# FERTILITY BUILD-UP OF RICE FIELD SOILS BY BLUE-GREEN ALGAE

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### INTRODUCTION

In the account that follows, the results of a study covering a period of five crop seasons to evaluate the practical and economical aspect of utilization of blue-green algae, chiefly for the nitrogen enrichment of the rice soils, are presented. Some of the results of fundamental and applied interest have already been published (Relwani, 1963; 1965; Relwani and Subrahmanyan, 1963; Subrahmanyan et al., 1964 a, b, c; Subrahmanyan and Sahay, 1964, 1965). The actual quantum of nitrogen uptake by the crop as a result of blue-green algal application as well as interaction of different soil types to blue-green algal inoculation are discussed in other papers in the *Proceedings* (Sahay, 1965; Subrahmanyan et al., 1965).

The necessity for these investigations has been felt for a long time as our soils are deficient in nitrogen and supply of nitrogenous fertilizers falls far short of even the bare minimum required. Hence all possible sources are worth examining and in our investigations we have attempted to explore the possibility of tapping the abundant nitrogen of the atmosphere using the blue-green algae as the tool, for, these algae thrive best in the water-logged environment of the rice crop.

### **METHODS**

The lay-out and methods adopted have been described in detail by one of us (Relwani, 1963). Certain pertinent procedures have been referred to in the text. All the usual agronomical practices have been adhered to. It may be emphasized here that no nitrogen has been given to any of the treatments except to one, in the form of ammonium sulphate, to assess the nitrogen supply by the algae in terms of this popular fertilizer for the rice crop.

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#### EXPERIMENTAL

### I. 1961-62

Pot house and field experiments were initiated in 1961 in the second season (December-April), Dalua, with rice variety Ptb. 10 to study the rôle of blue-green algae on crop yield. The treatments consisted of lime (2000 kg./ha.), superphosphate (60 kg.  $P_2O_5/ha$ .) and sodium molybdate (0.28 kg./ha.) in different combinations without and with blue-green algae. Ammonium sulphate was applied at two levels, 16.8 and 33.6 kg. N/ha. to compare the yield responses of this popular fertilizer for rice crop. Randomised block design of lay-out was employed with four and five replications, in the field and pot house respectively.

Top soil from fields with blue-green slimy formations consisting of species of Nostoc, Anabaena, Microcystis, Cylindrospermum, Phormidium and Aphanothece, were scraped, air-dried and inoculated at the rate of 3.0 gm. per 15 kg. of soil per pot and 340 gm. per plot of 1/187.06 ha. The development of algal forms in the pots was found to be quite vigorous from the second fortnight of November to early December (mean maximum temperature  $27.7^{\circ}$  C. and mean minimum temperature  $14.3^{\circ}$  C.) but slowed down afterwards up to the end of January (mean maximum temperature  $26.2^{\circ}$  C. and mean minimum temperature  $11.6^{\circ}$  C.). Active and visible growth of algae re-started in the month of February and continued up to the end of March (mean maximum temperature  $30.8^{\circ}$  C. and mean minimum temperature  $18.4^{\circ}$  C.). The setback to the growth during the cold season has been observed in the laboratory cultures also (Subrahmanyan and Sahay, 1964).

In the ammonium sulphate treatments, green forms (Spirogyra sp. and Euglena sp.) dominated in such great abundance that frequent stirring of the top soil in the pots had to be done to prevent smothering of the rice plants. This observation has a practical bearing for rice cultivators in that they should interculture their crop more frequently when using nitrogen fertilizers in large quantities.

Pot cultures.—In the pot house, blue-green algae or lime application individually increased rice yield by 15.72% over check (no treatment and no algae); their combination significantly increased the yield by 81.25% (Table I). This may possibly be due to the direct rôle of lime in the nutrition of blue-green algae as well as in raising the pH of soil-water medium to alkaline reaction which is conducive to the development of the blue-greens,

TABLE I

Yield of grain and percentage increase over control under various treatments 1961–62 second crop

		_	Po	ot exp	periment	Field ex	xperiment
Treatment	ts		ir	eld n m.	% over control	Yield in kg./ha.	% over control
Control	• •	I	10	-56		291.57	
C + Lime at 224 kg./ha.	• •	I	E 12	·22	15.72	332.93	19 · 13
C + algae + lime	• •	1	N 19	14	81.25	379 • 29	30.29
C + algae		F	12	22	15.72	347 · 74	14.19
C + lime + sodium moly superphosphate	bdate +	(	3 16·	18	53.27	474 · 88	62.87
C + lime + sodium moly superphosphate + algae			19.	10	80.87	512.67	75.83
C.D. at 5%	• •	••	4-	-51	••	160 · 74	• • •

<sup>\*</sup> For more details vide L. L. R., 1963.

Superphosphate and sodium molybdate, like lime alone, produced small increases but the lime + superphosphate + sodium molybdate mixture produced an appreciable increase of 53·22% over the check. It appears that apart from the nutrition of the rice plant, this mixture exercises a beneficial effect on the nitrogen-fixing activity of the indigenous species of bluegreen algae present in the soil. In combination with inoculated blue-green algae, there was a further improvement and the yield increased by 80·87%.

