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Prospective of Green Waste Compounds Against Dengue Vector *Aedes aegypti* and Filariasis Vector *Culex quinquefasciatus*

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ABSTRACT

One of the main objectives of Sustainable development goals (SDGs) is to protect the environment and public health by substantially reducing waste generation through prevention, reduction, recycling, and reuse by 2030. Another current concern is the overuse and misuse of synthetic larvicides which are the primary drivers in the burgeoning of insecticide-resistant vector species. Eutrophic water, lack of sanitation, and inadequate infection prevention and control promote the spread of these vectors that are responsible for various severe diseases like Malaria, Dengue, Chikungunya, Filariasis, etc. Mosquito species not responding to synthetic larvicides have developed resistance to them and the changing environment and this is an alarming situation if the proliferation is not regulated. The green waste comprising leaves, grass, flowers, non-woody plants, etc. contains a high amount of lignocellulosic material. They can be converted into mulch in a short period. Following the investigations, we examine the natural products as larvicides. Green waste collected and segregated belonged to several families. They were extracted with two solvents and their larvicidal activities were observed on two most common mosquito species viz; *Aedes aegypti and Culex quinquefasciatus.* The plant materials extracted with solvents manifest various degrees of larvicidal activities, the most promising results were drawn for acetone extracts of *Alastonia scholaris* against *C. quinquefasciatus* (LC_{50} = 1.76 mg/L, LC_{90} = 3.89 mg/L). For *A. aegypti* the best results were drawn from Ethyl acetate extracts of *Azadirachta indica* (LC_{50} = 1.91 mg/L, LC_{90} = 3.96 mg/L). Green waste is one of the best alternatives to synthetic larvicides as well as an innovative way to induce SDGs.

Keywords: Green waste, Larvicides, Phytochemicals, Sustainable development goals (SDGs).

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INTRODUCTION

For the protection of the environment and public health, one of the most essential public services includes waste collection and management. The current global policy frameworks suggest that waste services mainly target commitments to prevent, reduce, recycle, reuse, and properly collect waste (urban solid waste) by 2030 [1]. Green waste which is mainly generated from the yards as well as public places is a section of urban solid waste that goes dumped unused. Yard waste term is used for biodegradable waste which is carbon-based. Dry leaves, floral waste, grass cuttings, twigs, paper, hay, sawdust, pines, and needles all come under yard waste. The brown waste and green waste (Fresh leaves; rich source of nitrogen) together are recycled by composting. Since the brown waste is needed only in a small amount for composting the rest is dumped [2]. The present scenario in India is dealing with solid waste. Composting is one of the options to tackle the issue of handling solid waste [3]. Despite recycling the waste as compost a major amount remains in the environment, which is further dumped in the dumpsites. The green waste consists of leaves, grass, flowers, stems, non-woody plants, and straw which contain a higher amount of lignocellulosic material that draws a longer time for the

composting process and can be converted into mulch in a short period. In sites where huge amounts of yard waste are generated especially during the rainy, spring, and autumn seasons, the recycling of these materials can be workable [4]. Another current concern is the improper and exaggerated use of synthetic larvicides which are the primary drivers in the burgeoning of insecticide-resistant vector species. Eutrophic water, lack of sanitation, and inadequate infection prevention and control promote the spread of these vectors which are in turn responsible for various severe diseases like Malaria, Dengue, Chikungunya, Filariasis, etc. [5]. The brown and green wastes together called yard waste, can be used for their secondary metabolites as larvicides which are efficient for severe diseases like Dengue, Malaria, Chikungunya, and Japanese Encephalitis [6].

