Trop J Nat Prod Res, July 2025; 9(7): 3267 - 3270 **ISSN 2616-0684 (Print)**
ISSN 2616-0692 (Electronic)

Tropical Journal of Natural Product Research

Available online at https://www.tjnpr.org

Original Research Article

Activity of Three Nigerian Mistletoes against *Culex quinquefasciatus* Larvae

Funmilayo G. Famuyiwa*, Maryam O. Oredola

Department of Pharmacognosy, Faculty of Pharmacy, Obafemi Awolowo University, Ile-Ife, Nigeria

ARTICLE INFO

Article history:
Received 20 May 2024
Revised 14 May 2025
Accepted 17 June 2025
Published online 01 August 2025

ABSTRACT

The development of resistance to the available larvicidal agents by mosquitoes has necessitated continued search for larvicidal compounds from plants. Therefore, in this study, the larvicidal activity of the extracts and fractions of flowers, leaves, and stems of *Tapinanthus bangwensis*, *Phragmanthera incana*, and *Tapinanthus globiferus* were investigated against *C. quinquefasciatus*. The leaf, flower, and stem of *T. bangwensis*, *P. incana*, and *T. globiferus* were collected, authenticated, air-dried, powdered, and extracted with methanol for 72 hours. The extracts were filtered and concentrated *in vacuo* and tested against *C. quinquefasciatus* fourth *instar* larvae. The most active extract of each plant was successively partitioned into *n*-hexane and ethyl acetate to obtain their corresponding fractions which were similarly tested. The methanol extract of *Nicotiana tabacum*, a known insecticidal plant, was used as the positive control. After 48 hours of exposing the test organism to the extracts, the leaf of *T. bangwensis* (LC_{50} 3.68 mg/mL), the flowers of *T. globiferus* (LC_{50} 7.55 mg/mL) and *P. incana* (LC_{50} 4.00 mg/mL) were the most active. After partitioning, the *n*-hexane fraction of *T. bangwensis* (LC_{50} 1.28 mg/mL), ethyl acetate fraction of *P. incana* (LC_{50} 1.91 mg/mL), and the aqueous fraction of *T. globiferus* (LC_{50} 2.24 mg/mL) had the highest activity for each extract. This is the first report of the larvicidal activity of these plants against *C. quinquefasciatus*.

Keywords: larvicidal activity, ethyl acetate, *n*-hexane, *Tapinanthus bangwensis*, *Phragmanthera incana*, *Tapinanthus globiferus*

Introduction

Lymphatic filariasis (LF) is usually transmitted to humans in childhood when bitten by *Culex quinquefasciatus* and certain *Anopheles* species of mosquitoes infected with the roundworms, *Wuchereria bancrofti* of the family Filariodidea. The infection damages the lymphatic system and kidney while disrupting the body's immune response. Additionally, it can result in abnormal swelling of the arms and legs, leading to pain, significant disability, and social stigma. Mosquito control has been supported by WHO as a supplementary strategy to reduce transmission of LF and other mosquito-borne infections.[1] Controlling mosquitoes at the larva stage has been reported to be efficient as they are relatively immobile and a large population can be killed in their breeding sites with little effort. However, the development of resistance to currently available synthetic larvicides, especially in the tropics, and environmental concerns have stimulated continued efforts to develop plant-based larvicides.[2] In this study, three mistletoes were screened for larvicidal activity against *C. quinquefasciatus*. The Nigerian mistletoes (Loranthaceae) are hemi-parasitic plants found growing on tree crops of high medicinal and economic importance including, *Hevea brasiliensis*, *Azadirachta indica*, Citrus spp, *Theobroma cacao*, *Spondias mombin*, *Vitellaria paradoxa*, *Morinda lucida*, *Rauwolfia vomitoria*, *Cola nitida*, *Ficus exasperata*, etc.[3-10]

*Corresponding author. Email: samidan@ouaife.edu.ng
Tel: +234 8137701974

Citation: Famuyiwa FG, Oredola MO. Activity of three Nigerian mistletoes against *Culex quinquefasciatus* larvae. Trop J Nat Prod Res. 2025; 9(7): 3267 – 3270 https://doi.org/10.26538/tjnpr/v9i7.54

