Natural finishes on textiles to combat the mosquitoes: A pilot study

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Abstract
Mosquitoes are one of the most significant vectors in transmitting diseases like malaria, dengue, chikungunya and many other deadly diseases. Traditionally, many substances have been used in repelling mosquitoes and the most common of them are plant extracts, oils, smoke, etc. Several plants growing in the vicinity may contain essential oils and compounds that are found to be effective insect repellents and are often considered as weeds. The present study focused on the development of eco-friendly, non-hazardous mosquito repellent fabrics using thirteen different plant sources collected based on ethnobotanical review. Fabrics treated using aqueous extracts of thirteen different plant sources namely leaves of holy basil, sweet basil, neem, lemongrass, eucalyptus, custard apple, moringa, betel, wild sage and spearmint, flowers of French marigold and rind of lemon and orange were tested using modified cage test method. Three plant sources namely flowers of french marigold, leaves of sweet basil and eucalyptus were proven to have excellent mosquito repellent property compared to the rest of the tested sources.

Keywords: Mosquito repellents, eco-friendly, mosquitoes, natural sources, modified cage test

1. Introduction
With the rising number of mosquitoes borne diseases, the control of mosquitoes is something of utmost importance in the present day. Specialty products like mosquito repellents stood as the main focus of research these days to combat the nuisance of mosquitoes [1]. Chemical mosquito repellents have a remarkable safety profile in terms of protection, but they are proven to have toxic effects against the human skin and nervous system like eye irritation, rashes on skin and the worse effects even includes breathing problems in children, anaphylactic shock, and low blood pressure [2]. Hence, the researchers from couple of decades were determined on natural mosquito repellents. The act of repelling biting insects with natural sources is not a modern approach. Attempts for prev

2. Materials and Methods
2.1 Selection of plant sources
Most plants contain chemical compounds like repellents, feeding deterrents, toxins, and growth regulators in order to prevent the attack from phytophagous (plant eating) insects [6]. The foremost functions of these compounds is to defend against plant eating insects, many of these compounds are also operative against mosquitoes and other biting Diptera, especially those volatile components released as a consequence of herbivory [7]. Green leaf volatiles were commonly produced by plants when leaves are damaged by herbivores, in order to deter them [8]. Strong responses of mosquito odour receptors to volatiles such as geranyl acetate, citronellal, 6-methyl-5-hepten-2-one and geranyl acetone were experimented by several authors [9]. As a part of this study, wide information on plants having mosquito repellent property was collected from an extensive list of ethno-botanical review. Thirteen plant sources were shortlisted depending on the source of availability and cost effectiveness.
Study was carried out with screened plant sources namely leaves of tulsi (Ocimum tenuiflorum L.), sweet basil (Ocimum basilicum L.), neem (Azadirachta indica), lemon grass (Cymbopogon schoenanthus (L.) Spreng.), eucalyptus (Eucalyptus globulus Labill.), moringa (Moringa oleifera Lam.), custard apple (Annona squamosa L.), betel (Piper betle L.), lantana (Lantana camara L.), mint (Mentha spicata L.), flowers of French marigold (Tagetes Patula L.), peel of orange (Citrus sinensis) and lemon (Citrus limon) (L.). These sources were procured from in and around the Agricultural University campus of Hyderabad in fresh form.

2.2 Selection of Fabric
Cotton was preferred for this research because of its inherent properties like resistance to wear, fading and pilling. 100 per cent plain weave cotton fabric with 122 GSM and 200 thread count was selected for the development of mosquito repellent finishes on textiles.

2.3 Scouring and Desizing
The fabric selected for the study (cotton) was de-sized and subjected to scouring in order to remove the impurities that might hinder the absorption of mosquito repellent finish.

2.4 Extraction
Aqueous extraction was employed using freshly collected leaves, flowers and rind of selected sources. The selected sources were surface cleaned with running tap water, followed by de-ionized water to remove climatic impurities. Thoroughly washed sources were boiled using distilled water at 60°C for 1 to 2 hrs depending on the source with MLR of 1:5. This extract was filtered through what man no. 41 filter paper into conical flask and used for further experiment.

