

## EFFECT OF ALGAE AND AQUATIC MACROPHYTES ON NITROGEN DYNAMICS IN WETLAND RICE FIELDS\*

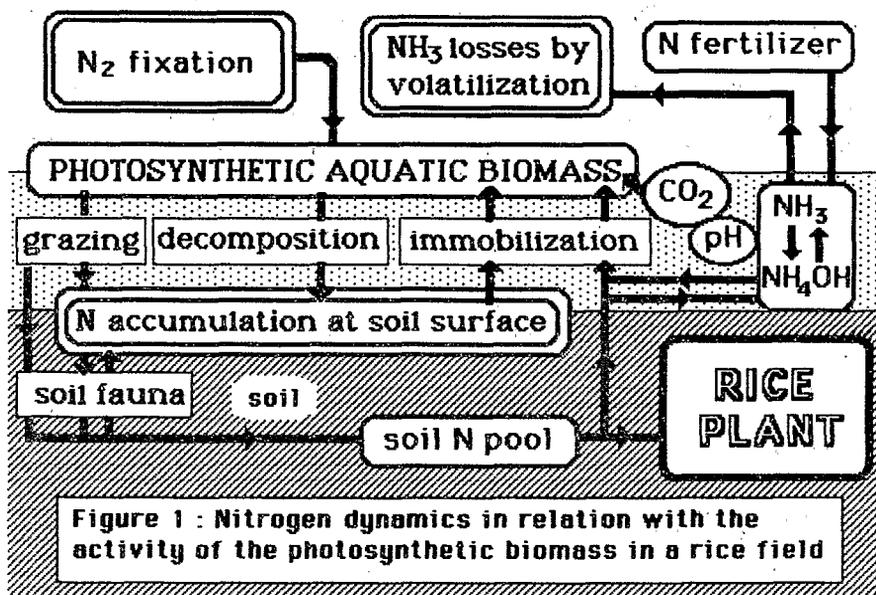
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Algae and aquatic macrophytes that develop in the floodwater of wetland rice fields are primary producers contributing significantly to the fertility of the ecosystem. This paper summarizes their major characteristics and activities regarding the nitrogen cycle : biological N<sub>2</sub> fixation (BNF) by free living blue-green algae (BGA) and *Azolla*, N immobilization, N recycling by grazing, N accumulation at the soil surface, N supply to the rice crop, and N losses by NH<sub>3</sub> volatilization (in relation to pH increase due to photosynthetic activity by the aquatic biomass) (Figure 1 ). Emphasis has been given to applied aspects.



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### 1) MAJOR CHARACTERISTICS

The photosynthetic aquatic biomass in rice fields is composed of planktonic, filamentous and macrophytic algae, and vascular macrophytes. Their development depends on the availability of nutrients and light ; largest biomasses are recorded in fallow plots and in fertilized fields when the rice canopy has not become too dense. Biomass value is usually a few hundred kg d.w./ha and rarely exceeds 1ton d.w./ha (Table 1). Planktonic algae generally have lower productivity than macrophytes (Roger and Watanabe, 1984).

