FORMULATION AND EVALUATION OF MOSQUITO REPELLENT SPRAY

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ABSTRACT
Mosquitoes are the blood sucking insects afflicting human beings and several mosquito species belonging to genera Anopheles, Culex and Aedes are responsible for mosquito-borne diseases. Therefore, the control of mosquitoes is an important public health concern around the world. As most of the synthetic mosquito repellent products and devices available in the market are reported to have harmful effects on human beings, the objective of the present study is to formulate effective plant-based mosquito repellent products with potent mosquito repellent activity with less side effects. The herbal mosquito repellent spray containing citronella as main mosquito repellent agent with tulsi essential oil and solvent ethanol and distilled water, emulsified with aid of Tween-80 is a formulation which spare its role in protecting the subjects from varieties of mosquitoes by providing the repellency layer over the skin of application with no skin irritancy were as bio-efficacy was tested by rubbing volunteer’s forearm with 1 ml test solution was exposed to open space of environment and to the areas between the lakes, where blood-seeking mosquitoes are found and the number of mosquitoes that aligned on the lower leg was recorded in each 10 minutes of interval for 1 hour. Analysis was carried out as control group treated with ethanol, standard group treated with marketed odomos cream and test group treated with formulated herbal mosquito repellent spray and mosquito repellent activities of test group were found to be effective as similar as of marketed formulation of odomos, as per the determined percentage protection (%P).

KEYWORDS: Mosquito repellent, Citronella, Spray, Emulsion, Essential Oil, %Protection.
INTRODUCTION

Cosmetic is defined as per the “Drugs and Cosmetics Act, 1940 as, any article intended to be rubbed, poured, sprinkled or sprayed on, or introduced into, or otherwise applied to, the human body or any part for cleansing, beautifying, promoting attractiveness or altering the appearance, and includes any article intended for use as a component of cosmetic.

Mosquitoes are the blood sucking insects afflicting human beings and several mosquito species belonging to genera Anopheles, Culex and Aedes are responsible for mosquito-borne diseases such as malaria, chikungunya, yellow fever, rift valley fever, dengue fever, and filariasis Japanese encephalitis producing significant morbidity and mortality in humans and livestock. According to one World Health Organization (WHO) report in 1990, there were approximately 270 million cases of malaria and approximately one million deaths annually. Therefore, the control of mosquitoes is an important public health concern around the world.

As most of the synthetic mosquito repellent products and devices available in the market are reported to have harmful effects on human beings, the objective of the present study is to formulate effective plant-based mosquito repellent products with potent mosquito repellent activity with less side effects. The herbal mosquito repellent spray containing citronella as main mosquito repellent agent with tulsi essential oil and solvent ethanol and distilled water, emulsified with aid of Tween-80 is a formulation which spare its role in protecting the subjects from varieties of mosquitoes by providing the repellency layer over the skin of application with no skin irritancy were as bio-efficacy was tested by rubbing volunteer’s forearm with 1 ml test solution was exposed to open space of environment and to the areas between the lakes, where blood-seeking mosquitoes are found and the number of mosquitoes that aligned on the lower leg was recorded in each 10 minutes interval for 1 hour. Analysis was carried out as control group treated with ethanol, standard group treated with marketed odomos cream and test group treated with formulated herbal mosquito repellent spray and mosquito repellent activities of test group were found to be effective as similar as of marketed formulation of odomos, as per the determined percentage protection (%P).

Benefits of herbal mosquito repellant

- Safe for pregnant women.
- As effective as chemical repellent.
- Useful for those who are vulnerable towards chemical repellants.
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Fig. 1

1. Citronella

✓ Characteristics of citronella:

- Citronella oil is an essential oil obtained from the leaves and stems of different species of *Cymbopogon nardus* (lemongrass)
- The citronella belongs to the family Poaceae.
- It is widely cultivated in Southeast Asia and grown commercial in India.
- Its Characteristics includes a tall, tufted perennial, clump-forming tropical grass with narrow leaf blades. They grow to a height of 5–6 ft, with greyish green, flat, leaves about 3 ft long, and 1 inch wide.

