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The effect of straw application on chemical and microbiological properties of a wetland rice soil.

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Summary

A field experiment was conducted to study the effects of two methods of straw application on (1) the kinetics of chemical and electrochemical properties of the soil, soil solution and floodwater, (2) the bacterial populations in the rhizosphere and nonrhizosphere soil, plant roots, and decomposing straw, (3) algal populations, and (4) acetylene reduction activity (ARA) associated with soil and decomposing straw.

Straw application into the floodwater decreased O_2 concentration and pH in the floodwater for about 3 weeks. Incorporation of straw that had been predecomposed in the floodwater for three weeks kept pH and O_2 content of the floodwater lower for about 6 weeks after incorporation.

Straw amendments enhanced reduction of the puddled layer, resulting in higher concentrations of NH_4^+, Fe, P, Mg, Ca, Si, and HCO_3^- in soil solution.

Straw application significantly stimulated N_2-fixing and phototrophic bacteria. N_2-fixing BGA biomass was lower when straw was incorporated but higher when straw was surface applied after transplanting. Application of straw into the floodwater resulted in higher ARA. Phototrophic bacteria and BGA fixed more N_2 than heterotrophic bacteria.

Straw application increased total soil N. A flush of total N and NH_4^+ occured in all treatments after about 100 days but was highest when straw was applied into the floodwater.

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1. INTRODUCTION

Straw incorporation is an agricultural practice which has been primarily used to add nutrients and organic matter to the soil. Rice straw contains about 0.6% N, 0.1% P, 0.1% S, 1.5% K, 5% Si, and 40% C. Because it is available on site in amounts varying from 2 to 10 t/ha, it is a convenient source of plant nutrients. However, a survey by Tanaka (1973) indicated that straw incorporation is uncommon and that burning is the most frequent straw removal-use practice. Straw incorporation is widely practiced only in China and before the summer rice crop on Vietnam. Recycling of nutrients from rice straw may play an important role in maintening soil fertility and decreasing the need for chemical fertilizers.

Straw application in wetland soils leads to a burst of biochemical and microbiological activities; soil reduction and associated electrochemical changes, N immobilisation and N_2 fixation, production of organic acids and release of CO_2, CH_4, and H_2S (Yoshida 1981, Neue 1988). Straw has stimulatory effects on various groups of bacteria such as the N_2-fixing heterotrophs and the anoxygenic phototrophic purple-non sulfur bacteria (Ladha & Boonkerd, 1988). The average efficiency of heterotrophic N_2-fixation associated with straw is 2.4 mg N fixed per gram of substrate added, in about 1 month (Roger and Watanabe 1986). Straw application also reportedly favors photodependant N_2 fixation and blue-green algae (BGA) growth (Roger et al. 1982) but topdressing induced higher photodependant ARA and better rice growth than incorporation (Matsuguchi and Yoo 1981).

These processes directly and indirectly affect the availability and uptake of nutrients and therefore the value of straw as a source of nutrients for wetland rice. In a 5-year drum study with 3 soils, N increase from straw incorporation was computed to be 40 kg/ha per season, about 10 kg/ha per season more than the straw's N content. The extra N probably came from N fixation by heterotrophs and phototrophs (Ponnamperuma 1984).

On the other hand, straw incorporation might have detrimental effects. Adding straw to acid sulphate soils and degraded saline soils may favor formation of H_2S highly toxic to rice. Adding straw to neutral soils may cause net immobilization of N and induce Zn deficiency. There are reports on detrimental effect of straw incorporation on BNF in relation to the release of toxic organic acids and phenolic compounds (see Roger and Watanabe 1986).

The purpose of the experiment was to study the effect of two methods of straw application on:

(1) the kinetics of chemical and electrochemical properties of the soil, soil solution and floodwater;
(2) the bacterial populations and the diversity of the anoxygenic phototrophs in the rhizosphere and nonrhizosphere soil, plant roots, and decomposing straw;
(3) algal populations;
(4) acetylene reduction activity (ARA) associated with soil and decomposing straw.

In the context of a Saturday Seminar only part of the data are presented and discussed.

2. MATERIALS AND METHODS

2.1. Field layout

The experiment was conducted from April 1st to August 30 1986 (Table 1) at block H14 of the IRRI farm. Major chemical properties of the soil (Typic Tropaquept) are shown in Table 2. The experimental design was a complete randomized block of 16 plots (8.5 x 4.7 m) with four treatments and four blocks. Treatments were as follows:

Control 1 (C1) : No staw applied, 30 kg N/ha broadcast as urea at panicle initiation

Control 2 (C2) : No staw applied, N broadcast as urea at transplanting (50 kg/ha) and at panicle initiation (30 kg/ha). No measurement performed in this treatment except algal biomass, acetylene reducing activity, and yield.

Treatment 1 (T1): Surface application of straw before transplanting: 5 tons of chopped (10-15 cm) straw was surface applied in floodwater 21 days before transplanting and incorporated 2 days before transplanting. Thirty kg N/ha was broadcast as urea at panicle initiation.

Treatment 2 (T2): Surface application of straw after transplanting : 5 tons of whole length straw was applied three weeks after transplanting in parallel rows between rice hills. Thirty kg N/ha was broadcast as urea at panicle initiation.

IR29723-88-2-3-3, a 90 days variety, was used. Superphosphate was broadcast at 15 kg P/ha before transplanting and at10 kg P/ha at 21 DT . No potassium was applied. Root dipping in 2% ZnO_2 solution was done before transplanting. Insecticide was applied when needed. No herbicide was used, plots were handweeded when needed. 5-8 cm of floodwater was maintained but because of rain and maintainance problems, water level reached 10 cm in some occasions.

Daily values of air temperature (maximum, minimum, average), rainfall , solar radiation, and sunshine hours during the experiment were obtained from the climate unit at IRRI

2.2. Soil and straw sampling

Soil samples were collected at 0 - 2 cm and 2 - 10 cm depths using PVC tubes. The tubes had slots at 2 and 10 cm levels. A metal sheet was used to separate the two depths. Small holes were bored just above the two 10 cm mark to enable floodwater to pass through. The samples were transferred into plastic bags, kept in an ice box between sampling and analyses, mixed and analyzed. Chemical analysis were performed on wet soil samples except for N and P.

2.3. Electrochemical and chemical analyses

2.3.1. Soil

Soil pH at 2 cm and soil temperature at were recorded *in situ* at 06:00, 09:00, 12:00, 15:00 and 18:00 hrs. Plantinum electrodes were installed in the field at 5 and 10 cm to measure Eh. Soil pH and Eh were measured using digital pH meter (DIGI-SENSE) and pH DIGI 500 (WTW), respectively.

Total N was determined using a Digestion System 12 1009 Digester and a Tecator Kjeltec 1030 auto analyzer. Ammonium was extracted with 2N KCl and determined with the same auto analyzer.

Available (Olsen) P was extracted with $NaHCO_3$ and determined colorimetrically using the molybdate blue method (Black, 1965). Available Zn was estimated with 0.05 N HCl (Katyal and Ponnamperuma, 1974) and determined by atomic absorption spectrohotometry.

2.3.2. Soil solution

Piezometer tubes were installed in the puddled layer at 6-8 cm and in the traffic pan. Soil solutions were collected in Erlenmeyer flasks previously filled with N_2 and analyzed for ammonium, bicarbonate, P, Zn, Fe, Mg, Ca and Si. Bicarbonate was determined by a potentiometric titration of an aliquot with standard H_2SO_4 to pH 3.9 (IRRI, 1964). P was analyzed by a modification of the benzene-isobutanol extraction method. Iron, zinc, magnesium and calcium were analyzed by atomic absorption spectrometery. Silicon was analyzed colorimetrically using tartaric acid.

2.3.3. Floodwater

pH, and dissolved O_2 concentration in the floodwater were measured at 1-5 days intervals between 7 and 7.30 in the morning and between 1. 00 and 1. 30 in the afternoon. Four measurements were performed in each plot. Oxygen was measured with a YSI Oxymeter.

Floodwater was analyzed for ammonium, bicarbonate, P, Zn, Fe, Mg, Ca and Si using the same methods as for soil solution.

2.4. Microbiological methods

2.4.1. Sample preparation for microbial studies

Decomposing straw was sampled using a 19 x 19 cm metal frame at 1, 7, and 14 days after straw application (DASA).

Soil adhering to the roots of the rice plants was removed by slow squeezing the soil and roots and was referred as rhizospheric soil. The roots of the rice plant, were separated from the stem by cutting 5 mm below the basal region and cleaned with running water to remove any adhering soil particles as described by Watanabe (1979). The microorganism adhering to the roots were removed by shaking roots with glass beads as described by Rovira et al. (1974). The washed roots were further trimmed into small pieces (1 cm in length) and macerated in a Waring blender jar for 1.5 min.

2.4.2. Algal populations

Algal counts. Algal populations were estimated in Control 1 and Treatment 1 by plating ten-fold serial soil suspension-dilutions in triplicate onto agarized BG11 medium (Stanier et al. 1971) to enumerate total algae and onto BG11o medium (BG11 witout N) to enumerate N_2-fixing BGA. Plates were incubated for three weeks at 28 ± 2 ºC under continuous illumination with white fluorescent lamps (about 800 lux) before counting and identification of the colonies.

Direct measurement of the algal biomass.. Algae were collected at 42, 54, and 67 DT from 16 squares (20 x 20 cm) aligned on a transect through each plot. The 16 subsamples were combined and homogenized before measuring the dry weight, ash, C, N, and P contents.

2.4.3. Bacterial counts

Bacterial populations were enumerated using ten-fold serial dilutions prepared from soil, root and decomposing straw samples. Heterotrophic bacteria were enumerated on 0.1% Tryptic soy agar (TSA) by spread-plate technique; cellulolytic bacteria were counted by pour-plate method on 0.5% cellulose-agar; spore-forming anaerobic bacteria were counted by roll-tube technique of Hungate (1966) on glucose-anaerobic agar after thermal destruction of vegetative cells at 80 ° C for 10 min.; N_2-fixing bacteria were enumerated by most probable number (MPN) techniques using glucose-mineral semi-solid agar; nitrifying bacteria wrere enumerated by microplate technique of Rowe et al (1971) and phototrophic bacteria by the roll-tube technique of Hungate (1966) using PBM #2 medium. Heterotrophs were incubated in dark at 31 °C and phototrophs were incubated at 31 °C with fluorescent and incandescent light sources (6 Klux). Counting was made on the following days of incubation: 3 days for heterotrophs, 4 days for anaerobes, 6 days for phototrophs, 21 days for cellulolytic bacteria, and 30 days for nitrifying bacteria.

2.4.4. Bacterial cell volume and biomass measurement

Ten grams of moist soil was added to 90 ml of buffered water. The soil suspension was homogenised with Waring Blendor for 2 min. One hundreth millimeter of the suspension was spread over an area of 1 cm^2 on a microscopic slide. The smear was allowed to dry. The soil suspension and smears were prepared by the methods of Babuik & Paul (1970) and stainings were made with phenol-aniline blue as described by Jones & Mollison (1948). At least 50 random measurements per slide were made using a Olympus Microscope. Cell diameter was measured with a calibrated ocular micrometer and used to calculate the volumes of the cells, assuming a sphaerical shape for cocci and a cylindrical shape for rods. Bacterial biomass was derived from the biovolumes and counts.

2.4.5. Isolation and characterization of phototrophic bacteria.

Isolation. Phototrophic bacteria were isolated from roll-tube cultures under anaerobic condition. The isolates were purified by repeated isolations of single colony from agar plates of PBM #2 in anaerobic jar under an atmosphere of 90% H_2 + 10% CO_2 and phototrophic condition. The basal medium used was similar to that of PBM #2 except that the C sources were omitted.

Cultural characteristics. The isolates were characterized from the following characters: color of the photopigment; morphology of the cell and colony on agar plates and broth; presence of cytochrome oxidase, poteases, lipase; gelatin liquefaction; starch and cellulose hydrolysis; utilization of glucose, acetate, proponic acid, citrate, glycerol, tartrate, and malic acid as carbon sources, and arginine, aspartate and glutamate as nitrogen sources; and determination of maximum absorption of the photopigment of intact cells at 320 nm to 900 nm.

Absorption spectra of pigments . Bacteria grown on the plates under light and anaerobic condition were picked-up and inoculated to screw-cap test tubes filled up to the neck with PBM #2. Pure N_2 gas was flushed into the medium before and after inoculation. The cultures were incubated under light in an anaerobic jar with pure N_2 gas and allowed to grow for 6 days. The cells were washed and suspended in 30% bovine serum-albumin using the method of Sojka et al (19). The photopigment absorption spectra of the intact cells were measured by a Shimadzu UV-Visible 240 scanning spectrophotometer.

Cellular fatty acid analysis. Phototrophs were grown in liquid PBM #2 as described earlier. The cells were harvested by centrifugation, washed and lyophilized. Methylation, extraction, and separation of polar and non-polar fatty acids by thin-layer chromatography were carried out as described by Watanabe et al. (1987). Total fatty acid methyl esters, polar and non-polar methyl esters were analyzed by Hitachi Gas Chromatograph model 365-30 using 5% diethylene glycol-succinate (DEGS) PS on 100/120 "Supelcoport" under the following conditions: column temperature, 190 °C isothermal or 160 - 200 °C for gradient temperature ; injector and detector temperature, 250 °C and carrier gas, N_2 (40 mL/min flow rate for isothermal or 20 mL/min for gradient temperatures). Another column, Sp 2100 D0H in 100/120 "Supelcoport", was also used with the same condition as to the other column except for the maximum temperature was 240 °C for gradient and 230 °C for isothermal. Fatty acid methyl esters peaks were identified by comparing retention times with those of reference bacterial mixed standards (Supelco, U.S.A.) and individual authentic standards (Alltech, Inc., U.S.A.; Supelco, U.S.A. and Gasukuro Kogyo, Japan).

2.5. Acetylene reduction assays

2.5.1. ARA associated with straw

Acetylene reduction activities associated with decomposing straw were measured according to the method described by Ladha et al (1986a).

2.5.2. ARA of soil core samples

Ten soil-water cores were collected from each plot with glass tubes (1.8 cm in diameter, 10 cm in length). Floodwater (but not algal colonies suspended in it) was removed. Cores from each plot were enclosed in a perspex cylinder. The incubation, in an atmosphere of10% acetylene in air (V/V), was performed in a light chamber (20 klux, 24 ºC). ARA was calculated from the ratio of acetylene to ethylene measured by gas chromatography in 0.5ml gas samples collected from the cylinders after 45 and 135 min of incubation.

3. RESULTS AND DISCUSSION

3.1.Electrochemical analysis of floodwater and soil

3.1.1.Water pH and dissolved oxygen

Concentration of O_2 in the floodwater results from an equilibrium among production by the photosynthetic aquatic biomass (PAB), diffusion between air and water, and consumption by respiration and oxidation. As partial pressures of CO_2 and O_2 are inversely proportional, O_2 concentration and pH of the floodwater are positively correlated (cf. Roger and Kurihara, 1988). Therefore results regarding floodwater pH and O_2 concentration are presented simultaneously.

Range and general trends. The pH of the floodwater ranged from 6.7 to 8.4 in the morning (Fig 1) and from 7.6 to 10.4 in the afternoon (Fig 2). Oxygen content of the floodwater ranged from 2 to 7 ppm in the morning (Fig 3) and from 3 to 16 ppm in the afternoon (Fig 4). The general trend for both variables was a continuous decrease during the crop cycle as shown by a highly significant negative correlation between 1) pH and O_2, and 2) days after flooding (Table 3).

The study of the correlations between O_2, pH, and climatic parameters, showed no significant correlations except for maximum temperature. This indicates that the intensity of incident light was not the main limiting factor for photosynthetic activity in the floodwater.

Rainfal lowered the pH. The positive correlations observed with maximum temperature was mostly indirect, resulting from a strong negative correlation of O_2, pH, and maximum temperature with time (days after flooding)(Table 3 and 4).

We found higly significant positive correlations between pH and O_2 content of the floodwater (Fig 5). Regression curves for the four treatments did not significantly differ from each others indicating that the regression between pH and O_2 of the floodwater was mostly influenced by floodwater and soil properties rather than by the treatments and biological activities.

Effect of straw on pH and O_2 content of the floodwater. Straw application in the floodwater decrease O_2 concentration by 6-10 ppm (Fig. 6) and pH by about 1 unit (Fig.7) in T_1 and T_2.

In treatment T_2, where straw was not incorporated, pH and O_2 of the floodwater became similar to that in the control C_1 about 3 weeks after straw application.

In treatment T_1, where straw was incorporated 3 weeks after its application, O_2 concentration and pH remained lower (2-4 ppm and 0. 5-0.3 units, respectively) than in the control for about 2 months after incorporation. This prolonged decrease of O_2 and pH might have resulted first from the incorporation of the algal population into the soil together with straw. This was followed by a high grazer pressure that refrained algal growth. We observed that snails proliferated in plots when straw was applied and remained abundant after straw incorporation. The high ratio between snail and algal biomass after straw incorporation did not allow epipelic and epiphytic algae to multiply.

3.1.2. Soil pH and Eh

Straw treated plots had slightly lower pH at 6:00 a.m. (Fig. 8). In the afternoon soil pH was higher while soil Eh was lower (Fig. 9).
In general Eh values were lower with straw addition. The diurnal changes of soil pH and Eh are interrelated to floodwater properties and probably of soil fauna.

3.1.3. Correlation between water and surface soil pH

We found significant ($p< 2\%$) negative correlations in the morning and at noon between floodwater pH and the pH of the upper (0-2 cm) soil layer (Fig 10). High floodwater pH are caused a high consumption/extraction of CO_2 from the floodwater by algae. The diffusion rate of CO_2 from the soil

increases with decreasing partial pressure of CO_2 in the floodwater. Thereby the pH of the top soil decreases.

3.2. Soil chemical analysis

3.2.1. Nitrogen

Total soil N was higher in 0-2 cm and in straw treatment 2 plots (Fig. 11). Total N in straw treatment 1 plots remained approximately constant while in straw treatment 2 it gradually increased after straw application. Straw incorporated to rice fields enriches their N status because it stimulates activities of both heterotrophic and phototrophic N_2- fixing organisms (Matsuguchi, 1979, Rajarama Mohan Rao, 1976, Yoneyama et al, 1977, Ladha et al. 1986, 1987a).

Ammonium in soil increased to 27 mg 1^{-1} at 40 days after flooding (DAF) and declined (Fig. 12). A second peak was reached 79 DAF coinciding with urea fertilizer addition to plots. No net immobilization of N was detected but there was an increased release of ammonium at end of the season. Placing the straw for 3 weeks in the floodwater before incorporation might have caused both effects. The ammonium flush, however, occured too late to benefit the rice crop.

The pattern of exhangeable ammonium reflects the N-dynamic most clearly with peak concentrations after flooding, at panicle initiation due to topdressing and at ripening stage. High CEC and high plant uptake may explain why topdressing of urea had hardly any effect on ammonium concentration in the soil solution (Fig. 13).

Soil solution ammonium in S_1 increased to 1.4 $m1^{-1}$ 24 DAF while a maximum concentration of 1.1 $mg1^{-1}$ was attained in the control at 18 DAF (Fig. 13). Straw treated plots showed slightly higher ammonium concentration.

3.2.2. Phosphorus

Corresponding peaks at the time P fertilizers were added and the last peak at ripening stage were noted. Available P is slightly higher in top layer and in straw applied plots either incorporated or in surface (Fig. 14).

3.2.3. Iron

Iron reduction is enhanced by the addition of rice straw (Fig. 15). Concentrations of water soluble Fe were higher in straw treated plots, especially from 100 DAF onward coinciding with the maximum reduction.