**Table 1 : Biomasses of algae and aquatic macrophytes in rice fields**

Nature	Fresh weight (kg/ha)	Dry Weight (kg/ha)	Location	Reference
BGA	7500	375 <sup>a</sup>	China	Acad. Sinica ... <sup>c</sup>
Green algae	60/6000 <sup>a</sup>	3/300	India	Mahapatra <i>et al.</i> 1971 <sup>c</sup>
BGA	800 <sup>a</sup>	32	India	Mahapatra <i>et al.</i> 1971 <sup>c</sup>
Algal biomass	16000	640 <sup>a</sup>	UzbSSR	Muzafarov, 1953 <sup>c</sup>
Algal biomass	2/6000	0/240 <sup>a</sup>	Sénégal	Reynaud & Roger 1978 <sup>c</sup>
BGA	2/2300	0/92 <sup>a</sup>	Sénégal	Reynaud & Roger 1978 <sup>c</sup>
BGA	50/2850 <sup>a</sup>	2/114	Philippines	Saito & Watanabe 1978 <sup>c</sup>
BGA ( <i>Aulosira</i> )	12000 <sup>a</sup>	480	India	Singh 1976 <sup>c</sup>
BGA	125/2625 <sup>a</sup>	5/105	India	Srinivasan 1979 <sup>c</sup>
BGA ( <i>Gloeotrichia</i> )	24000	117	Philippines	Watanabe <i>et al.</i> 1977 <sup>c</sup>
<i>Chara</i> sp.	9000/15000	720/1200 <sup>b</sup>	India	Misra <i>et al.</i> 1976 <sup>d</sup>
<i>Chara, Nitella</i>	5000/10000	400/800 <sup>b</sup>	India	Mukherjy & Laha, 1969 <sup>d</sup>
<i>Najas, Chara</i>	5000 <sup>b</sup>	400	Philippines	Saito & Watanabe 1978 <sup>d</sup>
<i>Chara</i> spp.	2500/7500 <sup>b</sup>	200/600	France	Vaquer, 1984 <sup>d</sup>
<i>Marsilea</i>	25000	2000 <sup>b</sup>	India	Srinivasan, 1982 <sup>d</sup>
Total biomass				
fallow field	1000/3000	80/240 <sup>b</sup>	Philippines	Kulasooriya <i>et al.</i> 1981
planted field	7500	600 <sup>b</sup>	Philippines	Kulasooriya <i>et al.</i> 1981
fallow field	1250/2500 <sup>b</sup>	100/200	Philippines	Inubushi & Watanabe,
planted field	1250/6250 <sup>b</sup>	100/500	Philippines	in press
<b>Average</b>	<b>6000</b>	<b>350</b>		

a : extrapolated on the basis of 4% dry weight. b : extrapolated on the basis of 8% dry weight. c : quoted in Roger & Kulasooriya, 1980. d : quoted in Roger & Watanabe, 1984

The average composition of aquatic macrophytes is about 8% dry matter, 2 to 3% N (d.w. basis), 0.2 to 0.3% P, and 2 to 3% K. Planktonic algae have a lower dry matter content (averaging 4%) and a higher N content (3 to 5%). Components of the photosynthetic aquatic biomass usually have low dry matter content and high ash content (Roger and Watanabe, 1984; Roger *et al.*, 1986).

Biomass measurements and data on the composition of algae and aquatic macrophytes indicate that the N content of spontaneously growing photosynthetic aquatic biomass in planted rice fields rarely exceed 10-20 kg/ha but might attain 30-40 kg/ha in flooded fallow fields, when large populations of aquatic macrophytes develop.

Reported productivities of 50-60 g C/m<sup>2</sup> in 90 days (Saito and Watanabe, 1978), 70 g C/m<sup>2</sup> in 144 days (Yamagishi *et al.*, 1980), and 0.5 to 1 g C/m<sup>2</sup> per day (Vaquer, 1984) correspond to 10-15% of that of the rice crop and are similar to productivity values reported in eutrophic lakes.

## 2) N<sub>2</sub> FIXATION

### 21) SPONTANEOUS PHOTODEPENDENT N<sub>2</sub> FIXATION

Photodependent N<sub>2</sub>-fixing microorganisms in wetland rice field consist of photosynthetic bacteria, free-living BGA, and symbiotic BGA in *Azolla*.

The presence of photosynthetic bacteria has been recorded in rice soils, but their contribution in terms of kg N/ha is low (Roger and Watanabe, 1986).

Despite the lack of direct measurements of nitrogen fixed by BGA, there is enough indirect evidence to conclude that BGA have a moderate potential. In a review on BGA and rice, Roger and Kulasooriya (1980) reported that the average of 38 evaluations, mainly from acetylene reducing activity (ARA) measurements, was 27 kg N/ha per crop; maximum value was 50-80 kgN/ha per crop. Recent studies of BGA blooms and crusts (IRRI, 1986; Roger *et al.*, 1985; Roger *et al.*, 1986) indicate that: 1) a visible growth of BGA usually corresponds to less than 10 kg N/ha, 2) a dense bloom may correspond to 10-20 kg N/ha, and 3) higher values (20-45 kgN/ha) are recorded only under artificial conditions as in experimental microplots or in BGA soil-based inoculum production plots. More than two blooms of N<sub>2</sub>-fixing BGA is a rare occurrence during a crop cycle, therefore 27 kg N/ha per crop seems a reasonable estimate of photodependent BNF when BGA growth is visible.

