



COMPARATIVE STUDY OF THE EFFECT OF DIFFERENT CONCENTRATIONS OF MEDIA PHOSPHOROUS OF TWO SPECIES OF AZOLLA IN TWO SEASONS

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ABSTRACT

Two species of *Azolla* (*A.pinnata* and *A.filiculoides*) were selected to grow in different concentrations of phosphorus in two different seasons in tropical plains of the district of Burdwan, West Bengal to compare its productivity. Phosphorus in different concentrations was used uniformly for both the seasons. By absorbing phosphorus, fresh *Azolla* bio-mass was collected, which has been shown in this context. A correlation coefficient calculation amongst two consecutive seasons were done following Panse and Sukhatme (2005). It had a remarkable difference between two different seasons allowing them different light intensities. The total biomass weight and size of the plant materials were increased due to all those factors and it was observed better in the summer months than the winter months. Difference in optimum concentrations of media-P was noted in case of both the species in two different seasons. All meteorological informations were noted properly.

The main aims and objectives were to establish the research results which may help the poor farmer by utilizing this species for their crop field in easy way. Indeed, our moto is to propagate these activities in a mega scale practice for developing a green world.

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INTRODUCTION

***Azolla* (mosquito fern, duckweed fern, fairy moss, water fern)**

It is a genus of seven species of aquatic ferns in the family Salviniaceae. They are extremely reduced in form and specialized, looking nothing like other typical ferns but more resembling duckweed or some mosses. Members of the genus *Azolla* are utilized throughout the world for a wide variety of purposes besides its widespread uses as an ornamental in fish ponds and tanks (Lumpkin and Plucknett, 1980; 1982). The major fundamental constraints of growth of *Azollasp* are limitation of water supply and phosphorus and susceptibility to temperature, pests and pathogens. Phosphorus represents a major limiting factor in the field for the growth of the *Azolla-Anabaena* symbiotic nitrogen-fixing system.

Azollafiliculoides is one of just two fern species for which a reference genome has been published (Evrard and Hove, 2004) and it is a native to the America and has invaded many places in Europe and South Africa (Hill, 2003), *Azollafiliculoides* is a small, free floating freshwater fern, green to reddish-brown or purplish orange or red at the edges, branching freely, and breaking into smaller sections as it grows.

The adult plant is approximately 25-35 mm long, with the length of the individual frond ("leaves") being approximately 1-1.5 mm. Plants can change colour from green to brown and red as a result of changes in sunlight intensity (and shade) as well as ambient temperature. *Azollafiliculoides* is able to undergo rapid vegetative production throughout the year by the elongation and fragmentation of the small fronds. Under ideal conditions an infestation can double in area every 4-5 days. At such growth rates it is capable of completely covering pond and lake surfaces in a matter of weeks or months. Under favourable environmental conditions, *A. filiculoides* fern undergoes sexual reproduction through spores (Henderson and Cilliers, 2002a and 2002b). It can also be dispersed on the feet and feathers of water birds and on/by mammals such as hippos and otters. It has been spread by the garden and aquaria trade and can find its way into water bodies through discarded garden or pond waste and flood events.

The species has been introduced to many regions of the Old World, grown for its nitrogen-fixing ability which can be utilised to enhance the growth rate of crops grown in water like paddy rice, or by removal from lakes for use as green manure. It is also used as an ornamental plant in ponds.

On the other hand, *A. pinnata* is small, 1.5-2.5 cm long, with an almost straight main axis with pinnately arranged side branches, progressively longer towards the base, thus roughly

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triangular in shape; the basal branches themselves becoming pinnate and eventually fragmenting as the main axis decomposes to form new plants. Roots have fine lateral rootlets, giving a feathery appearance in the water. Leaves minute, 1-2 mm long, overlapping in two ranks, upper lobe green, brownish green or reddish, lower lobe translucent brown; minute, short, pillae, more or less cylindrical unicellular hairs often present on the upper lobes. When fertile, round sporocarps 1-1.5 mm wide can be seen on the under-side at the bases of the side branches. The leaves often have a maroon-red tinge and the water can appear to be covered by red velvet layer.

The use of aquatic plants for removal of nutrients from the waste water has been studied by a large number of workers (Yount and Crossman, 1970; Boyd, 1949, 1955, 1969, 1970, 1971a, 1971b; Steward and Dinges, 1976a, 1976b; Ehrlich, 1966; Jagadeesh and Lakshminarayana, 1971; Wooten and Dodd, 1976; Wolverton and McDonald, 1979; Reddy, 1981; 1982). A large amount of nutrients having access to water as pollutants is absorbed through the roots of aquatic plants (Dymond, 1948; Boyd, 1970). The distribution in this summary table is based on all the information available. When several references are cited, they may give conflicting information on the status. Further details may be available for individual references in the Distribution Table Details section which can be selected by going to Generate Report.

Thus the harvested biomass of this plant can play important role in the rice-field as an useful green manure rich in N P K.

MATERIALS AND METHODS

Plant Material

Two species of *Azolla* such as *A. filiculoides* and *A. pinnata* were taken for the present experiment. The genus possesses intrinsic interest in that its members are capable of assimilating atmospheric nitrogen with the help of a symbiont within the cavities of their leaves.

- Open atmosphere and partial shade areas
- Distilled water
- Reagents used to prepare IRRI's media as followed by I. Watanabe, 1977.
- Net house
- Culture vessels

Chemical balance

Necessary glass wares etc.

All the experimental studies were conducted in the net house, Department of Botany, The University of Burdwan, Burdwan, West Bengal, India.

Table of Meteorological Data (MD) recorded by District Seed Farm Kalna Road, Burdwan in 1994

Season	Atmospheric Temperature (°C)[mean]			Solar radiation (μmol m ⁻² s ⁻¹)		Relative humidity (%) [mean]			Rainfall (mm)
	Max.	Min.	Mean	O. A.	P.S.	Mean Max.	Mean Min.	Mean	
Jan-Feb	25.20	15.40	20.30	437.36	203.77	91	53.9	72.45	0
Mar-Apr	35.65	23.07	29.36	532.98	252.62	91	32.18	61.59	0

Table 1.A-i Correlations between light intensities of *A. filiculoides* in 0 ppm **Table 1.A-ii** Correlations between light intensities of *A. filiculoides* in 5 ppm

Sl. No	X	Y	X ²	Y ²	XY	Sl. No	X	Y	X ²	Y ²	XY
1	2.71	2.61	7.3441	6.8121	7.0731	1	4.61	4.31	21.2521	18.5761	19.8691
2	2.73	2.63	7.4529	6.9169	7.1799	2	4.63	4.27	21.4369	18.2329	19.7701
3	2.70	2.64	7.29	6.9696	7.128	3	4.64	4.3	21.5296	18.49	19.952
4	2.74	2.60	7.5076	6.76	7.00	4	4.60	4.28	21.16	18.3184	19.688
5	2.72	2.62	7.3984	6.8644	7.1264	5	4.62	4.29	21.3444	18.4041	19.734
6	2.74	2.59	7.5076	6.7081	7.00	6	4.64	4.31	21.5296	18.5761	19.9984
7	2.71	2.60	7.3441	6.76	7.046	7	4.60	4.28	21.16	18.3184	19.688
8	2.71	2.61	7.3441	6.8121	7.0731	8	4.61	4.30	21.2521	18.49	19.823
9	2.69	2.58	7.2361	6.6564	6.9402	9	4.60	4.32	21.16	18.6624	19.872
10	2.75	2.66	7.5625	7.0756	7.315	10	4.65	4.26	21.6225	18.1476	19.809
Σ	27.2	26.14	73.9874	68.3352	71.1023	Σ	46.20	42.92	213.447	184.216	198.203

