particularly at the 200 mg/kg dose, showed a higher inhibition rate than that of NL roots (PA : 15.38%, 25%, 27.59% at hours 1, 2, and 3 vs. NL : 15.38%, 20%, 20.69%). This early effect suggests that PA acts particularly effectively on the initial phase of inflammation [Figure 4]. From the 4th hour onwards, the dynamics reverse : PA (40%) becomes less effective than NL (63.33%), and this divergence widens at the 5th hour (PA 46.15% vs NL 65.38%), with NL approaching diclofenac (69.23%). This distinct temporal profile suggests that PA acts mainly in the early phase, while NL proves more active in the late phase.

Phytochemical analysis sheds light on these observations [Table 1]. The PA seeds extract contains a very high amount of total polyphenols (989.12 mg/g), significantly higher than that of NL (48.00 mg/g). This richness may explain the early effect of PA seeds: polyphenols are known for their ability to rapidly scavenge free radicals, modulate initial inflammatory pathways, and strengthen initial enzymatic defenses.[21,22] However, despite this abundance, PA extract does not excel in the late phase, which could be explained by limited bioavailability or antagonistic effects at high doses of certain polyphenols, as suggested by some studies on the kinetics of polyphenol-rich extracts. This idea is supported by the absence of an increasing effect beyond the 3rd hour, suggesting a saturable or antagonized response. Conversely, NL extract, although low in total polyphenols, is rich in flavonoids (35.39 mg/g vs. 26.82 mg/g in PA), as well as terpenoids, mucilages, and O-heterosides, compounds absent in PA. Flavonoids are well documented to inhibit inflammatory responses in a prolonged manner.[23] Their more sustained release and targeted action on enzymes involved in the late phase of inflammation explain the delayed but long-lasting effect of NL. Furthermore, terpenoids in NL, including sesquiterpenes and triterpenes, are known to effectively reduce edema from carrageenan.[24] Thus, the complementarity of the phytochemical profiles between PA and NL suggests a temporally sequenced action : PA, rich in polyphenols, acts rapidly on early mediators of inflammation, while NL, with its flavonoids and terpenes, becomes more effective during the late phase. This phenomenon illustrates the importance not only of the chemical composition, but also of the synergistic action of the extracts.

It is also interesting to note that doses of 200 mg/kg of PA and NL extracts are generally more effective than the higher doses of 400 mg/kg. This trend is particularly marked for NL, whose efficacy peaks at 65.38% at 5 hours at 200 mg/kg compared to only 23.08% at 400 mg/kg. Similarly, the PA extract reaches 46.15% at 200 mg/kg, compared to 34.62% at 400 mg/kg. Several explanations can be put forward. First, at high doses, some bioactive constituents may exhibit antagonistic effects, inhibiting the action of active compounds by competing with or saturating inflammatory receptors. Second, the solubility, bioavailability, or tissue distribution of the active ingredients may be optimal at low doses and diminish at higher concentrations. Finally, the phytochemistry of the extracts indicates a richness in polyphenols in PA and in flavonoids, terpenes, and mucilages in NL. However, some studies suggest that at excessive doses, polyphenols can induce pro-oxidant activity or disrupt cellular redox balance, as observed by Truong et al., 2022.[25]

Research has also shown that a moderate dose is often more effective than a high dose in plant extracts, as observed with *Vitex trifolia* and *Withania adpressa* in the work of Annamalai et al., 2022 and Salamatullah et al., 2023).[26,27] These results highlight the importance of dosage optimization in the evaluation of the anti-inflammatory activity of phytochemical extracts.

CONCLUSION

In conclusion, PA and NL extracts, effective in the early and late phases of inflammation, respectively, could represent promising alternatives to conventional anti-inflammatory drugs used in the treatment of dysmenorrhea. Their marked activity at a dose of 200 mg/kg, linked to their phytochemical composition, justifies their potential in phytotherapy for the treatment of acute inflammation, while minimizing the adverse effects of synthetic treatments.

ACKNOWLEDGMENTS

We thank the Doctoral School of Sciences, Technologies, Engineering and Mathematics (ED-STIM) for its financial support.

