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# Wastewater Treatment by Azolla: A review

**Prabhasha Jayasundara**

Department of Chemical and Process Engineering, Faculty of Engineering, University of Peradeniya, Sri Lanka

Corresponding author: prabhashajayasundara@gmail.com

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**Abstract**

Water is vital for humans and other living things, but water pollution has become a significant issue today. Various anthropogenic agricultural, industrial, and home activities produce multiple organic and inorganic substances dissolved or suspended in water. The goal of wastewater treatment is thus twofold: to reduce water pollution while also maintaining the water supply to demand. It is based on the three 3 Rs: reduce, reuse, and recycle. However, many of the methods used are ineffective or expensive. As a result, water purification is a matter of great interest. Eco-friendly approaches are essential among the new technologies and techniques tested in wastewater treatment. Aquatic macrophytes treat water by accumulating harmful metals and nutrients. Like water filters, a variety of aquatic floats can be suggested. Azolla is one among them, and it has been used for decades. Researchers have discovered that Azolla can reduce Chemical Oxygen Demand (COD), Biochemical Oxygen Demand (BOD), nitrogen, phosphorus, and heavy metal concentrations in wastewater; therefore, it can be utilized for wastewater treatment. Azolla also produces a lot of biomasses in wastewater. The use of Azolla in reducing eutrophication in lakes and streams and other benefits is discussed in this study. According to the literature, Azolla has a high growth rate, with a doubling date of 2-4 days. Azolla's cell wall is composed of pectin, which has a high affinity for the adsorption of organic substances. Azolla serves as a "biofilter" during wastewater treatment in this way.

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## Introduction

Environmental pollution has been discussed since the industrial revolution, and today, it is an essential topic all over the world in economy and politics. The pollutants in the form of organics from herbicides, insecticides, and inorganics like trace elements, heavy metals, and metalloids are being discharged into different compartments of the earth- soil, water, and air (Soman and Arora, 2018). Water pollution can be divided into groundwater and surface water pollution. Surface water pollution happens for many reasons; releasing organic and inorganic nutrients to the water discharging untreated sewage, heavy metals, and chemicals to water bodies. While water is a limited resource, the quality and quantity decrease day by day. Unless environmentally and economically sustainable management technologies are developed, the accumulation of these wastes may cause disposal and contamination difficulties. As a result, the development of cost-effective wastewater treatment methods should be studied (Amuda and Ibrahim, 2006).

Some plants can absorb and accumulate high concentrations of nutrients and heavy metals; this process is known as phytoremediation, and this process can be used in wastewater treatment. So many researchers have found that aquatic macrophytes have a high potential to absorb nutrients and heavy metals in water, with high biomass yield. Researchers have found that among the fern used in constructed wetlands, such as duckweed cattails, Azolla is more efficient than others. For decades, the symbiotic relationship has been used as green manure in rice fields because of its habitat and nitrogen-fixing capabilities. Because of its ability to extract phosphorous

and nitrogen from the water medium, there is growing interest in employing the fern for additional applications, such as a decontaminant plant in low-cost wastewater treatment systems (Forni *et al*., 2001).

The findings of A.A.Deshmukh et al. demonstrated that the treatment of sewage Azolla does not require the installation of a complicated unit or any elaborate mechanism. As a result, it is effective and should be used, particularly in developing nations. Because the treated water compares favorably to untreated water, it can be discharged into the environment without causing harm to the environment or human health. As a result, a low-cost, environmentally friendly, and effective treatment method has been developed. Azolla is a beneficial plant for controlling the number of nutrients in sewage and for helping to tackle the problem of overcrowding. Eutrophication and phytoremediation are environmentally friendly sewage treatment methods that are particularly effective when combined with other treatment methods (Deshmukh *et al*., 2013).