3.2.4. Magnesium

Water soluble magnesium was initially high, gradually decreasing with time. Magnesium in soil solution and in floodwater was slightly higher in straw treated plots (Fig. 16).

3.2.5. Calcium

Water soluble calcium was initially high, decreased sharply 7 DAF, reached a peak coinciding with that for maximum reduction 100 DAF and levelled off. Calcium in soil solution and floodwater was slightly higher in straw treated plots (Fig. 17).

3.2.6. Silicon

Silicon in the soil solution sharply increased 14-21 DAF and decreased thereafter. Plots with straw had higher silicon in soil solution and floodwater (Fig. 18).

3.2.7. Bicarbonate

Bicarbonate reached its peak at 14-20 DAF, maintained a common level until 80 DAF (panicle initiation) and decreased (Fig. 19). The increase is due to $C0_2$ released by the anaerobic respiration of heterotrophic microorganisms owing to decomposition of organic matter. Straw plots exhibited slightly higher bicarbonate.

The following were highly correlated, showing interaction of soil solution and floodwater:

soil solution bicarbonate vs floodwater ammonium at C1
soil solution bicarbonate vs floodwater ammonium at S2
soil solution ammonium vs floodwater bicarbonate at S1

3.2.8. Phosphate and bicarbonate dynamics

The soil solution data were processed with the Equilibrium Geochemical Model (EGM). EGM is a speciation program that solves individual ion activities of soil solution components. EGM takes into account the effects of ion-pairing and temperature. EGM uses an interative technique to arrive at the activities of the derived species of a given set of components and uses the Davies equation to calculate for activity coefficients.

The dynamics of the different nutrients analyzed show similar trends regardless of treatment.

The dynamics of P in soil solution (Fig. 20) show that P recurringly increased and decreased throughout the flooding period. Soil solution phosphorus decreased at 21, 63, 91, 112 and 140 DAF after reaching maxima at 14, 56, 84, and 126 DAF. The maximum at 56 is due to fertilizer P application at 50 DAF. The soil solution was oversaturated with respect to apatite (calcium phosphate) at points of maxima. This means that at these periods, precipitation of apatite is possible.

Soil solution bicarbonate started with an initial high value, dipped suddenly 14 DAF and returned to a high value 21 DAF. From 21 DAF onwards, bicarbonate decreased with time (Fig. 19). Iron started with an initial low value and gradually increased with time (Fig. 15). The concentration of soil solution Fe is controlled by chelation-reduction and absorption-precipitation as siderite (iron carbonate). The concentation of bicarbonate is controlled by decomposition processes, diffusion, plant uptake, and precipitation of carbonates.

3.3. Algal population and biomass

Because of the methods of sampling adopted, plate counts mostly record the algae growing at the soil-water interface (epipelic algae) and the algal propagules in the soil, while direct biomass measurements record floating algae.

Visual and microscopic observation showed that algal biomass during the whole experiment was largely dominated by a unicellular mucilaginous N_2-fixing BGA belonging to the genus *Aphanothece*. Major characteristics of this unique N_2-fixing genus are :

- a very low and variable dry matter content that can be lower than 1%,
- a high ash content as most field-grown mucilaginous BGA,
- a low N content,
- a quite unpredictable N_2-fixing activity because this genus, which has no heterocysts, alternates periods of photosynthesis and N_2-fixing activities, and
- resistance to grazing due to the mucilaginous nature of its large colonies.

During the 2-3 first weeks of the experiment, sparse colonies of *Aphanothece* were observed in most of the plots but no visible algal bloom developed. Algal biomass estimated at 67, 79, and 92 DAF showed a large variability among treatments and within replicates, with values ranging from 4 to about 500 kg dw ha^{-1}. Average values (Table 5) show a consistently lower biomass in plots where straw was incorporated (T_1). The highest average biomass (18 t f.w. ha^{-1} or 291 kg d.w. ha^{-1}) was

recorded at 67 DAF (42 DT), in plots where straw had been surface applied (T_2). This value was not significantly different from that in the control (C_1) (188 kg d.w. ha^{-1}).

However, when interpreting the data, the high inter-plot variability should be considered. Table 6 gives an example of this variability and shows that the relatively large average biomass reported for C_1 resulted from an exceptionally high biomass in one of the four replicates. On the contrary, algal biomasses in plots where straw had been surface applied (T_2) had relatively consistent values with a coefficient of variation of about 30% as compared with coefficients of about 100% or more in the other treatments. This may indicate a beneficial effect of surface application of straw on the development of blooms of N_2-fixing BGA. However, the first straw application (T1) did not had such an effect.

None of the treatments had a significant effect on the chemical composition of the *Gloeotrichia* bloom (Table 7) but composition changed during the cropping season. Carbon and N content increased, probably because of an higher pigment content under a denser canopy. Phosphorus content decreased from 0.24% to 0.14% in about one month.

Algae counts in C_1 and T_1 are presented in Fig. 21. No significant variations of heterocystous BGA were observed during the crop cycle, indicating that this counts corresponded mostly to spores or propagules of heterocystous BGA present in the soil. This agree with the observation that unicellular BGA were dominant when blooms were present.
Total algal population started to decrease in T_1 before the incorporation of straw. This probably partly resulted from an increasing grazer population of snails due to straw application. An increase in total algae was observed in both C1 and T1 after the second urea application.

3.4. Bacterial populations and biomass

3.4.1. Bacterial counts in decomposing straw and soil

Viable counts of different physiological groups of bacteria are shown in Table 8. Heterotrophs, cellulolytic, anaerobic-sporeforming and nitrifying bacteria showed no significant difference between control and straw amended soil, while N_2-fixing bacteria and phototrophic bacteria were significantly ($P < .05$) higher in the soil with straw amendment than in control. The viable counts of various physiological group of bacteria between surface and sub-surface layers were almost the same, irrespective of the treatment. Heterotrophs and cellulolytic bacteria were significantly ($P < .05$) higher in the decomposing straw than in the soil, while counts of N_2-fixing and anerobic sporeforming bacteria

showed no significant differences between soil and decomposing straw. The probable reason for the higher bacterial counts of various physiological group of bacteria in decomposing straw than in the soil may be that the nutrients released from decomposing straw were easily accessible to the microorganisms.

3.4.2. Soil bacterial biomass

The measurement of bacterial biomass were based on the direct microscopic counts and determination of the biovolume of the bacteria. The conversion of the biovolume to the biomass was based on average values of 1.09 g/ cm^3 and 30% as bouyant density and dry matter, respectively, as proposed by Bakken & Olsen (1983). The total bacterial biomass measured in surface and sub-surface soils from different treatments are shown in Table 9 It was significantly higher in surface soil of straw treatment than in control and straw treatment 2. The increase in biomass of treatment 1 can be attributed to the incorporation of the straw rather than surface application. Incorporating the straw at 21 days after straw application (DASA) increased the biovolume of the rod-shaped bacteria significantly than the cocci-shaped bacteria (data not shown). The rod-shaped bacteria constituted an average of 30% of the total bacterial population in soil of straw treatment 1, while 12 and 18% bacteria in control and treatment 2, respectively. The higher concentration of rod-shaped bacteria in straw treatment 1 is probably due to the incorporation. In sub-surface soil, the bacterial biomass in straw treatment 2 was lower than in straw treatment 1 and control. It is already known that the paddy soil amended with organic manure had higher bacterial biomass than without organic manure (Hasebe, 1984).

The bacterial biomass-C, N and P was calculated based on the assumption that the carbon, nitrogen, and phosphorus of bacterial cell was 50, 15 and 3.2% of the dry matter, respectively (Luria, 1960). The calculated bacterial biomass-C, N and P in surface and sub-surface soil is shown in Table 9 The bacterial biomass-C, N and P were higher in straw treatment 1 than in control and straw treatment 2 in the surface soil. In sub-surface soil, bacterial biomass-C, N and P values were similar to control and straw treatment 1. Unexpectedly, lower values of bactrial biomass-C, N and P was noted in straw treatment 2.

3.5. Phototrophic bacteria

It was earlier believed that phototrophic bacteria play a minor role in N_2-fixation in wetland rice soils (Roger and Watanabe 1986). However, recently this potential has been reevaluated (Ladha and Boonkerd 1988).

Because of the high populations of phototrophic bacteria recorded in straw treated plots and their possible contribution to N_2-fixation, special attention was paid to this group of microorganism.

3.5.1. Phototrophs in rhizosphere and root of rice

The populations of heterotrophs and phototrophs in rhizosphere soil and rice roots are shown in Table 10 . Straw treatment did not significantly increase the viable count of heterotrophs in the rhizosphere soil, while the phototrophic counts were higher in straw 1 treatment than in control and straw 2 treatment. We can not ascertained if straw or the exudates from the plants influenced the proliferation of the phototrophs in the rhizosphere. High population of phototrophic bacteria were fond in rice roots, however there was no significant effect due to straw application. There does not seem to be any published information on the observation of phototrophic bacteria with rice roots.

3.5.2. Phototrophic bacteria and environmental variables

Environmental factors, such as temperature, dissolved oxygen, pH of the floodwater and solar radiation play important roles for the survival and growth of the phototrophic bacteria. The increase in temperature usually enhanced the decomposition rate of straw which may increase the populations of bacteria. Surface application of straw on the floodwater markedly depressed the concentration of disolved O_2 which favors the proliferation of phototrophic bacteria.

Solar radiation an important environmental variable affect significantly the growth of phototrophic bacteria in the aquatic environment (Caldwell & Tiidjie 1975; Parkein & Brook, 1980; Truper and Genovese. 1968, Alio et al 1983 and Montesinos et al 1983). Although the phototrophic bacterial counts did not significantly correlate with the solar radiation when straw was not applied but there was a significant correlations in straw applied treatment (Fig 22). There is a probable interrelationship between solar radiation, temperature and presence of straw for the proliferation of phototrophic bacteria. When light intensity increases, temperature also increases, which enhances straw decomposition and the release of substrates that permits the increase of phototrophic bacterial population.

3.5.3. Phototrophic bacterial diversity

Phenotypes. Ninety six strains of phototrophic bacteria were isolated from decomposing straw, non-rhizosphere and rhizosphere soil,

and plant roots. Sixty three morphological, cultural, physiological and biochemical characters were used in calculating the Euclidean distance coefficients between strains and strains were clustered by unweighted-pair group method with arithmetic average (UPGMA). The Euclidean distance values were transformed into percentages of similarity. Using the similarity coefficient of 0.80 in the phenogram, 18 phena, comprised from 2 to 16 strains, were formed (Fig. 23) .

The common features of the strains was that all were Gram-negative, positive for catalase and cytochrome oxidase, unable to grow in alkaline pH, with 0.5% NaCl , cellulase negative, and all contained bacteriochlorophyll (Bchl) a (Fig 24) which is one of the important characteristics of photosynthetic purple non-sulfur bacteria.

Phena 2, 3, 4, 7 and 8 were the major ones and comprised of 6 to 16 representative strains.

Based on the abilities of strains to utilize different C and N sources, and maximum absorption spectra, the strains of phenon numbers 2, 4 and 7 have been tentatively assigned to *Rhodopseudomonas acidophila*, *Rhodocuclus gelatinosus* and *Rhodopseudomonas palustris* respectively. Strains belonging to other minor groups haveing different phenotypic features could not be assigned to other known photosynthetic purple non-sulfur bacteria and need further investigations.

Cellular fatty acid composition. The cellular fatty acid (CFA) composition has been successfully used in the identification of several Gram-positive as well as Gram-negative bacteria (Kumagata and Suzuki 1987). We employed this characteristic to identify several of our strains representing different phenon (Table 11). The classification of photosynthetic purple non-sulfur bacteria is based on 3 major types of CFA, such as type A (consists of C16:0 and C16:1), type B (C18:1) and type C (consists of C16:0, C16:1 and C18:1) and the presence of 3-hydroxy fatty acid (Urakami and Komagata, 1988). Type A fatty acid (Fig. 25) predominated in our strains of all the phena while type B (Fig. 26) in phena 5 and 3 and type C (Fig. 27) in phena 2,10,13 and 16. Based on CFA composition, some strains belonging to phena 2,4 and 13 have been tentatively assigned to *Rhodopseudomonas palustris, Rhodocuclus gelatinosus* and *Rhodospirillum rubrum* respectively. The strains of phena 3 contain 3-OH C10:0 (Fig. 28) which no known species has been reported to have and therefore appear to be a new species of *Rhodopseudomonas.*

3.6. Acetylene-reduction activity

3.6.1. ARA and N_2-fixing organisms associated with straw

The heterotrophic bacterial count on field-dried straw prior to application was 1.6 x 10^3 colony forming unit (CFU)/g and increased about 6-fold in decomposing straw sampled at 1 DASA (Fig 29). Phototrophic bacteria were not found in dried straw up to 10^{-2} dilutions but were found at an average density of1.3 x 10^6 CFU/ g straw at 1 DASA. Heterotrophs and phototrophs further increased at 7 DASA and sustained at that level up to 14 DASA.

Counts of algae epiphytic on decomposing straw ranged from 10^6 to 10^8 CFU g^{-1} d.w (Table 12). Such values are high as compared with values observed for soil (about 10^5). Whereas unicellular N_2-fixing BGA were dominant in the water, heterocystous forms, especially *Nostoc*, were dominant on straw. In T_1, heterocystous BGA on straw increased more than 10-fold during the second week following straw application. In T_2 they increased by 5 times in the corresponding period and were significantly higher than in T_1. Several factors including competition between non fixing and N_2-fixing forms, light intensity, and P availability could explain the larger abundance of epiphytic BGA on straw in T_2, but no clear evidence is available.

ARA associated with decomposing straw were much higher under light than under dark conditions (Fig. 29). Activities were higher at 7 DASA than at 14 DASA. Straw treatment 1 exhibited higher ARA than straw treatment 2. The higher ARA under light was associated with an increase in the population of phototrophic bacteria. The synergistic effect of N_2 fixation activities of heterotrophs and phototrophs are one possible reason for the marked increase in ARA. The mixed culture of photosynthetic bacteria (*Rhodopseudomonas capsulatus*) and N_2-fixing heterotrophs (*Azotobacter vinelandii*) are reported to fix significantly more N_2 than the pure culture (Okuda 1959). Likewise, the phototrophic bacteria can fix N_2 more effectively under light than under dark conditions.

The high ARA of decomposing straw under light was also partly due to oxygenic phototophs (blue-green algae) as shown by increasing populations of BGA epiphytic on straw (Table 12). However counts of BGA indicate that contribution of BGA might have been much lower in straw treatment T_1 than in straw treatment 2.

3.6.2. ARA of soil cores

Because small tubes were used to sample it was not possible to include straw pieces in the sampling in T2. Total ARA in T_2 is therefore the sum of the activity measured separately on soil+water cores (Fig 30) and straw .

Average ARA during the cropping season was about 250 µmol $C_2H_2 \cdot m^{-2} \cdot h^{-1}$ in T_2, 190 µmol $C_2H_2 \cdot m^{-2} \cdot h^{-1}$ in C_1, and 60µmol $C_2H_2 \cdot m^{-2} \cdot h^{-1}$ in C_2 and T_1 . Nitrogen application at panicle initiation inhibited ARA in all the treatments. The higher average photodependant ARA observed in C1 and T_2 is in agreement with the increase in soil N observed during the second half of the experiment in the upper soil layer of C_1 and T_2, but not in T_1 (Fig 30). Both N content of upper soil layer and ARA were higher in T_2 than in C_1.

BGA biomass and ARA measurements were highly correlated ($p < 0.01$). The coefficient of correlation between ARA and biomass expressed as kg P ha^{-1} was higher than that between ARA and biomass expressed as fw, dw, C, or N ha^{-1}(Table 13). This together with low P contents measured in the algal biomass confirms that P is a major limiting factor for BNF by BGA.

Specific ARA ranged from 0 to 10 nanomoles acetylene mg $protein^{-1}$ $hour^{-1}$. A general decrease of the specific ARA was observed at high biomass (Figure 31).

4. CONCLUSIONS

- Straw application into the floodwater decreased O_2 concentration and pH in the floodwater for about 3 weeks. Incorporation of straw that had been predecomposed in the floodwater for three weeks kept pH and O_2 content of the floodwater lower for about 6 weeks after incorporation.
- Straw amendments enhanced reduction of the puddled layer, resulting in higher concentrations of NH_4^+, Fe, P, Mg, Ca, Si, and HCO_3^- in soil solution.
- Algal biomass was lower when straw was incorporated but higher when straw was surface applied after transplanting. In all treatments, algal biomass was dominated by *Aphanothece* , a unicellular N_2-fixing BGA. Straw application had no effect on the chemical composition of the algal biomass.
- Straw application significantly stimulated N_2-fixing and phototrophic bacteria. Bacterial biomass was significantly higher in straw treatment 1 than in treatment 2.
- Ninety six strains of phototrophic bacteria were isolated from decomposing straw, non-rhizosphere and rhizosphere soil, and plant roots. A new species of *Rhodopseudomonas* was identified.

- Application of straw into the floodwater resulted in higher ARA. Phototrophic bacteria and BGA fixed more N_2 than heterotrophic bacteria. Urea application inhibited phototrophic ARA in all treatments.
- Straw application increased total soil N. When straw was applied to the floodwater, the increase was mainly due to increased algal biomass. When the straw was incorporated, the increase was mainly due to bacterial biomass.
- Application of straw into the floodwater prior to incorporation prevented possible net immobilization of N. A flush of total N and NH_4^+ occured in all treatments but was highest when straw was applied into the floodwater (T_2).
- Floodwater and soil electrochemical characteristics were interlinked. Both showed diurnal variations, though they were more prononced in the floodwater.

5. REFERENCES

Alio, C. P., Montesinos, E. & Guerrero, R. 1983. Factors determining annual changes in Holomictic lake Cisio, Spain. Appl. Environ Microbiol 26:999-1006.

Bakken, L. and Olsen, R. A. 1983. Bouyant densities and dry matter content of microorganisms: Conversion of a measured biovolume in biomass. Appl. Environ. Microbiol.45:118.

Black, C. A., Evans, D. D., White, J. L., Ensminger, L. E. & Clark,F. E. 1965. Methods of soil analysis. Part 2. American Society of Agronomy. Madison, Wisconsin, USA.

Caldwell, D. E. and Tiidjie, J. M. 1975. The structure of anaerobic bacterial communities in the hypolimnia of several Michigan lakes. Can J. Microbiol. 21:377-385.

Hasebe, A., Kanazawa, S. and Takai, Y. 1984. Microbial biomass in paddy soil. I. Microbial biomass calculated from direct count using fluorescence microscope. Soil Sci. Plant Nutr. 30:175-187.

Hungate, R E 1966 The rumen and its microbes. Academic Press New York.

International Rice Research Institute (IRRI) 1964. Annual Report for 1963. Los Banos, Laguna.

Jones, P.C.T. and Mollison, J.E. 1948. A technique for the quantitative estimation of soil microorganism. J. Gen. Microbiol. 2:54-69.

Kato, S., Urakami, T. and Komagata, K.1985. Quinone systems and cellular fatty acid composition in speceis of *Rhodopsirillaceae* . J. Gen. Appl. Microbiol. 31:381-398.

Katyal, J. C. & Ponnamperuma, F.N. 1974. Zinc deficiency: A widespread nutritional disorder of rice in Agusan del Norte. Phil. Agriculturist 58:78-89.

Kumagata, K. and Suzuki K. (1987) Lipid and cell wall analysis in bacterial systematics pages 161-209 *in* Methods in microbiology R. R. Colwell and R. Grigorova eds. Vol 19 A P

Ladha, J. K. & Boonkerd, N. 1988. Biological nitrogen-fixation by heterotrophic and phototrophic bacteia in association with straw. First International Symposium on Paddy Soil Fertility, Chaing-Mai, Thailand, Dec. 6-13, pp.173-187.

