Extraction of potential mosquito repelling phytoconstituents from male papaya flower petals

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Abstract: *As the population of pest increases, necessity for pesticide production also increases day by day. One of the popular fast moving consumer goods based pesticide is mosquito repellent. Mosquito attacks both humans and plants. The commercial repellents used to kill mosquito are harmful to human health too. In the present investigation, experiments were carried out for the extraction of alkaloid carpaine from male papaya flowers. The extract of male papaya flower was extracted with water as the solvent using simple distillation. The pH of the water was adjusted to 7.0. The preheating temperature of the solid was maintained between 42 and 50°C. The solid and solvent were taken in the ratio of 1:2. Distillation was carried out at 80oC and the distillate obtained comprised of the extract and water. Water is separated by liquid-liquid extraction with hexane as the entrainer. The presence of alkaloid carpaine in the extract was confirmed by Wagnor and Mayer tests. The yield obtained was 3.6%.*

Keywords — *Alkaloid carpaine, Simple distillation, Male papaya flower petals, Vaporisation, Mosquito.*

I. INTRODUCTION

The uses of phytochemicals from various plants were known traditionally in India since rid vedic times (between 450 to 1600 BC) as unani, ayurvedic, sidha, herbal etc. Nutrition researchers estimate that more than 4,000 phytochemicals have been identified, but only about 150 have been studied in depth. Thus, more research is needed to find out in the field of phytochemicals. There are 48632 associations involved worldwide in phytochemical development and production. In India 1742 medicinal plants are there. Phytochemicals have precious value than chemical compounds derived from combinatorial chemistry and synthetic chemistry [1]. Phytochemicals are not only used as therapeutics, but also as poisons for targeted species like pathogenic microorganisms. Phytochemicals are classified into primary metabolites and secondary metabolites. Carbohydrates, amino acids and lipids fall under primary metabolites. Secondary metabolites are metabolites derived from primary metabolites and it includes alkaloids, terpenoids and phenolics. Alkaloid carpaine is present in various parts of papaya plant. It can be used to reduce blood pressure, kill mosquito, worms and amoeba [2]. Papaya plant is commonly called as paw-paw. It is also known as papaya, papita, tree melon, mamao [1]. The biological name of papaya is Carica papaya. The roots, flowers, fruits, unripe fruits, fibers, latex and leaves of the plants have good medicinal property, nutritive value and biological activity [4]. Some medicinal values of papaya are cancer cell growth inhibition [1], improving digestion, lowering cholesterol level, healing injuries [2], cleaning intestines, purifying blood, lowering blood pressure, controlling obesity, stopping urination, treating ringworm and curing blindness [3]. Papaya can also be used as abortifacient [1], nasal decongestant [3], antifertility, antiinflammatory, antiviral, antitumor, antihypertension [1], antioxidation [2], antiulcerogenics [5], antifungal, antimalarial, analgesic, anaesthetic, and antiseptic agents [3]. All the parts of the plant are enriched with various compounds. But they vary in composition from one part to another [3]. Some of the compounds are papaintha enzyme, vitamin A, vitamin E [2], vitamin C [3], alkaloids, phenolic compounds, carotenoids [1], ascorbic acids, panthothenic acids [6], minerals like magnesium [2], iron, calcium and phosphorous [3]. The bioactive constituents include folates [6], phenolics, terepenoids [2], lavanoids, tannins, saponins, flavanoids, cardiac glycosides, anthroquinones, phlobatinins, anthocyanosides and alkaloids.

The papaya flowers are tiny; funnel shaped; clustered in leaf axils [2] and are ivory white colored [1]. Papaya plant is classified into male staminate, female pistillate and bisexual hermaphroditics based on the flower which they are bearing [7]. Male flowers are mostly borne in an inflorescence with more than 10 flowers [1]. An advantage of male flowering plant over female is that, they are self fertilizing [4]. Male plant bears flowers for most part of the year [7]. Male flowers can be boiled for dietary consumption, jaundice treatment or can be processed

as herbal tea [4]. These male papaya flower petals are used here as the source of alkaloid carpaine. Alkaloid carpaine is naturally present in papaya as self defense compound against various insects. This compound never affects human because humans are naturally resistant to compounds like alkaloid carpaine, pseudo carpaine and dehydro carpaine [3]. The bitter smelling alkaloid carpaine is not favoured by vectors [12]. Vectors are agents which transmit pathogens from one animal or human to the other [9]. World Health Organization says 90% of death occurs to children under 5 [10]. Mosquitoes are one of those dangerous vectors.

