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## Tomato leaves, a possible solution to control *Aedes aegypti* at larval stage

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### Abstract

Vector-borne diseases (VBDs) are causing mortality of almost 2 million people including nearly a million children every year across the globe. Thus mosquito control assumes worldwide importance and because of indiscriminate use of artificial insecticides creates multifarious complications including environmental pollution, insecticide resistance and toxic unsafe to person. Whereas, biopesticides offer an alternative to synthetic insecticides because they purpose low environmental pollution, much less toxic to human beings, less-hazardous to non-target biota, and also cheaper due to innate biodegradability. For this reason biopesticides is an eco-friendly approach to be correctly used in insect control including mosquitoes. The goal of the present study is to investigate the larvicidal activity of phyto extracts of tender leaves of tomato (*Lycopersicon esculentum*) against first to fourth larval stages of dengue vector i.e. *Aedes aegypti*, under the laboratory conditions. Mortality was examined visually at time interval of 24 hours upto 72 hours of exposure. The leaf extract exhibited promising larvicidal activity exhibiting 93.1% mortality, achieved at 30 and 40 ppm concentration levels of leaf extract after 72 hours of exposure in the 1st instar larvae of *Aedes aegypti*. The negative control showed lesser mortality. Thus the results indicate that tomato leaf extract could plausibly be utilized as an alternative to synthetic insecticide against the various larval stages of *Aedes aegypti*.

**Keywords:** Tomato leaves, phyto extract, *Aedes aegypti*, larvicidal activity, mortality, LC<sub>50</sub>, probit analysis

### Introduction

*Aedes aegypti*, the primary vector for dengue fever, dengue hemorrhagic fever, and yellow fever is an immense life threat over considerable areas of the tropics and subtropics. Approximately 40% of the world's population is now liable to dengue and the best way to stay safe from dengue virus transmission is to combat the disorder-carrying mosquitoes (WHO, 2012) [32]. In India, respectable facts of the Union Health Ministry display that there was a big growth of dengue infection in 2012.

Till 2012, figure stood at 50,222 cases and 241 fatalities in calculation to 18,860 instances and 169 deaths in 2011 (NVBDCP, 2013). An epidemic of chikungunya virus, results in an outbreak that has troubled >1.5 million people, along with travellers who have visited those regions (Taubitz *et al.*, 2007) [26].

Mosquitoes unfold dengue fever by means of manner of biting. This form of dengue virus is known as Flavi virus. It has 4 types i.e. DENV 1, DENV 2, DENV 3, DENV 4. These are a few manners or different have other *Aedes aegypti* transmit dengue in huge range. *Aedes aegypti* prefers to breed only in still waters and bites human beings at day time and thereafter comes in to blood circulation for next 2-7 days in the infected man or woman. Mosquitoes can transmit dengue from one man or woman to some other within the manner of biting infected people. Consequently, emerge as a hard hassle to public health international, and it has a serious social and hazardous impact especially in tropical and subtropical countries.

The approach to combat those illnesses in large part relies on interruption of the disease transmission cycle through both destruction of the aquatic stages or through killing the adult mosquitoes the usage of chemical pesticides (Pani *et al.*, 2015; Kanis *et al.*, 2018) [13, 21]. However, persevered use of synthetic chemical insecticide primarily based measures for vector manage has resulted in lower efficacy of such pesticides and appearance of resistance in mosquito populace, had undesirable outcomes on non-goal organisms, and convey damages to environment and human health (Govindarajan *et al.*, 2016; Araújo *et al.*, 2021) [2, 9]. Important emphasis has been focused on the use of herbal plant-primarily based products as larvicides that could provide a change to synthetic pesticides. Plants are wealthy assets of bioactive

compounds that may be used to expand environmentally secure vector and pest-coping with agents. Some of plant life and microbes had been said as selective with little or no harmful effect on non-goal organisms and the environment (Govindarajan *et al.*, 2008; Govindarajan *et al.*, 2012) [10, 12].

Phytochemicals are botanicals, extracted from floral sources, can potentially replace chemical pesticides. They are extracted both from the complete vegetation of little herbs or from diverse parts like fruits, leaves, stem, barks, roots and many others of large flowers or bushes. Phyto extracts are rising as potential mosquito control dealers, with low-price, easy-to-administer and less hazardous compared to isolated or synthetic insecticides and therefore, be used efficiently in mosquito management (Shalan *et al.*, 2005; Luz *et al.*, 2020) [17, 24].

The tomato plant is a short lived perennial plant, grown as an annual plant, inside the Solanaceae or nightshade family, usually developing to at least one-3 m tall, with a weakly woody stem that generally scrambles over other vegetation. Medicinal properties of the tomato plant are because of the presence of secondary metabolites like phytofluene, lycopene, carotenoids, phytoene, ascorbic acid, pro-vitamin A, polyphenols, beta-carotenoid, nutrients like Vitamin A, Vitamin B, Vitamin E, Vitamin K, Vitamin C, Sulphur, Potassium, Calcium, Iron, Phosphorus, sugars like ketoses, disaccharides, polysaccharides, aldoses, amino acids, all of these contribute to the antioxidant property of the tomato (Neji *et al.*, 2018) [19]. It also shows numerous insecticidal, antifungal, and nematicidal activities (Raiola *et al.*, 2014; Afreen *et al.*, 2016) [1, 23].

