# GROWTH OF THE AZALEA AS INFLUENCED BY AMMONIUM AND NITRATE NITROGEN

by

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A THESIS

submitted to

OREGON STATE COLLEGE

in partial fulfillment of
the requirements for the
degree of

MASTER OF SCIENCE

June 1956

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Date thesis is presented June 28, 1955

Typed by Betty M. Cohen

## ACKNOWLEDGMENTS

The author wishes to express his appreciation to Dr. A. N. Roberts for assistance and guidance during the course of this study and to Dr. A. N. Roberts and Professor Henry Hartman for their constructive criticism of the manuscript.

M.S.C., Jr.

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# GROWTH OF THE AZALEA PLANT AS INFLUENCED BY AMMONIUM AND NITRATE NITROGEN

## INTRODUCTION

The plant family Ericaceae contains a large number of species, many of which are characterized by their adaption to acid, organic soil types. Of these plants, the azalea is one that has become economically important as a greenhouse plant for forcing and as a nursery item for landscape planting.

Although there are few papers to be found in research journals on azalea nutrition, many articles dealing with this problem have appeared in trade papers and other semi-popular publications. Such articles have discussed the problems involved in attempting to grow azaleas at near alkaline soil reactions and often give soil acidifying techniques. The importance of adequate drainage and aeration around the root system is also stressed. When these conditions are not provided, the azalea plant often exhibits an interveinal yellowing of the leaves, which in most cases is attributed to a deficiency of available iron and is termed "iron chlorosis."

In commercial greenhouse practice, acid peat moss with various amendments is commonly used as a growing medium for azaleas, since it comes closest to providing the required root environment essential for satisfactory growth. However, under these seemingly ideal conditions, problems of nutrition may still develop and iron sprays, or incorporations of iron into the growing medium, are used in an attempt to correct the chlorotic symptoms which often appear. The use of chelated iron compounds has been much publicized in this regard and these compounds have shown promise in at least temporarily correcting iron chlorosis by providing a readily available form of iron to the plant. However, it is questionable in many cases whether the chlorotic condition ascribed to a deficiency of iron is a direct result of a shortage of available iron at the root surface or is the indirect result of an unbalance of other nutrient-elements or soil conditions which render the iron non-functional in the plant tissues.

Recent research with the blueberry and other related plants, has shown that nitrate nitrogen may be harmful and ammonium nitrogen under some conditions may be essential for the growth of certain of these ericaceous species. The fact that blueberry plants grown on nitrate nitrogen develop typical iron chlorosis symptoms, while those on ammonium nitrogen do not, has been interpreted (Cain, 4, pp.161-166) as indicating a close relationship between nitrogen and iron nutrition. Cain concluded that the superiority of ammonium nitrogen in this regard is not an effect on the availability of iron in the solution or absorption by the roots, but on internal function, since plants receiving nitrate nitrogen and showing iron deficiency symptoms contain as much or more iron in their foliage as those showing no symptoms of deficiency. He was of the opinion that further work on the interrelationships between soil acidity, form of nitrogen and iron nutrition were needed before a clear picture of these factors could be formulated.

It was with this concept in mind that this study was undertaken for the purpose of reaching a better understanding of the influence of ammonium and nitrate nitrogen on the growth of the azalea plant. A series of experiments was conducted in an attempt to show the conditions that favor one source of nitrogen over the other as evidenced by growth response and the development of chlorotic symptoms. The factors considered in this regard were the form and concentration of nitrogen, the hydrogen ion concentration, the concentration of other ions and the interrelationship of these factors. The possible influence of light and root temperature on the expression of plant response to these conditions was also given consideration.

## REVIEW OF LITERATURE

Foret and Volz (9, pp.636-637) concluded that iron becomes unavailable in soil by reacting with phosphorous, when the hydrogen ion concentration is below pH 4.00, and results in chlorosis of the azalea. Nieuwdrop (20) found the Rhododendron catawbiense would not flourish above pH 5.00 or in dry, acid soils. Barnette and Mowry (2, pp.72-77) grew azaleas in soils adjusted to give a range in pH. They found that plants in soils below pH 5.00 made a slow but healthy growth while those grown between pH 5.00 and 6.00 made more vigorous growth. Plants grown above pH 7.00 made very little growth and the foliage was chlorotic.

Stuart (28, pp.210-214) observed that azaleas grown in sand culture became chlorotic when calcium nitrate was used as a source of nitrogen, but the chlorotic condition could be corrected by adding ammonium sulfate to the solutions. The chlorosis was attributed to the change in pH brought about by the residual ion associated with the nitrogen. Spencer and Shive (27, p.433) obtained similar results with Rhododendron ponticum when the nutrient solutions contained large amounts of calcium nitrate. Leaf analysis of azaleas by Twigg and Link (31, pp.374-375) showed that the requirements of these plants for phosphorous, potassium, calcium and magnesium were low compared to most horticultural plants.

Nutrition studies have been made on several other members of the family Ericaceae, which appear to have nutritional requirements similar to those of azaleas. Addoms and Mounce (1, p.665) found ammonium nitrogen produced more runner growth on cranberry plants than did nitrate nitrogen. Cain (4, pp.161-166), cited previously, has reviewed the nitrogen nutrition of the blueberry plant in conjunction with his study of the nutritional requirements of this crop.

## NOTE

Since the completion of these studies, Cain (5, pp.61-69) has presented data which show a relationship between blueberry leaf chlorosis, the accumulation of the basic cations calcium, magnesium and potassium and an increase in the pH of the leaf tissue. These results and Cain's interpretation of them are especially significant in the light of the results and interpretations presented in this paper as they apertain to the azalea.

# MATERIALS AND METHODS

The detailed procedure and the materials used in studying each phase of the problem are given at the beginning of each specific experiment reported. Variations in solution composition and experimental methods in the several experiments necessitated such an arrangement. However, certain materials and methods were common to all experiments and are treated under this heading.

The evergreen azalea Hexe was used in all the greenhouse experiments. In the experiment comparing ammonium and nitrate nitrogen at various root temperatures, deciduous Mollis seedlings, as well as Hexe, were used. Several other evergreen varieties were employed for certain preliminary investigations to compare varietal behavior to various treatments.

All the plants were grown in sand culture, with the exception of one field trial and a series of plants grown in sawdust. A rather coarse grade of sand known in the building trade as Del Monte No. 20 was used. A comparison of Del Monte sand with sand blast sand (40 mesh) was obtained in several experiments. The sand was placed in glazed crocks of 1.3 liter capacity, especially designed for sand culture work (cf. Plate 3). The glazed crocks designed for the root temperature tanks had a capacity of two gallons (cf. Plate 1). The construction and arrangement of the temperature tanks are described by Mellenthin (19, pp.6-8).

The nutrient solutions were prepared from Bakers' analyzed reagents. Hoagland's (13, p.31) four-salt, nitrate solution and a similar solution containing ammonium sulfate as the nitrogen source were used in several of the experiments. The composition of the two solutions are given in Table 1. The pH of these solutions ranged from 6.50 to 7.00. Variations of these two solutions were used in several experiments and are described under those specific experiments. Hoagland's (13, p.31) minor element and iron solution was applied with all nutrient solutions. Solution concentration and frequency of application are discussed under each individual experiment. Solutions were applied by the slop culture method. Night temperatures of 55 to 65°F. were maintained throughout all the greenhouse experiments.

The pH determinations were made with the glass electrode. The method used to determine pH of the leaf tissue was essentially the same as that used by Kramer (17, p.30). A weighed amount of fresh leaves plus a small amount of sand were ground in a mortar and diluted to five times the leaf weight with distilled water. The pH was read on the resulting slurry. All pH determinations of plant tissue were made between 7:00 and 10:00 p.m. In the case of the pH determinations on solution leachate, the first 100 ml. collected from the crock after the solutions were applied was used.