✓ Extraction method:

Citronella oil was extracted from *Citronella nardus* by steam distillation method which extract oil from semi-dried leaves by 4 hours of process the oil and water were separated.
Citronella essential oil

Citronella oil has a slightly sweet, lemony smell. It is pale greenish yellow in colour.

- **Properties of citronella essential oil**
  - **Color:** Cranny Yellow.
  - **Odour:** Characteristic.
  - **Solubility:** Insoluble in water.

- **Chemical constituents:**
The main chemical components of citronella oil are citronellic acid, geraniol, nerol, citral, borneol, camphene, citronellol, citronellal, dipentene, and limonene. It consists about 3.0% limonene; 35.3% citronellal; 12.0% citronellol, 24.9 % geraniol, 4.3% citronellyl acetate, 6.3% geranyl acetate, and 0.8% linalool.

Thulsi essential oil

- **Properties of thulsi essential oil:**
  - Moisturizes and nourishes skin.
  - Promotes healthier cells.
  - Removes dead skin cells.
  - Great soothing agent.
  - Brightens skin.
  - Even though it acts as a scrub, it doesn’t take away the essential minerals from the skin.
  - Ensure it leaves behind no rashes but just healthy glowing skin.
  - Anti-aging effect is also identified skin pores, makes skin firmer.
  - Promotes blood circulation to skin.
  - Removes scar marks and blemishes.
  - Deep cleanses the skin leaving it refreshed.
Phytoconstituent of tulsi essential oil

*Ocimum sanctum* has antioxidant such as cirsilineol, circimaritin, isothymucin, apigenin and rosameric acid and eugenol. Among 0.7% of volatile oil it has 71% eugenol and 20% methyl eugenol. It comprises of two flavonoids orientin and andvicenin.

Extraction method

The essential oil of tulsi were extracted by hydro-distillation by using Clevenger’s type apparatus. Two hundred gram of dried tulsi leaves were put in a 5 liter round-bottomed flask and were filled with distilled water and electrically charged mantle which separated oil in condenser after 4 to 5 hours of heating.

3. **Tween-80:**

Properties of tween-80:

- It is apoloxethyethylene (80) sorbitan mono-oleate a non-ionic surfactant.
- Its soluble in ethanol
- It acts as emulsifier and also get categorized as plasticizer and wetting agent
- It has antioxidant properties.
- Good diffusant agent.
4. Propyl paraben
   ✓ Properties of propyl paraben
      ▪ It prevents germ growth.
      ▪ It has antioxidant.
      ▪ It is antibacterial and anti-inflammatory properties, which may help calm and repair skin.

   ![Fig. 7](image)

   ➢ How to use mosquito repellant spray:

   1. Take the mosquito repellent, turn up cap from bottle.

   2. Before applying on skin, check the direction on label content of formulation and apply.

   3. Apply on skin by actuating 2 to 3 times on the area which get exposed to environment.

   4. Gently massage on the skin where you sprayed on skin in a circular motion for promoting its penetration through skin.

   5. In case of exposure to mosquitoes re-apply the preparation after 8 hours of previous application or after washing.
Literature review on the methods of evaluating mosquito repellency effect of formulation

- Field experiments using the human landing catch method.