***Figure 1:Azolla (Sood et al., 2012)***

Azolla can be used as a biofertilizer in rice agriculture, as green manure, in saline soil reclamation, as a component of space cuisine, mosquito repellant, weed control, and biogas production. The current review focuses on Azolla's phytoremediation potential in various wastewaters and biomass output. This review aims to analyze the phytoremediation potential of Azolla and to analyze the growth kinetics of Azolla for different kinds of wastewater.

**Characteristics and Uses of Azolla**

The genus Azolla was established by Lamarck in 1783. Azolla is derived from two Greek words, "azo," which means to dry, and "alloy," which means to kill. It has several advantages, the most important being the ability to grow in the least time, due to which it is referred to as a "super-plant" (Soman and Arora, 2018). Azolla is a small floating plant species. It is a genus name belonging to the family *Azollacea*. Azolla has different species such as *A.pinnata, A.caroliniana, A.filiculoides, A.microphylla, and A.imbricata*. It is widespread in aquatic bodies. Their leaves are water repellent and like small-scale roots hanging in the water. Azolla form symbiosis with Cyanobacteria, Namely *Anabaena azollae*. These bacteria can fix nitrogen. Therefore, the plant can quickly get nutrients. They have a higher growth rate. Phosphorous is one of the limiting minerals. Phosphorous abundance sometimes leads to Azolla blooms. Symbiotic bacteria transfer from one generation to the next.

The nitrogen sequestering potential of the organism makes it play a significant role in agriculture by increased rice productivity due to Its application as a dual crop, and a biofertilizer and biomass can be used as a bio-fertilizer or as a feed supplement for aquatic and terrestrial animals due to its protein; crude fiber and mineral content; Moreover, the growth of Azolla in natural water bodies forms a mat which can help reduce the evaporation of water from them thereby making water available year-round (Soman and Arora, 2018).

According to the results of previous continuous assays, using Azolla, an aquatic fern with a high growth rate and productivity, to improve treated urban wastewater quality appears to be very promising, primarily because of its phosphorus removal efficiencies (40-65 %) and heavy metal biosorption using living Azolla (phytoremediation) or dried biomass as a bioadsorvent material are both relatively new technologies for metal removal (Costa *et al*., 2010).

Research has been investigated Azolla can be used as a space diet and Space agriculture based on the menu with Azolla. According to the International Rice Research Institute reports, "The use of *Azolla* in controlling water pollution" is a major research priority.

## Phytoremediation Ability of Azolla

### Chemical oxygen demand (COD)

The COD value represents the extent of pollution of a water body; the more the COD, the more the pollution is. However, its value numerically depends on the number of compounds that can be oxidized present in the water sample only. Different physical, chemical, and biological methods in conventional wastewater treatment reduce COD load. Wastewater supplies macronutrients such as carbon, nitrogen, phosphorus, and micronutrients such as sodium, chlorine, and iron. When an aquatic plant is used for water treatment, it absorbs that nutrient and grows up. Azolla biomass has been used to reduce COD in wastewater by various researchers (Golzary *et al*., 2018; Amuda and Ibrahim, 2006; Soman and Arora, 2018). Table 1 shows their results. The petroleum wastewater mainly consists of organic compounds, and hence with time, the COD removal percentage increases as the biomass of Azolla increases.

Azolla is also efficient in the removal of nitrogen and phosphorous. Hence, the wastewater mixed with cow dung increased the nitrogen and phosphorus content of the sample and decreased the percentage COD removal rate than unmixed sewage water, for COD takes only an amount of organic substance into the calculation. A part of Azolla removes phosphorus and nitrogen, and thereby the percentage of COD removal rate decreases. After secondary treatment, the system's physiochemical parameters like temperature and pH have also changed, affecting the percentage COD removal rate.

### Growth kinetics

The laboratory studies showed that Azolla could double its biomass in 3.5 days and grow in a nitrogen-free solution since it does not need a nitrogen nutrient medium for its biomass growing (Golzary *et al*., 2018). Because of this fast-growing kinetics and the floating nature of Azolla help in its easy harvest for disposal or recovery of heavy metals from biomass.

