International Journal of Engineering Inventions
e-ISSN: 2278-7461, p-ISSN: 2319-6491
Volume 14, Issue 2 [February. 2025] PP: 145-148

Repellent Activity of Methanol Extract of *Tagetes erecta* Against Mosquitoes: A Potential Biopesticide

Nagma Mev [1] Varun Jain[1] and Manik Sharma[2]
[1-] *SAM Global University Raisen*
[2-] *Career College Autonomous Bhopal*

Abstract:
Mosquito-borne diseases remain a significant global health concern, necessitating the exploration of alternative and sustainable mosquito control strategies. Synthetic insecticides, while effective, pose environmental and health risks, driving the search for biopesticides. This study investigates the repellent activity of methanol extract of Tagetes erecta (marigold) against mosquitoes. The extract was prepared through maceration, and its chemical composition was assessed. Repellence assays were conducted using human landing catch and arm-in-cage methods to evaluate the efficacy of different concentrations of the extract against selected mosquito species. Results demonstrated significant mosquito repellence by the methanol extract, with the level of protection varying depending on concentration and mosquito species. This research suggests that Tagetes erecta methanol extract possesses promising repellent properties and can be considered as a potential environmentally friendly alternative for mosquito control.

Keywords: Tagetes erecta, *mosquito repellent, methanol extract, biopesticide, vector control, human landing catch, arm-in-cage.*

Date of Submission: 14-02-2025 Date of acceptance: 28-02-2025

I. Introduction:

Mosquitoes are vectors for numerous debilitating and life-threatening diseases like malaria, dengue fever, Zika virus, chikungunya, and West Nile virus, impacting millions of people worldwide, particularly in tropical and subtropical regions (WHO, 2023). Effective mosquito control measures are crucial for disease prevention. Traditionally, synthetic insecticides such as pyrethroids and organophosphates have been widely used. However, their overuse has led to the development of insecticide resistance in mosquito populations (Hemingway *et al.,* 2004) and raised concerns about their impact on non-target organisms and human health (Craddock *et al.,* 1997). This has fueled the urgent need for safer and more sustainable alternatives for mosquito control.

Biopesticides derived from plants offer a promising avenue for mosquito control due to their biodegradability, relatively low toxicity, and potential for local production (Pavela, 2016). Many plants synthesize secondary metabolites that exhibit insecticidal, repellent, or larvicidal properties.

Tagetes erecta L., commonly known as marigold, belongs to the Asteraceae family and is widely cultivated for its ornamental value. It is used in traditional medicine and has demonstrated various biological activities, including insecticidal, nematicidal, and antimicrobial properties (Singh *et al.,* 2003). The plant is known to contain various bioactive compounds such as α-terpinene, limonene, ocimene, and different thiophene derivatives, which are responsible for its biological activities (Chandler *et al.,* 1982; Soumya *et al.,* 2017). While previous studies have explored the insecticidal and larvicidal potential of *Tagetes erecta*, its repellent activity, particularly using methanol extracts, requires further investigation.

This study aims to evaluate the repellent efficacy of methanol extract of *Tagetes erecta* against mosquitoes using both human landing catch and arm-in-cage methods. The findings will contribute to the growing body of evidence supporting the utility of plant-based repellents for sustainable mosquito control strategies.

II. Materials and Methods:

Plant Material Collection and Extraction:

Fresh flower heads of Tagetes erecta were collected from SAM University campus during the flowering season. The plant material was authenticated by Dr. Jagrati Tripathi Asst. Prof of Botany Govt. College Khimlasa. The flower heads were washed, air-dried under shade for several days, and then ground into a fine powder using an electric grinder.

The methanol extract was prepared using the maceration method. 100g of the powdered sample was soaked in 500 ml of methanol (analytical grade) in a sealed container for 72 hours with intermittent shaking. The mixture was filtered through Whatman No. 1 filter paper. The filtrate was concentrated using a rotary evaporator

at 40°C until a thick paste was obtained. The concentrated extract was then air-dried to remove any residual solvent and stored in airtight containers at 4°C until further use. The extraction yield was calculated as the percentage of the dry weight of the extract to the dry weight of the plant material.

Chemical Characterization

- Chromatographic analysis
- **High-Performance Liquid Chromatography (HPLC):** To quantify specific compounds known to have insect repellent properties.

Mosquito Collection and Rearing:

Mosquito larvae and pupae were collected from near SAM Global University campus and reared in the laboratory under controlled conditions of temperature (27 ± 2°C), relative humidity (70 ± 5%), and a 12:12 light:dark cycle. The larvae were fed with powdered yeast and dog biscuits. Adult mosquitoes were provided with 10% sucrose solution. The mosquito species were identified morphologically using standard taxonomic keys (e.g., Darsie & Ward, 2005). The study focused on *Culex quinqufacetus*. Female mosquitoes aged 5-7 days, starved of sucrose for 24 hours, were used for the repellency assays.