In the field, the repellents were evaluated at locations that differ in their ecology. The areas usually display a relatively high abundance of mosquitoes. During the study, areas were neither treated with insecticides nor were they subject to any other vector control measure. The efficacy of the repellent formulations were assessed by using the human landing catch (HLC) method, observations were made hourly for 30 minutes over 6 hours. In the experiments, the 18 study participants were split into 3 groups of 6. Each group tested the 2 formulations, 15% PMD and 15% DEET, and the negative control containing 70% ethanol. Over 3 consecutive days each person received each treatment once and 2 participants tested the same treatment on a given day. The sequence of treatment allocation was organised in a Williams balanced Latin Square design to minimise any bias due to first-order carryover effects. In preparation of the field experiments, the test surface (i.e. the bare lower leg) of the study participants were washed with neutral soap, rinsed, dried and swabbed with Arixtra wipes containing 70% isopropanol. One of the lower legs was then treated with either one of the two repellent formulations or the negative control. Sixty minutes after application of the treatment, the study participants were assigned to one of 6 positions that were set at least 20 m apart, as recommended in the WHO guidelines, in order to avoid bias due to competition in attractiveness to the mosquitoes. With the exception of the treated lower leg the whole body was fully protected from mosquito bites by a white jump suit, a bee keeper’s hat and latex gloves through which mosquitoes could not bite. During an exposure period of 30 minutes the study participants sat on a stool and collected any mosquito alighting on the exposed lower leg using a mouth aspirator. Collected insects were transferred to 50 ml Falcon tubes covered with mesh. An acoustic signal from a horn indicated the participants the start and the
end of the 30 minutes exposure period. Following the exposure the study participants retreated from their position for 30 minutes before they moved to the next position, repeating the process above until they had 6 exposures over 6 hours. As with the treatment allocation, the sequence of rotation between the positions followed a Williams balanced Latin Square design. In addition to the mosquito collections by the study participants, mosquito traps were set in the study area to measure overall mosquito presence. The traps ran during the same 6 hours the study participants tested the repellents, yet the traps were set at least 20 metres away in order to avoid unwanted attraction of mosquitoes away from, or towards to, the study participants. All collected mosquitoes were identified with the aid of a stereo microscope using the identification keys. If it was not possible to identify a mosquito specimen on the basis of morphological characteristics, for example when it was damaged or a member of a species complex, the specimen was processed and sent for molecular identification. The molecular identification method used is based on matrix assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS), an emerging methodology for the identification of arthropods.

- **Laboratory Experiments Using The Arm-In-Cage Test**

The laboratory experiments were conducted by following the WHO guidelines for the arm-in-cage (AIC). In the AIC test, the protection time of a repellent is assessed by exposing a treated forearm to hungry mosquitoes at regular intervals. The cages measured 40 cm × 40 cm × 40 cm and were made of clear acrylic glass sides with an opening on the front side. At the bottom of the cage was a mirror positioned allowing for observation of mosquitoes landing on the lower side of the arm. The back side of the cage was made of a fine metal grid to ensure air supply during the experiments. The test cages contained 200 host seeking 5 to
10-day-old females of one of the 3 mosquito species; *Aedes aegypti, Anopheles stephensi or Culex quinquefasciatus*, species are WHO recommended model organisms. Adult mosquitoes were fed with 10% sucrose solution and water. Testing and rearing conditions for all mosquito colonies were 26.8 ± 1.2 °C (mean ± SD) and 64.8% ± 7.2% relative humidity (mean ± SD) and a 12:12 (light:dark) photoperiod. Male and female mosquitoes were kept in the same rearing cages to allow mating to occur. Experiments with Ae. aegypti were performed under artificial light at an intensity of 639 Lux, while tests with An. stephensi and Cx. quinquefasciatus were conducted under subdued light at an intensity of 49 Lux to mimic the conditions according to the mosquito species diurnal biting patterns. Twelve hours before the experiment, the sugar water was removed from the cage and the mosquitoes had only access to water. Before exposure to the mosquitoes the forearm was washed with odourless soap, dried and swabbed with an Arixtra isopropanol wipe and then dried again. Then, to assess the readiness of the mosquitoes to land, the forearm of a study participant was exposed in the experimental cage for 60 seconds or until 10 landings were counted. A landing was defined as a mosquito alighting on the skin and remaining for at least 2 seconds. After measuring the landing activity with the untreated forearm the forearm was treated from wrist to elbow with either 15% PMD. Thirty minutes after application of the repellent the participant exposed the treated forearm in the test cage for 3 minutes or until 10 mosquitoes landed. The procedure was then repeated every 30 minutes over 6 hours. The duration until the first, second and tenth landing of a mosquito on the treated forearm was noted. At the end of the experiment the arm was again washed and dried as before, and a second control measurement of the mosquitoes’ landing activity was taken. Data analysis. Raw data recorded on paper forms were entered into a Microsoft Excel 2010 spreadsheet. Statistical analysis was performed in the open source package R version 3.4.35 and graphs were produced with the R package. The endpoint measured in the experiments was the number of mosquitoes landing on the bare skin during each exposure period. Based on the number of landings and exposure times two outcome measures were estimated following the WHO guidelines: the complete protection time (CPT) and the percentage protection (%p) over time. Here, CPT is defined as the time elapsed between the application of the repellent and the first mosquito landing. Average CPTs (median and 95% confidence interval; 95% CI) over all study participants were estimated based on a Kaplan-Meier survival analysis implemented in the R package and compared between the two repellent formulations using the Mantel-Haenszel test, including a stratification term for study participants. %p was calculated as the
reduction in landings by the treatment when compared to the negative control over all exposures using equation: \(\%P = \frac{C-T}{T}\)

