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# EFFECT OF SOLUTION pH ON GROWTH AND MINERAL UPTAKE IN PLANTS UNDER CONSTANT pH CONDITION*

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Many automatic apparatus for maintaining constant pH have been proposed and used for solution cultures[2,5,9,10,11,12,17,18,28], because pH maintenance is difficult in the method of solution culture unless automatic regulations are provided[7,19,22].

Recently, Islam *et al.*[12] studied an optimal pH for plant growth using an apparatus for maintaining a constant pH of nutrient solution. They pointed out that the optimal pH range was from 5. 5 to 6. 5.

It has been said that the optimal pH for plant growth varies with nitrogen sources, *i.e.* higher in an ammonium type nutrient solution than in a nitrate type nutrient solution[6,13,24,26,27]. However, in preliminary experiments when an apparatus for maintaining constant pH in nutrient solutions was developing, we found that cucumber plants grew rapidly in an ammonium type nutrient solution when the pH was kept at 5. 0[17,18]; although cucumber plants had been considered to prefer nitrate to ammonium nitrogen[14,15,23].

Thus, experiments were undertaken on the optimal pH of both sources of nitrogen prior to the careful experiments on the effect of nitrogen sources on plant growth. In this paper, optimal pH for growth and mineral uptake in six plant species were re-evaluated by the constant pH solution culture.

## MATERIALS AND METHODS

The test plants were rice (*Oryza sativa* L. subsp. *Japonica*; cv. Akebono), barley (*Hordeum vulgare* L. ; cv. Akashinriki), corn (*Zea mays* L. ; cv. Nagano 1), bean (*Phaseolus vulgaris* L. ; cv. Masterpiece), cucumber (*Cucumis sativus* L. ; cv. Chihai) and tomato (*Lycopersicon esculentum* Mill. ; cv. Beiju). Culture experiments were carried out in a greenhouse under natural light conditions. Table 1 shows the season, days of culture and temperature conditions. Tomatoes were tested during off season (Tomato is a summer crop in Japan), because this plant suffers easily from ammoniacal disorders[8,16] and the growth was stunted in

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TABLE 1. The season, duration and temperature of the culture experiments

| Plants | N Source | Days in[*1] nursery | Initial and final days of experiment | Experimental period (days) | Temperature[*2] | |
|---|---|---|---|---|---|---|
| | | | | | Room | Solution |
| Rice | $NH_4$ | 10 | Aug. 14～Sept. 9 | 26 | 22-(32)-37 | |
| | $NO_3$ | 12 | Sept. 11～Oct. 8 | 27 | 13-(28)-38 | |
| Barley | $NH_4$[*3] | 10 | Dec. 21～Jan. 24 | 34 | 11-(18)-31 | 12-(17)-23 |
| | $NO_3$[*4] | 13 | Jan. 31～March 5 | 33 | 12-(19)-33 | 14-(19)-24 |
| Corn | $NH_4$[*5] | 6 | July 23～Aug. 10 | 18 | 21-(31)-38 | |
| | $NO_3$[*6] | 7 | July 3～July 21 | 18 | 21-(30)-37 | |
| Bean[*7] | $NO_3$ | 7 | July 4～July 18 | 14 | 24-(32)-38 | 24-(27)-32 |
| Cucumber | $NH_4$[*5] | 6 | July 23～Aug. 10 | 18 | 21-(31)-38 | |
| | $NO_3$[*6] | 7 | July 3～July 22 | 19 | 21-(31)-37 | |
| Tomato | $NH_4$[*3] | 12 | Dec. 21～Jan. 29 | 39 | 11-(18)-31 | 12-(17)-23 |
| | $NO_3$[*4] | 17 | Jan. 31～March 4 | 32 | 12-(19)-33 | 14-(19)-24 |
| Rice[*8] | $NH_4$-4 / $NO_3$-1 | 10 | July 20～Aug. 18 | 29 | 25-(33)-38 | 25-(28)-34 |
| Corn[*8] | $NH_4$-1 / $NO_3$-4 | 7 | June 30～July 19 | 19 | 23-(32)-37 | 24-(28)-33 |

*1 Days from seeding to transplanting.
*2 indicates Minimum-(Average at 9 : 00)-Maximum temperature during culture experiments. Room temperature was measured by a recording thermometer at 1 m height. Solution temperature was measured by a thermistor sensor dipped into the nutrient solution.
*3~6 Both plant species indicated the same number were cultured in the same apparatus. Periods of culture experiments are not always the same in both plants in the mixed culture, when harvest day is different.
*7 Bean plants were grown mixed with cucumber plants, though data on cucumber cultures were excluded from this paper.
*8 These are special culture experiments for growth and mineral uptake at the low pH range. Nutrient solutions for rice plants and for upland crops were used respectively.

summer with an ammonium type nutrient solution even though the constant pH solution culture was undertaken (confirmed by other experiments)[20,21].

Greenhouse temperature was controlled to 10°C or more in winter and to 35°C or less in summer by air conditioning. The temperature of the nutrient solution in the culture pots was maintained at 35°C or less with running tap water in summer.

Plants were grown in nutrient solutions at constant pH. The pH was adjusted stepwise every 0.6 pH units from 3.6 to 6.6. Constant pH was maintained by employing an apparatus that automatically set the pH of nutrient solution within the range of $\pm$ 0.1 pH unit throughout the experimental period. The details of the apparatus were described elsewhere[17,18].