The plant parts have been used since remote times as herbal medicines for healing and medicinal properties [7]. The plant consists of some bioactive compounds or secondary metabolites which are responsible for its medicinal properties. Some of the major bioactive compounds that are present in plant parts having medicinal values are Tannins, alkaloids, phenolic compounds, flavonoids, etc [8]. Since the plant parts consist of phytochemicals they are a blend of different compounds, and the composition may vary from plant to plant and can have different effects on target organisms as herbal medicine. Leaves and barks of *Alstonia sp*. Is known to have anti-inflammatory, antidiabetic, antidiarrhoeal, antifertility, and antifungal properties [9]. The plant parts (bark and leaves) of *Psidium guajava* are used to treat gastrointestinal disorders, diarrhea, swelling, toothache, and colds [10]. The leaves of the neem tree have several properties like prevention of Hepatitis, controlling diabetes, antibacterial, etc. [11]. Whereas burns and bruises can be healed with the help of marigold leaves [12]. Antimicrobial study of *C. procera* manifests the use of these plant parts as a comparative study has not been reported so far and the study focuses on the most effective plant part as antimicrobial [13]. A fresh approach to green waste as an antimicrobial compound is also being studied with fifteen different flora and the results showed the effectiveness of green waste as an antimicrobial compound [14]. Floral waste is always thrown away without any proper waste management. Whereas it can be reused or recycled for many value-added chemicals, biofertilizers, dyes and insense, generation of biofuels, food supplements, etc. [15] and besides this, it can also be used as an antimicrobial agent as well as a larvicide.

The consideration of larvicidal activities of various plants is of special engrossment nowadays because of the global issue of rising resistance of pesticides against different mosquito species. The continuous commercial use of these drugs has eventually led to the resistance of mosquitoes which eventually leads to the rise of vector-borne disease. The treatments of these vectors are important since they are dangerous to humans and can cause several diseases and infections if not controlled and treated. Novel medicines can be developed through phytochemicals as they are the blend of many bio-active compounds and therefore are hard for the mosquito species to get resistant.

For phytochemical extraction, studies were carried out with two solvent systems (Acetone, & Ethyl acetate) to produce a relative study against two disease-causing mosquito genera (*Aedes* and *culex*) to discover the larvicidal properties of some common trees which are a source of green waste, commonly found in India. Down to the literature survey carried out, there were no reports present on morphological, physiological, and larvicidal outturns of leaf extracts from different green waste. Therefore the present study was done to evaluate larvicidal attributes of green waste Acetone and Ethyl acetate extract against *Aedes* and *Culex* sp.

MATERIALS AND METHODS

Plant material

The major plant families that are found in green waste and can also be reused against multidrug-resistant species of mosquitoes are Apocynaceae, Meliaceae, Rutuceae, Asteraceae, Fabaceae, Malvaceae, etc. Therefore, concerning the mentioned families four different plants and plant parts were selected and further segregated as larvicides. The tree species which were selected for the study consisted of *Alastonia scholaris, Azadirachta indica, Plumera rubra,* only the leaves were selected for the study of larvicides, whereas floral waste was selected in which a mixture of several flower parts was

collected from the waste. All the selected plants were then specified for qualitative analysis**.** Different experiments were executed to investigate the presence of flavonoids, alkaloids, terpenes, etc. which are accountable for larvicidal activity [16]. The green waste was collected from the city of Nagpur, Maharashtra, India. Leaves were collected during the shedding and pruning of plants, as the floral waste was collected from religious places around Nagpur city, and gathered material was kept in reserve in low humidity conditions at room temperature.

Pre-processing of plant material

The plant materials were first segregated and then were finely chopped into small pieces and shade dried (green waste) thoroughly for 10-15 days, pulverized mechanically to prepare grainy powder. Each plant material powder was hoarded in plastic bottles labeled respectively and was stored at room temperature for further extraction. The selected plant parts were extracted with different solvents, such that all the polar and non-polar compounds were drawn out in the crude extract.

Preparation of different solvent extracts

The selection of solvents and extraction methods was carried out with the help of the literature survey and different evaluations, which concluded that maceration by acetone and ethyl acetate as the extraction solvents [17]. Each pulverized plant material was then extracted with Acetone and Ethyl acetate respectively by the method of cold maceration. Maceration of the plant materials (30 g) with 300 ml of Acetone and Ethyl acetate respectively with frequent shaking for 2 days at room temperature. After two days the macerated extracts were processed by using Whatmann No. 1 filter paper. The filtrates (extracts) were evaporated to dryness Using a rotary evaporator to yield the crude extracts and stored at 4°C until further analysis. The extracts were labeled accordingly. The Whatman filter paper was used to filter the supernatant while two more rounds of extracts were carried out with the leftover residues. The % extract yields of all the solvents and the mass of the extract in grams are given in **Table 1** [18].

$$\text{Percentage Yield (\%)} = \frac{\text{Dry weight of extract}}{\text{Dry weight of plant material}} \times 100 \quad (1)$$

