

CALCIUM NUTRITION OF TARO (COLOCASIA ESCULENTA
(L.) SCHOTT) AND ITS POSSIBLE RELATIONSHIP TO GUAVA SEED DISEASE

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ABSTRACT

Calcium deficiency symptoms on taro (*Colocasia esculenta* (L.) Schott cv. 'Lehua maoli') grown in nutrient solution culture for 7 weeks were leaf blade interveinal chlorosis and necrosis, failure of the leaf blades to unfurl, collapse of petioles, and die-back of roots. Death of the growing point occurred in severely Ca-stressed plants.

Sections of the leaf blades of taro grown in solutions varying in Ca concentration were sampled after 5 months to study the long-term effects of calcium deficiency on the ultrastructure of parenchyma cells. The disintegration of the plasmalemma, tonoplast, and all other membranous systems in the cytoplasm was observed in calcium deficient cells.

Taro was grown for 3 months in nutrient solution culture with 1, 2, 4, and 20 ppm of Ca and 21 and 100 ppm of nitrogen. Leaf dry weight increased significantly with increasing solution Ca and decreased significantly with increased solution nitrogen. Leaf Ca concentration increased linearly with increasing solution Ca, however the increase in leaf Ca was significantly less at the higher N treatment compared to the lower N treatment.

Calcium levels were increased to 2, 4, 8 and 40 ppm from month 4 through month six. The N treatments were both 42 ppm for the duration of the fourth month. For the fifth and six months, the 2 N levels were 42 and 84 ppm.

Dry weight of the corm plus cormels, dry matter content of the corm, and starch content of the corm at 6 months all increased with

increasing levels of solution calcium. A Mitscherlich type growth response to Ca was found at the lower N treatment but not at the higher N treatment.

The best tissue to sample for plant Ca levels is the third blade at 3 to 6 months of growth. The critical Ca concentration range for the third blade during this growth period was estimated to be 0.7 to 1.0% Ca on a dry weight basis.

Nutrient concentration in the solution was decreased for the final 2 months of the experiment. Starch content, dry matter and specific gravity of the corms as well as fresh weight of the corm plus cormels at 8 months all increased with increasing Ca in solution. The corms at the highest level of solution of Ca were mature as indicated by the specific gravity and dry matter percentage of the corms.

Guava seed disease was not found to be associated with any of the treatments at 6 or 8 months harvest. The disease was not directly caused by Ca deficiency, NH_4 toxicity, or N-induced Ca deficiency under the relatively sterile conditions of a solution culture experiment.

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INTRODUCTION

Guava seed disease of taro is a major problem of taro growers in Hawaii. It is characterized by the development of corky tissues in the corm (Parris, 1941). No disease organism has been found to be associated with this disease, despite repeated isolation attempts and microscopic examinations of diseased tissues (Parris, 1941; Trujillo, 1979). This disease reduces the marketable yield of taro because those portions of the corm affected by the disease are discarded.

Parris (1941) reported that liming at 2 T/A decreased guava seed disease incidence at two localities. However, he didn't recommend liming for disease control because of its inconsistent performance at other localities.

It was noted that guava seed disease of taro superficially resembled well-documented Ca deficiency disorders such as cavity spot of carrots and cork spot of pears and apples (Shear, 1975). This study was carried out with three objectives: (1) to demonstrate Ca deficiency symptoms of taro; (2) to determine whether a relationship existed between guava seed disease and Ca nutrition; and (3) to study the effects of excess N fertilization on the Ca nutrition of taro.

I. LITERATURE REVIEW

Botany and Culture of Taro

Taro (Colocasia esculenta (L.) Schott) is a succulent, perennial herb which produces edible corms (Kay, 1973; Plucknett et al., 1970). It is grown as a staple or subsistence crop throughout the tropics, subtropics, and warmer temperate regions. Taro is a commercial crop only in Hawaii, Egypt, Philippines, and certain islands in the South Pacific and Carribean.

Taro is grown primarily for its corms, but the leaves are sometimes used as a vegetable (Plucknett and de la Pena, 1971; Sickey, 1973). The leaves, when cooked, contain relatively high amounts of Ca, P, Fe, K, vitamin A and abscorbic acid (Plucknett and de la Pena, 1971). The leaves are a good quality source of protein, since most of the essential amino acids are present.