TABLE 2. Composition of nutrient solutions

| | | | |
|---|---|---|---|
| Composition of N, P and K | | | |
| 1. Ammonium type | | 2. Nitrate type | |
| [$NH_4$-N 5 mM] | | [$NO_3$-N 5 mM] | |
| $(NH_4)_2SO_4$ | 2.5 mM | $KNO_3$ | 4.0 mM |
| $KH_2PO_4$ | 1.0 | $NaNO_3$ | 1.0 |
| $K_2SO_4$ | 1.5 | $NaH_2PO_4$ | 1.0 |
| 3. for rice plants | | 4. for upland crops | |
| {$NH_4$-N 4 mM, $NO_3$-N 1 mM} | | {$NH_4$-N 1 mM, $NO_3$-N 4 mM} | |
| $(NH_4)_2SO_4$ | 2.0 mM | $KNO_3$ | 4.0 mM |
| $KNO_3$ | 1.0 | $NH_4H_2PO_4$ | 1.0 |
| $KH_2PO_4$ | 1.0 | | |
| $K_2SO_4$ | 1.0 | | |
| Composition of the other elements | | | |
| $CaCl_2$ 2.0 mM | $MgSO_4$ 1.0 mM | $SiO_2$*1 | 2.0 mM |
| Fe*2 | 1.0 ppm | Zn($ZnSO_4$) | 0.05 ppm |
| B($H_3BO_3$) | 0.5 | Cu($CuSO_4$) | 0.02 |
| Mn($MnCl_2$) | 0.5 | Mo($Na_2MoO_4$) | 0.01 |

*1 Silica was added to the nutrient solution when rice was planted.
*2 Iron (1 ppm) was added every other day as citrate.

Numbers 1 to 4 indicate the ammonium type nutrient solution, the nitrate type nutrient solution, the nutrient solution for rice plants and the nutrient solution for upland crops, respectively.

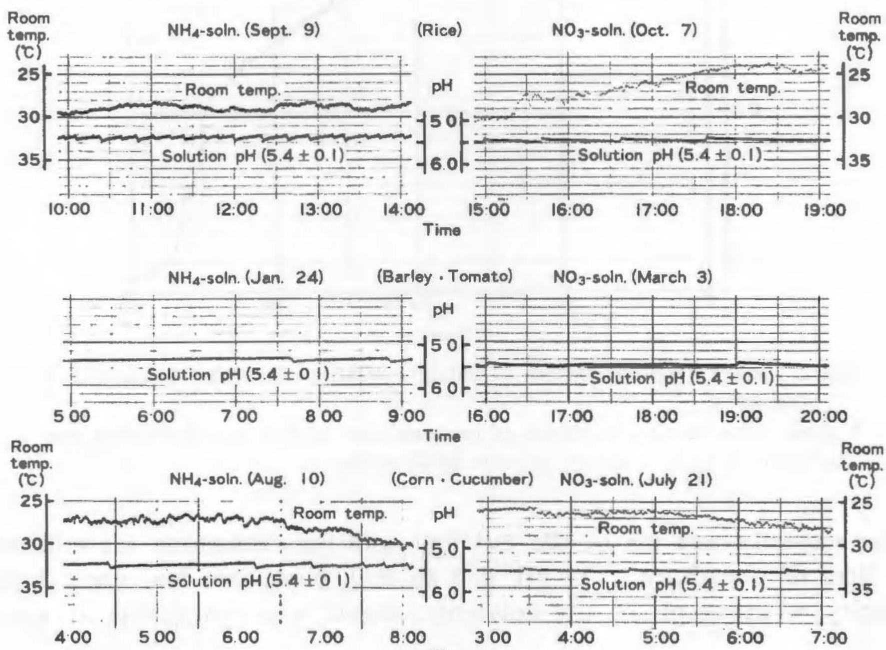


FIG. 1. Examples of pH regulation in latter periods of culture experiments.

Nutrient solutions used in this investigation are described in Table 2. They were ammonium type nutrient solution, nitrate type nutrient solution, nutrient solution for rice plants, and nutrient solution for upland crops.

Whenever rice plants were cultured, silica was added into all the types of nutrient solutions. Nutrient solutions were prepared with deionized water, and renewed once a week. The solutions were continuously aerated throughout the experimental period.

Fig. 1 shows examples of pH regulation at 5.4 by the apparatus. The accuracy of the regulation was satisfactory, even in the range of low buffer capacity of phosphate. Fig. 2 shows the relationship between

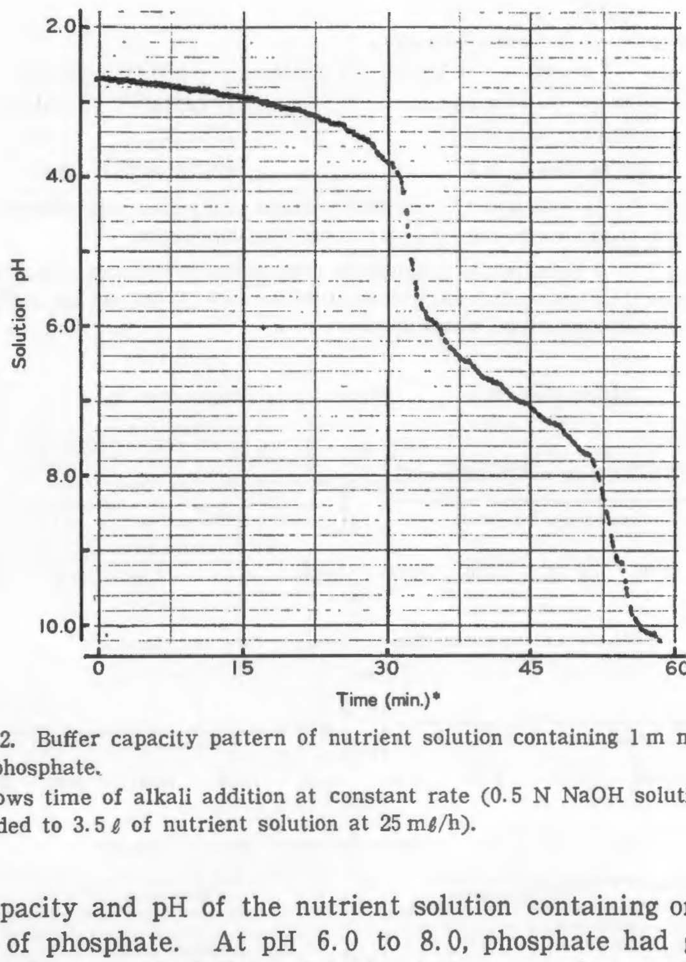


FIG. 2. Buffer capacity pattern of nutrient solution containing 1 m mol/$\ell$ of phosphate.