Therefore, the present study was carried out in the laboratory conditions to assess the larvicidal activity of leaf extract of *Lycopersicon esculentum* against the larval stages of *Aedes*

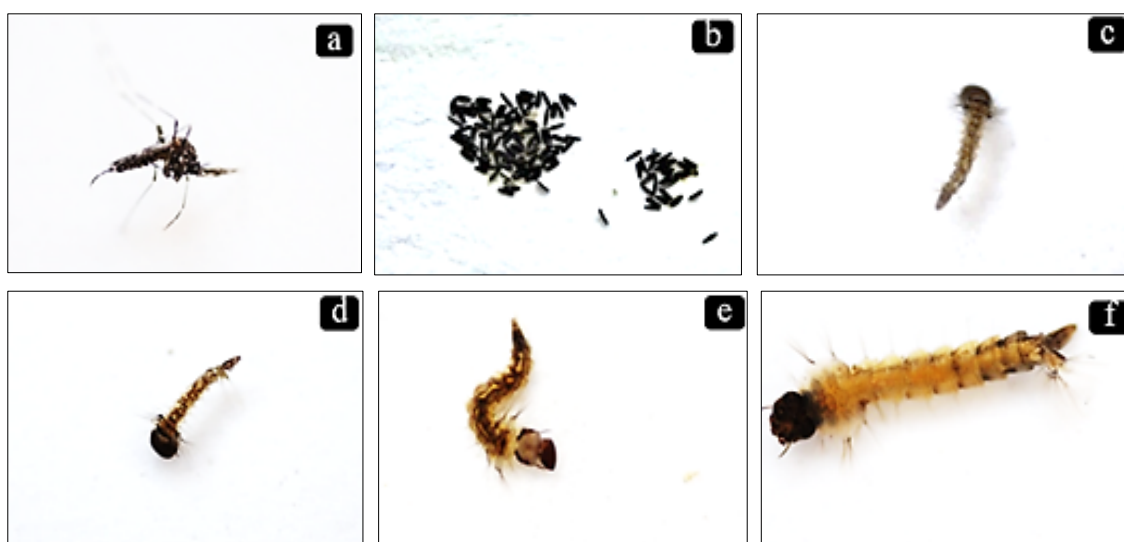
*aegypti*.

## 2. Materials and Methods

### 2.1 Mosquito rearing

The eggs of *Aedes aegypti* were procured from the different slum regions with hand net from local areas Prayagraj, UP, India. They were kept in a tray containing tap water and maintained at  $24\pm 2$  °C temperature,  $70\pm 30\%$  relative humidity. 5 mg of bovine liver powder suspension was dissolved in 375 ml distilled water and kept in a beaker. Then approximately 300 *Aedes* eggs were placed into liver powder nutrient suspension with the help of a piece of egg paper. The overcrowding of the eggs was maintained manually with the help of soft brush to avoid development of smaller mosquitoes.

After 24-36 hrs of incubation, the eggs hatched into first instar larvae and were moved with a transfer pipette to a large (3 L) bowl containing 1.5 L water and were fed on bovine liver powder which was checked every 1-2 days. The larvae grew rapidly from first larval stage to second, then third and fourth. The first to fourth larvae were used in the study. Once larvae become pupae they were transferred into a 500 ml plastic beaker containing 250 ml distilled water, which was placed into a rearing cage (50x50x50 cm<sup>3</sup>) covered with a fine mesh for trapping adults after their emergence. The adult males were provided with 10% sugar solution for feeding and the females were provided blood feed by placing a pigeon on top of the breeding cages and for oviposition petri dishes filled with 50 ml tap water lined with filter paper and kept inside the cages. Different beakers containing pupae or larvae, were used for collections of larvae from different growth stages for bioassay. Figure (A) portrays different phases of *Aedes aegypti*.



**Fig A:** Different phases of life cycle of *Aedes aegypti*. Adult (a); eggs (b); first instar larva (c); second instar larva (d); third instar larva (e); fourth instar larva (f).

### 2.2 Preparation of leaf extract from Tomato

The complete plant of tomato was procured from the agricultural grounds of SHUATS, Prayagraj, India. The identification of tomato plants were done by Department of Botany, SHUATS, Prayagraj for taxonomic identification and substantiation of the species. 50 gm of fresh green tomato leaves (figure B) were taken, washed with tap water and

cleaned thoroughly with a cloth. The leaves were cut into small pieces and immediately ground using a pestle and mortar. The grounded material was then filtered by cloth and then using the whatman no.1 filter paper kept in a funnel through which the grounded material passed. The filtrate of the crude leaf extract was stored in a clean brown bottle till further use.



**Fig B:** Tomato plant leaf

### 2.3 Bioassay for assessment of larvicidal activity

The mosquito larvae were exposed to four test concentrations (10, 20, 30 and 40 ppm each) and two controls (positive control and negative control) to find out the mortality of the materials under test in 24h, 48h, 72h and also to determine the LC<sub>50</sub> value. Batches of 100 first, second, third and fourth instar larvae are transferred by means of strainers to small disposable test cups, each containing 100 ml of water. The appropriate volume of dilution was added to 100 ml in the cups to obtain the desired target dosage, starting with the lowest concentration. Three replicates were set up for each concentration and an equal number of controls are set up simultaneously with tap water, each test was run three times on different days. For long exposures, larval food was added to each test cup, particularly if high mortality was noted in control. The test cups were kept at room temperature and preferably a photoperiod of 12 h light followed by 12 h dark (12L:12D). After 24 h exposure, larval mortality was recorded. Moribund larvae were counted and added to dead larvae for calculating percentage mortality. Dead larvae were those that cannot be induced to move when they were probed with a needle in the siphon or the cervical region, the results were recorded, from where the LC<sub>50</sub>, values, fiducial limits and slope and heterogeneity analysis were calculated.