Corm and cormels are rich in starch, and contain appreciable amounts of vitamin C (7-9 mg/100 g edible material), thiamine (0.18 mg/100 g) riboflavin (0.04 mg/100 g), and niacin (0.09 mg/100 g) (Kay, 1973). The starch grains in the corm are very small so that taro is easily digested. The composition of the corms are as follows:

(Kay, 1973)

63-85%	water
13-29%	carbohydrate
1.4 - 3.0%	protein
0.16- 0.36%	fat
0.60- 1.18%	crude fiber
0.60- 1.30%	ash

Corms are also rich in a mucilaginous material, which on hydrolysis yields d-galactose and l-arabinose (Kay, 1973). Most cultivars contain

oxalic acid (0.1-0.4% fresh weight), mainly in the form of raphides which are bundles of needle-shaped Ca oxalate crystals (Kay, 1973; Sakai and Hanson, 1974; Sakai et al., 1972; and Srivasta and Krishnan, 1959a, 1959b). The acidity of raw taro has been attributed to these Ca oxalate crystals (Black, 1910).

Anatomy of Taro Corms

In Colocasia esculenta, starch is the primary storage product in the corm, a modified underground stem. The outermost zone of the mature corm contains 24-30 cork layers, which form part of the periderm (Paliwal and Kavathekar, 1972). Cork cells are rectangular, relatively thin-walled, and arranged in tiers. To the interior of the periderm is a ring of isolated mucilage ducts (Paliwal and Kavathekar, 1972). The rest of the corm is composed of ground parenchyma cells and a large number of isolated lactifers.

According to Paliwal and Kavathekar (1972), the corm of Colocasia esculenta shares the following characteristics with food storage organs in carrot, sweet potato and manioc:

1. abundance of periderm;
2. food storage in large, thin-walled parenchymatous cells;
3. poorly developed vascular bundles that are few in number;
4. presence of latex cells, mucilage cells, and ergastic substances such as druses and raphides.

Taro Growing in Hawaii

Taro is propagated vegetatively by stem cuttings called 'hulis', which consist of 0.6 cm of upper corm plus 15-25 cm of lower petiole (Kay, 1973). Rapid vegetative growth occurs up to 4-6 months after

planting and is followed by a maturation phase where each successive leaf decreases in size (Plucknett and de la Pena, 1971; Plucknett et al., 1970). Maximum leaf canopy is achieved after 5-6 months, while corm formation begins at 3-5 months. After 12-15 months, very little canopy exists and the corms are ready for harvest.

In Hawaii, much of the commercial taro is grown in flooded paddies. Water continuously flows throughout the growing season except when the fields are drained for fertilization. Fertilizers are usually applied in 2 or 3 applications within the first 4-6 months after planting (Plucknett and de la Pena, 1971).

Watson (1970) stated that based on his experience of taro growing in Hawaii, the water requirement of paddy taro ranges from 15,000 to 60,000 gal/A/day. He estimated a general average requirement of 30,000 gal/A/day.

Fertilizer Requirements of Taro

de la Pena and Plucknett (1969) demonstrated increased yields of upland and lowland taro with increased fertilizer applications of N, P, and K. Chew (1971) found a response of Colocasia to magnesium lime on acid peat soils, and he attributed part of the increased tuber yields to the Ca and Mg supplied.

Taro has been reported to have a high Ca requirement and liming is beneficial in soils low in Ca (Kay, 1973; Plucknett et al., 1970). Cable (1973) and de la Pena and Plucknett (1969) have reported decreased plant Ca content with increased K fertilization.

Guava Seed Disease of Taro

Guava seed disease of taro has been reported only in Hawaii

(Plucknett et al., 1970; Sickey, 1973). Other names for this disease are hard rot and kalakoa, which in Hawaiian means "black and white", referring to the color difference between diseased and healthy tissue in the corm. The small, localized corky tissues in the corm characteristic of this disease, can be dug out, leaving a smooth, round hole which resembles the seed pockets of guava, and hence the name "guava seed" disease (Parris, 1941; Plucknett et al., 1970).