Ladha, J.K., Padre, A. T., Daroy, M. L.. G, Punzalan, G and Watanabe, I. 1987a. The effect on nitrogen fixation (C_2H_2 reduction), bacterial population and rice plant growth of two modes of straw application to a wetland rice field. Biol Fertil Soils 5:106-111.

Ladha, J. K., So, R. B. and Watanabe, I. 1987. Composition of *Azospirillum* species associated with wetland rice grown in different soil. Plant & Soil 102: 127-129.

Ladha, J.K., Tirol, A. C,. Daroy, M. L. G., Caldo, G., Ventura, W. and Watanabe, I. 1986a. Plant-associated nitrogen fixation (C_2H_2-reduction) by five varieties and relationship with plant growth characters as affected by straw incorporation Soil Sci Plant Nutr 32:91-106.

Matsuguchi, T. 1979. Factors affecting heterotrophic nitrogen fixation in submerged rice soils. in Nitrogen and Rice. The International Rice Research Institute. Los Banos, Laguna, Philippines pp. 465-484.

Matsuguchi T and Yoo (1981) Stimulation of phototrophic N2 fixation in paddy fields through rice straw application, pp 18-25, In Nitrogen cycling in South-East Asian Wet Monsoonal Ecosystems, Wetselaar R, Simpson JR and Rosswall T eds, Canberra, Austral Acad Sci.

Montesinos, E., Guerrero, R., Abella, C. & Esteve, I. 1983. Ecology and physiology of the competition for light between *Chlorobium limnicola* and *Chlorobium phaeobacteriodes* in natural habitats. Appl Environ Microbiol 46:1007-101.

Neue, H. U. 1988. Holistic view of chemistry of flooded soil. First International Symposium on Paddy Soil Fertility, Chaing-Mai, Thailand, Dec. 6-13, pp.21-53.

Okuda, A., Yamaguchi, M & Kobayash, M. 1956. Nitrogen-fixing microorganisms in paddy soils (Part 3). Distribution of non-sulfur photosynthetic bacteria in paddy soils. Soil & Plant Food 2:131-133.

Parkin, T. B. and Brock, T. D. 1980 The effects of light quality on the growth of phototropic bacteria in lakes. Arch Microbiol 125:19-27.

Ponnamperuma, F. N. 1984. Straw as source of nutrients for wetland rice. In Organic matter and rice. International Rice Research Institute, pp. 117-136.

Rao, V. R. 1976. Nitrogen fixation as influenced by moisture content, ammonium sulfate and organic sources in a paddy soil. Soil Biol Biochem 8:445-448.

Roger PA, Tirol A, Grant I and Watanabe I (1982) Effect of surface application of straw on phototrophic nitrogen fixation. Int Rice Res Newsl 7(3), 16-17

Roger PA, Watanabe I (1986) Technologies for utilizing biological nitrogen fixation in lowland rice : potentialities, current usage, and limiting factors. Fertilizer Research 9 : 39-77.

Rovira, A D, Newman, E. I., Bowen, H. J. and Campbell, R. 1974 Quantitative assesment of the rhizosphere microflora by direct microscopy. Soil Biol Biochem 6:211-216.

Rowe, R., Todd, R. & Waide, J. 1977. Microtechnique for Most-Probable Number analysis. Appl. Environ. Microbiol. 33: 675-680.

Sojka, G. A., H. H. Freeze and H. Gest. 1970. Quantitative estimation of bacteriochlorophyll in situ. Arch. Biochem. Biophys. 136:578-580.

Suzuki, K. & Komagata, K.. 1983. Taxonomic significance of cellular fatty acid composition in some coryneform bacteria. Int. J. Syst. Bacteriol. 33:188-200.

Tanaka A (1973) Methods of handling the rice straw in various countries. Int Rice Comm Newsl 22(2), I-20

Urakami, T. & Komagata, K. 1983. Grouping of *Pseudomonas* species in the basis of cellular fatty acid composition and their quinone system with special reference on the existence of 3-hydroxy fatty acids. J. Gen. Appl. Microbiol. 29:17-40.

Urakami, T. and Komagata, K. 1988. Cellular fatty acid composition with special reference to the existence of hydroxy fatty acids and the occurrence of squalese and sterols in species of *Rhodospirilliaceae* genera and *Erythrobacter longus.* J. Gen. Appl. Microbiol. 34:67-84.

Watanabe, I., Barraquio, W. L., Guzman M. de & Cabrera, D. 1979. Nitrogen-fixing (acetylene reduction) activity and population of aerobic heterotrophic nitrogen-fixing bacteria associated with wetland rice. Appl. Environ. Microbiol. 37:813-819.

Watanabe, I., So, R, Ladha, J.K., Fujimura, Y.K & Kuraishi, H. 1987. A new nirogen-fixing species of pseudomonad. *Pseudomonas diazotrophicus* sp. nov. isolated from the root of wetland rice. Can J. Microbiol. 33:670-678.

Yoneyama, T. , K. K. Lee, T. Yoshida. 1977. Decomposition of rice residues in tropical soils. Soil Sci. Plant Nutr. 23:287-295.

Yoshida, S. 1981. Fundamentals of rice crop science. International Rice Research Institute. Los Banos, Laguna, Philippines. p. 269.

Table 1 : Calendar of agricultural practices and stages of growth during DS 1986.

March 25 (0 DAF)	Plowing
April 1st	Harrowing. Initial soil sampling. Application of straw at 5 t/ha in S1
April 23rd	Straw incorporation in S1
April 24th	15 kg P_2O_5/ha broadcast in all plots. Urea broadcast at 50 kg N/ha in C2
April 25th (0 DT)	Transplanting in all plots
April 29th	Diagran added in all plots
May 19th	10 kg P_2O_5 applied in all plots
May 21th	Straw surface applied at 5 t/ha in S2
June 19th	Urea broadcast at 50 kg N/ha in all plots
June 20th	Panicle initiation
July 2nd	Heading
August 15th	Irrigation stopped (but it was raining)
August 25th	Harvest

Table 2. Major physical and chemical properties of the soil at Block H14

pH	6.7
Total N (%)	0.127
Exchangeable K (mmol kg^{-1})	10.06
Exchangeable Mg (mmol kg^{-1})	67.5
Exchangeable Ca (mmol kg^{-1})	94.5
CEC (mmol kg^{-1})	357
Olsen P (mg l^{-1})	8.1
Active Fe (%)	3.48
Active Mn (%)	0.189
Organic C (%)	1.41

Table 3. Correlation between climatic factors, and pH and O_2 content of the floodwater*.

	Days after flooding	Rainfal	Solar radiation	Sun hours	Maximum temperature	Minimum temperature
O2 morning	-(0.01)	ns	ns	ns	+(0.1)	ns
O2 noon	-(0.01)	ns	ns	ns	+(0.01)	ns
pH morning	-(0.01)	-(0.05)	ns	ns	+(0.01)	ns
pH noon	-(0.01)	-(0.1)	ns	+(0.1)	+(0.01)	ns

* The level of significance of the correlation is indicated in parenthesis. Levels > 0.1 are not presented

Table 4. Cross correlations between climatic parameters

	Days	Rainfal	Solar Rad.	Sun hours	Max temp	Min temp	Aver temp.
Days	1.000	0.366	-0.346	-0.406	-0.596	0.013	-0.557
Rainfal	0.366	1.000	-0.266	-0.355	-0.397	-0.061	-0.395
Solar Rad.	-0.346	-0.266	1.000	0.897	0.623	-0.501	0.354
Sun hours	-0.406	-0.355	0.897	1.000	0.586	-0.509	0.317
Max temp	-0.596	-0.397	0.623	0.586	1.000	-0.112	0.887
Min temp	0.013	-0.061	-0.501	-0.509	-0.112	1.000	0.358
Aver temp	-0.557	-0.395	0.354	0.317	0.887	0.358	1.000

Table 5. Effect of straw application on algal biomass (kg ha^{-1}) [a]

DAF[1]	Control 1	Control 2	Straw incorp.	Straw surface
67	188 ab	62 ab	23 b	291 a
79	123 ab	130 a	31 b	69 ab
92	39 ab	13 b	4 b	68 a
Average	120 ab	66 ab	19 b	142 a

[a] average of 4 replicates. [b] days after flooding.

Table 6. Inter plot variability of algal biomass 67 days after flooding.

Treat.	Biomass	Rep. 1	Rep.2	Rep.3	Rep.4	Mean	Stdev	C.V.
C1	f. w. (t/ha)	41	9	5	5	15	17	116
C1	d. w. (kg/ha)	463	122	73	93	188	184	98
C2	f. w. (t/ha)	11	0	5	0.1	4	5	132
C2	d. w. (kg/ha)	207	0	41	0	62	99	159
T1	f. w. (t/ha)	3	0	7	0.5	3	3	118
T1	d. w. (kg/ha)	12	0	69	12	23	31	133
T2	f. w. (t/ha)	24	13	14	23	19	6	30
T2	d. w. (kg/ha)	317	171	313	365	291	84	29

Table 7: Effect of the treatments and time of sampling on the composition of the algal biomass (*Aphanothece* sp. dominant).

	C%			N%			P%		
Days after flooding	67	79	92	67	79	92	67	79	92
Control 1[a] (0 N + 30 N)	30.1a	34.8a	nd	3.50a	4.08a	nd	0.24a	0.16	nd
Control 2 (50 N + 30 N)	29.7a	35.8a	35.5a	3.01a	3.85a	4.54a	0.24a	0.17a	0.14a
Straw 1 (incorporated)	34.2a	35.5a	36.6a	3.96a	3.82a	4.79a	0.26a	0.15a	0.14a
Straw 2 (surface applied)	30.8a	34.6a	nd	2.44a	3.50a	nd	0.23a	0.23a	0.15a
Average[b]	31.7b	35.3ab	36a	3.38b	3.79b	4.66a	0.24a	0.16b	0.14b

[a] Values on the same column followed by the same letter are not significantly different.

[b] For each set of data, values followed by the same letter are not significantly different.

Table 8. The viable counts of various physiological types of bacteria in soil and decomposing straw.

Depth/treatment	Bacterial viable count (Log_{10} CFU/gsoil)					
	Heterotrophs	Cellulolytic	Nitrogen fixing	Anaerobe Sporeformer	Nitrifying	Phototroph
Surface soil (0-2 cm)						
Control	7.50	5.231	4.58	5.43	3.16	2.99
Treatment 1	7.58(10.21)	5.26(7.59)	5.15**(5.76)	5.42(5.76)	3.02	3.39**
Treatment 2	7.52(11.03*)	5.35(9.93*)	5.09**(5.17)	5.45(6.00)	3.10	3.37**
Sub-surface (2-10 cm)						
Control	7.39	5.27	4.86	5.03	3.05	
Treatment 1	7.48	5.29	5.29**	5.4	3.06	
Treatment 2	7.55	5.24	5.04**	5.37	3.07	

Statistical signifance of difference between control and treatment, by t test; $P < .01$**; $P < .05$*
Figures in parenthesis are mean viable counts of decomposing straw (Log_{10} CFU/g. straw).

Table 9. Bacterial biomass of wetland rice soil with and without straw.

Depth/Treatment	Bacterial biomass (mg/100 g soil)			
	Total	Carbon	Nitrogen	Phosphorus
Surface soil (0-2 cm)				
Control	307 ± 15.9	154	46	9.8
Treatment 1	423 ± 32.00	212	63	13.5
Treatment 2	315 ± 60.01	158	47	10.1
Sub-surface soil (2-10 cm)				
Control	214 ± 39.93	107	32	6.8
Treatment 1	230 ± 27.30	115	34	7.4
Treatment 2	145 ± 12.80	73	22	4.6

Table 10. Counts of heterotrophic and phototrophic bacteria in rhizosphere soil and rice roots.

Sampling site/Treatment	Bacterial viable counts ($\log_{10}$ CFU g.soil)	
	Heterotroph	Phototroph
Rhizosphere		
Control	7.48	4.85
Treatment 1	7.45	5.79*
Treatment 2	7.54	5.30
Root		
Control	9.28	6.04
Treatment 1	9.01	6.45
Treatment 2	9.03	6.75

Statistical significance of difference between control and treatments, by t test; $P < .05$*

Table 11. Fatty acid composition of selected strain of phototrophic bacteria.

Strain	Phenon	Straight Chain Fatty Acids											Cyclo-propane acid	3-hydroxy fatty acid			Unknown	Fatty acid type
		12:0	12:1	14:0	14:1	15:0	16:0	16:1	17:0	17:1	18:0	18:1	19:0	10:0	12:0	14:0	16:0	
S1-10	1	3.8[a]	t	7.8	t	t	38.9	40.0			0.6	6.8					0.6		A
R3-1	2		0.6	0.8		t	19.3	7.5	t		3.7	66.1						0.95	C
S1-2	3	3.3	t	5.1	t	t	35.7	37.1	1		t	16.3				t	t		A
RH1-2	3	4.1	t	4.7	t	1.0	40.7	38.1	1	t	2.7	6.7					t		A
RH1-12	3	4.5	t	8.3	t	0.6	37.9	42.2			t	5.0							A
S1-4	4	3.5	t	6.0	t	1.1	39.9	39.9	t	t	2.2	6.1							A
RH1-9	4		0.9	1.3		0.5	23.1	7.8	t		3.9	60.1				t		0.88	B
R1-5	5	t	0.8	t		t	21.6	6.7	t		5.3	62.6				t			B
R3-5	5	t	t	t	0.5		24.2	5.1	t		8.1	59.0			1.66	t		1.05	B
C1-6	6	3.6	t	7.2	t	t	36.6	41.5	1	t	1.4	7.2					0.5		A
ST-4	7	4.7	t	4.8	t	1.5	39.5	38.8	t	t	3.5	5.7					t		A
ST-6	7	3.7	t	6.6	t	t	36.5	43.4			0.8	6.8					t		A
ST1-2	7	8.3	0.8	5.2	t	0.6	61.5	7.8	t		1.1	12.2					0.9		A
S4-3	8	4.6	t	4.6	t	1.4	39.2	40.4	1	t	1.4	5.1					1.5		A
S4-1	8	3.1	t	7.5	t	1.7	40.2	39.4	1		1.5	4.2							A
S3-11	9	4.3	t	2.7	t	t	40.5	44.0	t	t	t	5.4					1.2		A
S4-11	10	3.1	0.5	5.6	0.8	t	33.3	31.3	1			22.0						0.8	C
S3-5	10	4.1	t	6.5	t	t	45.1	39.3	t		0.7	2.9					t		A
Rh1-3	11	3.6	t	8.1	t	0.3	37.4	40.0	t		1.0	8.4							A
S3-1	12	3.9	t	6.1	t	1.4	35.2	39.3			t	11.2			0.7		1.0		A
R1-17	13	3.2	t	2.3	1.0	t	21.5	17.2	1	t	t	47.8	4.41		1.0				C
R1-2	14	3.5	t	7.0		1.1	40.0	39.9			2.2	5.1					t		A
R1-6	15	3.8	t	9.9	t	1.5	35.6	37.2				9.2							A
R1-9	16	4.3	t	8.7	1.7	0.9	39.0	32.2		7.37	t	3.4		0.89	1.89		t		A
S1-7	17	3.3	t	3.7	t	t	37.0	45.0	t		1.0	7.7					t		A
S3-10	18	3.4		7.8	t	1.1	37.6	38.7			1.7	1.3				t			A

[a]The figures refer to the percentage of fatty acid to the total fatty acids.
t = trace amount, less than 0.5%.

Table 12. Algal populations on decomposing straw (colony forming unit g^{-1} dw)

Treatment	Days after flooding	Heterocystous BGA	Unicellular BGA	Total algae
T1	8	$1.3\ 10^5$	$1.5\ 10^6$	$2.2\ 10^6$
T1	14	$2.3\ 10^6$	$< 1.0\ 10^4$	$7.8\ 10^6$
T1	22	$7.9\ 10^6$	$< 1.0\ 10^4$	$7.9\ 10^6$
T2	71	$6.0\ 10^7$	$2.4\ 10^7$	$1.2\ 10^8$
T2	78	$3.2\ 10^8$	$1.7\ 10^8$	$4.9\ 10^8$

Table 13: Coefficients of correlation between algal biomass and photodependant acetylene reducing activity (ARA)*.

Algal biomass	f. w.	d. w.	Carbon	Nitrogen	Phosphorus
ARA	0.73	0.70	0.67	0.66	0.83