* shows time of alkali addition at constant rate (0.5 N NaOH solution was added to 3.5 $\ell$ of nutrient solution at 25 m$\ell$/h).

buffer capacity and pH of the nutrient solution containing one millimole per liter of phosphate. At pH 6.0 to 8.0, phosphate had great buffer capacity, whereas pH of the nutrient solution was changeable at 4.0 to 6.0.

After each culture period, plants were harvested, rinsed with tap water and deionized water successively, then separated into tops and roots. The dry matter was weighed after oven-drying at 90° to 100°C, then ground and submitted for mineral analysis. After dry ashing, the ash was dissolved with dilute hydrochloric acid.

Potassium, calcium, magnesium, iron, manganese, zinc and copper were determined by atomic absorption spectrophotometry. Phosphate was measured by the molybdovanadophosphate method. Total nitrogen was determined by the Kjeldahl method. Nitrate-fed plants were digested by the Gunning modified Kjeldahl method.

As for rice and corn plants which were not stunted at pH 3.6, the pH range to be examined was lowered to 3.0 (low pH experiment). The nutrient solutions used in the low pH experiments were for rice plants and for upland crops in Table 2.

Mineral uptake was evaluated by their contents of plants. The contents of phosphate and micronutrients in plant roots were eliminated

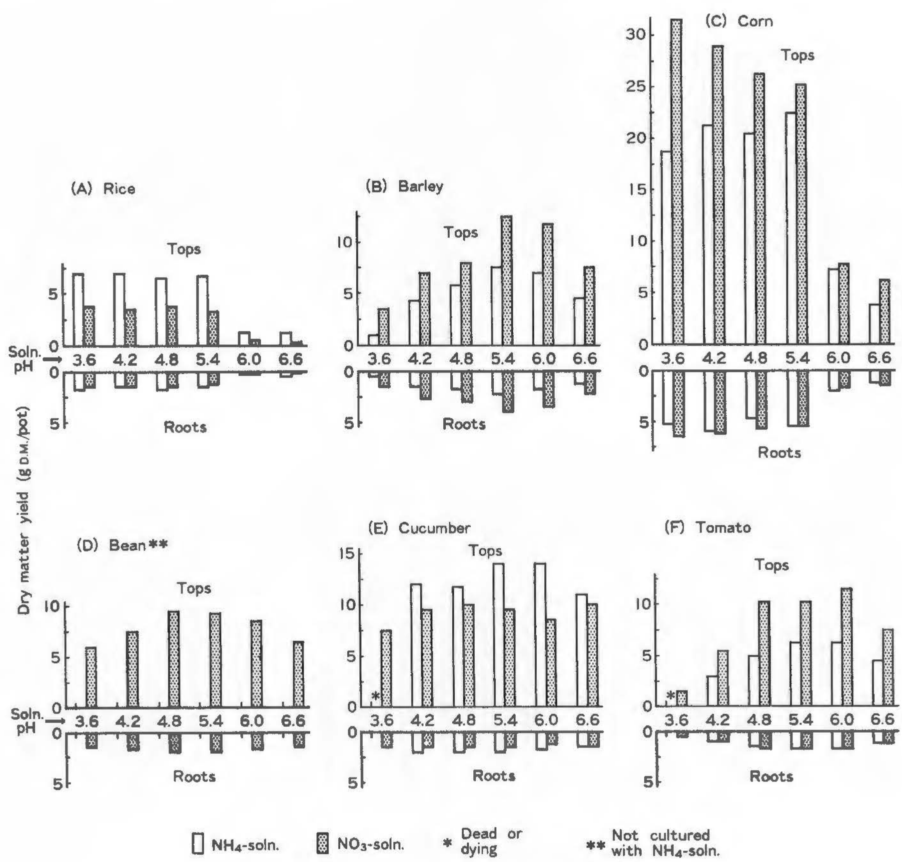


FIG. 3. Effect of solution pH on plant growth.


PLATE 1. Symptom of ammonium poisoning in cucumber leaves exposed to low pH and $NH_4$-N feeding.