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria

Most farmers in Western part of Africa see mistletoes as notorious and devastating parasites that depletes the economic values and compete with tree crops for nutrients. They are seen by farmers or gardeners as destructive plants that cause significant harm to fruit trees and plants with medicinal properties and economic values.[11-12] Ethnomedicinally, they have been used as therapeutic remedy for mental conditions, urinogenital diseases, sterility, rheumatism, pain, tumors, fever, hypertension, syphilis, fracture, diabetes, etc.[13-21] Pharmacological activities like hypotensive, antimicrobial, anti-oxidative, hypoglycaemic and anti-inflammatory have been reported for them.[22] However, a literature search showed a dearth of information on the activity of *Tapinanthus bangwensis*, *Phragmanthera incana*, and *Tapinanthus globiferus* against mosquito larvae, hence this study.

Materials and Methods

Preparation of Extracts and Fractions

The leaf, stem, and flower of *T. bangwensis*, *P. incana*, and *T. globiferus* growing on *Theobroma cacao*, *Leucaena leucocephala*, and *Theobroma cacao* respectively were collected at various locations in Ile-Ife, Osun state, Nigeria on 10th March 2022. They were all authenticated and voucher specimens deposited at the Herbarium of the Department of Pharmacognosy, Faculty of Pharmacy, Obafemi Awolowo University, Ile-Ife, under reference numbers FPI 2469, FPI 2471 and FPI 2470 respectively. They were separately air-dried, and blended to powder using a grinding machine. The powdered samples of each plant part were separately weighed and macerated in methanol for 72 hours. Each methanol solution was filtered and the solvent was evaporated under vacuum at 35°C. This process was repeated three times, and the combined extracts were stored for subsequent larvicidal activity testing against *C. quinquefasciatus* larvae. The most potent extract from each plant was suspended in water, then successively partitioned with *n*-hexane and ethyl acetate, and concentrated with the rotary evaporator to afford their corresponding fractions coded TBLa, TBLb, and TBLc for *T. bangwensis*, PIFa, PIFb and PIFc for *P. incana* and TGLa, TGLb and TGLc for *T. globiferus*. Each fraction was similarly tested for activity against the test organism.

 ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

Larvicidal Assay

The extracts and partitioned fractions were subjected to larvicidal assay by the World Health Organization's (2005) recommendations.[23] The extracts were first solubilized in Tween 80 at a concentration of 0.2 % (v/v) and there were five replicates for each concentration. A 10 mg/mL and 5 mg/mL stock solution were prepared for the extracts and partitioned fractions respectively. The concentration range used for the extracts was 1.0, 2.0, 3.0, 4.0, and 5.0 mg/mL while that for the fractions was 0.5, 1.0, 1.5, 2.0, 2.5 mg/mL. The negative control contains distilled water and Tween 80. *Nicotiana tabacum* leaf extract was used as the positive control.

Statistical Analysis

The ANOVA (One-way analysis of variance) and Student Newmann Keul's post hoc test were used to perform the statistical analysis of the data obtained and values of $p < 0.05$ were deemed statistically significant.

Results and Discussion

Culex quinquefasciatus mosquito is the vector for various zoonotic diseases affecting both humans and animals, including lymphatic filariasis, avian malaria, western equine encephalitis, St. Louis encephalitis, and West Nile fever. Control of this mosquito has been identified as an effective supplementary means of controlling/preventing the diseases it transmits. For example, control of this mosquito played a crucial role in eliminating lymphatic filariasis, even in the absence of widespread preventive chemotherapy.[24-25] The use of synthetics to eliminate or reduce mosquito populations has resulted in many environmental and health issues. However, the use of compounds of plant origin are effective alternative as they are non-toxic and biodegradable with broad-spectrum and target-specific activities against different mosquito species.[26] In this study, based on the fact that activity varies due to the morphological part used, three morphological parts of *T. bangwensis*, *P. incana*, and *T. globiferus* were investigated for activity against *C. quinquefasciatus* larvae. This will enable the identification of the part containing the active constituent(s) qualitatively or quantitatively.[27]

