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Activity of Polyherbal Formulated Mosquito Liquid Vaporizer Against *Culex quinquefasciatus*

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Abstract: This is the first report on larvicidal and repellent practises in polyherbal repellent (PHR) formulation BIHER01 (Bharath Institute for Higher Education and Research 01) and its components. Our findings showed that polyherbal (POLH) extracts are promising and noteworthy larvicides for *Culex quinquefasciatus* which can be linked to polyphenols such as flavonoids, phenols, tannins, alkaloids, saponins, terpenoids, tri-terpenoids, glycosides, cardiac glycosides, protein, carbohydrates, acids and steroids. Based on the time periods used, either a synergist or an antagonistic effect of the phytoconstituents was observed against larvae. On the other hand, owing to its high potential for repellence, PHR was known as an active repellent for *Culex quinquefasciatus*. Nevertheless, further studies are necessary to establish the contribution to mode of action and repellence of the major and minor elements of POLH extract.

Keywords: Polyherbal repellent, BIHER01, *Culex quinquefasciatus*, Phytochemical analysis, *Azadirachta indica*, Eucalyptus, *Curcuma longa*

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Introduction

In the tropics, mosquito (MQ)-borne disease, including malaria, yellow fever, Chikungunya, Filariasis, Japanese Encephalitis, Kala azar and dengue fever (DEN-1, DEN-2, DEN-3 and DEN-4), pose a significant danger to over 2 billion people. MQ bites can also induce allergic reactions to local skin, including local and systemic reactions including urticaria and angioedema (Benelli, 2016; Lee *et al.*, 2018; Tjaden *et al.*, 2018; Franklin *et*

al., 2019; Cameron and Ramesh, 2021). MQ's can vector multiple viruses and parasites that cause disease (Dahmana and Mediannikov, 2020). These species are carried by the infected mosquitoes from person to person without any symptoms (Massaro *et al.*, 2019). Dengue fever is DENV-14's fastest growing spore of antiviral infection (Jain and Kumar, 2018; Ludwig *et al.*, 2019; Lowe *et al.*, 2020). Home remedies such as Neem (*Azadirachta*

indica) leaves and Papaya (*Carica papaya*) juice are used for treating dengue fever (Uppala *et al.*, 2017; Hina and Rizwani, 2019; Ghani *et al.*, 2019; Dave *et al.*, 2020; Mohammadi *et al.*, 2021). Dengue has been the most significant infection of arboviruses (Nunkoo and Mahomoodally, 2012). Natural repellent produced no side effects (Pattanayak *et al.*, 2010; Benelli *et al.*, 2018). The above-mentioned reports inspired us to work with herbal repellent. Moreover, in public health, the use of various components of plants is popular. A cost efficient, beneficial way of dealing with illnesses is the use of natural cures and herbal medicines (Karunamoorthi and Tsehay, 2012; Yuan *et al.*, 2016; Chaudhari *et al.*, 2020).

In the treatment of multiple chronic disorders, POLH medicines play a key role. In ethanomedics as well as in the conventional medicines scheme worldwide, several POLH formulations are used (Sergon *et al.*, 2008; Coll *et al.*, 2015; Narang *et al.*, 2017; El-Naggar *et al.*, 2018). POLH formulation acts as the useful rational approach of researchers (Geethangili and Tzeng, 2011). POLH formulas also made a major effort to combat MQ-borne infections in their prehistoric formulation (Zhang *et al.*, 2011; Ashok *et al.*, 2018). While they are separately assessed for their repellent activity, four herbs are combined with little experimental research. The aim of this study is to test *Acorus calamus* powder, the eucalyptus oil (*Eucalyptus obliqua*), the turmeric powder (*Curcuma longa*) and the Neem oil (*Azadirachta indica*) as MQ liquid vaporizer for the use of the POLH formulation.

Materials and Methods

BIHER01 (Bharath Institute of Higher Education and Research 01) formulation of polyherbal repellent (PHR):

Specimens of the *C. quinquefasciatus* mosquitoes larvae were collected from and around Paruthipattu, Chennai and Tamil Nadu, India. The larvae were transferred into a 500 ml vessel and are fed glucose and yeast powder as a food at ratio of 3:2. The test box was used for emergencies with 100 adult pupae MQ's with the MQ net closed to

the open end of the test box. The MQ's are gathered from the attraction of used socks and cloths held under the net. Field assessment repellent POLH extract activity was performed in empty space. The different herbal formulations were procured from Thirumullaivoyal herbal shop such as *A. calamus* powder (Fig. 1A), *Eucalyptus* oil (*E. obliqua*) (Fig. 1B) and turmeric powder (*C. longa*) (Fig. 1D). From each different herbal powder, 10 g of *A. calamus* powder and *C. longa* powder were mixed with 50 ml of Eucalyptus oil and 50 ml of neem oil (*A. indica*) (Fig. 1C) and the mixtures were allowed to blend for 24 h and then the blended POLH extract was filtered using Whatman No.1 filter paper. It is easy for local people to follow this approach and is cheap. The preparation of the polyherb extract was then transformed into appropriate liquid vaporizers for commercial use, as shown in Figure 1E. The regulation consisted of three replicates of the same concentration. For control group, instead of POLH extracts, potable tap water blended with glucose was used. The MQ replacement device is attached to the electric vaporizer and is then held in the laboratory, and the electric vaporizer is kept next to the test box. At 6 h and 12 h the mortality rate was recorded.