Where T is the average number of mosquitoes landing on the surface per second in a test and C is the average number of mosquitoes per second landing on the skin surface treated with the negative control in the field experiment or the untreated forearm in the AIC test. Landing rates were estimated on the basis of generalised linear models (GLM) with a negative binomial distribution and a log link function.

- **Materials and Method of preparation of herbal mosquito repellant spray**

  **Materials:**

  The citronella essential oil and tulsi essential oil used in this project were purchased from local market Aroma Pharmaceuticals, Solapur. The other chemicals, including (Tween-80, Ethanol, Distilledwater) used for formulation of mosquito repellant were lab graded and obtained from college laboratory as listed in formulation table.

  **Method:**

  **Step 1:** -

  Required quantity of ingredients were weighed. By the aid of MORTAR and PESTLE the trituration of essential oil with ethanol was done with continuously stirring.

  **Step 2:** -

  At room temperature the Tween-80 was mixed with the distilled water and it was gradually added to above mixture with continuous stirring.

  **Step 3:** -

  Then few drops of rose oil were added to above herbal mosquito repellant and its volume is make up with distilled water up to 10 ml

  **Step 4:** -

  Packed in a suitable container with proper labelling.
Formulation table of herbal mosquito repellant

<table>
<thead>
<tr>
<th>Sr No.</th>
<th>Ingredients (quantity)</th>
<th>F1 50 ml</th>
<th>F2 50 ml</th>
<th>F3 50 ml</th>
<th>Uses</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Citronella</td>
<td>1.25</td>
<td>1.25</td>
<td>1.25</td>
<td>Mosquito repellent agent</td>
</tr>
<tr>
<td>2</td>
<td>Tulsi essential oil</td>
<td>0.375</td>
<td>0.375</td>
<td>0.375</td>
<td>Anti-septic agent</td>
</tr>
<tr>
<td>3</td>
<td>Ethanol</td>
<td>12.5</td>
<td>12.5</td>
<td>12.5</td>
<td>Solvent</td>
</tr>
<tr>
<td>4</td>
<td>Tween-80</td>
<td>3.75</td>
<td>3.75</td>
<td>3.75</td>
<td>Emulsifier</td>
</tr>
<tr>
<td></td>
<td>Ingredient</td>
<td>Quantity</td>
<td>Quantity</td>
<td>Quantity</td>
<td>Category</td>
</tr>
<tr>
<td>----</td>
<td>---------------------</td>
<td>----------</td>
<td>----------</td>
<td>----------</td>
<td>-----------------</td>
</tr>
<tr>
<td>5</td>
<td>Rose oil</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>Perfumery agent</td>
</tr>
<tr>
<td>6</td>
<td>Distilled water</td>
<td>Qs</td>
<td>Qs</td>
<td>Qs</td>
<td>Vehicle</td>
</tr>
<tr>
<td>7</td>
<td>Propyl paraben</td>
<td>2.5</td>
<td>2.5</td>
<td>2.5</td>
<td>Preservative</td>
</tr>
</tbody>
</table>

**Label of formulation**

- **Evaluation parameters**
  - **A) Physico-chemical parameters:**
    1. Appearance
    2. Viscosity
    3. pH
    4. Phase separation
    5. Wash ability
    6. Spray Angle
    7. Stability Testing
    8. Drying Time
    9. Amount emitted per actuation
  
- **B) Biological evaluation parameters:**
  1. Mosquito Repellency Test
  2. Skin Irritation Test
Evaluation of Physico-chemical properties of herbal mosquito spray:
The prepared herbal mosquito spray formulation is evaluated for various specifications.

1) Appearance:
The prepared herbal mosquito repellent was evaluated for its odour and color. The prepared formulations were inspected visually.