* $r = 0.35$ for $p = 0.01$

Fig. 1. Floodwater pH at 07:00-07:30

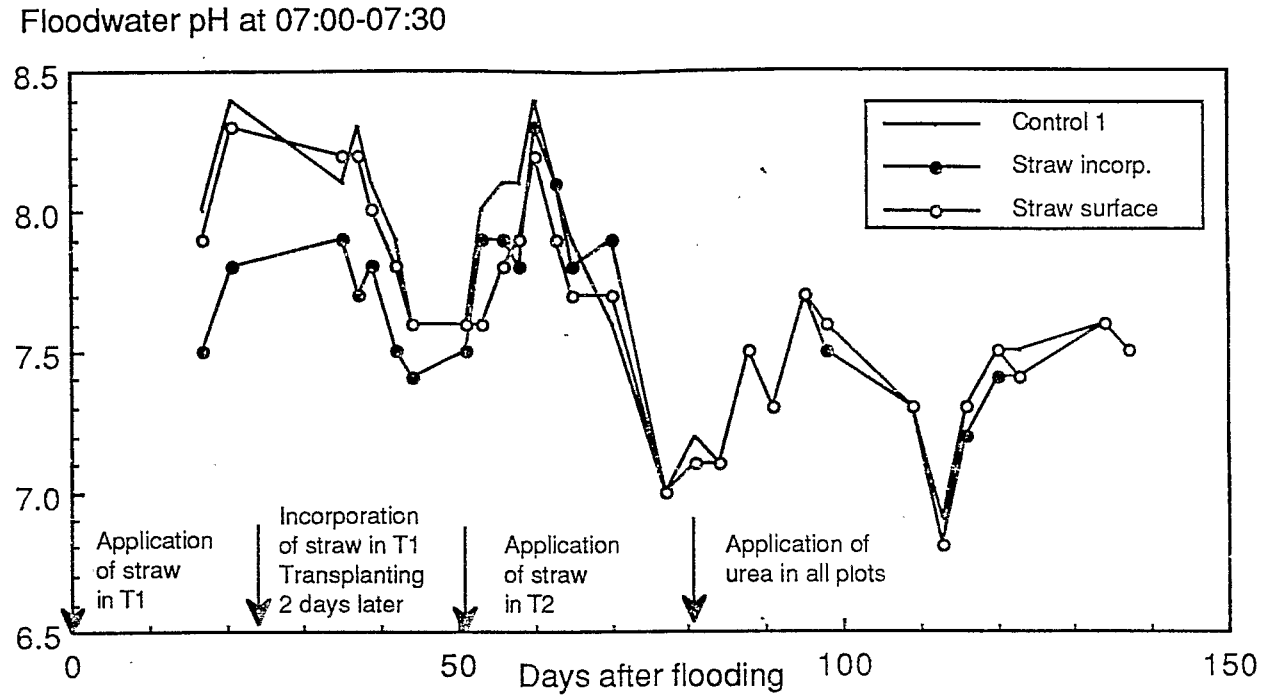


Fig. 2. Floodwater pH at 13:00-13:30

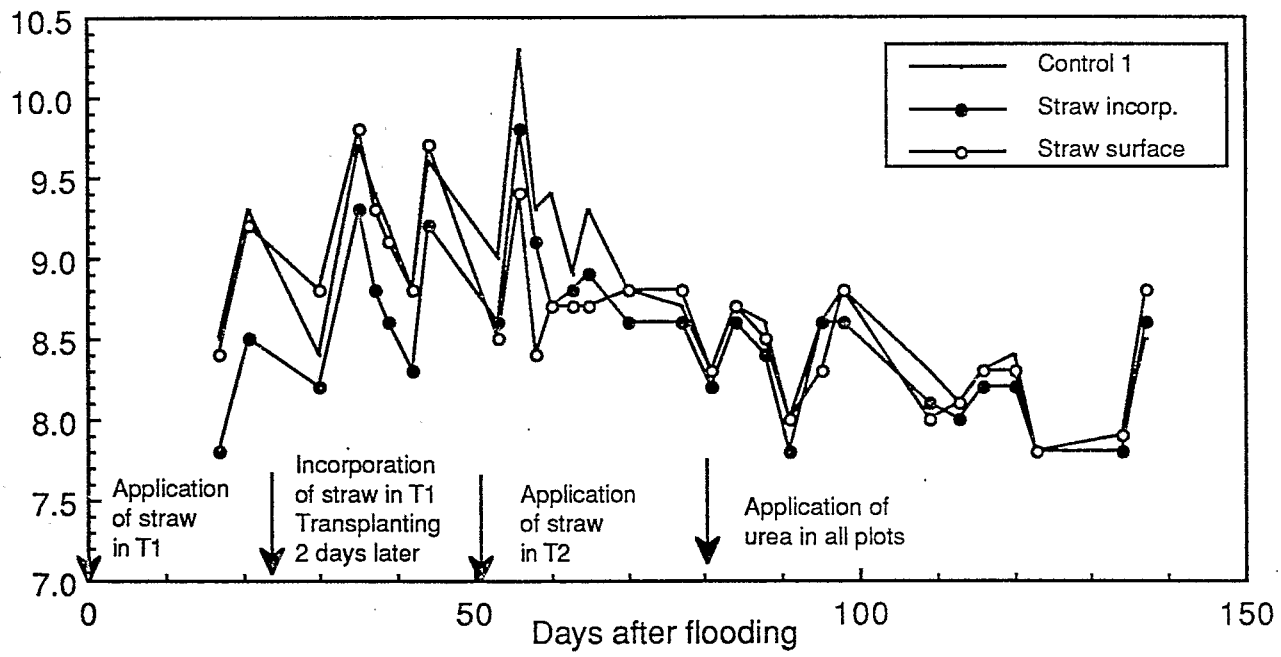


Fig. 3. Dissolved oxygen in the floodwater at 07:00-07:30

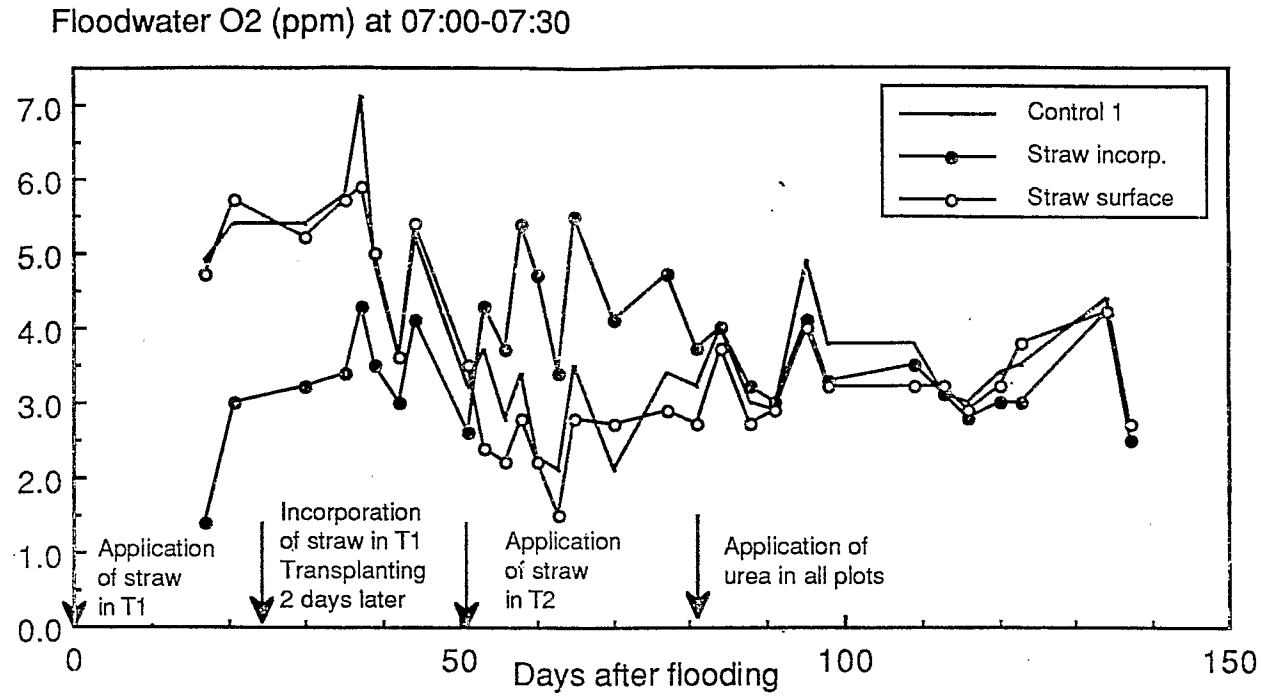


Fig. 4. Dissolved oxygen in the floodwater at 13:00-13:30

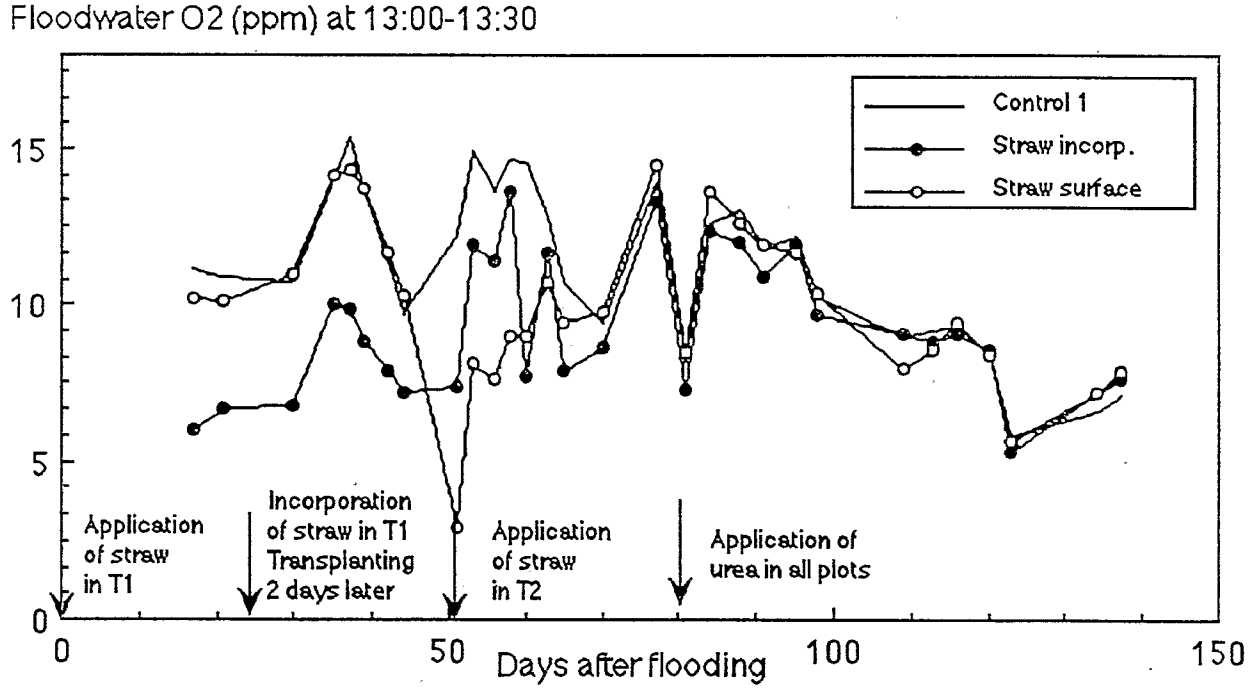


Fig. 5. Correlation between pH and dissolved oxygen in the floodwater

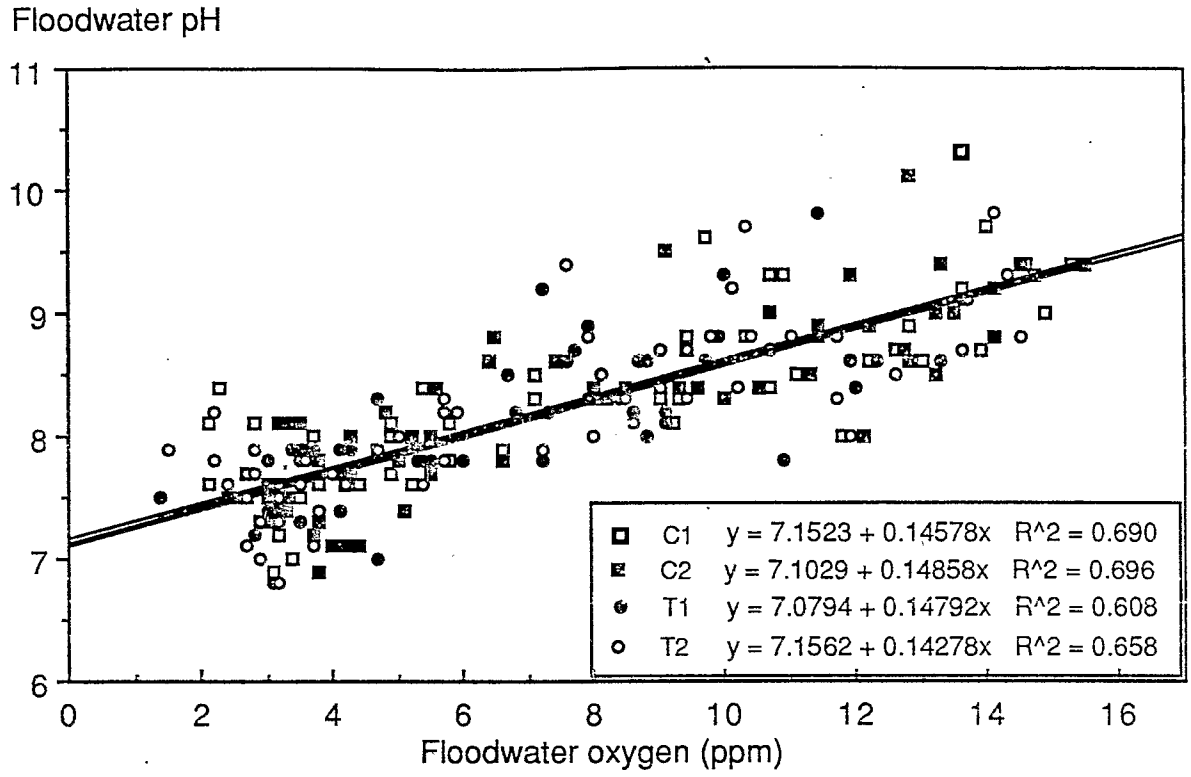


Fig. 6. Difference in dissolved O_2 concentration at 13:00-13:30 between straw treated plots (T_1,T_2) and C_1

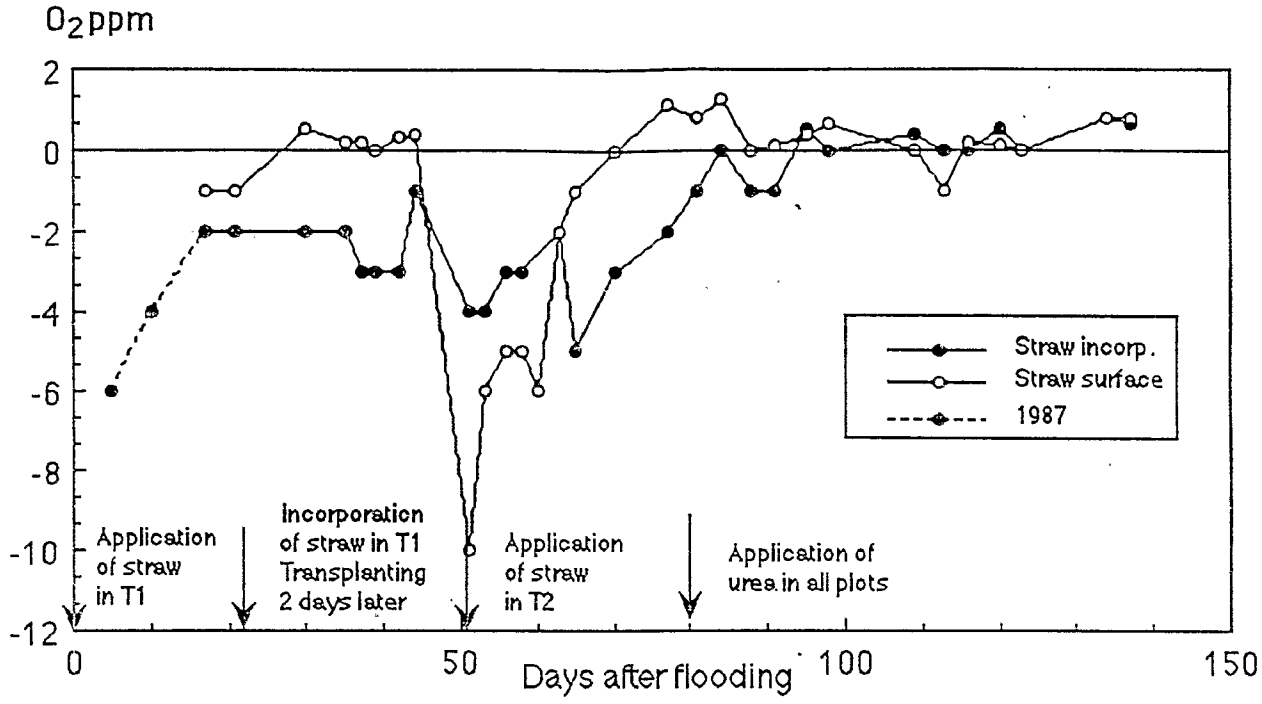
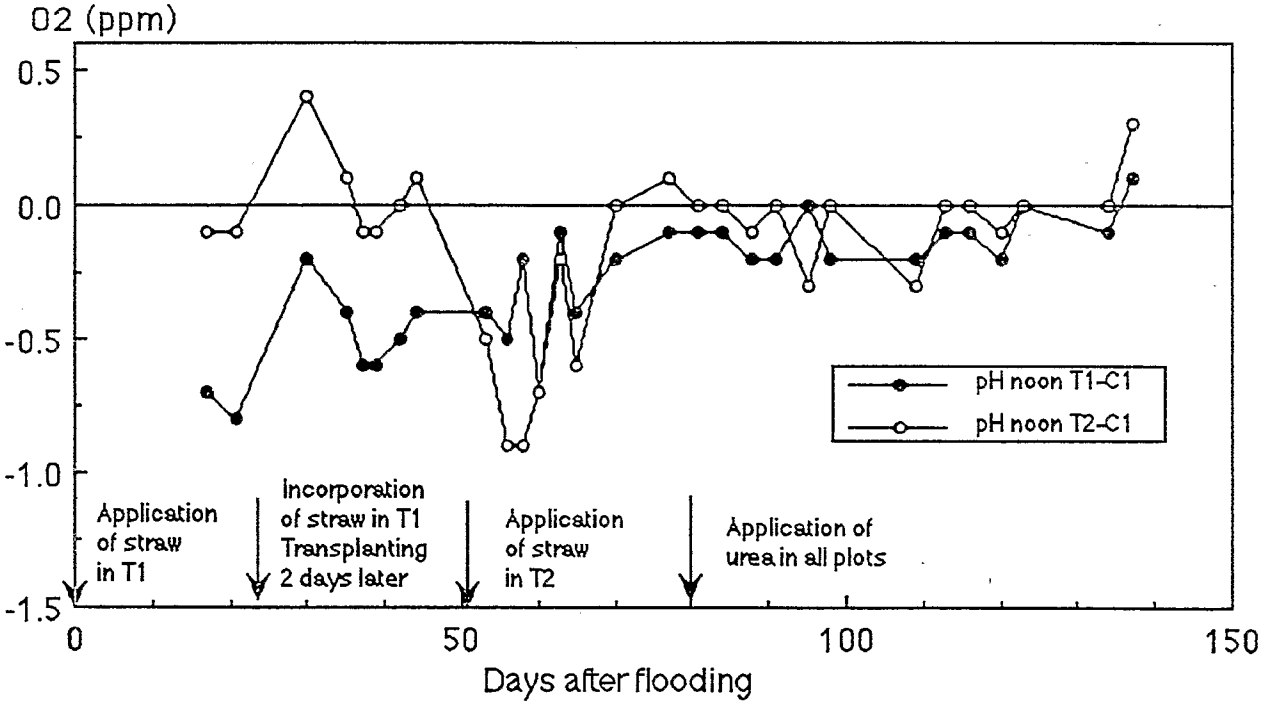


Fig. 7. Difference in flodwater pH at 13:00-13:30 between straw treated plots (T_1,T_2) and the control C_1



