

# Antibacterial and Mosquito Repellent Assay of Poly Herbal Oil

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## ABSTRACT

**Aim:** To evaluate the anti-bacterial and mosquito-repellant activities of herbal skin oil. **Materials and Methods:** The herbs used for the present study were washed well with tap water and weighed. 250 g each of the selected plant leaves was weighed and made into a paste using mortar and pestle. 500 mL of coconut oil was measured in a measuring cylinder transferred to a heating vessel and heated to low flame. The crushed leaf paste of *Salvia rosmarinus*, *Azadirachta indica*, and *Ocimum basilicum* were added to the coconut oil and heated to low flame for 15 min. The oil was cooled down to room temperature and filtered using a fine mesh filter. The filtered herbal oil was stored in glass bottles. **Results and Discussion:** *In vitro* antimicrobial activity of the Herbal oil was tested against 4 bacterial pathogens using the agar disc diffusion method. The zones of inhibition (in mm) were as follows: The highest inhibition was noted (19 mm) for *Streptococcus pyogenes*. The Medium inhibition was noted (17 mm) for *Staphylococcus aureus* and (15 mm) for *Aeromonas hydrophila*. The lowest inhibition was noted (14 mm) for *Pseudomonas aeruginosa*. The survey forms were collected from the randomly selected people after 10 days. About 40% of the surveyed people rated 5 out of 5 for the herbal oil. Another 40% of the surveyed people rated above 4.5 out of 5. The remaining 20% is rated above 4 for the herbal oil. From the survey, it is evident that the prepared herbal preparation is an effective mosquito repellent and is also 100% safe for the skin as a topical application. **Conclusion:** There are many chemical mosquito repellents seen in our market that cause side effects. This paper focuses on herbal mosquito repellent as well as poly herbal skin oil. In the present study, three herbs *Salvia rosmarinus*, *Azadirachta indica*, and *Ocimum basilicum* are selected to come up with an oil-based herbal formulation to be used as mosquito repellent and skin disease cure oil. The antibacterial activity of the herbal oil was good against pathogens causing skin infections and the mosquito-repellent efficiency of the poly-herbal oil is assessed using the Survey method. From the survey conducted, it is evident that the prepared herbal preparation is an effective mosquito repellent and is also 100% safe for the skin as a topical application.

**Keywords:** Antibacterial activity, Ethnobotanical, Medicinal plants, Mosquito repellent.

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## INTRODUCTION

Mosquitoes is an insect which causes threatening diseases like malaria, dengue etc, over 700 million people were affected by diseases caused by mosquito. Mosquito is cosmopolitan insects seen all over the world. There are 3541 species of mosquito were identified from the tropical and sub-tropical countries. Recently there is a report, about emerging of mosquito borne diseases which causes series, public health

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problem.<sup>[1]</sup> In past decades such mosquito-borne diseases were common in urban area due to open sewage and poor maintains of waste dumping, but now-a-days disease like malaria, dengue were reported in villages too, because of favorable breeding sites for the mosquito. More over because of over population and poor sewage management our country people also got affected by mosquito-borne disease like dengue, malaria, chikungunya. In India year by year there are many people infected by malaria, chikungunya. On other hand changes in epidemiology of such diseases in terms of strains, severity and geographic distribution is also common and there is also emerging insecticidal resistance mosquito varieties act as main causative for such dangerous disease spread.<sup>[2]</sup> WHO also reported every year 50-100 million dengue infections noticed worldwide is a series problem on mankind. WHO revealed that India bears mosquito-borne disease 34% of global dengue and 11% of global malaria cases.<sup>[3]</sup> To prevent the prevalence of mosquito-borne diseases it is important to prevent the man - mosquito contact to the most possible extent. Today's markets are flooded with chemical-based repellents which are scientifically proven to cause certain adverse effects to human health including respiratory disorders and skin problems. So the formulation of a more safe mosquito repellent has become the need of the hour. Since the prehistoric period, a large part of the population recognizes the remedial property of numerous plants and the same has been proved by medical science.<sup>[4]</sup> India a land of vast alters in soil along with weather conditions is an idyllic position meant for the gardening of a great number of plants with medicinal properties and which can be used in cosmetics, agrochemicals, and perfumery also in pharmaceutical industries.<sup>[5]</sup> Herbal preparations have been in use for centuries in traditional medicine Siddha and Ayurveda as effective mosquito repellents. Plants have also been burnt and used as fumigants to fend off mosquitoes.<sup>[6]</sup> Ethnobotanical evidence also quotes the efficacy of many indigenous herbs as good repellents. The present experiment is an attempt the preparation of an herbal oil-based formulation to be used as an effective mosquito repellent with additional benefits like enhancing skin texture and preventing skin disease. Three plants *Salvia rosmarinus*, *Azadirachta indica*, *Ocimum basilicum*, and coconut oil were selected for the preparation of the herbal oil. In ancient literature and folklore rosemary has been reported as emblem of recall and loyalty. It acts as a stimulator. In traditional medicine active principle of rosemary used as an ingredient in tonic and in ointments, it was a popular aromatic component of tonics and liniments. Now a