Mosquito culture

Two vector mosquito species *Aedes aegypti* and *Culex quinquefasciatus* which are responsible for diseases like Dengue, Yellow fever, Lymphatic filariasis, and Japanese encephalitis were selected according to their pharmacological and clinical importance [19]. The vector species were collected from the sewage water, cement tanks, stagnant drains, soakage pits, cesspools, wetlands, etc around CSIR-NEERI, Nagpur, India. The mosquito species were reared in CSIR-NEERI, laboratory Nagpur. The collected larvae were fed with a 3:1 ratio of dog biscuits and yeast powder. Whereas adults were fed by chick blood age of one week and sucrose solution of 1%. The reared mosquito larvae as well as adults were kept in a light period of 12 hours and 12 hours dark, maintaining the temperature at (28 ± 2) ˚ C, whereas relative humidity (RH) was maintained at 75-80%.

Larvicidal bioassay

The plant crude extracts were subjected to larvicidal activity according to the method developed by WHO [20]. The test was performed on all the instars in several batches and the results here are depicted for the early fourth instar larvae which were introduced in disposable cups filled with 200 ml of water in each container. The test was performed in three replicates, and different concentrations of the plant extract were added to these containers ranging from 0.005-5 mg/L. For each set of concentrations equal number of controls was also set simultaneously. For the control 1ml solvent was added to the containers and one control was kept without the addition of solvent as well as the plant extract. The bioassay was carried out for 24-48 hrs and the present mortality, as well as the lethal concentrations (LC50, LC90), was calculated by probit analysis.

RESULTS AND DISCUSSION

Extraction yields of the plant extract

The green waste is generated mainly during the shedding and pruning of the plants, whereas the floral waste is generated mostly from religious places and a large quantity is dumped without

recycling. The selected plant parts were first dried, ground into a coarse powder, and extracted with different solvents, such that all the polar and non-polar compounds were drawn out in the crude extract. The selection of solvents and extraction methods was carried out with the help of a literature survey and setting up different evaluations, which brought to a conclusion of selecting maceration as the extraction method, and acetone, and ethyl acetate were chosen as the extraction solvents. Different tests were carried out to investigate the presence of alkaloids, flavonoids, terpenes, etc. [16] which are accountable for larvicidal activity **(Table 1)**. The powdered plant materials were extracted by using the cold maceration method, the extract yield then went through the evaporation process by using a rotary evaporator under 80 to 100 r/min at 60 °C to evaporate the surplus solvent. The crude extract was stored in an oven at 37 °C for the further drying process. The highest yield was obtained from *A. scholaris* in ethyl acetate extract (10.63%) while the lowest yield was from *P. rubra* in ethyl acetate extract (0.45%) **(Figure 1).**

Table 1. Screening and Qualitative phytochemical analysis of a crude extract from acetone and ethyl acetate, solvents of segregated green waste

Solvent	Qualitative phytochemical analysis	*A. scholaris*	Floral waste	*P. rubra*	*A. indica*
Acetone	Alkaloids	-	+	-	+
	Flavanoids	+	+	-	+
	Terpenes	+	-	-	+
	Saponins	+	+	+	+
	Tannins	+	+	+	+
	Phenolic compounds	+	-	-	+
Ethyl acetate	Alkaloids	-	+	-	+
	Flavanoids	+	+	+	+
	Terpenes	+	-	-	+
	Saponins	+	+	+	+
	Tannins	+	+	+	+
	Phenolic compounds	+	+	-	-

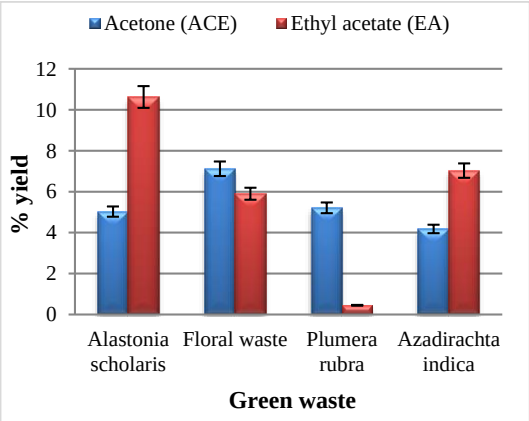

Figure 1. Extract yields for Acetone and ethyl acetate solvent extracts for segregated green waste.

