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Potential of *Azolla pinnata* for Phytoremidiation of Sewage waste water

Potencial de *Azolla pinnata* para la Fitorremediación de Aguas Residuales

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ABSTRACT

Phytoremediation offers an attractive and cost effective way for treatment of wastewater. Azolla , an aquatic pteridophyte is an excellent member for phytoremediation of wastewater and industrial effluents. In this study we investigated the ability of *Azolla pinnata* for improvement of water quality parameters like pH, TDS (Total dissolved solids), EC(Electrical conductivity) and Salinity in STP water collected from Khasra 359 Shantinagar site, Dayalbagh, Agra. Verification of phytoremediation capability of *A.pinnata* was done. STP water was collected , each tub filled with 8 L of STP water and inoculated with 50g, 100g 150g of fresh *A.pinnata* in separate tubs respectively along with a control. Experimental setup left undisturbed and Azolla was allowed to grow for 3 weeks. After 3 weeks of growth water sample collected from each tub. pH, TDS, EC salinity of the STP water was taken before and after treatment with *A.pinnata* along with the control.The pH, salinity, TDS and EC of water sample decreased in orderly manner from the initial analysis to after treatment and with increasing concentration of *Azolla* inoculated. The values of different parameters found to be decreased significantly.

Keywords: Phytoremidiation, *Azolla Pinnata,* Waste water, Shantinagar Dayalbagh, Bioremidiation, aquatic, pteridophyte.

RESUMEN

La fitorremediación ofrece una forma atractiva y rentable de tratar las aguas residuales. Azolla, una pteridofita acuática, es un excelente miembro para la fitorremediación de aguas residuales y efluentes industriales. En este estudio se investigó la capacidad de Azolla pinnata para la mejora de los parámetros de calidad del agua como pH, TDS (sólidos disueltos totales), EC (conductividad eléctrica) y la salinidad en el agua STP (estación depuradora de aguas residuales) recogidos de Khasra 359 Shantinagar, Dayalbagh, Agra. Se verificó la capacidad de fitorremediación de A. pinnata. Se recogió agua STP, cada cubeta se llenó con 8L de agua STP y se inoculó con 50g, 100g y 150g de A. pinnata fresca en cubetas separadas respectivamente junto con un control. El montaje experimental se dejó intacto y se permitió que la Azolla creciera durante 3 semanas. Después de 3 semanas de crecimiento se recogieron muestras de agua de cada cubeta. Se tomaron muestras de pH, TDS, EC y salinidad del agua STP antes y después del tratamiento con A.pinnata junto con el control. El pH, salinidad, TDS y EC de la muestra de agua disminuyeron ordenadamente desde el análisis inicial hasta después del tratamiento y con el aumento de la concentración de Azolla inoculada. Los valores de los diferentes parámetros disminuyeron significativamente.

Palabras clave: fitorremediación, Azolla pinnata, aguas residuales, Shantinagar Dayalbagh, biorremedación, acuatico, pteridofitas.

INTRODUCTION

Water is one of the most abundant and useful resource present on Earth. Although 70% of the earth surface is covered with water still there is lack of water with desired quality. Rapid industrialization, urbanization, and population growth in the last few decades have added huge loads of pollutants in the water resources (CPBC2008). The pollutants are increasing day by day and due to advancements in various fields there is a wide variety of pollutants. These pollutants maybe added to the water resources through different sources like Agriculture, Industries, Domestic sewage, solid wastes, inappropriate dumping practices, oil spill secretion. Number of industries including textile, printing, paper and pulp, iron–steel, electroplating, coke, petroleum, pesticide, paint, solvent, and pharmaceuticals etc. consume large volumes of water and chemicals differing in composition and toxicity.

Addition of these pollutants in water resources poses serious impact on biodiversity. The pollutants when dumped into the water resources they tend to accumulate and ultimately enters the food chain and cause serious health hazards. When such polluted water is either directly consumed by human beings or pollutants reaches in the body through some other way causes serious health risks and diseases in man including heavy metal toxicity.