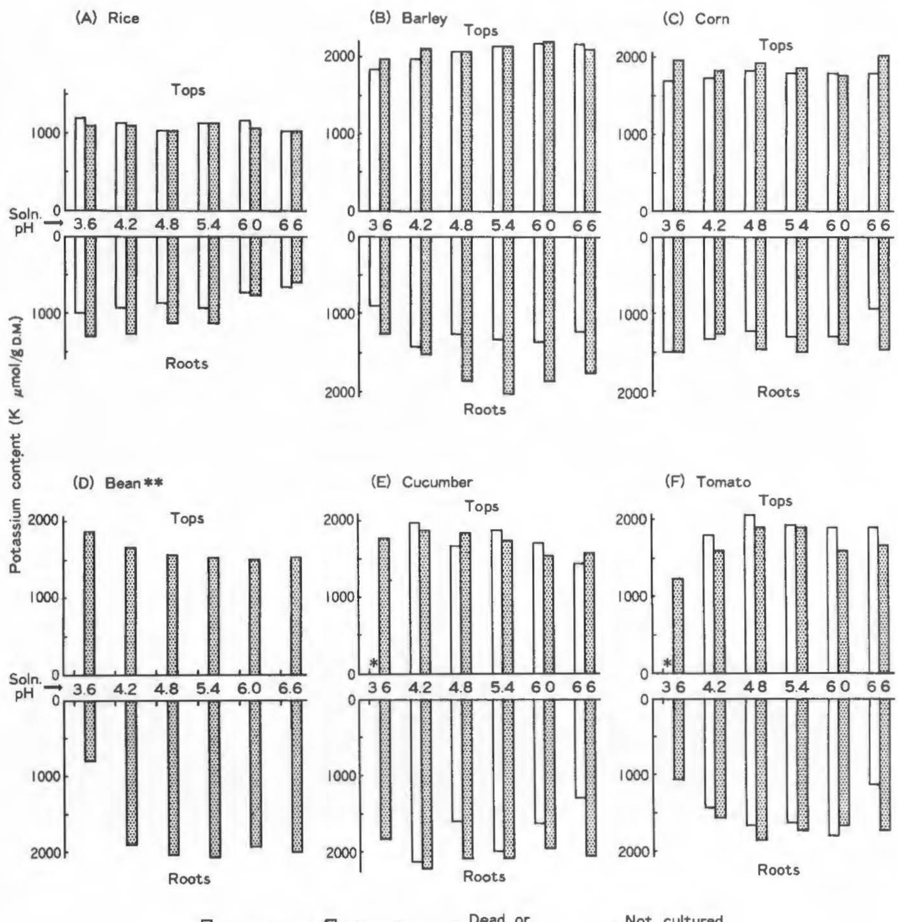


FIG. 4. Effect of solution pH on potassium content of plants.

from Fig. 7 to 11, because these elements were easily deposited on the surface of roots. The contents of total nitrogen in plant roots were also excluded from Fig. 12, because the test sample was not always sufficient for analysis.

## RESULTS

Fig. 3 shows the effect of solution pH on the growth of the test plants. The optimal pH for plant growth was about 5.5 for both sources of nitrogen, with the exceptions of rice and corn plants.

In rice and corn, pH 5.5 seemed to be the upper limit of the optimal pH for growth, because these plants suffered easily from iron deficiency at high pH. However, such plants were highly resistant to low pH, and the growth of such plants was rapid at pH 3.6 or above.

In other plants, growth was stunted at pH 3.6 especially in the ammonium type nutrient solution. Cucumber leaves show a characteristic appearance which looks like a cup or pan (Plate 1). This appearance has been reported as a symptom of ammonium injury by Matsumoto *et al.*[14].

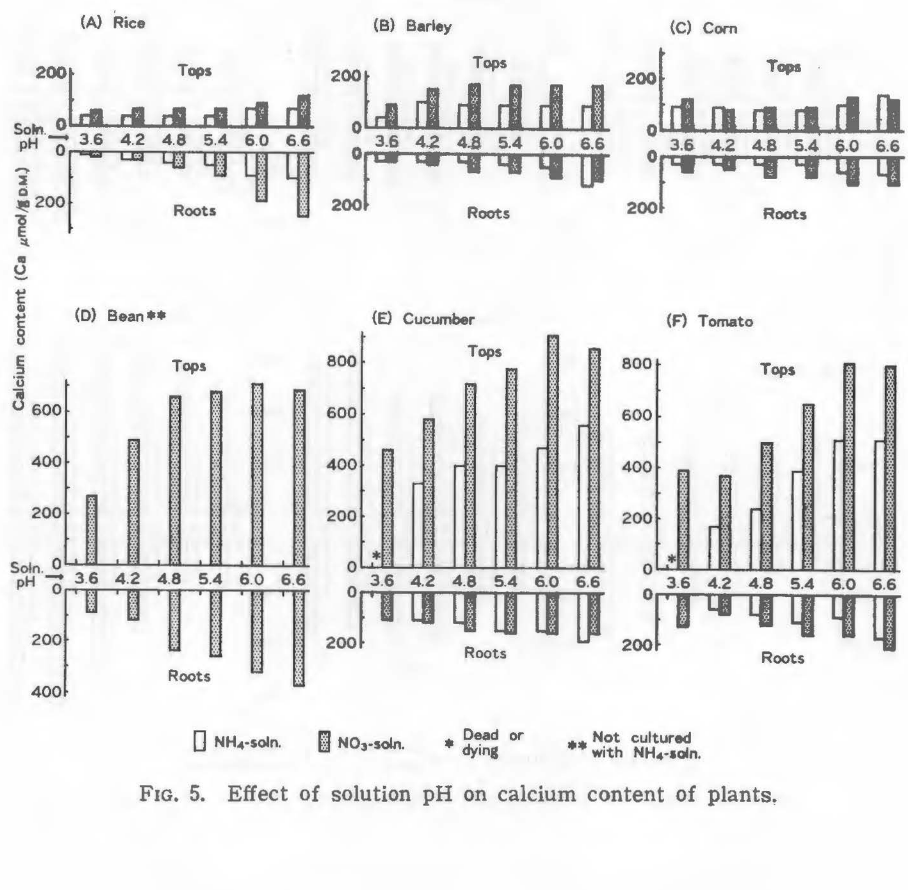


FIG. 5. Effect of solution pH on calcium content of plants.

Fig. 4 to 12 show the effects of solution pH on the contents of potassium, calcium, magnesium, manganese, zinc, iron, copper, phosphorus and total nitrogen in plants.

As indicated in Fig. 5 to 8, the pH of nutrient solution had a remarkable influence on the uptakes of manganese, zinc, calcium or magnesium in plants. Their uptakes were markedly depressed in the nutrient solution of low pH. The optimal pH for the uptake of these cations were higher than that for the plant growth.

Fig. 4 and 12 show the uptake of monovalent ions such as potassium, ammonium and nitrate. Their uptakes were less affected or almost unaffected with either nitrogen source within the pH range examined. The copper content in plants is shown in Fig. 10. The uptake of copper was less affected by the solution pH or the nitrogen sources. The uptake of phosphate also appeared to be not influenced in pH of nutrient solutions from 4.2 to 5.4 (Fig. 11). As indicated in Fig. 9, the optimal pH seemed to be a little lower for the uptake of iron than that for plant growth.

