International Journal of Botany Studies
www.botanyjournals.com
ISSN: 2455-541X
Received: 05-01-2022, Accepted: 20-01-2022, Published: 05-02-2022
Volume 7, Issue 2, 2022, Page No. 222-226

Azolla is a potential renevable bioresources in Indian agriculture

Thiyagarajan G[1], Sylvia Nithia Kumari[2], Rajasekaran S[3]
[1] Department of Botany, Government Arts College for Men, Kumbakonam, Tamil Nadu, India
[2] Department of Botany, A.V.C. College, Mannampandal, Tamil Nadu, India

Abstract
Azolla is a free floating fast growing freshwater pteridophytic fern plant which fixes atmospheric nitrogen by forming a symbiotic association with a prokaryotic cyanobacterium -*Anabaena Azollae*. *Azolla* biofertilizer has a tremendous ability to maintain suitable soil pH and fix organic C and N, improving mineralization, improving microbial activity, and status soil that can increase soil increase and ultimately enhance yield. Azolla is a cost effective, eco-friendly bio fertilizer in all over the world. As green manure in water logged soil, it enhances the rapid utilization of plant communities and mineralization of nitrogen, improves the physical and chemical properties of the soil and increases soil microbial activities. It increases the rice yield equivalent to that produced by 30-60 kg N/ha. Azolla application is considered as a good practice for sustaining soil fertility and crop productivity for our nation and small former easy to collect and distribute the plant field to achive more yield benefits.

Keywords: *Azolla*, biofertilizers, cyanobacteria etc

Introduction
Azolla is a floating aquatic fern and is commonly called as mosquito fern, duckweed fern, fairy moss and water fern. *Azolla* species are distributed throughout the world in temperate and tropical fresh waters. Azolla floats on the surface of water with the help of numerous small, closely overlapping scale-like leaves, with their roots hanging through the water and forms large mats. The habitats include ponds, ditches, canals and paddy fields. (Rev *et al* 1989) [19] By nature, they form a symbiotic relationship with the cyanobacterium *Anabaena Azollae*, which fixes atmospheric nitrogen, giving the plant access to the essential nutrient that helps to colonize areas of freshwater and grow at great speed. *Azolla* grow best at temperature range of 25-30°C, and water temperature for multiplication of Azolla is between 18-26°C. The relative growth rate of Azolla is high as it generally doubles its weight in 2-4 days period resulting in very high biomass in a short period of time. The ideal characteristics that make *Azolla* significant to agriculture and allied activities are its rapid growth rate and possession of various nutrients particularly protein.

Table 1

***Azolla* distribution in India**		
S. No	**Species**	**Distribution**
1.	*Azolla caroliniana*	1Canton, Hong Kong
2.	*Azolla filiculoides*	China, and Japan
3.	*Azolla pinnata*	, Japan, Korea, Malaysia, Nepal, New Caledonia, New Guinea, Pakistan, Philippines, Sri Lanka, Taiwan, Thailand, Vietnam Bangladesh, Burma, China, India, Indonesia

Source: (Lumpkin & Plucknett, 1980) [13]

Morphologyand life cycle of *Azolla*
Azolla fronds consist of leaf, rhizome (stem) and root. Leaves occur in two rows along the side of rhizome and each leaf has a thin ventral lobe and a thick dorsal lobe. The dorsal lobe contains chlorophyll and carotenoid pigments that carryout photosynthesis. Within the dorsal lobe the cyanobacterium, *Anabaena Azollae* is present in a specialized ovoid cavity (Lumpkin, 1985). The roots are adventitious and chlorophyllous in their early stage. *Azolla* fronds are triangular orpolygonal and float on water surface. The morphological characters are helpful to differentiate the six major Azolla species are *Azolla cristata* (*A. caroliniana*)-The fronds are very short and are about 5-10mm long, grow less than 1 inch tall, green to reddish in colour with tiny protuberances called trichomes that give them a velvety appearance.
The trichromes are septate; star shaped branched floating stem with highly imbricated leaves. The dorsal leaf lobe is round, acute angled and the ventral lobe has pink tint. Thin root hair is present throughout the root. It is a water fern having 2 surfaces of dorsal lobe and ventral lobe. Ventral side is brown in color, thin and dorsal side

is green in color (Photosynthesis). *Anabaena Azollae* present in the central cavity of Azolla which has a symbiotic relationship with it. There are around 80,000 symbiotic cyan bacteria present on its leaves. This unique property has drawn the attention of agriculturists for its utilization in agriculture (Kamalasamana *et al.*, 2002) [10].

