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REVIEW PAPER

A Review on Biology, Cultivation and Utilization of Azolla

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ABSTRACT

Azolla commonly known as mosquito fern, duckweed fern, fairy moss, and water fern, is a small free floating aquatic fern native to Asia, Africa, and the America. It grows in swamps, ditches, and even in lakes and rivers where the water is not turbulent (Lumpkin and Plucknett, 1980). The name *Azolla* is derived from the two Greek words, *Azo* (to dry) and *Ollyo* (to kill) thus reflecting that the fern is killed by drought. *Azolla-Anabaena* is a symbiotic complex in which the endophytic blue-green algae *Anabaena zollae* lives within the leaf cavities of the water fern *Azolla* (Lain). The endosymbiont, which is nitrogen-fixing, provides sufficient nitrogen for both itself and its host (Peters, 1978). The fern, on the other hand, provides a protected environment for the algae and also supplies it with a fixed carbon source. It has capability to fix atmospheric nitrogen as well as to produce biomass at a very high rate. A lot of research works was done on azolla during the past and recent decades. The objective of this paper is to review and synthesize those research results with the hope that it will be helpful for collaborative research on this 'green gold mine' in future. *Azolla* is commonly used as biofertilizer as well as green manure in the paddy field. Now a days *Azolla* (either fresh or in dried) is also used as a feed ingredient for ruminants and non-ruminants type of livestock. Besides its utilization as biofertilizer and livestock feed, azolla, the 'green gold mine' of the nature is also used as medicine, water purifier, human food and for production of biogas.

Keyword *Azolla*, biofertilizer, crude protein, amino acids, livestock feed

The term *Azolla* was first coined by Lamarck in 1783. The genus *Azolla* belongs to division Pteridophyta, class Polypodiopsida and order Salviniaceae. It also belongs to the family Salviniaceae and consisted of two subgenera and six living species (Lumpkin & Plucknett, 1980). The subgenus *EuAzolla*, characterized by three megaspore floats and septate glochidia, included

four species: *A. filiculoides* Lain., *A. caroliniana* WiUd., *A. microphylla* Kaulf., and *A. mexicana* Presl. The subgenus *Rhizosperma*, characterized by nine megaspore floats, included two species: *A. pinnata* R. Br., with simple glochidia, and *A. nilotica* Decne., with no glochidia.

Distribution

Azolla generally grows in freshwater in tropical, subtropical, and warm-temperate regions throughout the world. *Azolla caroliniana* is found in eastern North America and the Caribbean, *A. filiculoides* in the Americas from southern South America through western North America to Alaska, *A. microphylla* in tropical and subtropical America, *A. mexicana* in the Americas from northern South America through western North America, *A. nilotica* in East Africa from Sudan to Mozambique, and *A. pinnata* in most of Asia and the coast of tropical Africa (Sculthorpe, 1967; Lumpkin & Plucknett, 1980; Watanabe, 1982; Van Hove, 1989; Wagner 1997). Three *Azolla* sp. i.e. *A. caroliniana*, *A. microphylla*, and *A. pinnata* are commonly found all over the Indian subcontinent.

Morphology

The macrophyte of *Azolla* is called frond which ranges from 1 cm to 2.5 cm in length in species such as *A. pinnata* and 15 cm or more in the largest species like *A. nilotica* (Raja *et al.* 2012). It has a main rhizome which branches into secondary rhizomes, all of which bear small leaves alternately arranged. Numerous unbranched, adventitious roots hang down into the water from nodes on the ventral surfaces of the rhizomes. The roots absorb nutrients directly from the water and in shallow water they may touch the soil, deriving nutrients from it. Each leaf consists of two lobes: an aerial dorsal lobe, which is chlorophyllous, and a partially submerged ventral lobe, which is colourless and cup-shaped and provides buoyancy. Each dorsal lobe contains a leaf cavity which

*A. caroliniana**A. microphylla**A. pinnata*

houses the symbiotic *Anabaena Azollae* (Peters, 1977; Lumpkin & Plucknett, 1980).

