Larvicidal Activity of *Gliricidia sepium* (Jacq.) Kunth (Fabaceae) Leaf Extract Against A*nopheles* Mosquito: An Analytical Study

Dissertation

Submitted in partial fulfillment of the requirements for the award of the Degree of Bachelor of Science in Botany

BY

1. GOPIKA VARMA P

REG NO: AB22BOT020

DEPARTMENT OF BOTANY

ST. TERESA'S COLLEGE (AUTONOMOUS)

ERNAKULAM

2024-25

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CERTIFICATE

This is to certify that the dissertation entitled "Larvicidal Activity of *Gliricidia sepium (Jacq.) Kunth (Fabaceae)* Leaf Extract Against *Anopheles* Mosquito: An Analytical Study" submitted in partial fulfillment of the requirements for the award of the Degree of Bachelor of Science in Botany is an authentic work carried out by Gopika Varma P (Reg no: AB22BOT020) under the supervision and guidance of Dr. Arya P Mohan.

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Place: Ernakulam

Date: 2/5/2025

GOPIKA VARMA P

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CHAPTER 1
INTRODUCTION AND OBJECTIVES

1.1. INTRODUCTION

The mosquito subfamilies Anophelinae and Culicinae include several significant genera responsible for the transmission of life-threatening diseases. *Anopheles* mosquitoes are primary vectors of malaria and lymphatic filariasis, *Aedes* mosquitoes transmit chikungunya, dengue, lymphatic filariasis, Rift Valley fever, and Zika virus, while *Culex* mosquitoes spread Japanese encephalitis, lymphatic filariasis, and West Nile fever. Mosquitoes play a crucial role as vectors in disease transmission. Gibbons identified *Aedes aegypti* as the primary vector of arboviral dengue virus infections in tropical and subtropical regions (Baz et al. 2024). Globally, 50–100 million people contract dengue annually, with approximately 2.5% of cases resulting in death. Malaria incidence is estimated at 4–5 million cases per year in low-income countries, such as India, where around 236 million people live in filariasis-endemic areas. Additionally, diseases such as dengue, chikungunya, acute encephalitis syndrome, Japanese encephalitis, and scrub typhus have been consistently documented in Kerala state from 2011 to 2019 (Raj et al. 2022).

Since vaccines are unavailable for most mosquito-borne diseases, effective mosquito control strategies are essential for reducing transmission (Valarmathi et al. 2022). Chemical repellents such as N,N-diethyl-meta-toluamide, Picaridin, ethyl butylacetylaminopropionate, and Permethrin pose risks to human health, causing skin irritation, respiratory issues, and neurological effects. Additionally, their widespread use negatively impacts biodiversity and disrupts ecosystems. Targeting mosquito populations, particularly at the larval stage, through insecticide application remains one of the most effective preventive strategies (Yacoob et al. 2013). However, traditional mosquito control methods are becoming less effective due to the increasing resistance of mosquito populations to chemical insecticides (Pavela et al. 2019; Sonali et al. 2023). This growing resistance highlights the urgent need for innovative and sustainable solutions that can effectively manage mosquito populations while minimizing environmental and health risks (Onen et al. 2023).

Phytochemicals have emerged as a promising alternative to synthetic insecticides. These natural compounds can affect mosquitoes in multiple ways, including oviposition deterrence, developmental inhibition, hatching suppression, adulticidal and ovicidal activity, and emergence inhibition. Recent research has increasingly focused on plant-based insecticides due to their environmentally friendly nature and effectiveness. Several studies have demonstrated the mosquito-repellent properties of plants such as neem (*Azadirachta indica*), basil (*Ocimum basilicum*), peppermint (*Mentha piperita)*, and lemon eucalyptus (*Corymbia citriodora*) (Sharma et al. 1993; Ansari et al. 2000; Trigg et al. 1996). These natural substances provide a safer alternative to synthetic chemicals, reducing harm to non-target organisms and ecosystems.

One such plant with potential mosquito control properties is *Gliricidia sepium* (Jacq.) Kunth, a tropical, fast-growing leguminous tree from the family Fabaceae. This medium-sized, semi-deciduous tree, reaching heights of 10–12 meters, has been widely used for ecosystem restoration and agroforestry (Nair et al. 1984). The bark of *G. sepium* varies in colour from whitish-grey to deep red-brown. The twig of *Gliricidia sepium* (Fig. 1) exhibits pinnately compound leaves and smooth bark, while its inflorescence (Fig. 2) consists of pink to lilac-colored flowers that play a significant role in attracting pollinators and enhancing biodiversity.

Kingdom	Plantae
Phylum	Streptophyta
Class	Equisetopsida
Order	Fabales
Family	Fabaceae
Genus	Gliricidia
Species	*Gliricidia sepium*

Fig 1. Gliricidia sepium woody twig with compound leaves.

(Photo by Dinesh Valke , licensed under flickr)

Fig 2. Inflorescence : cluster of flowers in bright pink to lilac color with white tinge.

(Photo by Joao Medeiros , licensed under flickr)

Given its multiple ecological benefits, this study aims to evaluate the bio-efficacy of *G. sepium* against the fourth instar larvae of *Anopheles* mosquitoes.

This study aligns with several United Nations Sustainable Development Goals (SDGs), particularly SDG 3 (Good Health and Well-being), SDG 13 (Climate Action), and SDG 15 (Life on Land). By exploring plant-based mosquito control, it promotes healthier communities through reduced vector-borne disease transmission, thereby supporting public health initiatives. The use of *Gliricidia sepium* as a biocontrol agent reflects a sustainable approach that avoids the ecological damage associated with chemical insecticides, contributing to the protection of terrestrial ecosystems and biodiversity. Furthermore, the tree's natural abundance and multipurpose ecological role enhance its potential as a climate-resilient resource for integrated pest management. This positions the study within a broader framework of sustainable innovation, integrating environmental stewardship with health-focused outcomes.