In view of an agronomist, it is used as bio fertilizers which increases organic matter, improves soil and supply fixed nitrogen, increases the uptake nutrient element such as Ca, Mg & K. It also helps to build up the micro-flora and in turn, the soil health. *Azolla–anabaena* relationship can fix almost three times more atmospheric nitrogen than legumes. Typical rates for legumes are 400 kg of nitrogen per hectare per year those for Azolla-anabaena are 1100 kg of nitrogen per hectare per year. *Azolla filiculoides* grows up to 2.5 cm and is a large floating fern. Star shaped branched floating stem with highly imbricated leaves. Dorsal and ventral leaf lobes grow on upper side of the rhizome. The dorsal leaf lobe is slightly acuminated and acute angled. The ventral leaf lobe is pink tinted.

The trichomesare unicellular thereby could be distinguished from *Azolla cristata. Azolla mexicana* differs from *Azolla cristata* and *Azolla filiculoides* by having multicellular trichomes. Dorsal and ventral leaf lobes grow on upper side of the rhizome. Star shaped branched floating stem with highly imbricated leaves. The dorsal leaf lobe is slightly acuminated and acute angled. The ventral leaf lobe is pink tinted. *Azolla microphylla*—Star shaped branched floating stem with highly imbricated leaves. The dorsal leaf lobe is highly acuminated, acute angled and the ventral lobe is with pink tint. Thin root hair is present throughout the root. *Azolla pinnata*—Triangular shaped branched floating stem with slightly imbricated leaves. The dorsal leaf lobe is highly acuminated, acute angled with translucent ventral leaf lobe. Thick root hair is present throughout the root. *Azolla rubra*—Dorsal and ventral leaf lobes grow upper side of the rhizome. Star shaped branched floating stem with highly imbricated leaves. The dorsal leaf lobe is highly acuminated and acute angled. The ventral leaf lobe is pink tinted. Azolla has sporophytic and gametophytic phase of life cycle.

In sporophyte phase when the fronds reach a certain size, depending upon the species (1-2cm) the older secondary stems detach themselves as a result of formation of abscission layer and give rise to new frond. This is the common mode of reproduction in Azolla. Gametophytic reproduction (sexual) is essential for survival under adverse condition. In gametophytic phase, micro (male) and mega (female) sporocarps are produced on the same plant. The sporocarps are formed at the lateral end of the ventral lobe. The micro and mega sporocarps after maturation get detached from the mother sporophyte and sink to the bottom of the water column. The mega sporocarp germinateinto a female gametophyte containing one or more archegonia containing one egg cell called the oocyte. The antherozoids move through the massulae and fertilize the oospores. The zygote is formed within the megaspore apparatus under the water body. The germination of sporocarp is triggered by light and temperature. As the embryo grows, the new Azolla plantlet is released.

Ecological importance of *Azolla*

Azolla is a highly productive plant. It doubles its biomass in 1.9 days or more, depending on conditions, and yield can reach 8–10 tonnes fresh matter/ha in Asian rice fields. 37.8 t freshwww.justagriculture.in Page 2 weight/ha (2.78 t DM/ha dry weight) has been reported for Azolla pinnata in India. The daily nitrogen-fixing rate of the Azolla-algae (anabaena Azollae) complex is 3-7 kg N/ha. Azolla contains 4% nitrogen on a dry-weight basis (dry weight is 5% of fresh weight); 0.5-0.9% phosphorous; and 2-4.5% potassium. There are six species of Azolla *viz*., *Azolla Carolina, Azolla nilotica, Azolla filiculoids, Azolla mexicana, Azolla microphylla and Azolla pinnata*. The *Azolla pinnata* is a common species in India.