Losses from guava seed disease vary from 5 to 100%, with an average loss estimated to be 30% throughout the islands (Parris, 1941; Takahashi, 1953). Takahashi (1953) suggested that further losses are indicated by the fact that yields of 60,000 lbs/A are possible under ideal conditions and the absence of disease, while the state average yield is only 13,457 lbs/A.

Characteristic Symptoms of Guava Seed Disease

The disease appears in two forms: (1) a woody core of coalesced corky tissue, and (2) scattered corky spots (Takahashi, 1953). The scattered form of this disease is much more prevalent than the coalesced form. Takahashi (1953) believed that the coalesced form is the more advanced stage of the disease, while the scattered form is the relatively early phase.

The characteristic corky spots are rings of periderm around dead tissues (Hawaii Agr. Exp. Sta., 1938). Takahashi (1953) demonstrated that the healthy region of the corm next to the infected area was reduced in starch content relative to a disease-free corm.

There are no foliar diagnostic symptoms of this disease (Hawaii Agr. Exp. Sta., 1936; Takahashi, 1953). Diseased corms can be

distinguished from healthy corms on the basis of the external appearance of the periderm. The periderm of healthy corms is smooth and paperlike in texture and thickness, whereas that of diseased corms is rough, barklike, and about one-eighth to one-fourth of an inch thick (Parris, 1941; Takahashi (1953). Takahashi (1953) claimed that 100% of all dumbbell-cylindrical-shaped corms have guava seed disease; however, although there may be a correlation, the writer has seen many corms with constrictions that didn't have the disease. One cause of growth restrictions is an erratic moisture supply, which results in a slow-down in growth during dry periods (Kay, 1973).

Cause of Guava Seed Disease

The cause of guava seed disease of taro is unknown. No organism has been found to be associated with this disease, despite repeated isolation attempts and microscopic examinations of diseased tissues (Parris, 1941; Sickey, 1973; Takahashi, 1953; Trujillo, 1979).

Guava seed disease is essentially a disease of taro grown under wetland culture. Parris (1941) showed that only healthy corms developed from hulis of diseased corms planted in non-submerged soils. However, all of the corms produced from diseased hulis planted in sterilized soil under submerged conditions had guava seed disease.

Parris (1941) found no correlation between pH of the soil and incidence of this disease. He also found no correlation between susceptibility of taro to this disease and N-P-K fertilizer treatments (Parris, 1941).

Takahashi (1953) stated that the following faulty agricultural practices are conducive to the development of such taro corm diseases

as guava seed disease:

1. inadequate field preparation and lack of a fallow period;
2. monoculture;
3. overly close plant spacings;
4. insufficient water movement in the paddy;
5. use of diseased or poor quality hulís;
6. poor field sanitation; and
7. excessive delay in harvesting after plants are mature.

According to Takahashi (1953), environmental factors such as water and air temperatures, relative humidity, and amount of sunlight all influence the prevalence and severity of corm diseases. As a result, losses from guava seed disease vary considerable from year to year (Takahashi, 1953).

Many investigators have suggested possible causes of guava seed disease (Hawaii Agr. Exp. Sta., 1936, 1937; Takahashi, 1953; Trujillo, 1967). The following is a list of speculative causes:

1. lack of aeration under submerged conditions;
2. salt damage to roots;
3. root damage by Pythium;
4. nutritional imbalance; and
5. inadequate irrigation after 8-9 months.

Disease Control

Lime at 2 T/A caused appreciable reductions in guava seed disease at two localities. However, Parris (1941) didn't recommend liming for disease control because its effects were inconsistent. His only recommendation to farmers was to plant disease-free material (Parris,

1941). Parris (1941) did suggest that breeding for resistance to corm rot might be the solution since taros of the Kai group seemed to possess resistance to both hard and soft rot.

Calcium Nutrition of Plants
and its Possible Relationship to Guava Seed Disease

Calcium is required by all plants and is one of the most immobile of the essential plant nutrients. It is necessary for normal growth and elongation of roots. Sorokin and Sommer (1940) found that apical cells of garden pea roots rapidly degenerated in the absence of calcium. Since excess Ca isn't remobilized readily, root surfaces must be continuously supplied with adequate Ca levels in the soil solution (Loneragan and Snowball, 1969).