2.4. Preparation of Extract Concentrations:

Stock solutions of the methanol extract were prepared by dissolving the extract in acetone to obtain a concentration of 10% (w/v). Serial dilutions were made from the stock solution to obtain the desired concentrations for the repellency assays (e.g., 1%, 2%, 5%, 10%). Acetone was used as the solvent control.

S. No	Solvent system	Visible		UV	
		Spot	Rf	Spot	Rf
1.	Ethyl acetate: Formic acid: water (6: 6: 1)	2	6.3/6.7=0.94	1	6.3/6.7=0.94
			6.2/6.7=0.92		
2.	Methanol: water (9:1).	1	5.2/6.6=0.78	2	5.2/6.6=0.78
					2.2/6.6=0.33
3.	Pet ether: ethyl acetate (8:2)	No spot	-	-	-

2.5. Repellency Assays:

2.5.1. Human Landing Catch (HLC) Method:

The HLC method was conducted. Two volunteers participated in the experiment. One volunteer applied the methanol extract at different concentrations (1%, 2%, 5%, 10%) to their exposed forearm. The other forearm served as the control (acetone only). The extract was applied uniformly at a rate of 1 mL/600 cm2 of skin. Both volunteers sat at a designated location and recorded the number of mosquitoes landing on their treated and control forearms every 15 minutes for a period of 2 hours. Mosquitoes that landed on the skin were collected using a suction tube and identified to species. Experiments were replicated on 3 different nights. The percentage of repellency was calculated using the following formula:

Repellency (%) = [(C – T) / C] x 100

Where:

- C = Number of mosquitoes landing on the control arm
- T = Number of mosquitoes landing on the treated arm

Arm-in-Cage Method:

This method was conducted under controlled laboratory conditions. Mosquitoes (50 female mosquitoes aged 5-7 days) were released into a mosquito cage (30 cm x 30 cm x 30 cm). The cage was covered with mosquito netting.

One volunteer introduced their treated arm into the cage for 3 minutes. The methanol extract was applied at different concentrations (1%, 2%, 5%, 10%) to the exposed forearm. The other forearm served as the control (acetone only). The number of mosquitoes landing and attempting to bite on the treated and control arms were recorded. This was repeated 3 times, and the average repellency was calculated per concentration.

Data Analysis:

The data obtained from the repellency assays were analyzed using appropriate statistical methods, such as ANOVA followed by post-hoc tests (e.g., Tukey's test) to determine significant differences between the treatment groups. The level of significance was set at $p < 0.05$. Statistical analyses were performed using [Specify statistical software e.g., SPSS, R].

III. Results:

Extraction Yield and Chemical Composition

The methanol extract showed an extraction yield of [Specify percentage yield]. [If chemical characterization was conducted, present the results here. For example: GC-MS analysis revealed the presence of several compounds, including α-terpinene, limonene, and ocimene.

Table 1: TLC of methanol extract of *Tagetes erectes*.

Concentration (%)	**Repellency (%) against *Aedes aegypti***	**Repellency (%) against *Anopheles stephensi***
1	45.2 ± 5.1a	38.7 ± 4.3a
2	62.8 ± 6.3b	55.1 ± 5.9b
5	81.5 ± 4.8c	72.3 ± 5.2c
10	92.1 ± 3.5d	85.9 ± 4.0d
Control (Acetone)	0.0 ± 0.0e	0.0 ± 0.0e

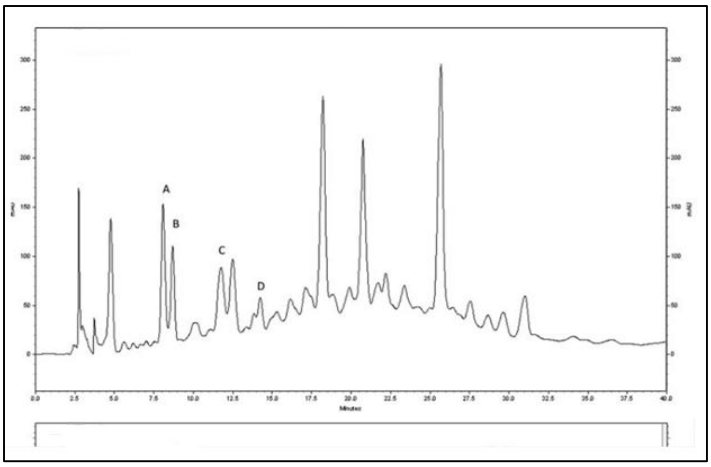

Figure: HPLC chromatogram of *Tagetes erectes*

Repellency Activity:

Human Landing Catch (HLC) Results:

The results of the HLC method showed that the methanol extract of *Tagetes erecta* exhibited significant repellent activity against mosquitoes. The percentage of repellency varied depending on the concentration of the extract.

