

## Effects of four biopesticides on three different species – A Comparative Study

Gayathri P K\*, Arun D, Sripriya R, Soniya E, Vennila R, Felcitta Aarthi R

Department of Biotechnology, Vel Tech High Tech Dr. Rangarajan Dr. Sakunthala Engineering College,  
Avadi, Chennai - 600 062.

\*Corresponding author: E-Mail: k.gayathri @velhightech.com

### ABSTRACT

Biopesticides are under high demand owing to the side effects caused by chemical pesticides. The present study aims to obtain aqueous extract from 4 different species viz *Chrysanthemum sp.*, *Gracilaria edulis*, *Enteromorpha intestinalis*, *Chaetomorpha linum* and its respective toxicity effect on live species (Artemia larvae, Rice beetles and *Abelmoschus esculentus* seeds). All the experiments were done in triplicates. *E. Intestinalis* showed best germination rate and pesticidal activity at lower concentration (20%). The best pesticidal activity and larvicidal activity was also observed in *Chrysanthemum sp.* *C. Linum* showed good mortality rate against Artemia followed by *G. edulis* and *E. Intestinalis*. The best activity was obtained in all the toxicity study and it shall be concluded that the biopesticides may be diluted to a larger extent to prevent unwanted cidal effects. This may also ensure the cost effectiveness of the biopesticide production.

**KEY WORDS:** *Chrysanthemum sp.*, seaweeds, larvicidal, pesticidal, phytotoxicity.

### 1. INTRODUCTION

Pesticides are considered as potential molecules to combat insects, pests, diseases and weeds in agricultural, horticultural cropping system as well as health management systems. Pesticides are substances meant for attracting, seducing and then destroying or mitigating any pest (US environmental, 2007). The term chemical pesticide also applies to herbicides, fungicides and various other substances used to control pest. Due to the use of pesticides, it is possible to combat pests and produce larger quantities of food. Pesticides which was discovered recently include 1080 (sodium fluorate), 2,4,5-Trichlorophenoxyacetic acid, 2,4-dichlorophenoxy acetic acid, DDT, endosulfan, methyl bromide. The use of pesticide has been in common practice from ancient times for their yield rate and to get rid of harmful organism and insects. Pesticides of chemical origin are highly effective against the target organism. However, the chemical pesticides have toxic effects on several non-target species and the growing environmental concerns raises the issues of safety and evaluation of toxicity. According to US EPA, more than 70 ingredients known to cause cancer in animal test were allowed for use. Phenoxy compound tend to selectively kill broad weed leaves rather than grasses. The phenoxy and benzoic acid herbicides function similar to plant growth hormones and grow cells without normal cell division, crushing the plant nutrient transport system (Kamrin, 1997). Alternatives to chemical pesticide include method of cultivation, use of biological controls, genetic engineering and methods of interfering with insect breeding (Miller, 2004). Application of composted yard waste has also been used as a way of controlling pest (Mcsorley, 1996). Biopesticides are certain types of pesticides that are derived from natural materials like plants (Botanical origin), bacteria, fungi, virus (Microbial origin) and certain minerals. Biopesticides are less toxic than conventional chemical pesticides. Biopesticides affect only the target and closely related pests, in contrast to chemical pesticides that may affect organisms as different as birds, insects and mammals. Biopesticides are often effective in very small quantities and decompose rapidly which results in lower exposures and avoid large pollution caused by conventional pesticides. Biopesticides can greatly replace the use of conventional pesticides, while crop yields remain high. Growth of biopesticide is projected to outpace the chemical pesticide with compounded annual growth rate of more than 15% (Pamela, 2014). Recently *Moringa oleifera* & *Annona muricata* seed oil has been effectively used as biopesticide (Nwankwo, 2015). In the current study, the toxicity effect of *Chrysanthemum sp.* is compared to that of three different seaweeds and the results are reported.

### 2. MATERIALS AND METHODS

**Materials:** *Chrysanthemum sp.* (marigold), *Abelmoschus esculentus* (lady finger) seeds, Petriplates, Distilled water, Whatman No. 1 filter paper, Artemia eggs, Rice beetles.

**Sample collection and identification:** Fresh seaweeds were collected from Pulicat Lake, Tamilnadu, India, during December 2013. They occur in all seasons. They were hand-picked and washed thoroughly with seawater to remove all the unwanted impurities, adhering sand particles, epiphytes etc. *Chrysanthemum sp.* was collected from the local market by gathering the left out biomass. Both were packed in a plastic bag and transferred to the lab. In the lab, the collected samples were again washed in fresh water to remove the surface salt and then blotted to remove excess water. The seaweeds and the *Chrysanthemum sp.* were identified by the experts (Marine biologists and Plant biologists) in Vel Tech High Tech Dr. Rangarajan Dr. Sakunthala Engineering College.

**Preparation of aqueous extract:** All the samples were shade dried to remove excess moisture content and the samples were either powdered or were cut into small pieces depending on the species. The final products were stored

in air tight container to avoid contaminants and moisture absorption for future use. The coarse powder was boiled with distilled water (1:20 (w/v)) for 60 min, filtered through 4 fold white muslin cloth and the filtrate obtained is considered as 100%. From this mother extract different concentrations (20, 40, 60, 80) were prepared using distilled water.