$\Sigma=(x-x\bar{x})^2=73.9874-(27.2)^2=0.0034$, $\Sigma=(y-y\bar{y})^2=68.3352-(26.14)^2=0.0052$, $\Sigma=(x-x\bar{x})(y-y\bar{y})=71.1023-(27.2 \times 26.14)/10=0.0015$; Hence, $Y=0.06504$
 $\Sigma=(x-x\bar{x})^2=213.447-(46.20)^2=0.0032$, $\Sigma=(y-y\bar{y})^2=184.1476-(42.92)^2=0.0052$, $\Sigma=(x-x\bar{x})(y-y\bar{y})=198.2036-(46.20 \times 42.92)/10=-0.0868$; Hence, $Y=6.0166$

Table 1.A-iii Correlations between light intensities of *A. filiculoides* in 10 ppm

Table 1.A-iv: Correlations between light intensities of *A. filiculoides* in 15 ppm

Sl. No	X	Y	X ²	Y ²	XY	Sl. No	X	Y	X ²	Y ²	XY
1	4.94	4.31	24.4036	18.5761	21.2914	1	5.2	4.99	27.04	24.9001	25.948
2	4.9	4.27	24.01	18.2329	20.923	2	5.22	4.97	27.2484	24.7009	25.9434
3	4.93	4.30	24.3049	18.49	21.199	3	5.23	4.96	27.3529	24.6016	25.9408
4	4.91	4.28	24.1081	18.3184	21.0148	4	5.19	4.99	26.9361	24.9001	25.8981
5	4.92	4.29	24.2064	18.4041	21.1068	5	5.21	4.98	27.1441	24.8004	25.9458
6	4.91	4.33	24.1081	18.7489	21.2603	6	5.00	5.00	25.00	25.00	25.000
7	4.9	4.32	24.01	18.6624	21.168	7	5.22	5.1	27.2484	26.010	26.622
8	4.91	4.27	24.1081	18.2329	20.9657	8	5.25	4.97	27.5625	24.7009	26.0925
9	4.94	4.26	24.4036	18.1476	21.0444	9	5.19	4.99	26.9361	24.9001	25.8981
10	4.95	4.27	24.5025	18.2329	21.1365	10	5.19	4.98	26.9361	24.8004	25.8981
Σ	49.21	42.9	242.165	184.046	211.109	Σ	51.9	49.93	269.404	249.314	259.186

$\Sigma=(x-x\bar{x})^2=242.1653-(49.21)^2=0.0012$, $\Sigma=(y-y\bar{y})^2=184.0462-(42.9)^2=0.0052$, $\Sigma=(x-x\bar{x})(y-y\bar{y})=211.1099-(49.21 \times 42.9)/10=0.001$; Hence, $Y=0.400321858$
 $\Sigma=(x-x\bar{x})^2=269.4046-(51.9)^2=0.0436$, $\Sigma=(y-y\bar{y})^2=249.3145-(49.93)^2=0.01401$, $\Sigma=(x-x\bar{x})(y-y\bar{y})=259.1868-(51.9 \times 49.93)/10=0.0501$; Hence, $Y=2.027101169$

METHODS

The experiment was performed using original IRRI's medium having 20 ppm of phosphorus concentration. Not only 20 ppm, but different concentrations of phosphorus such as 5 ppm, 10 ppm, 15 ppm and 60 ppm, alongwith a control without media-P were prepared and two species of *Azolla* were allowed to grow in it to find out the differences in growth, of those two species after the 10 days of incubation, both in O.A. and P.S. conditions.

Bi-variate correlation co-efficient model as followed by Panse & Sukhatme (2005)

RESULTS AND DISCUSSION

Results

The bi-variate correlation data have been calculated of both the species of *Azolla* which have been given below:

Table 1.A-v Correlations between light intensities of *A. filiculoides* in 20 ppm

Sl.no.	X	Y	X ²	Y ²	XY
1	5	5.33	25	28.4089	26.65
2	4.9	5.32	24.01	28.3024	26.068
3	5	5.33	25	28.4089	26.65
4	4.8	5.31	23.04	28.1961	25.488
5	5.61	5.34	31.4721	28.5156	29.9574
6	5.62	5.35	31.5844	28.6225	30.067
7	5.63	5.36	31.6969	28.7296	30.1768
8	5.62	5.35	31.5844	28.6225	30.067
9	5.63	5.36	31.6969	28.7296	30.1768
10	5.62	5.35	31.5844	28.6225	30.067
Σ	53.43	53.4	286.669	285.158	285.368

$\Sigma(x-x\bar{c})^2 = 286.1586 - (53.43)^2 = 0.68211$; $\Sigma(y-y\bar{c})^2 = 285.1586 - (53.4)^2 = 0.0026$; $\Sigma(x-x\bar{c})(y-y\bar{c}) = 285.368 - (53.43 \times 53.4) / 10 = 0.5018$; Hence, $Y = 1.230030496$

Table 1.A-vi Correlations between light intensities of *A. filiculoides* in 60 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	5	4.71	25	22.1841	23.55
2	4.9	4	24.01	16	19.6
3	5	4.71	25	22.1841	23.55
4	4.8	4.69	23.04	21.9961	22.512
5	5.01	4.72	25.1001	22.2784	23.6472
6	5.02	4.73	25.2004	22.3729	23.7446
7	5.03	4.74	25.3009	22.4676	23.8422
8	5.02	4.73	25.2004	22.3729	23.7446
9	5.03	4.74	25.3009	22.4676	23.8422
10	5.02	4.73	25.2004	22.3729	23.7446
Σ	49.83	46.5	248.353	222.552	231.777

$\Sigma(x-x\bar{c})^2 = 248.3531 - (49.83)^2 = 0.05021$; $\Sigma(y-y\bar{c})^2 = 222.5526 - (46.5)^2 = 6.3276$; $\Sigma(x-x\bar{c})(y-y\bar{c}) = 231.7774 - (49.83 \times 46.5) / 10 = 0.679$; Hence, $Y = 1.204634107$

Table 1.B-i Correlations between light intensities of *A. pinnata* in 0 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	2.71	2.61	7.3441	6.8121	7.0731
2	2.73	2.63	7.4529	6.9169	7.1799
3	2.7	2.64	7.29	6.9696	7.128
4	2.74	2.60	7.5076	6.76	7.00
5	2.72	2.62	7.3984	6.8644	7.1264
6	2.74	2.59	7.5076	6.7081	7.00
7	2.71	2.60	7.3441	6.76	7.046
8	2.71	2.61	7.3441	6.8121	7.0731
9	2.69	2.58	7.2361	6.6564	6.9402
10	2.75	2.66	7.5625	7.0756	7.315
Σ	27.2	26.14	73.9874	68.3352	71.1023

