



## EFFICACY OF NEEM OIL FOR THE ERADICATION OF AQUATIC INSECTS IN FISH NURSERIES

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### AUTHORS' CONTRIBUTIONS

This work was carried out in collaboration between both authors. Author SS designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author ASR guide for work. Both authors read and approved the final manuscript.

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### ABSTRACT

Generally, the survival rate of carp spawn during the nursery phase is very low due to the presence of harmful aquatic insects in fish nursery pond, which prey heavily upon the spawn and early stages of fry. Insects usually found in large numbers in ponds over the greater part of the year especially during and after rains. They injure the spawn and some of them prey upon spawn. Insects should be eradicated prior to stocking to ensure maximum survival of the spawn. Aquatic insects and their larvae compete for food with the young fish and also cause large scale destruction of hatchlings in nurseries. A study was carried out by the authors to investigating the possible use of natural chemical Neem Oil for the control of aquatic insects in nursery ponds. Bioassay studies were conducted in 5 glass aquaria and using four concentration 0.003, 0.004, 0.005, 0.006 ppm of Neem Oil (natural chemical) on four experimental insects *Notonecta* (Backswimmer), Dragonfly Nymph, *Eretes* (Small beetle), *Nepa* (Water Scorpion). The  $LC_{50}$  value of Neem Oil for the experimental insects (*Notonecta*, Dragonfly Nymph, Small beetle, Water Scorpion) ranged 0.0033 ppm for *Eretes* and *Nepa* at 6 hrs and 0.0040 for *Notonecta* at 5 hrs. Further, the results on toll rate showed that Backswimmer, Small beetle, Water Scorpion killed within 6hrs when exposed to a concentration of 0.004 ppm Neem oil. This was the lowest dose of Neem oil which killed the predatory insects within the desired period of 6 hrs and the death rate of fish seed at this dose is zero. Therefore, the use of Neem oil @0.004 ppm is recommended for the eradication of predatory insects from fish nursery ponds.

**Keywords:** Aquatic insects; *Notonecta* (Backswimmer); dragonfly nymph; *Eretes* (Small beetle); *Nepa* (Water scorpion); mortality; neem oil;  $LC_{50}$ ; physico – chemical parameters of water.

### 1. INTRODUCTION

India is the second largest fish production country in the world accounting for 6.56% of global production and contribution about 1% to the country's Gross Value Added (GVA) and over 5.37% to the agricultural GVA, fisheries and aquaculture continue to be an important source of food, nutrition, income and livelihood to million people. Fish culture in the developing countries, especially in India, is

undoubtedly hundreds of years old. The survival of fish seed in nursery pond is one of the constraints in large scale aquaculture in the country. The importance of fish culture is very significant in our country as it can contribute a major part of proteinous food to the people and also generate employment opportunities. Generally, the survival rate of carp spawn during the nursery phase is very low, there are many factors which contribute to this, but the most important appears to be the presence to the large number of

