# Evaluation of algal bioassay for pesticide toxicity by agar well diffusion method

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# Manuscript details:

Available online on <a href="http://www.ijlsci.in">http://www.ijlsci.in</a>

ISSN: 2320-964X (Online) ISSN: 2320-7817 (Print)

**Editor: Dr. Chavhan Arvind** 

## Cite this article as:

Khobragade Harshwardhan and Nandkar PB (2016) Evaluation of algal bioassay for pesticide toxicity by agar well diffusion method, *Int. J. of Life Sciences*, A6: 86-88.

#### Acknowledgement:

We are obliged to Mr. D. L. Shirodkar, Botanical Survey of India, Industrial Section, Indian Museum, 1, Sudder Street, Kolkata for the opinion and for supply of certain literature.

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# **ABSTRACT**

In the present study, algal bioassay was developed on the basis of agar solid culture lawn. The blue green algae i.e. *Gloeocapsa montana* Kutz and *Microcystis aeruginosa* Kutz were isolated and its clonal culture also prepared in vitro in BG 11 medium. The influence of fungicide Bavistin was tasted on blue green algae to find the inhibition zone on solid agar lawn. The algal toxicity was measured in context with the various coloration and inhibition zone on different concentrations of bavistin. The selected algae *Microcystis aeruginosa* Kutz and *Gloeocapsa montana* Kutz were showed effective zones of inhibition on concentration 6 ppm and 10 ppm respectively.

ISSN: 2320-7817 | eISSN: 2320-964X

 $\textbf{Keywords:} \ \ \textbf{Bioassay,} \ \textit{Gloeocapsa montana, Microcystis aeruginosa} \ \ \textbf{,} \ \ \textbf{Bavistin,} \\ \textbf{toxicity.}$ 

## INTRODUCTION

Agriculture is core of Indian economy. Continuous increasing demand of foods which scared due to increases Global population in restricted natural resources. The economic status of farmers depends on productivity; for that fulfills the expectation they used the pesticide extensively for kill the pest. Farmer is unaware about adverse effect of the pesticide and their safe used to maintain the sustainability (Chowdhury, 2008). Pesticides are substance intended for preventing, destroying, repelling any pest. Pesticides are percolated in ground water and enter into ecosystem. So it is prior need to prevention from pesticide in an atmosphere through the bioremediation. Many scientists work on the algal liquid culture which have commonly used as bioassay for toxicological test on water sample for the assessment of herbicide action (Eberle and Gerber, 1976; Devez, 2004). Well agar diffusion method is an applicable to check the pesticide toxicity response towards the microalgae with their specificity.

## **MATERIALS AND METHODS**

Blue green micoalgae *Gloeocapsa montana* Kutz and *Microcystis aeruginosa* kutz. were collected from paddy field of Bramhapuri taluka, Chandrapur distict (MS). The identification was done of microalgae *Gloeocapsa montana* 

kutz.and *Microcystis aeruginosa* kutz. from monograph of Desikachary (1959). The selected strains were isolated, purified and culture in BG 11 liquid medium in vitro (Robert Andersen, 2005). BGA were cultured using BG-11 Agar solid medium according to the composition of nutrient media given by (Stainer *et al.*, 1971).

## **Prepration of Pesticide Standard:**

prepared the standards different concentration gradient of pesticide (fungicide) Bavistin viz. 2ppm, 4ppm, 6ppm, 8ppm, 10 ppm, 12 ppm in Distilled water.

#### **Sterilization and Culture Condition:**

The BG 11 media and glassware was sterilize in an autoclave for 1 hour at  $121^{\circ}$ C (15psi). Prepared were well on solid agar plate by cork borer.  $100~\mu$ l of Bavistin with selected concentration of 2ppm, 4 ppm, 6ppm, 8ppm, 10 ppm, 12 ppm were loaded on the separate agar well.  $100~\mu$ l Algal inoculums were poured on the agar plate. Provided was optimum laboratory condition for the algal growth maximum (Robert Andersen, 2005).

## **OBSERVATION**

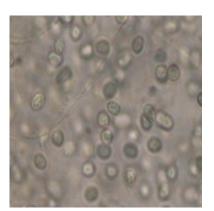
## Taxonomic enumerations of selected BGA:

*Microcystis aeruginosa* **Kutz.** (Desikachary, 1959, pp. 93, pl. 17, fig 1). The unicellular, Colonies when young round or slightly longer than board, solid, when old becoming clathrate, with distinct hyaline colonial mucilage; cells 3-7 u in diameter, spherical, generally with gas vacuole.