Field experiments (Table I).—Under field conditions, blue-green algae and lime treatments increased the yield by  $19 \cdot 13\%$  and  $14 \cdot 19\%$  respectively; their combination contributed to an increase of  $30 \cdot 29\%$ . The mixture of lime + superphosphate + sodium molybdate produced a still higher response of  $62 \cdot 87\%$  and with inoculated algae, the increase went up to  $75 \cdot 83\%$ . Thus, the trends in the pot house were also observed in the field.

### II. 1962 (Main), 1962-63 (Dalua) and 1963 (Main) Seasons

Based on the favourable results obtained in the previous season, described above, the trials were reoriented. Partial soil sterilization was introduced as a treatment to destroy the existing indigenous algal flora in order to get a more vivid picture about the response to introduced forms of blue-green algae. Partial soil sterilization was done by placing the soil for pots in the autoclave at 20 lb. pressure for 1 hour. In the field, top soil was heated by burning paddy straw. The nutrient mixture dose was reduced to 1,000 kg. lime/ha., 20 kg.  $P_2O_5/ha$ .; molybdate dose was kept the same.

The experiment was carried out for three seasons, main season 1962, second season (Dalua) 1962-63 and main season 1963. Medium duration variety C.R.T. 141 was used in the main seasons and short duration variety Ptb. 10 in the second season. In the field, heating of top soil was done for all the seasons, but in the pots, soil sterilization was done only once, in the main season of 1962, as the effect of this treatment on crop yield was found to be so high as to mask the beneficial rôle of blue-green algae.

In the 1962 main season, Anabaena sp. was inoculated in the pots and a mixture of species of Anabaena, Nostoc, Phormidium, and Aphanothece was used in the field at the rate of 2 gm./ha. dry weight (circa 200 gm. wet weight).

In the second season (1962-63), a mixture of species of *Nostoc*, *Scytonema*, *Anabaena* and *Tolypothrix* from the laboratory cultures of one of us (R.S.) was inoculated at the same rate; and a similar mixture was employed for the main season 1963 crop.\*

Periodical collections were made of the algal growth in the experimental plots. A consolidated list of algae thus recorded is given in Table II in the order of their abundance based on microscopical examination. It may be noted that most of them belong to the N-fixing genera. Some green algae, species of Spirogyra, Mougeotia, Zygnema, Oedogonium and Desmids as well as Diatoms were present, but in negligible quantities in some of the plots except in the ammonium sulphate treatments where they seemed to thrive better.

One interesting observation of great significance in rice culture may be mentioned here. It was noticed that in the plots with blue-green algal growth.

<sup>\*</sup>The algae were left on a layer of sand, allowed to dry and this algae + sand, mixed with more sand for uniform distribution was applied in equal quantities in the treatments concerned,

Consolidated list of blue-green algal species that appeared in the experimental plots during five crop seasons (in the order of their dominance) TABLE 11

Control	Rabbing	Control+ blue-green algae	Rabbing+ blue-green algae	Control+ fertilizer mixture	Rabbing+ fertilizer mixture	Control+ fertilizer mixture+ blue-green algae	Rabbing+ fertilizer mixture+ bluc-green algae	Ammonium sulphate at 20 kg./ N/ha.
Cylindro- spermum	Rivularia	Anabaena	Anabaena	Anabaena	Gloeotrichia Anabaena	Anabaena	Апавсепа	Anabaena
Anabaena	Gloeotrichia Nostoc	1 Nostoc	Cylirdi c- spermum	Cylindro- spermum	Cylir.dro- spermum	Ѕсуtопєта	Rivularia	Scytonema
Aulosira	Cylindro- spermum	Cylindro- spermum	Gloeotrichia	Gloeotrichia	Rivularia	Westiella	Gloeotrichia Aulosira	Aulosira
Scytonema	Scytonema	Scytonema	Rivularia	Rivularia	Anabaena	Gloeotrichia Scytonema	Scytonema	Gloeorichia
Gloeotrichia	Aulosira	Aulosira	Calothrix	Aulosira	Calothrix	Cylindro- spermum	Aulosira	Cylindro- spermum
Westiella	Anabaena	Westiella	Aulosira	Scytoriensa	Westiella	Lyngbya	Tolypothrix Hydrocoleus	Nostoc s
Aphanothece	Calothrix	Gloeotrichia	Gloeotrichia Scytoncma	Oscillatoria	Nostoc	Phormidium Westiella		Aphanothece
Oscillatoria	Tolypothrix	Tolypothrix Aphanothece Aphanothece	Aphanothece	Phormidiem	Aulosira	.:	Aphanothece Oscillatoria	Oscillatoria
•	We stiell a	Lyngbya	Lyngbya	•	Scytonema	:	Phormidium Lyngbya	Lyr.gbya
• 1	Aphanothece	Aphanothece Oscillatoria	Spirulir.a	:	Apharothece	:	Oscillatoria	Phormidium
**************************************	Phormidium	Phormidium Phormidium	Phormidium	:	Oscillatoria	:	Chroococcum	•
•	Lyngbya	•	Oscillatoria	:	:	•	Lyngbya	:
• • •	Oscillatoria	•	•	:	•	•	:	•

weeds such as *Panicum* sp., *Cyperus* sp., *Hydrolea* sp. and *Ludwighia* sp. common in the paddy fields, were almost absent. Similar observations have been made in a demonstration plot in a farmer's field as well. It would appear that, very probably, some external metabolites of the blue-green algae have a deleterious influence on their growth. Such phenomena brought about by planktonic blooms of algae are well known (*refer* Subrahmanyan, 1959). Thus, it would seem that promotion of blue-green algal growth, apart from N enrichment, serves to suppress weeds as well with benefit to the rice crop.