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aquatic insect which preys heavily upon the spawn. It has been reported that the soap oil emulsion application control most of surface breathing insects, but some subsurface breathers remain unaffected. At the same time the cost of soap oil emulsion is much higher (>Rs5000/ha) for the eradication of aquatic insects in nursery ponds. There has been a surging increase in the use of agricultural chemicals like to preserve the standing crops from the attack of pests and to boost up production, to meet the ever-increasing food demand of the human population [1]. Indian Pesticides may affect the population dynamics by increase death rate and controlling the reproduction rate of freshwater zooplankton and poses an ecological risk [2]. Aquatic insects also part of the aquatic ecosystem. Aquatic insects have wide feeding habits majority of them in their larvae or adult stages prey upon fish seeds and also compete with them for food. For the same reason increased uses of chemical insecticide for the eradication of aquatic insects in aquaculture for getting bulk of production of fishes. Mostly synthetic Pyrethroid used as an insecticide in aquaculture. Indian scenario India is the largest or bigger produce of pesticides in Asia and it got 12<sup>th</sup> rank in the world for the use of pesticides [3]. These pyrethroic acids are strongly lipophilic and they rapidly penetrate many insects and paralyze their nervous system. Pyrethroid has a large class of insecticides, which are effective against a wide range of insect pests of the orders Coleoptera, Diptera, Hemiptera, Hymenoptera, Lepidoptera, Orthoptera and Thysanoptera [4]. Toxicity effects of cypermethrin accrued in natural plankton community [5] and reduced zooplankton density in water [6]. Synthetic pyrethroids are extremely toxic to aquatic organisms, fish fry, and insects [4]. Acute toxicity of cypermethrin to some fresh water fishes and invertebrates and cypermethrin caused fact and biological effects in fresh water pond [7,8]. The photostable synthetic pyrethroid insecticides are quite toxic to many species of fish but very toxic to the early life stage of fish Pyrethroid caused many hazards due to its chemicals [9]. Aquatic insects constitute about 4% of the total insect fauna existing in world. Any water area is invariably explored by the large number of insects either in their adult or larval stages. Their morphological features are highly modified to enable them to adapt to an aquatic mode of life. Various adaptations found in insects and their larvae make them highly interesting from the academic point of view. Eradication of harmful insects from ponds plays a very important role in increasing fry survival rate.

Considering the present proposal has been undertaken to assess the effectiveness of Natural chemical Neem

Oil for the control of predatory insects from fish nursery ponds.

## 2. MATERIAL AND METHODOLOGY

### 2.1 Selection of Test Insects

For the present study, four harmful same aquatic insects were selected. These insects were selected based on of their abundance and distribution in fish farm. *Notonecta* (Backswimmer), Dragonfly Nymph, *Eretes* (Small beetle), *Nepa* (Water Scorpion) were selected for the Neem oil toxicity bioassay test. These insects are widely found in the Fish Farm, Udaipur. Insects were collected by netting in pond water.

### 2.2 Selection of Dose of Natural Plant Based Chemical Neem Oil Insect Eradicant for Investigation

The four concentrations 0.003, 0.004, 0.005, 0.006 ppm of Neem oil were selected for the static bioassay toxicity test on selected experimental insects. These doses were selected for the present study was based on their quick and assumed killing property. Raw Neem Oil can be obtained through the pressing of the Neem seed kernel and leaf both through cold pressing.

### 2.3 Experimental Techniques

**A) Experimental design:-** To select the optimum doses of selected natural chemical Neem oil a preliminary study will be conducted in 5 glass aquaria of 50 litres of water capacity were used for this purpose, for this purpose a wide range of selected insecticides were used their optimum dose was desired time period (i.e.96 hrs.). The static bioassay tests will be conducted under laboratory conditions. 5 glass aquaria of 50 litres were setup in the laboratory. Glass aquaria filled with water and optimum doses of insecticide dropped in glass aquaria by the using of the digital mini-micro pipette. 0.003, 0.004, 0.005, 0.006 ppm concentration of neem oil applied respectively in four water filled aquaria for experimental treatment and one water filled aquaria used as control treatment at 0.00 ppm. 10-10 specimen of *Notonecta*, *Dragon Fly Nymph*, *Eretes*, and *Nepa* were introduced in each experimentally treated glass aquaria and placed under observation for 96 hours and monitored continuously until the death of the insects. The insecticide concentrations gave 0-100% mortality during the bioassay test; the criterion for mortality was the absence of the movement of the insects when prodded by a glass rod and noticed percentage mortality of insects. This experiment was replicated three times.

**B) Studies on the effect of Neem oil on selected four aquatic insects:-** Percentage mortality was recorded at per hour till 100% mortality of experimental insects and  $LC_{50}$  value were calculated by Finney probit analyse method [10] in per hour.

**C) Monitoring of Physico -chemical water quality:** - Physico -chemical water quality parameters of each set of experimental water were monitored at different time intervals. Water temperature was recorded using a Celsius thermometer; pH was measured directly using digital pH meter (HI 991001, HANNA). Conductivity Measured by digital conductometer. Dissolve Oxygen, hardness, Total alkalinity, salinity were analyzed by following the standard method of APHA [10].