Table 2

S. No	Factors	Range
1.	Temperature	20°C - 25°C
2.	Light	50%
3.	Humidity	85 - 90%
4.	Water	5 - 12 cm
5.	pH	4-7
6.	Salinity	90-150 mg/Litre

***Azolla* for improve soil fertility**

The nitrogen-fixing capability of Azolla due to the presence of the cyanobiont *Anabaena Azollae* makes Azolla a popular bio-fertilizer, especially in parts of Southeast Asia. Azolla fixes 1100 kg nitrogen per hectare per year and Azolla is considered as a bio source of nitrogen. It can be easily multiplied in wet lands, cement tanks or pits lined with silpaulin sheet. The multiplied biomass can be collected and applied to soil as green manure. For paddy, it can be used as bio-fertilizer by a technique called dual culturing. In dual culturing Azolla @500kg per hectare is broadcasted in the standing water after seven days of transplanting the paddy seedlings. Azolla grows and would cover the surface. This prevents the growth of weeds in paddy fields4 (Bhuvaneshwari and Kumar. 2013) [5]. With the help of the cyanobiont, it converts the atmospheric nitrogen to ammonia that is released to the standing water. The released ammonia is utilized as a source of nitrogen by paddy (Ray *et al.*, 2008) [18]. When

the entire paddy field gets covered with Azolla, decomposition takes place and Azolla is added as organic manure to soil that adds upto nine tonnes of protein per hectare per year5 (Rev *et al*, 1989) [19].

Azolla contains 2-5% nitrogen, 1-2% phosphorus, 0.5- 6% potassium on dry weight basis (Ana 2018) [8]. Also, contains calcium, magnesium, silica, zinc, copper, iron, sulphur, sodium and plant growth promoting substances like auxins and gibberellins (Kannaiyan and Rajeswari 1983, and Kannaiyan and subramani 1992) [11, 12]. Upon decomposition these nutrients are released in to soil and help to improve soil physio-chemical and biological properties particularly the organic carbon content of soil.

Benefits of incorporating *Azolla* in rice cultivation

The thick Azolla mat in rice fields suppresses weeds. Since Azolla floats at the water surface, it does compete with rice for light and space. In most climates, Azolla grows best under a partial shade of vegetation which is provided by the rice canopy during early and intermediate stages of growth. When the rice approaches maturity, Azolla begins to die and decompose due to low light intensities under the canopy and a depletion of nutrients, thus releasing its nutrients into the water. Because Azolla decomposes rapidly, its nitrogen, phosphorus and other nutrients are rapidly released into the water and made available for uptake by rice during grain development. Azolla has a greater ability than rice to accumulate potassium in its tissues in low-potassium environments, providing rice with potassium after Azolla‟s decomposition. When grown in a rice field, Azolla reduces the ammonia volatilization that occurs following the application of inorganic nitrogen fertilizers by 20% to 50% (Roy *et al* 2016) [20]. This is due to the fact that the Azolla cover reduces light penetration into the floodwater, thus hindering the rise of pH which normally stimulates ammonia volatilization in an Azolla free rice field.

Various useful facts of *Azolla*

a. Larvicide - Mosquito control: Azolla can be used to control mosquitoes because a thick Azolla mat on the water surface helps to prevent breeding and adult emergence. In a survey of pools, ponds, wells, rice fields, and drains, Ansari and Sharma (1991) found that breeding by Anopheles was almost completely suppressed in water bodies that were covered with Azolla. Azolla extracts also has Larvicide effect over larvae of Aedes aegypti mosquito vectors.
b. Weed Control-The Azolla cover reduced light intensity by about 90%, reducing photosynthesis in the floodwater and thus reducing oxygen concentration of the water by more than 50%. Besides reducing light intensity, an Azolla cover alters light quality, the green leaves having a filter effect that increases the relative amount of infrared rays which reduces the germination of light-sensitive seeds.
c. Reduction of methane emission-Radha Prasana and a group of colleagues at various institutes in India have shown that methane production from rice growth is inhibited by the presence of Azolla. This is relevant to reducing greenhouse gases (Prasana *et al.*, 2004 and 2008). They investigated the role of cyan bacteria and Azolla in methane production and oxidation in laboratory simulation experiments using soil samples from rice fields (Raja *et al* 2012). All seven cyan bacterial strains that were tested and Azolla produced a significant decrease in the headspace concentration of methane in flooded soil, incubated under light.
d. Removal of diesel contaminants & heavy metal-Azolla has the potential to biodegrade and remove diesel contaminants from soil and water. the diesel degradation was due to the release of bacteria (bio augmentation) and physiochemical improvement of the soil and water (bio stimulation). It can also used to purify water polluted by metals and has potential applications for both industrial and mining operations, as well as space exploration.
e. Livestock feed-Azolla has enormous potential as a livestock feed due to its high content in proteins, essential amino acids, vitamins (A, B12, and Beta Carotene), growth promoter intermediaries and minerals. It has been used for many years throughout Asia and parts of Africa to feed pigs, ducks, chickens, cattle, fish, sheep and goats and rabbits.