$\Sigma(x-x\bar{c})^2 = 73.9874 - (27.2)^2 = 0.001$; $\Sigma(y-y\bar{c})^2 = 68.3352 - (26.14)^2 = 0.032996$; $\Sigma(x-x\bar{c})(y-y\bar{c}) = 71.1023 - (27.2 \times 26.14) / 10 = 0.0015$; Hence, $Y = 0.237289508$

Table 1.B-ii Correlations between light intensities of *A. pinnata* in 5 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	4.61	4.31	21.2521	18.5761	19.8691
2	4.63	4.27	21.4369	18.2329	19.7701
3	4.64	4.3	21.5296	18.49	19.952
4	4.60	4.28	21.16	18.3184	19.688
5	4.62	4.29	21.3444	18.4041	19.734
6	4.64	4.31	21.5296	18.5761	19.9984
7	4.60	4.28	21.16	18.3184	19.688
8	4.61	4.30	21.2521	18.49	19.823
9	4.60	4.32	21.16	18.6624	19.872
10	4.65	4.26	21.6225	18.1476	19.809
Σ	46.20	42.92	213.4472	184.216	198.2036

$\Sigma(x-x\bar{c})^2 = 213.4472 - (46.20)^2 = 0.0032$; $\Sigma(y-y\bar{c})^2 = 184.216 - (42.92)^2 = 0.0036$; $\Sigma(x-x\bar{c})(y-y\bar{c}) = 198.2036 - (46.20 \times 42.92) / 10 = -0.868$; Hence, $Y = 25.57369939$

Table 1.B-iii Correlations between light intensities of *A. pinnata* in 10 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	4.94	4.31	24.4036	18.5761	21.2914
2	4.9	4.27	24.01	18.2329	20.923
3	4.93	4.30	24.3049	18.49	21.199
4	4.91	4.28	24.1081	18.3184	21.0148
5	4.92	4.29	24.2064	18.4041	21.1068
6	4.91	4.33	24.1081	18.7489	21.2603
7	4.9	4.32	24.01	18.6624	21.168
8	4.91	4.27	24.1081	18.2329	20.9657
9	4.94	4.26	24.4036	18.1476	21.0444
10	4.95	4.27	24.5025	18.2329	21.1365
Σ	49.21	42.9	242.165	184.0462	211.109

$\Sigma(x-x\bar{c})^2 = 242.1653 - (49.21)^2 = 0.68211$; $\Sigma(y-y\bar{c})^2 = 184.0462 - (42.9)^2 = 0.0052$; $\Sigma(x-x\bar{c})(y-y\bar{c}) = 211.1099 - (49.21 \times 42.9) / 10 = 0.001$; Hence, $Y = 0.25795827$

Table 1.B-iv Correlations between light intensities of *A. pinnata* in 15 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	5.2	4.99	27.04	24.9001	25.948
2	5.22	4.97	27.2484	24.7009	25.9434
3	5.23	4.96	27.3529	24.6016	25.9408
4	5.19	4.99	26.9361	24.9001	25.8981
5	5.21	4.98	27.1441	24.8004	25.9458
6	5.00	5.00	25.00	25.00	25.00
7	5.22	5.1	27.2484	26.01	26.622
8	5.25	4.97	27.5625	24.7009	26.0925
9	5.19	4.99	26.9361	24.9001	25.8981
10	5.19	4.98	26.9361	24.8004	25.8981
Σ	51.9	49.93	269.4046	249.314	259.1868

$\Sigma(x-x\bar{c})^2 = 269.4046 - (51.9)^2 = 0.0436$; $\Sigma(y-y\bar{c})^2 = 249.3145 - (49.93)^2 = 0.01401$; $\Sigma(x-x\bar{c})(y-y\bar{c}) = 259.3145 - (51.9 \times 49.93) / 10 = 0.0501$; Hence, $Y = 2.027101169$

Table 1 B-v Correlations between light intensities of *A. pinnata* in 20 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	5	5.33	25	28.4089	26.65
2	4.9	5.32	24.01	28.3024	26.068
3	5	5.33	25	28.4089	26.65
4	4.8	5.31	23.04	28.1961	25.488
5	5.61	5.34	31.4721	28.5156	29.9574
6	5.62	5.35	31.5844	28.6225	30.067
7	5.63	5.36	31.6969	28.7296	30.1768
8	5.62	5.35	31.5844	28.6225	30.067
9	5.63	5.36	31.6969	28.7296	30.1768
10	5.62	5.35	31.5844	28.6225	30.067
Σ	53.43	53.4	286.669	285.1586	285.368

$\Sigma=(x-x\bar{c})^2=286.6691-(53.43)^2=1.19261$; $\Sigma=(y-y\bar{c}\bar{c})^2=285.1586-(53.4)^2=0.0026$; $\Sigma=(x-x\bar{c})(y-y\bar{c})=285.368-(53.43 \times 53.4)/10=0.0518$; Hence, $Y=0.930237553$

Table 1.B-vi Correlations between light intensities of *A. pinnata* in 60 ppm

Sl.no.	X	Y	X ²	Y ²	XY
1	5	4.71	25	22.1841	23.55
2	4.9	4	24.01	16	19.6
3	5	4.71	25	22.1841	23.55
4	4.8	4.69	23.04	21.9961	22.512
5	5.01	4.72	25.1001	22.2784	23.6472
6	5.02	4.73	25.2004	22.3729	23.7446
7	5.03	4.74	25.3009	22.4676	23.8422
8	5.02	4.73	25.2004	22.3729	23.7446
9	5.03	4.74	25.3009	22.4676	23.8422
10	5.02	4.73	25.2004	22.3729	23.7446
Σ	49.83	46.5	248.3531	222.5526	231.7774

$\Sigma=(x-x\bar{c})^2=248.3531-(49.83)^2=0.05021$; $\Sigma=(y-y\bar{c}\bar{c})^2=222.5526-(46.5)^2=6.3276$; $\Sigma=(x-x\bar{c})(y-y\bar{c})=231.774-(49.83 \times 46.5)/10=0.0679$; Hence, $Y=1.204634831$

Table 2.A-i Correlations between light intensities of *A. filiculoids* in 0 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	2.71	2.61	7.3441	6.8121	7.0731
2	2.73	2.63	7.4529	6.9169	7.1799
3	2.7	2.64	7.29	6.9696	7.128
4	2.74	2.60	7.5076	6.76	7.00
5	2.72	2.62	7.3984	6.8644	7.1264
6	2.74	2.59	7.5076	6.7081	7.00
7	2.71	2.60	7.3441	6.76	7.046
8	2.71	2.61	7.3441	6.8121	7.0731
9	2.69	2.58	7.2361	6.6564	6.9402
10	2.75	2.66	7.5625	7.0756	7.315
Σ	27.2	26.14	73.9874	68.3352	71.1023

$\Sigma=(x-x\bar{c})^2=73.9874-(27.2)^2=0.0034$; $\Sigma=(y-y\bar{c}\bar{c})^2=68.3352-(26.14)^2=0.00524$; $\Sigma=(x-x\bar{c})(y-y\bar{c})=71.1023-(27.2 \times 26.14)/10=0.0015$; Hence, $Y=0.355374446$