The chlorosis and nitrogen-deficiency index are an average of the several plants in each treatment. These are arbitrary ratings from 1 to 5 with increasing severity of leaf deficiency symptoms. Plants designated 1 showed no symptoms and those designated 5 showed severe deficiency symptoms.

Analysis of variance was used, as needed, in determining the significance of data presented in the various tables.

Table 1. Composition of full strength (1.0) ammonium and nitrate solutions (in ppm).

| Solution | N | P | K | Ca | Mg | $SO_4$ | Cl |
|---|---|---|---|---|---|---|---|
| $NO_3$ | 210 | 31 | 234 | 200 | 49 | 192 | 0 |
| $NH_4$ | 210 | 31 | 234 | 200 | 49 | 864 | 567 |

Hoagland's complete minor element solution was applied at the rate of 1 ml. per liter of solution. One ml. of a 0.5% ferric tartrate solution was added to 1 liter of nutrient solution at time of application.

## PROCEDURE AND RESULTS OF SPECIFIC EXPERIMENTS

Experiment I. The influence of ammonium and nitrate nitrogen on growth and on the development of chlorotic symptoms in azaleas grown at several root temperatures.

This experiment was designed to determine the response of azalea plants to ammonium and nitrate nitrogen, as it might be influenced by root temperature. The general color and condition of the foliage and the production of dry matter in the form of roots, stems, and leaves were used as a measure of response.

One-year-old Hexe and Mollis azaleas were used for this study. The roots were washed and the plants weighed, January 6, 1954, prior to planting in crocks containing Del Monte sand. The fresh weight of the Hexe plants ranged from 30 to 80 grams and the Mollis azaleas from 25 to 47 grams. The temperature tanks were adjusted to 13°, 18°, and 24° C.

Twelve plants of both varieties were grown at each root temperature. Six plants of each variety at the three root temperatures were supplied the nitrate solution and the remaining six were supplied the ammonium solution. The composition of the two solutions are given in Table I. The frequency of solution application was gradually increased and solution strength decreased so that by April each plant was receiving 500 ml. of a 0.25 concentration of solution daily. During the summer months the solutions were applied twice a day.

The plants were removed from the temperature tanks between September 18 and 22, 1954, and separated into stems, leaves and roots. Dry weight determinations were made after drying these samples for two days at 80°C.

Growth response to ammonium and nitrate nutrition, expressed as dry weight production, is presented in Table 2. These data show that the dry weight of leaves, stems and roots of Hexe azaleas receiving ammonium nitrogen was approximately double the weight of plants supplied nitrate at root temperatures of 18° and 24°C. Plants supplied nitrate at 13°C. produced slightly more leaf and stem weight than those supplied ammonium nitrogen, however the latter produced more root growth than the plants supplied nitrate at this temperature. As indicated in Table 2, the Mollis variety showed the same but an even greater response to differences in form of nitrogen and root temperature than did the variety Hexe.

The plants supplied ammonium nitrogen showed a marked response to root temperature. As root temperature was increased from 13° to 24°C., a corresponding increase in growth was obtained with both azalea varieties. The average total dry weight of Hexe azaleas grown at 24°C. was three times that of plants grown at root temperatures of 13°C. In the case of the Mollis variety, this difference amounted to approximately eight times in favor of the highest root temperature.

Plants receiving nitrate nitrogen, on the other hand, showed very little response to root temperature. The dry weight of plants grown at 24°C. was only slightly greater than that of plants grown at the lower root temperatures. Plants supplied nitrate at 13°C. showed a slight advantage in dry weight over those grown at 18°C. The differences in dry weight of plants supplied nitrate at the three root temperatures, however are of doubtful significance.

The development of iron chlorosis was closely related to the form of nitrogen supplied, but only slightly if at all to root temperature. The plants supplied nitrate nitrogen at root temperatures of 13° and 18°C. showed faint symptoms of chlorosis by the first of April. By late June all plants supplied nitrate were chlorotic. This condition was most severe at 18°C. and least severe at 13°C. A series of three iron chelate sprays applied to half the plants had little effect in correcting the chlorotic symptoms. The chlorosis was not evident in plants supplied ammonium nitrogen. A comparison of Plates 1 and 2 will show the effect of nitrogen source and root temperature on the growth and leaf color of the Hexe azalea.

Table 2. Dry weight (grams) production of azalea plants growing at three root temperatures and supplied with ammonium or nitrate nitrogen.*

| Treatment | Hexe Leaves | Hexe Stems | Hexe New Roots | Hexe Total dry weight | Mollis Leaves | Mollis Stems | Mollis New Roots | Mollis Total dry weight |
|---|---|---|---|---|---|---|---|---|
| 13° $NH_4$ | 6.74 | 5.44 | 3.11 | 15.29 | 2.83 | 2.98 | 0.89 | 6.70 |
| $NO_3$ | 7.27 | 6.46 | 1.47 | 15.19 | 3.29 | 3.81 | 0.43 | 7.53 |
| 18° $NH_4$ | 12.39 | 8.29 | 6.96 | 27.64 | 7.08 | 5.61 | 6.49 | 19.18 |
| $NO_3$ | 5.31 | 4.57 | 1.83 | 13.71 | 1.69 | 2.32 | 0.33 | 4.34 |
| 24° $NH_4$ | 20.81 | 13.50 | 10.70 | 45.01 | 23.66 | 16.61 | 14.93 | 55.20 |
| $NO_3$ | 8.53 | 7.37 | 5.87 | 21.77 | 2.38 | 3.02 | 2.77 | 8.17 |

*Each figure is an average of 6 plants


Plate 1. Growth response of Hexe azaleas to nitrate nitrogen at root temperature of 13°, 18° and 24°C.


Plate 2. Growth response of Hexe azaleas to ammonium nitrogen at root temperatures of 13°, 18° and 24°C.

Experiment II. The influence of ammonium, nitrate, and hydrogen ion concentration on growth and on the development of chlorotic symptoms in the azalea plant.

This study was undertaken to determine the influence of and the interrelationship between ammonium, nitrate and hydrogen ion concentration on growth and on the development of chlorotic symptoms in the azalea plant. The possible relationship of light intensity to the expression of these responses was also considered.

Rooted cuttings of Hexe azaleas were planted July 6, 1954, in the 1.3 liter glazed crocks filled with Del Monte white sand. The ammonium and nitrate solutions given in Table 1 were diluted to 0.25, 0.50 and 0.75, and solutions of each concentration were adjusted to pH values of 3.50, 5.00 and 6.50. This made a total of 18 treatments. The pH of the solutions was adjusted with sulfuric acid and checked periodically with the glass electrode. Difficulty was encountered in maintaining the solutions pH at 5.00, and therefore data are presented only as regards the influence of solution concentration in the case of these plants. Fluctuations in hydrogen ion concentration in the solutions maintained at pH 3.50 and 6.50 were kept within ±0.30 of a pH unit.

Eight plants were used in each treatment. Three plants of each treatment were placed under a double layer of cheesecloth, and the remaining five plants were grown in full light intensity. The plants were randomized in each of the two blocks.

The nutrient solutions were applied daily at the rate of 100 ml.

per pot until the first of August from which time the applications were made twice daily for the remainder of the summer. During the fall and winter the plants were again supplied the solution once a day but at a rate of 200 ml. per pot. The final measurements of plant response were made between March 22 and 24, 1955.

The effects of ammonium and nitrate nitrogen supplied at the two hydrogen ion concentrations on growth and leaf color are summarized in Table 3. From this table it will be seen that the fresh weights of plants receiving ammonium nitrogen were significantly greater than those supplied nitrate at either solution pH. No significant differences in fresh weight were obtained between plants receiving ammonium solutions at a high pH and those at a low pH, or between plants supplied nitrate at the two hydrogen ion concentrations. However, the data shows that when the ammonium solution pH was reduced from 6.50 to 3.50 a slight decrease in fresh weight of the roots occurred.