The contents of manganese, zinc, copper and phosphate were higher

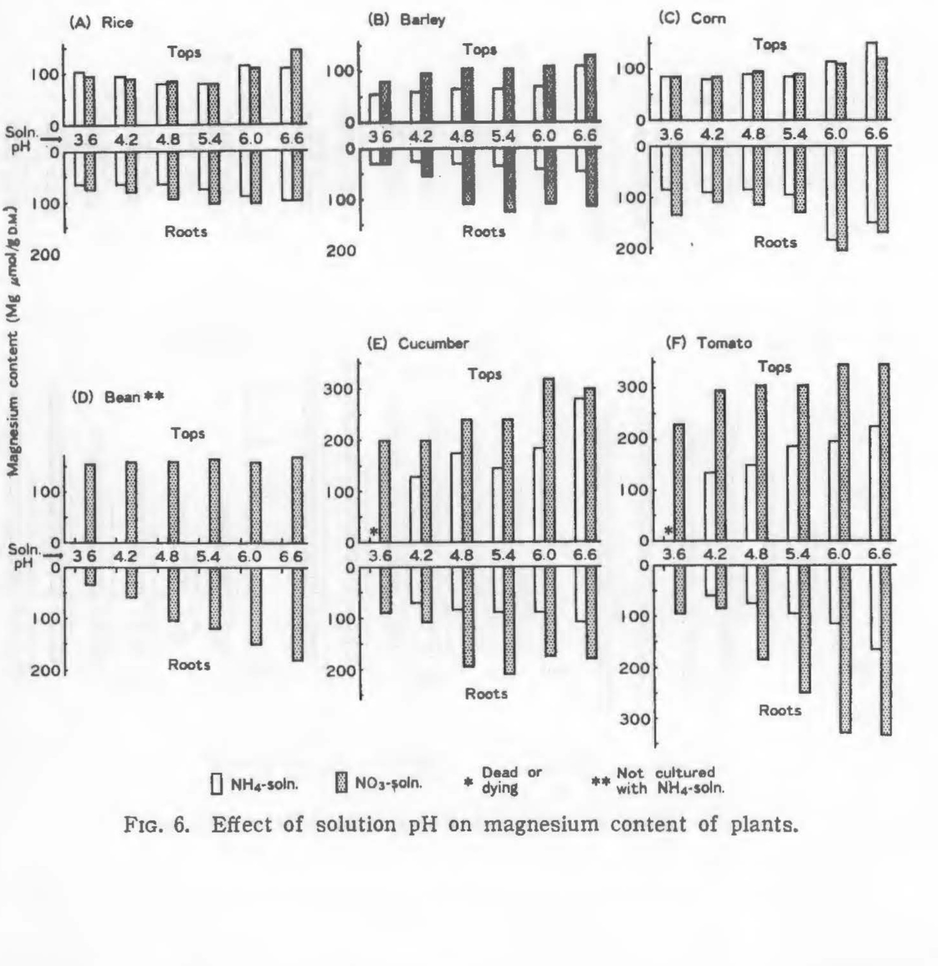


FIG. 6. Effect of solution pH on magnesium content of plants.

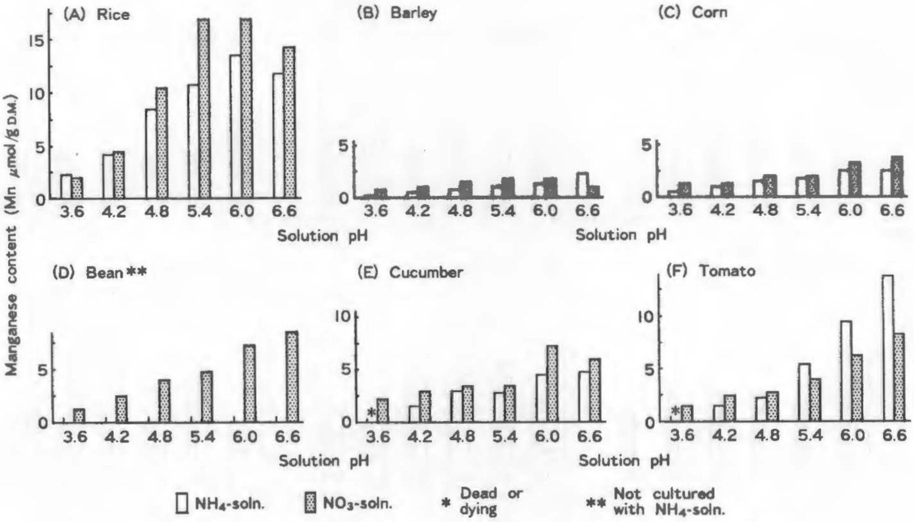


FIG. 7. Effect of solution pH on manganese content of plant tops.

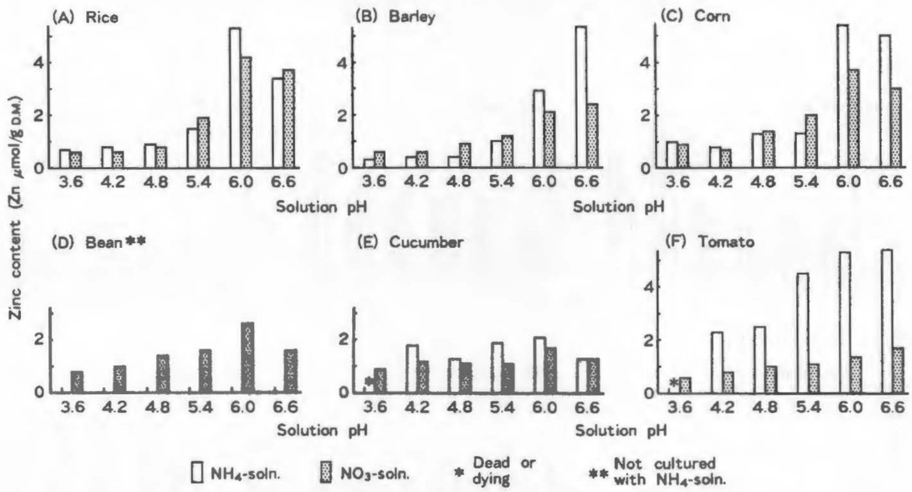


FIG. 8. Effect of solution pH on zinc content of plant tops.

in roots than in tops. An example is shown in Table 3. The iron content of the roots reached about 100 times that of the tops.