Many authors maintain that the quantity of Ca required for normal growth of plant cells is in the micro range (Christiansen and Foy, 1979; Jones and Lunt, 1967; Loneragan and Snowball, 1969; Ludders, 1979; Wallace and Frohlich, 1966). Very low levels of Ca (3 to 50 μM) have been found to be adequate for normal development of roots, provided that toxic levels of Mg, Cu, Fe, Mn, and Zn are not present in the nutrient or soil solution. The requirement for Ca in either the soil or nutrient solution appears to be related to the ratio of Ca to total salts or total cations in solution, rather than Ca concentration alone (Geraldson, 1957a, 1957b; Gerard, 1971).

Calcium Deficiency

The typical symptoms of Ca deficiency found with plants grown in nutrient solution culture are stunting of the leaves, marginal blade chlorosis and necrosis, a die-back of the roots, and finally death of the growing shoots (Arnott and Pautard, 1970). In the field, Ca

deficiency is restricted to plant parts low in Ca, such as fruits, shoot apices, and storage organs (Kirkby, 1979).

Calcium accumulates in older tissue because once deposited at a sink, it is rarely remobilized within the plant. There is a lack of correlation between Ca levels in apple leaves and fruit Ca deficiency disorders because of the localized nature of deficiency symptoms (Millaway and Wiersholm, 1979). Calcium within the plant may be adequate on a total plant basis, but if the Ca isn't available at the newer root and shoot tissues or expanding storage organs, localized deficiency symptoms will occur. Wiersum (1966) maintained that fruit and storage organs were particularly susceptible to Ca deficiency because of preponderance of the water supply via the phloem to these organs, while Ca moves exclusively in the xylem. Examples of Ca related disorders are cavity spot of carrots, internal browning of brussel sprouts, poor filling of peanuts, cork spot of pears and apples, blackheart of celery, blossom end rot of tomatoes, and tipburn of lettuce and sugarbeets (Shear, 1975; Woodbridge, 1971).

Conditions Influencing Calcium Deficiency

Calcium deficiency can be induced by applying fertilizers high in soluble cations other than calcium (Pearson, 1975). Gerard (1971) showed that the solution Ca requirement for root elongation increased with increasing salt concentration.

Enhanced growth rates can result in Ca deficiency symptoms, especially when Ca levels in the soil or nutrient solution are low. Maynard and Barker (1972) found that internal browning of brussel sprouts occurred only when growth was enhanced at low Ca levels.

Restricted growth and low Ca levels didn't result in internal browning symptoms.

Varietal effects on susceptibility to Ca deficiency were studied by Brumagan and Hiatt (1966). They found that tobacco varieties susceptible to Ca deficiency had higher oxalic acid concentrations relative to non-susceptible varieties. They hypothesized that precipitation of Ca oxalate in the stalk resulted in Ca deficiency in the inflorescence.

Walker (1969) examined temperature effects on occurrence of Ca deficiency. He showed that Ca deficiency symptoms in maize increased with increasing soil temperatures from 22 to 35°C. The Ca concentration in the top half of the blades were found to be in the deficiency range. Chang et al. (1968) found similar results for tobacco and hypothesized that Ca was immobilized in tobacco stems at higher temperatures, resulting in Ca deficiency in the terminal leaves and meristem.

Role of Ca in Membrane Stabilization

Calcium is reported to be involved in at least 20 specific biochemical processes in plants (Marschner and Gunther, 1964). Its primary function is generally agreed to be maintenance of membrane integrity which is necessary for ion uptake and retention (Christiansen and Foy, 1979; Epstein, 1972).

Calcium is involved in stabilization of membranes against such stress factors as low pH, heat, cold, oxygen deficiency, and toxic ion levels (Christiansen and Foy, 1979). Calcium deficiency results in deficiency, and toxic ion levels (Christiansen and Foy, 1979).

Calcium deficiency results in disintegration of the plasmalemma and tonoplast, destruction of all cytomembranes, and accumulation of degenerative vesicles (Hecht-Bucholz, 1979; Marinos, 1962; Marschner and Gunther, 1964).