Table 2.A-ii Correlations between light intensities of *A. filiculoids* in 5 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	4.61	4.31	21.2521	18.5761	19.8691
2	4.63	4.27	21.4369	18.2329	19.7701
3	4.64	4.3	21.5296	18.49	19.952
4	4.60	4.28	21.16	18.3184	19.688
5	4.62	4.29	21.3444	18.4041	19.734
6	4.64	4.31	21.5296	18.5761	19.9984
7	4.60	4.28	21.16	18.3184	19.688
8	4.61	4.30	21.2521	18.49	19.823
9	4.60	4.32	21.16	18.6624	19.872
10	4.65	4.26	21.6225	18.1476	19.809
Σ	46.20	42.92	213.4472	184.216	198.2036

$\Sigma=(x-x\bar{c})^2=213.4472-(46.20)^2=0.0032$; $\Sigma=(y-y\bar{c}\bar{c})^2=184.216-(42.92)^2=0.00336$; $\Sigma=(x-x\bar{c})(y-y\bar{c})=198.809-(46.2 \times 42.92)/10=-0.0868$; Hence, $Y=26.47129145$

Table 2.A-iii Correlations between light intensities of *A. filiculoids* in 10 ppm

Sl.no	X	Y	X ²	Y ²	XY
1	4.94	4.31	24.4036	18.5761	21.2914
2	4.9	4.27	24.01	18.2329	20.923
3	4.93	4.30	24.3049	18.49	21.199
4	4.91	4.28	24.1081	18.3184	21.0148
5	4.92	4.29	24.2064	18.4041	21.1068
6	4.91	4.33	24.1081	18.7489	21.2603
7	4.9	4.32	24.01	18.6624	21.168
8	4.91	4.27	24.1081	18.2329	20.9657
9	4.94	4.26	24.4036	18.1476	21.0444
10	4.95	4.27	24.5025	18.2329	21.1365
Σ	49.21	42.9	242.1653	184.0462	211.1099

$\Sigma=(x-x\bar{c})^2=73.9874-(27.2)^2=0.0012$; $\Sigma=(y-y\bar{c}\bar{c})^2=68.3352-(26.14)^2=0.0052$; $\Sigma=(x-x\bar{c})(y-y\bar{c})=211.1099-(27.2 \times 26.14)/10=-0.001$; Hence, $Y=0.400320416$

Table 2.A-iv Correlations between light intensities of *A. filiculoids* in 15 ppm

Sl.no	X	Y	X ²	Y ²	XY
1	5.2	4.99	27.04	24.9001	25.948
2	5.22	4.97	27.2484	24.7009	25.9434
3	5.23	4.96	27.3529	24.6016	25.9408
4	5.19	4.99	26.9361	24.9001	25.8981
5	5.21	4.98	27.1441	24.8004	25.9458
6	5.00	5.00	25.00	25.00	25.00
7	5.22	5.1	27.2484	26.01	26.622
8	5.25	4.97	27.5625	24.7009	26.0925
9	5.19	4.99	26.9361	24.9001	25.8981
10	5.19	4.98	26.9361	24.8004	25.8981
Σ	51.9	49.93	269.4046	249.3145	259.1868

$\Sigma=(x-x\bar{c})^2=269.4046-(51.9)^2=0.0436$; $\Sigma=(y-y\bar{c}\bar{c})^2=249.3145-(49.93)^2=0.01401$; $\Sigma=(x-x\bar{c})(y-y\bar{c})=259.1868-(51.9 \times 49.93)/10=0.0501$; Hence, $Y=2.027101169$

Table 2.A-v Correlations between light intensities of *A. filiculoids* in 20 ppm

Sl.no	X	Y	X ²	Y ²	XY
1	5	5.33	25	28.4089	26.65
2	4.9	5.32	24.01	28.3024	26.068
3	5	5.33	25	28.4089	26.65
4	4.8	5.31	23.04	28.1961	25.488
5	5.61	5.34	31.4721	28.5156	29.9574
6	5.62	5.35	31.5844	28.6225	30.067
7	5.63	5.36	31.6969	28.7296	30.1768
8	5.62	5.35	31.5844	28.6225	30.067
9	5.63	5.36	31.6969	28.7296	30.1768
10	5.62	5.35	31.5844	28.6225	30.067
Σ	53.43	53.4	286.6691	285.1586	285.368

$\Sigma=(x-x\bar{c})^2=286.6691-(53.43)^2=1.19261$; $\Sigma=(y-y\bar{c}\bar{c})^2=285.1586-(53.4)^2=0.0026$; $\Sigma=(x-x\bar{c})(y-y\bar{c})=285.368-(53.43 \times 53.4)/10=0.0518$; Hence, $Y=0.930237553$

Table 2.A-vi Correlations between light intensities of *A. filiculoids* in 60 ppm

Sl.no	X	Y	X ²	Y ²	XY
1	5	4.71	25	22.1841	23.55
2	4.9	4	24.01	16	19.6
3	5	4.71	25	22.1841	23.55
4	4.8	4.69	23.04	21.9961	22.512
5	5.01	4.72	25.1001	22.2784	23.6472
6	5.02	4.73	25.2004	22.3729	23.7446
7	5.03	4.74	25.3009	22.4676	23.8422
8	5.02	4.73	25.2004	22.3729	23.7446
9	5.03	4.74	25.3009	22.4676	23.8422
10	5.02	4.73	25.2004	22.3729	23.7446
Σ	49.83	46.5	248.3531	222.5526	231.7774

$\Sigma=(x-x\bar{c})^2=248.3531-(49.83)^2=0.05021$; $\Sigma=(y-y\bar{c}\bar{c})^2=222.5526-(46.5)^2=6.3276$; $\Sigma=(x-x\bar{c})(y-y\bar{c})=231.7774-(49.83 \times 46.5)/10=0.0679$; Hence, $Y=1.2046341$

Table 2.B -i: Correlations between light intensities of *A. pinnata* in 0 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	2.71	2.61	7.3441	6.8121	7.0731
2	2.73	2.63	7.4529	6.9169	7.1799
3	2.7	2.64	7.29	6.9696	7.128
4	2.74	2.60	7.5076	6.76	7.00
5	2.72	2.62	7.3984	6.8644	7.1264
6	2.74	2.59	7.5076	6.7081	7.00
7	2.71	2.60	7.3441	6.76	7.046
8	2.71	2.61	7.3441	6.8121	7.0731
9	2.69	2.58	7.2361	6.6564	6.9402
10	2.75	2.66	7.5625	7.0756	7.315
Σ	27.2	26.14	73.9874	68.3352	71.1023

$\Sigma=(x-x\bar{x})^2 = 73.9874 - (27.2)^2 = 0.0034$; $\Sigma=(y-y\bar{y})^2 = 68.3352 - (26.14)^2 = 0.00524$; $\Sigma=(x-x\bar{x})(y-y\bar{y}) = 71.1023 - (27.2 \times 26.14)/10 = 0.0015$; Hence, $Y = 0.355374446$

Table 2.B -ii: Correlations between light intensities of *A. pinnata* in 5 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	4.61	4.31	21.2521	18.5761	19.8691
2	4.63	4.27	21.4369	18.2329	19.7701
3	4.64	4.3	21.5296	18.49	19.952
4	4.60	4.28	21.16	18.3184	19.688
5	4.62	4.29	21.3444	18.4041	19.734
6	4.64	4.31	21.5296	18.5761	19.9984
7	4.60	4.28	21.16	18.3184	19.688
8	4.61	4.30	21.2521	18.49	19.823
9	4.60	4.32	21.16	18.6624	19.872
10	4.65	4.26	21.6225	18.1476	19.809
Σ	46.20	42.92	213.4472	184.216	198.2036