1.2. OBJECTIVES

1. To evaluate the larvicidal property of *Gliricidia sepium* plant extract at various concentrations against *Anopheles* mosquito larvae.

2. To assess the potential for developing eco-friendly mosquito repellents from this plant source.

CHAPTER 2
REVIEW OF LITERATURE

2.1 *GLIRICIDIA SEPIUM*
2.1.1 HABIT AND MORPHOLOGY

Gliricidia sepium is a medium-sized, semi-deciduous leguminous tree from the Fabaceae family. It typically grows between 10 to 15 meters in height, with a broad canopy and a medium crown. The tree may be single-stemmed or multi-stemmed.

Native to Central America and possibly northern South America, its cultivation has now become pantropical. *Gliricidia sepium* thrives best in tropical, seasonally dry climates, preferring deep, well-drained soils, though it can tolerate shallow soils with high levels of available calcium.

The bark is smooth and varies in color, ranging from whitish grey to deep red-brown. The genus name *Gliricidia* translates to “mouse killer” in Latin, referring to its traditional use as a rodenticide. The species name *sepium* comes from the Latin saepes, meaning hedge (Marak and Wani, 2018).

Gliricidia is a genus of six to nine species of small, spreading, unarmed, fast-growing perennial shrubs or short-boled trees, reaching 5 to 15 meters in height. They are able to fix nitrogen, and nodulation has been observed and evaluated (Patil and Prasunamma, 1986). They nodulate readily, usually within three months of planting when grown from stakes, or even faster when established from seed (Chadhokar, 1982). They are deciduous, glabrous, and characterized by a spreading or pyramidal crown of foliage borne on long, irregular, feathery branches that often curve downwards. They seasonally bear numerous large, showy, pink to light purplish, pea-shaped flowers, which often appear before the leaves at the end of the dry season. The pods are glabrous, blackish at maturity, and measure up to 14 cm long and 1.5 cm wide (Smith and van Houtert, 1987).

Native to tropical America and the West Indies, *Gliricidia* species now enjoy worldwide distribution as multipurpose trees suitable for use as living fences, for shade and soil improvement, as fuelwood and pole material, and as sources of high-protein fodder and browse. *Gliricidia* establishes well in the tropics: it is found in lowlands and at altitudes of up to 2000 meters. Although it is best suited to the wetter areas in

the humid tropics where it is widely cultivated, it is native to the drier parts of Central America. Its considerable drought tolerance is due to a policy of avoidance, as it drops its leaves and becomes dormant during the driest parts of the year. The plant thrives on relatively acidic, infertile soils and shows some tolerance of short-term waterlogging, conditions which are unsuitable for some of the other popular tree legumes such as *Leucaena leucocephala* (Chadhokar, 1982).

The common names of *Gliricidia sepium* In various languages include:

Malayalam	Sheemakkonna
Tamil	Seemai Agathi,Vivasaaya Thegarai
Kannada	Gobbarada gida
Telugu	Madri
Marathi	Giripushpa
Bengali	Sharanga

2.1.2 USES OF THE PLANT

Gliricidia sepium (Jacq) Walp., commonly known as *Gliricidia*, is a highly versatile leguminous tree native to Central America and extensively cultivated across tropical regions, including India. It serves multiple purposes across agriculture, animal husbandry, medicine, and environmental management (Alamu et al., 2023). Recognized as a truly multifunctional species, Gliricidia offers substantial ecological, economic, and social benefits. Its wide-ranging applications-in soil fertility enhancement, fodder provision, natural pest control, traditional medicine, and ecological conservation-position it as a key component of integrated rural development in tropical areas.

In Kerala, *Gliricidia* is commonly used as green manure in paddy fields and plantations, contributing to improved soil fertility and reduced dependence on chemical fertilizers. It is also used as a live fence, shade tree in plantations (especially coconut and pepper), and as fodder for livestock during dry seasons. Its adaptability to local agro-climatic conditions makes it a valuable species for sustainable agricultural practices in the region.

One of the most significant contributions of *Gliricidia sepium* lies in its ability to enhance soil fertility. As a legume, it forms symbiotic relationships with nitrogen-fixing bacteria in its root nodules, converting atmospheric nitrogen into forms readily available to plants. This natural fertilization reduces reliance on synthetic nitrogen inputs. Furthermore, its rapidly decomposing leaf litter returns essential nutrients such as potassium, phosphorus, and calcium to the soil. When used in alley cropping or intercropping systems, *Gliricidia* enriches soil organic matter and microbial biomass, thereby improving soil structure and long-term fertility. Its green biomass can be incorporated as green manure, significantly increasing nitrogen levels and boosting crop productivity in a sustainable manner (Alamu et al. 2023).

The species also contributes to Increased crop yields. Intercropping *Gliricidia* with key staples like maize, cassava, and tomato has demonstrated improvements in plant growth and output. Its leaf biomass not only serves as a rich nutrient source but also helps in suppressing weed growth and retaining soil moisture. For instance, a field experiment utilizing *Gliricidia* leaf compost alongside urea fertilizer on tomato crops showed enhanced fruit size, yield, and soil pH levels. This integrated approach proves especially valuable for small-scale farmers coping with declining soil fertility (Keya et al. 2021).

Gliricidia sepium is a cornerstone of agroforestry systems and erosion control strategies. Its extensive root network stabilizes soil on slopes, making it a suitable choice for contour hedgerows and live fences in tropical farming. The tree acts as a natural windbreak and supports terracing practices, aiding water conservation and reducing surface runoff.