Table 3: Nutrient composition of Azolla

S No	Constituents	Dry matter (%)
1.	Ash	10 2
2.	Calcium	0.4-1.0 3
3.	Chlorophyll	0.34-0.55
4.	Crude fat	3.3-3.6
5.	Crude protein	14.0-30.0
6.	Iron	0.06-0.26
7.	Magnesium	0.5-0.65
8.	Nitrogen	4.0-5.0
9.	Phosphorus	0.5-0.9
10.	Potassium	2.0-4.5
11.	Soluble sugars	3.4-3.5
12.	Starch	6.5

Source:(Salma, 2020)

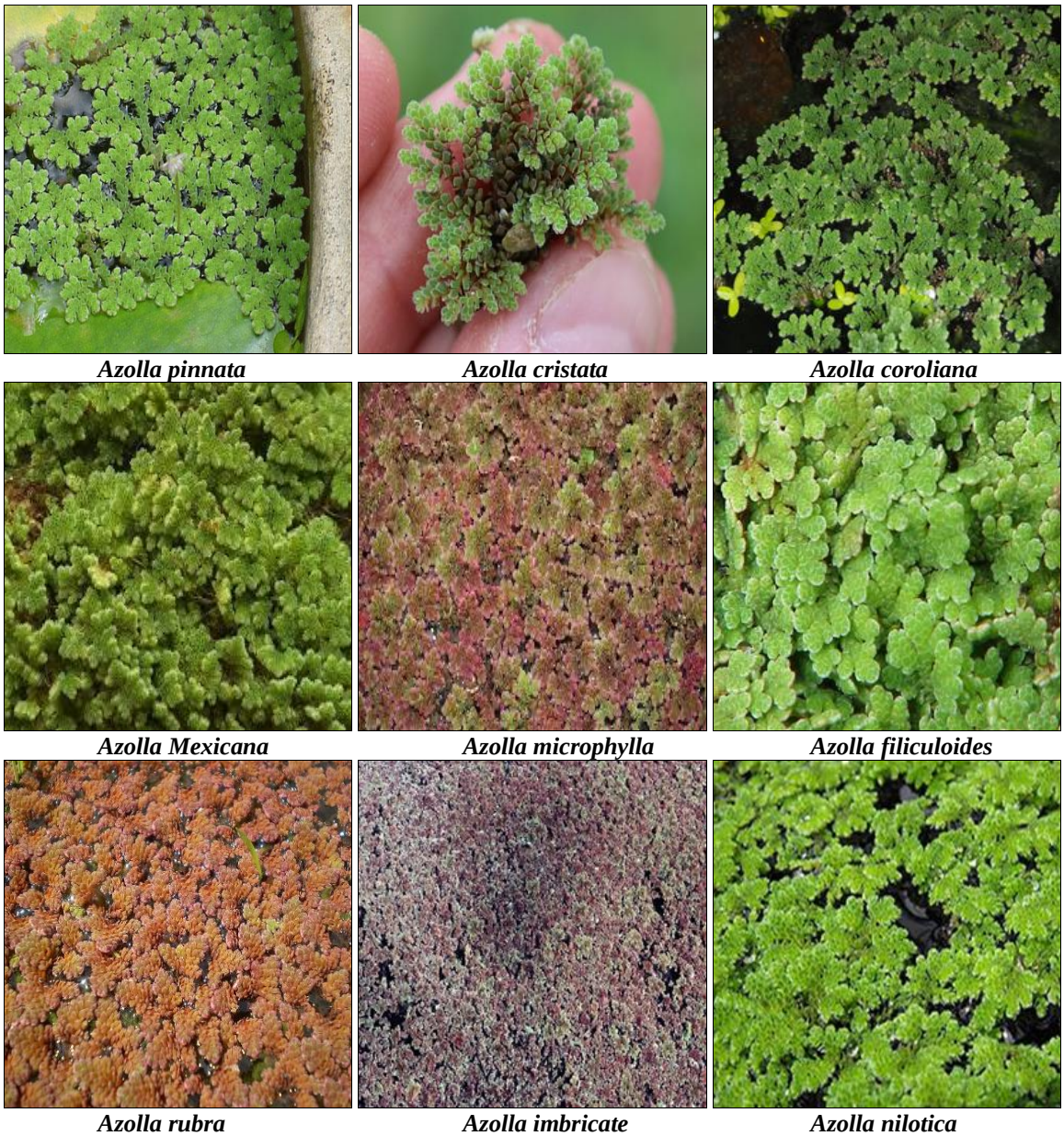
Specially for weed suppression in *Azolla*

