Review Article

Mosquito Repellent Textiles and their Evaluation: A Review

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Abstract
Mosquitoes pose a major public health risk by spreading many infectious and life-threatening diseases to millions of citizens living in tropical climates. They can spread diseases like Dengue, Malaria, Chikungunya, Filariasis, and also Zika, which kill millions of people each year. Therefore, it has become a necessity and a need of the hour to prevent mosquito-borne diseases. Protective clothing is the most effective and easiest means of protection against biting insects since it covers the majority of the human body. This paper discusses the pros and cons of several types of mosquito repellents. The various types of popular techniques like direct, nanoemulsion, and microencapsulation of incorporating the mosquito repellent agents into the textile substrates are also discussed. This paper also highlights the evaluation methods of mosquito repellency on textiles.

Keywords: *Essential oils, Microencapsulation, Mosquito-borne diseases, Mosquito repellents*

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1. Introduction

The vast majority of people have been bitten by a mosquito at some or the other point in their lives. Mosquitoes are really tiny in size but their power of spreading illness into the lives of humans should not be under-estimated. This is rightly believed that monsoon and mosquitoes have a deadly affair. Though every year, people rejoice at the advent of monsoon season but there lies a terrible part of steeping in of those uninvited guests; the threatening and ghastly mosquitoes. Mosquitoes serve no purpose other than to annoy humans by buzzing around at night, causing itchy swelling. However, these mini mosquito bites can infect humans with countless illnesses and transmit serious diseases which are sometimes incurable and can result in long-term problems or even death. Many tropical countries, including India, are infested with mosquito-borne diseases that have become a serious public health issue. As a result, keeping a check on their breeding is crucial, as "early discovery is the greatest defence".

Anopheles, Aedes, and Culex species of mosquitoes are vectors for dreadful diseases like Dengue, Malaria, and Chikungunya. The diseases caused by a different genus of mosquitoes are mentioned in **Table 1** [1].

Table 1- The diseases caused by a different genus of mosquitoes

Vector	Genus of Mosquito	Disease caused	Type of Pathogen
Mosquito	Culex *Culex quinquefasciatus*	Japanese encephalitis	Virus
		Lymphatic filariasis	Parasite
		West Nile fever	Virus
	Aedes *Aedes aegypti*	Chikungunya	Virus
		Dengue	Virus
		Lymphatic filariasis	Parasite
		Rift Valley fever	Virus
		Yellow Fever	Virus
		Zika	Virus

	Anopheles	Lymphatic filariasis	Parasite
	Anopheles stephensi	Malaria	Parasite

Mosquitoes have a range of sensors that can detect the presence of their prey, including;

- **Chemical Sensors**

Mosquitoes get attracted to humans because of the carbon dioxide and lactic acid found in our perspiration. Mosquitoes have chemoreceptors in their antennae that detect the odour of sweat [2].

- **Heat Sensors**

Mosquitoes are drawn to their hosts when they are warm. It has been postulated that mosquitoes react to changes in temperature and host thermal cues by combining their signals from these two sensilla [2].

- **Visual Sensors**

Mosquitoes are generally led toward their hosts by visual cues. Adult mosquitos have compound eyes that are susceptible to light intensity variations. Photoreceptors of night-biting mosquitos, such as Anopheles gambiae, are said to adjust to variable light intensity by modulating rhodopsin levels. In low-light situations, this could improve visual responsiveness to a possible host [2].

It is rightly known that prevention leads to protection, therefore, mosquito control and self-protection against mosquito bites are the most efficient strategies now being implemented to tackle the increasing number of mosquito-borne diseases. Mosquito management strategies include environmental modification, biological control, physical control, and chemical control. One of the strategies for preventing mosquito-borne diseases is to stop disease transmission by eradicating or stopping mosquitos from attacking humans. As a result, distinctive products such as **"mosquito repellents"** are being utilized to fight mosquitoes.

- **Mosquito Repellents**

A mosquito or any insect repellent is a material that is applied to the skin, apparel, or other surfaces to prevent mosquitos from settling on them [3].