In the realm of animal husbandry, *Gliricidia* serves as a valuable fodder crop. Its leaves contain approximately 20–25% crude protein, making them an excellent supplement for ruminants such as cattle, goats, and sheep. When combined with lower-quality forages, *Gliricidia* improves the overall diet quality, leading to better feed efficiency and weight gain. For example, feeding trials with Bali cattle showed improvements in weight gain, digestibility, and overall feed utilization, especially during dry seasons when fresh forage is scarce (Rusdy et al. 2019).

Research also suggests that *Gliricidia* leaf meal can be incorporated into the diets of poultry and rabbits. It contributes to better weight gain, enhances immune responses, and lowers feed costs, highlighting its potential in cost-effective animal nutrition strategies.

Traditional medicine has long valued *Gliricidia sepium* for its therapeutic properties. Its leaves and bark are used in treating wounds, ulcers, and skin infections due to their antimicrobial and anti-inflammatory effects. In experimental models, ointments prepared from *Gliricidi*a extracts showed significant tissue regeneration and reduced inflammation in rats with induced wounds, validating its traditional use (Aulanni'am et al. 2021).

In addition, the plant holds promise in the management of sickle cell disease. In West African medicine, its aqueous extracts have been used to mitigate red blood cell sickling. Scientific investigations confirm that *Gliricidia* reduces hemoglobin polymerization and oxidative stress under low-oxygen conditions, indicating its potential for further phytotherapeutic development (Oduola et al. 2016).

Pest and vector control is another domain where *Gliricidia sepium* excels. Its bioactive compounds-particularly flavonoids and tannins-are toxic to mosquito larvae, notably *Aedesaegypti*, a vector for dengue and Zika viruses. Methanolic extracts of the plant have demonstrated significant larvicidal effects within 24 to 48 hours, showcasing it as a viable eco-friendly solution for vector control. Additionally, *Gliricidia* extracts are used topically on livestock to repel ticks and fleas, providing a safe, plant-based alternative to chemical antiparasitics.

From an energy and environmental standpoint, *Gliricidia* is a favored source of fuelwood and charcoal. Its wood has a high calorific value, burns cleanly, and regenerates quickly, making it ideal for rural energy needs. Moreover, due to its rapid growth and substantial biomass production, *Gliricidia* serves as a carbon sink. By sequestering atmospheric carbon dioxide and improving local microclimates, it contributes significantly to climate adaptation and sustainable land management practices.

Lastly, *Gliricidia sepium* is extensively used as a living fence. Its ability to grow quickly from cuttings, along with its low maintenance needs, makes it an affordable and practical fencing solution for farmers. These living fences help demarcate land, control livestock movement, reduce erosion, and serve additional roles as sources of fodder, fuelwood, and organic mulch. As a nitrogen-fixing species, *Gliricidia* further enhances the fertility of surrounding soils, supporting the growth of adjacent crops.

In conclusion, *Gliricidia sepium* exemplifies a multipurpose tree that supports sustainable agriculture, improves soil and crop health, enhances livestock productivity, and contributes to environmental conservation. Its wide range of applications makes it a cornerstone species in agroecological systems across the tropics.

CHAPTER 3
MATERIALS AND METHODS

3.1 Collection of Plant Material

Leaves of *Gliricidia sepium (Jacq.) Kunth* (family Fabaceae), commonly known as sheemakkonna , were collected from Ernakulam, Kerala, India identified by Department of Botany, St. Teresa's college, Ernakulam, Kerala. The plants were identified using flora Gamble, 1925 (Gamble,1925). The collected leaves were thoroughly washed with distilled water to remove dust and debris. The fresh leaves were grinded using motor and pestle, and stored in containers for subsequent processing.

3.2 Preparation of Plant Extract

100 grams of crushed *G. sepium* leaves were extracted using distilled water as the solvent. For extraction, the crushed leaf material was mixed with 100 mL of distilled water in a glass vial. The mixture was stirred for five minutes and then filtered to separate the plant residue from the extract. The supernatants were collected and stored in 4 degree celsius unit further use.

3.3 Collection and Identification of Mosquito Larvae

Mosquito larvae were collected from natural habitats, specifically water-logged areas in Kumbalangi, Ernakulam, Kerala (Latitude: 9.8760584° and Longitude: 76.2870781°) and were maintained in the laboratory. Larval identification was performed according to standard identification literature. (Atting et al. 2016)

3.4 Larvicidal Bioassay

The larvicidal bioassay was conducted following the standard protocol outlined by the World Health Organization (WHO) (Mukhtar et al., 2015). The efficacy of *G. sepium* extract (water) was evaluated against the fourth instar larvae of Anopheles Mosquito.

The stock solution was taken to be 100% (w/v), as 100 g of plant extract was taken in 100 ml of distilled water.

The percentage concentration (% w/v) was determined using the following formula:

$$\text{Concentration (\% w/v)} = \frac{\text{Mass of solute (g)}}{\text{Total volume of solution (mL)}} \times 100$$

Various concentrations (0.1%,1%,10%, 20%, 30%, 40% and 50%) were made by diluting the stock solution with distilled water with the help of the formula

$$C1V1 = C2V2$$

Where C1 is the stock concentration (100%), V1 is the volume of stock solution taken, C_2 is the desired concentration, and V2 is the final volume after dilution. The necessary dilutions were prepared accordingly for application in the bioassay test.

A range of extract concentrations (0.1%,1%,10%,20%,30%,40% and 50%) was prepared, with 5 larvae exposed to each concentration in separate test tubes containing 10 mL of the extract solution. A negative control was established using distilled water without any plant extract.