### 2.4 Data analysis

The mortality of treated groups were corrected according to Abbott's formula,

$$\text{Mortality (\%)} = \frac{X - Y}{X} \times 100$$

Where  $X$  = percentage survival in the untreated control and  $Y$  = percentage survival in the treated sample.

Data from all replicates was pooled for analysis. LC<sub>50</sub> values were calculated from a log dosage-probit mortality regression line using computer software programs, or estimated using log-probit paper. Bioassays were repeated for three times, using new solutions or suspensions and different batches of larvae each time.

## 3. Results

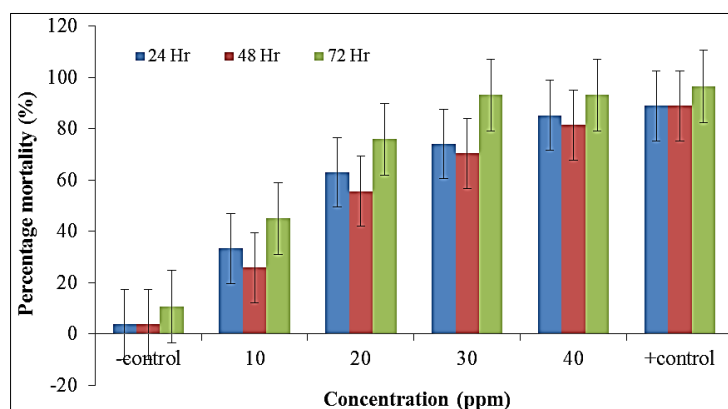
### 3.1 Effect of tomato leaf extract on larval mortality

#### 3.1.1 Larvicidal properties of leaf extract of *Lycopersicon esculentum*

The crude leaf extract of tomato was divided into four different concentrations; and the mortality of the *Aedes aegypti* larvae was examined, visually. The entire test concentration reported considerable amount of larvicidal activity against all the instar larvae of *Aedes aegypti*.

#### 3.1.2 Larvicidal activity of crude leaf extracts

The larvicidal activity of different concentrations of green leaf extracts were shown in Figure 1. It was visually observed that 93.1% mortality was achieved in 40 ppm and 30 ppm concentration level of leaf after 72 hours of exposure in the 1<sup>st</sup> instar larvae of *Aedes aegypti*. Whereas, 85.2% mortality was visually examined at 40 ppm concentration level after 24 hours of exposure in the 1<sup>st</sup> instar larvae and lowest mortality percent was found being 25.9% at 10 ppm concentration at 48 hours of exposure in the first instar larvae of *Aedes aegypti*.



**Fig 1:** Larvicidal activity of leaf extract of tomato against 1<sup>st</sup> instar larvae of *Aedes aegypti* shown through percentage mortality. Larvae cultured in the absence of tomato leaf extract were treated as – control whereas, larvae cultured in full strength tomato leaf extract were treated as + control. (The bars above the graph show standard error of the mean).

The probit analysis and regression analysis were done in Table 1 for the mortality rates of 1<sup>st</sup> instar larvae in the crude leaf extract at different concentrations at 24, 48 and 72 hours interval and found that the highest mortality rate (93.1%) at 40ppm concentration after 72 hours exposure.

Statistical analysis confirmed the lowest value of LC<sub>50</sub> (158.87) through visual examination after 48 hours of

exposure. Likewise 95% fiducial limits (FL) for LC<sub>50</sub> after 48 hours is 72.33-348.97 and the slope is 1.21. Followed by the LC<sub>50</sub> (184.53) after 72 hours of exposure with 95% fiducial limits for LC<sub>50</sub> being 68.33- 498.40 and the slope is 0.90 and then after 24 hours of exposure LC<sub>50</sub> (197.39) was visually examined with 95% fiducial limits for LC<sub>50</sub> after 24 hours being 73.23 - 532.03.

**Table 1:** Probit analysis and regression analysis of mortality rates of 1<sup>st</sup> instar larvae of *Aedes aegypti*.

Hours	LC <sub>50</sub>	95% Fiducial CL	Slope	Intercept
24	197.39	LFL (73.23)	0.91	2.90
		UFL (532.03)		
48	158.87	LFL (72.33)	1.21	2.34
		UFL (348.97)		
72	184.53	LFL (68.33)	0.90	2.96
		UFL (498.40)		

Where LC<sub>50</sub> stands for Lethal concentration at 50%.

LFL stands for lower fiducial limit.

UFL stands for upper fiducial limit.

**Table 2:** Probit analysis and regression analysis of mortality rates of 2<sup>nd</sup> instar larvae of *Aedes aegypti*.