The toxic substances present in water may be organic or inorganic in nature. Inorganic pollutants include metalloids, heavy metals, trace elements, non-metallic salts and radioactive elements. Organic toxic substances mainly come from the agricultural activities like application of chemical fertilizers, herbicides, pesticides, soil conditions and by the domestic sewage waste water.

Various remediation methods are used for the removal of these pollutants from water but those are proved to be in effective when the concentrations of toxic substances are either too high or low and are very costly. Bioremediation and phytoremediation is a cost effective way with low inputs for in-situ remediation (Saltetal.1995). Phytoremediation has several advantages over conventional procedures like low cost input, effectiveness and easy applicability. The term phytoremediation or bioremediation are first the use of methodologies where living organisms are used (especially lower organisms like algae, fungi, bacteria & pteridophytes etc.) for the reduction or elimination of toxic substances from the water or for any other waste treatment. Phytoremediation process involves various mechanisms like absorption, degradation, sequestration, accumulation, assimilation or transpiration of toxic substances by the organism used. This process utilizes the natural capability of the organism to reduce or eliminate a particular pollutant. The organisms used for bioremediation maybe genetically modified through biotechnological approaches to increase their efficiency of remediation.

Analysis of water quality

The suitability of water for various uses like irrigation, drinking and diverse domestic purposes etc. depends upon its quality. The quality of water is determined by its physical, chemical and biological properties.

Purposes of water quality analysis

- To measure concentration of the constituents in quantity for characterization of water for different uses
- To safeguard human health and protection of aquatic life.
- To maintain the permissible limits of different components.
- To prevent possible impact on health risks, damage of biodiversity & aquatic environment.

There are a number of parameters used for the analysis of water quality. It includes-

- Physical parameters- pH, color, temperature, turbidity, Total dissolved solids, suspended solids, conductivity etc.
- Chemical parameters- BOD (Biochemical oxygen demand), COD (Chemical oxygen demand), DO (Dissolved oxygen), total nitrogen content, phosphorus, chloride, pesticides etc.
- Biological parameters- Test for coliforms and quantity of microorganisms.

Table:1 Water quality parameters and their associated risks

S.No.	Parameter	Potential health effect	
1	pH	Affects mucous membrane; bitter taste; corrosion	Patil et.al., 2012

2	Dissolved oxygen	D. O. corrodes boilers, water lines and heat exchangers.	Li, et.al., 2018
3	Total Hardness	Poor lathering with soap; deterioration of the quality of clothes; scale forming	Patil et.al., 2012
4	Total Alkalinity	Embrittlement of boiler steel. Boiled rice turns yellowish	Patil et.al., 2012
5	TDS	Undesirable taste; gastro-intestinal irritation; corrosion or incrustation	Saravanakumar, K., & Kumar, R. R., 2011
6	Biochemical Oxygen Demand (B.O.D.)	High BOD decreases level of dissolved oxygen.	Susilowati, S., et.al., 2018
7	Electrical conductivity	Conductivity is due to ionizable ions. Corrosive nature of water is increased by higher EC.	Kumar., et. al., 2017
8	Turbidity	Higher level of turbidity are associated with disease causing bacteria.	Igbinosa et. al., 2012

We have analyzed the following parameters:

1. pH

pH is one of the most important factor in determining the corrosive nature of water. Low pH value makes water more corrosive in nature. pH is found to be positively correlated with electrical conductance (EC) and alkalinity (Gupta 2009). The reduced rate of photosynthesis and assimilation of carbon dioxide and bicarbonates are ultimately responsible for increase in pH. The high temperature coincided with low oxygen values during the summer months.

A number of factors bring about changes in the pH of water. The higher pH values suggested that carbon dioxide, carbonate-bicarbonate equilibrium is affected due to change in physicochemical condition (Karanth 1987).

2. EC (Electrical Conductivity)

Conductivity is found to have significant correlation with the ten parameters such as temperature , pH value , alkalinity , total hardness , calcium , total solids, total dissolved solids(TDS) , chemical oxygen demand(COD) , chloride and iron concentration of water.