**D) Effect of insect effective dose on indian major carp seed:-** The insect effective dose of 0.004ppm neem oil was selected on the basis of preliminary experiments conducted using a wide range of concentrations. The effective dose of 0.004ppm neem oil in triplicate were applied in water filled 50 litre glass aquaria than 10 spawn and 10 fry were stocked in each glass aquaria respectively at different time intervals called here after as “application time”. This time duration or time of application was fixed at 0-6, 6-12, 12-18 and 18-24 hours. The percentage of mortality was calculated at the end of the time duration. Physico – chemical parameters Water temperature, pH, Conductivity, Dissolve Oxygen, hardness, Total alkanity, salinity of each set of experimental water was monitored at different time intervals.

### 3. RESULTS

#### 3.1 Selection of the Optimum Dose of Neem Oil

The optimum dose of Neem Oil the control of aquatic insects viz. *Notonecta*, *Eretes*, *Nepa* and Dragonfly nymph has been selected on the basis of 100% insect mortality in bioassay studies. The results related to

insect mortality with different doses of Neem oil and  $LC_{50}$  values are presented in Tables. 1 to 5 and the values of water quality parameter are presented in Table 6 and Fig 1.

*Notonecta* showed 30% mortality at lowest concentration 0.003ppm in 2 hours. In this concentration mortality increases to 60%, 80% in 3 and 4 hours respectively and 100% mortality observed by the end of 5 hours. In case of 0.004 ppm concentration of Neem – oil for *Notonecta* mortality observed 10% in 1 hour and then mortality increases to 30%, 70%, 90% in 2, 3, 4 hours respectively and 100% mortality obtained at the end of 5 hours. 20% mortality of *Notonecta* observed in 1 hour at 0.005 ppm concentration and mortality increases to 60% in 2 hours and 100% mortality is recorded at the end of 3 hours. In the case of 0.006 ppm concentration 40% mortality observed in 1 hour of *Notonecta* and 100% mortality in 2 hours at highest concentration 0.006 ppm of Neem-oil.  $LC_{50}$  values were calculated 0.0059, 0.0038, 0.0041, 0.0035 and 0.0040 ppm at 1, 2,3,4,5 hours respectively for *Notonecta* in neem oil toxicity test.

In the case of *Eretes* 20% mortality observed at lowest concentration 0.003 ppm in 1 hour. The mortality increases to 50%, 60%, 70% in 3, 4, 5 hours respectively and 100% mortality recorded at the end of 6 hours. 10% mortality of *Eretes* recorded at 0.004 ppm concentration in 1 hour. In this concentration minority increases to 30%, 50%, 90% in 2, 3, 4 hours respectively and 100% mortality noticed at the end of 5 hours. In the case of 0.005 ppm Concentration of Neem oil for *Eretes* mortality observed 20% in 1 hour and 40% mortality increases in 2 hours and then mortality increases to 70% in 3 hours and 100% mortality exposed at the end of 4 hours. 20% mortality of *Eretes* observed in 1 hour at 0.006 ppm Concentration and then mortality increases to 50% in 2 hours and 100% mortality observed at the highest concentration .006 ppm of Neem oil in 3 hours.  $LC_{50}$  values were calculated 0.0060, 0.0059, 0.0034, 0.0042, 0.0037, and 0.0033 at 1, 2, 3,4,5,6 hours respectively for *Eretes* in neem oil toxicity test.

**Table 1. Mean mortality (%) rate of *Notonecta* in selected concentration of neem oil**

Exposure time (in hours)	Neem oil doses				
	0.0 ppm	0.003 ppm	0.004 ppm	0.005 ppm	0.006 ppm
1	0	0	10	20	40
2	0	30	30	60	100
3	0	60	70	100	
4	0	80	90		
5	0	100	100		

**Table 2. Mean mortality (%) rate of *Eretes* in selected concentration of neem oil**

Exposure time (in hours)	Neem oil doses				
	0.0 ppm	0.003 ppm	0.004 ppm	0.005 ppm	0.006 ppm
1	0	0	10	20	20
2	0	20	30	40	50
3	0	50	50	70	100
4	0	60	90	100	
5	0	70	100		
6	0	100			