Hours	LC <sub>50</sub>	95% Fiducial CL	Slope	Intercept
24	179.52	LFL (69.12)	0.94	2.88
		UFL (466.29)		
48	244.02	LFL (76.74)	0.77	3.16
		UFL (775.96)		
72	239.69	LFL (81.66)	0.84	3.00
		UFL (703.55)		

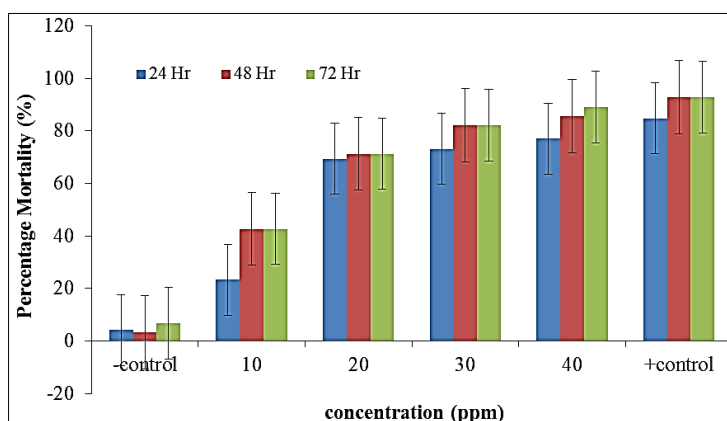
Where LC<sub>50</sub> stands for Lethal concentration at 50%.

LFL stands for lower fiducial limit.

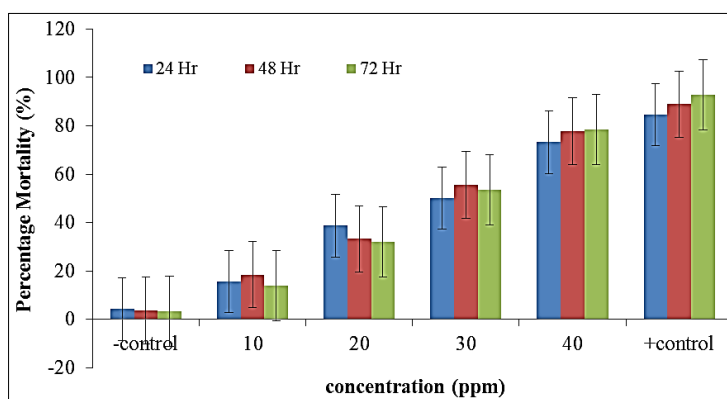
UFL stands for upper fiducial limit.

Crude extract tomato leaf (10, 20, 30 and 40 ppm) was tested against second instar of *Aedes aegypti* larvae (figure 2). It was visually examined that 89.2 percentage mortality was achieved at 40 ppm concentration level after 72 hours of exposure followed by 85.7% mortality at 40 ppm

concentration level after 48 hours of exposure. Whilst lowest mortality percent was found to be 23.4% it at 10 ppm concentration after 24 hours of exposure.



**Fig 2:** Larvicidal activity of leaf extract against 2<sup>nd</sup> instar larvae of *Aedes aegypti* shown through percentage mortality. Larvae cultured in the absence of tomato leaf extract were treated as – control whereas, larvae cultured in full strength tomato leaf extract were treated as + control. (The bars above the graph show the standard error of the mean)



**Fig 3:** Larvicidal activity of leaf extract against 3<sup>rd</sup> instar larvae of *Aedes aegypti* shown through percentage mortality. Larvae cultured in the absence of tomato leaf extract were treated as – control whereas, larvae cultured in full strength tomato leaf extract were treated as + control. (The bars above the graph show standard error of the mean)



The probit and regression analysis were done for the mortality rates of 2nd instar larvae in the crude leaf extract at different concentrations at 24, 48 and 72 hours interval and found that the highest mortality rate (89.2%) at 40 ppm concentration after 72 hours exposure.

The lowest value of LC<sub>50</sub> (179.52) was visually examined after 24 hours of exposure. Likewise 95% fiducial limits for LC<sub>50</sub> after 24 hours is 69.12 - 466.29 and the slope is 0.94. Followed by the LC<sub>50</sub> (239.69) after 72 hours of exposure with 95% fiducial limits for LC<sub>50</sub> being 81.66- 703.55 and the slope is 0.84 and then after 48 hours of exposure LC<sub>50</sub> (244.02) was optically canvassed with 95% fiducial limits for LC<sub>50</sub> after 48 hours being 76.74 - 775.96.

Crude extract tomato leaf (10, 20, 30 and 40 ppm) was tested against third instar of *Aedes aegypti* larvae. It was visually canvassed that 78.5% mortality was achieved in 40 ppm concentration level after 72 hours of exposure, followed by 77.8% mortality at 40 ppm concentration level after 48 hours of exposure, whereas lowest mortality percent being 14.0% at 10 ppm concentration after 72 hours of exposure in third instar larvae of *Aedes aegypti*.

The probit analysis and regression analysis were done for the mortality rates of 3<sup>rd</sup> instar larvae in the crude leaf extract at different concentrations at 24, 48 and 72 hours interval and found that the highest mortality rate (78.5%) at 40ppm concentration after 48 hours exposure.

The lowest value of LC<sub>50</sub> (111.76) was observed after 72 hours of exposure (table 3). Likewise 95% fiducial limits for

LC<sub>50</sub> after 72 hours is 59.98 - 208.25 and the slope is 1.55. Followed by the LC<sub>50</sub> (135.66) after 48 hours of exposure with 95% fiducial limits for LC<sub>50</sub> being 70.24- 262.01 and the slope is 1.52 and then after 24 hours of exposure LC<sub>50</sub> (157.25) was observed with 95% fiducial limits for LC<sub>50</sub> after 24 hours being 74.96 - 329.90.

**Table 3:** Probit analysis and regression analysis of mortality rates of 3<sup>rd</sup> instar larvae of *Aedes aegypti*.