The average hydrogen ion concentration of the leachate shows the physiological acidity and alkalinity resulting from the two forms of nitrogen nutrition. Over a 24-hour period plants supplied ammonium nitrogen reduced the pH of the solution in contact with the roots to approximately 3.00. The azaleas supplied nitrate increased the pH toward 6.00.

Significant differences in the pH of the leaf tissue were obtained between plants supplied ammonium nitrogen at pH 3.50 and those supplied nitrate at either 3.50 or 6.50. The pH of chlorotic leaf tissue was significantly greater than that of healthy leaf tissue, regardless of the form of nitrogen supplied. The data indicate a correlation between hydrogen-ion concentration of the plant tissue and fresh weight production in the aerial portions of the plant.

The effect of solution concentration on plant growth is shown in Table 4, which shows that a significant reduction in growth resulted with each increase in ammonium solution concentration. Increasing the concentration of the nitrate solution, however, had no effect on fresh weight of the plant material. The data show that fresh weight production was significantly greater at all three concentrations of ammonium than that of plants supplied nitrate nitrogen. Comparative plant size, as influenced by these treatments, is shown in Plates 3, 4, and 5.

Again there appears to be a relationship between plant growth and pH of the leaf tissue. Plants supplied ammonium nitrogen had a significantly lower leaf tissue pH than those supplied nitrate. As the ammonium concentration was increased a reduction in plant growth and hydrogen ion concentration of the leaf tissue occurred. Plants supplied the highest nitrate concentration produced the least amount of growth and had the highest leaf tissue pH.

Solution concentration had little effect on the development of chlorotic symptoms, although the data show that an increase in the concentration of nitrate was followed by a slight increase in the severity of chlorosis. No symptoms of chlorosis developed in plants supplied ammonium nitrogen, regardless of solution concentration.

During the first three months of this experiment, chlorosis was evident in all plants supplied nitrate nitrogen, regardless of solution concentration and/or pH. However, by November, 1954, some of the plants receiving the nitrate solution adjusted to pH 3.50 began to produce vegetative growth and the chlorotic symptoms disappeared. By January, 1955, all plants supplied nitrate at that pH were green and showed evidence of new growth.

The day length or light intensity was apparently an important factor influencing the expression of chlorotic symptoms in this experiment. The nitrate solution at pH 3.50 was not effective in preventing the development of chlorosis during the summer months, but the same solution applied during the winter corrected chlorosis and brought about vegetative growth. The shaded plants showed little variation from the unshaded plants during the first two months of the experiment and the cheesecloth was removed in September, 1954. However, the first plants to show growth and leaf color response mentioned above were those that were shaded during the preceding summer. The influence of the reduced light intensity could still be detected in January, 1955. All plants that were previously shaded and supplied the nitrate solution at pH 3.50 had darker green foliage than the unshaded plants receiving the same nutrient solution. Plates 3 and 4 show the final appearance of plants supplied nitrate at pH 3.50 and 6.50 respectively.

Plate 3. The effect of nitrate solutions adjusted to pH 3.50 on growth of the Hexe azalea. (Left to right: Concentration 0.25, 0.50, and 0.75)


Plate 4. The effect of nitrate solutions adjusted to pH 6.50 on growth of the Hexe azalea. (Left to right: Concentration 0.25, 0.50, and 0.75)


Plate 5. The effect of ammonium concentration on growth of the Hexe azalea. (Left to right: Concentration 0.25, 0.50, and 0.75)

Experiment II

Table 3. The effect of nitrogen nutrition and hydrogen ion concentration on growth, pH of leaf tissue, and leaf color of the azalea.*

| Treatment | | | | Fresh Weight (Grams) | | | |
|---|---|---|---|---|---|---|---|
| Form of N | pH of Solution | pH of Leachate | pH of Leaf Tissue | Tops | Roots | Chlorosis Index | Nitrogen Deficiency |
| $NH_4$ | 3.50 | 2.97 | 4.04 | 32.9 | 49.2 | 1 | 1 |
| $NH_4$ | 6.50 | 3.34 | 4.13 | 27.1 | 64.5 | 1 | 1 |
| $NO_3$ | 3.50 | 5.56 | 4.27 | 7.8 | 15.7 | 1 | 1 |
| $NO_3$ | 6.50 | 6.34 | 5.34 | 2.5 | 4.5 | 4.6 | - |
| L.S.D. - 5% | - | - | 0.15 | 9.3 | 20.0 | - | - |

*Average of 12 plants.

Experiment II

Table 4. The effect of ammonium and nitrate ion concentration on the growth, pH of leaf tissue, and leaf color of the azalea.*

| Treatment | | | | Fresh Weight (Grams) | | | |
|---|---|---|---|---|---|---|---|
| Form of N | Solution Concentration | pH of Leachate | pH of Leaf Tissue | Tops | Roots | Chlorosis Index | Nitrogen Deficiency |
| $NH_4$ | 0.25 | 2.94 | 4.02 | 45.4 | 76.6 | 1 | 1 |
| $NH_4$ | 0.50 | 3.17 | 4.09 | 25.9 | 58.9 | 1 | 1 |
| $NH_4$ | 0.75 | 3.40 | 4.16 | 15.3 | 28.0 | 1 | 1 |
| $NO_3$ | 0.25 | 5.78 | 4.68 | 5.5 | 12.1 | 1.8 | 1.1 |
| $NO_3$ | 0.50 | 5.76 | 4.62 | 6.7 | 10.6 | 2.3 | 1.3 |
| $NO_3$ | 0.75 | 5.68 | 4.82 | 4.3 | 7.2 | 2.5 | 1.5 |
| LSD - 5% | - | - | 0.34 | 5.80 | 14.6 | - | - |

*Average of 11 plants.

Experiment III. The effect of varying the proportion of ammonium to nitrate in the nitrogen nutrition of azaleas.

The object of this experiment was to determine the effect of various proportions of ammonium to nitrate nitrogen in the nutrient solution on the growth and development of chlorotic symptoms in the azalea. The development of iron chlorosis was followed to determine the proportion of ammonium to nitrate nitrogen that was essential for preventing chlorosis under the conditions of these experiments. The influence of light on the expression of plant response to the several treatments was also considered.

Rooted cuttings of the azalea variety Hexe were planted July 16, 1954, in Del Monte white sand. The nutrient solutions, which contained ammonium to nitrate nitrogen ratios of 4:0, 3:1, 2:2, 1:3, and 0:4 (composition given in Table 5) were applied daily at the rate of 100 to 150 ml. per pot. There were 10 plants in each of the five treatments. Half the plants were placed under cheesecloth for shading and the remaining 25 plants were exposed to full greenhouse light intensity. The plants were randomized within the shaded and unshaded blocks. The unshaded plants were removed from the pots March 27, 1955, and the various pH and growth measurements were made. Leaf and root samples were dried for two days at 80°C. for ash determinations. Samples and crucibles of known weight were placed in a muffle furnace for eight hours at 450°C. The ashed samples were then cooled in a desiccator and reweighed. The ash content was calculated as the percent of the dry sample.

The effects of decreasing the proportion of ammonium to nitrate nitrogen in the nutrient solution on plant growth, ash content and pH of the leaf tissue are presented in Table 6. The data on fresh weight production show that no significant differences were obtained by reducing the ammonium nitrogen content of the solution from 100 to 75 per cent, or from 25 per cent to a complete nitrate solution. However, significant reductions in fresh weight resulted when the percentage of ammonium nitrogen was reduced from 75 per cent to 50 per cent. A further significant reduction in fresh weight resulted from decreasing the ammonium nitrogen from 50 to 25 per cent.