Mosquito repellents include active ingredients that block mosquitos' chemosensory, thermal, and optical senses, causing them to repel.

The mechanism of repellent substances can be divided into two categories: those that work on the 'olfactory senses' (Transpiration Repelling) and others that work on 'tactile senses' (Direct Contact Repelling) [4].

i. Transpiration Repelling

Transpiration repelling refers to a repellent's influence on the sense of smell. This has the advantage of deterring insects without causing them to strike or die against a repellent-treated surface. The repellant chemical limits insect humidity sensory receptors by suppressing the function of detecting moisture, making humans undetectable to insects [4].

ii. Direct Contact Repelling

Direct-contact repelling is the action of a repellent that stimulates the sensation of touch, and it pushes insects away from the treated surface before blood-sucking [4].

- **Mosquito repellents applied as topical solutions**

Historically, the use of repellents to protect individuals from mosquito bites has been recognized as part of a larger integrated insect-borne diseases control strategy. Various natural and synthetic compounds have been found to be effective insect repellents. Repellents are practical and cost-effective tools for preventing mosquito-borne diseases. As a result, personal protective measures against mosquito bites must be performed. Therefore, this enabled the development of advanced and more effective mosquito repellents which are applied as a topical application on the skin in the form of creams, lotions, sprays, etc.

Several studies reported using natural and synthetic mosquito repellents in the form of sprays, candles, coils, sticks, and other topical applications including creams and lotions which are mentioned in **Table 2**.

Table 2: Recent studies reporting the use of natural and synthetic mosquito repellents for application in different dosage forms

Name of Ingredient/s (Essential oils/extracts/synthetic components)	Mosquito species	Dosage form	References no.
Curcumin, Embelin, Lemon Oil, Eucalyptus Oil, Citronella Oil	Field test	Cream, Candle	[3], [5]
Sweet Orange (Citrus Sinensis), Lime (Citrus Aurantifolia), Lemon (Citrus Limon)	Camponotus nearcticus (Carpenter ants)	Topical	[6]
Curcuma longa, Eucalyptus globulus, Citrus aurantium, Bourbon geranium, cedar wood, clove, peppermint, and thyme	Aedes aegypti and Anopheles dirus	Topical	[7], [9]
Azadirachta indica oil (10-40% v/v)	Anopheles gambiae	Topical	[8]
N, N-Diethylphenylacetamide (DEPA, 20%), Essential oils (Catnip, Citronella, Basil, Chamomile, Galbanum, Rosemary, Juniper, Jasmine, Litsea, Cinnamon- 20% each)	Aedes aegypti	Topical	[10]
Eucalyptus Oil DEET (5%, 10%, 15%)	Culicidae	Topical	[11]
Nepeta cataria (catnip) plants and Isolated Nepetalactone Isomers (10%)	Aedes aegypti	Topical	[12]
Clove oil, Olive oil, Coconut oil, Citronella oil, Lemongrass oil, Ginger, Eucalyptus, Orange, Sweet Basil, Ylang-Ylang oil (0.1ml each)	Aedes aegypti, Anopheles dirus and Culex quinquefasciatus	Topical	[13]
DEET, Citronella Oil, Fennel Oil	Culex pipiens, Aedes togoi, and Aedes albopictus	Topical	[14]
Pine Oil	An. culicifacies and Cx. Quinquefasciatus	Topical	[15]

According to several studies, most repellents in the form of lotions, creams, essential oils, sprays, or solutions need a direct application to the human skin, which many people dislike due to the scent, skin itching, breathing problems, and other factors.

Repellents such as lotions, coils, and creams have limited effectiveness and durability; therefore they have to be re-applied or replaced frequently. When applied to human skin, these formulations and lotions can cause irritation and skin dryness [16]. This has necessitated the advancement of mosquito repellent clothing.

- **Mosquito Repellent Textiles**

Mosquito repellent clothing aids in the protection of humans from mosquito-borne diseases [24]. They are commonly found in the form of nets, home textile fabrics, curtains, apparel, and other products. Mosquito repellent textiles are a novel approach in the protective textiles domain to evolve the textile field by delivering much-needed properties such as mosquito repellency, which is especially important in tropical locations. Therefore, mosquito repellent clothing is an effective approach to protect humans from serious mosquito-borne diseases.