**Phytotoxicity assay:** The crop plant, selected for the present study was *Abelmoschus esculentus* commonly known as Lady Finger belonging to the family Malvaceae. The seeds were bought from Seed Works India Pvt.Ltd. The seeds belongs to F1 Hybrid US7902. The seeds with uniform size, colour and weight were chosen for the experimental purpose. Seeds were soaked in each aqueous extracts (3/extract) for 24 hours and were placed in petridish to observe the rate of germination of the seeds. The experiment was done in triplicates.

**Pesticidal activity:** Petri dishes were cleanly washed and autoclaved at 15 lbs. Initially the extracts were poured into each of the petridishes with filter paper and methanol was poured into one petridish which acts as a control. Two ml of the extracts was poured into each plate and the plates were labelled. The rice weevils (*Sitophilus oryzae*) were cultivated for two months in laboratory for determining the pesticidal activity of the plants. These weevils were collected from the rice that was stored in rice bags. They were then stored in small plastic containers with some rice for their feed. Ten rice weevils were isolated from the mass weevil cultivation and placed in each of the petriplates along with some rice which act as their feed. They were then placed under observation after sealing the sides with tape. After 24hr and 48hr, reading of the weevils mortality rate was recorded and tabulated. The experiment was repeated twice to get the mean value.

**Larvicidal activity:** A set of four test tubes for each extract was taken and washed cleanly and dried for their usage. All the test tubes were labelled according to their plant name in four dilutions of 1:10, 1:25, 1:50, and 1:100. The dilutions were made by adding salt water to the plant extracts. The larvae were isolated by culturing artemia seeds in salt water. About ten larvae were inoculated into each test tube. All the test tubes were placed in a test tube holder and left overnight and the readings were taken for 24 hrs and 48 hrs for the larvicidal activity. The mortality rate of the larvae was observed and taken as readings and tabulated.

### 3. RESULTS AND DISCUSSION

The aqueous extract of *G. edulis*, *E. intestinalis*, *C. linum* and *Chrysanthemum sp* showed best germination at 40%, 20%, 80%, 40% concentration respectively. In almost all the extracts, germination was absent at the highest concentration (100%). This might be due to the extract of some powerful antioxidants at higher concentration of the species in the aqueous phase. So it is advisable that the biopesticides may be diluted at lower concentration to avoid the phytotoxic affect against healthy plant species. This is in good agreement to the results discussed by Gayathri (2014). The number of dead insects out of 5 in each petriplate was monitored after 24 hours and the results are reported in Table 2. It was observed that considerable amount of insects died in the aqueous extract of *E. intestinalis* and *Chrysanthemum sp*. even at low concentration. *G. edulis* gave better results when at higher concentration whereas *C. linum* have no considerable pesticidal effect. The activity of *C. linum* almost ceased at highest concentration.

The mortality rate of the artemia larvae were calculated and reported in Table 3. The *C. linum* and *Chrysanthemum* extract showed the best larvicidal activity at lowest concentration (20%) whereas the extracts of *G. edulis* and *E. intestinalis* showed good activity at moderate concentrations (60%). All the extracts showed 100% mortality after 48 hrs of test period. Zero percent mortality was observed in control.

The bio pesticides were considered as an alternative for many chemical pesticides due to its adverse effects for environment and human being. The use of bio pesticides shall present a better option for the current scenario. Insect pests were responsible for most of the crop death and destruction. In long term usage chemical pesticides may become toxic to human consumption and may reduce the fertility of the soil. Many widely available plants and seaweeds shall be adopted for the production of bio pesticides. In order to justify the usage of plant and algal species as bio pesticides it is mandatory to test the extracts for phytotoxicity, larvicidal and pesticidal activity. It is desirable that the bio pesticides are harmful only to the live pests and harmless to the seeds under study. It was observed that all the bio pesticides showed good seed germination as well as larvicidal and pesticidal activity.

**Table.1.Germination rate of the aqueous extract**

Name of the species	Concentration of extract				
	20	40	60	80	100
<i>G. edulis</i>	-	100%	80%	-	-
<i>E. intestinalis</i>	100%	80%	-	-	-
<i>C. linum</i>	-	-	60%	100	-
<i>Chrysanthemum sp</i>	40%	60%	-	-	-
Control (Water)	0	0	0	0	0

**Table.2. Insecticidal activity\* of the extracts.**

Name of the species	Concentration of extract				
	20	40	60	80	100
<i>G. edulis</i>	0	0	3	3	3
<i>E. intestinalis</i>	3	3	3	3	3
<i>C. linum</i>	1	1	1	1	0
<i>Chrysanthemum sp</i>	3	3	3	3	3
Control (water)	0	0	0	0	0

\*Note: The numbers of dead rice beetles are provided

**Table.3. Mortality rate of the extracts**

Name of the species	Concentration of extract				
	20	40	60	80	100
<i>G. edulis</i>	0 %	0%	80%	80%	70%
<i>E. intestinalis</i>	0%	70%	70%	60%	50%
<i>C. linum</i>	90%	85%	80%	50%	0%
<i>Chrysanthemum sp</i>	80%	80%	70%	65%	10%
Control (water)	0%	0%	0%	0%	0%

#### 4. CONCLUSION

Many investigations have been made using various plant species belonging to the family Asteraceae (Sharma, 2006; Saxena, 1992; Raj Mohan, 2007), Euphorbiaceae (Rahuman, 2007; Yadav, 2002), Solanaceae (Chowdary, 2008), Leguminosae etc. Similar observations were observed in the present study and the safety of the bio pesticides prepared is not compromised. Further investigations are needed to identify the bioactive compounds in all the samples studied which is responsible for the pesticidal activity. Scale up of the production shall also be considered for vast application of bio pesticides using field trials.

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