Mosquitoes are tiny [9] hematophagous insects having mouth part that can penetrate the skin and affect the host with viruses [8]. Blood feeding female mosquitoes transmits malaria, yellow fever, dengue, filariasis, Japanese encephalitis [8], zika virus, chikungunya [9] and causes millions of death per year. 700 million people over the world are affected by mosquitoes [11]. Aedes aegypti sp. is one of those mosquitoes. It belongs to the diptera order, culicidae family, aedes genus and aegypti species [8]. Synthetic insecticides can ovicidally block the egg hatchings of aedes aegypti sp.. For control of aedes aegypti sp., synthetic insecticides yields immediate results but once on resistance, higher concentration is needed further. This may kill non target organism and also leads to increase in cost [8]. US Environmental Protection Agency states these synthetic chemicals further leads to water pollution [10]. So, bioinsect repellents like alkaloid carpaine can be preferred to synthetic insecticides. They are less toxic, ecofriendly, easily biodegradable and also does not build resistance [8,9]. Febriyanto et al., 2017 carried out experiments with aedis aegypti sp. larvae. Using glucose, the eggs were hatched out in tap water and the fourth instar larvae were taken as test species. They exposed larvae to the vapor of extract solution and concluded that when mosquito smelled the papaya extract containing bitter compounds like alkaloid carpaine, it paralysed before death. They also observed that the body of the larvae darkens, movement slowed and the size decreased. The optimum mortality efficiency was 75 % in 500 ppm concentration of the solution for 2 hours exposure.

This research work focuses on the extraction of alkaloid carpaine using simple distillation [14].

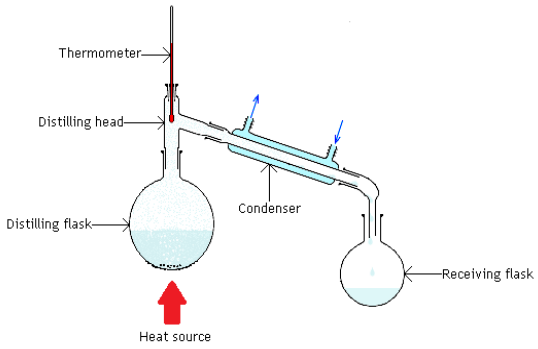

Fig. 1 Simple Distillation Setup

II. MATERIALS AND METHODS

Fresh male papaya flowers were collected from the Botanical garden of Annamalai University. The petals were separated and washed well. They were cut into pieces and preheated in hot air oven between 42 and 50ºC. Then it was mixed with solvent in the ratio of 1:2. The pH of the solvent was adjusted to 7.0. The distillation was carried out at an optimum temperature of 80ºC for 1 hour. The distillate collected was the mixture of extract and water [Fig. 1]. Water was separated by liquid-liquid extraction with hexane as entrainer. Hexane was then separated from the extract by boiling the mixture at 60ºC.

III. CONFIRMATORY TESTS

The extract of male papaya flower was evaporated to syrup consistency. While evaporating, 10 mL of 2M HCl was added. After 5 minutes it was allowed to cool using tap water. Then 0.5g of NaCl was added to the solution and mixed well. The solution was filtered and the filtrate obtained was divided into 2 portions. On addition of Mayer reagent and Wagner reagent in each test tube, a yellowish white precipitate and brown precipitate appeared respectively and confirmed the presence of alkaloid carpaine.

IV. RESULTS AND DISCUSSION

The extract containing alkaloid carpaine was weighed and found to be 1.8g. Thus the yield obtained was calculated with the formula

$$Yield = \frac{Desired\ product}{Feed} X100 = \frac{1.8}{50} X\ 100$$

= 3.6%

These findings were in agreement with the study conducted by Kumar et al., 2010 with papaya flowers. 300 g of flowers and 750 mL of solvent were distilled at 100ºC for 80 minutes and they obtained 10 mL of extract. In current research, distillation was carried out at 80oC for 1 hour and the yield was 3.6%. Alkaloid carpaine was also extracted from payaya leaves by Vaijayanthi et al., 2016. They carried out experiments in Soxhelt apparatus with acetone as the solvent in 1:4 (w/v) ratio and obtained the yield of 1.6%. Similar confirmatory results were also reported

by Bergonio et al., 2016 in their work on herbal tea production from male papaya flowers. They soaked 50 g of petals in 80% ethanol and filtered the contents. They further concentrated the filtrate using vacuum evaporator at 40ºC and 70 cm Hg pressure. After phytochemical screening of the extract, they confirmed the presence of alkaloids, flavanoids, polyphenols and cardiac glucosides. The confirmatory results were also correlated with the results obtained by Ukpobi et al., 2015. In their phytochemical analysis of dried papaya flower powder, they also found the presence of alkaloid carpaine about 0.05%/SEM.

V. CONCLUSIONS

The alkaloid carpaine extract yield of 3.6 % was obtained from male papaya flower petals. It could be concluded that instead of other synthetic chemicals which make pollution, affect human and build resistance, this phytoconstituent could be used as a bioinsecticide. Moreover, alkaloid carpaine is an insect neurotoxin. Thus it does not affect human being when inhaled. It is biodegradable and also it does not develop resistance of the target species.

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