Qualitative phytochemical analysis of PHR:

Qualitative phytochemical profiling of PHR was done according the method of Ashok *et al.* (2018).

Results and Discussion

The mortality rate for 6 h and 12 h was 30% (laboratory) and 70% (open field), respectively. No deaths were recorded in the control group. These investigations show that pesticides dependent on PHR do not quickly result in death, but can lead to certain activity changes. The poly-extractor biological activity could be caused by active polyphenolic compounds in *A. calamus*, *Eucalyptus*, *C. longa* and *A. indicica*. The PHR vaporizer begins to release PHR vapours that prevent MQ's from flying and falls down in 20 min, but did not kill mosquitoes. At the end of 25 min of PHR treatment some MQ's found to be active. In



Fig. 1: PHR formulation.

open area tests, the result shows that the MQ's are repelled within 1 h and certain improvements in the MQ's, and that some mosquitoes exhibit swirling activity over a short time. The results show that direct interaction with mosquitoes with PHR vapours increases deaths as treatment time increases (Fig. 2).

The pesticides dependent on PHR do not quickly knock insects can be a benefit if it is used as an alternative pesticide without side effects for the prevention of vector-borne diseases and it is environmentally friendly. Future repellent marks a departure since it can be used in the time release scheme that puts active ingredients. The principal action of poly-herbal extract has been observed to deter the host from landing or biting vectors. POLH extract has shown promising insecticide efficacy against *C. quinquefasciatus* MQ's in this study. Different extraction techniques have limits and benefits of their own. Essential oils, on the other hand, are intensely concentrated compounds derived from plant parts. Oils such as *Eucalyptus* oil and neem oil are also used for their flavour and therapy or odour. Essential oils of volatile ingredients with small components contain strong scent which causes mosquito repellence and inhibit bleeding insects' orientation (Benelli *et al.*, 2017; Orchard and van

Vuuren, 2017; Maggi and Benelli, 2018; Pavela *et al.*, 2018). Various considerations such as MQ-species, consistency of POLH extract and essential oils blending may be due to the difference in research results, i.e. the attractiveness to mosquitoes (Ganjewala, 2009).

PHR are determined by several influences, such as plant types, growing conditions, plant maturity, plant storage, essential oil storage plant preparation and extraction methods (Michaelakis *et al.*, 2009; Dua *et al.*, 2010; Maciel *et al.*, 2010). Further research is required to elucidate the effectiveness of the PHR formulation for a broad spectrum of mosquito born diseases, as well as classify active compounds for MQ repellents for use in the preparation of commercial products to be used as MQ repellents if necessary. Major phytochemical compounds present in PHR namely flavonoids, phenols, tannins, alkaloids, saponins, terpenoids, triterpenoids, glycosides, cardiac glycosides, protein, carbohydrates, acids and steroids has potential MQ larvicides and repellents for *C. quinquefasciatus* (Abdul *et al.*, 2018; Alharbi *et al.*, 2018; Biswas *et al.*, 2020).

Figure 3 shows the MQ repellent refilling (PHR). The liquid is used in a plastic container, a POLH mixture, and these compounds are

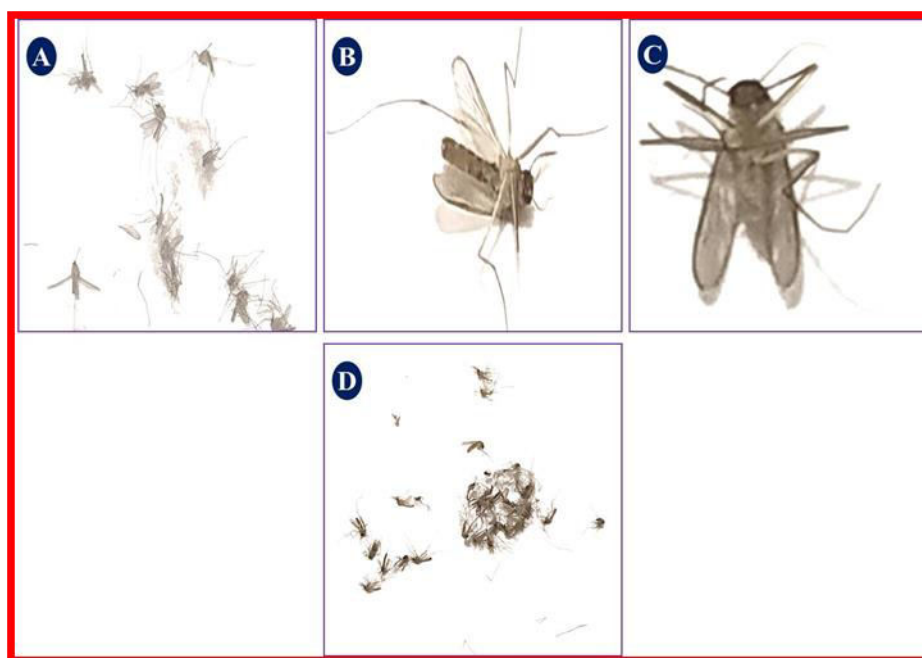


Fig. 2: A- MQ's death in laboratory; B and C- MQ's death in empty room; and D-MQ's death when exposed to PHR vapours.