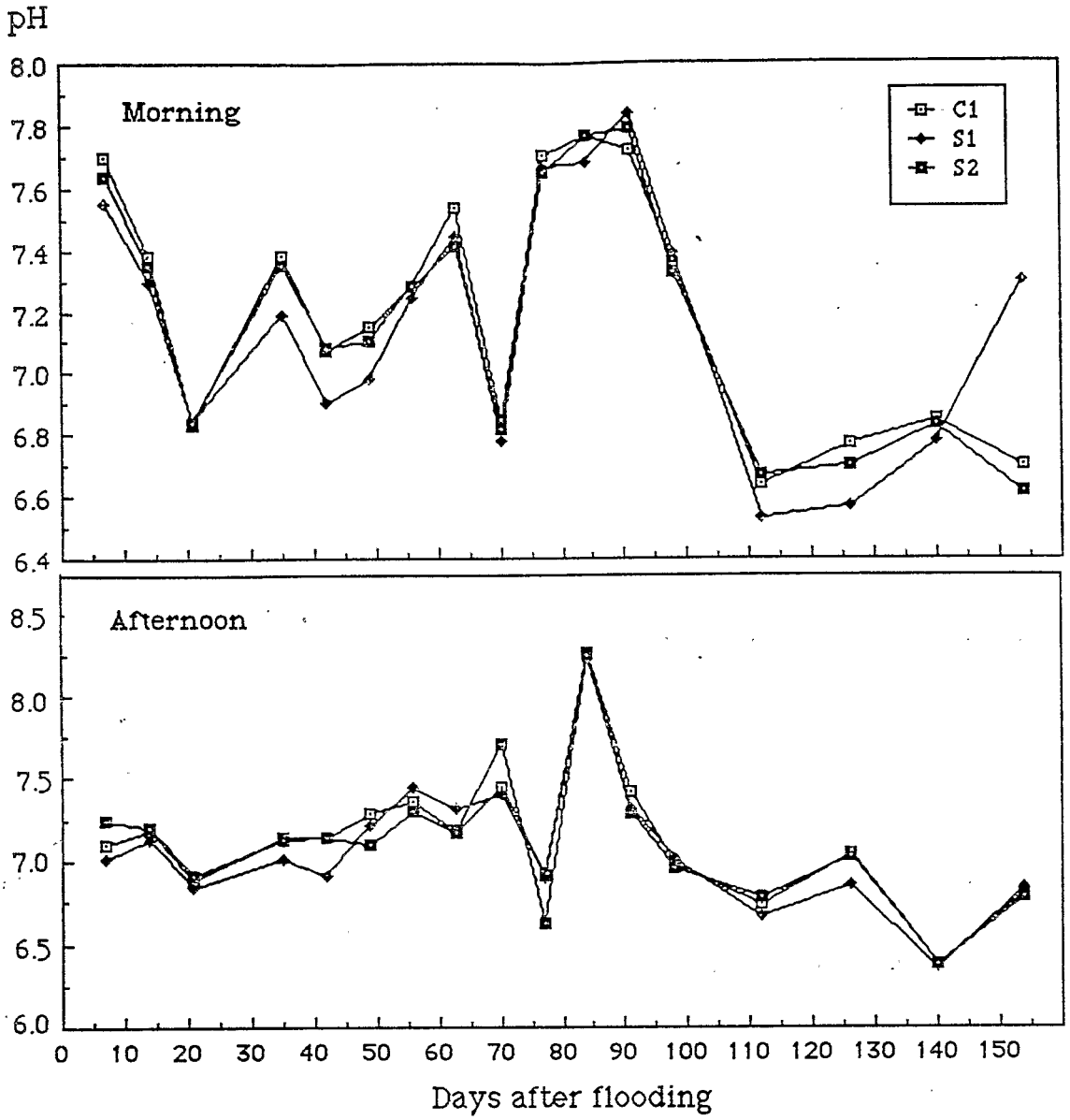


Fig. 8. Kinetics of soil pH measured at 2cm depth in situ

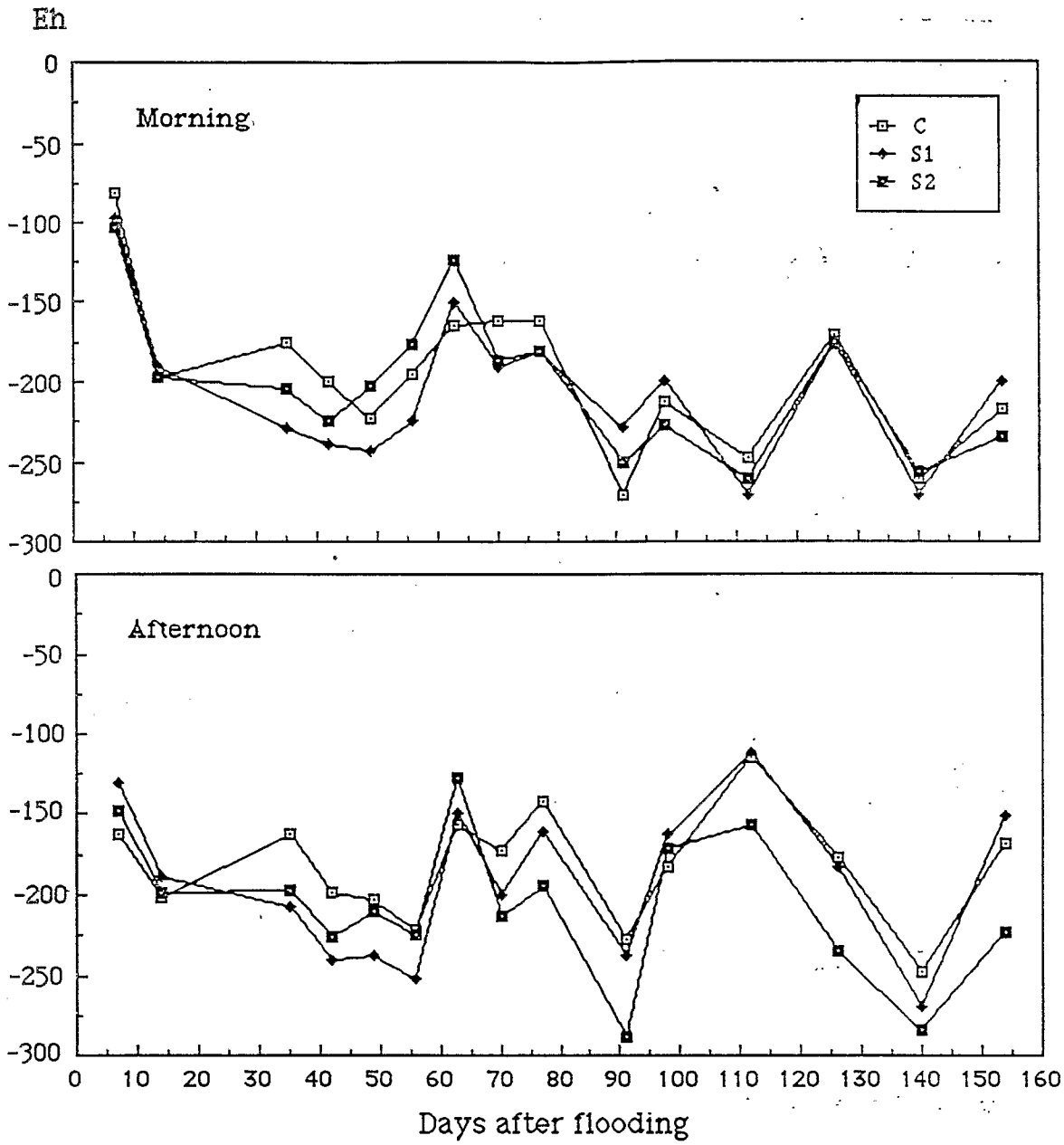
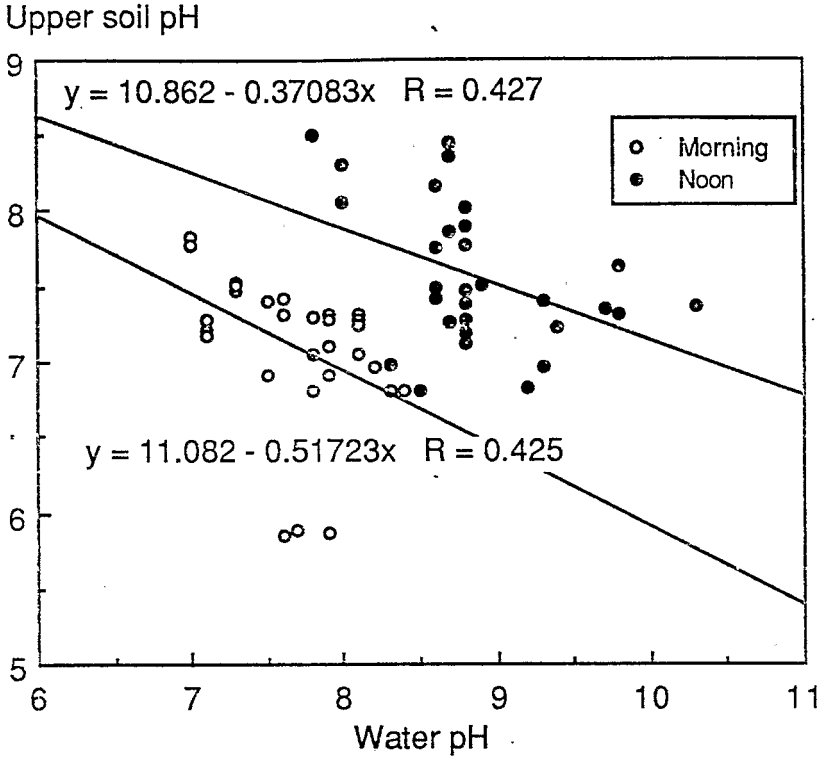


Fig. 9. Kinetics of soil Eh (mean of measurements at 5 and 10 cm)

Fig. 10. Correlation between water pH and pH of the 0-2 cm soil layer.