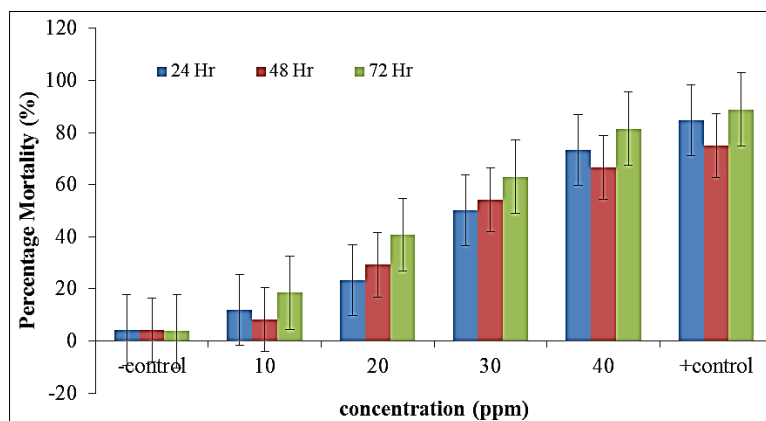
Hours	LC <sub>50</sub>	95% Fiducial CL	Slope	Intercept
24	157.25	LFL (74.96)	1.32	2.10
		UFL (329.90)		
48	135.66	LFL (70.24)	1.52	1.76
		UFL (262.01)		
72	111.76	LFL (59.98)	1.55	1.83
		UFL (208.25)		

Where LC<sub>50</sub> stands for Lethal concentration at 50%.

LFL stands for lower fiducial limit.

UFL stands for upper fiducial limit.

Crude extract tomato leaf (10, 20, 30 and 40 ppm) was tested against 4<sup>th</sup> instar of *Aedes aegypti* larvae (figure 4). The results revealed that 81.5 percent mortality was achieved in 40 ppm concentration level after 72 hours of exposure, followed by 73.2% mortality at 40 ppm concentration level after 24 hours of exposure. Whereas, lowest mortality was observed being 8.3% at 10 ppm concentration at 48 hours of exposure in the 4<sup>th</sup> instar larvae of *Aedes aegypti*.



**Fig 4:** Larvicidal activity of leaf extracts against 4<sup>th</sup> instar larvae of *Aedes aegypti* shown through percentage mortality. Larvae cultured in the absence of tomato leaf extract were treated as – control whereas, larvae cultured in full strength tomato leaf extract were treated as + control. (The bars above the graph show standard error of the mean)

**Table 4:** Probit analysis and regression analysis of mortality rates of 4<sup>th</sup> instar larvae of *Aedes aegypti*.

Hours	LC <sub>50</sub>	95% Fiducial CL	Slope	Intercept
24	112.72	LF (65.88)	2.00	0.90
		UFL (192.88)		
48	124.70	LFL (71.02)	1.96	0.89
		UFL (218.94)		
72	126.57	LFL (66.04)	1.51	1.83
		UFL (242.56)		

Where LC<sub>50</sub> stands for Lethal concentration at 50%.

LFL stands for lower fiducial limit.

UFL stands for upper fiducial limit.

The probit analysis and regression analysis were done for the mortality rates of 4<sup>th</sup> instar larvae in the crude leaf extract at different concentrations at 24,48 and 72 hours interval and found that the highest mortality rate (81.5%) at 40ppm concentration after 72 hours exposure.

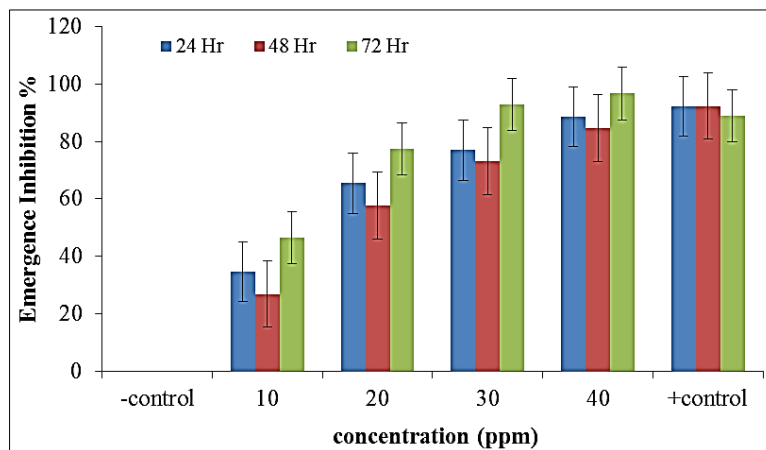
The lowest value of LC<sub>50</sub> (112.72) was optically canvassed after 24 hours of exposure (table 4). Likewise 95% fiducial limits for LC<sub>50</sub> after 24 hours is 65.88 - 192.88 and the slope is 2.0. Followed by the LC<sub>50</sub> (124.70) after 48 hours of exposure with 95% fiducial limits for LC<sub>50</sub> being 71.02-

218.94 and the slope is 1.96 and then after 72 hours of exposure  $LC_{50}$  (126.57) was visually examined with 95% fiducial limits for  $LC_{50}$  after 72 hours being 66.04 - 242.56.