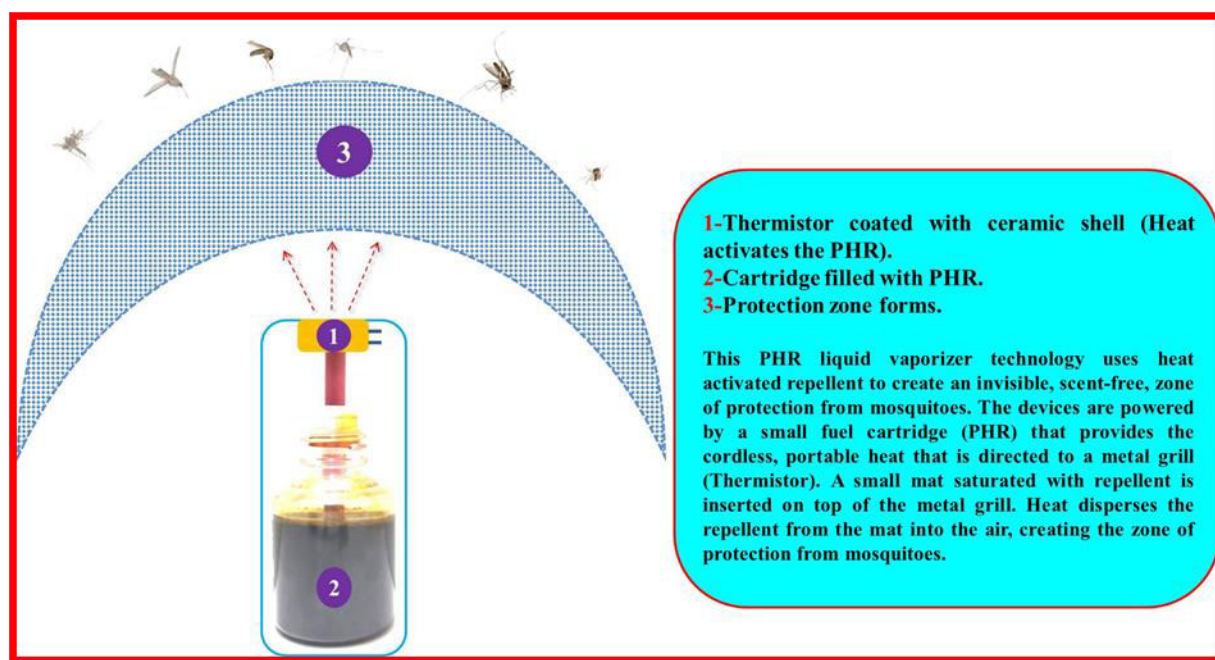


Fig. 3: Possible mechanism caused by liquid vaporizer when it is fuelled by PHR.

dissolved in essential oils to be readily vaporised under temperature. The combination is nontoxic and does not induce a range of reactions like dizziness, headache, allergies to the eyes, inflammation and other fitness. The most

significant component of the combination is the poly-herbal combination. It enters the skin of the MQ's and causes paralysis, which slows MQ flight, thus destroying the nervous system. The wick is the only way the liquid gets through the repulsive

vaporising system. The wick collects the PHR and the top section heats up when put into the pump. It splashes the fluid and spreads it into the room.

The findings of physicochemical and phytochemical analysis can be slightly different for various reasons. It may be attributed to various geographic factors, environmental conditions, cultivation and harvest time, plant age, collection methods, etc. The pulverisation and extraction process can also lead to variance. This research was therefore carried out in conjunction with the pharmacopoeia criteria for determining the quality of raw drugs, as they are used in preparation of herbal remedies. The standardization criteria conducted in the phytochemical analysis offer valuable knowledge on raw material recognition and authentication. The study of the raw materials used in the preparation of PHR will use the findings as appropriate control measures of consistency. The presence of saponins and tannins in POLH extract contribute to the susceptibility of MQ larvae and act as a killing agent (Abou-Elnaga, 2015; Goc and rath, 2016; Sharifi-Rad *et al.*, 2017; Villaba *et al.*, 2017; Akhtar *et al.*, 2019; Nayak *et al.*, 2020). POLH extract's mortality effect on MQ larvae is because of the presence of saponins and tannin molecules in our study.

Conclusion

It has been concluded that the PHR MQ repellent is safe, cheaper and not toxic than the currently available chemical MQ repellents on the market and ensures a stable, MQ free atmosphere for society.

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