After 24 hours of exposure, larval mortality was recorded. Larvae were considered dead if they showed no movement upon gentle probing. Mortality data were corrected using Abbott's formula to account for natural mortality in the control group. The percentage mortality for each concentration was calculated, and lethal concentration values were determined using probit analysis. (Banupriya et al. 2015)

$$\text{Corrected mortality} = \frac{\text{Observed mortality in treatment} - \text{Observed mortality in control} \times 100}{100 - \text{Control mortality}}$$

$$\text{Percentage mortality} = \frac{\text{Number of dead larvae}}{\text{Number of larvae introduced}} \times 100$$

3.5 Survey conducted

A survey was conducted among 176 participants to assess public awareness and perception towards mosquito control and repellents, with a specific focus on eco-friendly alternatives.

CHAPTER 4
RESULTS AND DISCUSSIONS

4.1 RESULT

Bioassay was conducted to evaluate the effect of *Gliricidia sepium* leaf extract on the fourth instar larvae of Anopheles mosquitoes. Bioassays are scientific tests used to assess the biological activity, toxicity, or potency of a substance by observing its effects on living organisms under controlled conditions. Figure 3 presents a microscopic view of Anopheles mosquito larvae, highlighting their morphological characteristics. The mosquito larvae species and their developmental stage were identified as the 4th instar stage of Anopheles mosquito following standard literature guidelines. The primary distinguishing features include the two large palps next to their proboscis and wings with a scaly appearance and a pattern of pale and dark spots.

Fig 3. *Anopheles* mosquito larva under microscope

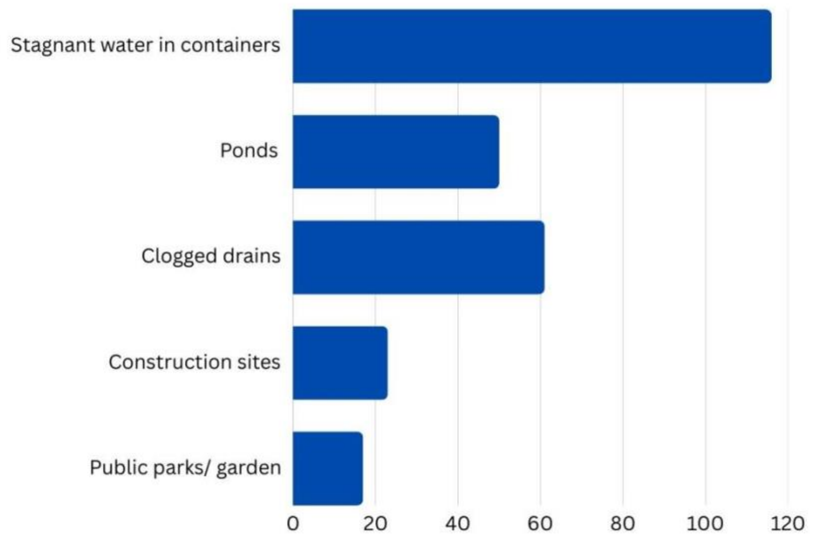
(Photo by Harry Weinburgh, licensed under Pixnio)

The conducted survey is used to analyze and interpret the mosquito menace and to understand public opinion regarding the introduction of natural mosquito repellents. The primary objective of this study is to assess public concern about mosquito breeding and the prevention measures adopted, as well as the willingness of people to switch from current repellents to plant-based alternatives.

By analyzing survey responses, we aim to identify trends in avoiding mosquito breeding grounds both within households and in the surrounding localities, along with the harmful effects experienced by the public from existing repellents.

Using various statistical tools and methods, we will examine key aspects such as public preferences for environmentally friendly products and the factors that drive or hinder the use of current repellents. The interpretation of the data is based on primary responses collected from 176 participants, ensuring a reliable representation of public perspectives.

1.What are the common mosquito breeding sites in your household?



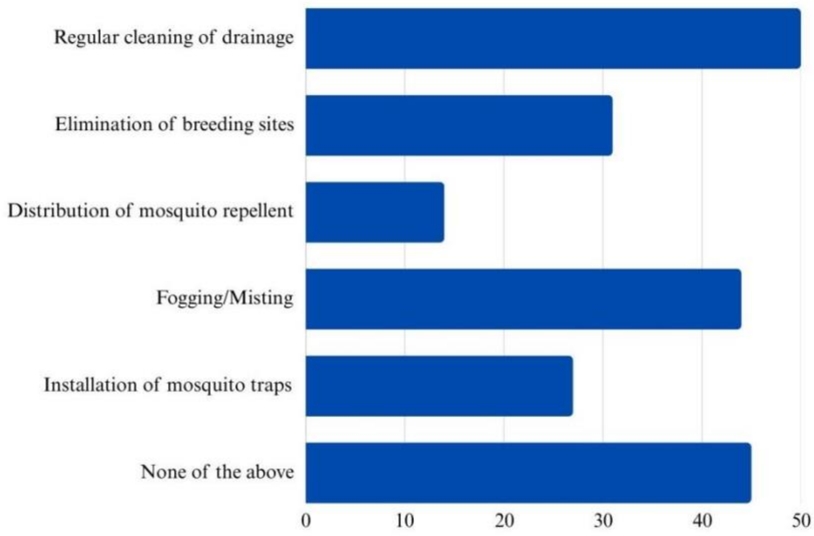
Graph 1: Common mosquito breeding sites in household

INTERPRETATION

The data shows that stagnant water in containers is the most common mosquito breeding site, reported by 65.9% (116 respondents). This is followed by clogged drains at 34.7% (61 respondents) and ponds at 28.4% (50 respondents). Construction sites were identified by 13.1% (23 respondents), while only 9.7% (17 respondents) cited public parks or gardens.

This indicates that household-level water storage and poor drainage systems are the primary contributors to mosquito breeding, underlining the importance of domestic preventive measures. The relatively low numbers for construction sites and public parks suggest that personal environments pose a greater risk compared to public or shared spaces.