Activity of the Methanol Extracts

The methanol extracts of these plant demonstrated different levels of activity against the test organism. After 24 hours of exposure, *T. bangwensis* leaf extract (TBL) had moderate activity (LC_{50}, LC_{90} 4.17±0.23, 6.28±0.54 mg/mL) and was the most active. The activities of the flower (TBF) and stem (TBS) were weak with LC_{50} greater than 4.20 mg/mL (Figure 1). At 48 hours, TBL maintained the highest activity while TBF and TBS were still weak-acting (Figure 2). The moderate activity (LC_{50} of 2.66 mg/mL) of the cyclohexane extract of the stem bark had been reported against *Sitophilus zeamais*.[28-29] The activity of TBL is comparable to the methanol extracts of *Ficus exasperata*, *Ficus vogelli*, and *Thevetia neriifolia* bark.[2] For *P. incana*, after 24 hours, extracts from all morphological parts displayed weak larvicidal activity. The flower extract (PIF) exhibited the lowest lethality, with LC_{50} and LC_{90} values of 8.99 ± 0.15 mg/mL and 14.96 ± 0.23 mg/mL, respectively. However, at 48 hours there was an improvement in their activities, with the leaf (PIL) and PIF becoming comparatively more active than the stem (PIS) with LC_{50} values 4.19 ± 0.17 mg/mL and 4.00 ± 0.42 mg/mL respectively (Figure 2). Similar to *P. incana*, the extract of all the morphological parts of *T. globiferus* displayed weak activity at 24 hours (LC_{50} 9.83 – 12.78 mg/mL). After 48 hours of exposure, there was a slight increase in activity (LC_{50} 7.55 – 9.10 mg/mL) with the leaf (TGL) and flower (TGF) extracts being comparable (Figure 2). However, TGL was partitioned due to the low weight of TGF extract. Generally, the activities of the extracts improved with a longer time of contact with the test organism. Although the activities of most of the extracts were better than that of a purified lectin from *Agaricus semotus* with LC_{50} 13.50 mg/mL,[30] throughout the test period, none of the extracts of the three mistletoes had comparable activity with *N. tabacum* extract used as the positive control (Figures 1 and 2).

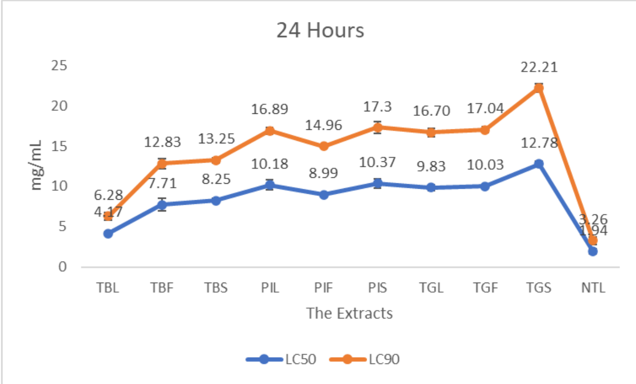

Figure 1: The Larvicidal Activities of the Morphological Parts of *T. bangwensis*, *P. incana* and *T. globiferus* at 24 hours. **KEY:** TBL: *T. bangwensis* leaf; TBF: *T. bangwensis* flower; TBS: *T. bangwensis* stem; PIL: *P. incana* leaf; PIF: *P. incana* flower; PIS: *P. incana* stem, TGL: *T. globiferus* leaf; TGF: *T. globiferus* flower; TGS: *T. globiferus* stem; NTL: *N. tabacum* leaf extract

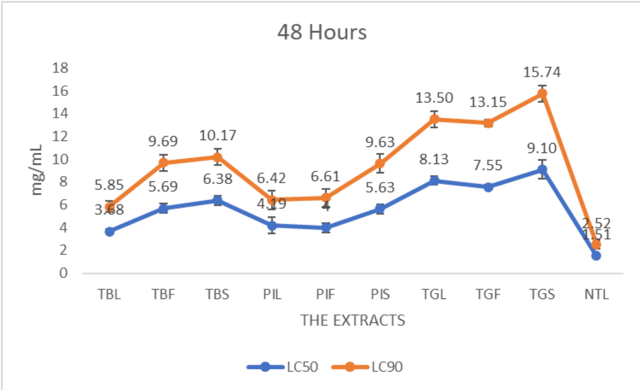

Figure 2: The Larvicidal Activities of the Morphological Parts of *T. bangwensis*, *P. incana* and *T. globiferus* at 48 hours.
KEY: TBL: *T. bangwensis* leaf; TBF: *T. bangwensis* flower; TBS: *T. bangwensis* stem; PIL: *P. incana* leaf; PIF: *P. incana* flower; PIS: *P. incana* stem, TGL: *T. globiferus* leaf; TGF: *T. globiferus* flower; TGS: *T. globiferus* stem; NTL: *N. tabacum* leaf extract