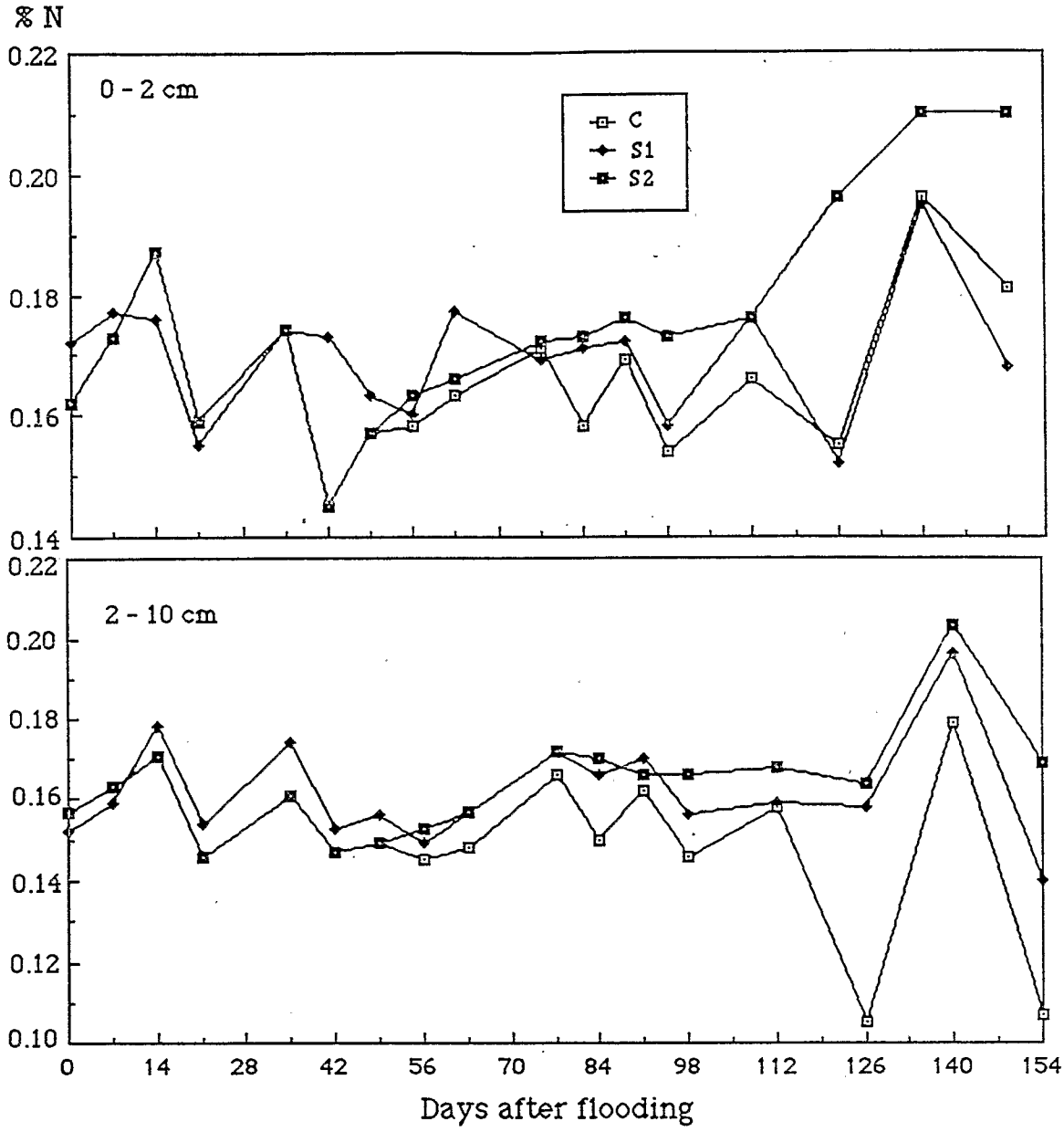


Fig. 11. Kinetics of soil total N at 0-2 and 2-10 cm depth

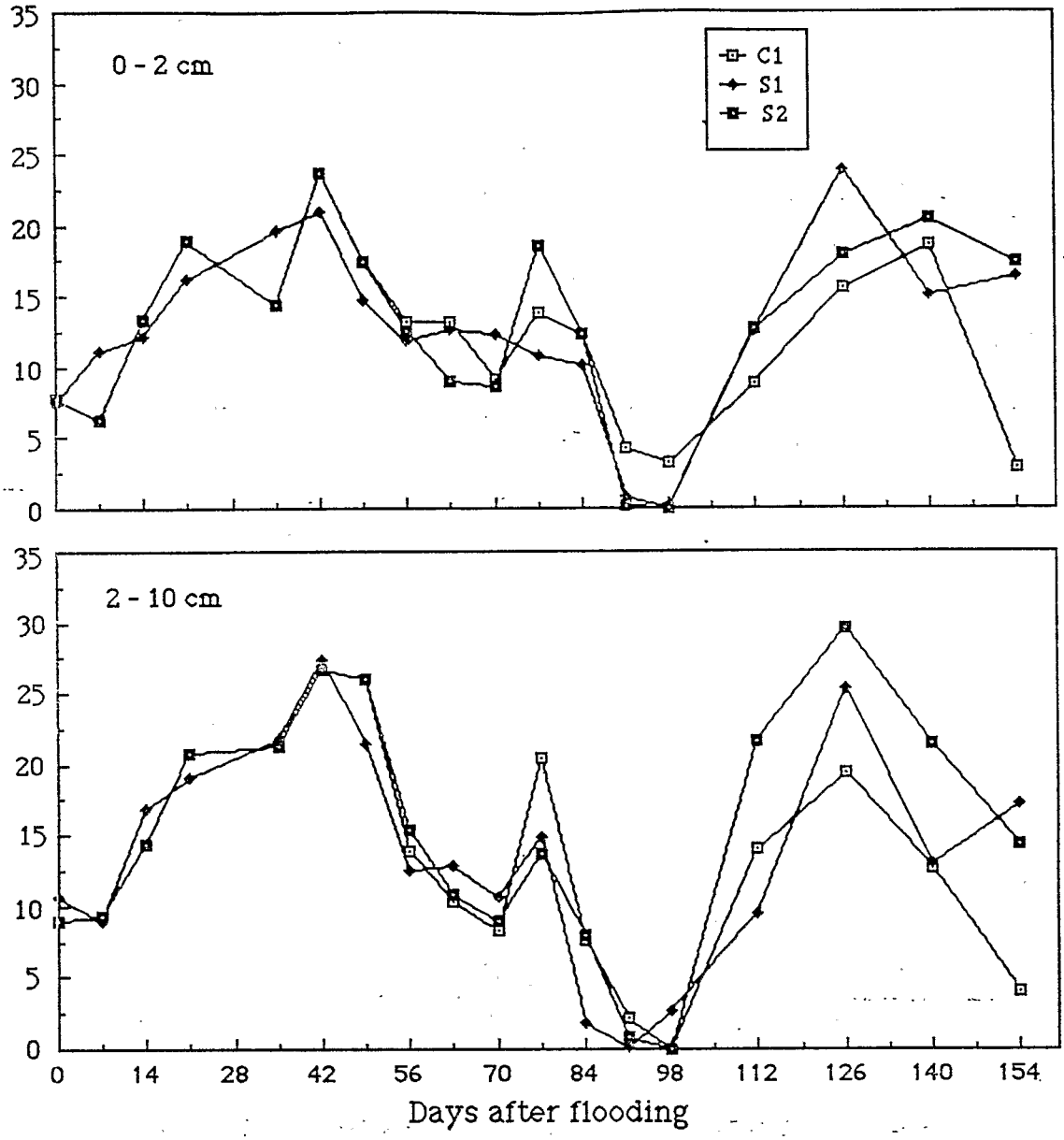


Fig. 12. Kinetics of ammonium-N at 0-2 and 2-10 cm soil depth

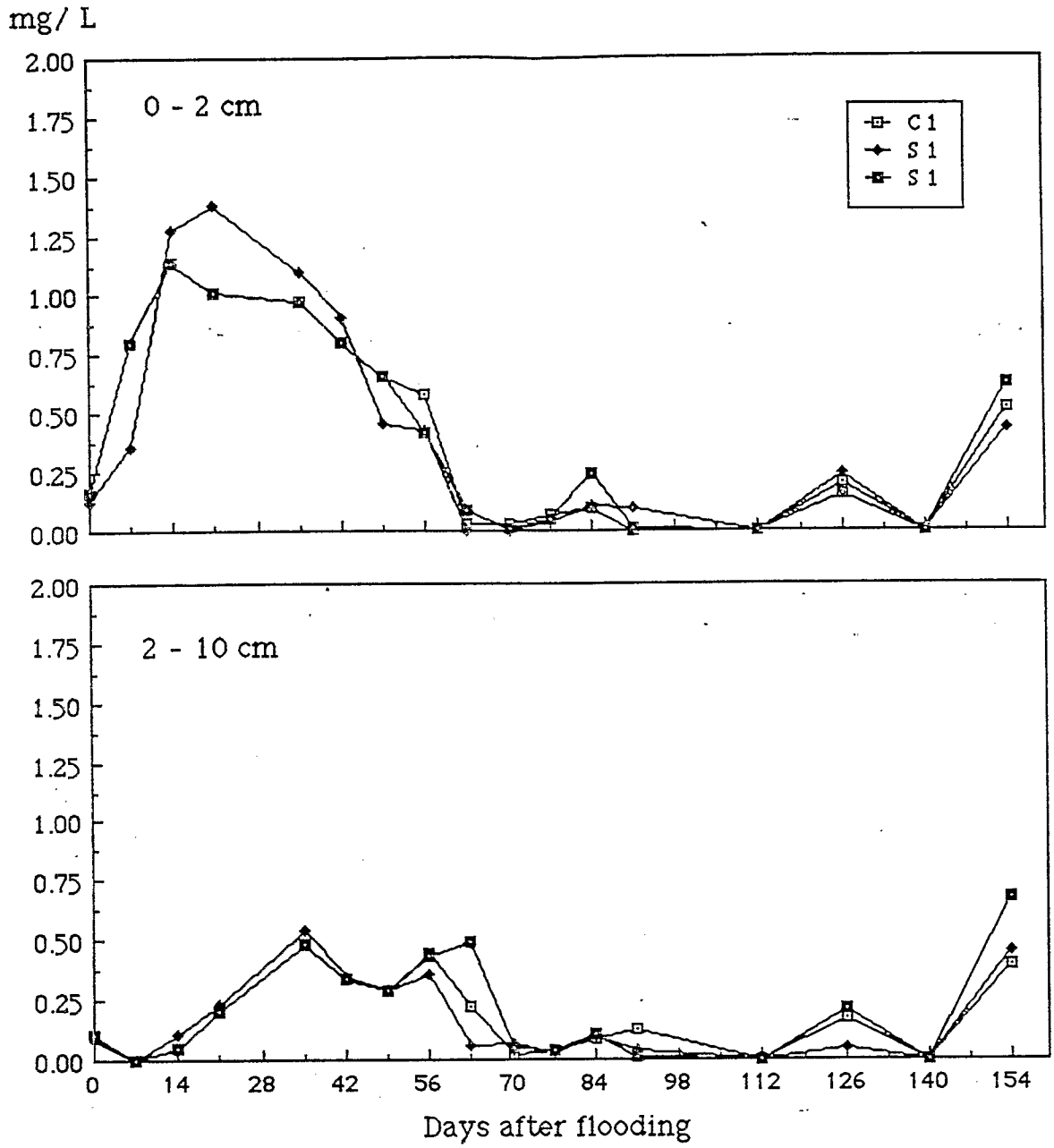


Fig. 13. Kinetics of ammonium-N in soil solution

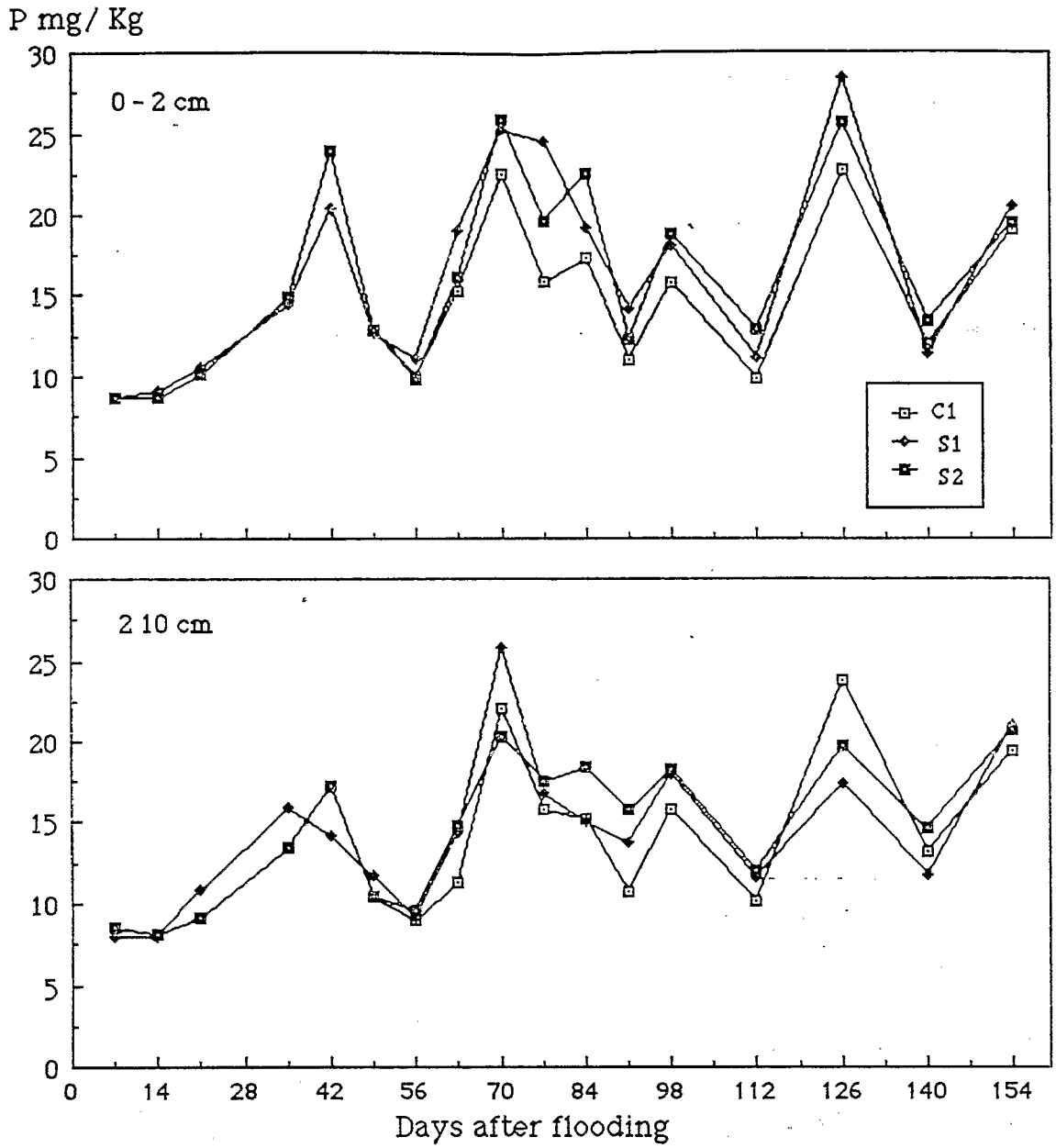


Fig. 14. Kinetics of Olsen P at 0-2 and 2-10 cm soil depth

Fig. 15. Kinetics of Fe in soil solution

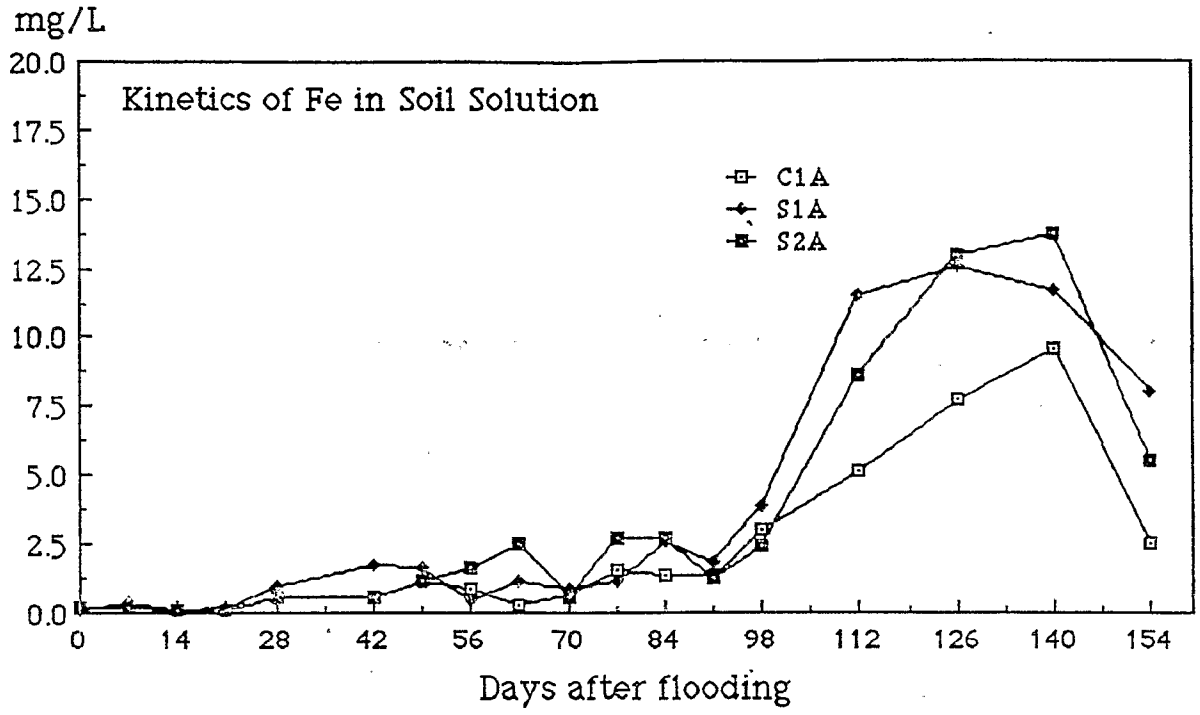


Fig. 16. Kinetics of Mg in soil solution

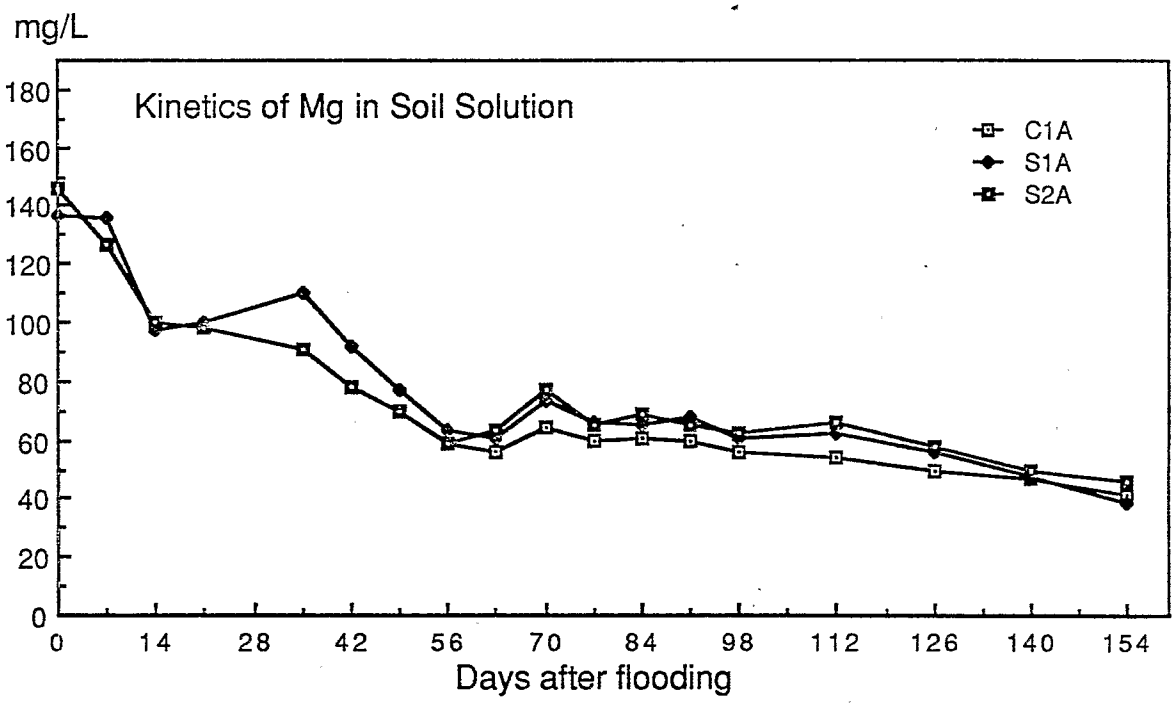


Fig. 17. Kinetics of Ca in soil solution

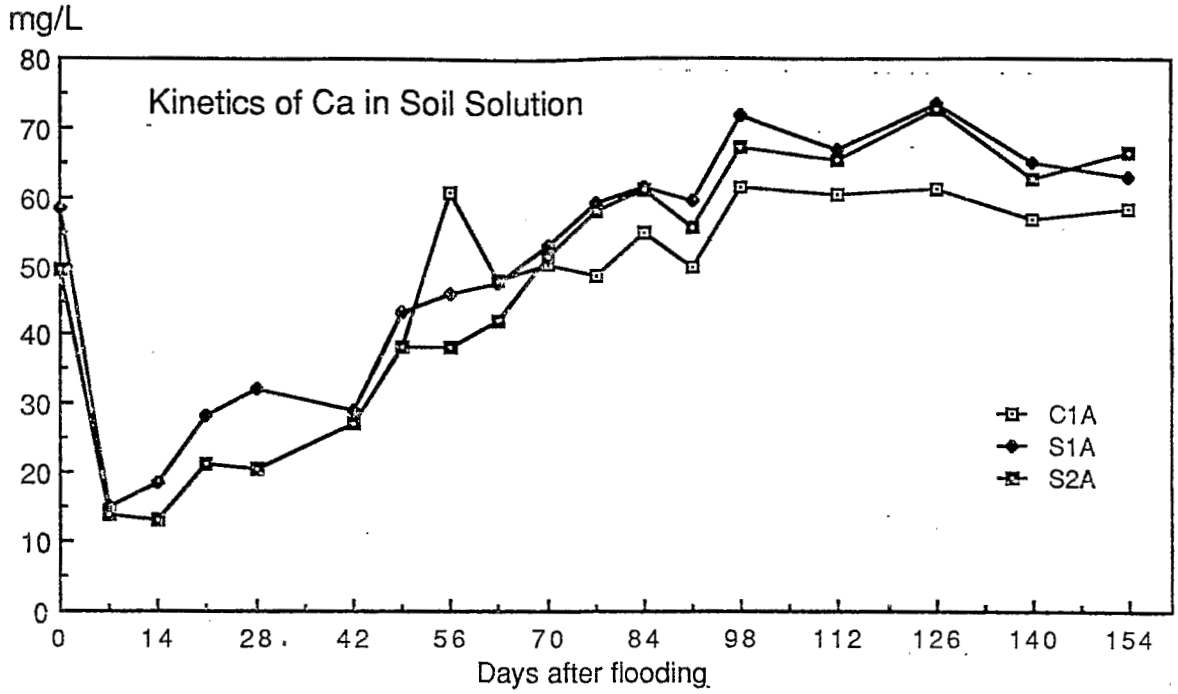
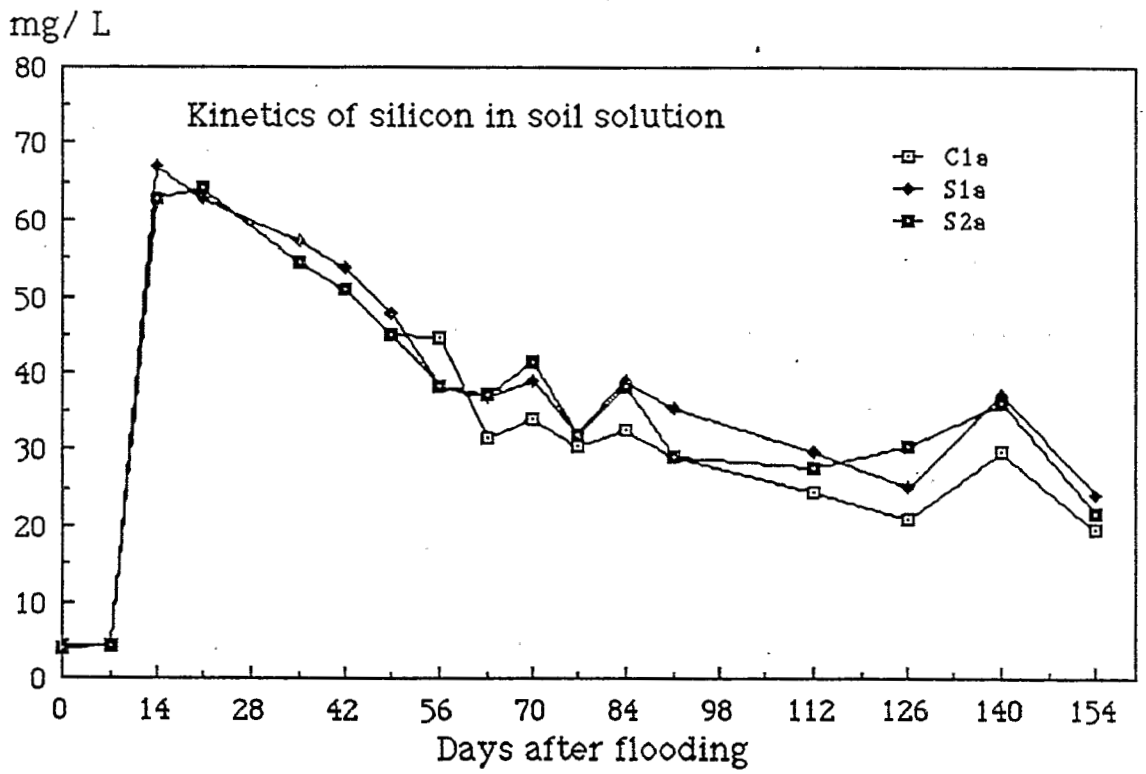


Fig. 18. Dynamics of Si in soil solution



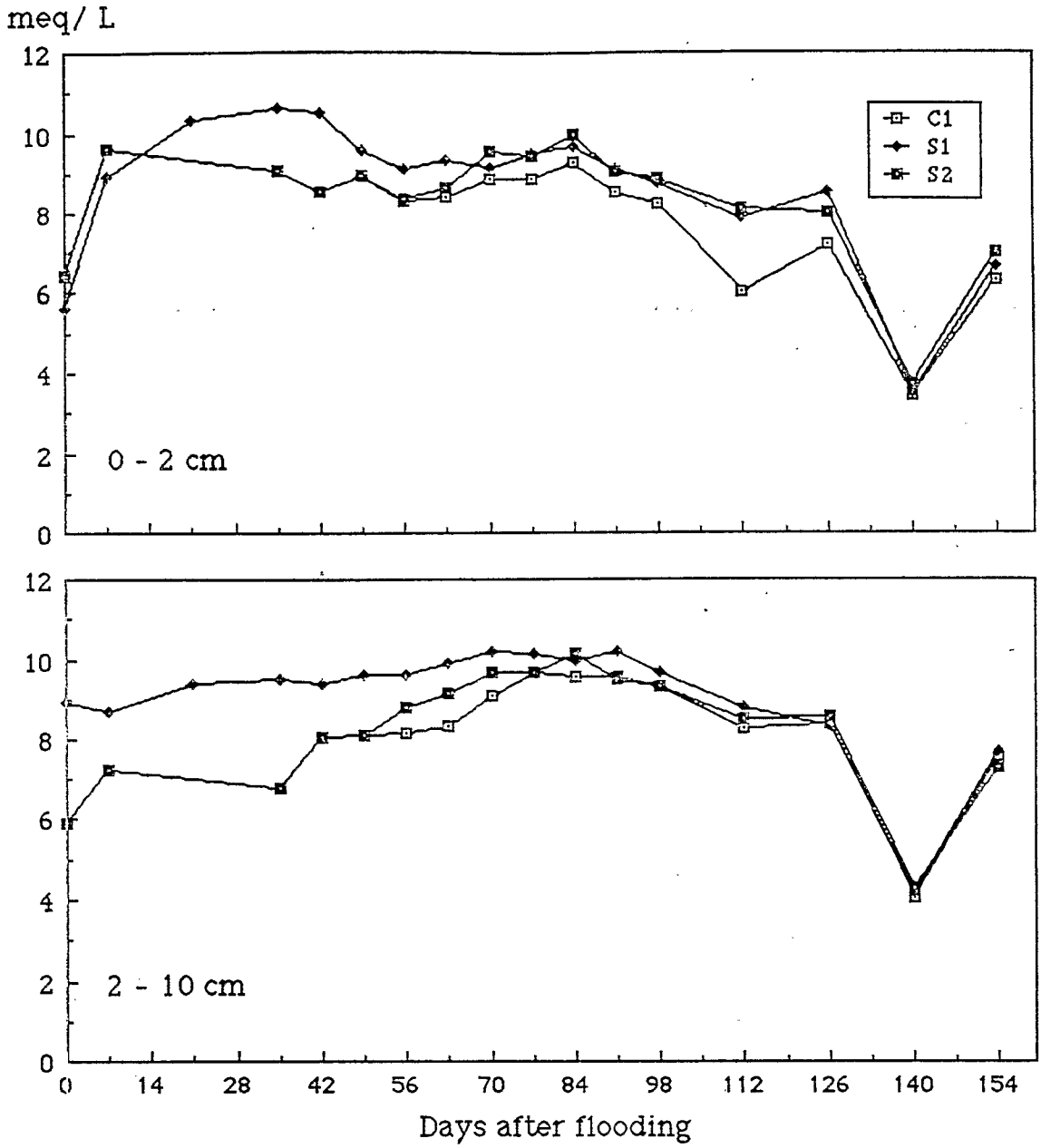


Fig. 19. Dynamics of bicarbonate in soil solution.