Pot culture experiments (Table III).—The data show that in the absence of nutrient mixture (Treatment LB), blue-green algae produce small increases of 3.6 and 0.9% in grain yield in the main season var. C.R. T141 in 1962 and 1963 respectively. The corresponding increases in straw yields were of the order of only 12.9 and 4.1% respectively. For the same treatment, second season (1962–63) var. Ptb. 10 showed a higher response of 20.3 and 26.4% in the yield of grain and straw respectively.

The yield in check, in the second season, was low compared with the main season. It is, therefore, quite possible that higher increases over check were recorded by a number of treatments although the absolute increases by these treatments were generally not so high compared to main season yield with var. C.R. T141.

Lime + superphosphate + sodium molybdate mixture by itself (Treatment MA) produced grain increases of 5.8, 45.2 and 54.4% in the first, second and third crops respectively. The progressive increases in yield may be due to the cumulative effect of this nutrient mixture (refer Abichandani and Patnaik, 1961) and the activity of the indigenous flora of blue greens in the soils. In combinations with introduced blue greens, increases of 50.2, 235.1 and 281.8% were obtained during the corresponding seasons. This confirms the rôle of lime, superphosphate and sodium molybdate in improving the efficiency of blue greens in nitrogen fixation.

Partial soil sterilization (Treatment NA) itself accounted for considerable increases: 181.8, 172.7 and 90.4% in grain yields during the three seasons in order. In combination with blue-green algae, the increases recorded during the respective seasons were of the order of 159.3, 154.9 and 146.3% respectively. Thus, the effect of blue-green algae becomes visible only in the third season when the beneficial effect of soil sterilization itself had wancd. Similar trends were observed in the case of straw as well.

The beneficial effect of soil sterilization appears to be due to the improvement in the physical properties of the soil, death of undesirable pathogens,

TABIE III

Yield of paddy grain and straw (kg. per hectare)—Average of four replications

			Grain	Grain yield					Straw	Straw yield		
Treatments	1962 (	1962 (T. 144)	1962–63	1962-63 (Ptb. 10)	.) 8961	1968 (T. 141)	1962 (T. 141)	(, 141)	1962-63	1962-63 (Ptb. 10)	1963 (	1963 (T. 141)
	A	В	· A	В	A	В	A	В	A	n D	A	В
L. Control (Check)	2379.5	2578·4 (8·4)	459.8	658.0 (43.1)	1796.8	2194·7 (24·0)	3795.9	4060.7	952.0	1616.1 (69.1)	1994.3	2275· <b>5</b> (14·1)
<ul><li>M. Lime + superphosphate</li><li>+ sodium molybdate</li><li>N. Soil sterilization</li></ul>	2942·1 (23·6) 2857·8 (20·1)	3441.2 (44.6) 2977.9 (25.2)	789·6 (71·7) 818·2 (78·0)	978.9 (112.9) 879.2 (91.2)	3000.9 (69.6) 2496.2 (41.0)	3979.1 (124.8) 2620.2 (48.0)	4767.0 (25.6) 4510.0 (20.2)	5561.4 (46.5) 4796.4 (26.4)	18 <b>6</b> 2·1 (95·6) 1960·1 (105·9)	2436·1 (155·9) 2240·1 (135·3)	3145·0 (57·7) 2762·2 (38·5)	4326.5 (116.9) 2953.6 (48.1)
O. Soil sterilization + lime + superphosphate +	3489.4 (46.7)	3661.3 (53.9)	1033·3 (124·7)	1070.8 (132.9)	3170·1 (79·1)	3833.8 (116.6)	56±9·7 (48·8)	5826·3 (58·0)	2520·1 (164·7)	2688.1 (182.3)	3397.0 (70.4)	$3870.4 \\ (94.1)$
Sodium motypdate  P. Ammonium sulphate at 20 kg. N/hectare	\$002.6 (26.2)		819·3 (78·2)	::	2463·1 (39·2)	::	4825.3	::	2002.1 (110.3)	::	2629.9	::
C.D. at 5%	13	139.6	15	153.3	35	352.3	398	363•5	48	484.0	43	437.8
C.D. at 1%	18	189.3	20	207.8	47	477.5	493	492.6	•	•	64	642.0

A-Without blue-green algae B-With blue-green algae

Figures in brackets represent per cent. increases over control.

parasites and weeds and to increased production of ammonia, nitrates and other available mineral plant nutrients (Relwani and Subrahmanyan, 1963; Kleimashevkiis et al., 1962). It may be mentioned here that the irrigation water in the pots in these treatments were always clear while in the others it remained turbid.

Soil sterilization in combination with the nutrient mixture (Treatment OA) further improved grain yields by  $218 \cdot 9$ ,  $203 \cdot 4$  and  $174 \cdot 1\%$  in the respective seasons. In combination with blue-green algae, the increases recorded went up to  $275 \cdot 6$ ,  $227 \cdot 1$  and  $317 \cdot 1\%$  respectively. In this instance also the effect of blue-green algae was most pronounced in the third season with the decline in the influence of soil sterilization.

It may further be observed that the effect of blue-green algae in combination with the nutrient mixture used was much higher than that of ammonium sulphate at 20 kg./N/ha. which was only 8.3, 72.7 and 108.2% respectively in the seasons concerned.