The mosquito repellent textiles are developed by introducing natural or synthetic components into the substrates Permethrin, a pyrethroid-based insecticide employed in military and army uniforms and nets. Synthetic repellents have great durability but have limitations due to their toxic effects on humans. The world is shifting towards herbal products that are eco-friendly. Therefore, a large number of plants and their parts such as eucalyptus, lemongrass, neem, clove, and many more are also assessed for their mosquito repellent characteristics. These plant parts and their essential oils are abundant in nature, are non-irritant and non-toxic to the skin, and can be used for skin application directly [25].

- **Methods of Incorporating Mosquito Repellent Agents into Textiles**

According to the literature review and analysis of findings, there are many methods for incorporating mosquito repellent agents into fabrics which can be broadly categorized into the direct application through the padding, formation of nanoemulsion, and microencapsulation.

a. Direct Application Method

The direct application includes the pad–dry–cure approach which is the most typical application method for easy care. Before the crosslinking reaction takes place during the curing step, the crosslinking reactant, emulsifier, core material, and other components are dried on the cloth.

The advantages of this method include its easy application, less time-consuming, and cheaper as compared to microencapsulation while the biggest disadvantage is the poor durability of the finish. The finish lasts for a maximum of one or two washes.

Several studies have reported the use of a pad-dry-cure method for applying mosquito repellent finish on fabrics. The mosquito repellency of modified DEET (N, N-Diethyl-3-methylbenzamide) was tested using the conventional method of pad-dry-cure. The treated textiles showed over 90% repellency against Aedes mosquitoes while between 70 and 80% was observed for 10 times washed treated fabrics [17]. Researchers [18] developed mosquito repellent textiles utilizing the herbal extract of tulsi, neem, and mint leaves using pad-dry and exhaust methods which exhibited 90% repellency in case of neem and mint while 70% in case of tulsi leaves extract until 9 washes. Another researcher [19] utilized the different methods of the pad-dry process to provide mosquito repellency to cotton fabrics using castor oil. In this experiment, the mosquito repellent property of the samples treated with 6% binder and 2% oil was efficient, indicating that the Pad-Batch-Dry (PBD) method should be used; however, the washed samples had low mosquito repellency.

b. Nano-Emulsion Method

Nano-emulsions can be made using a **low-energy emulsification** approach that promotes the generation of ultra-small droplets based on phase behaviour and characteristics. Self-emulsification, phase transition and phase inversion temperature emulsification procedures are examples of low-energy processes. However, **high-energy emulsification** using high-pressure homogenizers, high shear stirring, and ultrasound generators, is commonly utilized due to its simplicity of large-scale manufacturing and low cost, the most popular technology. Its application in the food, cosmetics, and pharmaceutical industries is most common due to its excellent transparency, stability, and low viscosity. A nano-emulsion is a dispersed system with uniform small droplet sizes ranging from 20 to 500 nanometres. Because of their small size, nano-emulsions frequently have better long-term physical stability than regular emulsions. Using a high-speed homogenizer and moderate surfactant concentrations, an essential-oil-loaded nano-emulsion system can be created [20].

Among the studies reported, two studies have used the nano-emulsion technique for encapsulating core material into textile substrates. According to [21] nylon net fabrics finished with chrysanthemum oil nano-emulsions using a layer-by-layer technique showed 100% mosquito repellency until 25 washes along with a 90% mortality rate. In the research [22] nano-emulsions of DEPA (N, N-diethyl phenylacetamide) were developed and compared with Bulk-DEPA for mosquito repellency on the Culex quinquefasciatus mosquito species. The results showed that Nano-DEPA has better mosquito repellency than Bulk-DEPA even at lower concentrations. As reported by [23], fabrics treated with nanoparticles loaded with Vitex Negundo leaf extract showed 100% mosquito repellency until 15 washes.