### 3.2 Effect of Emergence Inhibition (EI) on crude extract of leaf of *Lycopersicon esculentum*

Crude extract (10, 20, 30 and 40 ppm) of tomato leaf was tested against first instar larvae of *Aedes aegypti* for EI (figure

5). It was visually examined that the maximum emergence Inhibition (EI) was achieved 82.6% in 40 ppm concentration level having average survival (1.7) after 72 hours of exposure. Followed by 73.9% of EI in 30ppm concentration having average survival 2.3 after 72 hours of exposure. Whereas the lowest (9.1%) EI was recorded at 10ppm concentration after 24 hrs of exposure.



**Fig 5:** EI of leaf extract against 1<sup>st</sup> instar larvae of *Aedes aegypti* shown through percentage EI. Larvae cultured in the absence of tomato leaf extract were treated as – control whereas, larvae cultured in full strength tomato leaf extract were treated as + control. (The bars above the graph show standard error of the mean)

The lowest value of  $LC_{50}$  (1.63) was visually seen after 72 hours of exposure (table 5). Likewise 95% fiducial limits for  $LC_{50}$  after 72 hours is 0.59 - 4.50 and the slope is -1.08. Followed by the  $LC_{50}$  (2.10) after 48 hours of exposure with

95% fiducial limits for  $LC_{50}$  being 0.79- 5.56 and the slope is -1.06 and then after 24 hours of exposure  $LC_{50}$  (2.47) was visually seen with 95% fiducial limits for  $LC_{50}$  after 24 hours being (0.96 - 6.37).

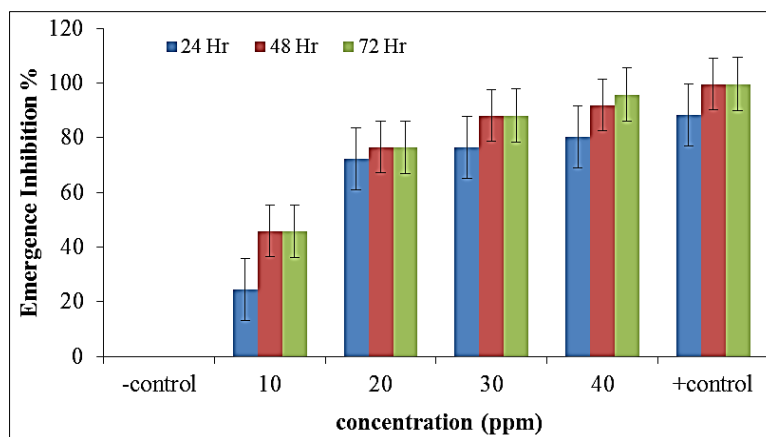
**Table 5:** Probit analysis and regression analysis of EI rates of 1<sup>st</sup> instar larvae of *Aedes aegypti*.

Hours	$LC_{50}$	95% Fiducial CL	Slope	Intercept
24	2.47	LF (0.96)	-1.06	5.41
		UFL (6.37)		
48	2.10	LFL (0.79)	-1.06	5.34
		UFL (5.56)		
72	1.63	LFL (0.59)	-1.08	5.24
		UFL (4.50)		

Where  $LC_{50}$  stands for Lethal concentration at 50%.

LFL stands for lower fiducial limit.

UFL stands for upper fiducial limit.



**Fig 6:** EI of leaf extract against 2<sup>nd</sup> instar larvae of *Aedes aegypti* shown through percentage EI. Larvae cultured in the absence of tomato leaf extract were treated as – control whereas, larvae cultured in full strength tomato leaf extract were treated as + control. (The bars above the graph show standard error of the mean)

Crude extract (10, 20, 30 and 40 ppm) of tomato leaf was tested against second instar larvae of *Aedes aegypti* for EI (figure 6). It was visually examined that maximum EI was achieved 82.6% in 40 ppm concentration level having average survival (1.7) after 72 hours of exposure. Followed by 78.7% of EI in 40ppm concentration level having average survival 2.0 after 24 hours of exposure.

Whereas lowest EI was checked visually being 8.7% at 10ppm concentration after 72 hours of exposure.

The probit analysis and regression analysis were done for the EI of 2nd instar larvae in the crude leaf extract at different concentrations at 24,48 and 72 hours interval and found that the highest EI rate (82.6%) at 40ppm concentration after 72 hours exposure.

The lowest value of LC<sub>50</sub> (2.41) was optically canvassed after 48 hours of exposure (table 6). Likewise 95% fiducial limits for LC<sub>50</sub> after 48 hours is 0.96 - 6.05 and the slope is -1.13. Followed by the LC<sub>50</sub> (2.82) after 24 hours of exposure with 95% fiducial limits for LC<sub>50</sub> being 1.22- 6.50 and the slope is 1.28 and then after 72 hours of exposure LC<sub>50</sub> (2.95) was optically canvassed with 95% fiducial limits for LC<sub>50</sub> after 72 hours being 1.30 - 6.67.