2.5 Fabric treatment
Mosquito repellent finish was applied on cotton fabrics using by simple dip and dry method after soaking the fabric in extraction for 1 hour with M:L:R of 1:7.

2.6 Mosquito Collection
Malaria is transmitted through bites of parasite–infected Anopheles mosquitoes. Anopheles is the major malarial vector in India [10]. Anopheles mosquitoes were identified based on morphological characteristics and were collected during the evening hours with the help of suction tube. They were put in a cage made of nylon net. All mosquitoes were starved of blood and sugar for 4 hours before the tests. A small filter paper wrapped inside the beaker containing water was kept inside the cage overnight.

2.7 Standard test method for mosquito repellency
Modified cage test method was employed for testing the mosquito repellent efficacy of treated samples. Cage test is the quick and cost-effective way to determine the mosquito repelling properties of treated materials [11]. A cage of 30×30×30 cm made out of acrylic was developed for testing the repelling rate percentage of treated samples. The room temperature was maintained at 25°C ± 2°C and 60 to 70 per cent humidity. For conducting the experiment, the opposite facets of the cage were hung with control sample and the other two sides with mosquito repellent treated sample. The bottom of the cage was even half filled with mosquito repellent fabric sample and half with control. Twenty mosquitoes were released into the cage and allowed to settle for 2 minutes. Mosquito were deprived of all the nutrition and water for a minimum of 4 hours before exposure. Laboratory tests were performed during daylight hours only [12,13]. The anti-mosquito effectiveness was noted by counting the number of mosquitoes that rested on the untreated and treated samples at 15, 30, 45 and 60-minutes intervals and repellence percentage was calculated using the formula

Mosquito Repellency (%) = \frac{\text{Number of specimens on controlled fabric}}{\text{Total exposed specimen in cage}} \times 100

Mosquito repellent percentages were analysed after each test.

3. Results and Discussion
3.1 Assessment of Mosquito Repellent Property
The treated fabrics were tested for mosquito repellent efficacy against untreated fabrics using modified WHO cage test method [14], while exposing 20 mosquitoes in cage. The mosquitoes were allowed to settle for two minutes. After 2 minutes, the number of mosquitoes resting on the treated and untreated fabrics were counted and notes at 15, 30, 45 and 60-minutes interval. The mosquito repellent percentages were calculated for each sample using the above formulae. The results of the study revealed that the treated fabrics using Tagetes patula (flowers) extracts displayed highest mosquito repellency with 90 per cent followed by Eucalyptus globulus (leaves) with 85 per cent efficacy and 80 per cent efficacy by Ocimum basilicum (leaves). The samples treated with lemon grass leaf extracts when tested, displayed 75 percent mosquito repellent efficacy followed by the samples treated with holy basil leaf and orange rind extracts. The leaf extracts of the wild sage and neem exhibited a mosquito repellent percentage of 65 whereas the samples treated with the extracts of Moringa oleifera leaves and Citrus limon rind showcased mosquito repellent percentage of 60 respectively as shown in figure 1.0 to 4.0 and table 1.0 respectively. The other sources in order of their mosquito repellency were spearmint leaf extracts, betel leaf extracts and custard apple leaf extracts with a mosquito repellent percentage of less than 50 and were reported to have lowest mosquito repellency among the sources.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Fabrics treated plant extracts</th>
<th>Common name</th>
<th>Plant Parts used for extraction</th>
<th>Mosquitoes repelled (nos.)</th>
<th>Percentage repellency (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Ocimum tenuiflorum L.</td>
<td>Tulsi/Holy basil</td>
<td>Leaves</td>
<td>14</td>
<td>70</td>
</tr>
<tr>
<td>2.</td>
<td>Ocimum basilicum L.</td>
<td>Great basil</td>
<td>Leaves</td>
<td>16</td>
<td>80</td>
</tr>
<tr>
<td>3.</td>
<td>Azadirachta indica</td>
<td>Neem</td>
<td>Leaves</td>
<td>13</td>
<td>65</td>
</tr>
<tr>
<td>4.</td>
<td>Cymbopogon schoenanthus (L.) Spreng</td>
<td>Lemon grass</td>
<td>Leaves</td>
<td>15</td>
<td>75</td>
</tr>
<tr>
<td>5.</td>
<td>Eucalyptus globulus Labill.</td>
<td>Eucalyptus</td>
<td>Leaves</td>
<td>17</td>
<td>85</td>
</tr>
<tr>
<td>6.</td>
<td>Moringa oleifera Lam.</td>
<td>Drumstick</td>
<td>Leaves</td>
<td>12</td>
<td>60</td>
</tr>
<tr>
<td>7.</td>
<td>Annona squamosa L.</td>
<td>Custard apple</td>
<td>Leaves</td>
<td>6</td>
<td>30</td>
</tr>
<tr>
<td>No.</td>
<td>Plant Source</td>
<td>Part</td>
<td>Repellent Percentage</td>
<td></td>
<td></td>
</tr>
<tr>
<td>-----</td>
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<tr>
<td>8</td>
<td><em>Piper betle</em> L.</td>
<td>Betel</td>
<td>70</td>
<td></td>
<td></td>
</tr>
<tr>
<td>9</td>
<td><em>Lantana camara</em> L.</td>
<td>Wild-sage</td>
<td>80</td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td><em>Mentha spicata</em> L.</td>
<td>Spearmint</td>
<td>65</td>
<td></td>
<td></td>
</tr>
<tr>
<td>11</td>
<td><em>Tagetes Patula</em> L.</td>
<td>Flowers</td>
<td>40</td>
<td></td>
<td></td>
</tr>
<tr>
<td>12</td>
<td><em>Citrus sinensis</em></td>
<td>Rind</td>
<td>13</td>
<td></td>
<td></td>
</tr>
<tr>
<td>13</td>
<td><em>Citrus limon</em> (L.)</td>
<td>Lemon</td>
<td>18</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*= n = no. of mosquitoes exposed, *= Time of observation: 1 hr.