**Table 3. Mean mortality (%) rate of *Nepa* in selected concentration of neem oil**

Exposure time (in hours)	Neem oil doses				
	0.0 ppm	0.003 ppm	0.004 ppm	0.005 ppm	0.006 ppm
1	0	0	10	20	30
2	0	10	30	50	60
3	0	30	50	80	100
4	0	50	90	100	
5	0	70	100		
6	0	100			

**Table 4. LC<sub>50</sub> of neem oil at different time intervals for experimental insects**

Name of insects	Time intervals in hrs.					
	1	2	3	4	5	6
Notonecta	0.0059	0.0038	0.0041	0.0035	0.004	-
Eretes	0.006	0.0059	0.0034	0.0042	0.0037	0.0033
Dragon Fly	*	*	*	*	*	*
Nepa	0.0058	0.0051	0.0036	0.0029	0.0036	0.0033

\* Nil mortality

In the case of *Nepa* 10% mortality noticed at lowest Concentration 0.003ppm in 2 hours. The mortality increases to 30%, 50%, 70% in 3, 4, 5 hours respectively and 100% mortality recorded at the end of 6 hours. 10% mortality of *Nepa* observed in 0.004 ppm concentration in 1 hour. In this Concentration mortality increases to 30% in 2 hours and then mortality increases to 50%, 90% in 3 and 4 hours respectively and 100% mortality observed at the end of 5 hours. In case of 0.005 ppm concentration of Neem-Oil for *Nepa* mortality noticed 20% in 1 hour and mortality increases to 50%, 80% in 2 and 3 hours respectively and 100% mortality exposed at the end of 4 hours. 30% mortality of *Nepa* observed in 1 hour at 0.006 ppm Concentration and mortality increases to 60% in 2 hours at highest Concentration and 100% mortality observed in 3 hours at 0.006 ppm Concentration of Neem-oil. LC<sub>50</sub> values were calculated 0.0058, 0.0051, 0.0036, 0.0029, 0.0036, 0.0033ppm at 1, 2, 3, 4, 5, 6 hours respectively for *Nepa* in neem oil toxicity test.

In case of Dragonfly No mortality observed at different concentration of Neem oil within 3 days. In the case of Control there was no mortality of insects observed.

The various parameter with reference to the concentration of neem oil are detailed here which shows the general temperature status which ranges control from 25 – 26.8°C and pH of the water here range from 7.3-7.8., 6.62-8.62ppm value to be selected for the dissolved oxygen factor of water here while hardness of water which is measured as 149.9-163.3 ppm and salinity range from 0.4-0.4 ppt, conductivity of the selected water here measured in between the range of 793.5-806.1µs/cm and alkanity of the experiment water here range from 161.30-172.01 ppm. At 0.003ppm here general temperature of selected water with reference to neem oil here range from 25.7-26.6° and pH value is consider here from 7.2-7.8, dissolved oxygen of the selected water here range from 5.89-8.38 ppm and hardness of water maintained as 151.2-168.2 ppm., the TDS value here measured between 382-399.6 ppm and salinity selected here as 0.4-0.4ppt, conductivity of the water measured from 796.48-807.3 µs/cm and alkanity of experimental water were maintained 148.78-159.6 ppm. At the 0.004 ppm concentration of neem oil, the temperature status maintained in between 25.8-26.3°C and pH value range from 7.2-7.9 where dissolved oxygen of the treated water here measured between 6.02-8.52 ppm and hardness of water is selected from