3. Salinity

Salinity represents the concentration of salts present in water. *It* refers to the measure of the amount of dissolved salts in *water*. It is usually expressed in percentage (%) or parts per thousand (ppt). Salinity affects the growth & development of aquatic flora & fauna and induces land degradation as well if used for irrigation making by making the soil saline.

4. TDS (Total Dissolved Solids)

Total concentration of dissolved substances in water represents TDS. It refers to the amount of organic and inorganic materials, such as metals, minerals, salts, and ions, dissolved in a particular volume of water; TDS are essentially a measure of anything dissolved in water that is not a water H_2O molecule. Dissolved solids can produce hard water.A high concentration of TDS is an indicator that harmful contaminants ,such as iron, manganese, sulphate , bromide & arsenic can also be present in water.

A number of procedures are used for removal of pollutants from water. Phytoremediation offers an attractive alternative for water treatment and is a cost effective method for in situ remediation (Salt et al. 1995). *Azolla,* an aquatic pteridophyte is considered to be an excellent member for phyto-accumulation of heavy metals and improving water quality parameters because it possesses qualities required for a plant to be used in phytoremediation. These properties which makes *Azolla* an ideal candidate for phytoremediation are high biomass production, fast growth rate, moderately extensive root system, easy harvesting and tolerance to wide range of heavy metals (Sood et.al.2012). *Azolla* sps. are found to be effective in improving the water quality parameters so it can be used for the treatment of wastewater.

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MATERIALS AND METHODS

Cultivation of *Azolla pinnata*

Cultivation of Azolla is an easier task as it does not require very specific nutrient composition and growing conditions. Azolla flourishes well in wild habitat and it can also be cultivated in controlled conditions. It requires a shallow pond to grow.

Figure-1 *Azolla* Cultivation Unit

Requirements:

- 4 plastic tanks of size 1*1 m with 1 feet depth
- Compost
- Water
- Fertile soil
- Azolla seeds / Inoculum

Procedure:

Took 5 kg of clean fertile soil and 5 kg compost powder for each tank, mixed it with water and uniformly spread across the pond/tank. Maintained water level upto 6 inches in the tank. Applied appropriate quantity of *Azolla pinnata* culture uniformly. The culture was allowed to grow for 2 weeks under natural climatic conditions.

Appropriate environmental conditions were provided-

- Kept the culture under shade to avoid direct contact with sunlight (Partial shade)

- Maintained water level upto 6 inches throughout the culturing period by adding required quantity of water.

Figure-2 *A.pinnata* growing

Analysis of Water Quality Parameters (pH, Salinity, TDS & EC)

Analysis of pH, Salinity, TDS & EC of STP water before treatment was done using Water analyzer 371.

- Water sample was collected from the flowing stream at Shantinagar site (Khasra 359).

- pH, Salinity,TDS & EC were measured using Water analyser 371.

➢ Treatment of STP water by *Azolla pinnata*

After control experimental design was used. Experimental setup was done where STP was inoculated with different quantities of *A.pinnata* along with a control.

- Took 4 tubs and filled 8 L of water in each of those.

- Tub 1 was left as such.

- Tub 2, 3& 4 were inoculated with 50g,100g, & 150g of *A.pinnata* respectively and left for growth.

- The setup was maintained for 3 weeks under natural environmental conditions.