At a particular experimental condition, Figure 2 shows the influence of temperature on the Azolla growth rate. Azolla growth was reported to be better at room temperature. The temperature influence on Azolla growth rate was studied at temperatures ranging from 10 to 50 C°. The optimal temperature for the Azolla growth rate was 30 C°. As can be seen, Azolla growth was reduced at temperatures above and below 30 C° (Zazouli *et al*., 2014).

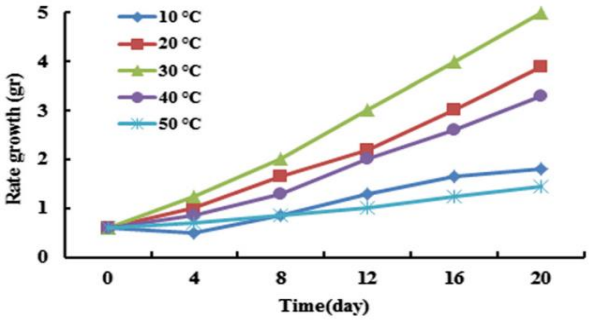


***Figure 2: Effect of temperature on Azolla growth (initial biomass = 0.9 g)***

### Phytoremediation of heavy metals

Naghipour *et al.* (2018) have studied the phytoremediation of heavy metals (Ni, Cd, Pb) by Azolla. They have followed the following method. "The Azolla was taken from a lake near Rasht city and rinsed in tap water before being weighed (0.2, 0.4, and 0.8g) and maintained in a 100 mL disposable container for 15 days in the presence of 5, 10, and 25 mg $L^{-1}$ of Lead, Nickel, and Cadmium ions." The samples were kept in polyethylene containers to determine the metal concentration using "ICP-OES."(Naghipour *et al*., 2018).

The maximum efficiencies they have found are listed in table 2.

***Table 1: Literature on COD removal percentages by Azolla***

| Type of wastewater | COD/ mg $L^{-1}$ | Number of days | COD removal percentage | Reference |
|---|---|---|---|---|
| Petroleum refinery wastewater | 4326<br>3042<br>1324<br>135<br>51 | 0<br>7<br>14<br>21<br>28 | <br>29.68<br>69.39<br>96.88<br>98.82 | (Golzary et al., 2018) |
| sewage water | 199<br><br>128<br>54 | 0<br><br>15<br>30 | <br><br>35.68<br>72.86 | (Amuda and Ibrahim, 2006) |
| Sewage water+ cow dung | 117<br>71<br>48 | 0<br>15<br>30 | <br>39.31<br>58.97 | |
| Secondary treated sewage water | 676<br>85 | 0<br>NA | 87.42 | (Soman and Arora 2018) |

**Table 2:** ***Literature on heavy metal removal efficiency by Azolla***

| Heavy Metal | Biomass of Azolla/g | Initial concentration/ $mgL^{-1}$ | contact days | Removal efficiency % |
|---|---|---|---|---|
| Cd | 0.8 | 5 | 15 | 92.84 |
| Pb | 0.8 | 10 | 15 | 97.12 |
| Ni | 0.8 | 25 | 15 | 76.82 |

But there is a problem with discharging dead Azolla after treatments as heavy metal should not be released directly to the environment.

Pandey has studied the efficiency of *Azolla caroliniana* for phytoremediation of fly ash (FA) pond because of its higher bioconcentration factor. The metal concentrations ranged from 175 to 538 and 86 to 753 mg $kg^{-1}$ in roots and fronds. Bioconcentration factor (BCF) values of all metals in root and frond ranged from 1.7 to18.6 and 1.8 to 11.0, respectively, which were greater than one and indicated the metal accumulation potential of Azolla caroliniana. Translocation factor (TF) ranged from 0.37 to 1.4 for various heavy metals (Pandey, 2012), and figure 3 shows heavy metals concentration in roots, fronds, and FA effluents.