142.2-166 ppm and TDS value range from 379.3-396.89 ppm while salinity is considered as 0.4-0.4 ppt, conductivity of the water here measured in between 789.3-800.68  $\mu\text{S}/\text{cm}$  and alkanity of water calculated in between 167.10-178.43 ppm which determines the basicity of water. For 0.005 ppm concentration, temperature is maintained as 25.7-26.5°C and pH of the water is selected from 7.1-7.9. Dissolved oxygen of the water here recorded in between 5.98-8.83 ppm while the hardness of water here maintained from 144-167.1 and TDS value here range from 383.8-401.2 ppm whereas salinity recorded at same 0.4-0.4ppt, conductivity of water to measured between 783-806.2 $\mu\text{S}/\text{cm}$  and alkanity of the experiment water calculated from 171.02-181.36ppm at 0.005ppm concentration of neem oil. At 0.006 ppm concentration the general temperature status which range control from 25.3-26.9°C and pH of the water here range from 7.2-7.8., 5.68-892 ppm value to be selected for the dissolved oxygen factor of water here while hardness of water which is measured as 143.2-169.2 ppm and salinity range from 0.4-0.4ppt, conductivity of the selected water here measured in between the range of 798.2-811.2  $\mu\text{S}/\text{cm}$  and alkanity of the experiment water here range from 170-177.57 ppm.

### 3.2 Effect of Insect Effective Dose on Indian Major Carp Seeds

Carp seeds were safe from the starting to treatment no mortality found till end of 24 hours and after 24 hours on effective dose 0.004 ppm of Neem oil. While the value of water quality parameters have been summarized in Table 6. It would be seen from this table that there was no significant alteration in the quality of the experimental water.

While within a one hour when just after applied Neem oil in experimental water while water parameter here where general temperature of Neem Oil treated water maintained here 25.8°C and pH of water 7.8, dissolved Oxygen which is 7.30 ppm while hardness of water here measured 152 ppm. TDS of experimental water considered in 380 ppm and salinity of treated water selected in 0.4 ppt, conductivity of the experimental water measure as 806 $\mu\text{S}/\text{cm}$  and alkanity of the water noticed in 146ppm after applying 0.004 ppm Neem Oil dose in water. After 6 hours of the experiment general temperature status here 25.8°C and pH of treated water was 7.8, Dissolved oxygen of the selected at 6 hours of treatment water here 7.36 ppm and hardness of water 152.40 ppm, TDS values of the water determine was 381 ppm. Salinity of water noticed 0.4ppt. Conductivity of the experimental water measured 807  $\mu\text{S}/\text{cm}$  and Alkanity of water 143ppm

was measured at the end of 6 hours. Whereas at 12 hours of treatment of Neem Oil (from the Table 6) water temperature was recorded 25.6°C, pH of water was 7.8. Dissolved oxygen of the selected after 12 hours of treatment water here 7.33 ppm and hardness of water 152 ppm, TDS values of the water determine was 380 ppm, Salinity of water noticed 0.4 ppt, Conductivity of the experimental water measured as 809  $\mu\text{S}/\text{cm}$  and Alkanity of water 146ppm were recorded. While at 18 hours of treatment water temperature was 25.7, pH of water was 7.8. Dissolved oxygen of the selected at 18 hours of treatment water here 7.26 ppm and hardness of water 152.80 ppm, TDS values of the water determine was 382 ppm. Salinity of water noticed 0.4ppt. Conductivity of the experimental water measured 804 $\mu\text{S}/\text{cm}$  and Alkanity of water 145ppm. But at 24 hours of the treatment water temperature was 25.8, pH of water was 7.9. Dissolved oxygen of the selected at 24 hours of treatment water here 7.28 ppm and hardness of water 152.80 ppm, TDS values of the water determine was 382 ppm. Salinity of water noticed 0.4ppt. Conductivity of the experimental water measured 804 $\mu\text{S}/\text{cm}$  and Alkanity of water 147 ppm.

