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FORMULATION OF MOSQUITO REPELLANT MOISTURISING CREAM USING THE OIL EXTRACT OF *AZADIRACHA INDICA* (NEEM) SEED



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ABSTRACT

The study aimed at formulating mosquito repellent moisturizing cream using the oil extract of *Azadirachta indica* seed, started with extracting the *A. indica* seed oil. The oil obtained was purified using micro column packed with silica gel of 60-120 mesh size. The purified seed oils was characterized using Gas Chromatography-Mass Spectrometry (GC-MS), which gave the indication of bioactive constituents like; oleic acid, hexadecanoic acid, octadecanoic acid etc. The efficacy the formulated of cream from the *A. indica* seed oil using 5% and 10% w/w as active ingredient were tested on adult mosquitoes and found to be effective.

1. Introduction

Synthetic Insecticides produced to kill mosquitoes in order to prevent these damages, also tend to have adverse effects on humans in various ways, most especially those produced from synthetic materials (Gou et al., 2020). These adverse effects in the short run include; headache, dizziness, catarrh etc. Maia and Moore (2011) reported that the use of plant materials as repellants have been exploited for thousands of years by man, mostly done by hanging bruised plants in houses, a practice that is still in wide used throughout the developing countries. The *A. Indica* is a well-known medicinal tree originating from India. It is regarded as a gift to nature, owing to its numerous medicinal properties. Most indigenous communities in Nigeria have been using the crude seed oil extracts for repelling mosquitoes and other pests. This work is aimed at using the *A. Indica* seed oil to formulate moisturizing cream to be used as mosquito repellent.

2. Materials and Methods

2.1 Collection of Plant Sample

Fresh *A. indica* fruits were collected in September 2019 from trees grown at Langtang North L.G.A Plateau state, Nigeria.

2.2. Preparation of sample and extraction

After obtaining the samples of the *A. indica* seed kernels, the oil was extracted and purified using the method described (Gushit et al., 2013).

2.3 GC-MS analysis

Gas chromatography-mass spectrometry (GC-MS) was used almost exclusively for the qualitative analysis of the oil constituents (Wang et al., 2018).

2.4 Preparation of Repellent Cream for Evaluation

An existing formulation of cream (Mohiuddin, 2019) was modified as used incorporating varying concentrations of the *A. Indica* seed oil in moisturizing cream indicated in Table 1. These concentrations include the following: 0% w/w(F0), 5% w/w(F1), 10% w/w(F2), 5% w/w + commercial repellent (F3) and Commercial repellent (F4).



Table 1: Different Formulations of moisturizing cream

Composition	F0	F1	F2	Function/Uses
<i>A. Indica</i> Seed oil	0%	5.0%	10.0%	Extract
Cetyl Alcohol	2.0%	2.0%	2.0%	An emulsion stabilizer, opacifying agent
Petroluem jelly	1.5%	1.5%	5.0%	Skin protectant.
Triethalonamie	1.0%	1.0%	1.0%	pH adjuster, alkalizing & emulsifying agent
Emmy wax	1.0%	1.0%	1.0%	Emulsifies water and oil interface
Paraffin Oil	1.5%	1.5%	1.5%	Improves skin softness & barrier function
Lanolin	1.0%	1.0%	1.0%	Emulsifying agent
Glycerin	2.0%	2.0%	2.0%	Humectant/Emollient
Propylene glycol	1.5%	1.5%	1.5%	Humectants and moisturizer
Carbopol	1.0%	1.0%	1.0%	Thickens/ensures consistency of products
Methyl Paraben	q.s	q.s	q.s	Preservative
Perfume	q.s	q.s	q.s	Gives the body a pleasant scent
Distilled Water	q.s100	q.s100	q.s100	Diluent and vehicle
	%	%	%	

2.5 Collection of mosquito larvae and Adult/Cage Assay

Iman et al. (2014) and WHO (1996) reported methods were used for the Assay.

2.6 Repellent Activity

Wannang et al. (2015) reported methods was modified and used for the repellency test.

2.7 Formulation properties, Evaluation of formulated Cream, storage stability test, Cold Emulsion stability test.

The methods reported by Waqas et al.(2010) and Purkait et al. (2019) were adapted.

2.8 pH of the Cream Formulation

The pH of the formulated samples (1% aqueous solution) was measured at 27°C using a calibrated pH meter (Model H198103).

2.9 Spreadability test

The spreadability was evaluated by reported methods (Shankar et al., 2016).

2.10 Irritancy test

The formulated cream was applied to the dorsal left hand surface. Irritancy, erythema, edema was checked up every hour for 24 hours and reported.