Estimation of larvicidal activity

The Mosquito vectors consist of several species that do not respond to synthetic larvicide treatment and have developed resistance to them and the changing environment. This was considered using the data of the World Health Organization in 2020. It is an alarming situation since vector proliferates and spread diseases quickly if not regulated. The synthetic larvicides used over a course of time consisting of only one active ingredient as one of the major reasons for developing resistance by the mosquito species. The application of green larvicides against such resistant vectors can be of great importance for the inhibition of their multiplication and resistance. Likewise, the green waste generated can also be reused and recycled in an eco-friendly way, thus sustainably targeting global policies.

The crude extracts were first dissolved in ethyl alcohol so that they could be miscible in water therefore, one control of ethyl alcohol was imposed as the control to ensure the similarity with the test solutions which possibly endure the solvent and do not influence the larvicidal activity.

The vector organisms are habitually noticed in spreading contagious diseases. The study

depicted that all plant extracts used in the study impose a diverse range of larvicidal activity against the *Culex* and *Aedes* species tested. At first, the larvae were exhibited to diverse concentrations, to obtain the effectiveness of the extracts. The range was 0.005-5 mg/L yielding maximal mortality in twenty-four hours of exposure. Thereafter, observation was done during the bioassays. Three replicates were kept respectively for every concentration in which the extracts were dissolved with every arranged experiment. The behavior of the larvae exposed to the phytochemicals was observed immediately after the application and mortality percentages as well as LC50 and LC90 were recorded after 24 h. The deformities in the larvae were observed under the microscope and was discovered that the treated larvae were underdeveloped and abnormally grown **(Figure 2).** The phytochemicals whose application manifests above mentioned symptoms were considered to have efficient larvicidal activity.

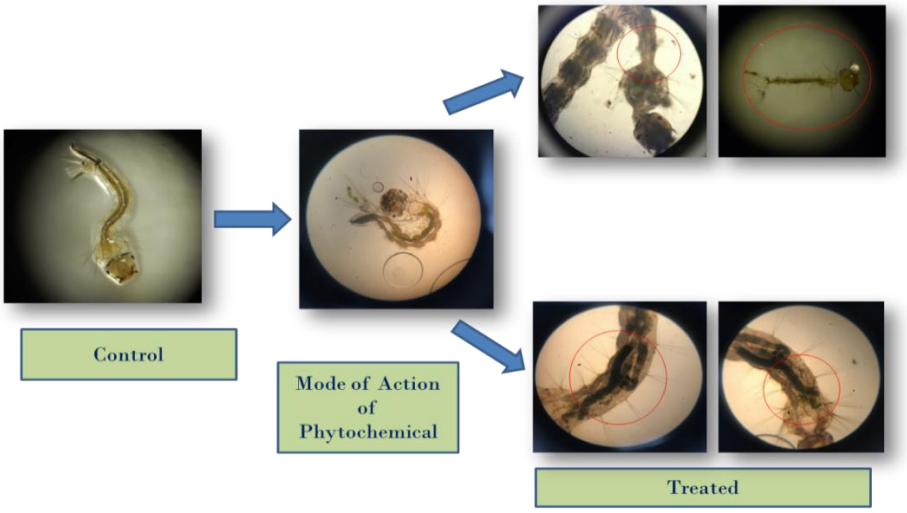

Figure 2. Mode of action of phytochemicals and deformities observed in larvae exposed to treatment.

Activity of treated larvae

A peculiar pattern was observed by the early fourth instar larvae of both mosquito species which were treated with different solvent extracts. The larvae exhibited an immediate effect on the treatment. Within an hour of exposure, the larvae exhibited agitation, excitation as well as restlessness. This was gradually followed by swift wriggling movements which continued for thirty minutes and eventually sank at the bottom of the containers in which the larvae were kept. The larvae were also observed to have a self-biting behavior of their anal papillae which gradually formed a ring-like structure and submerged in the water. After 5 hours of exposure to the treatment, the remaining larvae exhibited paralysis and eventually, 40% of the larvae population was found to be dead. The dead larvae were then examined under the microscope and the deformities in the morphology of the larvae were observed when examined against the control set of larvae. The damage was observed in the respiratory gills, and the anal papillae manifested as shrunken and damaged morphology. The larval instars were observed of consisting dark, dwarf, and damaged cephalothoracic regions **(Figure 2)**.

