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Growth and vegetative survival of green alga *Cladophora glomerata* in presence of auxins IAA, IBA and NAA

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Abstract

Growth of the green alga *Cladophora glomerata* (L.) Kuetzing, on the basis of the total chlorophyll content, was observed at all concentrations of indoleacetic acid and indolebutyric acid ranging between 0.01 to 1000 ppm but it was only at 0.01 to 100 ppm of naphthalene acetic acid. However, the alga died on 30 day of inoculation at 100 to 1000 ppm of IAA, on 45 day of inoculation at 1 to 1000 ppm of IBA and at 10 to 1000 ppm of NAA.

Key words *Cladophora glomerata*, IAA, IBA, NAA

Introduction

Plant hormones are reported to be present in various marine (Overbeek, 1940; Mowat, 1965) and freshwater algae (Shanab *et al*, 2003). Auxin induced stimulation in growth in *Chlorella vulgaris* (Yin, 1937), *Fucus evanescence*, *Ascophyllum nodusum* (Davidson, 1950) and *Caulerpa sertularoides* (Mishra and Kefford, 1969). IAA at 10^{-9} M concentration increased the dry weight of *Anacystis nidulans*, *Chlorogloea fritschii* and *Tolypothrix tenuis*, but at higher concentrations of 10^{-3} and 10^{-4} M, it was inhibitory (Ahmad and Winter, 1970).

The present study, deals with the effect of auxins, indoleacetic acid (IAA), indolebutyric acid (IBA) and naphthalene acetic acid (NAA) on the total chlorophyll content and vegetative survival of the green alga *Cladophora glomerata* (L.) Kuetzing.

Material and Methods

Algal material: The present alga was collected growing attached to the wall of a freshwater pond at Department of Botany, University of Allahabad campus. The alga was cultured in Bold's basal medium (BBM) (Cox and Bold, 1966) at a temperature of 25 ± 1˚C and light intensity 40 µmol m^{-2} s^{-1} from daylight fluorescent tubes for 16 h a day in a culture chamber.

With a view to see the responses of auxins, IAA (*Hi media laboratories*, India.), IBA (*Loba chemiie*, India) and NAA (*Central drug house (P) L*, India) on the growth of alga, the hormones were dissolved separately in minimal volume of 80% ethanol and mixed slowly in a known volume of BBM to prepare the desired concentrations of hormones which varied between 0.01 to 1000 ppm. For each hormone tested, a control was maintained containing an equal amount of ethanol. In each case the pH of the medium was adjusted to 7.5. The growth of the alga was estimated in terms of total chlorophyll content (Mckiney, 1941) on 3 and 5 day of inoculation with the help of spectrophotometer (EUROLAB, Gmbh, Colong, Germany). Data represented are mean of three replicates ± SD.

The percentage vegetative survival of alga was estimated by observing the percentage of live cells (healthy, dark green in colour) versus dead cells (hyaline, showing shrinkage of chloroplast) of the alga out of about 2000-2500 cells of the filaments counted from each of three replicates.

Result and Discussion

As compared to control, at all concentrations used between 0.01 to 1000 ppm of IAA, total chlorophyll content of the alga increased. Thus increase in the total chlorophyll content of alga was positively correlated with IAA concentration (Table 1). 10, 100, 500 and 1000 ppm of IAA, stimulated the amount of total chlorophyll content approximately 2 fold, 2.25 fold, 2.5 fold and 3 fold, respectively. As compared to control, total chlorophyll content of the alga increased with an increase in IBA concentrations. Maximum 1.5 fold increase in total chlorophyll content was observed at 500 and 1000 ppm IBA. NAA at concentrations between 0.01 to 100 ppm increased the total chlorophyll content of the alga; the maximum increase was observed at 1 and 10 ppm of NAA; 500 and 1000 ppm of NAA concentrations were inhibitory (Table 1).

IAA increased cell number in *Ulothrix subtillisma* (Conrad *et al*, 1959). Dawes (1971) reported that 5×10^{-6} and 1×10^{-6} M/ L of IBA stopped growth in *Caulerpa prolifera.* Hunt et al (2011) observed that NAA dissolved in EtOH (5ppm+500ppm) enhanced biomass productivity and chlorophyll content of *Chlorella sorokiniana*, *Haematococcus pluvialis* and *Pleurochrysis carterae*, but NAA at 10^{-6} M has an inhibitory effect on chlorophyll a and chlorophyll b content in *wolfia anhiza* (Czerpak *et al*, 2002).

C. glomerata died on 60 day after inoculation in control culture and at 0.01 to 1 ppm of IAA, on 45 and 30 day after inoculation at 10 and 100 ppm IAA and on 22 day after inoculation at 500 and 1000 ppm of IAA (Table 2). The alga died on 60 day after inoculation at 0.01 and 0.1 ppm of IBA and on 45 day after inoculation at 1 to 1000 ppm of IBA (Table 2). Alga died on 60 day after inoculation in NAA concentrations ranging from 0.01 to 10 ppm and on 45 day after inoculation at 100 to 1000 ppm NAA (Table 2). Thus increasing concentrations of IAA, IBA and NAA had an inhibitory effect on vegetative survival of *C. glomerata*.