Field experiments (Table IV).—Blue-green algae without the nutrient mixture (Treatment LA) produced non-significant increases of 8.4% grain and 7% straw in the first main season C.R. T141 crop. In the second crop (var. Ptb. 10) significant increases of 43.1% and 69.1% increase of grain and straw were obtained. In the third crop, a significant increase of 24.0% in the grain and a non-significant increase of 14.1% straw were recorded. The pattern of responses during the different seasons follow the same trends as in the pots described earlier.

The nutrient mixture (Treatment MA) produced increases of  $23 \cdot 6$ ,  $71 \cdot 7$ ,  $69 \cdot 6\%$  grain and  $25 \cdot 6$ ,  $95 \cdot 6$  and  $57 \cdot 7\%$  straw during the three seasons respectively. With inoculated algae the corresponding increases went up to  $44 \cdot 6$ ,  $112 \cdot 9$  and  $124 \cdot 8\%$  for grain and  $46 \cdot 5$ ,  $155 \cdot 9$  and  $116 \cdot 9\%$  for straw. In every season, the combination of blue greens and nutrient mixture produced increases which were significantly higher than that produced by either of the two separately.

The effect of rabbing (heating of top soil by burning straw) (Treatment NA) was less compared with the treatment in the pots where the entire quantity of soil had been sterilized in an autoclave. The increase obtained during the first, second and third seasons were of the order of  $20 \cdot 1,78 \cdot 0$  and  $41 \cdot 0\%$  respectively. Straw yields also followed similar trends. In combination with blue-green algae, the additional responses were small and not significant.

Rabbing in combination with nutrient mixture (Treatment OA) produce substantial increases of 46.7, 124.7 and 79.1% in the grain and 48.8, 164.7

TABLE IV

Yield of paddy grain and straw pots (gm./pot)—Average of five replications

Treatments	1962 (	(T. 141)	Grain 1962-63	Grain yield 1962-63 (Ptb. 10)	(1963	(1963 T. 141)	1962 (T. 141)	r. 141)	Straw 1962-63	Straw yield 1962-63 (Ptb. 10)	1963	1963 (T. 141)
	V	В	A	B	A	B	A	B	A	B	V	B
L. Control (Check)	10.43	10.81	6.30	7.58	13.26	13.38	14.25	16.09	6.37	8.05	14.80	15.4
	:	(3.6)	:	(20.3)	;	(10.9)	:	(12.9)	:	(26.4)	:	$(4\cdot 1)$
M. Lime + superphosphate + sodium molybdate	(5.8)	15·67 (50·2)	9.15 $(45.2)$	21.11 (235.1)	20.48 $(54.4)$	50.63	14.05	24·85 (74·4)	9.25 (45.2)	22·25 (24 <b>9·</b> 3)	24·1 (62·8)	86•1 (481•8)
Soil sterillzation	(181.3)	(159.3)	(172.7)	16·06 (154·9)	25. <b>25</b> (90.4)	32.66 (146.3)	41.72 (192.8)	34·82 (144·4)	(178.75)	$17.13 \ (168.9)$	33.2 (124.8)	$^{40.6}_{(174.3)}$
0. Soil sterilization + lime + superphosphate +	33.81	39·17 (275·6)	19·14 (203·4)	20·61 (227·1)	36.34 $(174.1)$	55.31	43.86 (207.8)	47.92 (2 <b>\$</b> 6.3)	$19.95 (213 \cdot 2)$	21·39 (235·8)	58.4 (294.6)	79.6 (437.8)
P. Ammonium sulphate at 20 kg. N./hectare	11.30 (8.3)	::	10.88	::	$^{27.61}_{(108.2)}$	::	18.07 (26.8)	::	11.47 (80.1)	::	36.8 (148.6)	::
C.D. at 5%	4	4.30	က်	3.59	4	4.65	7.58	. 82	က်	3.84	<b>-</b> :	1.63
C.D. at 1%	δ.	5.77	4.	4.83	.9	6.25	10.23	53	•		.4	2.19

A-Without blue-green algae. B-With blue-green algae.

Figures in brackets represent percent increases over control,

and 70.4% in the straw yields during the three seasons in order. The increases produced by this combination were generally found to be significant over either of the two treatments separately (MA, NA). With inoculated blue greens, there were further increases which were quite appreciable in the first and third seasons. Thus, rabbing, nutrient mixture and blue-green algae combination produced the highest yields. The effect of this combination was a little more than double the effect of ammonium sulphate at 20 kg./N/ha.

It may be mentioned in this connection, that in a series of pot house experiments conducted for three seasons Subrahmanyan et al. (1964 b) found that though in the initial stages the responses to partial sterilization of soil (autoclaving, top soil heating and application of formaldehyde) themselves were highly significant, the effect diminished in the succeeding seasons to non-significant levels while the algal inoculation in combination with nutrient mixture continued to maintain the fertility, obviously, by their rôle in N fixation and addition of organic matter as pointed out by Subrahmanyan and Sahay (1964).