The interior surface of each leaf cavity is lined with an envelope (Peters, 1976) and covered by a mucilaginous layer of unknown composition which is embedded with filaments of *A. Azollae* and permeated by multicellular transfer hairs

***Azolla* as Biofertilizer in Rice Cultivation**

Azolla is cultivated in the paddy field either as monocrop or as intercrop and incorporated into the mud/soil for increasing humus and nutrient content of the soil. This practice of *Azolla* cultivation is widely popular in the countries of south-east Asia like India, China, Phillipines, Indonesia etc. Peters (1978) reported that the use of *Azolla* increased rice yields by 112% over unfertilized controls when applied as a monocrop during the fallow season, by 23% when applied as an intercrop with rice, and by 216% when applied both as a monocrop and an intercrop. When *Azolla* used as a bio-fertilizer in paddy field it produces around 300 tons of green bio-hectare per year under normal subtropical climate which is comparable to 800 kg (1800 kgs of urea) of nitrogen (Wagner 1997). *Azolla* has quick decomposition rate in soil and thus it speeds up the efficient availability of its nitrogen to rice plant. The quick multiplication rate and rapid decomposing capacity of *Azolla* has become paramount important factor to use as green manure cum bio-fertilizer in rice field. The benefits of *Azolla* application in the rice field are the following: Basal application @ 10-12 tones/hectare increases soil nitrogen by 50-60 kg/ha and reduces 30-35 kg of nitrogenous fertilizer requirement of rice crop. Release of green *Azolla* twice as dual cropping in rice crop @ 500 kg/ha enriches soil nitrogen by 50 kg/ha and reduces nitrogen

requirement by 20-30 kg/ha. Use of *Azolla* increases rice yield by 20 to 30% (Raja *et al.*, 2012).

***Azolla* as Nutritional Supplement for Livestock**

Azolla is used as food supplement for variety of animals like cattle, goat, pigs, rabbits, chickens, ducks and fish. Seultrope, 1967 conducted an experiment and reported that *Azolla* can be utilized as fodder for cattle and pigs. It was also found that broilers feed with *Azolla* resulted in growth and body weight values similar to those resulting from the use of maize-soya bean meal. Das *et al.*, 1994 found that digested *Azolla* slurry remaining after biogas production was suitable as fish pond fertilizer.

Murthy, *et al.*, 2013 fed 2 kg fresh *Azolla* per day to the milking cows replacing 50% of concentrate for 3 months and reported that *Azolla* maintained good dairy performance while decreasing feed+labour costs by 16.5% and milk production costs by 18.5%. Incase of 'Black Bengal' goat, replacement of the concentrate up to 20% with sun-dried *Azolla* resulted better growth with no adverse effect on its health (Tamang, *et al.*, 1993).

Parthasarathy, *et al.*, 2002 reported that 5 % replacement broiler ration with dried *Azolla* was quite profitable and safe for broiler production. Ali, *et al.*, 1995 conducted an experiment with feeding broiler chicken with maize and soybean meal 10% replaced by dried *A. pinnata* and reported that feed cost significantly decreased without affecting the meat production resulting higher net return. Rai *et al.*, 2012 conducted an experiment and reported that layer birds fed with fresh *Azolla* had a higher body weight at 8 weeks or higher egg production at 40 and 72 days than control.

Nutritional qualities of *Azolla****Azolla* (*Azolla* spp.), as fresh**

1. Main analysis	Unit	Avg	Min	Max
Dry matter	% as fed	6.7	5.1	8.7
Crude protein	% DM	20.6	13.9	28.1
Crude fibre	% DM	15.0	11.3	22.8
NDF	% DM	43.8	35.4	52.3
ADF	% DM	31.8	24.0	38.9
Lignin	% DM	11.4	9.3	13.5
Ether extract	% DM	3.8	1.9	5.1
Ash	% DM	15.9	9.8	21.6
Starch (polarimetry)	% DM	4.1	2.7	5.5
Gross energy	MJ/kg DM	17.0		
2. Minerals	Unit	Avg	Min	Max
Calcium	g/kg DM	11.0	5.8	17.0
Phosphorus	g/kg DM	6.1	0.3	15.5
Potassium	g/kg DM	17.4	10.9	22.5
Sodium	g/kg DM	9.0	2.8	12.5
Magnesium	g/kg DM	5.0	3.9	6.1
Manganese	mg/kg DM	762	208	1429
Zinc	mg/kg DM	38	11	77
Copper	mg/kg DM	16	10	28
Iron	mg/kg DM	3900	711	8200
3. Amino acids	Unit	Avg	Min	Max
Alanine	% protein	6.4	5.3	7.4
Arginine	% protein	5.9	5.1	6.6
Aspartic acid	% protein	9.3	8.2	10.3
Cystine	% protein	1.6	0.7	2.3
Glutamic acid	% protein	12.6	11.6	13.5
Glycine	% protein	5.6	4.5	6.6
Histidine	% protein	2.1	1.6	2.4
Isoleucine	% protein	4.5	3.7	5.4
Leucine	% protein	8.4	7.0	9.2
Lysine	% protein	4.7	3.5	6.5
Methionine	% protein	1.4	1.2	1.9
Phenylalanine	% protein	5.4	5.2	5.6
Proline	% protein	4.9	3.5	6.8
Serine	% protein	4.5	3.9	5.6
Threonine	% protein	4.7	4.0	5.3
Tryptophan	% protein	1.8	1.5	2.0
Tyrosine	% protein	3.6	3.2	4.1
Valine	% protein	5.5	3.8	6.8

