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ASSESSMENT OF ALLELOPATHIC EFFECT OF *CALOTROPIS PROCERA* ON *CHROOCOCCUS MINUTUS*.

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ABSTRACT:
In allelopathy, soil microbial communities interfere with activity of plants secondary metabolites, so studies based on bioassays require the incorporation of microbes, as it can decrease the activity and concentration of chemicals decomposition of compound after entry in soil. The effect of *Calotropis procera* plant parts extract on a a cyanophycean alga *Chroococcus minutus* was studied. The leaves extract showed the significant increase in blue green algal cells growth where as the flower, stem and root extract affected the growth of *C. minutus* moderately. However the latex extract was found to be toxic.

Keywords: *allelopathy, secondary metabolites, cyanophycean alga.*

INTRODUCTION:

Cyanobacteria, or blue green algae, are widely distributed all over the world, in a great variety of ecosystems. They are especially interesting as they contribute to soil fertility in agroecosystems (Roger and Watnabe, 1986). *Chroococcus. minutus* is used as remedy for the biotransformation of pollutants, including nutrients (Sivasubramanian *et al.*, 2009). *Calotropis procera* (Ait) member of family Asclepediaceae, is an abundantly growing, laticiferous medicinal weed plant of tropical regions that contains ample amount of secondary metabolites. Thus keeping in this view allelopathic effect of *C. procera* was assessed on *C. minutes*, a soil habitant blue green algae.

METHOD AND MATERIAL:

Preparation of algal inoculums for assessment:-
For the preparation of algal inoculums, 500ml of culture was taken from the one month old unialgal liquid culture of *C. minutus* , and subsequently filtered with help of Watman Filter paper 40 to get a lump of alga to which, 100 ml of sterile distilled water was added and swirled in vortex cyclomixer to get the uniform inoculums. This homogenous culture was used as starter culture for the experiment.

Preparation of Calotropis plant parts extract:-
5 gm of plant material (fresh material was finely chopped), to make fresh extract (latex in v/v (ml) and dried material which was ground to powder added to 100ml of distilled water and kept overnight in rotatory shaker filtered with wattman filter paper. This was used as 100% concentrate. These extracts were further diluted with distilled water to make 50%, 25%, 10%, 5% and 1% concentration. Distilled water was used as control.

Evaluation of effect of Calotropis plant part extract (dried and fresh) on Alga:-

Assessment procedure: - 5 ml of starter culture was poured in test tubes to which 500µl dried/fresh extract of *Calotropis* plant parts was added in different concentration separately in triplicates and distilled water was used as control.

a) Initial assessment was through visual observation by the color intensity of extract treated cultures.

b) Effect was confirmed by counting the algal cells in Hemocytometer.

Counting the cells in Hemocytometer:-
Test tubes of liquid culture were swirled in vortex cyclomixer for 2 minutes, to prepare the homogenous algal cell suspension. 0.5ml Lugol's solution (KI3 + CH3COOH) was added the cell

suspension to kill and immobilize algal cells. Then10µl of this suspension was carefully loaded in grid and the number of cells was counted. The cell density in 1ml cell suspension was calculated from the following calculation (Stein, 1973)-

Density of cells/ml = Average number of cells counted × Dilution factor × 10^4

Statistical Analysis: All the data collected was analyzed using Analysis of Variance with Duncan's Multiple Range Test (Gomez et al, 1984).

RESULTS AND DISCUSSION

The effect of *Calotropis* on *Chroococcus* was observed by counting the number of cells in Hemocytometer. The total number of cells 311437/ml in fresh leaves extract showed significant (1%) increase. The total number of cells was found to be reduced in rest all the plant part extract treated on blue green algal culture at 1% level of significance. Plants producing secondary metabolites with a significant biological activity are subjected to cause stimulatory/inhibitory effect on plants and microbes in its vicinity. (Phillinger *et.al* 1995; Park *et al* 2006). *Calotropis* contains a number of biologically active components which are toxic to microorganism. This justifies the inhibitory effect of *Calotropis* on the alga with respect to both fresh and dried plant parts like flower, stem, root and latex. The inhibitory effect of latex might be due to the presence of toxic glycosides in latex. From the elemental analysis of *Calotropis* (Verma, 2014) leaves are found to be rich source of nutrition which might be the possible cause for enhancement in growth of alga (Sofowara, 1993). This is seen in the stimulatory growth of *Chroococcus* on being treated with fresh and dried leaf extracts Of *Calotropis*. Gerhenzon and Dudareva (2007) suggested that triterpenes are the most common group of secondary compounds which can stimulate or inhibit the growth of associated organism. In context to this Callaway and Aschehong (2000); Ehlers and Thompson (2004) suggested that the microbial community can potentially interact with allelochemicals and may degrade it.

CONCLUSION:

The fresh leaves of *Calotropis procera* can be used as an additive to soil, to enhance the growth of microalgal components and *Chroococcus minutus* not only assuages the also toxicity Secondary metabolites present in rhizosphere, but also improves the soil structure.

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Table No. 10 : Effect of *Calotropis* plant parts on *Chroococcus minutes*

S. No	Treatment	Fresh Plant parts No. of cells/ml	Dried Plant parts No. of cells/ml
1	Control	287977 ± 21.30	287947 ± 38.66
2	Leaf	311437 ± 40.90	289436 ± 19.30
3	Flower	277842 ± 26.25*	270125 ± 17.78*
4	Stem	268905 ± 2.16*	266670 ± 32.97*
5	Latex	112269 ± 39.36*	112215 ± 16.58*
6	Root	169026 ± 25.98*	113659 ± 48.33*
7	Seed	247458 ± 25.85*	208576 ± 94.71*
8	**Computed F**	**14041555**	**5840614.00**
9	**Coefficient of Variance**	**7.07%**	**11.99%**
10	**Standard Error**	**28.57**	**46.06**
11	**Critical Difference**	**50.27**	**81.06**

Note : Tabular F (i) at 0.05 = 2.85 and (ii) at 0.01= 4.46 and t-value at 5% = 1.76, a* = significant at 1 %