Figure-3 Treatment of STP water with *A. Pinnata*

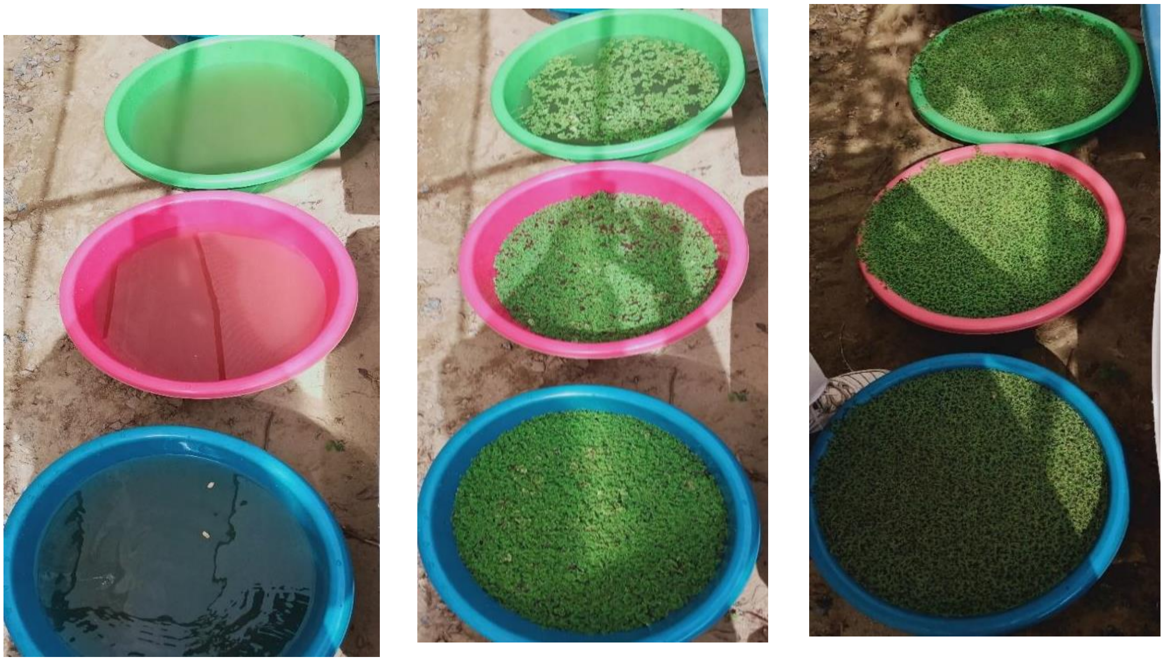

Fig. 3.A.Tubs filled with STP water

Fig. 3.B.Water inoculated with *A.pinnata* – 50g,100g & 150g of *A.pinnata* in tubs A ,B & C respectively.

Fig. 3.C. After 3 weeks of growth.

- Analysis of Water Quality Parameters after treatment

After 3 weeks Water sample was collected from each of the tubs along with the control and pH,Salinity, TDS & EC were analysed using water anlyser 371.

The water analyser was first calibrated and then the respective electrodes were dipped in each sample and readings were noted.