The reduction in fresh weight that was obtained by decreasing the proportion of ammonium nitrogen in the nutrient solution was probably not related to the pH of the medium. As shown in Table 6, only a slight increase in pH of the leachate occurred, so long as ammonium sulfate was present in concentrations as low as 13 ppm nitrogen.

The total ash content of the foliage failed to show any significant relationship. The high content of ash in the roots of plants receiving all their nitrogen as nitrate was, in part, due to the poor sample obtained, since these plants had made practically no growth during the course of the experiment.

No significant differences in pH of the leaf tissue occurred in plants that received nutrient solutions containing ammonium sulfate.

The development of chlorosis followed a pattern similar to that observed in the previous experiment. The plants supplied 25 per cent ammonium nitrogen and those supplied all their nitrogen as nitrate developed typical chlorotic symptoms within a month and a half after the experiment was started.

The shaded plants responded to the treatments in a similar manner, and the cheesecloth was removed in September, 1954. By January, 1955, the chlorotic plants supplied 25 per cent ammonium nitrogen started to produce new growth. As in the preceding experiment, plants that were previously shaded were the first to show this response and the leaves appeared darker green than unshaded plants of the same treatment. At the end of January, 1955, all plants were green and producing good vegetative growth with the exception of those plants supplied all of their nitrogen as nitrate. Plate 6 illustrates the final condition of the plants as they were influenced by the various proportions of ammonium and nitrate nitrogen.

Experiment III

Table 5. Composition of the nutrient solutions (ppm).

| % $NH_4$ | N | P | K | Ca | Mg | $SO_4$ | Cl |
|---|---|---|---|---|---|---|---|
| 100 | 52.5 | 8 | 59 | 50 | 12.2 | 216 | 142 |
| 75 | 52.5 | 8 | 59 | 50 | 12.2 | 182.5 | 100 |
| 50 | 52.5 | 8 | 59 | 50 | 12.2 | 138 | 66.3 |
| 25 | 52.5 | 8 | 59 | 50 | 12.2 | 92 | 32.5 |
| 0 | 52.5 | 8 | 59 | 50 | 12.2 | 48 | 0 |

Experiment III

Table 6. The effect of varying the proportion of ammonium to nitrate in the nutrient solution on pH of the leaf tissue, fresh weight produced and total ash content.*

| % $NH_4$ | pH of Solution | pH of Leachate | pH of Leaf Tissue | Fresh Weight (Grams) | | Total Ash | | Chlorosis Index | Nitrogen Deficiency |
|---|---|---|---|---|---|---|---|---|---|
| | | | | Tops | Roots | Tops | Roots | | |
| 100 | 6.80 | 3.45 | 4.11** | 30.9 | 28.9 | 6.05 | 11.61 | 1 | 1 |
| 75 | 6.80 | 3.65 | 4.05** | 34.5 | 24.6 | 5.74 | 8.01 | 1 | 1 |
| 50 | 6.80 | 4.01 | 4.21** | 17.1 | 15.5 | 6.26 | 6.33 | 1 | 1 |
| 25 | 6.80 | 4.22 | 4.11** | 7.6 | 7.5 | 6.00 | 6.91 | 1 | 2 |
| 0 | 6.80 | 6.43 | 5.07 | 2.8 | 1.9 | 6.41 | 46.45 | 4.6 | - |
| LSD - 5% | - | - | - | 9.2 | 6.2 | - | - | - | - |

*Average of 4 plants per treatment.

**No significant difference.


Plate 6. The effect of varying the proportion of ammonium to nitrate nitrogen in the nutrient solution on the growth of the Hexe azalea. (Total nitrogen 52 ppm)

Experiment IV. The effects of additional sulfate and chloride ions in the nitrate solution on growth and leaf color of the azalea.

The azaleas supplied ammonium nitrogen in the previous experiments consistently produced more growth than plants receiving nitrate nitrogen. When anion and cation nitrogen were used in separate solutions, however, difficulty was encountered in balancing the ions so that each solution had identical composition. The ammonium solutions in the previous experiments contained larger amounts of sulfate and chloride than the corresponding nitrate solutions, and it was questionable whether the superior growth of the azaleas was due entirely to the ammonium ion or in part to the additional sulfate and/or chloride ions. For this reason solutions were prepared which contained either chloride, sulfate, or chloride and sulfate in amounts equivalent to that found in the 0.25 strength ammonium solution. This was accomplished by using either chloride or sulfate salts and by the addition of hydrochloric and sulfuric acid. A fourth treatment consisted of 0.25 Hoagland's nitrate solution. The composition of the 4 solutions are given in Table 7. The solutions containing the additional anions had a lower pH than the standard nitrate solution.

Rooted cuttings of Hexe azaleas were planted August 12, 1954, in Del Monte white sand and leached with tap water for one week, after which time the solutions were applied daily at the rate of 150 ml. per pot. Ten plants were used for each treatment, and were arranged as a randomized block. Five plants in each treatment were supplied iron in the form of ferric tartrate and the remaining five plants were given chelated iron in equivalent amounts. This procedure was discontinued after two and one-half months, when no differences in growth or leaf color were observed, and ferric tartrate was used in all solutions. Four plants of each treatment were harvested April 10, 1955, and the final results are given in Table 8.

The solutions containing additional anions and hydrogen produced a significantly greater amount of growth than did the standard nitrate solution. The plants supplied additional sulfate produced significantly greater fresh weight of tops and roots than those supplied additional chloride, chloride plus sulfate, or the standard nitrate treatments. The plants receiving both anions produced significantly less fresh weight than the plants supplied additional chloride or sulfate alone.

There appears to be a correlation between the hydrogen ion concentration of the leachate and the fresh weight of plants supplied the additional anions. The pH of the leachate increased as fresh weight of the plants increased. The pH data show that the hydrogen ion concentration of leaf tissue was significantly higher when additional sulfate was added to the nitrate solution.

The addition of 142 ppm of chloride to the nutrient solution produced foliage of a lighter green color than that of plants supplied only sulfate. The appearance of the foliage suggested a lower rate of absorption or utilization of nitrate nitrogen. The chloride plus sulfate treatment produced a slight marginal leaf necrosis.

Although chlorosis did not develop in plants supplied the additional ions, the influence of light, as noted in the previous experiments, apparently reduced the rate of growth during the first two months of the experiment. Although the pH of the leachate was similar to that of plants supplied ammonium nitrogen, these plants were light green in color. However, in December, 1954, plants supplied additional sulfate began to produce growth and the foliage became dark green. By January, 1955, all plants receiving additional ions were producing vegetative growth. Plate 7 shows the effect of additional anions on plant growth and foliage color of the azalea.

Experiment IV

Table 7. Composition of nutrient solutions (in ppm).

| Treatment | N | P | K | Ca | Mg | $SO_4$ | Cl | Na |
|---|---|---|---|---|---|---|---|---|
| Cl | 52.5 | 8 | 59 | 50 | 12.2 | 0 | 142 | 6 |
| $SO_4$ | 52.5 | 8 | 59 | 50 | 12.2 | 216 | 0 | 29 |
| Cl + $SO_4$ | 52.5 | 8 | 59 | 50 | 12.2 | 216 | 142 | 29 |
| Standard | 52.5 | 8 | 59 | 50 | 12.2 | 48 | 0 | 0 |

Experiment IV

Table 8. The effect of additional sulfate and chloride ions to nitrate solutions on growth, leaf color, and pH of the leaf tissue of the azalea.*

| Treatment | pH of Solution | pH of Leachate | pH of Leaf Tissue | Fresh Weight (Grams) | | Chlorosis | Nitrogen Deficiency |
|---|---|---|---|---|---|---|---|
| | | | | Tops | Roots | | |
| Cl | 2.70 | 4.01 | 4.35 | 11.5 | 21.2 | 1 | 2 |
| $SO_4$ | 3.00 | 5.18 | 4.14 | 21.4 | 27.4 | 1 | 1 |
| Cl + $SO_4$ | 2.50 | 3.13 | 4.10 | 8.5 | 14.9 | 1 | 1.5 |
| Standard | 6.80 | 6.52 | 5.01 | 2.8 | 5.7 | 4.7 | - |
| LSD - 5% | | | 0.02 | 4.4 | 3.5 | | |

*Average of 4 plants per treatment.