Weed alone can reduce the Rice yield ranging from 15 − 20% and up to 50% in severe cases (Sureshkumar *et al* ., 2016). A thick Azolla mat in a rice field has the side benefit of suppressing weeds. Azolla covering water surface reduces light penetration of soil surface, resulting in the depreciation in the germination of weeds (70% of the weed). Thus, the growth of Azolla reduces aquatic weeds in flooded rice fields like Echinochloa crus-Galli, Cyperus sp., Paspalum sp. and so on and, therefore, lead to improved crop growth and productivity (Biswas *et al.*, 2005; Monajjem and Hajipour, 2010) [6, 14]. The degree of suppression increases with an increase in the percent of Azolla cover and water depth (Amanulla *et al.*,2015 and 2016) [1, 2]. Application of preassumed at 10 t ha-1 + Azolla at 1 t ha-1 recorded the least weed count and highest weed control index in rice crop, as the thallus growth formed a very thick mat on the surface of the water, curtailing the interception of light by weed seeds and seedlings (Gnanavel, 2015; Gnanavel and Kathiresan, 2002) [7, 8]. A study reported that weeds were suppressed by 69 – 100% at rice flowering and 86 – 95 % at harvest depending upon weed species due to the use of the Azolla (Janiya and Moody, 1984) [9].

***Azolla* species**

Scientific classification

Class: Polypodiopsida
Order: Salviniales
Family: Salviniaceae
Genus: Azolla



Azolla pinnata ***Azolla cristata*** ***Azolla coroliana***

Azolla Mexicana ***Azolla microphylla*** ***Azolla filiculoides***

Azolla rubra ***Azolla imbricate*** ***Azolla nilotica***

Fig 1

Conclusion

Nowadays, there is a major concern to meet the increasing demands of rice without degrading the environment and soil health in the long term. So, in recent times, most Asian countries governments have formulated policies advocating on use of biofertilizers. Considering these aspects, use of Azolla as a biofertilizers can be viable option for the rice producer as it increases rice productivity and also improves soil health sustainably.

Azolla has the potential to suppress weed, increasing the availability of N, P, K, and other mineral nutrients, which all contribute to increasing the yield. Considering the agronomic benefits and reducing the urea (N-fertilizer) demand in the rice cropping system, Azolla could develop low-input cropping systems for rice production. Azolla, the economics of using Azolla should be considered because technology is very labour-intensive.