days it is used as an ingredient in toilet soap bar and Landry powder for its aromatic fragrance. The essential oil content is from 0.3 to 2% essential oil of rosemary were extracted using steam distillation method, it is obtained by distillation; its component is borneol.<sup>[7]</sup> Another plant of this study is neem all parts including roots, barks, seed, fruit, leaves, buds, flowers of neem contain medicinal property. Neem is also used as main medicine in Indian Ayurvedic and Siddha Medicine. Azadirachtin and Nimbidine is an active principle which acts as anti-microbial agent in neem. Neem extracts also used as antimicrobial, insecticide, mite repellent in our country, because of high value neem extracts also used as an ingredient in soap, shampoo, cosmetics and ointment. In traditional medicine neem oil mainly used to treat skin diseases like psoriasis, skin infection, ulcer, wound etc. neem is also used to remove stomach worm infection, ascariis, tape worm infection. Another one is sweet basil more than 1000 years it is served as a minor ingredient in cookery and medicine. It is used to cure skin infection, loss of smell, insect bite.<sup>[8,9]</sup> Sharma reported that sweet basil is also effective against intestinal parasites the essential oil of this plant is used in aroma therapy.<sup>[10]</sup> In Indian traditional medicine coconut oil is served as a good moisturizer on skin. It also helps to remove skin dryness, psoriasis, and fungal infection. It act well on dry skin and help to retain skin moisture and help from causing skin infection. Using other oils like mineral oil on skin may cause side effects, but coconut oil makes better result as skin moisturizer.<sup>[11]</sup> It is proved that coconut oil can used on skin dryness, flaking of the skin.<sup>[12,13]</sup>

## MATERIALS AND METHODS

### Sample Collection

The leaves of *Salvia rosmarinus*, *Azadirachta indica*, and *Ocimum basilicum* are collected from the local area and identified by comparing with standard herbarium specimens.

### Preparation of Poly herbal Mosquito repellent skin Oil

The herbs used for the present study were washed well with tap water and weighed. 250 g each of the selected plant leaves were weighed and make it into a paste using mortar and pestle. 500 mL of coconut oil measured in a measuring cylinder and transferred to a heating vessel and heated to low flam. The crushed leaf paste of *Salvia rosmarinus*, *Azadirachta indica*, and *Ocimum basilicum* were added to the coconut oil and heated till low flame for 15 min. The oil was cooled down to room temperature

### Poly herbal mosquito repellent skin Oil

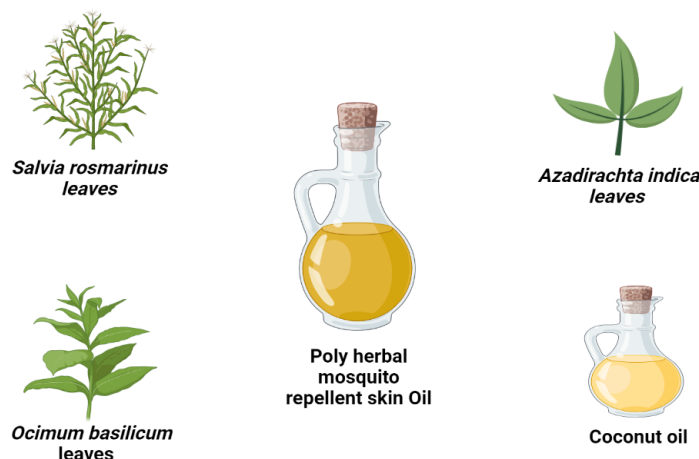


Figure 1: Ingredients of Poly Herbal Mosquito Repellent Skin Oil.

and filtered using a fine mesh filter. The filtered herbal oil was stored in glass bottles.