Figure-4 Water Samples before Treatment

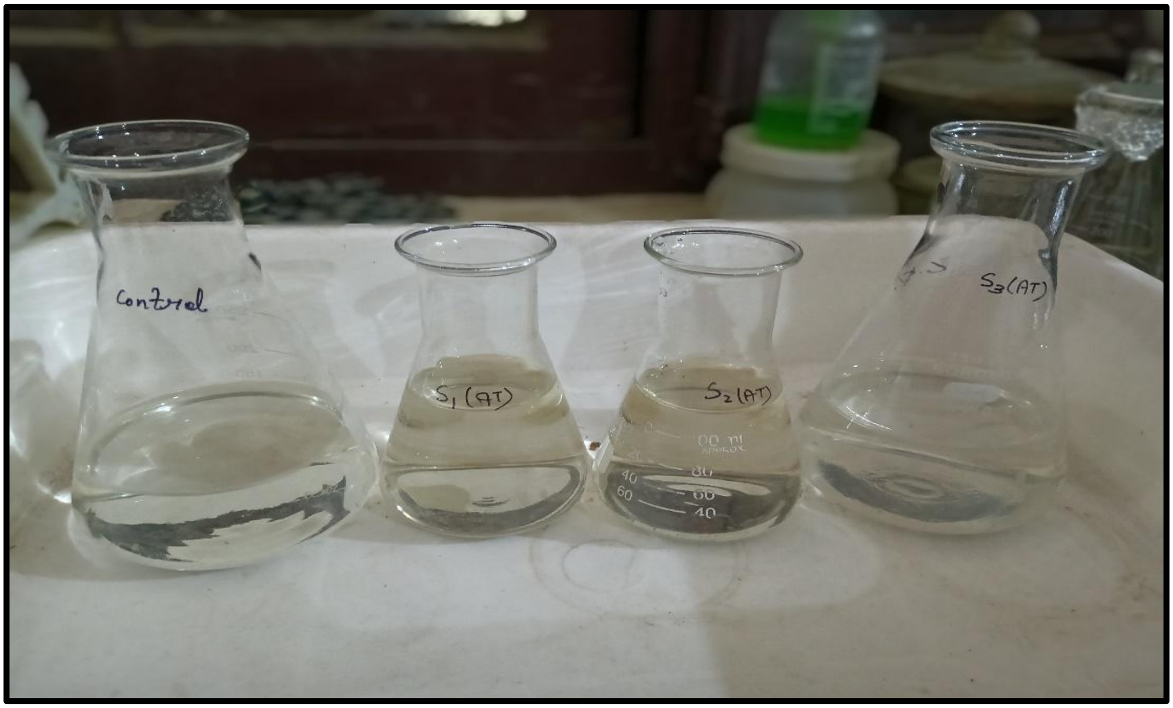

Fig- 4 Water samples after 3 weeks of Treatment -1. Control, 2. Sample 1(STP water inoculated with 50 g of *A.pinnata)*, 3. Sample 2(STP water inoculated with 100 g of *A.pinnata)*, 4. Sample 3(STP water inoculated with 150 g of *A.pinnata)*.

RESULT & DISCUSSION

Table-2 indicating the observed values of different parameters

S.No.	Sample	pH	Salinity	EC	TDS
1	STP water *	7.78	0.72 ppt	2.08 mS/cm	1.07 ppt
2	Control **	7.78	0.72 ppt	2.08 mS/cm	1.07 ppt
3	Sample 1**	7.73	0.43 ppt	1.38 mS/cm	0.65 ppt
4	Sample 2**	7.68	0.23 ppt	0.695 mS/cm	0.349 ppt
5	Sample 3**	7.53	0.02 ppt	0.159 mS/cm	0.036 ppt

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* - Before treatment and ** - After treatment

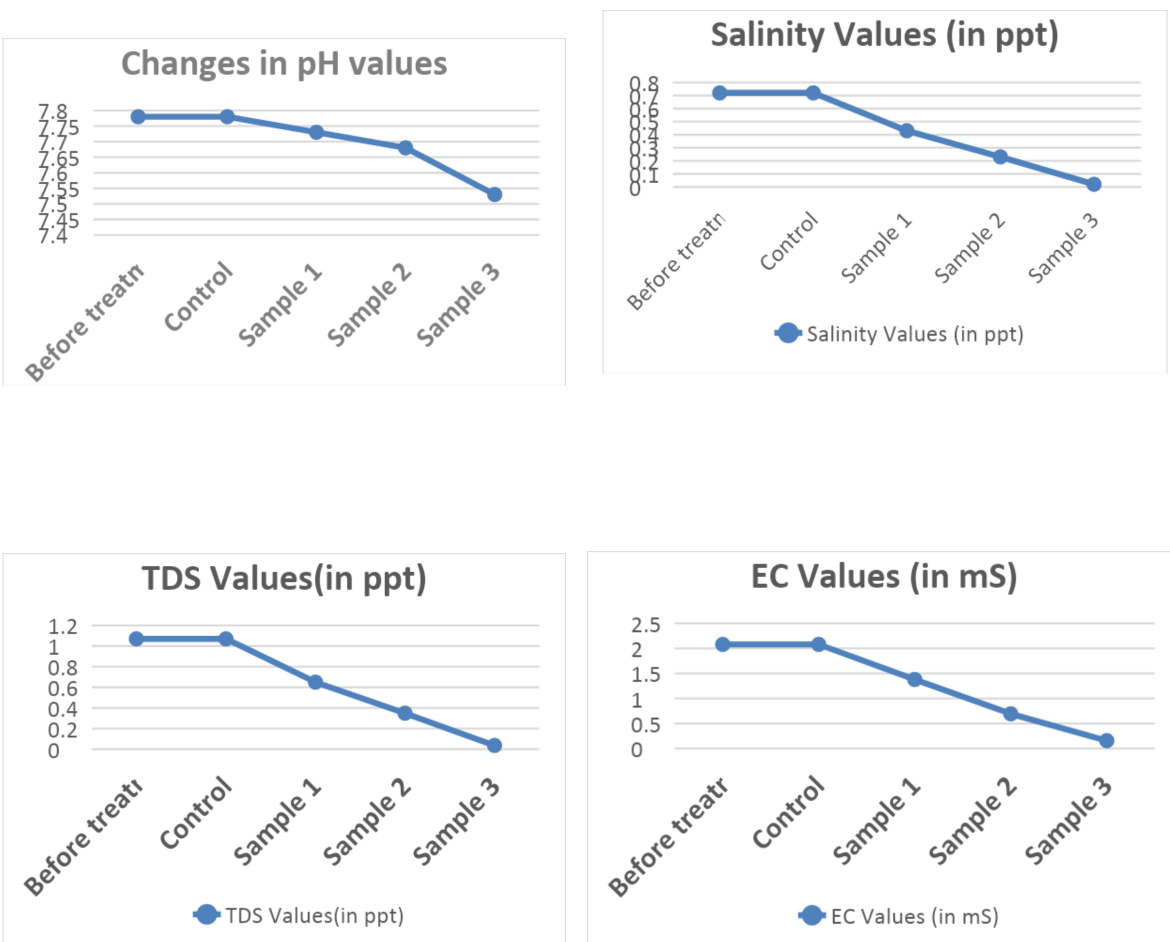
The results show changes in pH of STP water from 7.78 before treatment to 7.78 in control, 7.73 in sample 1, 7.68 in sample 2 and 7.53 in sample 3 respectively.