Factors that permit the development of a bloom are still poorly understood. These may include depletion of N in the floodwater, P availability, low CO<sub>2</sub> concentration due to an alkaline reaction, low grazer populations, or presence of BGA resistant to grazing, and optimal temperature and light intensity.

*Azolla* is an aquatic fern which harbors the symbiotic N<sub>2</sub>-fixing BGA *Anabaena azollae*. Spontaneous development of *Azolla* in rice fields is less frequent than that of BGA. *Azolla* usually needs to be inoculated and grown when used as green manure (Watanabe, 1982).

## 22) UTILIZATION OF FREE-LIVING BLUE-GREEN ALGAE.

Because of the belief that N<sub>2</sub>-fixing BGA were not common in many rice soils, research on methods for utilizing BGA in rice cultivation has focused on inoculation. However, recent soil surveys indicate that heterocystous BGA are present in most rice soils at densities ranging from a few dozen to more than 10<sup>6</sup> colony forming units (CFU)/cm<sup>2</sup> of soil. The median is about 5 x 10<sup>4</sup> CFU/cm<sup>2</sup> (IRRI, 1985 ; Roger *et al*, 1985).

In addition, a study of BGA inocula composition shows that the number of CFU of N<sub>2</sub>-fixing BGA in the quantity of inoculum applied is most frequently considerably smaller than that of indigenous BGA present in the inoculated soil ( Roger *et al*, 1985). This indicates that inoculation is not the only possible way to utilize BGA and that emphasis should also be placed on agricultural practices that enhance indigenous BGA growth.

### 221) Algal inoculation.

A bibliographic survey (Roger and Kulasooriya, 1980) showed an average rice yield increase of 14% in field experiments where application of BGA inoculum increased yield. There might also have been a number of "no-effect" results unreported since such experiments are seldom published. In most experiments, only grain yield was measured. Currently, no data regarding environmental conditions, BGA establishment, algal biomass, or N<sub>2</sub>-fixing activity in successful inoculation experiments are available. Reasons for the yield increase are still unclear, especially in cases when a beneficial effect was observed with high levels of N fertilizer, which reportedly inhibit BGA growth (Roger *et al*, 1980)

Recent experiments (IRRI, 1985, 1986) show that while BGA inoculated in five Philippine wetland soils persisted for at least one month in the soils, their growth as a bloom was very rare (1 out of 10 cases). This was possible with *Aulosira fertilissima* when grazers were controlled. Blooms of indigenous strains were observed in other cases.

Reports on adoption of algal inoculation are somewhat conflicting, but, it appears to be restricted to a limited hectareage in two Indian states (Roger *et al*, 1985 ) and, possibly, Burma (Roger and Watanabe, 1986).

### 222) Agricultural practices to enhance BGA growth.

Agricultural practices known to enhance BGA growth are : liming of acidic soils, P application, straw application (App *et al.*, 1984), deep-placement of N fertilizer (Roger *et al.*, 1980), and grazer control (Grant *et al.* 1985) . Recent experiments (Reddy and Roger, unpublished) show that split P application is more efficient than basal application in increasing photodependent ARA along the crop cycle. While the effectiveness of these practices in increasing BGA growth and/or the ARA has been establish, no field experiment has yet quantified the relative contribution of the increased BGA activity and the direct effect of the practice to the increase in rice yield, when some was observed.