2. How is mosquito breeding prevented in your locality?



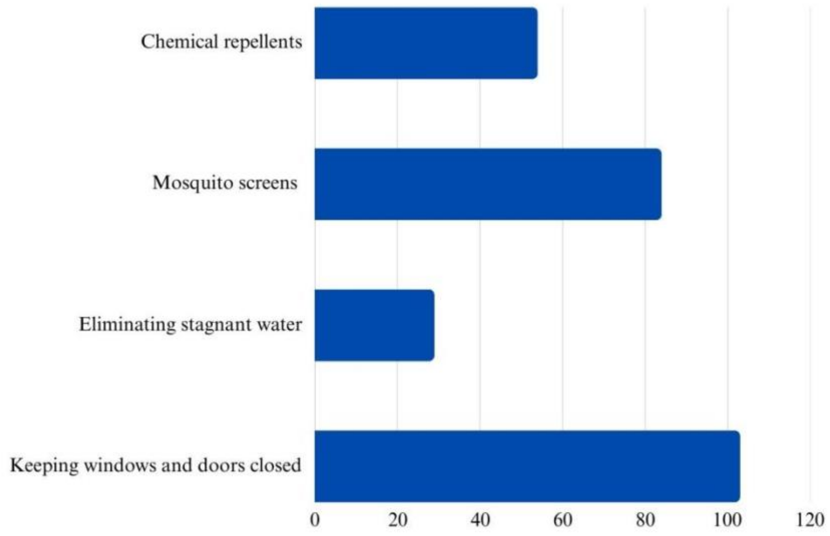
Graph 2 : Mosquito Breeding Prevented in Locality

INTERPRETATION

The data shows that regular cleaning of drainage is the most commonly reported preventive measure, cited by 28.4% (50 respondents). This is followed closely by fogging or misting at 25% (44 respondents) and the option “none of the above” at 25.6% (45 respondents), indicating a lack of visible or effective control measures in certain areas. Installation of mosquito traps was noted by 15.3% (27 respondents), while elimination of breeding sites was reported by 17.6% (31 respondents). Only 8% (14 respondents) mentioned the distribution of mosquito repellents.

This suggests that while some localities are actively engaging in control measures such as drainage cleaning and fogging, a significant portion of the population perceives that no proper mosquito control initiatives are being undertaken. The findings highlight a need for more consistent and visible public health interventions at the community level.

3. What methods do you employ in your household?



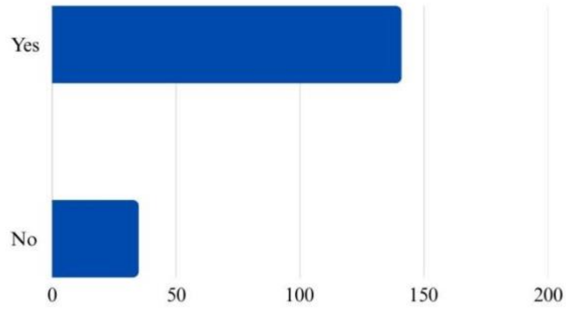
Graph 3 : Methods Employed in Households to Prevent Mosquito Entry

INTERPRETATION

The data shows that the most commonly adopted method for preventing mosquito entry is keeping windows and doors closed, reported by 58.5% (103 respondents). Mosquito screens are used by 47.7% (84 respondents), followed by chemical repellents at 30.7% (54 respondents). Only 16.5% (29 respondents) reported actively eliminating stagnant water as a preventive measure.

This indicates that passive barriers such as closed doors/windows and screens are preferred over active preventive actions like removing stagnant water or using repellents. The relatively low adoption of source elimination practices highlights a potential gap in awareness or effort toward addressing the root cause of mosquito breeding.

4. Have you noticed an increase in the mosquito population over the past period?



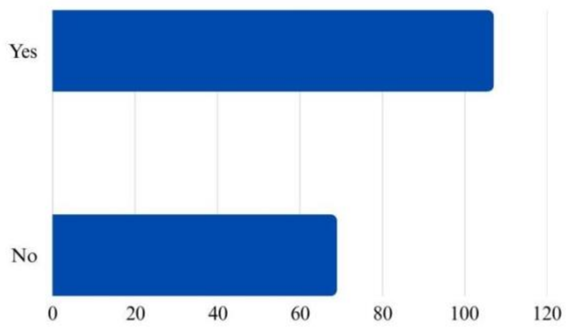
Graph 4 : Perception of Mosquito Population Increase

INTERPRETATION

The data reveals that a significant majority of respondents, 141 out of 176 (80.1%), have noticed an increase in the mosquito population over the past period. In contrast, only 35 respondents (19.9%) reported no noticeable change.

This indicates a growing public concern regarding the rising mosquito population, which may be linked to changes in environmental conditions, inadequate waste management, or increased breeding grounds. The strong perception of an increase highlights the urgency for more effective control measures and public health interventions.

5. Have you considered utilizing plant-derived mosquito repellent?



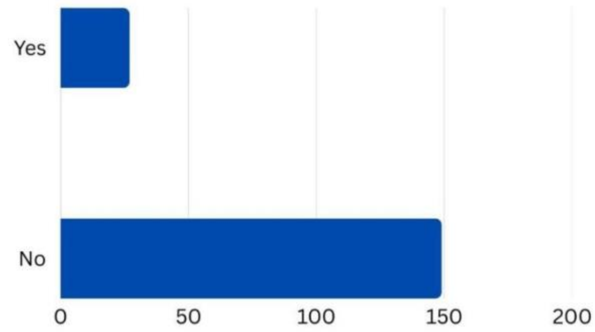
Graph 5 : Consideration of Plant-Derived Mosquito Repellents

INTERPRETATION

The data shows that 107 out of 176 respondents (60.8%) have considered using plant-derived mosquito repellents, while 69 respondents (39.2%) have not.