### III. Residual Effect of Blue-Green Algae Inoculation

The pot house and field experiments discussed above were continued in the two succeeding seasons (1963-64 second crop and 1964 main season). For this, seedlings were planted in the pots and the field after just puddling only and neither nutrient mixture nor blue-green algae were applied as for the earlier crops. Ammonium sulphate at 20 kg./N/ha. was, however, applied to the respective earlier similar treatments for comparison with the responses in the residual effect study. The object was to study whether earlier application of blue-green algae as well as nutrient mixture to promote their growth had contributed to the fertility build-up of the soil; in other words, to assess whether the above treatments during the three earlier seasons have in any way conditioned the soil favourably for the rice crop.

The results are presented in Tables V and VI. Under pot culture conditions the residual effect of former treatments MA and MB gave significantly higher yields over control (I). In the next crop also, main 1964, the trend of response remained the same; significantly higher yields were obtained from the earlier treatments MA and MB.

This confirms that blue-green algae not only have an immediate effect in increasing the yield during the season, but also leave a significant residual effect which is found sufficient to support at least two crops of rice without any manuring, the gross yield being not affected at all.

Yield of paddy grain and straw—Residual fertility effect of different treatment combinations with blue-green algae—Pots (gm. per pot)—Average of five replications

;		Grai	n yield			Stra	w yield	
Treatments	1963–64	4 (Ptb. 10)	1964 (	(T. 141)	1963–64	4 (Ptb. 10)	1964	(T. 141)
	A	В	A	В	A	В	A	В
I. Control	8.04	8·25 (2·6)	10.50	10·30 (-1·9)	6.54	7·13 (9·0)	10.24	10·24 (0·0)
M. Lime + super- phosphate + sodium molybdate	15·39 (91·4)	23·04 (186·6)	13·9 (32·4)	17·4 (65·7)	11·06 (69·1)	15·16 (131·8)	12·30 (20·1)	14·76 (44·0)
N. Rabbing	11·14 (38·5)	11·91 (48·2)	9·1 (-13·3)	8·9 (-15·2)	8·84 (35·2)	9·54 (45·9)	9·50 (-7·2)	9·12 (-10·9)
O. Rabbing+lime +superphos- phate+sodium molybdate	18·71 (132·8)	21·80 (171·1)	14·3 (36·2)	15·9 (51·4)	13·46 (105·8)	14·18 (116·9)	13·98 (36·5)	14·46 (41·2)
P. Ammonium sulphate 20kg. N/ha	11·26 a (40·0)	•••	11·20 (6·7)	• •	7·64 (16·8)	••	12·2 (19·1)	***
C. D. at 5%	4	·18	4.	90	5	- 50	4	10

A—Without blue-greeen algae. B—With blue-green algae. Figures in brackets represent percent increases over control.

It would appear that establishment of a good blue-green algal flora would serve as a perpetual source of enrichment of the soil; only, occasionally, nutrient mixture will have to be applied to promote algal growth and some rice nutrient other than nitrogen.

As regards 1964 main season data, it is necessary to draw attention to a fact to avoid confusion. A reference is invited to Tables IV and VI. It may be seen that control (IA) recorded an yield of 3,529 kg./ha. in 1964 as against an yield of 2,379·5 and 1,769·8 kg./ha. in 1962 and 1963 main crops of C.R. T141. In 1964, the highest yield of a control (no manure) crop in the Farm was only 2,500 kg./ha.; and, in an experiment with same variety conducted in a nearby plot using green manure, potassium sulphate, lime and

TABLE VI

Yield of grain and straw—Residual fertility effect of different treatment combinaions with blue-green algae—Yield (kg. per hectare)—Average of four replications

		Grain	yield			Straw	yield	
Treatments	1963–64	(Ptb. 10)	1964	(T. 141)	1963–64	(Ptb. 10)	1964	(T. 141)
	A	В	Α	В	A	В	A	В
I. Control (Check)	 957	934 (-2·6)	3529	3504 (-0·7)	1151	1065 (-7·5)	4575	4425 (-3·3)
M. Lime+superphosphate+sodium molybdate	853 (-10·9)	1044 (9·0)	3377 (-4·3)	3623 (2·7)	986 (-4·3)	1178 (2·3)	4125 (-9·8)	4763 (4·1)
N. Rabbing	923 (-3·6)	$923 \\ (-2.6)$	3825 (8·4)	3486 (-1·2)	1024 (-11·0)	1091 (-5·2)	5213 (13·9)	4763 (4·1)
O. Rabbing+lime+ superphosphate+ sodium+molybdate	1172 (22·5)	971 (1·5)	3520 (-0·3)	3827 (8·4)	1211 (5·2)	(-7.5)	4913 (7·4)	4763 :(4·1)
P. Ammonium sulphat 20 kg. N/ha.	te 1196 (25·0)	• •	3648 (3·4)	••	1406 (22·2)	••	5100 (11·5)	•••
C.D. at 5% C.D. at 1%	Not sig	nificant	2	45	Not sig	nificant	Not sig	gnificant

A—Without blue-green algae. B—With blue-green algae.