Activity of the Partitioned Fractions

Each plant's most active morphological part was suspended in water and its chemical constituents were separated into *n*-hexane and ethyl acetate using a liquid-liquid partitioning procedure. Larvae of *C. quinquefasciatus* were used to test the fractions. The *T. bangwensis* ethyl acetate fraction (TBLb) showed the maximum activity after 24 hours, with lower fatality values than the positive control. This is followed by *n*-hexane (TBLa) while aqueous fraction (TBLc) was the least active. By 48 hours, the activities of TBLa improved and became comparable ($p<0.05$) to TBLb and the positive control (Figure 3).
Partitioning of *P. incana* flower extract (PIF) gave *n*-hexane fraction (PIFa), ethyl acetate fraction (PIFb), and aqueous fraction (PIFc). Twenty-four hours after exposure of *C. quinquefasciatus* larvae to these fractions, all the fractions gave moderate activities.[25] This was a great improvement over that of PIF.[2] On further exposure, PIFb became highly active (LC_{50} 1.91 ± 0.50 mg/mL) and was the most active (Figure 4). The resulting fractions *n*-hexane (TGLa), ethyl acetate (TGLb), and aqueous (TGLc) of *T. globiferus* leaf extract (TGL) gave higher activities than the mother extract throughout the test period. TGLa and TGLb had comparably moderate activities while TGLc was highly active with LC_{50} 2.24 ± 0.24, 3.91 ± 0.20 mg/mL at 48 hours (Figure 5).

 ISSN 2616-0684 (Print)
ISSN 2616-0692 (Electronic)

This observation deviates from previous trend because lipophilic chemicals have a greater capacity to penetrate the cytoplasmic membrane and insect cell wall, making them generally more effective insecticides.[31] Furthermore, although it is rare, significant larvicidal activity in the aqueous fraction is not unheard of. For example, it has been observed that the aqueous fraction of *Ficus sur* leaf exhibits significant activity, with LC_{50} and LC_{90} values of 2.89 ± 0.21 mg/mL and 1.52 ± 0.11 mg/mL, respectively.[2]

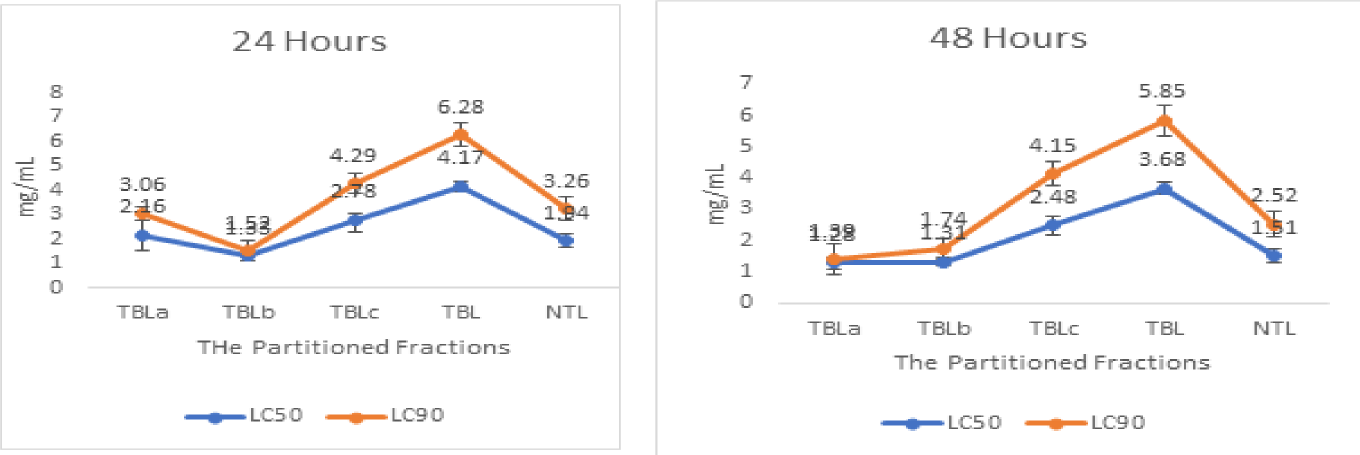

Figure 3: The Larvicidal Activities of Partitioned fractions of *T. bangwensis* Leaf at 24 and 48 hours. **KEY:** TBL_a: *n*-hexane fraction; TBL_b: ethylacetate fraction; TBL_c: aqueous fraction; TBL: methanol extract; NTL: *N. tabacum* leaf extract