This suggests a growing interest in natural and potentially safer alternatives to chemical repellents among the public. The majority's willingness to consider plant-based options reflects increased environmental awareness and a shift toward more sustainable health practices. However, a significant portion still remains hesitant, possibly due to concerns about effectiveness or lack of information.

6. Have you experienced any mosquito-borne diseases?



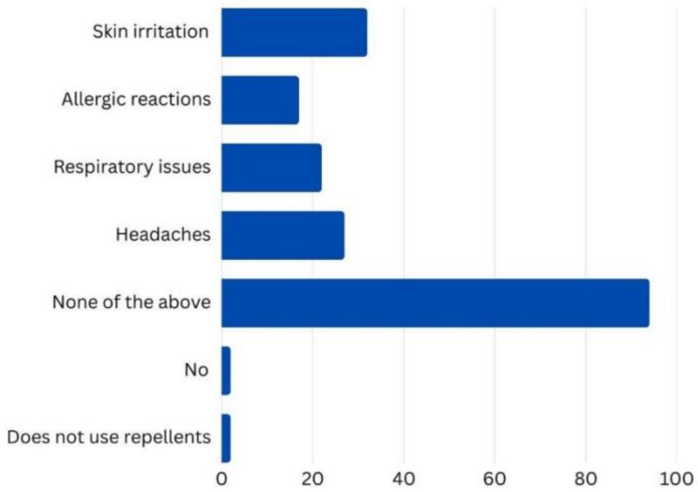
Graph 6 : Experience with Mosquito-Borne Diseases

INTERPRETATION:

The data shows that a large majority of respondents (87.7%) have not experienced any mosquito-borne diseases, while only 12.3% reported having been affected.

This suggests that although mosquito exposure may be common, actual disease incidence remains relatively low among the surveyed group, possibly due to effective preventive measures or limited exposure to high-risk areas.

7. Have you noticed any adverse effects from using your current mosquito repellent?



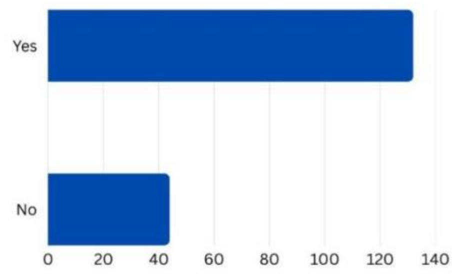
Graph 7 : Adverse effects from current mosquito repellents

INTERPRETATION

The data reveals that a majority (94 respondents) reported no adverse effects from using mosquito repellents. However, some users experienced issues, with 32% reporting skin irritation, 27% headaches, 22% respiratory issues, and 17% allergic reactions (multiple responses allowed).

This indicates that while most users tolerate repellents well, a significant minority do experience side effects, highlighting the need for safer, skin-friendly alternatives, especially for sensitive users.

8. Would you prefer using a natural mosquito repellent over conventional options ?



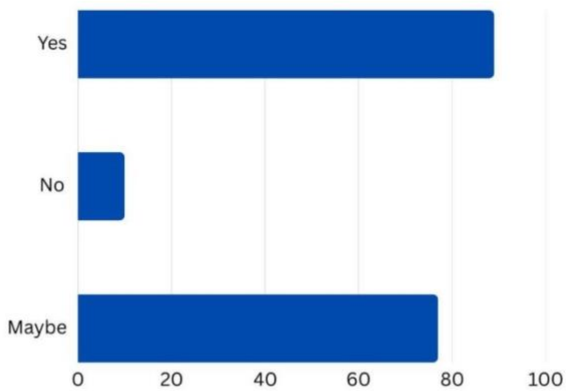
Graph 8 : Preference for Natural Mosquito Repellents

INTERPRETATION

A significant majority (76.6%) of respondents a preference for using natural mosquito repellents over conventional ones, while only 23.4% preferred not to.

This highlights a strong consumer inclination towards safer, eco-friendly alternatives, possibly due to concerns over side effects or chemical content in conventional repellents.

9. If provided with a sample of natural mosquito repellent would you be willing to try it?



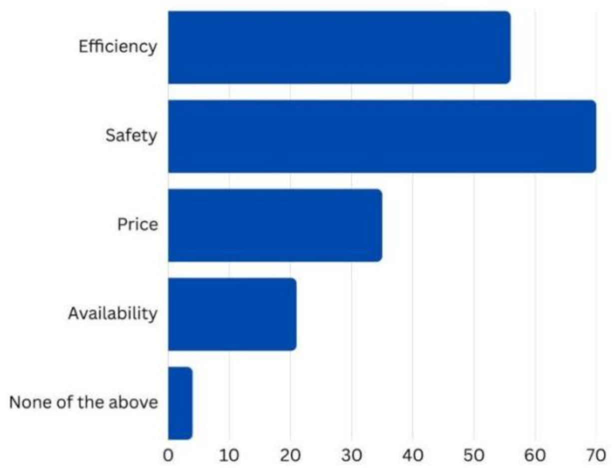
Graph 9 : Willingness to Try a Sample of Natural Mosquito Repellent

INTERPRETATION

A large majority (89%) of respondents showed interest in trying a natural mosquito repellent, with 44.5% saying “Yes” and another 44.5% choosing “Maybe.” Only 11% were not interested.

This indicates a strong preference for natural, eco-friendly solutions—likely influenced by concerns about chemical exposure in conventional repellents.