**Table 6:** Probit analysis and regression analysis of EI of 2<sup>nd</sup> instar larvae of *Aedes aegypti*.

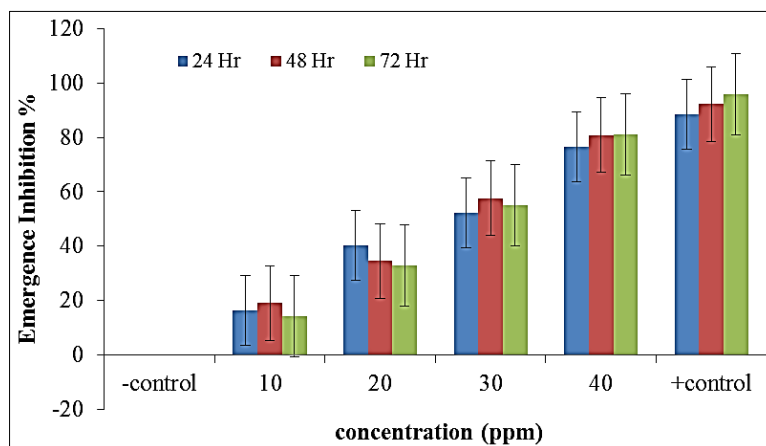
Hours	LC <sub>50</sub>	95% Fiducial CL	Slope	Intercept
24	2.82	LF (1.22)	-1.28	5.58
		UFL (6.50)		
48	2.41	LFL (0.96)	-1.13	5.43
		UFL (6.05)		
72	2.95	LFL (1.30)	-1.30	5.62
		UFL (6.67)		

Where LC<sub>50</sub> stands for Lethal concentration at 50%.

LFL stands for lower fiducial limit.

UFL stands for upper fiducial limit.

Crude extract (10, 20, 30 and 40 ppm) of tomato leaf was tested against third instar larvae of *Aedes aegypti* for EI (figure 7). It was visually examined that maximum EI was achieved 78.3% in 40 ppm concentration level having average survival (2.0) after 72 hours of exposure. Followed by 73.2% of EI in 40 ppm concentration level having average survival 2.3 after 24 hours of exposure. Next being 51.9% of EI in 40 ppm concentration level having average survival 3.7 after 48 hours of exposure.



**Fig 7:** EI of leaf extract against 3<sup>rd</sup> instar larvae of *Aedes aegypti* shown through percentage EI. Larvae cultured in the absence of tomato leaf extract were treated as – control whereas, larvae cultured in full strength tomato leaf extract were treated as + control. (The bars above the graph show standard error of the mean)

Whereas lowest EI was checked visually being 6.7% at 10ppm concentration after 24 and 48 hours of exposure.

The probit analysis and regression analysis were done for the EI of 3rd instar larvae in the crude leaf extract at different concentrations at 24, 48 and 72 hours interval and found that the highest EI rate (78.3%) at 40ppm concentration after 72 hours exposure.

The lowest value of LC<sub>50</sub> (1.14) was optically canvassed after

48 hours of exposure (table 7). Likewise 95% fiducial limits for LC<sub>50</sub> after 24 hours is 0.32 - 4.10 and the slope is -0.77. Followed by the LC<sub>50</sub> (1.75) after 24 hours of exposure with 95% fiducial limits for LC<sub>50</sub> being 0.61- 5.06 and the slope is -0.95 and then after 72 hours of exposure LC<sub>50</sub> (2.56) was visually examined with 95% fiducial limits for LC<sub>50</sub> after 72 hours being 1.03 - 6.41.

**Table 7:** Probit analysis and regression analysis of EI of 3<sup>rd</sup> instar larvae of *Aedes aegypti*.

Hours	LC <sub>50</sub>	95% Fiducial CL	Slope	Intercept
24	1.75	LF (0.61)	-0.95	5.24
		UFL (5.06)		
48	1.14	LFL (0.32)	-0.77	5.05
		UFL (4.10)		
72	2.56	LFL (1.03)	-1.10	5.46
		UFL (6.41)		

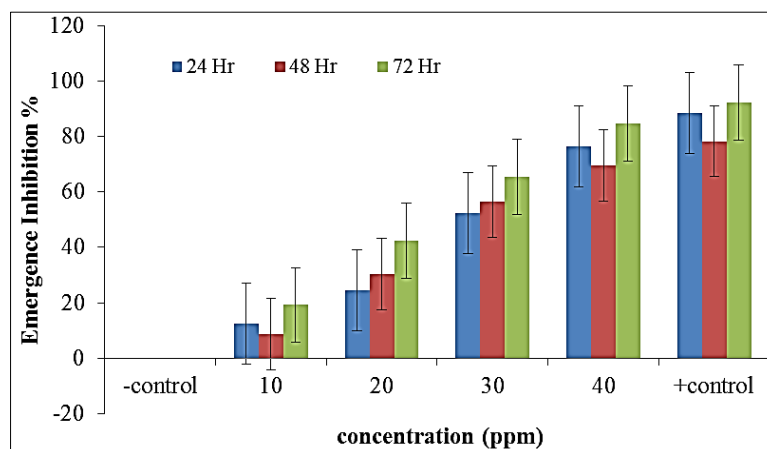
Where LC<sub>50</sub> stands for Lethal concentration at 50%

LFL stands for lower fiducial limit

UFL stands for upper fiducial limit

Crude extract (10, 20, 30 and 40 ppm) of tomato leaf was tested against fourth instar larvae of *Aedes aegypti* for EI (figure 8). It was optically canvassed that maximum EI was achieved 70.0% in 40 ppm concentration level having average

survival (2.7) after 72 hours of exposure. Followed by 65.7% of EI in 30 ppm concentration level having average survival 3.0 after 72 hours of exposure.



**Fig 8:** EI of leaf extract against 4<sup>th</sup> instar larvae of *Aedes aegypti* shown through percentage EI. Larvae cultured in the absence of tomato leaf extract were treated as – control whereas, larvae cultured in full strength tomato leaf extract were treated as + control. (The bars above the graph show standard error of the mean)

Whereas lowest EI was visually examined being 7.3% at 10ppm concentration after circadian of exposure.