$\Sigma=(x-x\bar{x})^2 = 213.4472 - (46.20)^2 = 0.0032$; $\Sigma=(y-y\bar{y})^2 = 184.216 - (42.92)^2 = 0.00336$; $\Sigma=(x-x\bar{x})(y-y\bar{y}) = 198.2036 - (46.20 \times 42.92)/10 = -0.0868$; Hence, $Y = 26.83725401$

Table 2.B -iii: Correlations between light intensities of *A. pinnata* in 10 ppm

Sl.No	X	Y	X ²	Y ²	XY
1	4.94	4.31	24.4036	18.5761	21.2914
2	4.9	4.27	24.01	18.2329	20.923
3	4.93	4.30	24.3049	18.49	21.199
4	4.91	4.28	24.1081	18.3184	21.0148
5	4.92	4.29	24.2064	18.4041	21.1068
6	4.91	4.33	24.1081	18.7489	21.2603
7	4.9	4.32	24.01	18.6624	21.168
8	4.91	4.27	24.1081	18.2329	20.9657
9	4.94	4.26	24.4036	18.1476	21.0444
10	4.95	4.27	24.5025	18.2329	21.1365
Σ	49.21	42.9	242.1653	184.0462	211.1099

$\Sigma=(x-x\bar{x})^2 = 242.1653 - (49.21)^2 = 0.00289$; $\Sigma=(y-y\bar{y})^2 = 184.0462 - (42.9)^2 = 0.0052$; $\Sigma=(x-x\bar{x})(y-y\bar{y}) = 211.1099 - (49.21 \times 42.9)/10 = 0.001$; Hence, $Y = 0.25795827$

Table 2.B -iv: Correlations between light intensities of *A. pinnata* in 15 ppm

Sl.no	X	Y	X ²	Y ²	XY
1	5.2	4.99	27.04	24.9001	25.948
2	5.22	4.97	27.2484	24.7009	25.9434
3	5.23	4.96	27.3529	24.6016	25.9408
4	5.19	4.99	26.9361	24.9001	25.8981
5	5.21	4.98	27.1441	24.8004	25.9458
6	5.00	5.00	25.00	25.00	25.00
7	5.22	5.1	27.2484	26.01	26.622
8	5.25	4.97	27.5625	24.7009	26.0925
9	5.19	4.99	26.9361	24.9001	25.8981
10	5.19	4.98	26.9361	24.8004	25.8981
Σ	51.9	49.93	269.4046	249.3145	259.1868

$\Sigma=(x-x\bar{x})^2 = 269.4046 - (51.9)^2 = 0.0436$; $\Sigma=(y-y\bar{y})^2 = 249.3145 - (49.93)^2 = 0.0096$; $\Sigma=(x-x\bar{x})(y-y\bar{y}) = 259.1868 - (51.9 \times 49.93)/10 = -0.0099$; Hence, $Y = 0.483900815$

Table 2.B -v: Correlations between light intensities of *A. pinnata* in 20 ppm

Sl. No	X	Y	X ²	Y ²	XY
1	5	5.33	25	28.4089	26.65
2	4.9	5.32	24.01	28.3024	26.068
3	5	5.33	25.00	28.4089	26.65
4	4.8	5.31	23.04	28.1961	25.488
5	5.61	5.34	31.4721	28.5156	29.9574
6	5.62	5.35	31.5844	28.6225	30.067
7	5.63	5.36	31.6969	28.7296	30.1768
8	5.62	5.35	31.5844	28.6225	30.067
9	5.63	5.36	31.6969	28.7296	30.1768
10	5.62	5.35	31.5844	28.6225	30.067
Σ	53.43	53.4	286.6691	285.1586	285.368