To examine the effect of low pH on growth and mineral uptake of rice and corn, the pH range was lowered. The results are illustrated in Fig. 13. Even in such acid-resistant plants which were not stunted at pH 3.6 (Fig. 3), growth was retarded at pH 3.0. The growth of corn was retarded more than that of rice under such conditions. In another culture experiment (Table 4), it was also confirmed that pH 2.7 was fatal condition to rice plants.

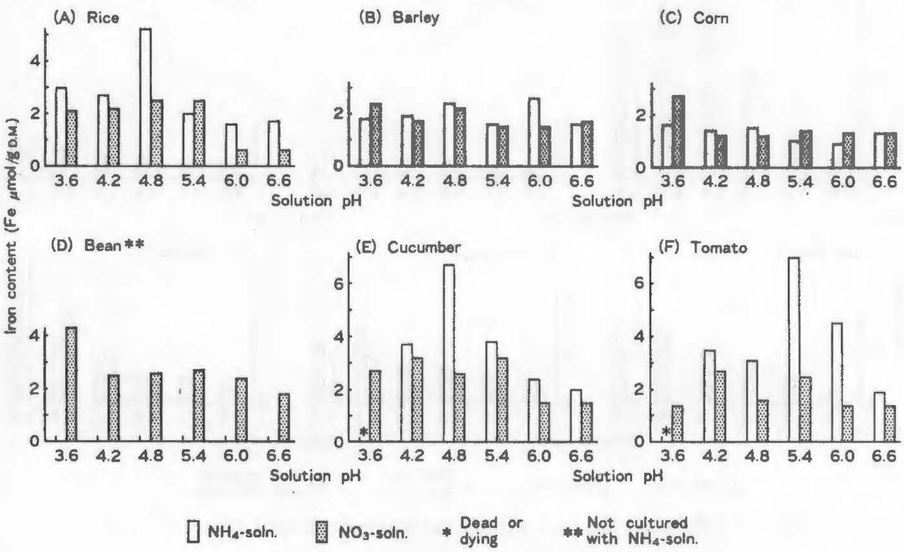


FIG. 9. Effect of solution pH on iron content of plant tops.

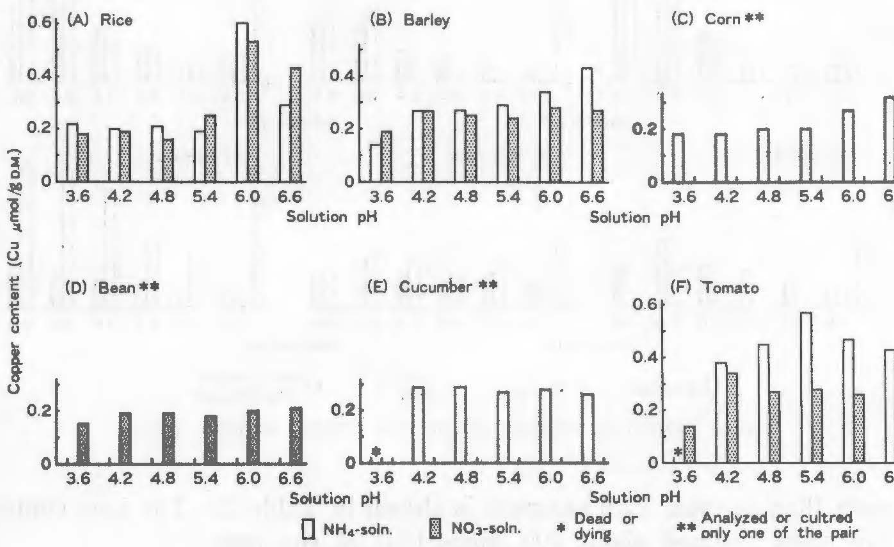


FIG. 10. Effect of solution pH on copper content of plant tops.

The uptake of calcium and magnesium decreased at pH 3.0, while the uptake of potassium did not change to pH 3.0. Phosphate uptake increased slightly in this pH rnage.

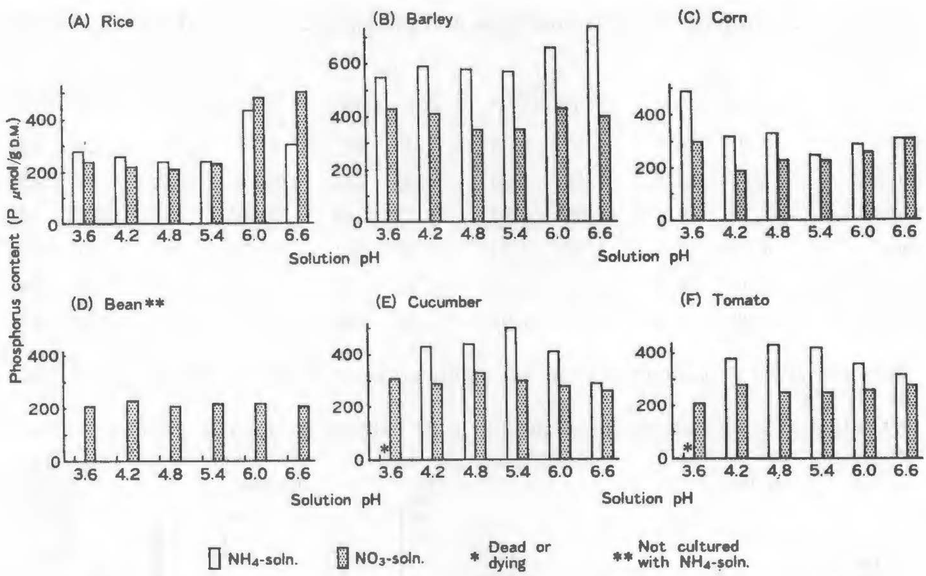


FIG. 11. Effect of solution pH on phosphorus content of plant tops.