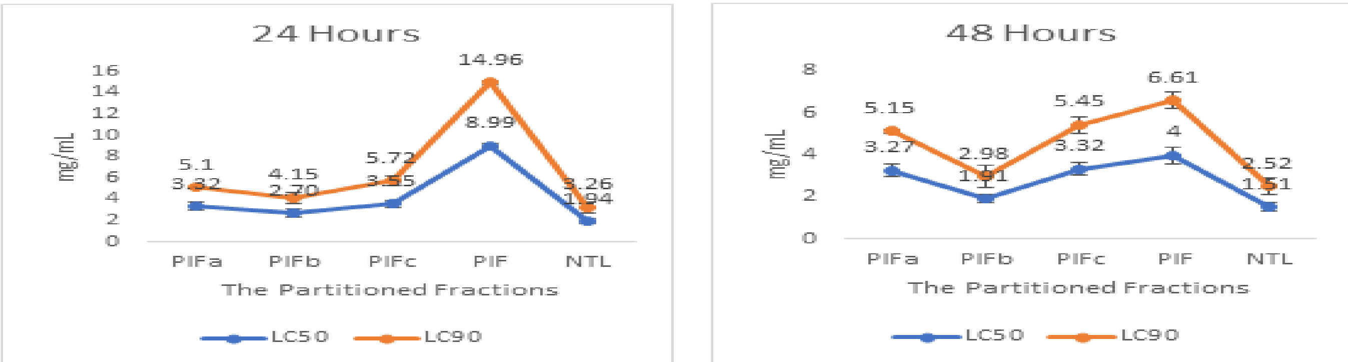

Figure 4: The Larvicidal Activities of Partitioned fractions of *P. incana* Flower at 24 and 48 hours. **KEY:** PIF_a: *n*-hexane fraction; PIF_b: ethylacetate fraction; PIF_c: aqueous fraction; PIF: methanol extract; NTL: *N. tabacum* leaf extract

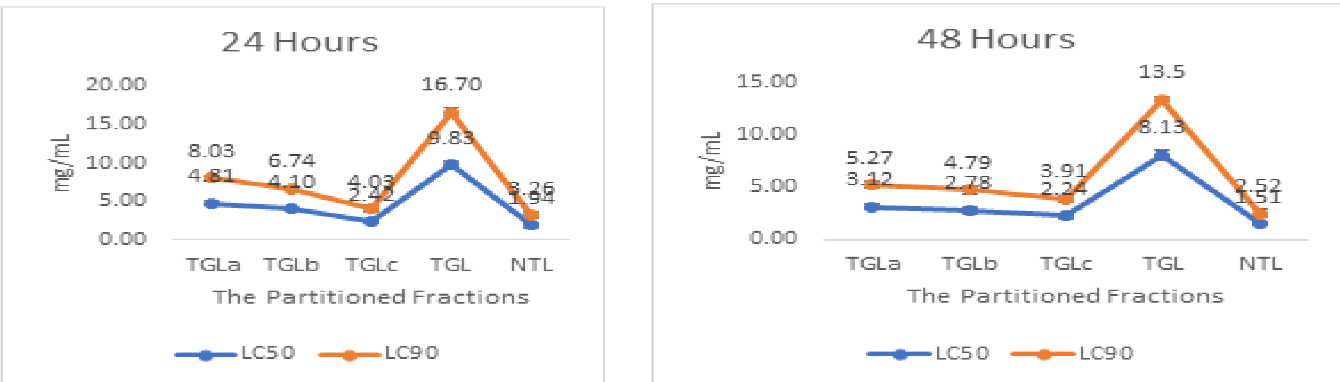

Figure 5: The Larvicidal Activities of Partitioned fractions of *T. globiferus* Leaf at 24 and 48 hours. **KEY:** TGL_a: *n*-hexane fraction; TGL_b: ethylacetate fraction; TGL_c: aqueous fraction; TGL: methanol extract; NTL: *N. tabacum* leaf extract

Conclusion

The methanol extracts of *T. bangwensis* leaf, *T. globiferus*, and *P. incana* flowers have larvicidal potential. Generally, after partitioning, their resulting fractions are more active than the methanol extracts. The *n*-hexane fraction of *T. bangwensis*, the ethyl acetate fraction of *P. incana*, and the aqueous fraction of *T. globiferus* demonstrated the highest activity. Efforts are ongoing to isolate the active compounds of these active fractions.