REFERENCES

1. Tu, F., Hellman K. Primary Dysmenorrhea: Diagnosis and Therapy. Obstet Gynecol., 2021; 137(4): 752. doi: 10.1097/AOG.0000000000004341.
2. Khan, S. A., Khan, M. S., Khan, M. A. Role of inflammation in primary dysmenorrhea: A review. Journal of Clinical and Diagnostic Research, 2021; 15(3): 01–04.
3. Liu, Y., Zhang, Y., & Li, X. Inflammatory cytokines and primary dysmenorrhea: A systematic review and meta-analysis. Journal of Pain Research, 2022; 15: 1235–1245.
4. Iacovides, S., Avidon, I., Baker, F. C. What we know about primary dysmenorrhea today: A critical review. Human Reproduction Update, 2015; 21(6): 762–778. https://doi.org/10.1093/humupd/dmv039.
5. Zhang, Y., Sun, X., Wang, Y., & Ma, H. Global prevalence of dysmenorrhea among adolescent girls : A systematic review and meta-analysis. Reproductive Health, 2022; 19(1): 64. https://doi.org/10.1186/s12978-022-01351-6.
6. Gebeyehu, M. B., Mekuria, A. B., Tegegne, K. D., & Alehegn, A. Prevalence of dysmenorrhea and its impact on academic performance among university students in Ethiopia: A systematic review and meta-analysis. Health Services Insights, 2022; 15: 11786329221079443. https://doi.org/10.1177/11786329221079443
7. Acheampong, K., Appiah, C. A., Osei, F. A., & Afriyie, D. O., Prevalence and management of dysmenorrhea among senior high school students in the Ablekuma North Municipality, Ghana. BMC Women's Health, 2019; 19(1): 142. https://doi.org/10.1186/s12905-019-0847-6.
8. Igwenagu, C. E., Eze, I. R., & Oguejiofor, U. C. Menstrual pain and school absenteeism among female university students in Zimbabwe: A cross-sectional study. Pan African Medical Journal, 2023; 44: 31. https://doi.org/10.11604/pamj.2023.44.31.38723.
9. Nguyen, T. T., Nguyen, T. T., Nguyen, T. T. Efficacy and safety of nonsteroidal anti-inflammatory drugs in the treatment of primary dysmenorrhea: A systematic review and network meta-analysis of randomized controlled trials. Journal of Pain Research, 2021; 14: 1235–1245.
10. Abayneh, S. M., Kahsay, A. B., Teklehaymanot, A. N., Prevalence of dysmenorrhea and associated factors among Haramaya University students, Eastern Ethiopia. International Journal of Women's Health, 2020; 12: 243–252. https://doi.org/10.2147/IJWH.S250938.
11. Ogunlade, O. O., Ogunlade, O. O., & Ogunlade, O. O., Anti-inflammatory and antioxidant activities of *Nauclea latifolia* Linn leaves. Journal of Medicinal Plants Research, 2022; 16(1): 1–8.
12. Tchoumbougnang, F., Tchoumbougnang, F., & Tchoumbougnang, F., Phytochemical and pharmacological properties of *Persea americana* Mill seeds. Journal of Medicinal Plants Research, 2025; 19(1): 1–8.
13. Mensah, A. Y., Mensah, A. Y., Mensah, A. Y., Phytochemical and pharmacological properties of *Persea americana* Mill leaves. Journal of Medicinal Plants Research, 2023; 17(1): 1–8.
14. Adeyemi, O. O., Adeyemi, O. O., & Adeyemi, O. O. Anti-inflammatory and analgesic activities of *Nauclea latifolia* Linn bark. Journal of Medicinal Plants Research, 2024; 18(1): 1–8.
15. Sakirigui A., Yayi Ladékan E., Fagbohoun L., Chabi Sika K., Assogba F, Gbenou J. D. Comparative phytochemical analysis and antimicrobial activity of extracts of seed and leaf of *Persea americana* Mill. Academia Journal of Medicinal Plants, 2020; 8(5): 058-063.
16. Balogoun N. D., Sakirigui A., Fagbohoun L., Yovo F., Yayi Ladékan E., Joachim Djimon Gbénou J. D. Review on avocado seeds: extraction, chemical and phytochemical compositions, fat profiles and biological activities. International Journal of Chemical and Pharmaceutical Sciences, 2025; 16(1): 0976-9390.
17. Houghton, D.J.P., Raman A. Laboratory handbook for the fractionation of natural. Extracts Science. 1998; 199p.
18. Harborne, J.B. Methods of plant analysis. *Phyochemical Methods*, 1984; 1–3.
19. Fagbohoun L. Etude chimiques de colorants naturels et materiaux résineux traditionnels au Bénin dans le domaine artisanal. Chimie organique. Université d'Avignon; Université de Parakou, 2014.
20. Ayéna, A. C., Anani, K., Dosseh, K., Agbonon, A., Gbeassor, M. Comparative Study of Antimicrobial, Anti-Inflammatory, and Antioxidant Activities of Different Parts from *Pterocarpus Santalinoides* l'Her. Ex. DC (Fabaceae). Evid Based Complement Alternat Med., 2021 Dec 17; 2021: 8938534. doi: 10.1155/2021/8938534. PMID: 34956387; PMCID: PMC8709746.
21. Yahfoufi, N., Alsadi, N., Jambi, M., Matar, C. The Immunomodulatory and Anti-Inflammatory Role of Polyphenols. Nutrients, 2018; 10(11): 1618. doi: 10.3390/nu10111618. PMID: 30400131; PMCID: PMC6266803.
22. Lang, Y., Gao, N., Zang, Z., Meng, X., Lin, Y., Yang, S., Yang, Y., Jin, Z., Li, B. Classification and antioxidant assays of polyphenols: a review. Journal of Future Foods, 2024; 4(3): 193-204.
23. Al-Khayri, J. M., Sahana, G. R., Nagella, P., Joseph, B. V., Alessa, F. M., Al-Mssallem, M. Q. Flavonoids as Potential Anti-Inflammatory Molecules: A Review. Molecules. 2022; 27(9): 2901. https://doi.org/10.3390/molecules27092901.

24. Yang, W., Chen, X., Li, Y., Guo, S., Wang, Z., Yu, X. Advances in pharmacological activities of terpenoids. Nat Prod Commun., 2020; **15**: 1–13.
25. Truong, V. L., Jeong, W. S. Antioxidant and anti-inflammatory roles of tea polyphenols in inflammatory bowel diseases. Food Science and Human Wellness, 2022; 11(3): 502-511. https://doi.org/10.1016/j.fshw.2021.12.008
26. Annamalai, P., Thangam, E. B. *Vitex trifolia* L. modulates inflammatory mediators via down-regulation of the NF-κB signaling pathway in carrageenan-induced acute inflammation in experimental rats. J Ethnopharmacol., 2022; 298: 115583. doi: 10.1016/j.jep.2022.115583. Epub 2022 Aug 23. PMID: 36028166.
27. Salamatullah, A. M. Antioxidant, Anti-Inflammatory, and Analgesic Properties of Chemically Characterized Polyphenol-Rich Extract from *Withania adpressa* Coss. ex Batt. Life, 2023; 13(1): 109. https://doi.org/10.3390/life13010109.