### 23) UTILIZATION OF AZOLLA

Because of its rapid growth and ability to grow together with rice, *Azolla* has been used as green manure for centuries in China and North Vietnam (Lumpkin and Plucknett, 1982 ; Watanabe, 1982). The reported maximum standing crops of *Azolla* range from 0.8 to 5.2 t d.w./ha (20-146 kg N) and average 2.1 t d.w./ha (70 kg N/ha) (Kikuchi *et al.*, 1984). Field trials, conducted for 4 consecutive years at 19 sites in 9 countries, showed that incorporating one crop of *Azolla* grown before or after transplanting was equivalent to a split application of 30 kg fertilizer N (IRRI, 1983). *Azolla* has a N potential similar to that of legume green manures. Several *Azolla* crops can be grown within a rice crop cycle and are easier to incorporate than legumes. However, the following environmental, technological, and economical constraints limit *Azolla* use to about two millions hectares of rice fields (estimate for 1982-83) (Roger and Watanabe, 1986) :

- *Azolla*, being sensitive to drought, requires a good water control that can be realized in only 20% of Asian rice fields.
- Propagated vegetatively, *Azolla* has to be maintained year round in a network of nurseries. Large quantities of inoculum are usually required (0.5 t/ha). A limited knowledge of conditions permitting sporocarp formation and the slow growth of newly germinated sporophytes (Watanabe, 1985) limit propagation through spores which could have alleviated problems related to inoculum conservation, multiplication and transport.
- Insects and fungi severely limit *Azolla* growth in humid tropics. Pesticide application is economically feasible in nurseries but not in the

field (Kikuchi *et al.*, 1984).

- Optimum temperature requirement for most *Azolla* species is below the average temperature in the tropics. This limitation can be reduced by selecting cold or heat tolerant strains (Watanabe and Berja, 1983).
- *Azolla* can grow without P application in soils rich in available P (Olsen P > 25 ppm.) and having a low sorption capacity (<1500 mg P<sub>2</sub>O<sub>5</sub>/100g). Phosphorus has to be applied in other soils (Watanabe and Ramirez, 1984).
- Labor cost may be limiting (Kikuchi *et al.*, 1984).

Among green manures, *Azolla* is still not widely utilized. However many countries are considering it for adoption (IRRI, 1984; Roger and Watanabe, 1986). Research on possible usage of *Azolla* in integrated production systems such as rice-fish-*Azolla* is conducted in China (Liu, in these proceedings).

### **3) NITROGEN IMMOBILIZATION**

Photosynthetic biomass prevents N losses by immobilization of N of the floodwater and returns it as organic N into the soil. This role is obvious but poorly documented. Using a gas lysimeter, Vlek and Crasswell (1979) estimated that, three weeks after N fertilizer application, immobilization in the algal biomass of N from fertilizers was 18-30% for urea and 0.4-6% for ammonium sulphate. These results were confirmed by <sup>15</sup>N experiments showing immobilization of 18-41% of N from urea applied in the floodwater three weeks before (Vlek *et al.* 1980). Low immobilization (< 5%) was observed with incorporated ammonium sulphate (Inubushi and Watanabe, personal communication).

### **4) N RECYCLING BY GRAZING**

Recent studies related with nutrient recycling from photosynthetic biomass deal with grazing of BGA by invertebrate populations. These were initiated because insecticide application was seen to increase algal growth and zooplankton was identified as a major limiting factor for BGA growth (Wilson *et al.*, 1980b; Grant *et al.*, 1985).

The rice field fauna directly responsible for the breakdown of the photosynthetic biomass consist of microcrustaceans and gastropods. These, together with the protozoans and rotifers, also recycle nutrients from decaying photosynthetic biomass. Translocation of photosynthetic biomass and breakdown products from the surface to the deeper soil layer is expedited by

tubificids worms (Grant and Seegers, 1985b).

Quantitative data on grazing are still very limited and estimates of regeneration rates cannot be proposed until the population dynamics of grazers and their diets have been elucidated. However, available data suggest a very significant activity of the zooplankton. Grazing rates of ostracods on BGA varies from 1 to more than 100  $\mu\text{g}$  d.w.alga/ostracod per day and diet preferences are exhibited (Grant et al., 1983). Ingestion and excretion rates of *Heterocypris luzonensis* (Ostacoda) determined in the laboratory by Grant and converted to BGA consumed by a field population (8700 / $\text{m}^2$ ) totalled 187 g N/ha per day, 118 g of which was excreted as  $\text{NH}_3$  (see Roger et al. in press).