Figures in brackets represent per cent. increases over control.

ammonium sulphate, the yield during the same season was only 3,551 kg./ha.\* (Dr. M. S. Chaudhury, Agronomist, CRRI, oral communication). In our experimental plots, soon after transplantation, in 1964, the crop was completely submerged due to heavy rains and a sheet of water covered the entire area. This led to algae from algal plots moving over to non-algal and control plots

<sup>\*(1)</sup> Green manure (Dhaincha crop grown with superphosphate at 40 kg./P<sub>2</sub>O<sub>5</sub>/ha.) about 5,000 kg. ploughed back;

<sup>(2)</sup> Basal dressing of 500 kg. lime, 40 kg. K<sub>2</sub>O/ha. potassium sulphate, 10 kg./N/ha. ammonium sulphate;

<sup>(3)</sup> Top dressing of 10 kg./N/ha. ammonium sulphate after establishment; and

<sup>(4) 10</sup> kg./N/ha. ammonium sulphate three weeks before flowering.

as well. Examination of the collections made (Table II) showed as many as 8 species of N fixing blue-green algae in the control and also ammonium sulphate plots which were not present either as many in number or quantity in the earlier seasons. The gross yield from the whole area had gone up over 20% in the third main crop. Obviously, the algae have enriched the soil and boosted the yield to this high level in the control plot also; this only further confirms that blue-green algae could be profitably used to enrich the soil for the rice crop.

The data in the preceding paragraph would suggest that blue-green algae alone are able to maintain the yield at a high level and also that, apart from nitrogen, the algae supply the crop with certain substances which help maintain the yield potential, The latter point finds support in a trial under progress where within 10 days after application of blue-green algae (fresh condition) the crop growth as evidenced by tillering and stand increased almost 100% over ammonium sulphate treatment at 30 kg./N. It is proposed in the coming seasons to apply nutrient mixture to the plots already rich in N fixing species to test whether the yields can be further enhanced.

# IV. Rôle of Blue-Green Algae in Combination with Conventional Fertilizers and Manures

(a) Role of blue-green algae compared with that of conventional fertilizers. The authors (1964 a) conducted a replicated field experiment during the main crop season of 1963 (July-December) with C.R. T141 to compare the efficacy of blue-green algae with conventional organic manures and fertilizers, viz., farm-yard manure, green manure (Sesbania speciosa), urea and ammonium sulphate at 20 kg. N/ha. A basal dressing of lime at 500 kg., superphosphate at  $20 \, \text{kg}$ .  $P_2O_5$  and sodium molybdate at  $0.28 \, \text{kg}$ . per hectare was applied. Four species of blue-green algae, Nostoc sphaearicum, N. amplissimum, Tolypothrix campylonemoides and Westiella sp. were used for field inoculation. Blue-green algae alone increased the grain yield significantly by about 30% over check plot and the response was statistically at par with 20 kg. N/ha. in the form of farm-yard manure, urea and ammonium sulphate. None of the fertilizers or manures enhanced the efficiency of the blue-green algae except urea. The application of green manure produced a response of 49% increase over control; this was significantly higher than that with blue green algae (Table VII).

The expression of algae in conjunction with organic manures and ammonium sulphate is not evident, presumably due to the fact that as nitrogen

TABLE VII

Iata (average of four replications) pertaining to the m

Yield and biometrical data (average of four replications) pertaining to the main crop season of 1963—Variety T. 141 (field experiment)

Treatment (over basal dressing of lime, super-	Grain (Kg.		Straw (Kg.			ight m.)	1	ve tillers hill
phosphate and sodium molybdate)	A	В	A	В	A	B	A	В
Check (No manure)	2615 (100)	3 <b>403</b> (130)	2496 (100)	3657 (147)	131·4 (100)	137·9 (105)	3·60 (100)	5·51 (153)
Farm-yard manure at 20 kg. N/ha.	3392 (130)	357 <b>3</b> (137)	3483 (140)	3657 (147)	137·6 (105)	139·2 (106)	<b>4·64</b> (129)	5·12 (142)
Green manure (Sesbania speciosa) at 20 kg. N/ha.	3907 (149)	3902 (149)	4238 (17 <b>0</b> )	4412 (177)	140·7 (107)	145•9 (110)	5·42 (151)	5·80 (161)
Ammonium sulphate at 20 kg. N/ha.	3431 (131)	<b>3</b> 472 (132)	35 <b>\$</b> 8 (144)	<b>3</b> 861 (155)	139·2 (106)	140·9 (107)	4·81 (134)	4·98 (138)
Urea at 20 kg. N/ha	3366 (129)	3585 (137)	3454 (138)	3715 (149)	136·0 (104)	141·2 (108)	4·74 (132)	4·93 (137)
C.D. (0.05) per hectare	21	.5	•	•	• •			•
C.D. (0·10) per hectare	29	)1		•	·• •	,	•	•

A—Without blue-green algae.

B-With blue-green algae.

Figures in brackets represent per cent. increases over control.

(After Subrahmanyan, Relwani and Manna, 1964).

in the form of ammonia is readily available for its growth, most forms do not fix atmospheric nitrogen. It may be noted here that ammonia is the key intermediate in nitrogen fixation by the blue-green algae (Fogg, 1963).

The significantly higher yield of crop with green manure may be attributed to the fact that though, on equal nitrogen basis, green manure produces similar yields as ammoniacal fertilizers (Nair, 1953) its efficacy is always more with superphosphate (Vivekanandan and Raja, 1964) and still more with the addition of lime which helps the mineralization process of green manure (Jochim *in* Mukherjee and Agarwal, 1950).