Plate 7. The effect of additional sulfate and/or chloride ions to the standard nitrate solution on the growth of the azalea. (Left to right: Cl, $SO_4$, Cl + $SO_4$, and standard nitrate solution)

Experiment V. The influence of total salt content of the nutrient solution as related to the concentration and form of nitrogen used on growth and development of chlorosis of the azalea.

The previous experiment indicated that large additions of chloride or sulfate to the nitrate solutions were not essential in preventing chlorosis. Azaleas receiving solutions lacking either anion produced healthy foliage. The increased hydrogen ion concentration was apparently the factor responsible for the satisfactory growth and absence of iron chlorosis, although the exact function of hydrogen in this respect was not understood. Experiment V was designed primarily to study the relationship between the total concentration of salts in the nutrient solution and the concentration and form of nitrogen used. No acidic or basic substances were used to control solution pH, but the concentration of hydrogen in the solutions and leachate was recorded periodically to determine more closely the effect of this ion on the development of iron chlorosis.

Rooted Hexe cuttings were planted as described in the previous experiments and leached with tap water for one month. Nutrient solutions were applied at the rate of 150 ml. per pot daily beginning November 24, 1954. The composition of the nutrient solutions is given in Table 9. Treatments 1, 2, and 3 contained 40, 26, and 13 ppm of ammonium nitrogen respectively, in the form of ammonium sulfate. Equal amounts of other salts were used in these treatments. Nitrate nitrogen supplied as nitric acid was used at the same decreasing rates in treatments 4, 5, and 6, with the total salt content the same in all treatments. In treatments 7, 8, and 9, nitrate was constant at 40 ppm nitrogen with total salt content varied in the solutions. The three treatments contained 305.0, 155.0, and 97.5 ppm total salt respectively. There were five single plant replications for each of the nine treatments, and the 45 plants were randomized in one block on the greenhouse bench.

The various measurements of plant response to the several treatments are given in Table 10. The fresh weight of the plant tops in treatment 1 (40 ppm $NH_4$ nitrogen with 471.5 ppm total salts) and 2 (26 ppm $NH_4$ nitrogen with 436.0 ppm total salts) was significantly greater than that of any of the other treatments. No significant differences in fresh weight occurred between plants supplied 40 ppm nitrate nitrogen (Treatment 4 and 9) and those receiving 13 ppm ammonium nitrogen (Treatment 3). The plants of treatment 4 (40 ppm $NO_3$ nitrogen with 359.0 ppm total salts) and treatment 9 (40 ppm $NO_3$ nitrogen with 97.5 ppm total salts) had a better leaf color score than plants of treatment 3 (13 ppm $NH_4$ nitrogen with 391.0 ppm total salts), which suggests an increase in nitrate nitrogen would have had little effect on fresh weight of the azalea. Treatment 6 (13 ppm $NO_3$ nitrogen with 332.0 ppm total salts) and 7 (40 ppm $NO_3$ nitrogen with 305.0 total salts) produced the least amount of growth as compared to the other treatments. The fresh weight of roots showed about the same growth pattern as the tops for each treatment.

The differences in growth response to the various treatments of this experiment were not significant in several instances, due to the rather short duration of the experiment and the relatively slow growth rate of the azalea plant. However, certain trends were evident. A reduction in growth occurred as a result of reducing the concentration of nitrate from 40 ppm to 13 ppm nitrogen (Treatment 4, 5 and 6). An increase in growth was obtained by decreasing the total salt content from 305.0 ppm to 97.5 ppm with nitrate constant at 40 ppm nitrogen (Treatment 7, 8 and 9).

The source of nitrogen determined the pH of the leachate, as was found in the previous experiment. The leachate from plants supplied ammonium nitrogen had a significantly lower pH than the leachate of all nitrate treatments. The several nitrate treatments produced no significant pH differences in the leachate, with the exception of treatment 4, which had a higher concentration of hydrogen ions.

The hydrogen ion concentration of leaf tissue showed that plants supplied ammonium at 40 and 26 ppm nitrogen had a significantly lower pH than plants supplied nitrate at 40 ppm nitrogen, with the exception of plants in treatment 9 (97.5 ppm total salts). The leaf-tissue pH of plants in treatment 9 was the lowest of all nitrate treatments in this experiment. Treatments 4, 5, and 6 show that as nitrate concentration is reduced the pH of the leaf tissue decreases.

The development of chlorosis in the azalea appears to be closely associated with the pH of the leaf tissue. The highest leaf-tissue pH occurred in the chlorotic plants of treatment 7.

The conditions that influence the pH of the leaf tissue, and evidently the development of iron chlorosis as well, are evident in the data presented in Table 10. The low total salts concentration of the nitrate solution in treatment 9 resulted in the lowest leaf-tissue pH of all nitrate treatments. Reducing the nitrate content of the nutrient solutions (Treatments 4, 5, and 6) resulted in a higher concentration of hydrogen ions in the leaf tissue. These two conditions (low salt or low nitrate) that apparently prevent iron chlorosis in azaleas supplied nitrate nitrogen are independent of the hydrogen ion concentration of the nutrient media. The third condition that lowered tissue pH and thereby reduced chlorosis was the high concentration of hydrogen ions in the nutrient media. This was shown in treatment 4, which produced a significantly lower pH in the leachate than all other nitrate treatments. However, this last condition appeared to be less effective in reducing leaf-tissue pH than the low concentration of salts or nitrate (cf. Treatments 4, 6, and 9). The influence of reduced salts content of the nutrient solution on growth and leaf chlorosis is illustrated in Plate 8.

Experiment V

Table 9. Composition of nutrient solutions (in ppm).

| Treatment | N | P | K | Ca | Mg | $SO_4$ | Cl | Nitrogen Source |
|---|---|---|---|---|---|---|---|---|
| 1 | 40 | 8 | 59 | 50 | 12 | 170.5 | 142 | $(NH_4)_2SO_4$ |
| 2 | 26 | 8 | 59 | 50 | 12 | 125 | 142 | $(NH_4)_2SO_4$ |
| 3 | 13 | 8 | 59 | 50 | 12 | 80 | 142 | $(NH_4)_2SO_4$ |
| 4 | 40 | 8 | 59 | 50 | 12 | 48 | 142 | $HNO_3$ |
| 5 | 26 | 8 | 59 | 50 | 12 | 48 | 142 | $HNO_3$ |
| 6 | 13 | 8 | 59 | 50 | 12 | 48 | 142 | $HNO_3$ |
| 7 | 40 | 8 | 59 | 50 | 12 | 48 | 88 | $HNO_3$ - $Ca(NO_3)_2$ |
| 8 | 40 | 4 | 30 | 25 | 6 | 24 | 26 | $HNO_3$ - $Ca(NO_3)_2$ |
| 9 | 40 | 2 | 15 | 12.5 | 3 | 12 | 13 | $HNO_3$ - $Ca(NO_3)_2$ |