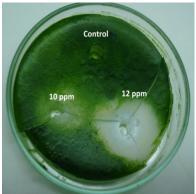
Gloeocapsa montana Kutz. (Desikachary, 1959, pp. 123, pl 24, fig 14). Thallus amorphous, broad, mucilaginous, pale yellowish or light green in color; cells spherical or sub spherical, with sheath 4-10 u broad and without sheath 2-5 broad, single or two together in a colony, colony 13-28 u broad; sheath lamellated, colourless, outer lamellae diffluent; contents more or less opaque, homogeneous, finely granular, pale blue-green.

# **Alga Lawn Formation:**

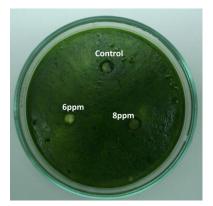
After 10 days observed algal lawn with uniformity where it used for determine the algal inhibition zone for Bavistin (observation table no.1 and plate no. 1).



Gloeocapsa montana Kutz.



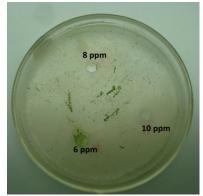
Gloeocapsa montana lawn Agar shows the bavistin inhibition zone



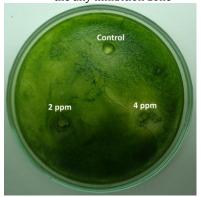
*Gloeocapsa montana.* Does not shows the any inhibition zone



Microcystis aeuroginosa Kutz.



Microcystis aeuroginosa Agar Lawn shows inhibition.



*Microcystis aeuroginosa* lawn shown luxuriant growth.

Table 1:

| Sr. | Bavistin      | Inhibition zone on algal lawn     |                                  |
|-----|---------------|-----------------------------------|----------------------------------|
| No. | concentration | Goleocapsa montana                | Microcystis aeuroginosa          |
| 1   | 2 ppm         | Not observed                      | Not observed                     |
| 2   | 4 ppm         | Not observed                      | Not observed                     |
| 3   | 6 ppm         | Not observed                      | Inhibition formed moderately     |
| 4   | 8 ppm         | Not observed                      | Inhibition zone formed (23 mm)   |
| 5   | 10 ppm        | Inhibition zone formation (14 mm) | Inhibition zone formation (26mm) |
| 6   | 12 ppm        | Inhibition zone formation (19 mm) | Not recorded                     |

#### **RESULTS AND DISCURSION**

The present study showed inhibition zone of lawn against the *Gloeocapsa montana Kutz.* 10 ppm and 12 ppm bavistin fungicide concentration recorded as 14 mm and 19 mm respectively. The Bavistin concentration at 2 ppm, 4ppm, 6 ppm and 8 ppm exhibit luxurious growth of alga *Gloeocapsa montana Kutz.* in dark green coloration.

Whereas in alga *Microcystis aeruginosa* Kutz. observed a growth at 2 ppm and 4 ppm and inhibition recorded at 6 ppm concentration of the fungicide bavistin. The inhibition zone is recorded in *Microcystis aeruginosa* Kutz. at 8 ppm and 10 ppm concentration of 23 mm and 26 mm respectively. The above results exhibited that blue green alga *Microcystis aeruginosa* Kutz. sensitivity towards bavistin than *Gloeocapsa montana* Kutz. in pale green coloration. Therefore, *Gloeocapsa montana* Kutz. may be preferable used for the bioremediation of bavistin due to their more tolerance against toxicity. The control test does not found anywhere inhibition whereas algal lawn formation to that moderate concentration of pesticide.

The coloration demarcation and inhibition zone indicated the sensitivity of blue green algal species against above mention concentration of pesticide. The toxicity, sensitivity and tolerances depend on concentration of pesticide and nature of stains used. Pesticide residue causes a serious environmental and health problem of human being. Herewith Bavistin showed antagonistic effect on non-tagged blue green algae i.e. *Gloeocapsa montana* Kutz. and *Microcystis aeruginosa* Kutz. Olfat *et al* (2014) developed the protocol including the modern technique for algal culturing method for decreases the cost of microalgae

production, present study also achieved same positive results.

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