c. Microencapsulation Method

One of the primary drawbacks of using natural or synthetic ingredients for mosquito repellent finishes is the lack of permanence. Most mosquito repellents get removed after washing because they have no attraction for textiles or are not permanently attached to them. One of the popular approaches for trapping active compounds is microencapsulation. Microencapsulation is a protective technique that encapsulates solid, liquid, or gas components into tiny particles with a diameter of 1–1000 m. It is widely utilized in pharmaceuticals, cosmetology, foods, textiles, and advanced materials. The core material is totally coated and insulated from the external environment, which is a unique advantage of microencapsulation. More importantly, if the right shell material and preparation procedure are used, microencapsulation will have no effect on the properties of the core components. As a result, microencapsulation is a great way to make thermochromic mixtures more stable [24].

Microencapsulation has the significant advantage of entirely coating and isolating the core material from external conditions. More importantly, if the right shell material and preparation procedure are used, microencapsulation will have no effect on the attributes of core materials. As a result, microencapsulation is an excellent way to increase the stability of thermochromic compositions. However, the disadvantage of microencapsulation is the material cost. The formulation process may be more expensive than typical formulations, and reproducibility may be compromised. In reaction to heat, hydrolysis, or biological agents, the influence of the polymer matrix, polymer additives, and their degradation products on the environment varies dramatically. Also, the stability of the core particle is modified by changes in process variables such as temperature, pH, solvent addition, and solvent evaporation.

Most of the recent studies reporting the microencapsulation of essential oils for mosquito repellency in textiles are mentioned in Table 3.

Table 3- Recent studies reporting the microencapsulation of essential oils for mosquito repellency in textiles

Name of Ingredient/s (Essential oils/extracts/synthetic components)	Mosquito species	Type of technique	Type of Fabric	Repellency Percentage	Protection time (hours/washes)	Reference no.
DEET (1%, 5%)	Aedes aegypti	Pad-dry-cure method	Cotton	70-80%	10 washes	[17]
Neem, tulsi, Mint leaves	Anopheles	Formulation (Pad-Dry-Cure)	Cotton (Woven and Knitted)	90% (Neem, Mint) 70% (Tulsi)	9 washes	[18]
Castor Oil (2%)	Field Mosquitoes	Pad-Dry-Cure	Cotton	2% oil and 6% binder gave better repellency	0 washes	[19]
Chrysanthemum oil (10g)	Laboratory-reared mosquitoes	Nano emulsion using Layer by Layer technique	Nylon net fabrics	100%	25 washes	[21]
Diethylphenylacetamide (Bulk-DEPA) (6.47% v/v)	Culex quinquefasciatus	Polymerization followed by Phase Inversion Temperature (PIT) Emulsification method	Cotton	-	-	[22]
Vitex Negundo leaf extract (0.06%, w/v)	Anopheles	Nanoparticles using Ionic Gelation Method	Cotton	100%	15 washes	[23]
Lemongrass Oil (30ml)	Anopheles	Microencapsulation- Ionic	Polyester	92%	15 washes	[25]

		Gelation Method				
Thyme oil, Cypress oil and Grapefruit oil (2:1:1 (50%: 25%: 25%))	-	Microencapsulation (Ionic gelation)	Bamboo/Tencel 50:50 blended fabric	60% (with Moringa oleifera)	30 washes	[26]
Citronella Oil (5:1 w/w oil to polymer ratio), (4:1 oil to wall materials ratio)	Aedes aegypti	Microencapsulation (Complex Coacervation)	Cotton	>90%, Citronella-80% Citriodiol-95%	Up to 21 days, 2 weeks 30 days	[27], [29]
Lemongrass (10ml)	Anopheles	Microencapsulation	Knitted Cotton	88-90%	20 washes	[28]
Eucalyptus Rosemary (10%, 30%, 50%)	Aedes, Anopheles and Culex	Microencapsulation	Cotton	Eucalyptus-100% Rosemary-40-49%	Eucalyptus- 15 wash cycles Rosemary-decreased	[30]