Experiment V

Table 10. The effect of nitrogen concentration and total salt content of the nutrient solution on certain growth responses of the azalea.*

| Treatment | | | | pH of | | | Fresh Weight (Grams) | | Chlorosis | Nitrogen |
|---|---|---|---|---|---|---|---|---|---|---|
| No. | ppm N | Form of N | ppm Salts | Solution | Leachate | Leaf Tissue | Tops | Roots | Index | Deficiency |
| 1 | 40 | $NH_4$ | 471.5 | 6.80 | 4.17 | 4.22 | 14.1 | 38.3 | 1.0 | 1.0 |
| 2 | 26 | $NH_4$ | 436.0 | 6.80 | 4.74 | 4.22 | 12.1 | 44.6 | 1.0 | 1.4 |
| 3 | 13 | $NH_4$ | 391.0 | 6.80 | 4.93 | 4.34 | 6.9 | 28.2 | 1.0 | 2.4 |
| 4 | 40 | $NO_3$ | 359.0 | 3.25 | 5.63 | 4.47 | 6.4 | 18.5 | 1.0 | 2.0 |
| 5 | 26 | $NO_3$ | 345.0 | 3.80 | 5.92 | 4.44 | 4.9 | 19.8 | 1.0 | 2.6 |
| 6 | 13 | $NO_3$ | 332.0 | 5.85 | 6.23 | 4.38 | 2.6 | 10.2 | 1.0 | 2.8 |
| 7 | 40 | $NO_3$ | 305.0 | 4.70 | 6.02 | 5.29 | 3.1 | 5.3 | 4.8 | - |
| 8 | 40 | $NO_3$ | 155.0 | 4.60 | 5.96 | 4.49 | 4.4 | 7.0 | 1.4 | 2.0 |
| 9 | 40 | $NO_3$ | 97.5 | 4.40 | 6.06 | 4.26 | 6.6 | 17.9 | 1.0 | 1.6 |
| | LSD - 5% | | | | 0.37 | 0.19 | 3.2 | 12.9 | | |

*Average of 5 plants per treatment


Plate 8. The influence of total salt concentration of nitrate solutions on growth and development of chlorosis symptoms in the azalea. (Left to right: 305.0, 155.0, and 97.5 ppm total salts)

Experiment VI. The effect of nitric acid and ammonium hydroxide on growth of azaleas planted in sand and sawdust.

A comparison of ammonium hydroxide and nitric acid as nitrogen sources for azaleas was obtained in an experiment with plants grown in sand and sawdust. The plants grown in sand were supplied a complete nutrient solution to which ammonium hydroxide or nitric acid was added at the rate of 50 ppm nitrogen. Thus, a comparison of ammonium and nitrate was made with solutions that had identical composition with the exception of the form of nitrogen and the concentration of hydrogen and hydroxyl ions. The comparison of the two forms of nitrogen was also obtained without the large amounts of sulfate and chloride ions present in the ammonium solutions used in the previous experiments. The pH of the ammonium and nitrate solution was 9.80 and 3.00 respectively. The solutions were applied daily at the rate of 150 ml. per plant.

The plants grown in sawdust were supplied nitric acid or ammonium hydroxide at the rate of 200 ml. per plant. No elements other than ammonium or nitrate nitrogen were added to these plants. This was done in an effort to obtain a direct comparison of the two forms of nitrogen without the influence of other nutrient elements, since they occur in very low concentrations in sawdust. The nitrogen concentration and frequency of application were continually increased during the first two months of the experiment. During the last two and one-half months the solutions were applied three times a week at the rate of 120 ppm nitrogen.

The plants supplied ammonium hydroxide in the sand culture produced more growth and were darker green than plants supplied nitric acid, although the differences were not very great. The plants receiving nitrate produced an elongated type of growth and fewer new shoots were formed than on plants supplied ammonium hydroxide.

The hydrogen ion concentration of the leachate from crocks supplied ammonium hydroxide ranged from pH 3.50 to 3.71, and from 5.12 to 5.46 in the case of crocks supplied the nitric acid solution. The pH of the leaf tissue of plants receiving ammonium hydroxide was consistently lower than that of plants supplied the nitric acid solution.

The growth responses of the plants grown in sawdust for six months are given in Table 11. These data show that ammonium hydroxide was superior to nitric acid in promoting growth and in increasing the green coloration of the foliage.

The pH of the medium, given in Table 11, was measured from samples of sawdust taken from the root system of several plants. The hydrogen ion concentration of samples taken from the surface layer of the ammonium hydroxide section showed a pH of 6.00, but was consistently lower in the vicinity of the root system. The pH of the sawdust in the nitric acid section showed little variation regardless of the depth the sample was taken.

The hydrogen ion concentration of the leaf tissue as reported here is an average of six measurements. Each measurement was a composite sample of leaves taken from five different plants. As shown in Table 11, no differences in pH of the leaf tissue occurred as a result of the two forms of nitrogen and in the absence of other nutrient elements.

## Experiment VI

Table 11. The effect of $HNO_3$ and $NH_4OH$ on growth and pH of the leaf tissue of azaleas grown in sawdust.

| Treatment | pH of Sawdust | pH of Plant Sap | New Growth in Cm.* | No. of Shoots* | Chlorosis Index* | Nitrogen Deficiency* |
|---|---|---|---|---|---|---|
| $NH_4OH$ | 4.20 | 4.23 | 86.5 | 8.1 | 1 | 1 |
| $HNO_3$ | 4.20 | 4.21 | 63.7 | 6.8 | 1 | 3 |

*Average of 30 plants.

Experiment VII. The effect of a single application of nitrogen on field-grown Mollis azaleas.

The results of a field trial with nitrogen fertilizers is mentioned here to show the similarity of azalea response to ammonium and nitrate nitrogen in the field to that observed in sand culture experiments. Nitrogen in the forms of ammonium sulfate, ammonium phosphate, ammonium nitrate and sodium nitrate were applied at random to four plots consisting of four plants each of three-year-old seedling Mollis azaleas in four separate blocks. Eight plots of the same number of plants served as a check on the treated plants. The plants were in a poor condition and many showed evidence of iron chlorosis at the time the fertilizers were applied. The rate of nitrogen application was 0.05 pounds of nitrogen or an equivalent of 0.25 pounds of ammonium sulfate per plant. This was applied in a single application on July 13, 1954. The azalea block was then mulched with sawdust and irrigated. Observations were made periodically on the condition of each of the 96 plants in the experimental block.

The response to the nitrogen additions and irrigation was rapid and rather outstanding. Ammonium sulfate and ammonium phosphate brought about the greatest increase in growth. Plants supplied ammonium sulfate produced the darkest green foliage of all the treatments. Ammonium nitrate was intermediate in its effect between the plants supplied all ammonium nitrogen and those receiving all nitrate nitrogen. Plants receiving sodium nitrate produced little growth and in many cases appeared inferior to the check plants.

Final observations were made October 10, 1954. Many of the plants supplied ammonium forms of nitrogen were observed to develop chlorosis on some of the new growth. This was possibly due to a lack of ammonium nitrogen, since oxidation to nitrate occurs rapidly under certain soil conditions. The high light intensity during this period may also have accelerated the development of chlorotic symptoms in these plants. Soil samples taken throughout the block for pH determinations showed little variation, ranging from 4.80 to 5.45. It is unlikely that these variations in hydrogen ion concentrations could account for the superiority of ammonium nitrogen in this experiment.

# DISCUSSION

Throughout these studies, an attempt was made to supply both ammonium and nitrate nitrogen under conditions that would produce satisfactory growth of the azalea. This was essential if a meaningful comparison of the two forms of nitrogen was to be attained. Although early in the studies it appeared that ammonium nitrogen was superior to that of nitrate as a source of nitrogen for this plant, it was not known whether the nitrate had been supplied under conditions satisfactory for growth. For this reason the various adjustments in the nitrate solutions were made in an effort to produce plant growth comparable to that obtained with ammonium nitrogen. These adjustments have shown the importance of concentration of nitrogen, hydrogen and certain other ions in achieving this comparable growth with nitrate. At the same time, the importance of light and temperature to the expression of these nutritional relationships on growth and the development of iron chlorosis has been established.

