

EFFECTS OF UREA ON VEGETATIVE SURVIVAL AND REPRODUCTION OF GREEN ALGA *Pithophora oedogonia*

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ABSTRACT

Urea an organic nitrogenous compound was supplemented in different concentrations to liquid bold's basal culture media from 0.1 to 1000 ppm. As the concentrations of urea increased from 0.1 to 1000 ppm, lower concentrations of urea (up to 100 ppm), increased the vegetative survival of alga while the higher concentrations of urea (from 250 to 1000 ppm) increased the percentage of akinete formations in *Pithophora oedogonia*.

Keywords: Akinete formation, *Pithophora oedogonia*, vegetative survival, urea

Nitrogen was found to be quantitatively most important nutrient affecting the biomass, growth and lipid production of various microalgae (Griffiths and Harrison, 2009). It had been found that not only the presence or absence of nitrogen, but its quantity (high and low concentrations) affects the vegetative survival and reproduction in algae. The lack of nitrogen enhanced zoosporogenesis in *Ophiocystium* (Pecora and Russell, 1973) while lack of nitrogen decreased the vegetative survival and reproduction in *P. Oedogonia* (Agrawal and Mishra, 2002). Different levels of a nitrogen compounds control different mode of reproduction in an alga. Low levels of nitrogen induced gametogenesis in *Ulvafasciata*, while high levels of nitrogen induced asexual reproduction in the same alga (Mohsen *etal.* 1974). Lots of work has been done on algal growth, pigment analysis, biomass calculations but very little is known about the effects of urea on the survival and reproduction of the green alga *P. oedogonia*.

The main objective of the present work was to examine the effects of urea an organic nitrogen sources vegetative survival and akinete formation in *P. oedogonia* under different concentrations.

MATERIALS AND METHODS

Test Organisms

For the present study, green algae *Pithophora oedogonia* was collected as growing and forming tangled mats in a fresh water pond at the Botany Department, University of Allahabad. The source of water of the pond is ground water.

Chemical used for experiment

Urea, also known as carbamide, is an organic compound with chemical formula NH₂CONH₂. There are two NH₂ groups joined by carbonyl functional group. It is colorless, odorless, crystalline solid (E. Merck India Limited, India).

Method of measuring chlorophyll content

Alga *P. oedogonia* was collected from their natural habitats and its equal amount (10 mg) measured with electronic balance (CONTECH, India) was inoculated separately into equal volumes (10 ml) of Bold Basal Medium as well as in desired concentrations of urea solution of 0.1, 1, 10, 100, 250, 500 and 1000 ppm. All inoculated culture tubes were placed in the culture chamber at control culture conditions with illumination of 2 K lux white Light intensity for 16 hours a day at temperature of 25 ± 1°C.

Estimation of percentage of Vegetative survival of algal filaments

Percentage vegetative survival of alga was determined by observing the percentage of live vegetative cells versus dead vegetative cells. About 1000-1500 vegetative cells of inoculated algal mass were counted under microscope from each of 3 replicates to determine the percentage vegetative survival of the alga. Values were mean of 3 replicates.

Estimation of percentage of akinete formation in *Pithophora oedogonia*

Percentage of akinete formation in *P. oedogonia* was determined by counting the number of akinetes formed with respect to total number of vegetative cells in algal filaments under microscope. Values were mean of 3 replicates of algal inoculants.

Statistical analysis

The results presented are the means ± standard error data obtained from each of three replicates. All data are subjected to one way analysis of variance (ANOVA) with SPSS 16.0 software. Duncan’s multiple range test was performed at the 0.05 level of significance.