**Fig 1:** Mosquito repellent efficacy of plant sources

**Fig 2:** Fabric treated with *Ocimum basilicum* leaf extract

**Fig 3:** Fabric treated with *Eucalyptus globulus* leaf extract

**Fig 4:** Fabric treated with *Tagetes Patula* flower extract
The results were supported with the results of phytochemical screening of the aqueous Ocimum basilicum extract which revealed the presence of saponins, tannins, cardiac glycosides that are responsible for the mosquito repellency [15]. According to another study, plant sources contain certain kind of phytochemicals like linoleic acid in Ocimum basilicum L, alkaloids, flavonoids, tannins, and saponins in Tagetes and Eucalytus leaves which were proven to act on the mosquito odour receptors thus helping in the mosquito repellency [16]. Therefore, on the basis of mosquito repellent percentages, three fabrics treated using Tagetes patula flowers, Eucalyptus globulus and Ocimum basilicum leaf extracts were reported to have highest mosquito repellent efficacy among the tested sources hence, can be used to protect the human being from the bites of mosquitoes and thereby promising safety from mosquito borne diseases.

4. Conclusion
In today’s world, the growing number of mosquito-borne diseases necessitates the discovery of new forms of mosquito repellents, in both synthetic and natural forms. In recent years, the trend is shifting towards the development of mosquito repellent finished textiles that provides a much-necessitated feature of repelling mosquitoes through use of natural active agents derived from plant extracts. The present study focused on “Development of eco-friendly mosquito repellent textiles”. The developed fabrics were assessed for mosquito repellent efficacy. From an extensive list of sources prepared based on ethnotanical review, thirteen sources were selected and tested for mosquito repellent efficacy using modified cage test method. It was evident from the test results that Tagetes patula (flowers) extracts displayed highest repellence with 90 per cent followed by Eucalyptus globulus (leaves) with 85 per cent efficacy and 80 per cent efficacy by Ocimum basilicum (leaves). This study proved that mosquito repellent finishes can be given to the textile materials in order to provide external protection in the form of door curtains, bed sheets, table and sofa covers, to prevent the mosquito bites.

5. References