Conflict of Interest
Authors declare no conflict of interest.

Authors' Declaration
The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

Acknowledgement
The authors appreciate Mr. I.I. Ogunlowo for his assistance during plant collection.

References

1. World Health Organization. Global programme to eliminate lymphatic filariasis. WHO Weekly Epidemiological report 2020. 81:221-232. (Accessed May 22, 2024).
2. Famuyiwa FG, Adewoyin FB, Oladiran OJ, Obagbemi OR. Larvicidal Activity of Some Plants Extracts and Their Partitioned Fractions against *Culex quinquefasciatus.* Int J.Trop Dis Health. 2020; 41(11): 23-34. DOI: 10.9734/IJTDH/2020/v41i1130332
3. Polhill R, Wiens D. Mistletoe of Africa. The Royal Botanic Garden, Kew, U. K. 1998. 370.
4. Bright EO, Okusanya BA. Infestation of economic plants in Badeggi by *Tapinanthus dodoneifolius* (DC) Danser and *Tapinanthus globiferus* (A. Rich) Van Tiegh. Nig J. Weed Sci. 1998; 11: 51-56.
5. Overfield D, Riches C, Samoah AM, Sarkodie O, Baah F. A farming system analysis of the mistletoe problem in Ghanian Cocoa. Cocoa Grower's Bulletin. 1998; 51: 42–50.
6. Gill LS, Onyibe HI. Mistletoes on rubber trees in Nigeria. Haustorium. 1990; 23: 1-2.
7. Begho ER, Omokhafe KO, Omo-Ikerodah EE, Akpaja EO. Some observations on the fruit set and incidence of Mistletoes on rubber trees in Nigeria. Am-Eurasian J Sustain Agric. 2007; 1(1): 13-18. https://www.researchgate.net/publication/291962010.
8. Simeon KA, Illoh HC, Imoh IJ, Imoh EJ. African Mistletoes (Loranthaceae); Ethnopharmacology, Chemistry and Medicinal Values: An Update. Afr J Tradit Complement Altern Med. 2013; 10(4): 161-170. doi: 10.4314/ajtcam.v10i4.26
9. Anka SA, Aliero AA. Preliminary Survey of African Mistletoe (Loranthaceae) on Host Trees Species in Sokoto Metropolis, North Western Nigeria. SJBA. 2019: 1(1): 46-50.
10. Tizhe TD, Alonge SO, Aliyu RE. Mistletoe presence on five tree species of Samaru area, Niger. Afri J. Plant Sci. 2016; 10(1); 16-22. DOI: 10.5897/AJPS2015.1335
11. Zewdie K, Eshetu T. Preliminary observation on feasibility of mechanical control of parasitic plants on trees. Proceedings of the 7th Annual Conference of the Ethiopian Weed Science Committee, Rezene Fesehale, Ethiopia, 1993; 41-42.
12. Jiofack RT, Dondjang JP, Nkongmeneck BA. The Loranthaceae of the Bafou area in Cameroon; identification, distribution, biology and eradication strategies. In: Burgt X van der, Maesen J van der, Onana JM (Eds). Systematics and Conservation of African Plants; Proceedings of the 18th AETFAT Congress, Yaounde, Cameroon; 2007. 229-235.
13. Adodo A, Nature PA. Christian Approach to Herbal Medicine. (3rd Ed.). 2004. Nigeria. Benedictine Publication 2004. 103-111.
14. Jadhav N, Patil CR, Chaudhari KB, Wagh JP, Surana SJ, Jadhav RB. Diuretic and natriuretic activity of two mistletoes species in rats. Phcog Research. 2010; 2(1): 50-57. doi: 10.4103/0974-8490.60576
15. Noumi E, Eloumou MER. Syphilis ailment; Prevalence and herbal remedies in Ebolowa subdivision (South region, Cameroon). Int J Pharm Biomed Sci. 2011; 2(1): 20–28.