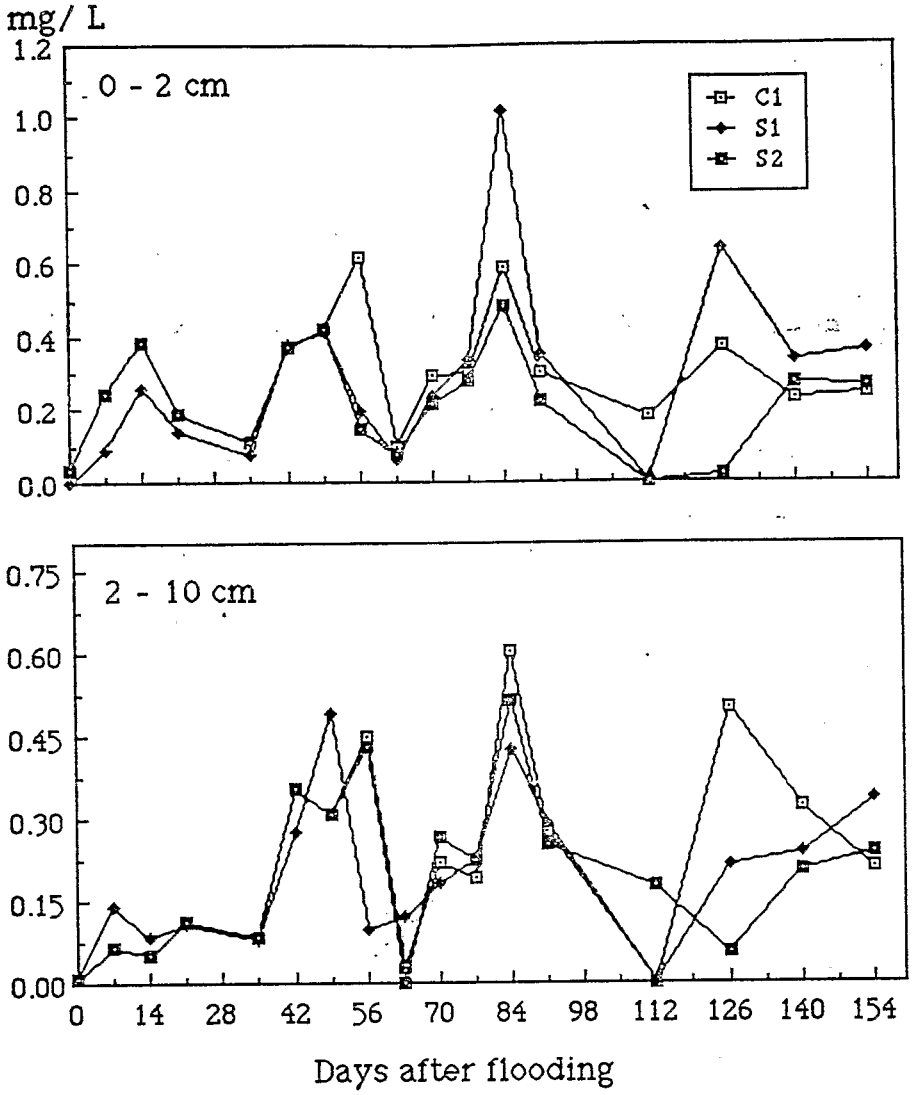
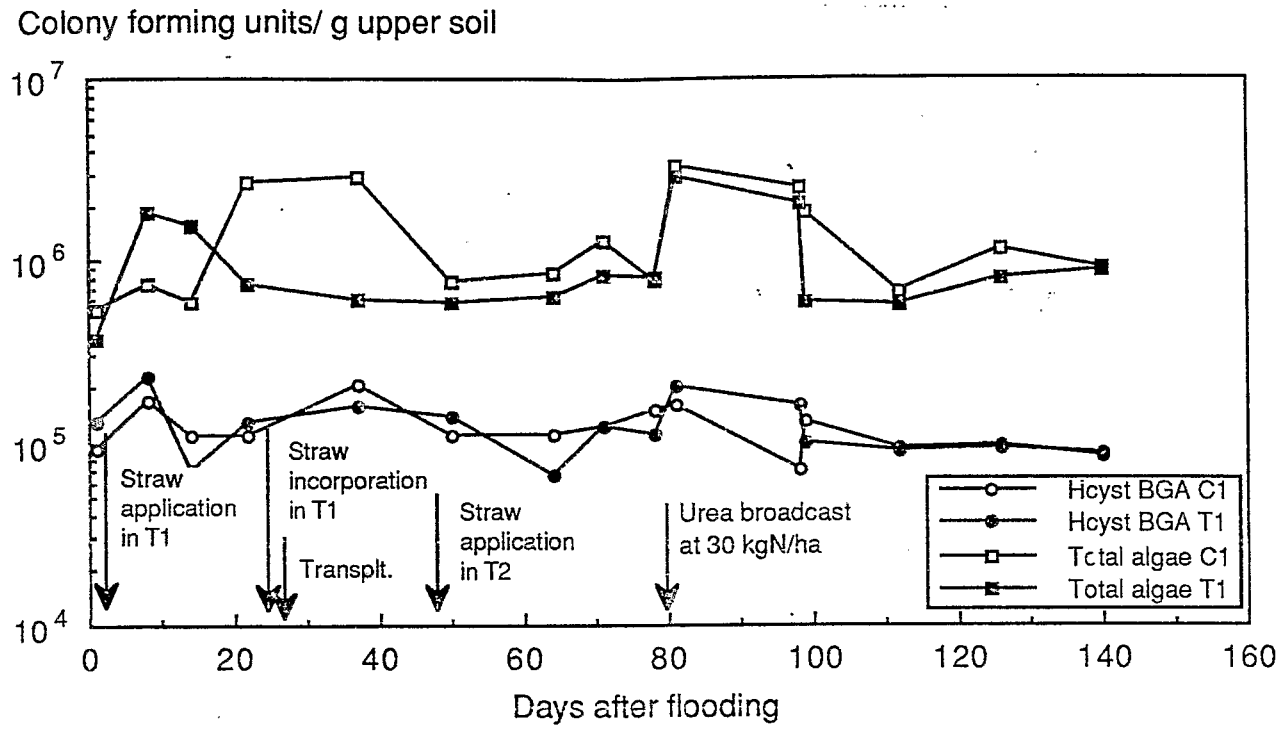


Fig. 20. Dynamics of P in soil solution.

Fig. 21. Algae counts in Control 1 and Treatment 1



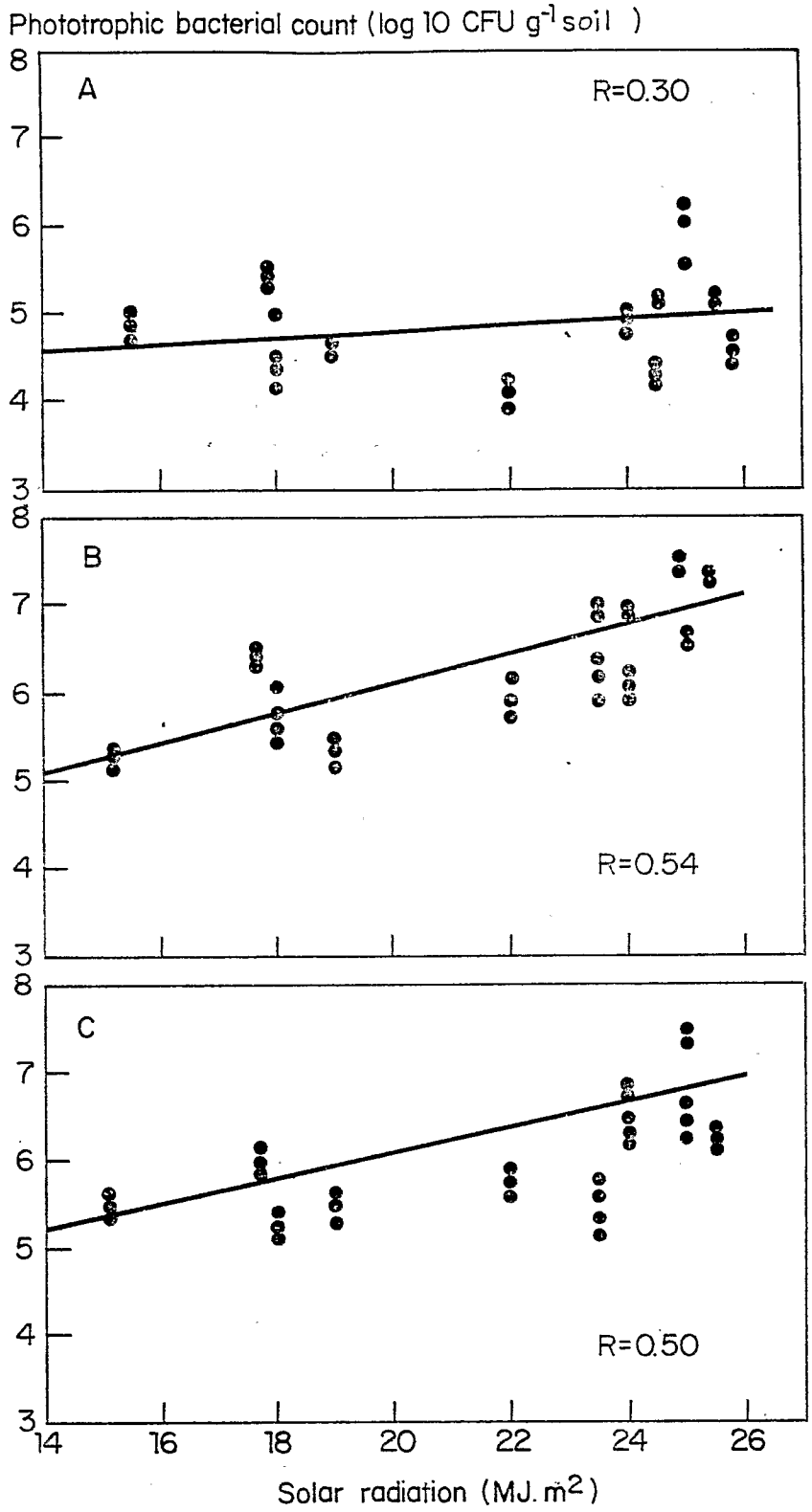


Fig. 22. Correlation between phototrophic bacterial counts and solar radiation in wetland rice soil of control (A), treatment 1 (B) and treatment 2 (C).

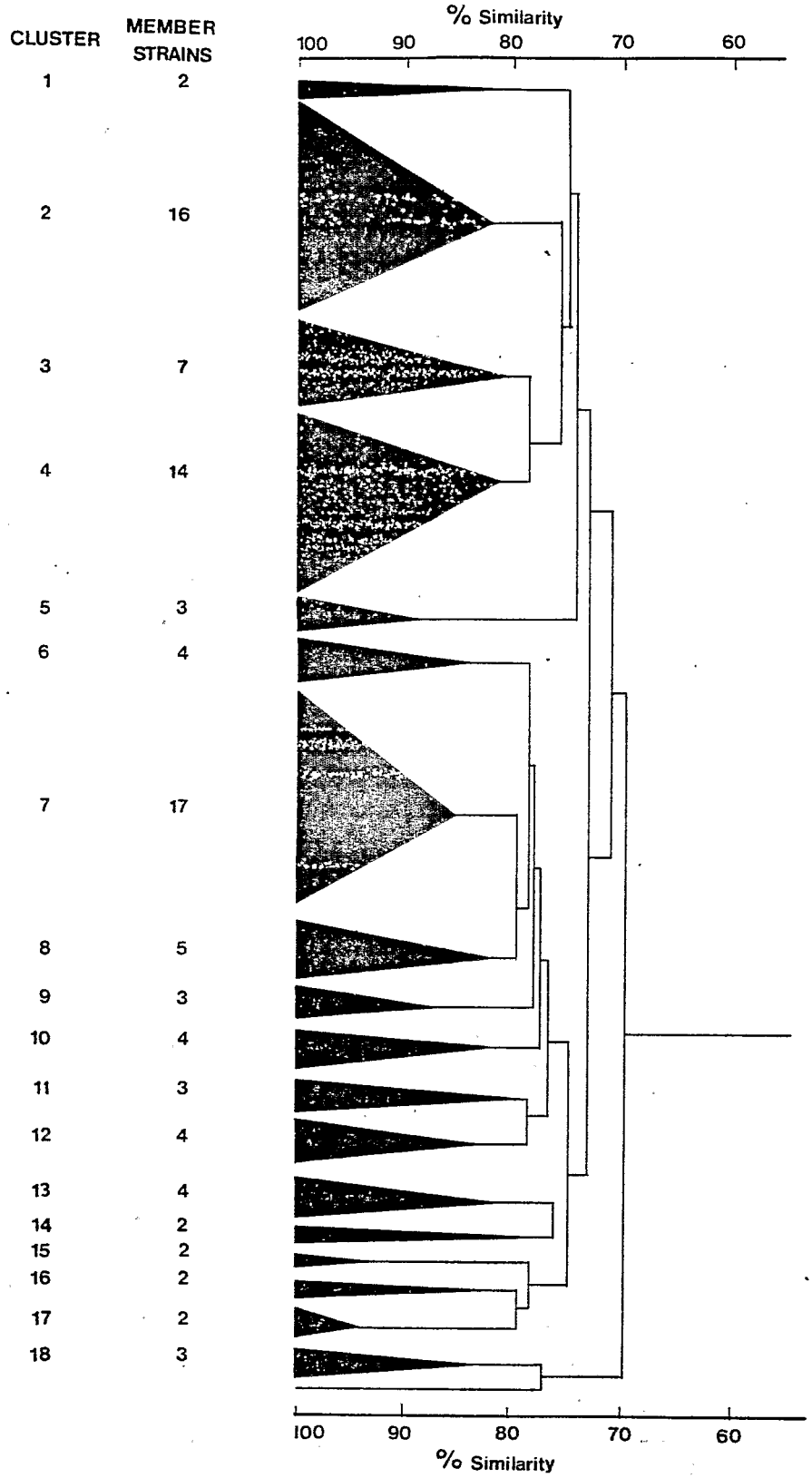


Fig. 23. Phenogram of phototrophic bacteria from nonrhizosphere, rhizosphere and decomposing straw.

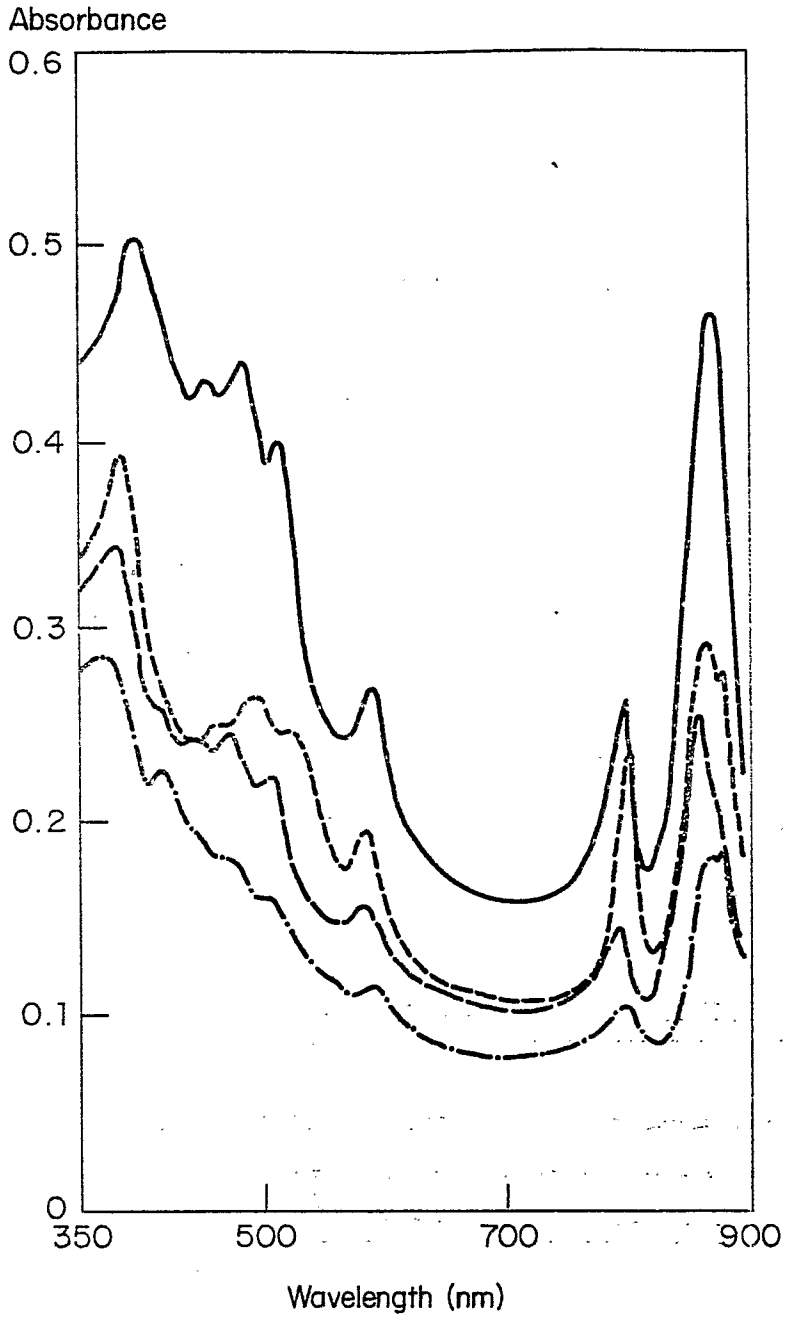


Fig. 24. Photopigment absorption spectra of intact cells of phototrophic bacteria isolated from decomposing straw (-), plant roots (--), rhizosphere (..) and nonrhizosphere (-.-).

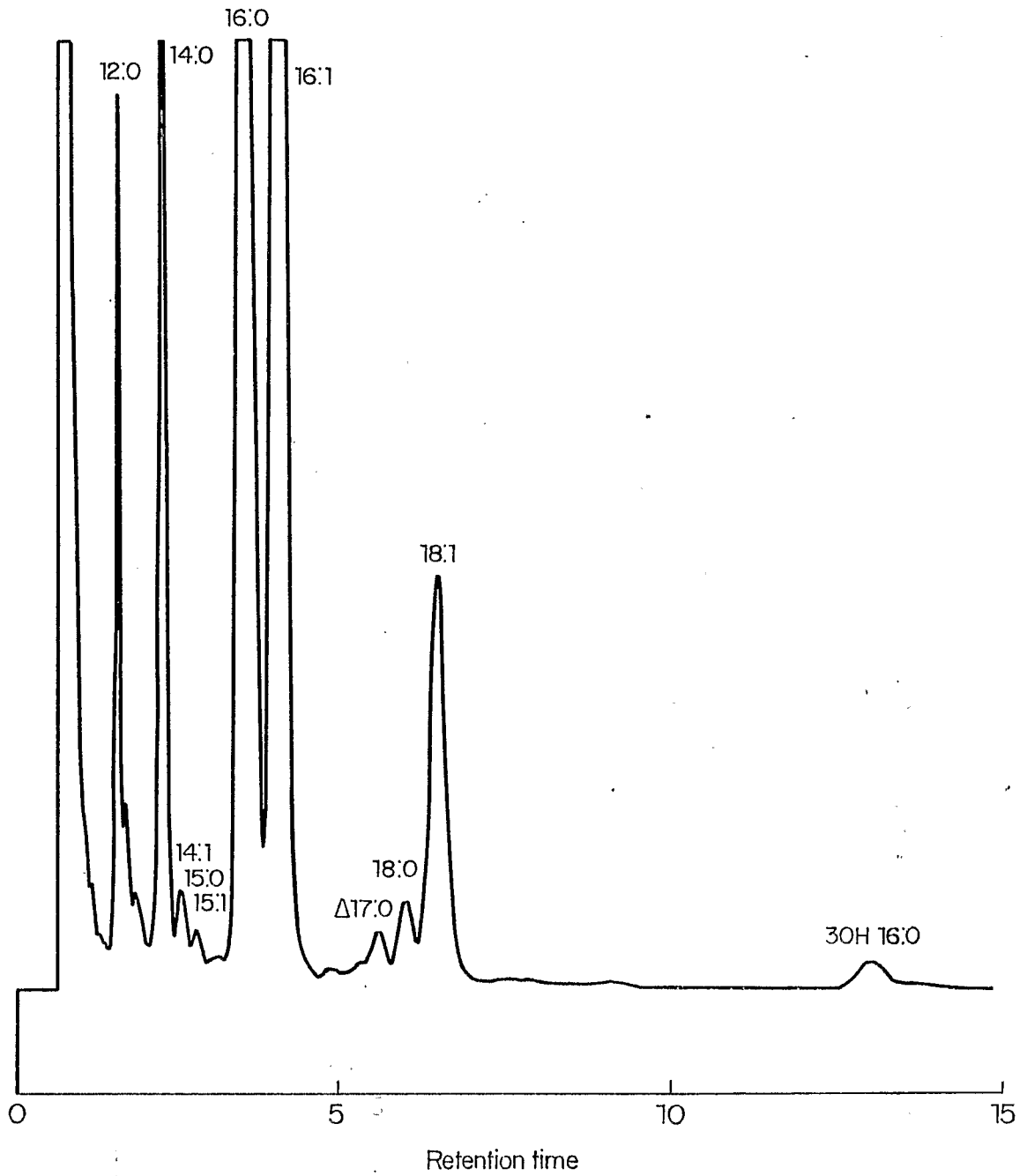


Fig. 25. Cellular fatty acid (CFA) profile of strain Rh1-2 with type A fatty acid.