10. Do you have any concerns or complaints about the mosquito repellents you currently use?



Graph 10 : Concerns or Complaints About Current Mosquito Repellents

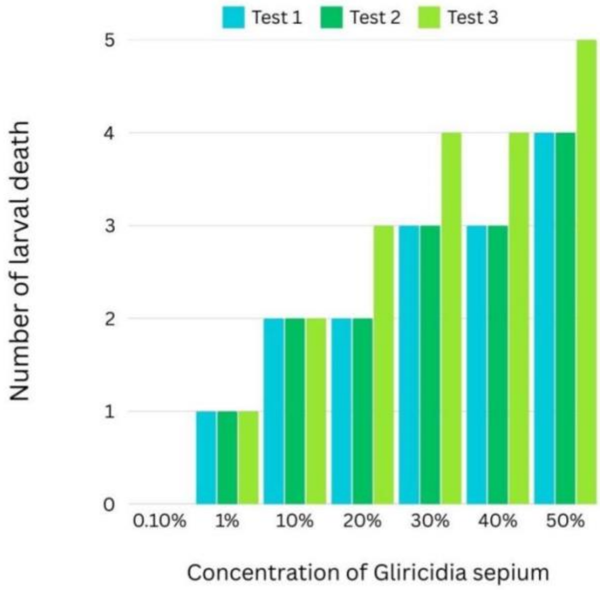
INTERPRETATION

The majority of respondents (66.3%) identified safety as their main concern regarding current mosquito repellents, followed by efficiency (58.7%) and price (35.9%). Fewer users raised concerns about availability (22%) or had no complaints (3.3%).

This highlights that users are primarily worried about the health impacts and effectiveness of the repellents they use, reinforcing the need for safer, reliable, and accessible alternatives in the market.

Anopheles mosquito larvae were exposed to seven concentrations of crude leaf extracts, which demonstrated activity likely due to a complex mix of active compounds. Initial screening after 24 hours revealed notable larvicidal potential in extracts prepared with water as the solvent. The various concentration percentages used in the bioassay test included (0.1%,1%,10%,20%,30%,40% and 50%). The mortality percentage, calculated using Abbott's formula, ranged from 0%, 20%, 40%, 40%, 60%, 60% & 80% against 4th instar larvae, respectively. The greatest mortality rate was observed at the 50% concentration of *G. sepium*, with 80% mortality obtained in the 24-hour assay. Higher concentrations (above 50%) were tested, resulting in 100% mortality. A clear dose-dependent response was observed, with mortality increasing as concentration increased. Beyond a certain level, mortality plateaued at 100%, indicating maximum efficacy. The leaf extract of *Gliricidia sepium* with water as a solvent showed high larvicidal activity. In the negative control (water), larvae remained viable after 24 hours.

Image 1. Larvicidal bioassay being conducted on *Anopheles* mosquito larvae using extract of Gliricidia sepium.



Graph 11: Larvicidal Bioassay Results

4.2 Discussion

The phytochemicals present In *G.sepium* includes Alkaloid, Flavonoids, Steroids, Flavones, Anthracene Glycoside and these compound results in the various insecticidal properties that contributes to the larvicidal activity against the developmental stages of A.aegypti. (Banupriya et al. 2015). The effectiveness of plant constituents is primarily due to their ability to regulate growth, rather than their toxic properties (Moore et al. 2003).

Bioactive compounds such as terpenoids, flavonoids, saponins, steroids, and tannins, may have contributed to the observed larval and pupal mortality due to their insecticidal properties (Zahran et al. 2010). According to Liu et al., alkaloids are among the bioactive compounds responsible for the toxicity to mosquito larvae (Liu et al. 2012). Botanical derivatives with mosquito larvicidal properties typically exert their effects through multiple mechanisms, including: direct attack on the nervous system, causing damage; disruption of the midgut epithelium, with secondary effects on the gastric caeca and malpighian tubules; acting as mitochondrial poisons; and interacting

with and disarranging the cuticle membrane of the larvae, ultimately leading to larval death.

Applying botanical substances in mosquito control, rather than synthetic insecticides, offers a promising approach to cost reduction and minimizing environmental pollution. The overuse of synthetic insecticides such as N,N-Diethyl-meta-toluamide, Picaridin, Ethyl Butylacetylaminopropionate, and Permethrin as mosquito repellents poses risks to human health, causing skin irritation, respiratory issues, and neurological effects. It also affects non-target organisms and disrupts ecosystems.

4.3 Conclusion

This study indicates the promising potential of *Gliricidia sepium* leaf extract as a natural larvicide against Anopheles mosquitoes. The extract was very effective in controlling mosquito larvae, offering a safer and eco-friendly alternative to chemical insecticides. Since *G. sepium* is easily accessible and easy to use, it could be a potential alternative for mosquito control in local communities. With more research and development, this plant-based solution could potentially contribute to sustainable mosquito management and reduce health and environmental risks.

5.1 Reference

Alamu, E. O., Adesokan, M., Fawole, S., Maziya-Dixon, B., Mehreteab, T & Chikoye, D. (2023). *Gliricidia sepium* (Jacq.) Walp Applications for Enhancing Soil Fertility and Crop Nutritional Qualities: A Review. Forests, 14(3), 635.

Ansari, M. A., Vasudevan, P., Tandon, M & Razdan, R. K. (2000). Larvicidal and mosquito repellent action of peppermint (Mentha piperata) oil. Bioresource Technology, 71(3), 267–271.

Aulanni'am, A., Ora, K. M., Ariandini, N. A., Wuragil, D. K., Permata, F. S., Riawan, W & Beltran, M. A. G. (2021). Wound healing properties of *Gliricidia sepium* leaves from Indonesia and the Philippines in rats (*Rattus norvegicus*). Veterinary World, 14(3), 820–824.

Banupriya, D., Umavathi, S & Cinthamani, M. (2015). Efficiency of *Nerium oleander* L. and *Gliricidia sepium* (Jacq.) Kunth ex Walp. As a larvicidal agent against aquatic stages of *Aedes aegypti* (L.),Int. J. Adv. Res. Biol.Sci. 2(1): (2015): 33–37.