Grazing by invertebrates permits nutrient recycling but limits algal growth. In microplot (0.5  $\text{m}^2$ ) experiments, N accumulation in the surface layer increased ( 1 to 3.5 times) when grazers were controlled, the rate depending on the soil type and algae growing on it. Nitrogen accumulation during two month ranged from 5 to 18 kg/ha in the control, and from 15 to 28 kg in insecticide treated plots (Roger et al, in press).

### 5) NITROGEN ACCUMULATION AT THE SOIL SURFACE

In wetland soils, N accumulates at the soil surface (App et al, 1984). Nitrogen may come from 1) the atmosphere, through  $\text{N}_2$  fixation, 2) the floodwater, through immobilization by the aquatic biomass, and 3) the soil, after absorption by rooted plants or ingestion by invertebrates. The process is mostly photodependent, as demonstrated by field experiments with a control placed in the dark (App et al, 1984) or a control that prevents exchanges between surface soil and deeper soil. Ono and Koga (1984) measured the accumulation of 35 kg N/ha per crop under normal field conditions and of 26 kg N when surface soil was isolated from deeper soil by placing it in Petri dishes.

### 6) AVAILABILITY OF N OF THE PHOTOSYNTHETIC BIOMASS TO RICE

Studies by Wada et al. (1982) and Watanabe and Inubushi (1985) show a positive correlation between the amount of chlorophyll-like substances in the soil and its N-supplying ability. This indicates that the photosynthetic biomass contributes significant quantities of available N and has an important role in maintaining the fertility of wetland soils.

Availability of algal N to rice has been quantified in  $^{15}\text{N}$  experiments with BGA by Wilson et al. (1980a), Tirol et al. (1982), and Grant and Seegers (1985a) ( Table 2 ). Recovery of BGA N in rice crop varied from 13 to 50 %, depending on the nature of the algal material (fresh vs. dried), the method of application (surface applied vs. incorporated), and the presence or absence of soil fauna.

Highest recovery (50%) was obtained when fresh material was incorporated in a soil depleted of fauna (Wilson *et al.*, 1980a). Lowest recovery was obtained when dried material was applied on the surface of a soil rich in tubificids (*Oligochaeta*) (Tirol *et al.*, 1982). Grant and Seegers (1985a) showed that tubificid activity reduced the recovery of algal N by rice by making the soil N available through a mineralization process. A residual effect of algal N was observed in the second rice crop where 4 to 7% of algal N was recovered (Tirol *et al.*, 1982; Grant and Seegers, 1985a).

Few data are available on the utilization of N from macrophytes by rice (Table 2). Shi *et al.* (1980) reported that 25% of the N from incorporated  $^{15}\text{N}$ -labeled water hyacinth was recovered in the rice crop. In a field experiment, Ito and Watanabe (1985) observed that when  $^{15}\text{N}$  labeled *Azolla* was placed at the surface of the soil, about 66% of *Azolla* N was lost and 12-14% was recovered in the rice plants. When *Azolla* was incorporated, losses were significantly reduced and availability increased to 26%.

**Table 2 : Availability of N of the photosynthetic biomass to rice.**

Material		N recovery (%)		Experimental	References
Nature	State	Surface applied	Incorporated		
Blue-green algae	fresh	37	52	? pot	Wilson et al 1980
Blue-green algae	dry	14	28	- pot	Tirol et al 1982
"	dry	23	23	+ field	" "
"	fresh	-	38	- pot	" "
Blue-green algae	fresh	24	44	- pot	Grant & Seegers 1985
"	fresh	25	30	+ pot	" "
Blue-green algae	dry	-	35-40	- pot	Mian & Stewart 1985
Water Hyacinth	fresh	-	25	+ field	Shi et al 1980
<i>Azolla pinnata</i>	fresh	-	26	+ field	Watanabe et al 1981
<i>A. caroliniana</i>	fresh	12/14	26	+ field	Ito & Watanabe 1985
<i>A. caroliniana</i>	dry	-	34	pot	Mian & Stewart 1985
<i>A. caroliniana</i>	fresh	-	32	+ field	Kumarasinghe 1986
<b>Average</b>		<b>21</b>	<b>31</b>		

This results indicate that N fixed or immobilized in the photosynthetic biomass is much more efficiently used by rice when incorporated into the soil.