A variety of procedures and techniques have been devised and adapted to make microcapsules with desired materials and target qualities. However, not all processes are appropriate for textile applications. Methods of making microcapsules for textiles are listed in **Figure 1** [31]:

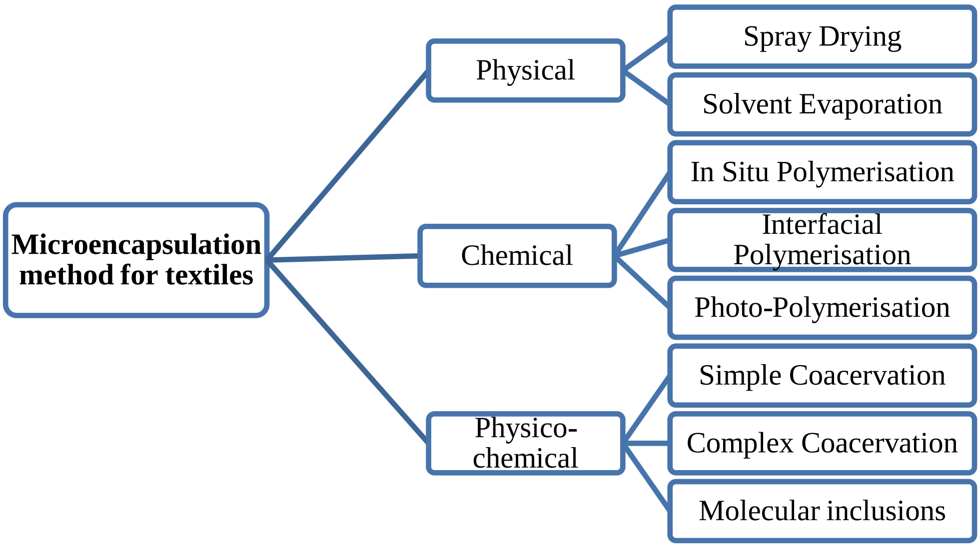

Figure1- Microencapsulation methods used in Textiles

Several studies reported the use of different techniques of microencapsulation for finishing textile substrates. Researcher [25] used ionic gelation method (coacervation) of microencapsulation using lemongrass oil for polyester fabric which exhibited 92% repellency until 15 washes. In another research [26], three shell materials, namely sodium alginate, Acacia arabica and Moringa oleifera gum and three core materials like thyme oil, cypress oil and grapefruit oils in combination of 2:1:1 and were microencapsulated and found that fabrics finished with Moringa oleifera showed maximum repellency of 60% until 30 washes. According to [27], microencapsulated citronella oil using complex coacervation technique demonstrated 90% repellency on cotton fabrics until 21 days.

In the study [28], microcapsules of lemongrass oil were prepared using the coacervation technique and found 88-90% repellency until 20 washes. A study conducted by [29], compared mosquito repellency of citronella and citridiol using complex coacervation technique and found that citronella oil gave 80% repellency for 2 weeks while citridiol showed 95% repellency for 30 days. In the study [22], nanoparticles of DEPA were prepared using polymerization followed by the Phase Inversion Temperature (PIT) emulsification technique. The Nano-DEPA was compared to Bulk-DEPA and found that Nano-DEPA indicated improved mosquito repellency. Another researcher [30] analysed the mosquito repellent efficacy of eucalyptus and rosemary microcapsules and found that eucalyptus indicated 100% repellency until 15 wash cycles while rosemary showed 40-49% repellency which decreased with washing.

2. Methods of Evaluating Mosquito Repellency on Textiles

i. Cage Test

It's also known as the Arm-in-Cage test, and it's a standard test method for determining mosquito repellent efficacy in treated and untreated substrates. The illustration of the cage test is mentioned in **Figure 2** [32]. The mosquitoes employed in the test must be pathogen-free because the human subjects involved in the test must be prevented from any infection.