### 4. DISCUSSION

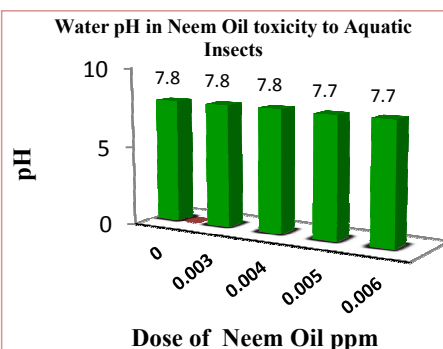
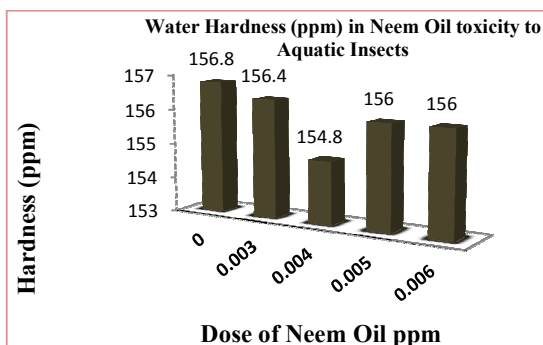
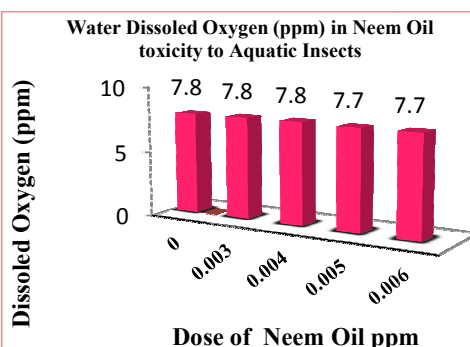
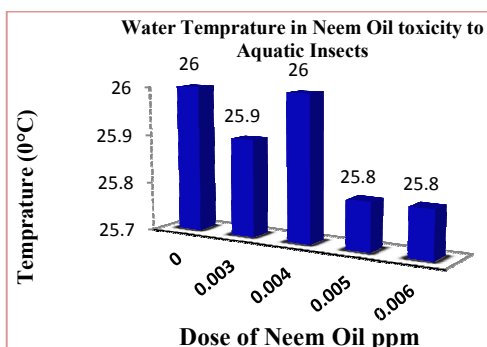
The discovery of natural chemical insecticide revolutionized the concept of aquatic insect control, thereby contributing to higher production from fish nurseries and aquaculture. The applicability of different insecticide in the aquatic realm is species dependent, besides, of course, their efficacy again depends on water quality for example, toxicity or the functional efficiency may increase with increasing temperature and insecticides also change water quality parameters after applying chemical insecticide in pond water [12]. However, the present study state that differences in water quality among different bioassay tests were very little in used Neem Oil toxicity test to showed in Fig. 1. The neem oil (*Azadirachta indica*) used the malaria vector *Anopheles* at the lower concentration of neem oil 6ppm and found a positive result [13,14]. Neem based insecticides products are economically cheaper and safe to the environment [15]. The plant based insecticide is less toxic compared to synthetic chemical insecticides to aquatic non-target organisms and reduce the chemical load of environment [16] and Natural pesticides can be easily isolated from plants comparative to chemical pesticides and they can rapidly bio-degradation in environment [17] Natural insecticide having an eco-friendly features and safety environmental issue and chemical insecticide caused a harmful effect in environment and living organisms [18]. Some plant oil and products can use for the eradication of aquatic insects from fish nurseries and they are economically cheap for fish farmers [1]. Neem oil is extracted from

**Table 5. The range and mean values of selected water quality parameters in neem oil toxicity to aquatic insect experiment**