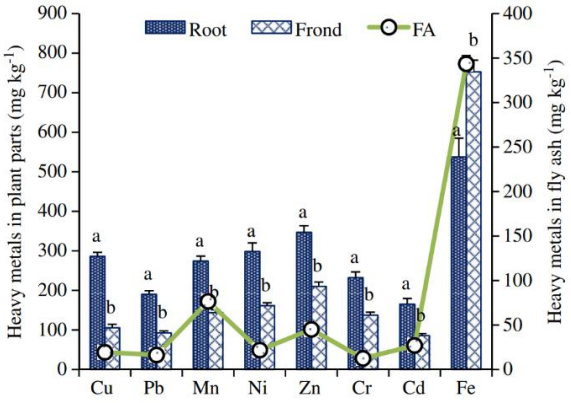


**Figure 3:** ***Heavy metals concentration in roots, fronds, and FA effluents (Pandey, 2012)***

## Nitrogen and phosphorous

The eutrophication of lakes and streams has been increased by nutrients, particularly nitrogen and phosphorus, released through wastewater treatment facilities. In eutrophic water, the growth of aquatic weeds and algae typically renders the water non-potable or unfit for recreational use. Overproduction of algae is frequently linked to oxygen deficiency and taste and odor issues in water. Because preventing

further eutrophication is costly, current pollution control research eliminates fertilizing ingredients from effluents before they enter natural rivers (Kitoh *et al.*, 1993).

Nitrogen and Phosphorous are the primary nutrients in wastewater and cause eutrophication. Azolla can absorb and accumulate nitrogen and phosphorous from wastewater. (Forni *et al.*, 2001; Amuda and Ibrahim, 2006; Golzary *et al.*, 2018; Soman and Arora, 2018) have studied the characteristics of Azolla. Figure 4 shows the different forms of nitrogen in wastewater that Azolla can remove.