Plant families like Cladophoraceae, Miliaceae, Asteraceae, Oocystaceae Solanaceae, and Rutaceae consist of secondary metabolites that have various types of microbial as well as larval, adulticidal, and repellent activities against different species of mosquitoes [21]. Many plants have described different groups of chemicals like terpenoids, alkaloids, essential oils, steroids, phenomic compounds, etc. as secondary metabolites that are used as a plant defense mechanism against various species of invertebrates and different microbes. Compounds like tannins are rich in protein content and can inhibit the regulation of protein synthesis by binding to proline-rich proteins [22]. The plant parts used in this study are rich in flavonoids, alkaloids, and terpenoids which the plants use as a defense mechanism against various microbial infections and hence are active against various groups of microbes [23].

Furthermore, it was observed that saponins can create leakage to certain proteins and enzymes from the cells which will eventually result in the breakdown of the cells [24]. One of the studies manifests that the plant *C. citrinus* methanol extract is remarkable as a larvicide against the *A. Aegypti* species [25]. One of the studies carried out on *Delonix elata* ethanolic extract against all the instars manifests an LC_{50} in 24 hrs yielding the values of 4.91%, 5.16%, 5.95%, and 6.87%, respectively [26]. When the ethanolic leaf extract of *Annona reticulata* was tested against *C. Quinquefasciatus* and *A. aegypti* it was observed that the ethanolic leaf extracts were more effective on *A. aegypti* and that, with time it was observed that the LC_{50} value decreased gradually with more exposure. The LC_{50} values of both the mosquito larvae species were found to be 6.883, 5.992, 14.57, and 19.88 ppm against *A. aegypti* larvae and 0.502, 2.937, 4.204, and 6.224 ppm against 1st-4th larvae of against *C. quinquefasciatus* respectively at 24hrs. When *A. aegypti* fourth instar larvae were examined with methanol leaf extract of Crimson bottlebrush, it was observed that the larvae after treatment exhibited restlessness, excitation, aggressive movement, etc. These behavioral changes can be due to the presence of nerve poison in the phytochemical extract [25]. Similarly, the current study is carried out by selecting different plants that are characterized as yard/green waste on two mosquito species at different concentrations to scrutinize the effective plants along with suitable solvent systems as effective larvicides, following are the results discussed of the studies carried out.

Amongst The segregated green waste extracts that were tested against the two mosquito species, the utmost larvicidal activity was depicted in the leaf acetone extract of *A. scholaris* against *C. quinquefasciatus* followed by leaf ethyl acetate extract of *A. indica* against *A. Aegypti*, comprising the LC_{50} and LC_{90} values of 1.76, 3.89 mg/L and 1.91, 3.96 mg/L, respectively. The 95% confidence limits LC_{50} (LCL-UCL) and LC_{90} (LCL-UCL) were also calculated and are depicted in **(Table 2)**. Whereas, the Percent mortality of *A. aegypti and C. quinquefasciatus* treated with varying concentrations of the solvent leaf extracts from segregated green waste is depicted in **(Table 3)**. The other solvent extracts also depicted promising results against the tested organisms, and their crude extracts showed up to be effective as larvicides. The floral waste Ethyl acetate extract against *C. quinquefasciatus* followed by leaf acetone extract of *P. rubra* against *A. Aegypti,* also depicted promising LC_{50} and LC_{90} values of 2.21, 4.28 mg/L and 2.34, 4.52 mg/L, respectively. The comparative values of LC_{50} and LC_{90} are depicted in **Figures 3 and 4**. Similar results were drawn out from the studies done on the larvicidal properties of *A. indica* Oil extracted from the seed kernel against *Anopheles gambiae* depicting the LC_{50} 1666.86 ppm and LC_{90} 2880.94 ppm. 100 percent larval mortality was observed within 3 days at 500 ppm [27].