Although IAA at 10 to 1000 ppm increased the total chlorophyll content of the alga, it resulted in a lower vegetative survivability. Auxin stimulated cell wall expansion in yeast by activating the gene controlling cell wall degrading enzymes, resulting in relaxation of the cell wall (Yanagishima and Shimoda, 1968; Shimoda and Yanagishima, 1971). Yau *et al* (1973) also observed marked cell wall expansion in *Coelastrum microporum* after exogenous application of IAA. In the present study, an increase in total chlorophyll content was probably because of the fact reported by Hansen and Grossman (2000) that plants treated with IAA and NAA at higher concentration increase the activity of ACC synthase (1- aminocyclopropane-1carboxylic acid synthase); the key regulator enzyme in ethylene biosynthesis, which in turn triggers the biosynthesis of abscisic acid (ABA). Increased levels of ethylene, ACC synthase and ABA, in close correlation, inhibit the growth of plants (Grossman, 2000). Jin *et al* (2008) also reported that auxin at higher concentration caused cell wall weakening.

References

Ahmad, M. R. & Winter, A. 1970 The effect of weak auxins on the growth of blue-green algae**.** *Hydrobiol.*, **36** (2) : 305-316.

Conrad, H., Saltman, P., & Eppley, R. 1959 Effects of auxin and gibberellic acid on growth of Ulothrix. *Nature* **184** : 556-557.

Cox, E. R. and Bold, H. C. 1966 Taxonomic investigation of *Stigeoclonium*, in (Eds.) University of Texas Publ. No. 6618, Austin (Texas). *Phycological studies* VII : 10.

Czerpak, R., Dobrzyń, P., Krotke, A. and Kicińska, E. 2002 The effect of auxins and salicylic acid on chlorophyll and carotenoid contents in *Wolffia arrhiza* (L.) Wimm. (Lemnaceae) Growing on Media of Various Trophicities. *Pol. J. Environ. Stud.* **11** (3) : 231-235.

Davidson, F.F. 1950 The effects of auxins on the growth of marine algae, *Am. J. Bot*. **37** : 502–510.

Dawes, C. J. 1971 Indole-3-acetic acid in the green algal coenocyte *Caulerpa prolifera* (Chlorophyceae, Siphonales). *Phycologia* **10** (4) : 375-379.

Grossmann K. 2000 The mode of action of auxin herbicides: A new ending to a long, drawn out story *Trends Plant Sci*, **5** : 506–508.

Hansen H., Grossmann K. 2000 Auxin-induced ethylene triggers abscisic acid biosynthesis and growth inhibition. *Plant Physiol.* **124** : 1437–1448.

Hunt, R. W., Chinnasamy, S. and Das, K. C. 2011 The effect of naphthalene-acetic acid on biomass productivity and chlorophyll content of green algae, coccolithophore, diatom and cyanobacteria cultures. *Appl. Biochem. and Biotech*., **164** (8) : 1350-1365.

Jin Q., Scherp P., Heimann K., Hasenstein K. H. 2008 Auxin and cytoskeletal organization in algae, *Cell Biol Int.* **32** (5) : 542-545.

Mckiney, G. 1941 Absorption of light by chlorophyll solution, *J. Biol. Chem*. **140** : 315-322.

Mishra, A. K. and Kefford, N. P. 1969 Developmental studies on the coenocytic alga, *Culerpa sertularoides. J. Phycol.* **5** (2):103–109.

Mowat, J. A 1965 A survey of results on the occurrence of auxins and gibberellins in algae. *Bot. Mar.* **8** : 149-155.

Overbeek, J. V. 1940 Auxin in marine algae. *Plant Physiol.* **15** : 291-299.

Shanab, S., Saker, M. M. and Abdel-Rahman, 2003 Crude extracts of some fresh water cyanobacteria have auxin like activity on potato tissue culture. *Arab. J. Biotech.* **6**, (2): 297- 312.

Shimoda, C. and Yanagishima, N. 1971 Role of cell wall-degrading enzymes in auxin induced cell expansion in yeast. Physiol. Plantarum. **24** (1) : 46–50.

Yau, P. C. and Chan, K. Y. 1973 Some physiological responses of two strains of *Coelastrum Microporum* Naeg. after auxin treatment, *New Asia College Acad. Annu*. **15** : 17-27.

Yanagishima, N. and Shimoda, C. 1968 Auxin induced expansion growth of cells and protoplasts of yeast, *Physiol. Plantarum.* **21** (5) : 1122–1128.

Yin, H. C. 1937 Effect of auxin on *Chlorella vulgaris*. *Proc. N. A. Sci.* **23** : 174-176.