Salinity (in ppt) changes from 0.72 before treatment to 0.70 in control, 0.43 in sample 1, 0.23 in sample 2 and 0.02 in sample 3 respectively after treatment.

EC (Electrical conductivity in mS) changes from 2.08 before treatment to 2.08 in control, 1.38 in sample 1, 0.695 in sample 2 and 0.159 in sample 3 respectively after treatment.

TDS (Total dissolved solids in ppt) changes from 1.07 before treatment to 1.07 in control,0.65 in sample 1, 0.35 in sample 2 and 0.036 in sample 3 respectively after treatment.

Figure-5



CONCLUSION

The aquatic pteridophyte *Azolla pinnata* is found to be effective as a potential agent for treatment of water by improving various water quality parameters.

The pH, Salinity, TDS & EC of STP water supplied to Khasra 359 shantinagar site were tested.

A.pinnata has been successfully cultivated at Khasra 359 Shantinagar site , Dayalbagh, Agra and experimental setup was placed there.

After experimentation the effect of growing *A.pinnata* on the quality of water was observed. Water quality parameters –pH, Salinity, TDS & EC were tested after treatment.

The values of different parameters found to be decreased significantly over a period of time after treatment.

SIGNIFICANCE & FUTURE PROSPECT

Water is required by all living organisms for their survival and growth. Maintenance of water quality to be used for different purposes is the most important task in today's world where a number of human activities are consistently adding harmful pollutants and contaminants in water resources which ultimately impacts different components of ecosystem.

Phytoremediation provides a cost effective way to maintain water quality by altering physicochemical or biological properties of water through different mechanisms like accumulation, degradation, absorption etc. of the pollutant. *Azolla*, a macrophyte possess the capacity to reduce the pollutants can be used in waste water treatment procedures to make water suitable for the purpose.

The results of this study indicates that water quality parameters have been successfully improved after treatment so *A.pinnata* can be effectively used for water treatment. It can be effectively utilized for improvement of water quality for irrigation, drinking, domestic and other purposes.

STP water is generally used for irrigation purpose in different localities. Its poor quality affects the soil quality and vegetation growth. This problem could be solved through phytoremediation in cost effective manner. Since the ability of *A.pinnata* to treat water is verified through this experiment it could be utilized in various fields and aspects where the remediation has to be done under natural conditions and in cost effective way.

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