## Form and Concentration of Nitrogen

The results obtained (Experiment V) indicate that the azalea requires less ammonium than nitrate nitrogen for good growth and foliage color. Similar results have been obtained by Tiedjens and Blake (30, pp.24-26) with apple trees and by Clark (6, p.18) with tomato plants. The more rapid absorption and assimilation by plants of the ammonium ion as shown by Prianishnikov (22, pp.84-85) and by

Tiedjens (29, p.53) probably accounts for the difference in quantitative requirements for the two forms of nitrogen.

The concentration of nitrate nitrogen was apparently not the factor limiting growth of the azalea in several of these experiments. Nitrate supplied as high as 158 ppm nitrogen gave no significant increase in growth or improvement in leaf color over that of plants receiving 50 ppm of nitrogen as nitrate (Table 4). Increasing the nitrate supply to azaleas grown in sawdust did not produce any noticeable change in growth or foliage color. The plants receiving 40 ppm nitrate nitrogen (Table 10) had a better leaf color score than plants supplied 13 ppm ammonium nitrogen, but produced less total growth. This indicates that factors other than the concentration of nitrogen were limiting growth in this experiment. Solutions containing 26 ppm nitrogen as ammonium produced as much growth as those supplied 40 ppm of ammounium nitrogen (Table 10). When the ammonium solution was increased to 105 ppm ammonium nitrogen, a significant reduction in plant growth resulted (Table 4).

A low concentration of nitrate nitrogen was apparently a factor in preventing chlorosis symptoms on plants in Experiment V and will be discussed further as it is related to the concentration of hydrogen ions.

### Hydrogen Ion Concentration

Of the several factors that influence the absorption and assimilation of nitrogen, the pH of the root media is usually considered of great importance. Results obtained with a number of plants, including tomato and apple (29, p.54), strawberry (7, pp.44-45), peach trees (8, pp.363-364), and certain other crop plants (22, pp.71-89) indicate that nitrate nitrogen is best absorbed and assimilated at a low pH and ammonium nitrogen at a high pH. In contrast to these results, good growth was obtained in these experiments with azaleas supplied ammonium sulfate solutions at pH 3.50 (Table 3). Since the effects of a high pH were not observed when ammonium nitrogen was applied to the sand cultures, a series of aerated solution cultures were prepared for this purpose. The 0.25 ammonium solution was used at a pH of 6.80. The 10 liters of solution was renewed daily to prevent any change in pH. The azaleas grew normally and without chlorosis under these conditions for a period of two months, indicating that ammonium nitrogen is absorbed and assimilated equally well by the azalea at a low or high pH.

The data also indicate that the azalea plant controlled to a considerable extent the pH of the media in which it was growing, depending upon which form of nitrogen was supplied. A decrease in hydrogen ion concentration of the media occurred when nitric acid was supplied as a source of nitrogen, whereas an increase occurred with ammonium hydroxide. Tiedjens and Blake (30, p.7) obtained similar results with ammonium hydroxide as a source of nitrogen for apple trees.

Plants supplied the standard, nitrate solution did not grow and developed severe chlorosis symptoms, unless the hydrogen ion concentration was very high (Experiment II and IV). However, a low pH was not essential to normal growth, if the nitrate content was low, or if the total concentration of salts was low (Table 9). The function of the large amounts of hydrogen in this regard is apparently related to the absorption of other elements. Hoagland and Broyer (14, pp.178-179) found a substantially greater accumulation of cations and anions in barley roots at a high rather than a low pH. Under such conditions calcium was not absorbed and in some cases was lost from the plant. Prianishnikov (22, p.78) has also found an antagonism between hydrogen and calcium.

The hydrogen ion is apparently similar to the ammonium ion in its effect on absorption of other bases. The results and interpretations of a number of research workers offer an explanation in this regard. That nitrate nitrogen increases the absorption of the bases calcium, magnesium and potassium in comparison to similar solutions containing ammonium nitrogen has been demonstrated by Sideris and Young (24, pp.267-268) with pineapple, and Holley (15, pp.5-10) with cotton. Clark (6, pp.16-17) found a lower organic acid content in tomatoes supplied ammonium nitrogen, which indicates a lower amount of bases absorbed. Wadleigh and Shive (34, p.282) show that the ammonium ion has a greater influence in reducing the base content of corn plants than has hydrogen. They state, however, that the high concentration of hydrogen ions on the root film may be partially responsible for the lower uptake of cations.

Concentration of Other Ions

Variations in the proportion of sulfate plus chloride to the total amount of calcium, potassium and magnesium in the nitrate solution had a significant effect on plant growth (Experiment IV), but had no effect in preventing the development of iron chlorosis at a high pH. Chlorotic symptoms and poor growth occurred when the 0.25 nitrate solution was used, whether total anion content was 64.0 or 144.0 ppm with total bases constant (121.0 ppm). The increased rate of growth obtained from additional sulfate may have resulted from an increased absorption of the nitrate ion. This was reported by Prianishnikov (22, pp.90-99) as occurring in the case of corn plants. Nightingale (21, p.191) reports an antagonism between chloride and nitrate ions which could account for the poor rate of growth and nitrogen deficiency symptoms of plants supplied additional chloride in Experiment IV. The sulfate and chloride ions had little effect on plant growth and chlorosis when ammonium was used as the source of nitrogen (Experiment VI).

Evidence showing that chlorosis development and poor growth are results of total base content of the nitrate solution was first obtained in Experiment V. Reducing total salts from 305.0 to 97.5 ppm increased growth and prevented chlorosis development. Thus, the total amount of bases in relation to hydrogen ion concentration of the root media appears to be directly related to the occurrence of iron chlorosis in the azalea.

Of the bases essential for plant growth, calcium has frequently been associated with the development of iron chlorosis in plants. In these studies, the 0.25 standard nitrate solution with 121.0 ppm total bases always produced chlorotic plants (Experiments I, II, and V) unless the hydrogen ion concentration was high or the nitrate content low. In order to determine the relative importance of specific cations as against total base content in the nutrient solution, an experiment, which is still in progress, was designed to compare the influence on chlorosis development of high (75 ppm) levels of calcium or potassium in a low-base, nitrate solution (96 ppm total bases) with results obtained from the normal concentration of calcium (50 ppm) and potassium (59 ppm) in the standard 0.25 nitrate solution containing a relatively high (121.0 ppm) total base content. The effects of the increased cations are being made in solutions of pH 3.45 and 6.00. After 14 weeks of nutrient application, there is no evidence of chlorosis in those plants supplied the high-calcium solution at either pH. Some of the plants supplied the high-potassium solution at pH 6.00 are showing faint symptoms of chlorosis, although this condition is not as severe and did not develop as rapidly as in the case of plants supplied the standard nitrate solution. These chlorotic symptoms may be the result of the rapid absorption and accumulation of potassium in the plant tissue which are a characteristic plant response to this ion. In contrast to azaleas supplied the nitrate solutions, plants receiving ammonium hydroxide at the rate of 35 ppm nitrogen are producing satisfactory growth and foliage color although the total base content of the solution is high (160 ppm).

The data presented by Cain (4, pp.162-164) show a relationship to exist between the total base content of the foliage of blueberry plants and the form of nitrogen applied. His data show that as ammonium nitrogen was replaced in the nutrient solution by calcium nitrate, the calcium content of the foliage increased. When ammonium nitrogen was reduced so that the nutrient solution contained 240 ppm calcium as calcium nitrate, a sharp increase in potassium content of the foliage resulted. Total base content of the foliage was highest when ammonium nitrogen was completely replaced in the nutrient solution by calcium nitrate. A relationship appears to exist in his experiment between the total base content of the foliage, the amount of plant growth and the amount of chlorosis that developed.

The work of Vickery et al. (32, p.75) offers an explanation for the similarity in ash content of the azaleas supplied varied ratios of ammonium and nitrate nitrogen in these experiments (Table 6). These authors obtained similar results with tobacco, but their analysis of the ash showed that as ammonium nitrogen was increased in the solution the anion content of the ash increased with a corresponding decrease in bases.