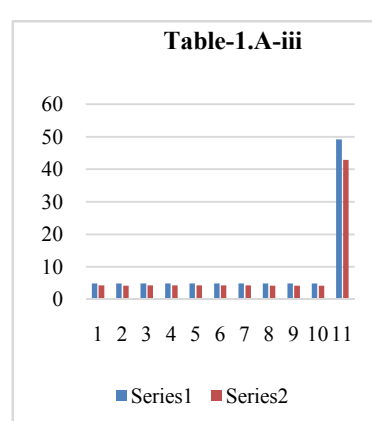
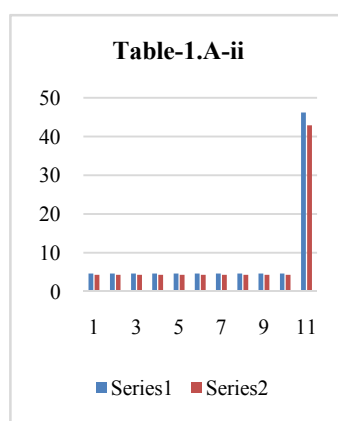
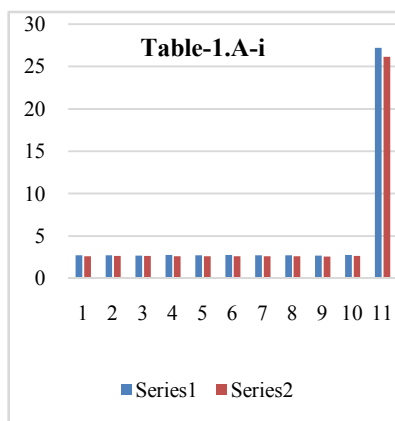
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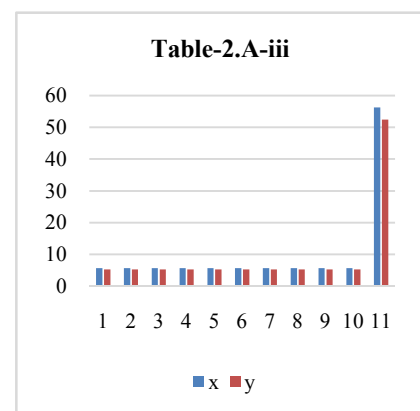
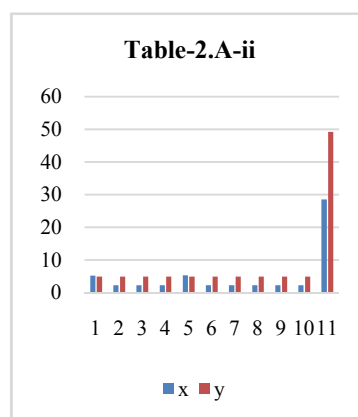
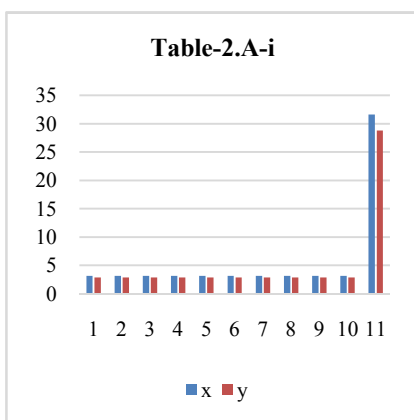
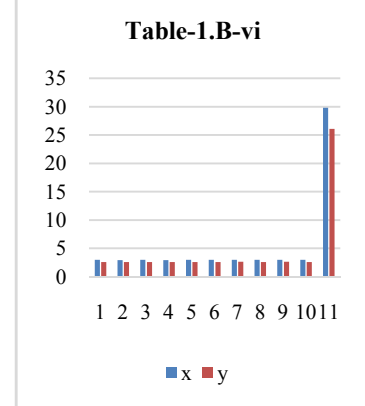
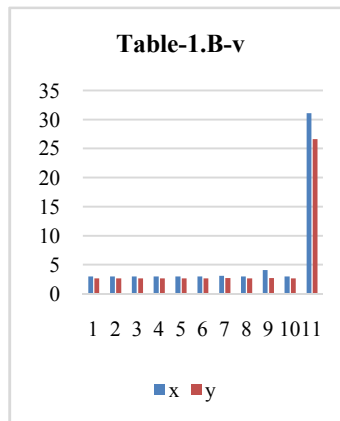
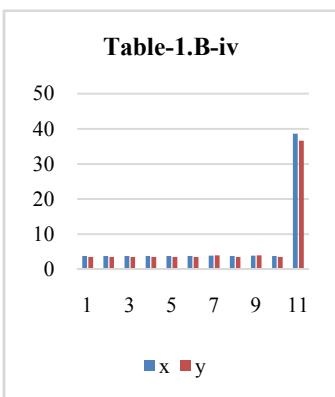
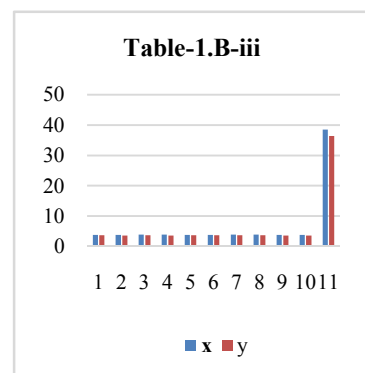
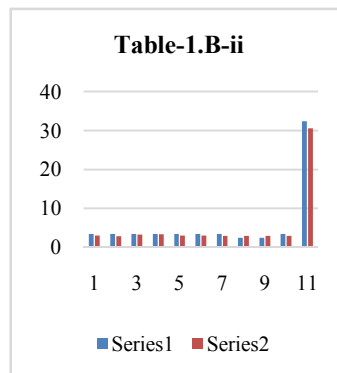
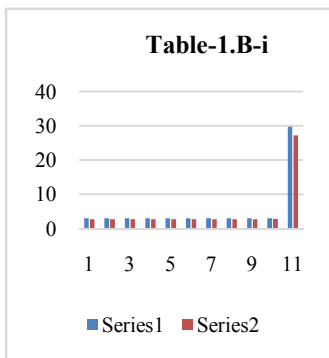
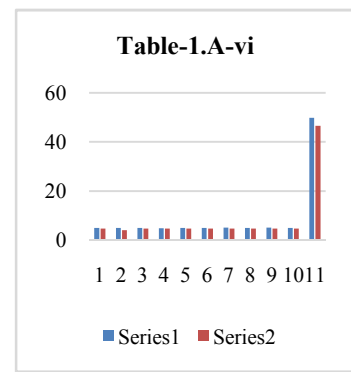
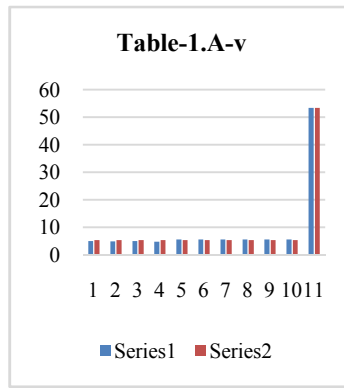
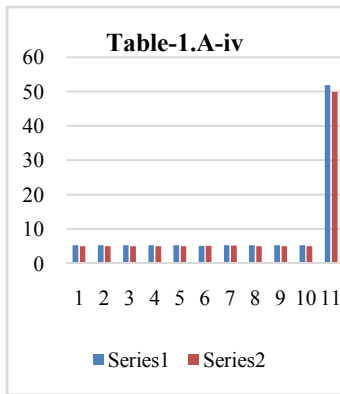
Table 2.B -vi: Correlations between light intensities of *A. pinnata* in 60 ppm

Sl. no	X	Y	X ²	Y ²	XY
1	5	4.71	25	22.1841	23.55
2	4.9	4	24.01	16	19.6
3	5	4.71	25	22.1841	23.55
4	4.8	4.69	23.04	21.9961	22.512
5	5.01	4.72	25.1001	22.2784	23.6472
6	5.02	4.73	25.2004	22.3729	23.7446
7	5.03	4.74	25.3009	22.4676	23.8422
8	5.02	4.73	25.2004	22.3729	23.7446
9	5.03	4.74	25.3009	22.4676	23.8422
10	5.02	4.73	25.2004	22.3729	23.7446
Σ	49.83	46.5	248.3531	222.5526	231.7774

$\Sigma=(x-x\bar{x})^2 = 248.3531 - (49.83)^2 = 0.05021$; $\Sigma=(y-y\bar{y})^2 = 222.5526 - (46.5)^2 = 6.3276$; $\Sigma=(x-x\bar{x})(y-y\bar{y}) = 231.7774 - (49.83 \times 46.5)/10 = 0.0679$; Hence, $Y = 0.12046341$