According to WHO norms, the cage must be loaded with 200 mosquitoes that have been deprived overnight and only fed sucrose solution. The usage of fewer mosquitoes in the cage (as few as 30 mosquitoes) has been updated because reduced density gives more accuracy and better reflects the average biting environment seen during most indoor and outdoor activities, as well as providing a comfortable atmosphere for volunteers. Volunteers should not smoke and should refrain from using scent or repellent products for the duration of the study. This variable may modify a person's appeal to mosquitoes, affecting the repellency test's outcome. The volunteer's hand must be cleansed with fragrance-free soap and rinsed with water. The participants' arms, protected in gloves or treated materials is put into the cage.

The left arm is used as a control, whereas the right arm is as a treated sample. Both forearms with treated and untreated components are subjected to the mosquito population for 3 minutes at the same time. The test continues if at least two mosquitos land or bite within the first three minutes. If no mosquitos land in 3 minutes, the hand is withdrawn from the cage. The exposition is repeated every 30 minutes for the next 8 hours or until the repellency disappears. Each test sample is performed in triplicate at 28±2 °C and 80±5% RH, with a 5-minute interval between duplicates.

The number of mosquitoes on the samples and the number of bites on the control are used to determine the effectiveness of the samples.

$$\% \text{ Mosquito protection: } (U-T) / U \times 100$$

Where 'U' represents the number of mosquitoes on control samples or untreated samples and 'T' corresponds to the number of mosquitoes on treated samples [32].
As per the studies reported, four studies [29], [19], [17], [30] used cage testing in their mosquito repellent investigation.

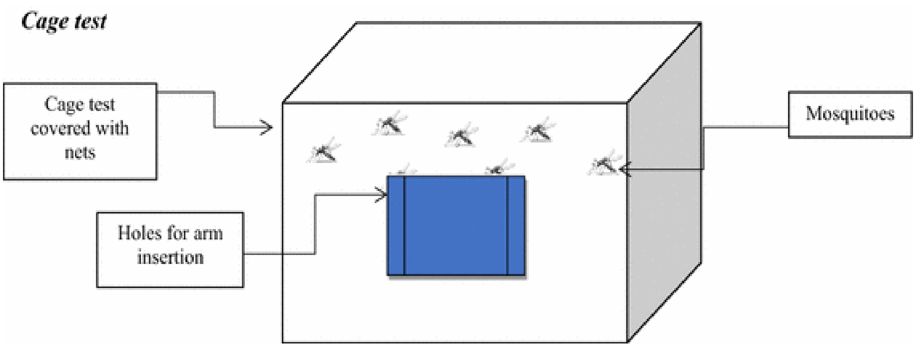

Figure 2: Cage Test

ii. Cone Bioassays Test

This is a standard test method of assessing the toxicity of finished mosquito repellent substrates as shown in **Figure 3** [32]. The test follows the WHO-approved Cone Bioassays procedure. A masking tape is used to adhere to the standard WHO plastic cone on top of the sample's treated surface. The aspirator is utilised to blow five to ten female mosquitoes into the cone, and the mosquitoes are exposed to the treated surface. The low mosquito density used in this strategy allows for simple monitoring of mosquito behaviour. Within three minutes of exposure, the number of mosquitos resting on the treated samples is counted.

$$\% \text{ Mosquito mortality: } (MR - MC) / (100 - C) \times 100$$

In the above formula, 'MR' corresponds to the mosquito's mortality in test replicate while the 'MC' represents the mosquito's mortality in control samples [32].
Only one study [22] used Cone Bioassays test for assessing the toxicity of the mosquito repellent (DEPA).