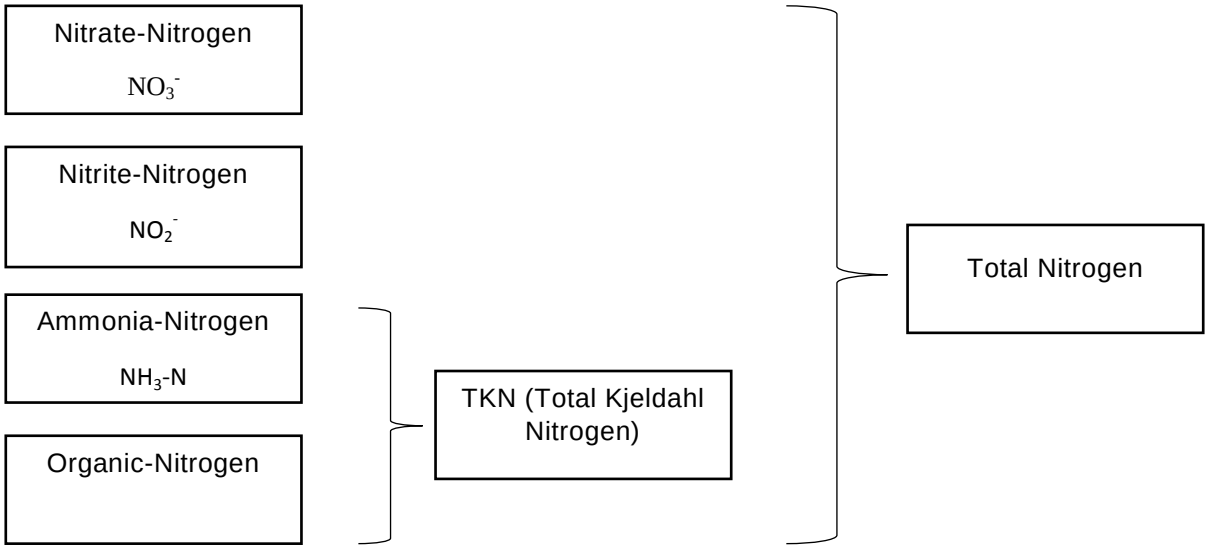


***Figure 4: Different types of nitrogen in wastewater***

Researchers have concluded the following essential points regarding Azolla. Azolla algae can remove nutrients (phosphorus and nitrogen) from aqueous solutions; therefore, it can be used for the advanced treatment of wastewater to improve the quality of effluent. However, it is unsuitable for nitrogen removal at low nitrogen content wastewaters. (Golzary et al., 2018). The ability of *Azolla* to carry out sewage bioremediation was analyzed. The study has shown promising results in removing total organic carbon, phosphorous, and nitrogen and efficiently removing ammonia (Soman and Arora, 2018). Azolla's performance after two weeks, particularly in the summer, revealed stress conditions caused primarily by high temperatures. As a result, it is advised that the fern be used to remove nitrogenous substances from aquaculture effluent for a short period in the spring and summer (Forni *et al.*, 2001).

The naturally growing aquatic fern Azolla algae are an excellent approach for wastewater treatment in the experimental tests. Its primary function is to reduce metabolic and chemical oxygen demand. This method can also reduce other nitrogen, phosphate, and potassium variables. Azolla alga may store contaminants in its tissues by consuming water for development. (Amuda and Ibrahim, 2006).

Muvea et al. (2019) have tabled the nitrogen and phosphorus fixing abilities of Azolla, as shown in Table 3.

**Table 4. Nutrients removal efficiency by the Azolla** (Muvea *et al.*, 2019)

| Nutrient | Removal efficiency (%) |
|---|---|
| Nitrite | 63.16 |
| Nitrate | 89.89 |
| Ammonia | 16.29 |
| Total phosphorous | 46.45 |
| Soluble reactive Phosphorous | 69.66 |
| Total nitrogen | 25.57 |
| Average | 68.67 |

## Benefits and Applications

Figure 5 represents the benefits and different applications of Azolla:

Experiments have demonstrated that Azolla can help plants overcome nutritional limitations by improving nutrient absorption. Azolla provides different benefits to plants and the environment depending on the plant species and growing practices and conditions, such as increased yields and crop quality, reduced disease occurrence, improved flowering and fruiting, increased plant establishment, and survival at seedling or transplanting, and so on. Produce plants that are more vigorous and healthier. Drought tolerance is improved, reducing watering (Raja *et al.,* 2012). Improve soil salinity tolerance and contribute to soil quality and nutrient cycling through optimizing fertilizer use, particularly nitrogen (Raja *et al*., 2012).