## **7) N LOSSES BY NH<sub>3</sub> VOLATILIZATION**

The poor efficiency of N fertilizer utilization by rice is partly due to N losses by NH<sub>3</sub> volatilization which range from 2 to 60% of N applied (Fillery *et al.*, 1984; Simpson and Freney, in these proceedings). Water pH is a major factor in determining the rate and extent of losses (upto pH 9, NH<sub>3</sub> concentration increases by a factor of 10 per unit increase in pH). Therefore, aquatic photosynthetic organisms have a key role in NH<sub>3</sub> volatilization. They deplete CO<sub>2</sub> in floodwater during the day, and replenish it partly at night through respiration, thus causing diurnal changes in floodwater pH which may reach values as high as 10 by midday and decrease by 2-3 units at night (Mikkelsen *et al.*, 1978). Practices decreasing algal growth, such as Cu application (Mikkelsen *et al.*, 1978) and deep-placement of N fertilizer (Zhi-Hong Cao *et al.*, 1984) decrease diurnal variations of pH and N losses.

Fillery *et al.* (1986) estimated the photosynthetic biomass in fields where N losses were evaluated. One week after fertilizer application, a limited and uneven growth of algae (about 100 kg fresh weight/ha) was observed in N treated plots where pH at noon time ranged from 7.8 (no visible algal growth) to 10.5 in the vicinity of algal colonies. Despite the low algal biomass, significant N losses (30 - 40%) occurred, suggesting that large algal populations are not required to increase floodwater pH to levels which support rapid N losses. Apparently, the most unfavorable situation seems to be at the beginning of the crop cycle, when there is almost no canopy and the resulting high light availability permits a high photosynthetic activity of a low algal biomass sufficient to induce a significant pH increase in the floodwater but not to limit N losses through immobilization.

Measurements in IRRI showed that submerged macrophytes (*Chara*, *Najas*, etc.) significantly increased floodwater pH whereas it was fairly stable under floating macrophytes *Azolla* and *Lemna* (Roger *et al.*, in press). Such result indicates that there is a potential for combined use of *Azolla* and chemical N.

## **8) CONCLUSION**

When considering the relationship between photosynthetic biomass and N management in rice cultivation, the two most obvious methods of practical utilization are 1) enhancing BNF and 2) decreasing N fertilizer losses due to NH<sub>3</sub> volatilization.

BNF technologies currently adopted by rice farmers (green manuring with legumes or *Azolla*) are labor intensive and most often used under socioeconomic conditions where labor intensive practices are economically feasible or where economics is not a major factor. Utilization of free-living

BGA should not be as labor intensive as green manuring but has moderate potentialities and is still limited by methodological problems. BGA are competitive under unfavorable environments as in less productive problem soils where farmers tend to apply less fertilizers. Under such conditions the BGA's moderate contribution to yield increase could be of value. However, it is unlikely that BNF could be an exclusive N source for attaining high rice yields under the most economical conditions (Roger and Watanabe, 1986). The future of BNF in rice cultivation most probably lies in integrated management.

A better knowledge of rice field ecology will contribute to high yields with reduced inputs through a more efficient use of chemical fertilizers and the simultaneous utilization of BNF. Deep-placement of nitrogen fertilizer (De Datta *et al.*, 1983) significantly decreases N losses by volatilization and permit photodependent BNF by BGA, (Roger *et al.*, 1980). Deep-placement of nitrogen fertilizer, coupled with practices that enhance BGA growth, is a good example of a technology that has to be developed to take advantage of the potentialities that the photosynthetic aquatic biomass in rice fields has to offer.

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