Graphical representation of each and Table has been cited hereunderin





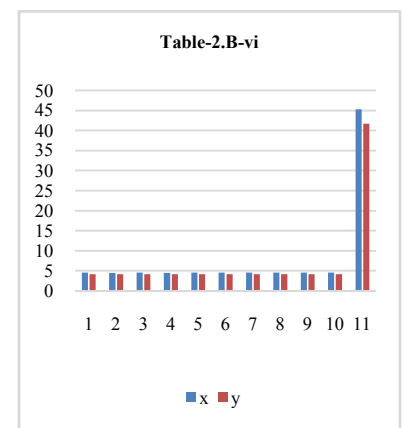
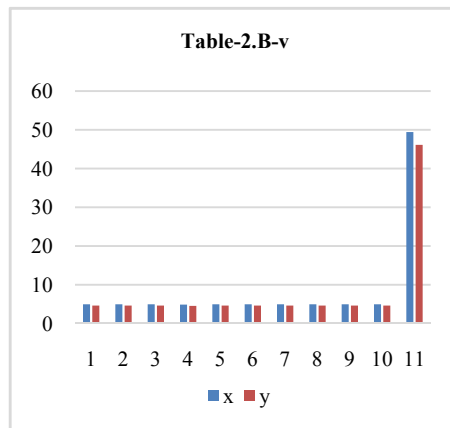
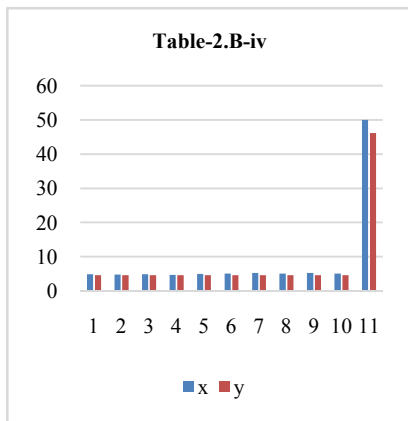
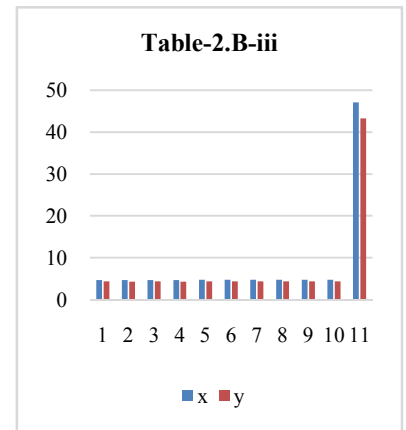
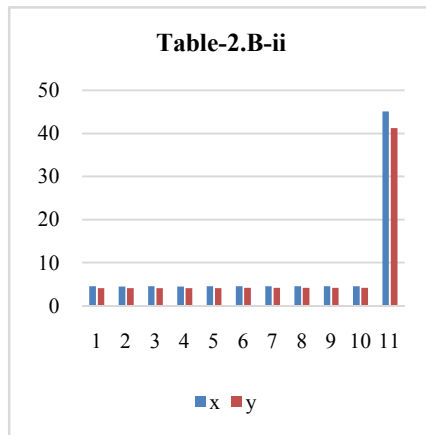
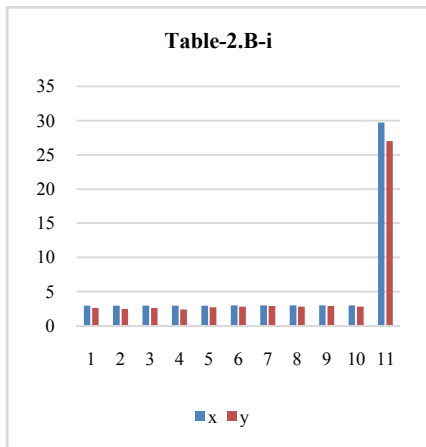


Table 3 Correlation Values at a Glance of Biomass production (Fr. wt. in g) of *Azolla filiculoides* and *Azollapinnata* at different concentrations of media-P, under two different light intensities, during the months of Jan- Feb. (O.A. = Open Area; P.S. = Partial Shade) (initial fr. wt. being 2 g in each case).

Concentrations of media-P (ppm)	Light intensities	<i>Azolla filiculoides</i>		<i>Azollapinnata</i>	
		Mean Value	Correlation value	Mean Value	Correlation value
0	O.A.	2.720	0.3553	2.970	0.2379
	P.S.	2.626		2.701	
5	O.A.	4.620	6.0166	3.445	25.5736
	P.S.	4.291		3.020	
10	O.A.	4.920	0.4003	3.856	0.0167
	P.S.	4.642		3.647	
15	O.A.	5.210	1.2300	3.860	2.0271
	P.S.	4.980		3.580	
20	O.A.	5.610	1.2033	2.990	0.9302
	P.S.	5.340		2.660	
60	O.A.	5.010	1.2046	2.980	1.2046
	P.S.	4.727		2.615	

Graphical representation of Table- 3 has been cited below:

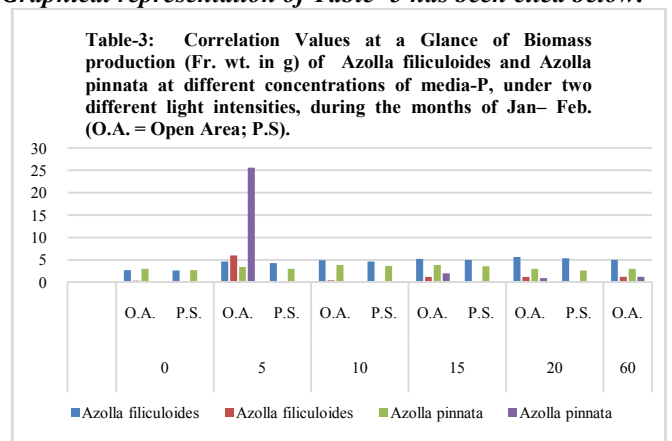
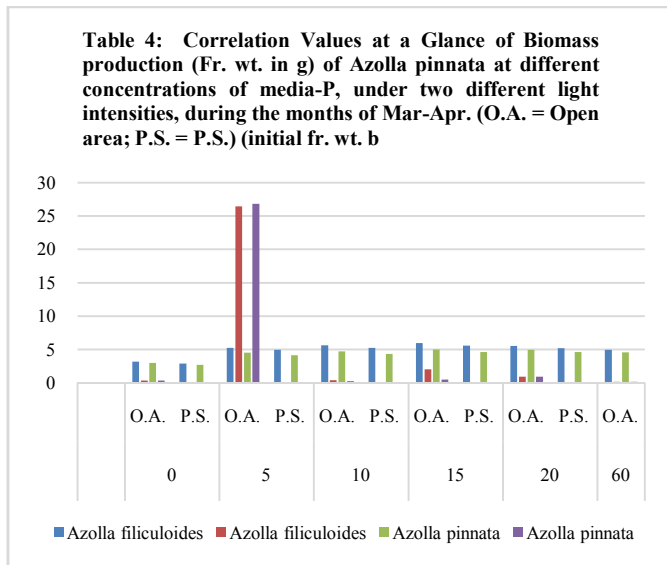


Table 4 Correlation Values at a Glance of Biomass production (Fr. wt. in g) of *Azollapinnata* at different concentrations of media-P, under two different light intensities, during the months of Mar-Apr. (O.A. = Open area; P.S. = P.S.) (initial fr. wt. being 2 g in each case)

Concentrations \of media-P (ppm)	Light intensities	<i>Azolla filiculoides</i>		<i>Azollapinnata</i>	
		Mean Value	Correlation value	Mean Value	Correlation value
0	O.A.	3.160	0.3553	2.970	0.3553
	P.S.	2.881		2.701	
5	O.A.	5.252	26.4712	4.518	26.8372
	P.S.	4.920		4.120	
10	O.A.	5.630	0.4003	4.710	0.2579
	P.S.	5.240		4.330	
15	O.A.	5.930	2.02710	5.000	0.4839
	P.S.	5.555		4.617	

20	O.A.	5.521	0.9230	4.944	0.9248
	P.S.	5.170		4.617	
60	O.A.	4.945	0.1204	4.538	0.1204
				4.175	

Graphical representation of Table-4 has been cited below



DISCUSSION

In the above experiments it has been found that in control set in case of both the species of *Azolla* i.e. *A. filiculoides* and *A. pinnata*, the biomass production as measured, was lowest in the months of Mar-Apr, but in the months of Jan-Feb, *A. pinnata* produced more or less same biomass at the control set as well as at 60 ppm of media-P. Both the species were found to produce the highest production of biomass at 15 ppm of media-phosphorous concentration during the months of March -April, but in Jan- Feb *A. filiculoides* showed the maximum production at 20 ppm of media-P whereas, *A. pinnata* showed at 10-15 ppm of media-P.