References

1. Amanullah Khan I, Jan A, Jan MT, Khalil SK, Shah Z, Afzal M. Compost and Nitrogen Management Influence Productivity of Spring Maize (Zea mays L.) under Deep and Conventional Tillage Systems in Semi-arid Regions. Communications in Soil Science and Plant Analysis,2015:46(12):1566–1578. https://doi.org/10.1080/00103624.2015.1043462
2. Amanullah Khan SUT, Iqbal A, Fahad S. Growth and productivity response of hybrid rice to application of animal manures, plant residues and phosphorus. Frontiers in Plant Science,2016:7(16):1-10. https://doi.org/10.3389/fpls.2016.01440
3. Ana LP, The Unique Symbiotic System between a Fern and a Cyanobacterium, Azolla-Anabaena Azollae: Their Potential as Biofertilizer, Feed, and Remediation. In Everlon Cid Rigobelo Ed. Symbiosis, 2018. DOI: 10.5772/ intechopen.70466.
4. Ansari MA, Sharma VP. Role of Azolla in controlling mosquito breeding in Ghaziabad District villages (U.P.). Indian J. Malariol.,1991:28:51-54.
5. Bhuvaneshwari K, Kumar A. Agronomic potential of the association Azolla – Anabaena. Science Research Reporter,2013:3(1):78-82.
6. Biswas M, Parveen S, Shimozawa H, Nakagoshi N. Effects of Azolla species on weed emergence in a rice paddy ecosystem. Weed Biology and Management,2005:5(4):176-183.
7. Gnanavel I. Eco-friendly weed control options for sustainable agriculture. Science International (Dubai),2015:3(2):37-47
8. Gnanavel I, Kathiresan RM. Sustainable weed management in ricerice cropping system. Indian Journal of Weed Science, 34 (3and4),2002:192-196.
9. Janiya JD, Moody K. Use of Azolla to suppress weeds in transplanted rice. International Journal of Pest Management,1984:30(1):1-6.
10. Kamalasanana Pillai, Premalatha S, Rajamony S. Azolla – A sustainable feed substitute for livestock. Leisa India, 2002, 15-17.
11. Kannaiyan S, Rejeswari N. Comparative effect of fertilizer nitrogen and Azolla biofertilizer on tiller production of rice. Science and Culture, 1983.
12. Kannaiyan S, Subramani S. Use of Azolla as biofertilizer for rice crop, Cyanobacterial Nitrogen Fixation. Indian Agricultural Research Institute, New Delhi, 1992, 281-289.
13. Lumpkin, Thomas A, Plucknett DL. Azolla: botany, physiology, and use as a green manure. Economic Botany,1980:34(2):111–153.
14. Monajjem S, Hajipour A. The role of Azolla in improving of rice fields stability. In Proceedings of 5th National Conference on Sustainable Agriculture and Healthy Products. Isfahan Research Center for Agriculture and Natural Resources, 2010, 303-307.
15. Prasanna P, Gunaratne LHP, Withana W. Economic analysis of paddy threshing methods. Sri Lankan Journal of Agricultural Economics,2004:6(1381-2016–115718):51–66.
16. Prasanna R, Jaiswal P, Singh Y, Singh P. Influence of biofertilizers and organic amendments on nitrogenase activity and phototrophic biomass of soil under wheat. Acta Agronomica Hungarica,2008:56(2):14-159.
17. Raja W, Rathaur P, John SA, Ramteke PW. Azolla-Anabaena Association and Its Significance In Supportable Agriculture. Hacettepe Journal of Biology and Chemistry,2012:40(1):1-6.
18. Ray TB, Peters GA, Toia RE, Mayne BC. Plant Physiol.2008:62:463-467.
19. Rev A, Physiol P, Mol P, Downloaded B, Peters GA. The AzollaAnabaena Symbiosis, 1989, 193-210.
20. Roy DC, Pakhira MC, Bera S. A review on biology, cultivation and utilization of Azolla. Adv Life Sci, 2016:5(1):11-15.
21. Salma SU. I Med Pub Journals Azolla: A Boon or Bane Abstract Azolla as a Fish Nourish Limitation of nitrogen volatilization, 2020, 1–7. https://doi.org/10.36648/2581-804X.4.4.35
22. Sureshkumar R, Reddy YA, Ravichandran S. Effect of weeds and their management in transplanted rice–a review. International Journal of Research in Applied, Natural and Social Sciences,2016:4(11):165–180.