2) Viscosity:
The prepared formulation was measured for its viscosity by using lab scale calibrated BROOKFIELD VISCOMETER

3) pH of the formulation:
The PH of aqueous solution of formulation was measured by using pH paper.

4) Phase separation:
The formulated herbal mosquito repellant was kept intact in a closed container at 25- 300C not exposed to light. Phase separation was observed carefully for every 24hr for 30days.

5) Wash ability:
Little quantity of mosquito repellant was applied over the skin and washed with water.

6) Measurement of spray angle:
The sprays were actuated in horizontal direction onto activated form of TLC aluminium sheet polyamide 11 F -254 mounted at distance of 15 cm from the nozzle. The diameter of the circle were observed and measured under UV light and Spray angle (Θ) = tan -1 (l / r) where l is the distance of sheet from nozzle, and r is the average radius of the circle.

7) Stability testing:
The 3 batches of formulation was made and the pH was recorded on the day of formulation prepared and after the interval of 3 days it was measured and it was observed that there was no any significant increment or either decrement in the pH of formulation by aid of pH paper which was analyzed for up to 2 weeks. It was carried out in triplicate and average values taken.

8) Drying time:
Evaporation time is the time required for the spray to dry and it was estimated by spraying the formulation on filter paper and noting down the drying time in minutes.

9) Amount sprayed per actuation:
There was no significant difference in the amount emitted per each actuation indicating the effectiveness of spray system in delivering reproducible amount of the formulation per each actuation.
C) Biological parameters:

1) Mosquito repellency test
2) Skin irritation test

- Mosquito repellency test

In the open space of environment that is in the field, the repellents were evaluated at two locations in Solapur that differ in their ecology; the Killa Garden a restored park area in the Lucky Chowk and in the Kambarr Talav, area around the lake. Both areas had relatively high abundance of mosquitoes, while the killa garden also shown a variety of mosquito species diversity,. During the study, both areas were neither treated with insecticides nor any other vector control measure.

The efficacy of the repellent formulations was first assessed in the Kambarr Talav area between 12th April and 14th April 2023 and then in the Killa Garden between 18th and 20th May 23, 2023. By visual inspection the observations were made hourly with 10 minutes of time interval.

In the experiments, the 9 study participants were split into 3 groups of 3. Each group tested the 3 formulations, Standard Odomos Cream (Standard group), Lab graded Ethanol (Control Group) and Citronella herbal spray (Test Group). In preparation of the field experiments, the test surface (i.e. the bare lower leg) of the study participants were washed with neutral soap, rinsed, dried and swabbed with, herbal mosquito repellant spray(test), marketed odomos preparation (standard), lab graded ethanol (control). With the exception of the treated lower leg the whole body was fully protected from mosquito bites by a full sleeves suit, a hat and hand gloves. During an exposure period of 10 minutes the study participants were in the field found mosquito on the exposed lower leg which was treated with the control ethanol solution. Whereas the lower leg which was treated with the test herbal mosquito repellent spray and was not found to be attractive for mosquitoes.

Based on the number of landings the outcome measures were estimated following the WHO guidelines: the percentage protection (%p) over time.

%p was calculated as the reduction in landings by the treatment when compared to the control group over all exposures with in 1 hour of 10 minutes of time interval using following equation;
\[
\frac{P = C - T \times 100}{C}
\]

where \( T \) is the average number of mosquitoes landing on the surface per second in a test and \( C \) is the average number of mosquitoes per minute landing on the skin surface treated with the negative control in the field.

Fig. 7: Study participants in field of experiment.
Fig. 8: Participant with standard application.
Fig. 9: Participant with test application. Fig. 10: Participant with control application.

- **Skin irritation test**

Small amount of herbal mosquito repellant was sprayed on the skin kept for few minutes and it was non-irritated.
RESULT AND DISCUSSION

The herbal mosquito repellant was prepared and evaluated. The herbal mosquito repellant was prepared in the laboratory was found to be compared with various parameters such as Appearance, pH, Phase separation, Viscosity, Irritancy, Washability, Spray angle measurement, and was found to be satisfied with all required characterization. The bio-efficiency was evaluated and compared with the marketed mosquito repellent formulation and comparison was made on the basis of %P(Percentage protection) value. On the basis of outcomes the developed formulation can be used as an effectively against mosquitoes, with 100% protection.