Source: FAO, 2015

***Azolla* in Reclamation of Saline Soils**

Although, *Azolla* is relatively sensitive to salt, cultivation in saline environment for a period of two consecutive years decreased salt content from 0.35-0.15 and desalinate rate (71.4%) was 1.8 times faster than through water leaching and 2.1 times faster than *Sesbania* and also reduced the electrical conductivity, pH of acidic soil and increased calcium content of soil (Anjuli *et al.*, 2004).

***Azolla* in Bioremediation**

It was found that *A. pinnata* and *Lamna minor* removed the heavy metals iron and copper from polluted water (Jain *et al.* 1989). The pollutants at low concentration could be treated by passing it through ponds and can be reused for Agriculture purpose. Recently Arora *et al.* (2006) found that tolerance and phyto-accumulation of chromium by three *Azolla* species and also results found by Cohen-Shoel *et al.* 2002 shows bio-filtration of toxic elements by *Azolla* biomass. *Azolla* exhibits a remarkable ability to concentrate metals Cu, Cd, Cr, Ni, Pb and nutrients directly from pollutants or sewage water.

***Azolla* as Mosquito Repellent**

Azolla can also be used in the control of mosquitoes, for a thick *Azolla* mat on the water surface can 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 spp.* was almost completely suppressed in water bodies that were completely covered with *Azolla*.

***Azolla* in Production of Biogas**

Anaerobic fermentation of *Azolla* (or a mixture of *Azolla* and rice straw) results in the production of methane gas which can be utilized as fuel and remaining effluent can be used as a fertilizer because it contains all the nutrients originally incorporated in plant tissues except for a small percentage of nitrogen lost as ammonia as suggested by Van Hove 1989. Das *et al.* 1994 mixed cow dung and *Azolla* residues and found that best ratio was 1:0.4, which gave a gas production 1.4 times that of cow dung alone.

***Azolla* and Bioenergy**

A non-polluting, high energy fuel when *Azolla-Anabaena* is grown in a nitrogen- free atmosphere and or a water medium containing nitrate in the symbionts, instead of fixing nitrogen evolves

hydrogen, using water as the source [Peters 1975]. Hall *et al.*, 1995 shows that rate of hydrogen production can be increased by exposure to a micro aerobic environment, a partial vacuum or argon-enriched or carbon dioxide enriched atmosphere or by immobilization of cells of *Anabaena-Azollae* isolated from the fern.

Medicinal use of *Azolla*

In Tanzania, *Azolla* is used for preparing cough medicine (Raja *et al.*, 2012)

***Azolla* as a human food**

Azolla is widely used as a livestock feed in India and the Far East, but its potential as a food for people is less well known. This is now changing and some of the impetus has come from studies into diets that could be used for space stations, space travel, and habitation on the Moon and Mars. *Azolla's* protein content is close to that of soybean. It is a rich source of minerals (10-15% of dry weight), essential amino acids (7-10% of dry weight), vitamins and carotenoids. 20 – 30% of *Azolla's* dry weight is protein, which is a lot for a vegetable. The quality of the protein in *Azolla* is good.

***Azolla* as a Component of Space Diet**

Recent research by Katayama *et al.*, 2008 in collaboration with Space Agriculture Task Force suggested *Azolla* as a component of the space diet during habitation on Mars and found that *Azolla* was found to meet human nutritional requirements on Mars.

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