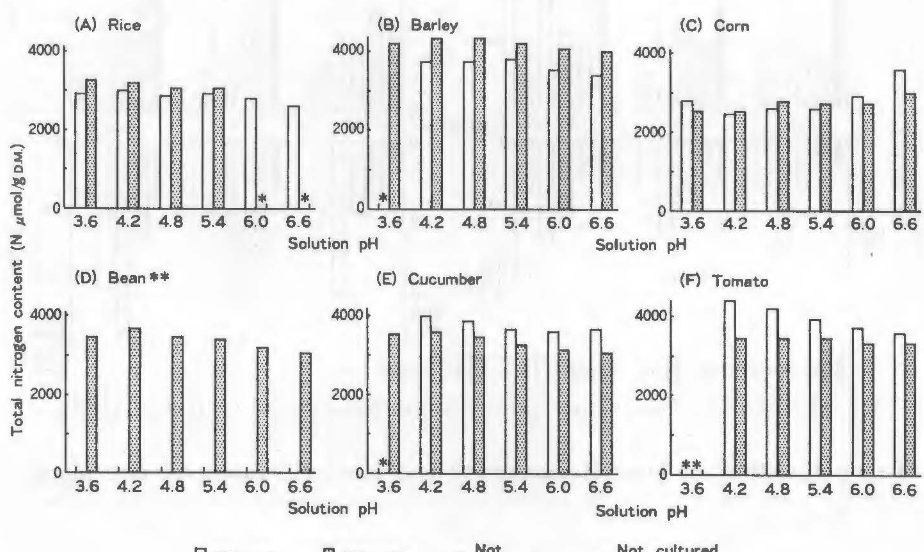


FIG. 12. Effect of solution pH on total nitrogen content of plant tops.

## DISCUSSION

The optimal pH of the nutrient solution for plant growth has been demonstrated to be dependent on the nitrogen sources, *i.e.* higher in the ammonium type nutrient solution and lower in the nitrate type nutrient solution[6,13,24,26,27]. However, in the present investigation, the

TABLE 3. Examples of micronutrients and phosphate content of tops and roots

| | Mn | | Zn | | Fe | | Cu | | P | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Tops | Roots | Tops | Roots | Tops | Roots | Tops | Roots | Tops | Roots |
| Rice | 17.0 | 16.9 | 1.93 | 2.40 | 2.46 | 402 | 0.25 | 3.76 | 226 | 496 |
| Barley | 1.71 | 11.1 | 1.18 | 2.30 | 1.46 | 232 | 0.24 | 1.44 | 348 | 420 |
| Corn | 1.98 | 8.3 | 1.95 | 4.53 | 1.42 | 60 | 0.20* | 1.01* | 232 | 386 |
| Bean | 4.76 | 22.5 | 1.62 | 9.83 | 2.68 | 181 | 0.18 | 1.34 | 216 | 742 |
| Cucumber | 3.57 | 6.4 | 1.11 | 5.86 | 3.15 | 81 | 0.27* | 2.34* | 303 | 404 |
| Tomato | 3.98 | 18.5 | 1.07 | 5.01 | 2.45 | 244 | 0.28 | 1.72 | 251 | 455 |

Data are given as $\mu$ moles/g D. W. for plants grown in nitrate type nutrient solution at pH 5.4.

* Content in plants grown in ammonium type nutrient solution at pH 5.4.

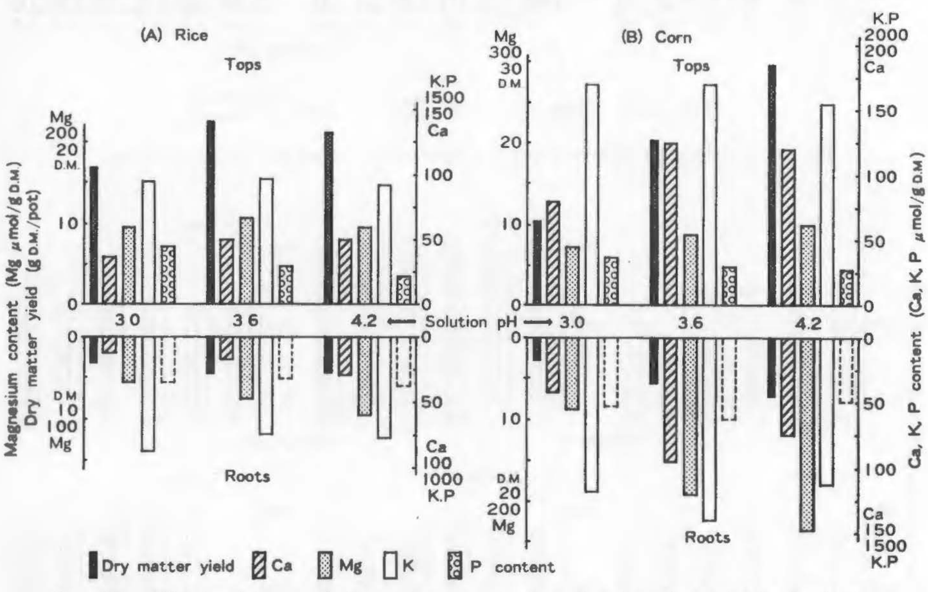


FIG. 13. Effect of low solution pH on growth and mineral content of plants.