Physical evaluations results

Fig. 11: pH of F1, F2, F3.  
Fig. 12: pH range.

pH measurement of 3 batches was found to be:

Table No. 1

<table>
<thead>
<tr>
<th></th>
<th>F1</th>
<th>F2</th>
<th>F3</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>5.8</td>
<td>6</td>
<td>5.8</td>
</tr>
</tbody>
</table>

Fig. 3: Radius value of herbal spray.
Spray angle of 3 batches was found to be:
Table No. 2

<table>
<thead>
<tr>
<th></th>
<th>F1</th>
<th>F2</th>
<th>F3</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.</td>
<td>83.9 deg</td>
<td>83.9 deg</td>
<td>83.9 deg</td>
</tr>
</tbody>
</table>

![Fig. 14: Brookfield viscometer.](image)

Viscosity was found to be all 3 batches
Table No. 3

<table>
<thead>
<tr>
<th></th>
<th>F1</th>
<th>F2</th>
<th>F3</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.</td>
<td>18cps</td>
<td>19cps</td>
<td>18.5cps</td>
</tr>
</tbody>
</table>

![Fig. 15: Drying results.](image)

The drying time was as follows:
Table No. 4

<table>
<thead>
<tr>
<th></th>
<th>F1</th>
<th>F2</th>
<th>F3</th>
</tr>
</thead>
<tbody>
<tr>
<td>No.</td>
<td>2.17 min</td>
<td>2.02 min</td>
<td>1.55 min</td>
</tr>
</tbody>
</table>

- Biological efficacy testing results

Lower-leg treated with ethanol: Presence of mosquito
Fig. 16 and Fig. 17: Lower-leg treated with ethanol: Presence of mosquito.

Lower-leg treated with Herbal Mosquito Repellent Spray: Absence of mosquito

Fig. 18: Lower-leg treated with Herbal Mosquito Repellent Spray: Absence of mosquito.

Application of standard; Marketed formulation of odomos cream

Fig. 19 and Fig. 20: Application of standard; Marketed formulation of odomos cream.
Table No. 5: (%P of Control group of Ethanol and Test group of herbal mosquito repellent spray).

<table>
<thead>
<tr>
<th>Sr. No</th>
<th>Time interval (min)</th>
<th>F1 C</th>
<th>T</th>
<th>%p</th>
<th>F2 C</th>
<th>T</th>
<th>%p</th>
<th>F3 C</th>
<th>T</th>
<th>%p</th>
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<tr>
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<td>2</td>
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<td>0</td>
<td>100</td>
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<td>0</td>
<td>100</td>
</tr>
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<td>2.</td>
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<td>100</td>
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<td>0</td>
<td>100</td>
<td>1</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td>3.</td>
<td>30</td>
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<td>0</td>
<td>100</td>
<td>2</td>
<td>0</td>
<td>100</td>
<td>2</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td>4.</td>
<td>40</td>
<td>3</td>
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<td>100</td>
<td>1</td>
<td>0</td>
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<td>0</td>
<td>100</td>
</tr>
<tr>
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<td>1</td>
<td>0</td>
<td>100</td>
<td>1</td>
<td>0</td>
<td>100</td>
<td>2</td>
<td>0</td>
<td>100</td>
</tr>
<tr>
<td>6.</td>
<td>60</td>
<td>2</td>
<td>0</td>
<td>100</td>
<td>2</td>
<td>0</td>
<td>100</td>
<td>1</td>
<td>0</td>
<td>100</td>
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</table>

Table No. 6: (%P of standard marketed odomos cream).