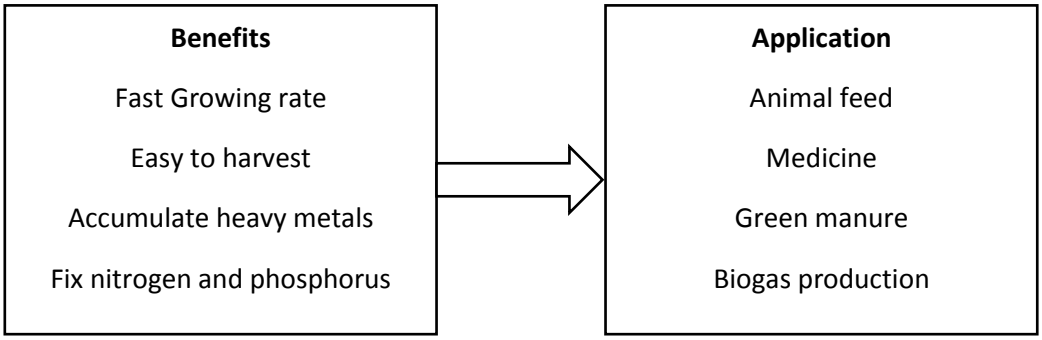


***Figure 5: Benefits and application of Azolla***

## Conclusion

Due to their cheap construction, operation, and maintenance costs compared to existing conventional systems, water treatment systems with aquatic plants are an efficient and cost-effective choice for wastewater treatment, eliminating microorganisms and physical-chemical pollutants. Removing suspended particles and organic matter in macrophyte-dominated systems is attributed to sedimentation, absorption, and removal mechanisms. Experimental results show that Azolla can be used differently for wastewater treatment, such as nutrient remover and heavy metal absorber. But most researchers haven't been forced into real situations and have studied only laboratory scale. Some practical problems will occur in real cases because Azolla may not grow well in deep tanks, such as treatment tanks used in wastewater treatment. Azolla needs a large surface area for its growth; Providing a large surface area may not be a practical situation in wastewater treatment. As a result, more research is needed to improve Azolla's efficiency and gain a more profound knowledge of this symbiosis.

## Conflict of interest

The author declares that the publishing of this article does not include any conflicts of interest. Furthermore, the author has strictly adhered to ethical problems such as plagiarism, informed consent, misconduct, data fabrication and falsification, multiple publishing and submission, and redundancy.

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## References

Costa, M. L., Santos, M. C. R., and Carrapiço, F. (2010). Potential uses of Azolla filiculoides' biomass grown in natural ecosystems and urban wastewater. In Proceedings of the 14th RAMIRAN International Conference.

Deshmukh, A. A., Bandela, N. N., Chavan, J. R., and Nalawade, P. M. (2013). Studies on potential use of water hyacinth, Pistia and Azolla for municipal waste water treatment. Environment, 3(11), 226-228.

Forni, C., Chen, J., Tancioni, L., and Caiola, M. G. (2001). Evaluation of the fern Azolla for growth, nitrogen and phosphorus removal from wastewater. *Water research*, *35*(6), 1592-1598.

Golzary, A., Tavakoli, O., Rezaei, Y., and Karbassi, A. (2018). Wastewater treatment by Azolla filiculoides: A study on color, odor, COD, nitrate, and phosphate removal. Pollution, 4(1), 69-76.

Kitoh, S., Shiomi, N., and Uheda, E. (1993). The growth and nitrogen fixation of Azolla filiculoides Lam. in polluted water. Aquatic botany, 46(2), 129-139.

Muvea, F. M., Ogendi, G. M., and Omondi, S. O. (2019). Nutrient removal efficiency by floating macrophytes; Lemna minor and Azolla pinnata in a constructed wetland. *Global Journal of Environmental Science and Management*, *5*(4), 415-430.

Naghipour, D., Ashrafi, S. D., Gholamzadeh, M., Taghavi, K., and Naimi-Joubani, M. (2018). Phytoremediation of heavy metals (Ni, Cd, Pb) by Azolla filiculoides from aqueous solution: a dataset. Data in brief, 21, 1409-1414. https://doi.org/10.1016/j.dib.2018.10.111

Pandey, V. C. (2012). Phytoremediation of heavy metals from fly ash pond by Azolla caroliniana. Ecotoxicology and Environmental Safety, 82, 8-12. https://doi.org/10.1016/j.ecoenv.2012.05.002

Amuda, O. S., and Ibrahim, A. O. (2006). Industrial wastewater treatment using natural material as adsorbent. African *journal of Biotechnology*, *5*(16).

Raja, W., Rathaur, P., John, S. A., and Ramteke, P. W. (2012). Azolla: An aquatic pteridophyte with great potential. Int. J. *Res. Biol. Sci*, *2*(2), 68-72.

Soman, D., Anitha, V., and Arora, A. (2018). Bioremediation of municipal sewage water with Azolla microphylla. International *Journal of Advanced Research*, *6*(5), 101-108.

Sood, A., Uniyal, P. L., Prasanna, R., and Ahluwalia, A. S. (2012). Phytoremediation potential of aquatic macrophyte, Azolla. Ambio, 41(2), 122-137.

Zazouli, M. A., Mahdavi, Y., Bazrafshan, E., and Balarak, D. (2014). Phytodegradation potential of bisphenolA from aqueous solution by Azolla Filiculoides. Journal of Environmental Health Science and Engineering, 12(1), 1-5.