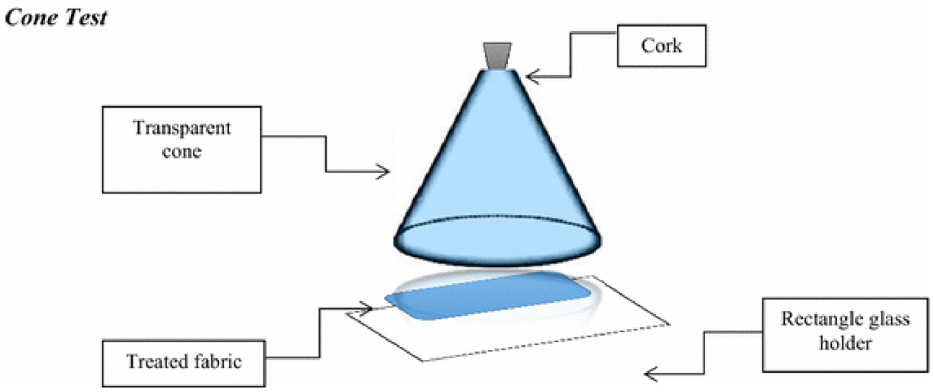

Figure 3: Cone Test

iii. Excito Chamber Test

The excito chamber technique is a modified bespoke approach for observing mosquito behaviour change as illustrated in **Figure 4** [32]. The box has one front and exit panel, both of which are occupied by a single escape portal. The screened inner chamber, glass holding frame, and door cover are all part of the construction. The mosquitos are starved for at least 4 hours before the test, preferably overnight. The behaviour of mosquitoes is studied in terms of the number of escaped mosquitoes to another space and the number of insects remaining inside the treated product chamber. After 10 and 30 minutes of exposure, the results are recorded. The experiment was carried out in broad daylight and was repeated four times.

The following formula is used to compute the percentage of mosquito repellency:

$$\% \text{ Mosquito repellency: } (NES+NDE)/\,(NEX) \times 100$$

where 'NES' represents to the number of mosquitoes escaped, while the 'NDE' corresponds to the number of mosquitoes dead and 'NEX' represents the number of mosquitoes exposed.

Among the studies reported six studies [25], [21], [23], [26], [18], [28] conducted excito chamber test method for testing the mosquito repellence.

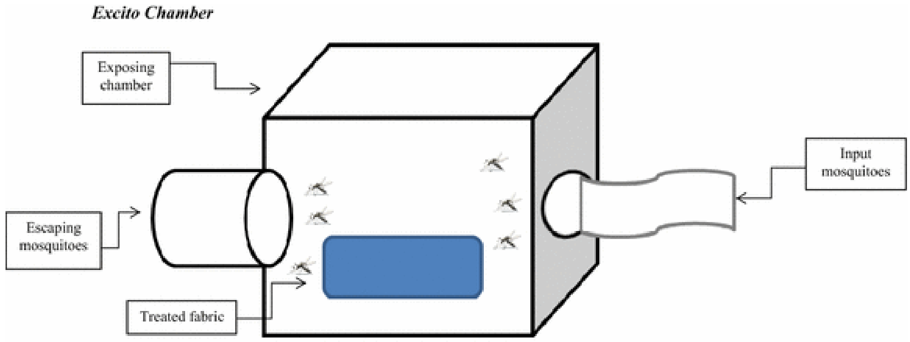

Figure 4: Excito Chamber Test

3. Conclusion

There are no vaccines to prevent mosquito and tick-borne infectious diseases apart from few exceptions and the only approaches to prevent disease transmission are to use insect repellents, and insecticides. Since mosquito-borne diseases are one of the major concerns, clothing can act as a physical barrier between human skin and the blood-sucking mosquitoes, preventing the transmission of diseases caused by mosquitoes. The findings of this study revealed that essential oils and extracts from certain plants have strong repellent action against various species of mosquitoes. The researchers have been looking for novel natural repellents, and certain plants and their essential oils have shown to be very effective. It also demonstrated the numerous approaches for imparting repellent compounds into the textile substrates, with microencapsulation and nano-emulsions being the most effective methods due to their increased durability and effectiveness. The method of mosquito repellency assessment used to perform the efficacy of the impregnated cloth are also summarised in this review. As a result, if appropriate materials with a suitable repellent ingredient and proper techniques of imparting mosquito repellency are practiced, it can give rise to new domain of protective textiles in apparel industry. Therefore, this review urges for the consideration and emphasis of the researchers working in the field of mosquito-borne diseases and protective textiles to acknowledge the potential involvement of plant-derived repellents in disease prevention and developing new domain of mosquito repellent textiles which has less influence on human health and the ecosystem and use techniques of applying the mosquito-repellent agents to the textiles.

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