Chadhokar, P.A. and Lecamwasam, A. (1982). Effect of feeding *Gliricidia maculata* to milking cows: a preliminary report. Tropical Grasslands, 16:46-48.

Gamble J. S. (1925). Flora of the Presidency of Madras (Vol. 3, pp. 1351–1371). Adlard & Son Ltd.

Inyang A. Atting, Mfonobong E. Akpan (2016). Morphological and Molecular Characterization of *Anopheles* Mosquitoes and Their Behavioural Patterns in Uyo, Nigeria. International Journal of TROPICAL DISEASE & Health 14(2): 1-11

Jishnu Raj, Soorya V, Anitha K.R (2022): Trends and seasonality of vector borne diseases in Kerala. VOL. 9 NO. 10. International Journal Of Community Medicine.

Keya, S. S., Miah, M. G., Rahman, M. A & Islam, M. T. (2021). Application of Gliricidia sepium Tree Leaves and Nitrogen Fertilizer to Improve Tomato Production and Soil Properties. Annals of Bangladesh Agriculture, 24(1), 77–87.

Liu, Z. L., Liu, Q. Z., Du, S. S & Deng, Z. W. (2012). Mosquito larvicidal activity of alkaloids and limonoids derived from Evodia rutaecarpa unripe fruits against Aedes albopictus (Diptera: Culicidae). Parasitology Research, 111(3), 991–996.

Mayona K. Marak and Afaq Majid Wani. (2018). Monitoring of Vegetative and Floral Characters of Gliricidia sepium. ISSN: 2319-7706 Volume 7 Number 09. International Journal of Current Microbiology and Applied Science

Mohamed M Baz, Abdelfattah M Selim, Ibrahim Taha Radwan, Abeer Mousa Alkhaibari, Hattan S Gattan, Mohammed H Alruhaili, Saeed M Alasmari, Mohammed E Gad (2024): Evaluating larvicidal, ovicidal and growth inhibiting activity of five medicinal plant extracts on Culex pipiens (Diptera: Culicidae), the West Nile virus vector , Scientific Reports 14 (1), 19660

Mohamed Yacoob, S., Sundaram, R & Beula, J. (2013). Mosquito larvicidal activity of seaweed extracts against Anopheles stephensi, Aedes aegypti, and Culex quinquefasciatus. Asian Pacific Journal of Tropical Disease, 3, 196–201.

Moore, S., Camoron, M & Hill, N. (2003). Low-technology approaches for the use of neem extract for mosquito control. In M. Cole & R. Strang (Eds.), Science and application of neem (pp. 40–42).

Mukhtar, M., Mushtaq, S., Arslan, A., ki, A., Hammad, M & Bhatti, A. (2015). Laboratory study on larvicidal activity of different plant extracts against Aedes aegypti. International Journal of Mosquito Research, 127, 127–130.

Nair PKR, Fernandese ECM, Wombugn (1984): Multipur-Pose leguminous trees and shrubs for agroforestry. Agro-Forestry Syst 2: 145–163.

Oduola, T., Dallatu, M. K., Muhammed, A. O., Ndakotsu, M. A., Adebisi, I. M & Hassan, S. W. (2016). Gliricidia sepium Aqueous Leaf Extract Possesses Antisickling Property. International Blood Research & Reviews, 5(3), 1–6.

Onen, H., Luzala, M. M., Kigozi, S., Sikumbili, R. M., Muanga, C.-J. K., Zola, E. N., Wendji, S. N., Buya, A. B., Balciunaitiene, A., Viškelis, J., Kaddumukasa, M. A &

Memvanga, P. B. (2023). Mosquito-borne diseases and their control strategies: An overview focused on green synthesized plant-based metallic nanoparticles. Insects, 14(3), 221.

Patil, S.D and Prasunaμμα, K. (1986). A preliminary evaluation of nodules from six nitrogen fixing trees. Nitrogen Fixing Tree Research Reports, 4 20-21.

Pavela, R., Maggi, F., Iannarelli, R & Benelli, G. (2019). Plant extracts for developing mosquito larvicides: From laboratory to the field, with insights on the modes of action. Acta Tropica, 193, 236–271.

Rusdy, M., Baba, S., Garantjang, S & Syarif, I. (2019). Effects of supplementation with Gliricidia sepium leaves on performance of Bali cattle fed elephant grass. Livestock Research for Rural Development, 31(6).

Sharma, V. P., Ansari, M. A & Razdan, R. K. (1993). Mosquito repellent action of neem (Azadirachta indica). Journal of the American Mosquito Control Association, 9, 359–360.

Smith, O. B., Van Houtert, M. F. J., (1987). The feeding value of Gliricidia sepium. A Review. World Animal Review, 62: 57-68.

Sonali, K., Poonam, K & Ankita, S. (2023). Review on mosquito repellent incense sticks using herbs. IJARIIE-ISSN(O)-2395-4396

Trigg, J. K & Hill, N. (1996). Laboratory evaluation of a eucalyptus-based repellent against four biting arthropods. Phytotherapy Research, 10(4), 313–316.

Valarmathi, D. S., SenthilKumar, D. M., Chithra, E., Nisha, R. C. C., Prabu, L. A. N., Shibin, J. A & Abzal, M. (2022). Design and evaluation of mosquito repellent incense sticks using herbals. Volume 7, Issue 1, pp: 873-876. International Journal of Pharmaceutical Research and Applications.

Zahran, H. E. M & Abdelgaleil, S. A. M. (2010). Insecticidal and developmental inhibitory properties of monoterpenes on Culex pipiens L. (Diptera: Culicidae). Journal of Asia-Pacific Entomology, 14, 46–51.