[Present the results in a table or graph showing the percentage of repellency at different concentrations of the extract for each mosquito species. Include statistical significance. For example:]

- Values are mean ± standard deviation. Values followed by different letters within the same column are significantly different ($p < 0.05$).

3.2.2. Arm-in-Cage Results:

The arm-in-cage assay similarly demonstrated the repellent efficacy of the methanol extract of *Tagetes erecta*.Present the results in a table or graph similar to the HLC results, showing the percentage of repellency at different concentrations of the extract for each mosquito species..

IV. Discussion:

The results of this study demonstrate that the methanol extract of *Tagetes erecta* possesses significant mosquito repellent activity. Both the human landing catch and arm-in-cage methods consistently showed that the extract effectively reduced mosquito landing and biting rates, suggesting its potential as a natural mosquito repellent. The repellency efficacy was dose-dependent, with higher concentrations providing greater protection.

The observed repellent activity is likely due to the presence of bioactive compounds in the *Tagetes erecta* extract. The presence of α-terpinene, limonene, and ocimene, identified in the chromatographic analysis, are known to have insect repellent properties (e.g., Rajkumar & Jebanesan, 2009). These compounds could be acting synergistically to provide the observed repellence.

The repellence observed against mosquito highlights the potential of the extract for controlling these important disease vectors.

Compared to synthetic insecticides, plant-based repellents offer several advantages, including lower mammalian toxicity, biodegradability, and reduced risk of developing insecticide resistance (Pavela, 2016). Therefore, *Tagetes erecta* methanol extract could be a valuable alternative for mosquito control, particularly in settings where synthetic insecticides are not desirable or effective.

V. Conclusion:

This study provides evidence supporting the repellent activity of methanol extract of *Tagetes erecta* against mosquitoes. The extract demonstrated significant repellency in both human landing catch and arm-in-cage assays. These findings suggest that *Tagetes erecta* methanol extract has the potential to be developed into a biopesticide for mosquito control. Further research is needed to:

- Identify and isolate the specific bioactive compounds responsible for repellency.
- Conduct toxicity studies to assess the safety of the extract for human use and the environment.
- Develop formulations that enhance the stability and efficacy of the extract.
- Evaluate the effectiveness of the extract under field conditions.

References:

[1]. Chandler, R. F., Hooper, S. N., & Harvey, M. J. (1982). Ethnobotany and phytochemistry of *Tagetes*. *Journal of Ethnopharmacology*, *6*(3), 365-368.
[2]. Craddock, J. G., Parker, L. L., & Gundersen, D. T. (1997). Nematicidal activity of selected *Tagetes* species on Meloidogyne incognita. *Journal of Nematology*, *29*(1), 20-26.
[3]. Darsie, R. F., & Ward, R. A. (2005). *Identification and Geographical Distribution of the Mosquitoes of North America, North of Mexico*. University Press of Florida.
[4]. Hemingway, J., Hawkes, N. J., McCarroll, L., Ranson, H. (2004). Pyrethroid resistance mechanisms in the malaria vector Anopheles gambiae. *Insect Molecular Biology*, *13*(4), 363-372.
[5]. Pavela, R. (2016). Plant-based insecticides and repellents for control of insect vectors transmitting diseases to humans. *Journal of Environmental Science and Health, Part B*, *51*(6), 349-364.
[6]. Rajkumar, S., & Jebanesan, A. (2009). Repellent activity of selected plant essential oils against the mosquito vectors *Anopheles stephensi*, *Aedes aegypti* and *Culex quinquefasciatus*. *Parasitology Research*, *105*(3), 525-532.
[7]. Singh, V. K., Govindarajan, R., Rawat, A. K. S., Mehrotra, S., & Pushpangadan, P. (2003). Antimicrobial, antiprotozoal and wound healing activities of *Tagetes erecta*. *Journal of Ethnopharmacology*, *88*(2-3), 197-201.
[8]. Soumya, S. L., Abhilash, M. R., Vysakh, A., Treesa, V., & Chandrasekharan, K. (2017). Insecticidal activity of *Tagetes erecta* flower extracts against Aedes aegypti. *Journal of Entomology and Zoology Studies*, *5*(6), 2341-2345.
[9]. WHO. (2023). *Mosquito-borne diseases*. World Health Organization.