TABLE 4. Effect of low pH on growth and mineral content of rice plants

| Plant parts | | Tops | | | Roots | | |
|---|---|---|---|---|---|---|---|
| pH | | 3.3 | 3.9 | 4.5 | 3.3 | 3.9 | 4.5 |
| Dry matter yield (g/pot) | | 8.31 | 12.0 | 12.6 | 1.97 | 3.10 | 3.23 |
| Content ($\mu$moles/g D. M.) | K | 978 | 915 | 856 | 923 | 866 | 885 |
| | Ca | 41.7 | 55.6 | 53.6 | 12.7 | 18.8 | 28.7 |
| | Mg | 124 | 97.2 | 75.9 | 66.9 | 88.2 | 93.9 |
| | P | 569 | 263 | 230 | 506 | 392 | 429 |

The culture experiment was carried out from September 8 to October 13. Rice plants were also examined at pH 2.7, but were not grown and died for strong acidity within 3 weeks.

optimal pH was approximately 5.5 in either nitrogen source, except for rice and corn plants (Fig. 3). These results seem to be similar to those obtained by Islam *et al.*[12] with ginger and other plants.

The results shown in the present paper are clearly different from the data in many reports described above[6,13,24,26,27]. Consequently, the use of an automatic apparatus for maintaining a constant pH is important in culture experiments.

Rice and corn plants were stunted in the nutrient solutions of high pH, because these plants suffered easily from iron deficiency. The effect of iron sources may be checked in plants grown in high pH with a more stable iron sources (*e.g.* EDDHA salt)[12] in comparison with citrate. However, the similarity of iron efficiencies between citrate and cheletes in high pH range has been confirmed by recent experiments (unpublished data). The cause for the small difference in optimal pH range in the present investigation and in the work by Islam *et al.*[12] may be attributed to the difference in experimental conditions.

In the low pH range, plant growth in the nitrate type nutrient solution is not retarded so much as that in the ammonium type nutrient solution (Fig. 3). This agrees with the results that nitrate nitrogen has a wide optimal pH in plant growth as compared with ammonium nitrogen[24,27]. In cucumber and tomato plants, growth was highly stunted in the ammonium type nutrient solution at pH 3.6; while, in the nitrate type nutrient solution at pH 3.6, these plants increased in size slightly, though very low rate (Fig. 3). The explanation might be that in these plants, the assimilation of ammonium to organic compounds was retarded, and that the ammonium concentration in low pH plants fed with ammonium was increased, then the growth was retarded more owing to the toxicity of ammonium.

On cation uptake, an optimal pH was found in both nitrogen sources at pH higher than 5.5. The cation uptake was generally depressed in the nutrient solution of lower pH, except for potassium (Fig. 4 to 8). However, in iron uptake, the optimal pH fell to lower than 5.0.

Solution pH has an important effect on the uptake of manganese and zinc (Fig. 6 and 7). These results were similar to the results which showed increased manganese content in high pH nutrient solution[29,30,32], intensified manganese poisoning in the nutrient solution of high pH[31], or depressed zinc uptake at low pH range[3,25].

Nitrogen uptake was found to be constant within the pH range tested (Fig. 12). This result clearly indicates that the uptake of nitrogen was similar in both nitrogen sources in long-term constant pH solution culture. On the contrary, it has been generally thought that the uptake of ammonium was more rapid than that of nitrate in short-term experiments[34], or that the uptake of ammonium was rapid in high pH medium and that of nitrate was rapid in low pH medium in long-term culture experiments[4].

However, it has been also demonstrated that the effect of external pH was minimized with both nitrogen sources when concentrations were low[34], or that the difference in nitrate uptake between pH 4.4 and 7.0 is very small[1]. These results are similar to the results of the present experiments.

The low pH culture experiments show that rice plants were more resistant to low pH than corn plants (Fig. 13). This is contrary to the results of Tanaka *et al.*[28] who also used an apparatus for constant pH solution culture. Such disagreements might be due to differences in culture methods. Rice and corn were grown in separate apparatus in the present investigation, while they were grown in the same apparatus with many other species of plants in the experiment of Tanaka *et al.*[28]. Such a difference in culture system brings about differences in competition among plant species in mineral uptake and in light-receiving.

In mineral uptake of rice and corn plants at low pH range, potassium uptake was not depressed at pH 3.0 (Fig. 13). This is in contrast to the results which demonstrated that the uptake of potassium decreases from pH 4 to 3 even when nitrate was the sole source of nitrogen[33].

## SUMMARY

Rice, barley, corn, bean cucumber and tomato plants were grown in cultures under constant pH condition to re-evaluate the optimal pH for growth, by use of an apparatus for automatically controlled the pH of the nutrient solution. The solution pH was set at six stages that rose stepwise at 0.6 intervals from pH 3.6 to 6.6 ($\pm$0.1 pH). Plant growth and mineral uptake were measured and compared to solution pH and nitrogen sources. The results were as follows:

1) The optimal solution pH for plant growth was about 5.5 for both ammonium and nitrate nutritions.

2) The optimal pH for rice and corn plants was slightly lower, because those plants were sensitive to iron deficiency at high pH. However, the optimal pH range of these plants did not differ in the presence of ammonium or nitrate ion.

3) The effect of solution pH on mineral uptake was examined by mineral contents of plants. The solution pH had little influence on the uptake of ammonium, nitrate, potassium or phosphate ions.

4) The uptake of manganese, zinc, calcium and magnesium ions were markedly depressed in the low pH conditions.

5) The growth and mineral uptake of rice and corn plants were examined in the lower pH conditions (3.0 to 4.2), because these plants were highly resistant to low pH conditions. When the solution pH was 3.0, growth was slightly retarded in both plants. Rice plants were more resistant to low pH than corn plants.

6) The calcium and magnesium uptakes of rice and corn plants were depressed in the nutrient solution at pH 3.0, whereas the potassium uptake of either of these plants was not depressed, even at pH 3.0. In this pH range, phosphate uptake was slightly stimulated.

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