#### Antibiotic Susceptibility Test (Disk diffusion)

The disk diffusion test is a test of the antibiotic sensitivity of bacteria. Preparation of pathogen broth 24 hr old pathogens like, *Aeromonas hydrophila*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Streptococcus pyogenes* were prepared. Nutrient agar plates were prepared and kept for drying. Pathogens were taken in a sterile swab. Each swab was incorporated separately in separate nutrient agar plate. Sterile disk (6 mm) were placed in the agar plates. Sample (Poly Herbal Skin Oil) were impregnated into the sterile disk, control were mentioned using solvent. Kept for overnight incubation at 37°C. After Overnight the plates were taken and zone of inhibition were measured using ruler.

#### Assessment of mosquito repellent activity of the prepared herbal formulation (Survey method)

The prepared herbal formulation was filled in bottles of 50 mL capacity. 10 people from the Samathanapuram, Vellaram, Tirunelveli Junction, VM Chathram and KTC Nagar (Tirunelveli district, Tamil Nadu, India) locality were selected at random to test the repellent activity of the prepared herbal formulation. Outdoor and indoor field trials were conducted by separately applying the mosquito repellent oil on volunteers' hands and legs. Trials which were carried out for 6 hr each day for 10 days. 10 people per area selected randomly for the study were instructed to apply the herbal oil on either of their hands and were asked to note the mosquito

#### ANTIMICROBIAL ACTIVITY (DISC DIFFUSION TEST)



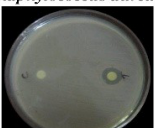

S.NO	BACTERIAL SP	ZONE OF INHIBITION	
		CONTROL	TEST (HERBAL OIL)
1	<i>Aeromonas hydrophila</i> 	-	15
2	<i>Pseudomonas aeruginosa</i> 	-	14
3	<i>Staphylococcus aureus</i> 	-	17
4	<i>Streptococcus pyogenes</i> 	-	19

Figure 2: Antimicrobial activity test of Poly Herbal Skin oil.

biting frequency on both hands and were asked to fill the survey form.

## RESULTS

#### Antimicrobial activity test (Poly Herbal Skin oil)

*In vitro* antimicrobial activity of the Herbal oil was tested against 4 bacterial pathogens using the agar disc diffusion

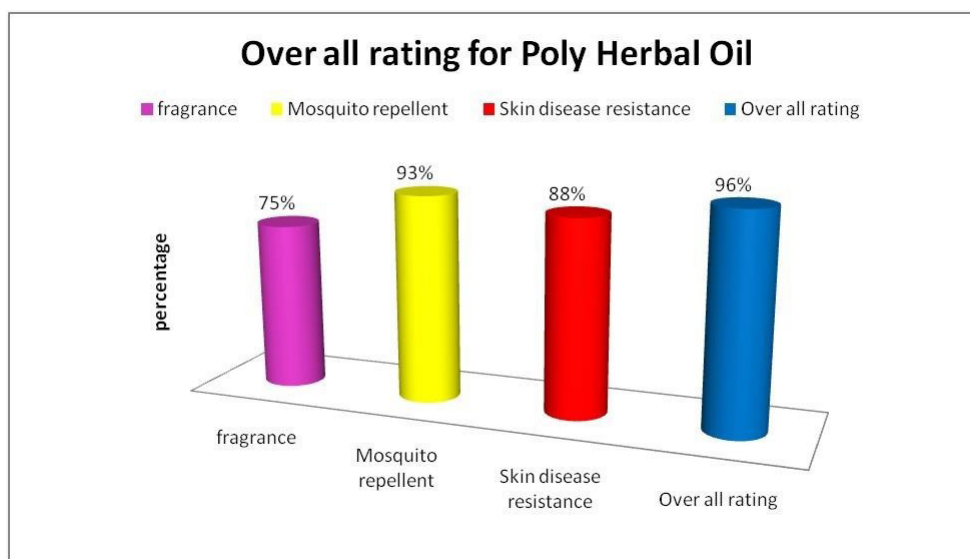


Figure 3: Over all rating of Poly Herbal Skin Oil.

method. The means of the zones of inhibition are shown in Table. Inhibitory effect was detected on four pathogens, Including Two Gram-positive (*Staphylococcus aureus* and *Streptococcus pyogenes*) and Two Gram-negative bacteria (*Aeromonas hydrophila* and *pseudomonas aeruginosa*). The zones of inhibition (in mm) were as follows: The highest inhibition was noted (19 mm) for *Streptococcus pyogenes*. The Medium inhibition was noted (17 mm) for *Staphylococcus aureus* and (15 mm) for *Aeromonas hydrophila*. The lowest inhibition was noted (14 mm) for *Pseudomonas aeruginosa*.