<b>Doses of neem oil ppm</b>	<b>Parameters</b>							
	<b>Temperature (0°C)</b>	<b>pH</b>	<b>Dissolved oxygen (ppm)</b>	<b>Hardness (ppm)</b>	<b>TDS (ppm)</b>	<b>Salinity (ppt)</b>	<b>Conductivity (µS/cm)</b>	<b>Alkalinity (ppm)</b>
0	25.6 - 26.8 (26)	7.2 - 8.0 (7.8)	6.62 - 8.62 (7.83)	149.9 - 163.3 (156.8)	382.1 - 397.2 (392)	0.4 - 0.4 (0.4)	793.5 - 806.1 (802)	161.30 - 172.01 (168)
0.003	25.7 - 26.6 (25.9)	7.3 - 7.8 (7.8)	5.89 - 8.38 (7.86)	151.2 - 168.2 (156.4)	382 - 399.6 (391)	0.4 - 0.4 (0.4)	796.48 - 807.3 (801)	148.78 - 159.63 (154)
0.004	25.8 - 26.3 (26)	7.2 - 7.9 (7.7)	6.02 - 8.52 (7.9)	142.2 - 166 (154.8)	379.3 - 396.89 (387)	0.4 - 0.4 (0.4)	789.3 - 800.68 (794)	167.10 - 178.43 (172)
0.005	25.7 - 26.5 (25.8)	7.1 - 7.9 (7.7)	5.98 - 8.83 (7.93)	144 - 167.1 (156)	383.8 - 401.2 (390)	0.4 - 0.4 (0.4)	783 - 806.2 (799)	171.02 - 181.36 (176)
0.006	25.3 - 26.9 (25.8)	7.2 - 7.8 (7.7)	5.68 - 8.92 (7.93)	143.2 - 169.2 (156)	381.2 - 397.2 (390)	0.4 - 0.4 (0.4)	798.2 - 811.2 (800)	170 - 177.57 (174)

the Neem tree and has insecticidal and medicinal properties due to which it is used as pest control. [19] Neem oil and neem cake can be used in insect control and Neem borne products have effective and eco – friendly features and get by easy production at low cost and neem products are ideal insecticides which deal with insects control and Neem oil, neem cake and their fractions acts as promising ovicides, larvicides, pupicides and oviposition deterrents against important arbovirus and malarial mosquitoes vectors [20]. The result of present study gives positive results of Neem oil against aquatic insects. In the case of *Notonecta* LC<sub>50</sub> 0.0059 ppm was at 1 hour which is the highest LC<sub>50</sub> value recorded and the least LC<sub>50</sub> value 0.0038 ppm was at 2 hours. LC<sub>50</sub> value 0.0041, 0.0035 and 0.0040 was at 3, 4 and 5 hours respectively observed for suitable Neem oil dose. Where for *Eretes* the least LC<sub>50</sub> value of Neem oil is 0.0033 ppm found at 6 hours while the highest LC<sub>50</sub> 0.0060 ppm at 1 hour. LC<sub>50</sub> 0.0059 was at 2 hour and LC<sub>50</sub> value of Neem oil for 3, 4 and 5 hours is 0.0034, 0.0042 and .00037 ppm respectively recorded. *Nepa*, where the highest LC<sub>50</sub> value on Neem oil 0.0058 ppm is at 1 hour and the lowest 0.0029 ppm was at 4 hours LC<sub>50</sub> values 0.0051 and 0.0033 ppm were at 2 and 6 hours respectively and 0.0036 ppm LC<sub>50</sub> value is at 3 and 5 hours simultaneously. In case of Dragonfly No mortality observed at different concentration of Neem oil within 3 days. Dragonfly

Nymph may not be killed due to specific breathing habits. Dragonfly Nymph living in bottom of freshwater pond and adult fly are fast agile flyers and migrate across the pond however *Notonecta* and beetles are presents in large number in freshwater fish nurseries and heavily predator on fish larvae. *Notonecta*, *Eretes* and *Nepa* are sensitive to Neem oil. *Notonecta* (backswimmer), *Eretes* (small beetle), *Nepa* (water scorpion) were showed 100% on 0.004ppm doses of neem oil within 6 hours. Higher doses at more than 0.004ppm of neem oil caused 100% mortality within 3hours that are more toxic while at lower concentration of neem oil 0.003ppm caused 100% mortality within 7 hours. In view of mortality rates and exposure time, it can be considered that a doses of 0.004ppm. However Carp seeds were safe from the starting to treatment no mortality found till end of 24 hours and after 24 hours on effective dose 0.004 ppm of Neem oil and there was no significant alteration in the quality of the experimental water and as such the water quality fit for the survival of fish seeds of addition of 0.004 ppm dose of raw Neem Oil. The cost of treatment for nursery pond with Neem Oil comes to 300₹ against 5000₹ per hector in conventional method. Neem oil is more suitable for the control of predatory aquatic insects especially, *Notonecta* sp., *Eretes* sp. and *Nepa* sp.