The probit analysis and regression analysis were done for the EI rates of 4th instar larvae in the crude leaf extract at different concentrations at 24, 48 and 72 hours interval and found that the highest EI rate (70.0%) at 40ppm concentration after 72 hours exposure.

The lowest value of LC<sub>50</sub> (0.72) was optically canvassed after 48 hours of exposure (table 8). Likewise 95% fiducial limits for LC<sub>50</sub> after 48 hours is 0.17 - 3.11 and the slope is -0.68. Followed by the LC<sub>50</sub> (2.36) after 72 hours of exposure with 95% fiducial limits for LC<sub>50</sub> being 0.93- 5.96 and the slope is -1.11 and then after 24 hours of exposure LC<sub>50</sub> (2.73) was visually examined with 95% fiducial limits for LC<sub>50</sub> after 24 hours being 1.09 - 6.83.

**Table 8:** Probit analysis and regression analysis of EI of 4<sup>th</sup> instar larvae of *Aedes aegypti*.

Hours	LC <sub>50</sub>	95% Fiducial CL	Slope	Intercept
24	2.73	LFL (1.09)	-1.08	5.47
		UFL (6.83)		
48	0.72	LFL (0.17)	-0.68	4.90
		UFL (3.11)		
72	2.36	LFL (0.93)	-1.11	5.41
		UFL (5.96)		

Where LC<sub>50</sub> stands for lethal concentration at 50%

LFL stands for lower fiducial limit

UFL stands for upper fiducial limit

#### 4. Discussion

Dengue is one of the deadly VBDs transmitted by female *Aedes aegypti*. It has been reported that more than 2 billion people around the world are at the risk of dengue virus. Dengue pyrexia, dengue hemorrhagic pyrexia, and dengue shock syndrome which may sometime affect the central nervous system involution are the more rigorous forms of diseases (Hag *et al.*, 1999; Pluempanupat *et al.*, 2013; Dhiman *et al.*, 2010). Mosquitoes control, postulates ecumenical paramountcy. So to achieve immediate results utilization of puissant synthetic insecticides were utilized

particularly in situations of epidemic outbreak and sudden increase of adult mosquitoes (Nathan *et al.*, 2006) [18]. In spite of, their pesticidal efficiency, they created many quandaries, such as insecticide resistance (Subramanian and Kovendann, 2012) [25] hazardous to human health and cause water pollution (WHO, 1992). The dangerous vector-borne diseases could be efficaciously dealt by the botanical insecticides at the individual as well as at the community level (Liu *et al.*, 2005) [16]. Plant based herbicides basically contain secondary metabolites that serve as defence mechanism of plants to protect them from herbivore predators and other environmental factors. Several phytoextracts like, steroids, phenolics, alkaloids, terpenoids and essential oils from different plants have been reported for their insecticidal activities (Shalan *et al.*, 2005) [24]. The plant based insecticide have been given consequentiality due to non-toxic, biodegradable and eco-convivial nature to overpower the synthetic insecticides in the control of various VBDs (Vohora and Kumar, 1971; Yadav and Agarwal, 2011; França *et al.*, 2021) [6, 29, 33]. Tomato has a large number of secondary metabolites such as lycopene, phytoene, phytofluene, ascorbic acid and polyphenols including quercetin, kaempferol, naringenin (George *et al.*, 2010; Afreen *et al.*, 2016) [1, 10]. Nutrients like pro-vitamin A, vitamin C, vitamin E, vitamin K, vitamin B, along with phosphorus, Sulphur, potassium, calcium, iron present in significant amounts. Sugars like polysaccharides, disaccharides, aldoses, ketoses, mainly starch, proteins and amino acids and small quantities of fats. All of these contribute significantly to the antioxidant activity of tomato (Omodamiro and Amechi, 2013; Bhowmik *et al.*, 2012; Kumar *et al.*, 2021) [3, 20].

The present study evaluates the effect of crude leaf extract against all the four larval stages of *Aedes aegypti*. It was optically canvassed that the highest mortality being 93.1% was achieved in 40ppm and 30ppm concentration level of leaf extract after 72 hours of exposure in the 1st instar larvae of *Aedes aegypti*. Whereas the lowest mortality percent was found being 25.9% at 10 ppm concentration at 48 hours of exposure in the first instar larvae of *Aedes aegypti* which



designates its potential to be used as an ideal biopesticide for the control of *Aedes aegypti*.

## 5. Conclusion

In the present study the effect of environmentally benign and renewable source, tomato leaf extract against larvae of *Aedes aegypti* was found to be highly effective as high mortality rates were observed probably due to the presence of the active metabolites possessing phenolic compounds, carotenoids and many vitamins which are used as antioxidants to neutralize free radicals and remove the toxins from the body. Besides biological activity of the crude extract, it contains potential cytotoxic activity (Raiola *et al.*, 2014; Afreen *et al.*, 2016; Trease and Evans 1996; Tiwari *et al.*, 2011) [1, 14, 23, 28]. The present investigation revealed that the leaf of *Lycopersicon esculentum* has a potential source of serviceable drugs due to the presence of phyto extracts and can be utilized in the treatment of many diseases. It is a boon for the development of non-toxic, environmental acceptable pesticide. The future scope studies are required to isolate the active component involved in the mode action from the crude plant extract for opportune effective anti-mosquito control programme.

**Conflict of Interest:** None.

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