Plants comprise secondary metabolites which can act as natural agents to treat micro-organisms. Their efficacy of natural larvicidal agents has been more effective as compared to synthetic insecticides, by which the vectors have now become resistant. Brown and green waste are generally used for composting, but they can also be used as antimicrobial and larvicidal agents. The commonly available plants should be targeted for these activities. Such plants should be toxic to the target organisms causing various diseases. According to previous reports, and the larvicidal studies carried out at present of different extracts from solvents of saptaparni, neem leaves, marigold, Frangipani, drumstick, asthma weed, tulsi, etc. demonstrated larvicidal, antimicrobial activity against different species. All the plant materials extracted with solvents (Acetone and Ethyl acetate), delineated various degrees of larvicidal activities, but the most promising results were detected for the acetone extracts of *Alastonia*, *Azadirachta, Plumera*, against *C. quinquefasciatus,* followed by the ethyl acetate extracts for both the species. The plant parts which are being wasted or dumped can be used as natural products for these species, and can also be applied to the other mosquito species as well as other micro-organisms which can be a threat to humans. The plant extracts can be further studied for phytochemical analysis which gives favorable yields, the studies will further give a clear picture of the secondary metabolites or the bioactive compounds that are responsible for larvicidal activity.

Table 2. 3 Dose-response larvicidal bioassays (24 hours) of Acetone and Ethyl acetate solvent of leaf extracts of segregated green waste against *A. aegypti* and *C. quinquefasciatus.*

Green waste	Larval specie	Solvent	LC_{50}(mg/L) (24hrs) (LCL-UCL)	LC_{90}(mg/L) (24 hrs) (LCL-UCL)	r^2	χ2 df(10)	P-value significant
A. scholaris (P1)	*A. aegypti*	Acetone	2.53 (0.757-4.302)	4.42 (2.647-6.193)	0.9669	8.760	0.55
		Ethyl acetate	2.33 (0.557-4.102)	4.33 (2.557-6.103)	0.9611	3.935	0.95
	C. quinquefasciatus	Acetone	1.76 (-0.012-3.532)	3.89 (2.117-5.663)	0.8784	1.975	0.99
		Ethyl acetate	1.86 (0.087-1.772)	3.88 (2.107-5.653)	0.8776	1.455	0.99
Floral waste (P2)	*A. aegypti*	Acetone	2.50 (0.727-4.272)	4.45 (2.677-6.223)	0.9764	1.89	0.99
		Ethyl acetate	2.46 (0.687-4.232)	4.34 (2.567-6.113)	0.9713	2.396	0.99
	C. quinquefasciatus	Acetone	2.87 (1.097-4.642)	4.96 (3.187-6.733)	0.9488	17.25	0.068
		Ethyl acetate	2.21 (0.437-3.982)	4.28 (2.507-6.053)	0.9704	0.70	0.99
P. rubra (P3)	*A. aegypti*	Acetone	2.76 (0.987-4.532)	4.74 (2.967-6.513)	0.9817	2.984	0.98
		Ethyl acetate	2.34 (0.567-4.112)	4.52 (2.747-6.293)	0.9267	1.578	0.99
	C. quinquefasciatus	Acetone	2.64 (0.867-4.412)	4.65 (2.877-6.423)	0.9783	2.706	0.98
		Ethyl acetate	2.31 (0.537-4.082)	4.34 (2.567-6.113)	0.9526	1.698	0.99
A. indica (P4)	*A. aegypti*	Acetone	2.59 (0.817-4.362)	4.46 (2.687-6.233)	0.9747	4.866	0.89
		Ethyl acetate	1.91 (0.137-3.683)	3.96 (2.187-5.733)	0.9135	1.37	0.99
	C. quinquefasciatus	Acetone	3.64 (1.867-5.413)	4.80 (3.027-6.573)	0.9047	7.046	0.72
		Ethyl acetate	2.26 (0.487-4.033)	4.75 (2.977-6.523)	0.951	1.616	0.99

LCL – Lower Confidence Limits; UCL – Upper Confidence Limits; χ2 – Chi square.r2 regression

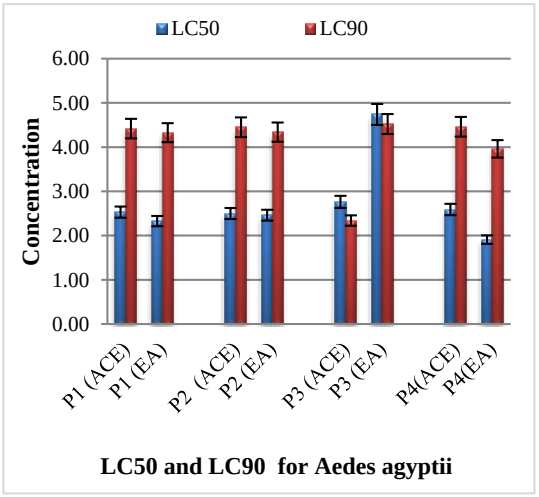

Figure 3. LC_{50} and LC_{90} values (24 hours) of Acetone and Ethyl acetate solvent of leaf extracts of segregated green waste against *A. aegypti.*