During the months of Jan-Feb, when the temperature ranged from 15.40°C to 25.20°C, relative humidity varied from 53.9% to 91% with a solar intensity of 203.77 $\mu\text{mol m}^{-2} \text{s}^{-1}$ in P.S. and 437.36 $\mu\text{mol m}^{-2} \text{s}^{-1}$ in O.A. and having no rainfall, maximum biomass was found to be produced by *A. filiculoides* i.e. 5.610 g in O.A. 5.340 g in P.S., at 20 ppm of media-P and *A. pinnata* producing 3.860 g in O.A. at 15 ppm of media-P, 3.647 g in P.S. at 10 ppm of media-P. The maximum decrease in fresh biomass at limiting concentration of media-P (5 ppm of media -P) was found in *A. pinnata*.

Again, during the months of March -April, when the temperature ranged between 23.07°C and 35.65°C relative humidity was 32.18% to 91% with a solar intensity of 532.98 $\mu\text{mol m}^{-2} \text{s}^{-1}$ in open area, 252.62 $\mu\text{mol m}^{-2} \text{s}^{-1}$ P.S., with no rainfall, *A. filiculoides* produced 5.930 g in O.A. and 5.555 g in P.S. at 15 ppm of media-P, and *A. pinnata* producing 5.000 g and 4.617 g in O.A. and P.S. respectively at 15 ppm of media-P.

During the months of March-April, it has been observed that, the tolerable concentration of media-P was 15-20 ppm of media-P, but the tolerable limit of *A. filiculoides* was 15 ppm of media-P. Though *A. pinnata* preferred lower concentration of media-P, but when the temperature arose, then the species showed the tendency to tolerate higher concentration of media-P. Again, *A. filiculoides* was a high media-P requiring species, it

has been found to utilize lower concentration of media-P as compared to winter season. O.A. was found to be more favourable than the P.S. for biomass production in both the cases.

The maximum biomass obtained by the two species at their respective optimum concentration were found to remain stable upto 20 ppm of media-P and 15 ppm of media-P during the months of Jan-Feb and Mar-April respectively in case of *A. filiculoides* but it was 15 ppm of media-P during the months of Jan-Feb, whereas, 20 ppm of media-P during the months of Mar-Apr in case of *A. pinnata*, indicating the tendency to utilize higher concentration of media-P during summer by *A. pinnata* and lower concentration of media-P by *A. filiculoides* as compared to winter season.

The graphical curve was falling down linearly up to the 60 ppm phosphorous concentration and was observed less than control set in case of the control set-up experiment run during January-February and March-April months. It has been observed that experiments run during January-February months i.e. during winter month was stable in biomass production which indicates the acceptability of phosphorous media.

The effect of different concentrations of phosphorous which have been cited in tables and graphs. Indeed, phosphorous has proved its positive role to enhanced the stable growth of the plant population in 20 ppm concentration. Further, it does require in any higher concentrations up to 60 ppm. Some relevant references of this findings also supported this hypothesis. Our results are consistent with the hypothesis that temperature might be an important factor determining the fitness of floating macrophytes (Janes 1998; van der Heide *et al.* 2006; Netten *et al.* 2010; Szabo *et al.* 2010; Peters *et al.* 2013; Watanbe *et al.*, 1977, 1981). Warm temperatures during the winter might open “windows of opportunity” that promote the fast growth of *Azolla* mats before the spring establishment of submerged macrophytes. A positive increase in the fitness of floating species (*Azolla* among others) in response to local warming has also been described in temperate areas, such as in a thermal stream in Slovenia (Sajna *et al.* 2007), and in a portion of the River Erft (Germany) which has been abnormally warmed as a consequence of opencast mining water discharges (Hussner and Löscher 2005). Several studies have pointed out that a major consequence of increasing nutrient loading in water bodies is the displacement of functional groups responsible for primary production from submerged to floating macrophytes, some of which may be invasive and outcompete floating macrophytes (Morris *et al.* 2003; Meerhoff *et al.* 2007; Netten *et al.* 2010; Szabo *et al.* 2010; Scheffer *et al.* 2003). In water ecosystems these shifts can affect species assemblages, sediment biogeochemistry and water quality. Therefore, one major consequence of dense blooms of *Azolla* is the decrease of submerged macrophyte cover (Janes *et al.* 1996). Although we did not study its effect on submerged macrophytes, threshold irradiance for maintaining autotrophic communities dominated by submerged macrophytes have been identified in Doñana (Geertz-Hansen *et al.* 2011). The effect of this important nutrient on the overgrowth of *Azolla* has also been confirmed in Anzaliwetland (Sadeghi *et al.*, 2012a, 2012b). In laboratory experiments, Jane (1998) found that increasing phosphorous supply led to increase sporulation. Kushari and Watanabe (1991), Kushari and Watanabe (1992), observed the optimum

growth of different species belong to *Azolla* genus which responded to different concentrations of phosphorous.

Biomass production of both the species of *Azolla* were shown as tabulated form in the Table-3 for the month of January - February and in the Table-4 for the month from March - April. It has been evident clearly that the correlation value of *A. pinnata* seems to be greater than that of *A. filiculoides* in both the seasons. Fannah (1987) reported a completed life cycle of *Elophila africalis* on *A. pinnata* in Sierra Leone which was followed up by Roberts *et al.* (1998). Sands and Kassulke (1986) reported oviposition by females of *Paulinia acuminata* after feeding on *A. pinnata*. Therefore, it is unlikely that it is an important constraint on *A. pinnata* Stewart *et al.* (1968, 1976, 1977, 1980, 1982).

Singh *et al.* (2010) studied the effect of micronutrients (e.g. Mo⁶⁺, Mn²⁺, Zn²⁺, Cu²⁺ and Fe²⁺) on cellular and extracellular activities of two *Azolla* species (*A. microphylla* and *A. filiculoides*) exposed to a P-deficient, saline (20mM NaCl) medium. At lower concentrations (0-0.01mM), the micronutrients showed a significant enhancement in the given activity, whereas higher concentrations (e.g. at 10 mM) played an inhibitory role. Sadeghi *et al.* (2012b) reported a moderate effect of Fe on the growth of *A. filiculoides* in the Anzali wetland.

CONCLUSION

In order to have a successful wetland restoration and conservation management program one has to get acquainted with the habitat requirements of invasive aquatic fern species such as *Azolla*. However, this mosquito fern has many benefits (e.g. nitrogen fixation, phosphorus removal from wastewater, or use as green fertilizer), until now, little is known about the negative impacts of *Azolla* (as an invader or alien species) on a new environment. This paper reviewed the most important structural habitat variables in order to meet the habitat requirements of *Azolla* including water, light intensity, air and water temperature, relative humidity, wind velocity and waves. Moreover, the importance of physical-chemical variables for *Azolla* has been confirmed from the cited literature. Phosphorus is considered to be the most important macronutrients to induce the growth of *Azolla*.

On the other hand, some micronutrients (e.g. molybdenum, cobalt and vanadium) are well known to stimulate the growth of *Azolla*. The structural habitat variables probably have a more important effect on growth of *Azolla* compared to the physical-chemical ones. Among the biological factors covered, insects, bacteria, fungi and viruses have been shown to affect growth and development of *Azolla*. As a final conclusion, getting more insight into abiotic and biotic factors affecting growth of *Azolla* will help future research and management of this aquatic fern. On the contrary, it has also been found the growth was better in open area condition than that of the partial shade condition of cultivation in both the species supported by the evidence of research work of

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