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Table I: The effects of IAA, IBA and NAA on the total chlorophyll content (x 10^{-3} mg/ml) of *C. glomerata

Concentration of hormone (ppm)	IAA		IBA		NAA	
	3 d	5 d	3 d	5 d	3 d	5 d
0 (Control)	**1.789** ± 0.10	**2.082** ± 0.35	**1.821** ± 0.12	**2.023** ± **0.16**	**1.423** ± 0.19	**3.297** ± 0.09
0.01	**0.766** ± 0.15	**2.408** ± 0.06	**2.608** ± 0.42	**2.026** ± 0.34	**1.804** ± 0.29	**4.281** ± 0.05
0.1	**0.600** ± 0.09	**2.622** ± 0.43	**2.985** ± 0.12	**2.168** ± 0.17	**2.767** ± 0.08	**5.635** ± 0.11
1	**2.693** ± 0.14	**3.601** ± 0.08	**3.363** ± 0.23	**2.234** ± 0.12	**2.648** ± 0.13	**7.162** ± 0.15
10	**2.330** ± 0.18	**4.080** ± 0.27	**3.533** ± 0.48	**2.513** ± 0.22	**2.644** ± 0.19	**7.039** ± 0.30
100	**2.335** ± 0.26	**4.492** ± 0.16	**2.535** ± 0.31	**2.596** ± 0.24	**2.204** ± 0.12	**4.605** ± 0.05
500	**2.364** ± 0.12	**5.250** ± 0.33	**2.189** ± 0.24	**3.049** ± 0.05	**1.285** ± 0.49	**2.600** ± 0.09
1000	**3.208** ± 0.35	**6.735** ± 0.76	**2.193** ± 0.16	**3.157** ± 0.08	**1.137** ± 0.07	**2.567** ± 0.09

*** The alga had fresh blot-dry weight of 0.025 gm at the time of inoculation; the total chlorophyll content of the alga was determined by the method of Mckiney (1941); the data represent mean of three replicates ± SD; the total chlorophyll content of 0.025 gm blot dry weight of the alga, at the start of experiment was 1.272 ± 0.2**

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Table II: Percentage vegetative survival of *Cladophora glomerata* in presence of different concentrations of IAA, IBA and NAA

Hormone	Concentration (ppm)	*C. glomerata*						
		Day after inoculation						
		2	7	15	22	30	45	60
IAA	0 (control)	89 ± 1	66 ± 1	36 ± 3	23 ± 2	17±1	08 ± 2	00 ± 00
	0.01	92 ± 2	78 ± 1	56 ± 1	32 ± 1	21±0	06 ± 2	00 ± 00
	0.1	90 ± 0.9	78 ± 1.2	38 ± 2	23 ± 2.3	11±1	02 ± 1	00 ± 00
	1.0	98 ± 0	82 ± 2	46 ± 1	25 ± 2	07±1	09 ± 0.8	00 ± 00
	10.0	99 ± 1	81 ± 1	42 ± 1	35 ± 2	14±1	00 ± 00	__
	100.0	100 ± 2	85 ± 1	33 ± 1	14 ± 1	00±00	__	__
	500.0	100 1	84 ± 0.6	42 ± 1	00 ± 00	__	__	__
	1000.0	100 ± 0.8	97 ± 1	34 ± 2	00 ± 00	__	__	__
IBA	0 (control)	74 ± 1	64 ± 2	47 ± 3	44±1	37±1	23±2	07 ±0.4
	0.01	75 ± 2	49 ± 3	29 ± 2	22±1	14±1	08±0	03±1
	0.1	78 ± 3	55 ± 2	23 ± 1	12±2	09±1	02±0	00±00
	1.0	82 ± 3	62 ± 1	44 ± 1	25±2	16±2	00±0	__
	10.0	87 ± 2	59 ± 1	26 ± 1	11±0.8	06±00	00±0	__
	100.0	94 ± 1	71 ± 1.2	36 ± 2	25±1	04±1	00±0	__
	500.0	96 ± 1.5	76 ± 2	56 ± 1	33±3	14±1	00±0	__
	1000.0	97 ± 1	88 ± 2	57 ± 1	30±1	22±2	00±0	__
NAA	0 (control)	84±1	75±2	54±1	41±1	33±1	14±0.3	00±0
	0.01	87±1	70±2	40±1	34±0.7	17±1	04±1	00±0
	0.1	89±1	67±1	28±1	13±1.2	09±1	06±1	00±0
	1.0	98±3	65±1	42±2	31±1	11±0.2	07±0.5	00±0
	10.0	96±2	63±1	29±1	12±1	08±1	00±0	00±0
	100.0	94±3	66±2	25±1	13±2	03±1	00±0	__
	500.0	92±1.8	36±1	11±2	09±0.6	05±0.8	00±0	__
	1000.0	86±2	42±2	13±1	08±0.7	02±0.3	00±0	__

*** Percentage vegetative survival of alga was determined by counting percentage of live cells (dark green) versus dead cells (hyaline) out of about 2000-2500 cells of filaments from each of three replicates; data represents mean of three replicates ± SD.**