(b) Effect of blue-green algae in combination with urea.—Relwani and Manna (1964) studied the rôle of blue-green algae in combination with urea, another popular fertilizer for rice. Urea was applied at 10 kg. and 20 kg. N/ha. as soil application at transplanting and as foliar application 50 days after transplanting. A mixture of blue greens consisting of the same four

forms mentioned above was inoculated at transplanting. A basal dressing consisting of 500 kg. lime + superphosphate at 20 kg.  $P_2O_5 + 0.28 \text{ kg}$ . sodium molybdate per hectare was applied to each pot before planting the seedlings. The experiment was conducted during the main crop season of 1963 using popular rice variety C.R. T141 as test crop.

The application of blue-green algae alone to check pot accounted for an increase of 109% in grain yield. The increases resulted mainly from a higher number of ear-bearing tillers (43% over check). Soil application of urea at 10 and 20 kg. N/ha. produced 19.9 and 65.6% increases respectively over check pot and in combination with blue-green algae, the corresponding increases went up to 142.8% and 175.9%. In combination with urea spray, 50 days after transplanting, the additional responses were generally lower in magnitude (Table VIII). It is, therefore, evident that small soil application

TABLE VIII Effect of blue-green algae in combination with urea (1963 Main crop) (gm. per pot.—Average of three replications)

	. L	Average yield	l per pot (gr	<b>a.)</b> .				
Treatment	G	rain	Si	raw	No. of e	ear-bearing per pot		e length of micle
	A	В	A A	В	A	В	   A	В
	5·23 (IOO)	10·93 (209·0)	3·83 (100)	8·00 (208·9)	3·70 (100)	5·30 (143·2)	16.93	19.22
Urea at 10 kg. N/ha. (Soil application)	6·27 (119·9)	*12·70 (242·8)	4·9I (128·2)	9·10 (237·6)	3·30 (89·2)	6·00 (162·2)	18·17 (107·3)	(113 · 6
Urea at 20 kg. N/ha. (Soil application)	8 • 66 (165 • 6	14·43 (275·9)	6·33 ( <i>165</i> ·3)	11·80 (308·1)	5·00 (135·1)	8·00 (216·2)	I8·32 (108·2)	17.95
Urea at 10 kg. N/ha. (Foliar application)	5·73 (109·6)	*11·53 (220·5)	4·27 (111·5)	9·20 (240·2)	3·30 (89·2)	6·50 (175·7)	17-87	17·63
Urea at 20 kg. N/ha. (Foliar application)	4·90 (93·7)	13·03 (249·1)	4·77 (124·5)	9·55 (249·3)	3·30 (89·2)	6.00	(105·6) 18·28	(104·1) 19·13
A—Without blue	(117.8)	12·54 (239·8)	4·82 (125·8)	9·5 <b>3</b> (248·8)	3·73 (100·8)	6·36 (171·9)	(108•0) 17•91 (105•8)	(113·0) 18·67 (110·3)

A-Without blue-green algae. \* Varietal mixture was observed in these pots.

Figures in brackets represent treatment rating with check as 100, (After Relwani and Manna, 1964).

of urea in combination with blue-green algae can push up rice yields to appreciable extent.

## V. Response of Paddy Varieties and Methods of Propagation to Blue-Green Algae

In yet another experiment, Relwani (1965) investigated response of paddy varieties, viz., C.R. T141, GEB 24, FR 43B and J×I—431 and methods of propagation [seed and tiller (vegetatively) propagated] to blue-green algae under late transplanting conditions. A mixture of Anabaena, Nostoc, Tolypothrix, Scytonema and Westiella species was inoculated in the field. An average significant response of 13.8% to application of blue-green algae was obtained. The method of propagation of seedlings and the interactions algae×variety and algae×method of propagation were not found to be significant but variety×method of propagation interaction was found to be significant at 1% level of significance. Thus, the response to algae, although appears to diminish under late planting conditions, was found to be independent of variety or method of propagation (Table IX).

#### GENERAL CONCLUSIONS

The investigations so far made with blue-green algae lead to some interesting and useful conclusions:

- 1. Promotion of the growth of the indigenous flora is possible by supplying calcium, phosphates and trace element molybdenum and also by raising pH to alkaline level. This can be accomplished by application of lime, superphosphate and sodium molybdate. On a conservative estimate, this alone has accounted for 45% grain yield.
- 2. A few such applications, e.g., for three seasons as in our experiments, strengthens the blue-green algal flora in the field with beneficial results for the crop.
- 3. Introduction of nitrogen-fixing species further enhances the beneficial effects of blue-green algae. Yields up to 125% over control have been obtained in field trials.
- 4. The manuring schedule described here leads to the establishment of a good flora of nitrogen-fixing species; this is revealed by the periodical examination of the algal growth in the field. A list of such species is given in Table II,

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Table IX

Yield of paddy grain in kg. per hectare

V	ariety		Treatment	Seed crop	V.P. crop	Mean	Increase %	Variety mean
T. 141	••	••	No algae	3446	2729	3087	• •	
۰			Algae	3995	2911	3453	11.8*	3270
GEB 24	••	••	No algae	2566	3710	3138	• •	
			Algae	2906	4300	3603	14.8	3371
FR 43 B	• •		No algae	1073	1328	1200	••	-
		•	Algae	1331	1410	1370	14.2	1285
$J \times I-431$	••	· • •	No lagae	3263	2586	2925	• •	
			Algae	3701	2996	3349	14.5	3137
Method of (mean)	propag	ation	••	2785	2746	••	••	, .
Algae (mea	ın)	••	No algae	Al:	gae			
			2588	29	14			

V. P.=Vegetative propagation.

- 5. Such strengthening of the blue-green algal flora contributes to the enrichment of the soil is confirmed by the consistently good yields obtained for two seasons in succession when neither nutrient mixture nor algae were applied.
- 6. The algal mat left over in the field after the second such harvest would indicate that one or more crops could still be grown without application of nutrient mixture and algae.
- 7. It may be necessary to apply the nutrient mixture periodically to enable the algae to thrive and enrich the soil. This method of utilization

<sup>(1)</sup> C.D. (0.05) to compare any two varieties for a particular method of planting (seed or V.P. crop)—530 kg./ha.