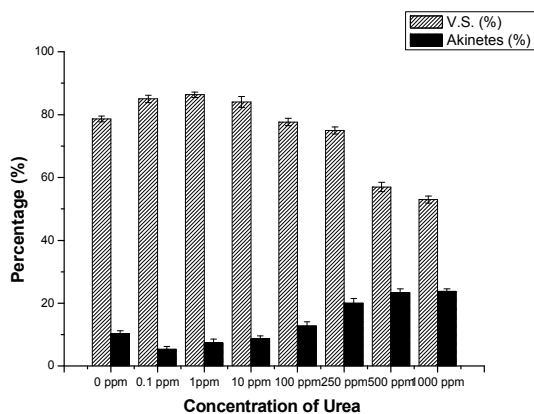
RESULTS AND DISCUSSIONS

Effect of urea on vegetative survival and akinete formation in *P. oedogonia*

The vegetative cells of live algal filaments of algae were light green to dark green in color, full of chlorophyll, turgid, while dead cells of the filaments were hyaline, had shrinkage of cytoplasm, low turgidity and no greenery in the cell sap (Fig. 1).

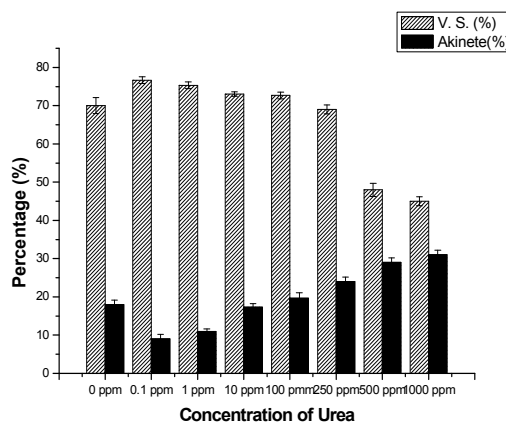
The akinete formation was started to appear after fifth to sixth day of inoculation of green healthy vegetative filaments of *P. oedogonia* in liquid Bold’s basal medium. During the formation of akinetes, the major part of the protoplasm of the vegetative cell was contracted towards one end of the cell and thickening of cell wall started. Matured akinetes were dark green to blackish in color (Fig. 2).

Urea at concentrations of 250, 500 and 1000 ppm increased the akinete formation in *P. oedogonia* than at lower concentrations of 0.1, 1, 10, and 100 ppm of urea. At 10th day of inoculation the rate of formation of akinete was lower or at initial stage and it continuously increased up to 15th day of inoculation. The percentage of akinete formation in the alga and the percentage of vegetative survival of the alga both are positively linked with each other in *P. oedogonia*. For more akinete formation, the percentage vegetative survival of the alga must also be more.



Graph A*

Graph A and Graph B represents the percentage of vegetative survival and akinete formation in *P. oedogonia* in presence of different concentrations of urea on 10th day and 15th day of inoculation respectively; All data were subjected to one way analysis of variance (ANOVA) with SPSS



Graph B*

16.00 software and Duncan’s multiple range tests was performed at the 0.05 level of significance, readings were mean of three replicates with standard error; V. S. (vegetative survival) and 0 ppm was the urea free control culture.



Fig 1

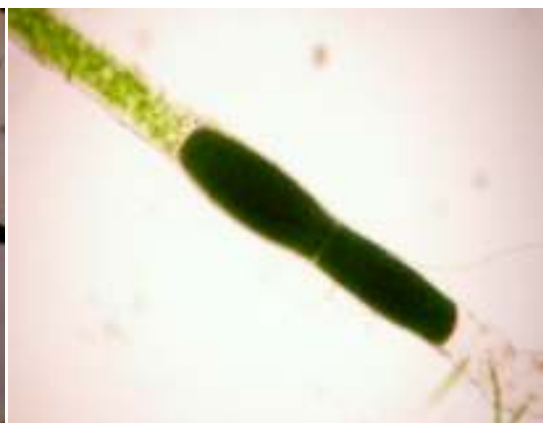


Fig. 2

Fig 1: Dead (hyaline cells without chlorophyll) and healthy vegetative (turgid, chlorophyll bearing green cells) filaments of *Pithophora oedogonia*

Fig 2: Mature, thick walled akinete bearing filament of *Pithophora oedogonia*

CONCLUSIONS

Addition of urea (a nitrogen source), increased the vegetative survival of *P. oedogonia*. The percentage vegetative survival of alga was higher in urea containing culture media than in control culture media containing 0 ppm urea and higher concentrations of urea (i.e. at 250, 500, and 1000 ppm), stimulated the akinete formation in *P. oedogonia* (Graph A and B).

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