3.Results

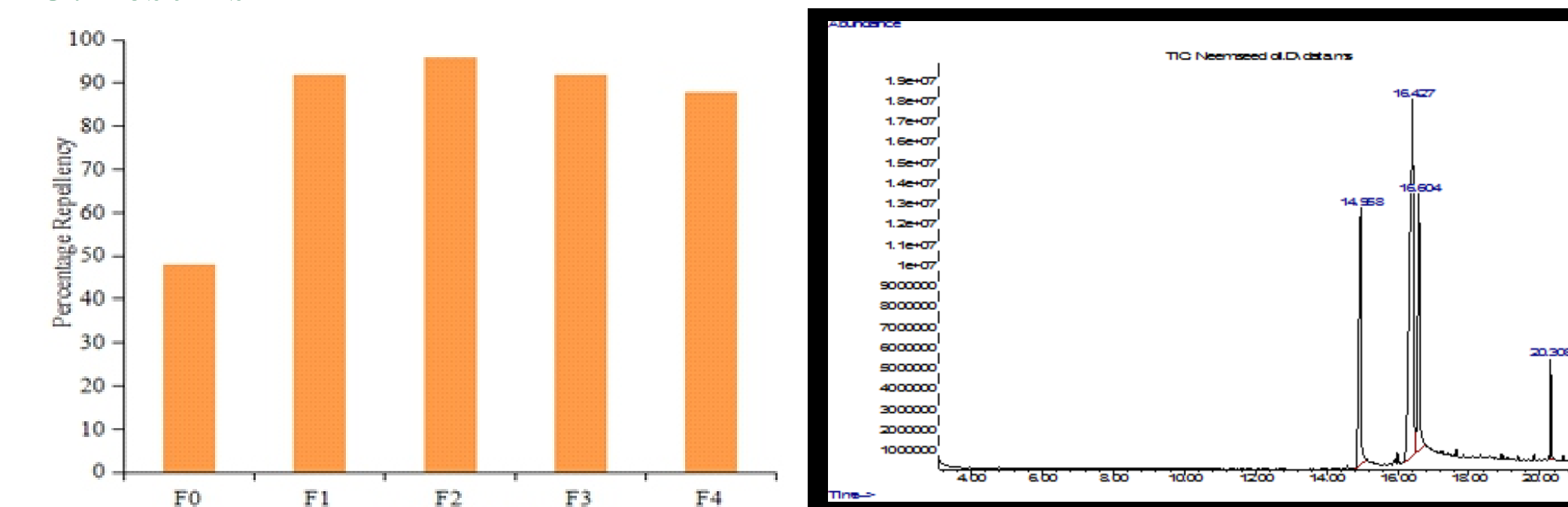


Figure 1: Percentage Repellency from the Arm-in-cage test

Figure 2: GC Chromatogram of *A. indica* seed oil

Table 2 GCMS of the Fatty Acid composition of the *A. indica* seed oil

Fatty Acid	Chemical Formular	m/z	RT
Hexadecanoic acid		43(BT), 60, 73, 129, (256)	14.96
Oleic acid		41(BT),55,29,69,83,(280)	16.43
Octadecanoic acid		43(BT), 3,60,129,69,(284)	16.60
Squalene		69(BT),81,41,95,121,(410)	20.31

Table 3: Arm-in-cage Assay on Number of bites between minutes by the mosquitoes Treatment using the different cream formulation

Formulations	Number of mosquitoes exposed	Repellency time interval (mins) on treated arm				% protection
		0-5	35-40	70-85	115-120	
F0	25	4	3	4	2	48
F1	25	0	0	2	0	92
F2	25	0	0	0	1	96
F3	25	0	0	0	2	92
F4	25	0	0	1	2	88

4. Discussion

Seed oils (5% and 10%) from the plant *A. indica*, were formulated into a mosquito repellent cream (Table 1). This was achieved by modifying the formulation by Mohiuddin (2019). The high protection rate of 92–96% exhibited by *A. Indica* oil in the formulated moisturizing cream in this study is an indication that *A. Indica* oil in comparison to some commercial synthetic repellent creams (*Odemos*) could be an alternative repellent ingredients for formulating environmentally friendly mosquito repellent creams (Figure 1). The presence of bioactive substances like oleic acid, octadecanoic acid and hexadecanoic acid were detected in this work (Figure 2 & Table 2). This relate to a study by Gurunathan et al., (2016), in which Oleic acid was isolated, and when tested against mosquitoes it was found to have insecticidal properties showing good larvicidal activities against *Aedes* and *Culex* mosquitoes. Anane-Adjei (2015) suggested that oleic acid smell also may indicate danger to living insects, prompting them to avoid places where the smell comes from. Similarly, squalene which is detected in the seed oil is a terpenoid, it is a natural organic compound, a unique triterpene compound that is one of the main components of the skin surface lipids (Passi et al., 2002). It widely known as insect repellants. The presence of these compounds in the *A. Indica* oil gives it the repellent properties with effects reported in Tables 3. Similarly the creams were evaluated and found to be stable