<sup>(2)</sup> C.D. (0.05) to compare any two varieties—393 kg./ha.

<sup>(3)</sup> C.D. (0.05) to compare algae effects-192 kg./ha.

<sup>(4)</sup> Method of propagation—F test not significant. S. Ed.  $\pm$  132 kg./ha.

of blue-green algae appears to be an economical method to obtain increased yields.

- 8. This method appears to be an effective, more efficient and a suitable substitute for the popular ammonium sulphate. It may be mentioned in this connection that repeated application of ammonium sulphate has a tendency to deplete the soil of calcium. The deficiency of calcium, it may be pointed out, has a deleterious effect in the development and functioning of the root-hairs as has been found for other crops (Cormack et al., 1963); and this may apply to the rice plant as well; this probably accounts for the impoverishment of the soil when ammonium sulphate is repeatedly used in the absence of organic manures.
- 9. In the pot and field experiments, it has been noticed that the crop has a tendency to ripen earlier in instances where blue-green algae had been applied. Such observations as also superiority of a biological source of nitrogen have been recorded (Dorosinskii, 1962) for other crops and it is interesting to note that this applies to the rice crop as well.
- 10. These observations lead us to the hypothesis that besides supplying nitrogen fixed from the atmosphere, these algae which are known to liberate several extra-cellular substances, the nature of all of which is still unknown, also have in them certain growth-promoting substances which stimulate the rice crop. This finds confirmation in a recent field experiment in progress where within 10 days of the application of algae the crop showed vigorous growth.
- 11. Another interesting observation during the field experiments at CRRI and in a farmer's field was that the field, where the blue-green algae had been applied or their growth promoted, were comparatively free from the common rice field weeds mentioned in the text earlier. It would appear that the manuring schedule adopted as well as growth of blue-green algae act as a deleterious influence on the weeds concerned. It has also been recorded elsewhere that blue-green algae suppress the development of some saprophytic flora (Gaukhman and Rybov, 1962).
- 12. Experiments conducted to compare efficacy of blue-green algae with conventional fertilizers such as farm-yard manure, urea, green manure and ammonium sulphate showed that blue-green algae supply in the first crop season nitrogen up to 20 kg./ha. applied in terms of farm-yard manure, urea and ammonium sulphate. It would appear that green manure in the present experiment showed a better response because of lime and superphosphate

in the basal dressing. Soil application of urea in conjunction with bluegreen algae was better than foliar spray of urea.

- 13. Trials to assess effect of blue-green algae on different rice varieties as well as methods of propagation showed that under late planting conditions, though response tended to diminish, an average significant response of 13.8% to application of algae occurred and that response to algae was independent of variety and/or methods of propagation.
- 14. The experiments conducted at CRRI, the results achieved and the preliminary results of trials in progress in the different regions in the States, would indicate that the method adopted here could be employed profitably in regions where soil is not too acidic and where a certain amount of indigenous flora also exists. Trials with soils from several regions are in progress at present in this regard. Some preliminary results are embodied in another paper (Subrahmanyan et al., 1965). Blue-green algae not only serve to augment our nitrogen resources but also help to build up the fertility status of our impoverished soils considerably. The blue-green algae are a living green manure plus something for the rice crop.

### SUMMARY

The results of large-scale trials covering five crop seasons with blue-green algae to exploit their faculty to fix atmospheric nitrogen in the rice fields are described. The approach was to exploit the indigenous flora of the soil by conditioning the soil for their growth. Effect of inoculation of known nitrogen-fixing species has also been studied. Based on preliminary trials with various combinations of fertilizer mixtures, manures, with and without blue-greens and so on, a schedule of manuring has been arrived at to promote blue-green algal growth, viz., application of lime, superphosphate and sodium molybdate and this has been adopted in the experiments. Ammonium sulphate was used for comparison. The results show that blue-green algae can be used effectively and with considerable benefit for obtaining increased yields of rice and as a substitute for ammonium sulphate to which they are superior and they are as good as some of the other conventional manures and fertilizers. Their action is not related to variety of rice crop or methods of propagation. It has been found that the adoption of this schedule for three seasons leaves a beneficial residual effect on account of which two crops have been grown with the yield remaining consistent. Apart from this, it was noticed that the gross yield from the whole experimental area where blue-green algae had established themselves had gone up by over 20%, due, obviously, to the fertility build-up of the soil. The blue-green algae appear to function as a

living green manure, enriching the soil by their activity. Some interesting observations made during the trials are set out, viz., the possible presence of a growth-promoting substance in the extra-cellular products liberated by the blue-green algae and the almost total absence of common paddy field weeds in the plots with blue-green algae. It is suggested that the schedule adopted here could be profitably employed in certain regions of the country to increase rice production where the soil contains a good indigenous blue-green algal flora.

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