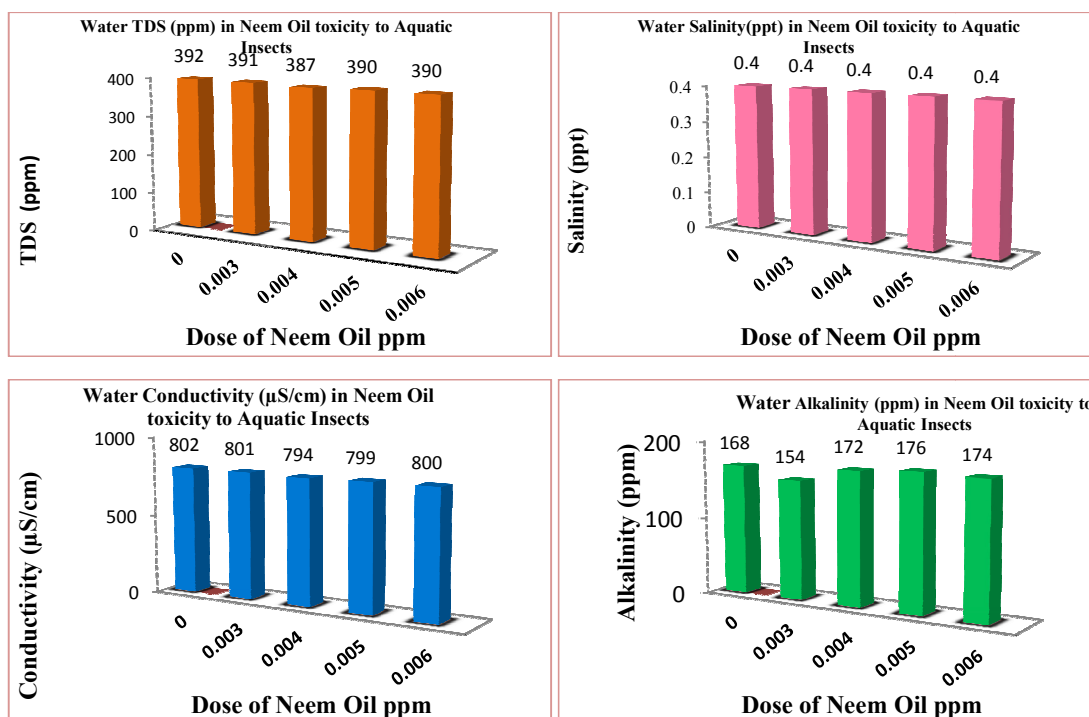


Fig. 1. Graphical representation of different quality parameters in neem oil extract

Table 6. The values of selected water quality parameters in Neem oil toxicity to carp seed experiment

Time duration (in hours)	Parameters							
	Temperature (°C)	pH	Dissolved oxygen (ppm)	Hardness (ppm)	TDS (ppm)	Salinity (ppt)	Conductivity (μS/cm)	Alkalinity (ppm)
0	25.8	7.8	7.30	152	380	0.4	806	146
6	25.8	7.8	7.36	152.40	381	0.4	807	143
12	25.6	7.8	7.33	152	380	0.4	809	146
18	25.7	7.8	7.26	152.80	382	0.4	803	145
24	25.8	7.8	7.28	152.80	382	0.4	809	147

## 5. CONCLUSION

The present investigation indicated that the application of Neem Oil has no adverse effect on water quality (Fig. 1) as there was no significant change in water quality over control. However, the application of neem oil has discovered the significant role of it in the eradication of aquatic insects. 0.004ppm concentration is useful to insect eradication from fish nurseries and the use of Neem Oil is much cheaper than that of conventional method and can be easily obtained through the pressing of the Neem seed kernel and leaf both through cold pressing. A Newer biological insecticide is developed to replace deleterious chemical insecticide in fish nurseries.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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