<table>
<thead>
<tr>
<th>Sr. no.</th>
<th>Time interval</th>
<th>Standard group of marketed formulation odomos cream</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>C</td>
<td>S</td>
</tr>
<tr>
<td>1.</td>
<td>1</td>
<td>10</td>
</tr>
<tr>
<td>2.</td>
<td>2</td>
<td>20</td>
</tr>
<tr>
<td>3.</td>
<td>3</td>
<td>30</td>
</tr>
<tr>
<td>4.</td>
<td>4</td>
<td>40</td>
</tr>
<tr>
<td>5.</td>
<td>5</td>
<td>50</td>
</tr>
<tr>
<td>6.</td>
<td>6</td>
<td>60</td>
</tr>
</tbody>
</table>

DISCUSSION

The %P (percentage protection) of standard and test solution was found to be 100 %. On the basis of the above outcome, the formulated herbal mosquito repellent spray was equally effective in the treatment as the marketed preparation odomos cream.

Review on the prepared herbal mosquito repellent spray formulation

1. Name-Ahraz khan:
Review- I have applied your (herbal mosquito repellent spray). I found it as non-irritant and it is better to use spray rather than creams because, I found oily texture after use of mosquito repellent cream but, your spray seems to be nice.

2. Name-Onkar bhingoli:
Review- I have applied your product, it seems to be quite effective and non-irritant.

3. Name-Rehan mujjawar:
Review- I have applied your herbal mosquito repellent spray. It is non-irritant and it is effective.
4. Name- Laxmikant javalkote:
Review- I have used your (herbal mosquito repellent spray). It gave better result. It is non-irritant and it is best to use against various species of mosquitoes.

5. Sanket swami:
Review- I have used your formulated herbal mosquito repellent spray which was found to be more effective than other marketed mosquito repellent formulation.

➢ CONCLUSION

Table No. 7: Summary of all evaluation parameters results.

<table>
<thead>
<tr>
<th>Sr. no.</th>
<th>Evaluation Parameter</th>
<th>F1</th>
<th>F2</th>
<th>F3</th>
<th>STD</th>
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<tr>
<td>1</td>
<td>Appearance Test;</td>
<td></td>
<td></td>
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<tr>
<td></td>
<td>: Color : Odour</td>
<td>Whitish Characteristic</td>
<td>Whitish Characteristic</td>
<td>Whitish Characteristic</td>
<td>Whitish Pungent</td>
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<tr>
<td>2</td>
<td>Drying Time</td>
<td>2:17 min</td>
<td>2:02 min</td>
<td>1:55 min</td>
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</tr>
<tr>
<td>3</td>
<td>pH of the formulation</td>
<td>5.8</td>
<td>6</td>
<td>5.8</td>
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<tr>
<td>4</td>
<td>Phase separation</td>
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<tr>
<td>5</td>
<td>Spray Angle Test</td>
<td>83 degree</td>
<td>83 degree</td>
<td>83 degree</td>
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</tr>
<tr>
<td>6</td>
<td>Irritancy</td>
<td>Non irritant</td>
<td>Non irritant</td>
<td>No irritant</td>
<td>No irritant</td>
</tr>
<tr>
<td>7</td>
<td>Wash ability</td>
<td>Easily washable</td>
<td>Easily washable</td>
<td>Easily washable</td>
<td>No easily washable</td>
</tr>
<tr>
<td>8</td>
<td>Actuation Of Spray</td>
<td>Uniform</td>
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<td>Uniform</td>
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<tr>
<td>9</td>
<td>Viscosity</td>
<td>18 cps</td>
<td>19 cps</td>
<td>18.5 cps</td>
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<tr>
<td>10</td>
<td>%P(Percentage protection)</td>
<td>100%</td>
<td>100%</td>
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</table>

➢ CONCLUSION

The present study was attempted to make an herbal mosquito repellant. The prepared herbal mosquito repellant was studied with various parameters such as appearance, pH, phase separation, viscosity, skin- irritancy, wash ability, Spray angle and it was found to be satisfied with all required specification. The bio-efficacy was determined and was compared with the marketed mosquito repellant formulation odomos cream. It was found that the formulated herbal mosquito repellant spray was equally effective as odomos cream. Also, on the basis of %P (percentage protection) value the developed herbal mosquito repellant formulation can be used as effective against mosquitoes. But as the marketed odomos mosquito repellent cream is chemical based formulation (N,N-Diethyl benzamide) it has concern side effects. The
present herbal formulation has all naturally derived ingredients. Thus, they had no side effects or fewer side effect.

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REFERENCES