That the role played by ammonium nitrogen in preventing chlorosis and promoting good growth of the azalea is possibly through its antagonistic action toward the bases calcium, magnesium and potassium, is supported further by the low requirement of the azalea for the basic elements. Twigg and Link (31, pp.374-375) established critical levels of 0.22, 0.17, and 0.80 per cent of dry foliage for calcium, magnesium and potassium respectively. These are quite low compared to those of many cultivated plants. It is interesting to note that the total amount of bases found by Twigg and Link for azaleas (1.19) is the same amount found by Cain in blueberry foliage, when the plants were supplied ammonium sulfate (4, p.164).

A close relationship was found in these experiments between the concentration of nitrate, total bases, and hydrogen ions in the nitrate solution, and the pH of the leaf tissue of the azalea. When the concentration of nitrate or total bases were increased in the nitrate solution, a corresponding increase in pH of the plant tissue occurred as compared with plants supplied ammonium nitrogen. Similar results have been reported by Wadleigh, Robbin, and Beckenbach (33, pp.167-171) working with corn plants, and by Vickery et al. (32, p.92) with tobacco. The high pH of the leaf tissue reported in these experiments with azaleas showed a close relationship with the development of iron chlorosis.

The inactivation of iron in plant tissue has been found to be closely related to the pH of the plant sap in other plants. Loehwing (18, pp.296-297) found that limed soils increased the pH of the plant sap as much as 0.82 of a pH unit as compared to that of unlimed soils. Ingalls and Shive (16, p.121), using nine different species of plants, found an increase in soluble iron content as the hydrogen ion concentration of the sap increased. They showed that plants with a high hydrogen ion concentration in the sap had a lower total but a higher soluble iron content than plants with a higher pH of the plant sap. Rogers and Shive (23, pp.237-250) found that iron accumulations occurred in tissues having a high pH when these were adjacent to tissues of low pH. Small (26, pp.38-51) has reviewed these and other experiments concerning the hydrogen ion concentration of plants and concludes that the explanation of lime induced chlorosis has apparently been settled.

The relationship between the form of nitrogen applied and the ratio of free organic acids to salts of organic acids with respect to plant sap reaction is discussed by Bonner (3, pp.159-161). Nitrate has been shown to increase the total organic acids of plants. A corresponding increase in salts of organic acids occurs as a result of an increased rate of base accumulation. Hoagland (12, pp.128-142) states that the proportion of unknown organic acids in plants increases with ammonium nutrition. The pH of the plant sap is evidently determined by the types of organic acids present and the dissociation constants of such acids associated with cations.

## Environmental Factors

The various adjustments of the nitrate solution in these experiments had little effect on plant growth during periods of long day length and/or high light intensity. Of interest in this regard is the work of Hibbard (11, pp.20-27). He found that full light intensity favored the absorption of calcium, potassium and iron in the plants studied, while short days decreased total salt uptake.

The translocation of iron in the aerial parts of the plant was more rapid under short day conditions. Gericke (10, pp.106-107) observed that wheat plants growing in solutions lacking iron were more etiolated and smaller than shaded plants. Similar results were obtained by Loehwing (18, pp.269-297).

Sideris, Young and Chun (25, pp.43-47) found that the hydrogen ion concentration and titratable acidity of the sap taken from pineapple plants decreased in light and increased during dark periods. This was a result of the breakdown of organic acids during the light period. Small (26, p.50) found that *Rhododendron ponticum* had a more acid sap during the winter than in the summer. The influence of light as a factor increasing the absorption of bases and reducing the pH of the plant sap could account, in part at least, for the results obtained in these experiments with azaleas supplied nitrate nitrogen.

## CONCLUSION

A knowledge of the natural environment of the azalea seems to provide an explanation for the results obtained in these experiments. The azalea occurs for the most part in acid soil types, fairly high in organic matter and usually in shaded situations. Such soils would be low in exchangeable bases, a factor found desirable in the nutrition of this ericaceous plant. Under field or greenhouse conditions pH is, in part, a measure of bases in the media. In other words, the high concentration of hydrogen ions is the result of a low base saturation on the soil complex. The beneficial effect of such a condition for azalea growth has been shown to be due to the low concentration of bases and not the high concentration of hydrogen. Although such conditions, as low hydrogen and low base concentration, would not be encountered in most soils, it can be demonstrated in sand culture.

On the basis of results recorded in the literature concerning many plant species, nitrate would be expected to be superior to ammonium nitrogen at a low pH. Although a low pH under certain conditions was shown to be essential where nitrate was used in these experiments, plant growth and color with nitrate were inferior to plants supplied ammonium nitrogen at low pH values. The nitrate solutions containing large amounts of sulfate and hydrogen produced the most desirable plants. The presence of the ammonium ion evidently promotes conditions ideal for growth of the azalea and appears to be superior to nitrate, even when nitrate is supplied under supposedly ideal conditions.

The successful plant growth obtained by commercial greenhouse operators using organic medias and fertilizers is evidently due, in part, to the low but constant source of reduced nitrogen available to the plant from such materials. Under field conditions, ammonium nitrogen may be rapidly oxidized to nitrate. The rate of oxidation would depend on soil type, pH, temperature, and biological factors. Frequent applications of small quantities of ammonium nitrogen would be necessary to maintain a constant supply to the plant under some conditions.

The experiments indicate that iron chlorosis often observed in these plants is a result of an increased accumulation of bases in the plant. The high base content within the plant reduces growth and increases the pH of plant tissue which results in the inactivation of iron with its accompanying chlorosis. Factors influencing the uptake of bases, such as nutrition with nitrate nitrogen, high total base content of the nutrient solution at pH values of 4.00 and above, and the effect of light as it increases base absorption and the breakdown of organic acids tends to increase plant tissue reaction and hasten the appearance of chlorotic symptoms in the azalea.

## SUMMARY

Several experiments have been conducted with azaleas grown in sand, sawdust and soil cultures, to determine the conditions that promote good growth and foliage color when supplied either ammonium or nitrate forms of nitrogen. The relationship of certain environmental factors to growth response was also considered in these studies.

Ammonium nitrogen, supplied as sulfate or hydroxide, produced more plant growth and better foliage color, and was required in smaller amounts than nitrate nitrogen.

The standard nitrate solution used in these experiments resulted in poor growth and chlorosis of the azalea. However, good growth and good foliage color was obtained when large additions of hydrogen and sulfate ions were added to the solution, or when total base content was reduced to a low level. Large additions of hydrogen and chloride ions prevented the development of chlorosis, but plants appeared deficient in nitrogen.

Reducing the concentration of nitrate nitrogen reduced plant growth, but chlorosis did not develop in these plants, although the pH and base content of these solutions usually resulted in chlorotic symptoms at the higher nitrate levels.

Chlorosis development was closely associated with the pH of the leaf tissue. Plants supplied ammonium nitrogen had a lower tissue pH than those receiving nitrate nitrogen, and chlorotic plants had the highest leaf tissue pH. The solution adjustments that promoted

good growth with nitrate lowered tissue pH. Reducing the nitrate content of the nutrient solution also lowered the pH of the leaf tissue.

The hydrogen ion concentration of the root media was determined to a considerable extent by the form of nitrogen supplied the plant. Plants receiving the ammonium forms of nitrogen decreased pH, while those supplied nitrate increased media pH, as determined by leachate measurements.

Increasing the root temperature to plants supplied ammonium nitrogen resulted in an increase in plant growth, but had little effect when nitrate nitrogen was used.

The nitrate solution adjustments that were made in an attempt to promote growth with this form of nitrogen had little effect during periods of high light intensity and/or long day length.

It is concluded that the ammonium ion evidentaly reduces the uptake of other cations which reduces plant tissue pH. Nitrate apparently increases base absorption and iron is precipitated in the tissue as a result of the higher tissue pH.

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