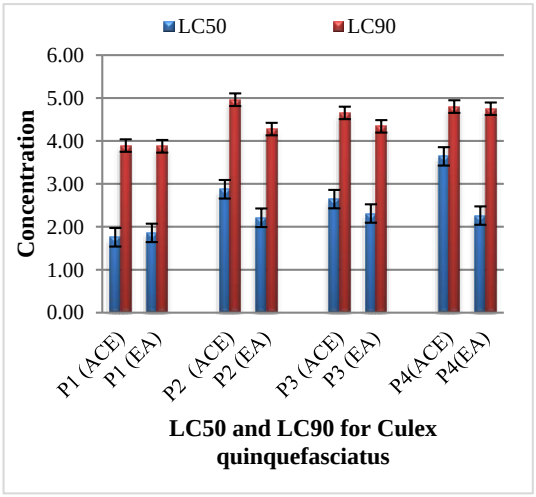

Figure 4. LC_{50} and LC_{90} values (24 hours) of Acetone and Ethyl acetate solvent of leaf extracts of segregated green waste against *C. quinquefasciatus.*

Table 3. Percentage mortality of mosquito larvae of *A. aegypti and C. quinquefasciatus* exposed to different concentrations of different solvent leaf extracts of segregated green waste.

Concentration (mg/L)			Mortality (%) (Mean ± SD)										
		Solvents	0.005	0.025	0.625	1	1.5	2	2.75	3.5	4	4.5	5
P1	*A. aegypti*	Acetone	1.33 ± 0.58	0.00 ± 0.00	4.00 ± 1.00	1.33 ± 0.58	36.00 ± 1.00	48.0 ± 0.00	53.3 ± 1.15	68.0 ± 0.00	81.3 ± 0.58	94.7 ± 0.58	100.0 ± 0.00
		Ethyl acetate	1.33 ± 0.58	1.33 ± 0.58	1.33 ± 0.58	28.0 ± 1.00	42.7 ± 2.08	54.7 ± 2.08	62.7 ± 0.58	73.3 ± 2.52	77.3 ± 1.53	93.3 ± 1.53	100.0 ± 0.00
	C. quinquefasciatus	Acetone	0.0 ± 0.00	5.33 ± 2.31	20.00 ± 1.73	53.33 ± 3.06	60.0 ± 2.65	72.0 ± 1.00	74.67 ± 2.08	84.00 ± 2.00	90.67 ± 1.53	96.00 ± 1.00	98.67 ± 0.58
		Ethyl acetate	1.33 ± 0.47	4.00 ± 0.82	16.00 ± 2.83	30.67 ± 2.49	56.0 ± 1.41	77.33 ± 1.25	86.67 ± 0.47	86.67 ± 0.47	88.00 ± 0.00	93.33 ± 0.47	98.67 ± 0.47
P2	*A. aegypti*	Acetone	2.67 ± 0.58	2.67 ± 0.58	5.33 ± 0.58	6.67 ± 0.58	36.00 ± 1.00	46.67 ± 0.58	53.33 ± 1.15	68.00 ± 0.00	81.33 ± 0.58	93.33 ± 0.58	100.00 ± 0.00
		Ethyl acetate	0.00 ± 0.00	4.00 ± 1.0	4.00 ± 0.00	6.67 ± 0.58	29.33 ± 0.58	50.67 ± 0.58	61.33 ± 0.58	66.67 ± 0.58	85.33 ± 0.58	96.00 ± 1.00	100.00 ± 0.00
	C. quinquefasciatus	Acetone	0.00 ± 0.00	1.33 ± 0.58	0.00 ± 0.00	1.33 ± 0.58	21.33 ± 0.58	32.00 ± 0.00	65.33 ± 0.58	65.33 ± 0.58	66.67 ± 0.58	77.33 ± 0.58	90.67 ± 0.58
		Ethyl acetate	0.00 ± 0.00	4.00 ± 1.00	14.67 ± 3.79	30.67 ± 4.93	41.33 ± 3.51	60.00 ± 2.00	61.33 ± 1.53	74.67 ± 2.89	81.33 ± 3.06	94.67 ± 1.53	98.67 ± 0.58
P3	*A. aegypti*	Acetone	0.00 ± 0.00	1.33 ± 0.58	4.00 ± 1.00	4.00 ± 0.00	22.67 ± 0.58	38.67 ± 0.58	53.33 ± 0.58	62.67 ± 0.58	77.33 ± 0.58	82.67 ± 1.53	94.67 ± 0.58
		Ethyl acetate	0.00 ± 0.00	1.33 ± 1.00	5.33 ± 0.00	38.67 ± 0.58	46.67 ± 0.58	57.33 ± 0.58	58.67 ± 0.58	66.67 ± 0.58	72.00 ± 0.58	89.33 ± 1.00	98.67 ± 0.00
	C. quinquefasciatus	Acetone	0.00 ± 0.00	0.00 ± 0.58	2.67 ± 0.00	8.00 ± 0.58	33.33 ± 0.58	44.00 ± 0.00	54.67 ± 0.58	69.33 ± 0.58	70.67 ± 0.58	86.67 ± 0.58	97.33 ± 0.58
		Ethyl acetate	0.00 ± 0.00	0.00 ± 0.00	5.33 ± 0.58	28.00 ± 5.20	44.00 ± 4.58	58.67 ± 1.53	65.33 ± 2.52	69.33 ± 1.53	77.33 ± 2.08	93.33 ± 1.53	98.67 ± 0.58