### Survey

The survey forms were collected from the randomly selected people after 10 days. The survey revealed that the herbal formulation is safe to use on skin as none of the people reported any irritation, edema or erythema. About 40% of the surveyed people rated 5 out of 5 for the herbal oil. Another 40% of the surveyed people rated above 4.5 out of 5. The remaining 20% rated above 4 for the herbal oil. From the survey it is evident that the prepared herbal preparation is an effective mosquito repellent and is also 100% safe to skin as topical application.

### DISCUSSION

Nowadays use of plant essential oils as an ingredient in mosquito repellent, such type of herbal insecticide can be served as eco-friendly and better solution for synthetic repellents.<sup>[14]</sup> Unlike synthetic one does not have any harmful effects. This is one of the main reason demand of organic repellents as vector control agents<sup>[15]</sup>

they are mixtures of various compounds with different modes of action and so resistance development can be observed.<sup>[16,17]</sup> Chokechajaroenporn proved that volatile oil of several basil species can act against *Anopheles aegypti* likewise, Guina Bissau reported that fresh *Ocimum canum* (*Ocimum americanum*) can reduce *anopheles mosquitoes* by about 63.6%. Essential oil from rosemary leaf (*Salvia rosmarinus*) can served as a good repellent against *Aedes aegypti* if proved it can work effectively on aedes mosquito power of 92.15% for 4 hr with a formula III, 24% concentration.<sup>[18]</sup> *Azadirachta indica* leaves having bioactive compounds with antimicrobial effects are useful in primary health care.<sup>[19]</sup> Koon and Budida discussed that the active compounds from *Azadirachta indica* contain antimicrobial effects are useful in primary health care. It is also used as traditional medicine in India to cure and inhibit the growth of several pathogens proved by Raja sekar.<sup>[20]</sup> In this study the herbal oil is tested for its antimicrobial activity against pathogens like *Aeromonas hydrophila*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Streptococcus pyogenes*. Why because those pathogens were found to be the reason of many skin infection.<sup>[21]</sup> It is also proved that Neem leaves contain antibacterial activity against *Aeromonas hydrophila*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, and *Streptococcus pyogenes*.<sup>[22]</sup> The anti-bacterial test for this study in disc diffusion method which was already followed by other reference work.<sup>[23]</sup> The phytochemical components of *Salvia rosmarinus*, *Azadirachta indica*, *Ocimum basilicum*, and coconut oil. These components have properties to the repellent activity of mosquitoes. From the results, it is observed that such component mixture herbal oil contain Mosquito repellent property and hence

it can be used as the protection against mosquito borne infection. There are also several research have established many herbal repellants after formulation with a fixative materials results must effective oil report from Songkro who also reported the effect of glucam P-20, vanillin, and fixolide on the mosquito repellent property of citronella oil lotions and found that the lotion containing emulwax and 5% vanillin was the most effective.<sup>[24]</sup> This study reinforced the assumption that ethnomedicinal plants could be a promising alternative resource for therapeutic compounds and mosquito-repellent activity. The results of this study clearly showed that *Salvia rosmarinus*, *Azadirachta indica*, *Ocimum basilicum*, and coconut oil mixed Poly Herbal mosquito repellent skin oil has high potency to control vector mosquitoes. Hence, the results may contribute to a reduction in the application of chemicals in mosquito repellents, which in turn increases the opportunity for natural products to control vector-borne disease. This study also helps other researches to focus on eco- friendly natural base research.

## ACKNOWLEDGEMENT

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## CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

## FUNDING

No funds, grants, or other support was received.

## AVAILABILITY OF DATA AND MATERIALS

The raw/processed data required to reproduce these findings cannot be shared at this time as the data also forms part of an ongoing study.

## SUMMARY

Mosquitos are the major insect that causes many deadly diseases. There are many chemical mosquito repellents seen in our market that cause side effects. This paper focuses on herbal mosquito repellent as well as poly herbal skin oil. Due to worst pollution and reason for emerging several skins infection really affects people's daily life even though usage of many chemical base skin cream, lotion, and ointments it may cause some side effects. For skin disorders, many of our traditional

herbs harbor a wide range of therapeutic properties. In the present study, three herbs *Salvia rosmarinus*, *Azadirachta indica*, and *Ocimum basilicum* are selected to come up with an oil-based herbal formulation to be used as a mosquito repellent and skin disease cure oil. The antibacterial activity of the herbal oil was good against pathogens causing skin infections and the mosquito-repellent efficiency of the poly-herbal oil is assessed using the Survey method. From the survey conducted, it is evident that the prepared herbal preparation is an effective mosquito repellent and is also 100% safe for the skin as a topical application.

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