5. Conclusion

Azadirachta indica seed oil cream has proven to be very valuable as insect repellent particularly at higher concentration of 10.0%w/w. This will be a good alternative to synthetic repellants that has been associated with certain skin reactions. This study reaffirms the possibility of using indigenous Nigerian plants with insecticidal property for the control of Mosquitoes and other insect pests.

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REFERENCES

- Anane-Adjei, B., 2015. Extraction of Oils from Azadirachta Indica Seeds using Different Techniques and the Effects of these Techniques on the Acid, Saponification and Peroxide Values of the Oils. *Researchgate*. 280022234. Gou, Y., Li, Z., Fan, R., Guo, C., Wang, L., Sun, H., Li, J., Zhou, C., Wang, C., & Wang, C., 2020. Ethnobotanical survey and evaluation of traditional mosquito repellent plants of Dai people in Xishuangbanna, Yunnan Province, China. *J. Ethnopharmacol.*, 262, 1-20. <https://doi.org/10.1016/j.jep.2020.113124> Gurunathan, A., Senguttuvan, J. & Paulsamy S., 2016. Evaluation of Mosquito Repellent Activity of Isolated Oleic Acid, Eicosyl Ester From Thalictum Javanicum. *Indian J. Pharm. Sci.* 78(1), 103-110. Gushit, J. S., E. O. Ekanem, O. J. Abayeh, H. M. Adamu, I. Y. Chindo, 2013. Analysis of herbicide residues and organic priority pollutants in selected root and leafy vegetable crops in Plateau State, Nigeria. *World J. Anal. Chem.*, (1)2, 23-28. Iman, H., Nigar, Z., Sofi, G. & Aziz, S., 2014. The basic rules and methods of mosquito rearing (*Aedes aegypti*). *Trop. Parasitol.* 4(1), 53-55. Maia, M.F. & Moore S.J., 2011. Plant-based insect repellents: a review of their efficacy, development and testing. *Malar. J.* 10 (1). <https://malariajournal.biomedcentral.com/articles/10.1186/1475-2875-10-S1-S11>. Mohiuddin, A.K., 2019. Skin Care Creams: Formulation and Use. *Dermatol. Clin. Res.*, 238. 5(1), 238-271. Passi, S., De Pita, O., Puddu, P. & Littarru, G.P.(2002). Lipophilic antioxidant in human sebum and aging. *Free Radic. Res.* 36, 471-477. Purkait A., Biswas S., Saha S., Hazra D.K., Roy K., Biswas P.K., Ghosh X.Z., Kole R.K., 2019. Formulation of plant based insecticides, their bio-efficacy evaluation and chemical characterization. *Crop Prot.* 125, 104907. doi: 10.1016/j.cropro.2019.104907. Shankar, B.S., Saravanan, T., Ragavi M., Kaviya, G., Anushree, A., Samraj, A. & Tennyson, S., 2013. Screening of Local Plants for Their Repellent Activity against Mosquitoes (Diptera: Culicidae). *J. Mosq. Res.* 3(14), 97-104. Wang, Y., Li, X., Jiang, Q., Sun, H., Jiang, J., Chen, S., Guan, Z., Fang, W., Chen, F., 2018. GC-MS analysis of the volatile constituents in the leaves of 14 compositae plants. *Molecules* 23s, 166. <https://doi.org/10.3390/molecules23010166>. Wannang, N.N., Ajayi, V.F., Ior, L. D., Dapar, L.M.P., Okwori, V. A. & Ohemu, T., 2015. Mosquito Repellent Property of *Azadirachta Indica* Extract (Fruit Bark and Seed Kernel). *Sci. Res. J.* 3 (6). Waqas M.K., Akhtar N., Ahmad M., Murtaza G., Khan M.S., Iqbal M., Rasul, A. And Bhatti, N.S., 2010. Formulation and Characterization of A Cream Containing Extract of Fenugreek Seeds. *Acta Pol. Pharm.*, 67 (2), 173-178. World Health Organization [WHO], 1996. Report of the WHO informal consultation on the evaluation on the testing of insecticides. Geneva: World Health Organization [Online] Available from: http://apps.who.int/iris/bitstream/10665/65962/1/CTD_WHOPES_IC_96.1.pdf

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