P4	*A. aegypti*	Acetone	0.00 ± 0.00	0.00 ± 0.00	1.33 ± 0.47	6.67 ± 0.47	22.67 ± 1.70	34.67 ± 0.94	60.00 ± 0.82	78.67 ± 0.94	82.67 ± 0.47	90.67 ± 0.47	94.67 ± 0.47
		Ethyl acetate	0.00 ± 0.00	2.67 ± 1.15	14.67 ± 3.79	44.00 ± 1.00	56.00 ± 2.00	68.00 ± 1.00	73.33 ± 1.55	81.33 ± 1.53	88.00 ± 1.73	98.67± 0.58	98.67 ± 0.58
	C. quinquefasciatus	Acetone	0.00 ± 0.00	0.00 ± 0.00	1.33 ± 0.58	2.67 ± 1.15	9.33 ± 0.58	10.67 ± 1.15	24.00 ± 1.00	38.67 ± 0.58	54.67 ± 1.15	66.67 ± 1.53	90.67 ± 0.58
		Ethyl acetate	0.00 ± 0.00	1.33 ± 0.58	4.00 ± 0.00	29.33 ± 4.62	46.67 ± 4.51	58.67 ± 1.53	65.33 ± 2.31	70.67 ± 1.15	80.00 ± 1.00	94.67 ± 1.53	100.00 ± 0.00
	Control	Ethanol	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.0 0 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00

± SD = standard deviation; P1 = *A. scholaris*; P2 = Floral waste; P3 = *P. rubra*; P4 = *A. indica*

CONCLUSION

By the present investigation, it can be concluded that all the phytochemicals present in green waste are the best alternatives to synthetic drugs and multidrug-resistant pathogens as well as an innovative way to recycle, reuse, and reduce the waste generation in the environment. Plants consist of various degrees of inhibitory effects against the pathogens tested. The most significant larvicidal activity was demonstrated by *A. scholaris* and *A. indica*, etc. showing potential larvicidal components that can be further used by pharmaceutical industries against various diseases caused by these vectors. The phytonutrient procedures can be accomplished in the future to segregate and separate secondary metabolites which can be separately produced as new drugs. By this method, green and brown waste can also be managed and is a great option apart from composting, dumping, and burning, to fulfill the

sustainable development goals of reducing, and reusing recycling of waste generated in the environment.

Guidelines for the study of plants and mosquito larvae in the research

All the studies were performed with the plants that are the least concerned according to IUCN and are part of yard waste. No harm was done to any plant parts which are vulnerable or fall into other such categories. All the experiments were carried out according to the WHO guidelines for laboratory and field testing of mosquito larvicides (2005).

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