16. Ken'ichi M, Kazuhiro N, Norimasa N, Dawa D, Laximi T, Atsushi W, Fumi M, Toshiro B, Gaku M. Report of investigation for Wild Edible Plants and their Traditional Knowledge in Bhutan. J Faculty Agric Shinshu University. 2006; 42(1–2): 37-47.
17. Sher H, Alyemeni MN. Pharmaceutically important plants used in traditional system of Arab medicine for the treatment of livestock ailments in the kingdom of Saudi Arabia. Afr. J. Biotechnol. 2011; 10(45): 9153–9159. doi: 10.5897/AJB10.1570
18. Yineger H, Yewhalaw D. Traditional medicinal plant knowledge and use by local healers in Sekoru District, Jimma Zone, Southwestern Ethiopia. J Ethnobiol. Ethnomed. 2007; 3: 24. doi:10.1186/1746-4269-3-24
19. Matthes H, Schad F, Buchward D, Schenk G. Endoscopic ultrasound-guided fine needle injection of *V. album* L. (mistletoe; *Helixor M*) in the therapy of primary inoperable pancreas cancer; a pilot study. Gastroenterol. 2005; 128(4 Suppl. 2): 433.
20. Urech K, Scher JM, Hostanska K. Apoptosis inducing activity of viscin, a lipophilic extract from V. album. L. J Pharm Pharmacol. 2005; 57: 101–109. doi: 10.1211/0022357055083
21. Kienle GS, Kiene H. Review article: Influence of *V. album* L. (European mistletoe) extracts on quality of life in cancer patients; a systematic review of controlled clinical studies. Integr Cancer Ther. 2010; 9(2): 142-157. doi: 10.1177/1534735410369673
22. Adesina SK, Iloh HC, Johnny II, Jacobs IE. African Mistletoes (Loranthaceae); Ethnopharmacology, Chemistry and Medicinal Values: An Update. African J. Tradit. Complmentary Altern Med. 2013; 10(4): 161-170. doi: 10.4314/ajtcam.v10i4.26
23. Guidelines for Laboratory and Field Testing of Mosquito Larvicides. Available:http://whqlibdoc.who.int/hq/2005/WHO_CDS-WHOES-GCD2005.13.df?ua=1
24. World Health Organisation Fact Sheet on Filariasis 2023. Available from: https://www.who.int/health-topics/lymphatic-filariasis#tab=tab_1
25. Centre for Disease Control and Prevention Fact Sheet on Filariasis 2020. Available from: https://www.cdc.gov/parasites/lymphaticfilariasis/index.html
26. Ghosh A, Chowdhury N, Chandra G. Plant extracts as potential mosquito larvicides. Indian J Med Res. 2012; 135(5): 581–598.
27. Sukumar K, Perich MJ, Boobar LR. Botanical derivatives in mosquito control: a review. J Am Mosq Control Assoc. 1991; 7(2): 210-237.
28. Adebajo CA, Famuyiwa FG, Aliyu FA. Properties for sourcing Nigerian larvicidal plants. Molecules. 2014; 19: 8363-8372; doi:10.3390/molecules19068363
29. Diouf EHG, Diop EG, Sène M, Samb, A. Phytochemical Screening and Insecticidal Activity of Three Extracts of *Tapinanthus bangwensis* (Engl. & Krause) on *Sitophilus zeamais* (Maize Weevil). Open Access Lib. J. 2016; 3: 2962. http://dx.doi.org/10.4236/oalib.1102962
30. Adedoyin, I. O., Adewole, T.S., Agunbiade, T.O, Kuku A. and Adewoyin FB. (2021). A purified lectin with larvicidal activity from a woodland mushroom, *Agaricus semotus*. Acta Biol Szeged, 65(1):65-73. DOI: 10.14232/abs.2021.1.65-73
31. Famuyiwa F.G, Famuyiwa SO, Aladesanmi AJ. (2018). Activity of the compounds isolated from *Blighia sapida* (Sapindaceae) stem bark against *Aedes aegypti* larvae. IJS. 20(3): 601-605.