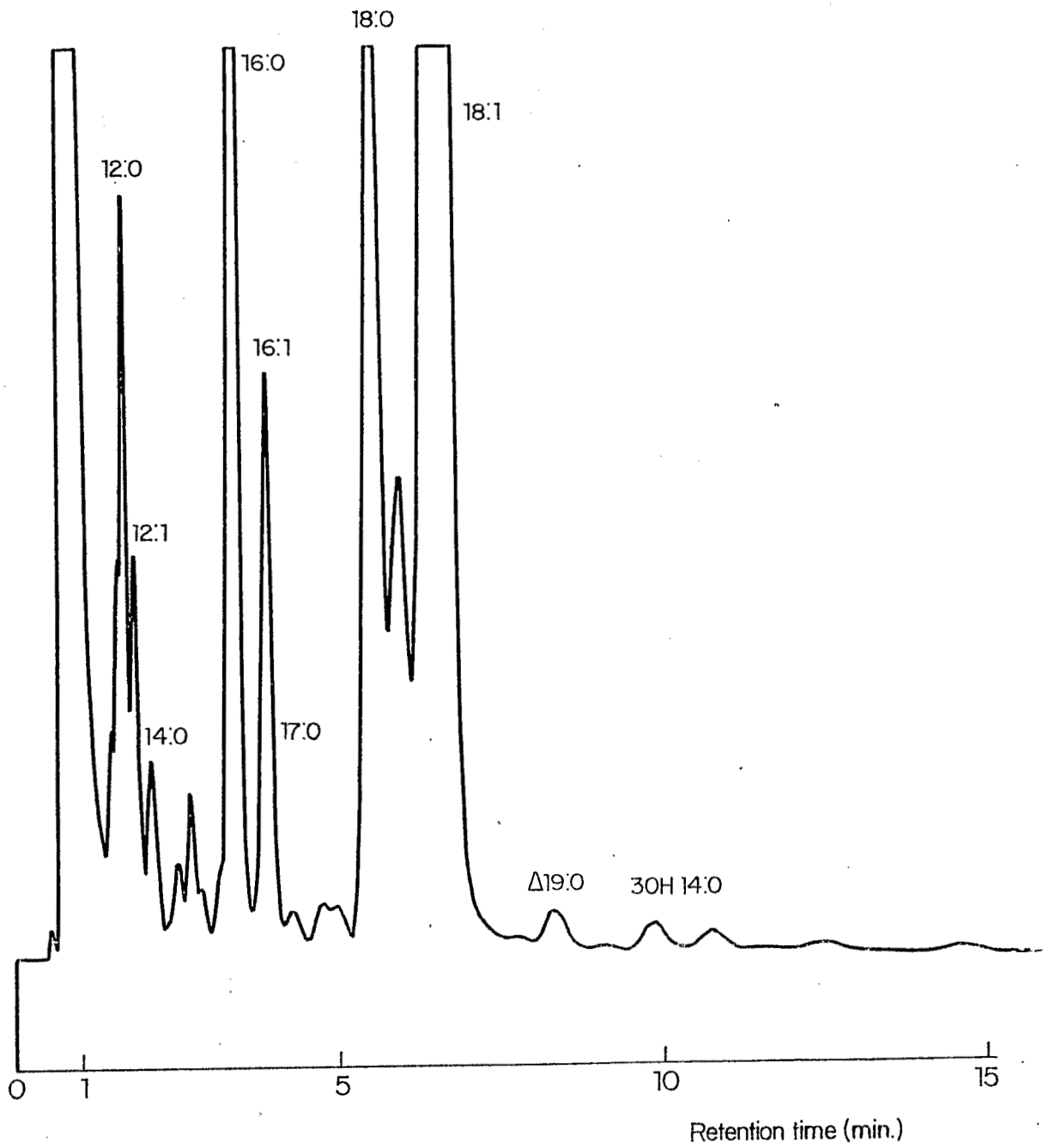


Fig. 26. Cellular fatty acid (CFA) profile of strain R3-5 with type B fatty acid.

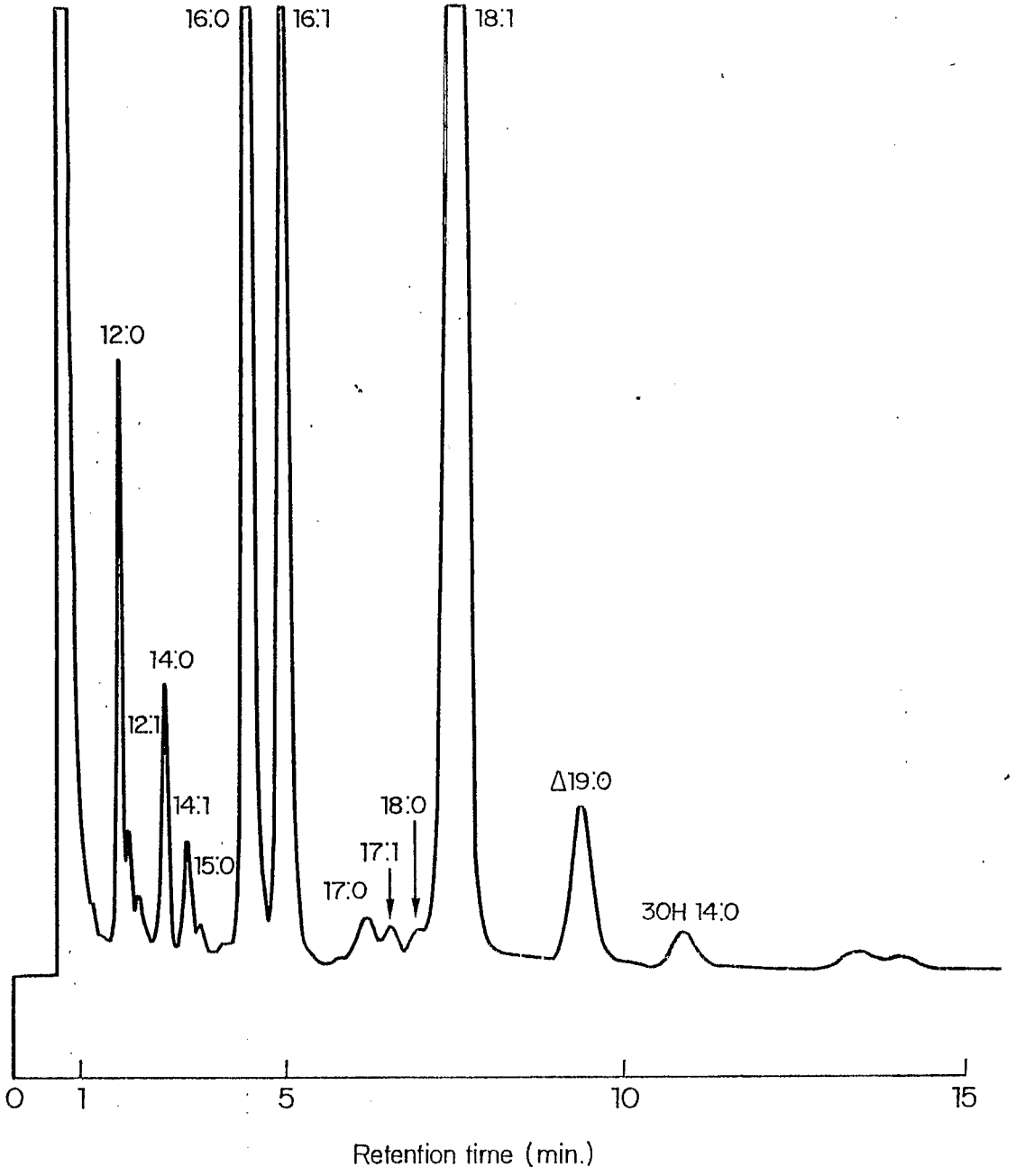


Fig. 27. Cellular fatty acid (CFA) profile of strain R1-17 with type C fatty acid.

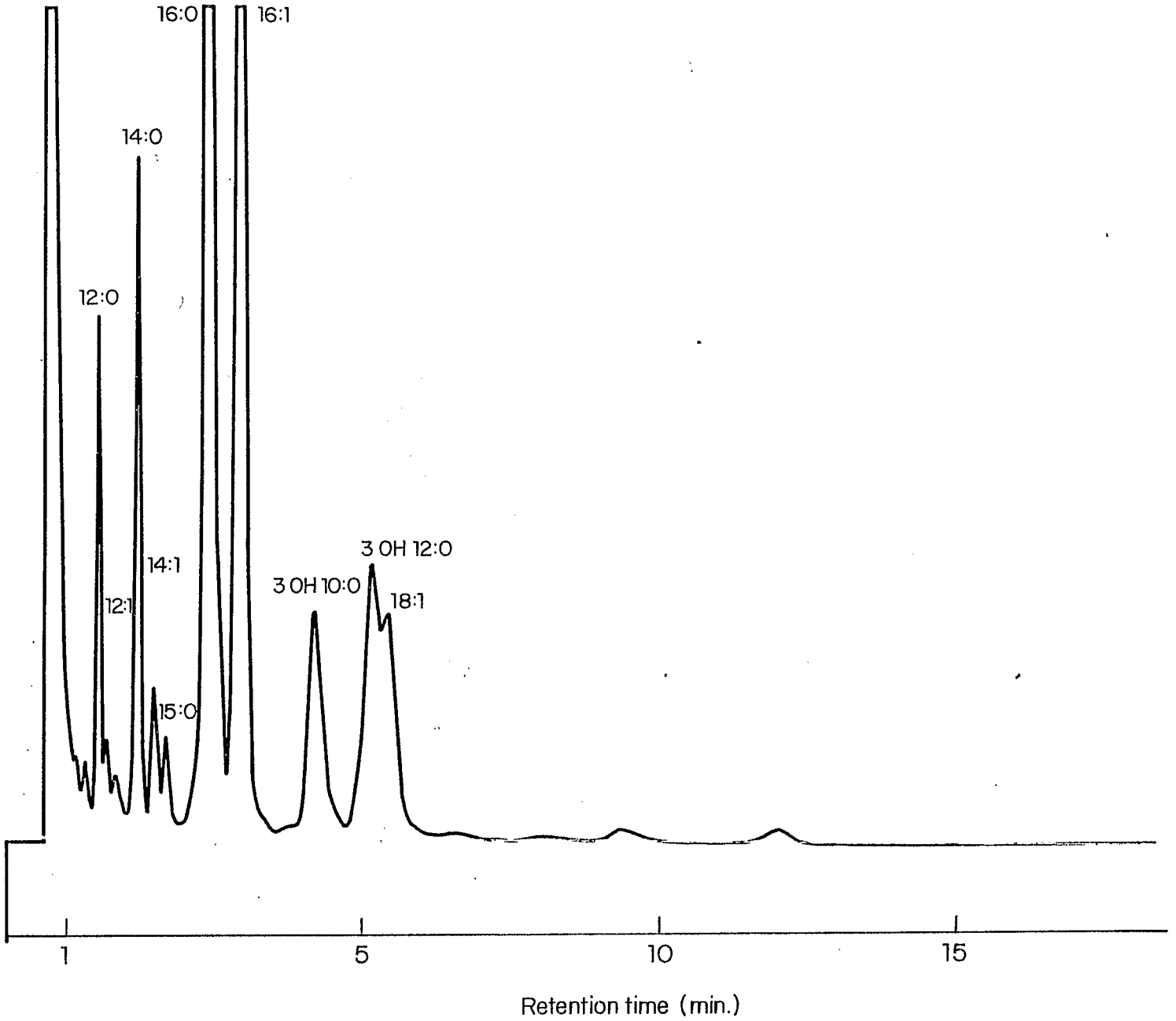


Fig. 28. Cellular fatty acid (CFA) profile of a new strain of Rhodopseudomonas sp. R1-9 with type A fatty acid.

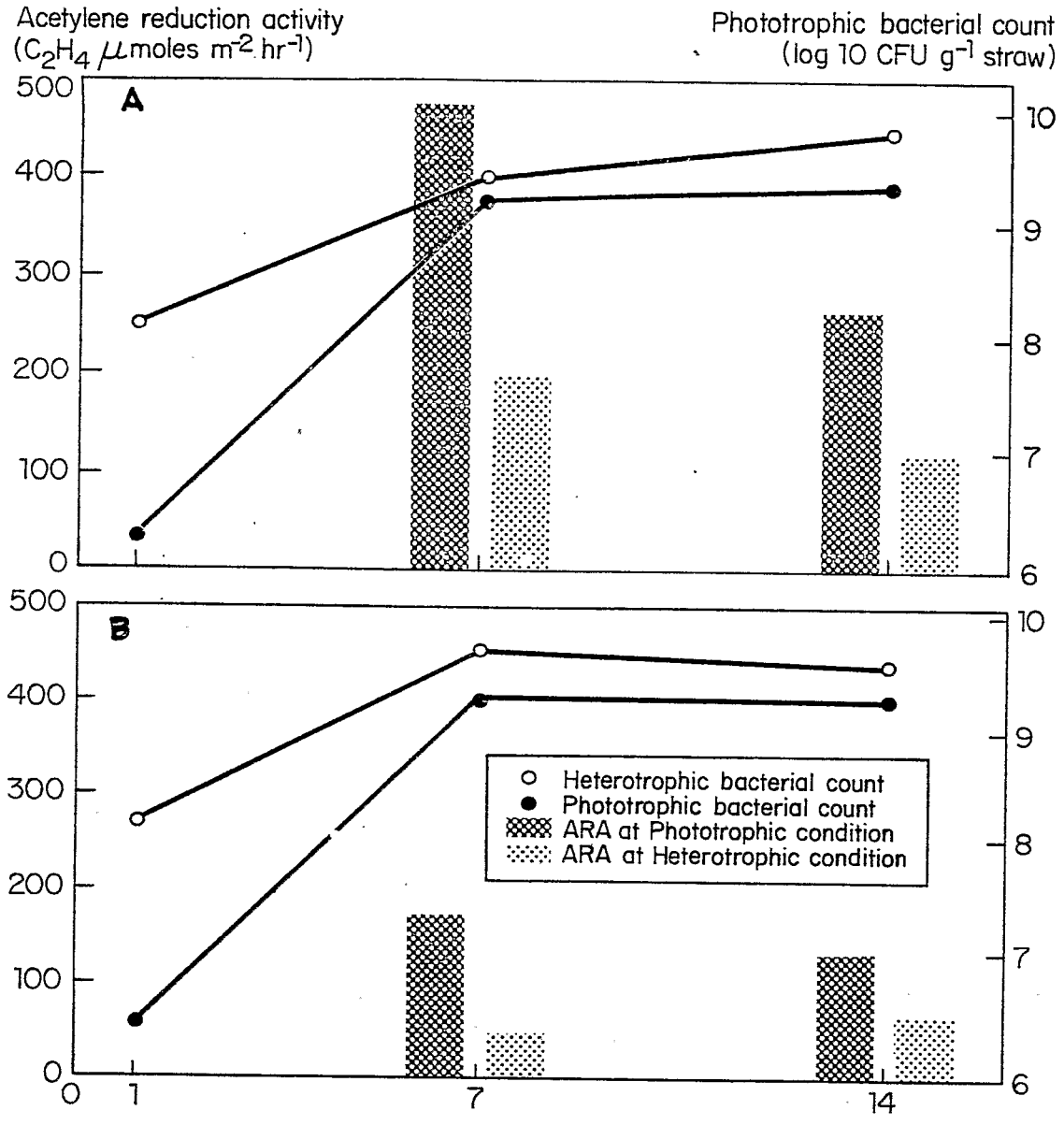


Fig. 29. Heterotrophic and phototrophic bacterial counts in decomposing straw from treatment 1 (A) and treatment 2 (B) under phototrophic and heterotrophic incubation conditions.

Fig. 30. Acetylene reducing activity of soil+water cores and straw.

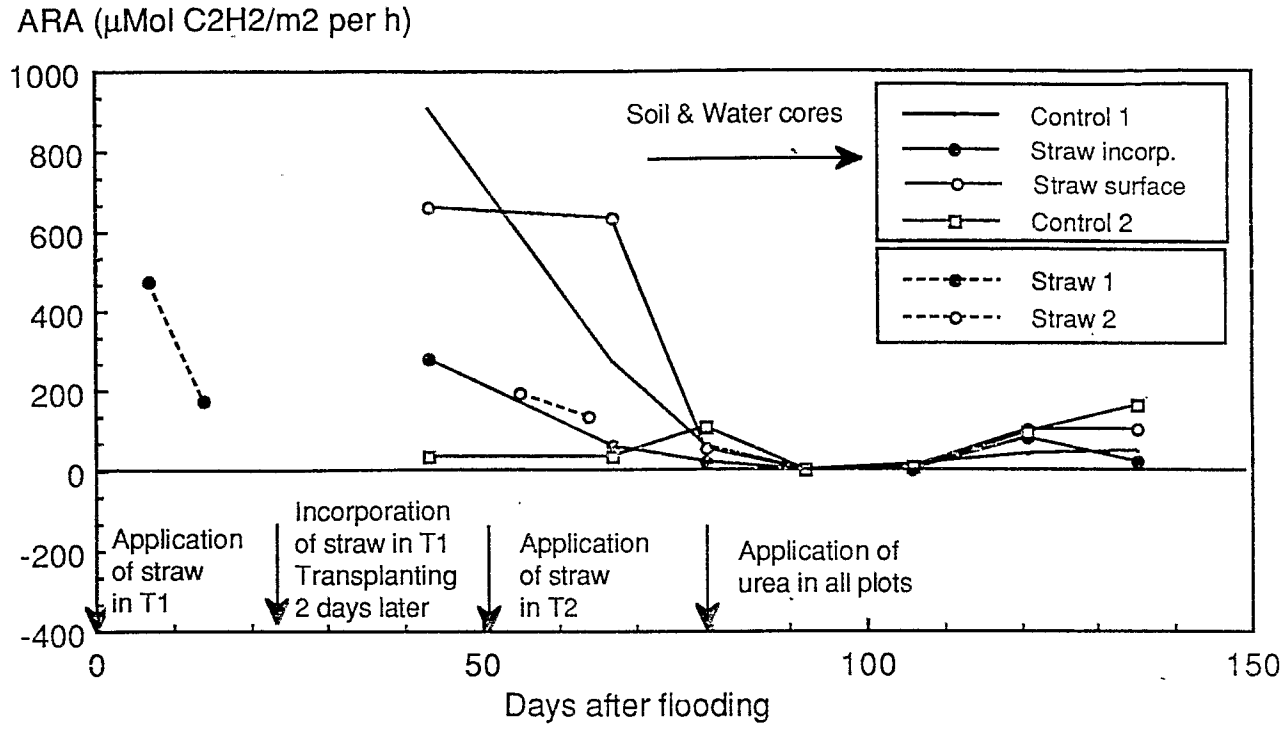


Figure 31. Specific ARA of *Aphanothece* at various biomasses.