Morre and Bracker (1976) found that a conformational change in the plasma membrane occurred in the presence of CaCl_2 and auxin. The plasma membranes treated with CaCl_2 were thicker than controls while those treated with auxin were thinner than controls. The CaCl_2 effect occurred at $0-4^\circ\text{C}$, suggesting that Ca had a physical effect on membrane structure rather than a metabolic one. The nonmetabolic stabilization of membranes by Ca ions has been supported by the work of other researchers (Christiansen et al., 1970).

Gary-bobo (1970) showed that in the presence of Ca, osmotic flow of water across artificial phospho-lipid membranes decreased and he attributed this effect to decreased membrane porosity. Garrard and Humphrey (1967) hypothesized that Ca links certain membrane anionic groups together to form bridges between structural units. Since the permeability properties of membranes is determined by the size of the pore and the size of the molecules, it is possible that Ca affects the pore radius of membranes.

Poovaiah (1979a, 1979b) showed that Ca delayed leaf senescence in corn leaf discs and prevented membrane leakage in beet root slices treated with ethephon. He interpreted these effects as being due to Ca maintenance of cell wall structure and membrane integrity.

Paul and Goff (1973) found that Ca deficiency blocked cell plate formation in onion roots. Since they found a very similar effect with

caffeine, they postulated that Ca, ATP, and a Ca or Mg-activated ATPase are necessary for membrane fusion. Caffeine is known to release membrane-bound Ca and to inhibit a Mg-stimulated ATPase. The release of Ca and ATP from the membrane results in membrane destabilization which is a prerequisite for membrane fusion.

Role of Ca in Maintenance of Cell Wall Structure

Calcium is a structural part of the cell wall and is present in the form of calcium pectates, which cross-link to form a rigid lattice-work (Millaway and Wiersholm, 1979). This process of cross-linkage increases the resistance of the tissue to infection, probably by increasing the resistance of the walls to degradation by pathogen-produced polygalacturonidase (Jones and Lunt, 1967). Field studies have shown that Ca fertilizer reduces the incidence of certain pathogenic diseases in cotton and tomatoes (Millaway and Wiersholm, 1979).

Gerard and Hinojosa (1973) found that Ca was essential to the development of thick cell walls in the stele of cotton roots. Dickinson and McCollum (1964) showed that Ca had a strengthening effect on tomato fruit tissue and they attributed the firming effect to the formation of Ca pectates.

Arnott and Pautard (1970) stated that Chlorella has little pectin present in the cell wall and this algae can grow on very small amounts of calcium. Spirogyra has a high Ca requirement as well as large amounts of pectin in the cell wall. Perhaps Ca is a macro-nutrient only in plants with cell walls composed of Ca pectates.

Other Roles of Ca in Plant Nutrition

Calcium has also been found to be necessary for the proper functioning of growth hormones (Cooil and Bonner, 1956). It has been recently demonstrated that auxins require attachment to a specific membrane site and Ca operates in this binding of auxin (Poovaiah and Leopold, 1976).

Calcium is also postulated to have a role in binding to ribonucleic protein (Jones and Lunt, 1967). Sorokin and Sommer (1940) found chromosome abnormalities associated with Ca deficiency. Steffensen (1958) found that decreased Ca levels resulted in increased frequency of x-ray induced chromosome breaks.

Calcium Uptake and Translocation

It is generally agreed that Ca moves primarily with water into the root tissues (Millaway and Wiersholm, 1979). The plant transpiration rate and Ca uptake are highly correlated (Marschner, 1974).

Calcium movement in the plant is unidirectional, moving from the roots to the young shoot tissues (Hanger, 1979). Once deposited in the leaf tissue, Ca isn't recycled in most annual plants even under Ca stress (Arnott and Pautard, 1970; Hanger, 1979).

Ferguson and Clarkson (1976) showed that the pattern of uptake and translocation of Ca and Mg was very similar. Both cations appeared to move across the root cortex primarily in the free space. Suberization of the endodermis greatly reduced uptake of either ion. Thus, the main site of Ca and Mg uptake is at the growing root tip where the suberized endodermis is absent (Kirkby, 1979).

Calcium is thought to move up the xylem by a series of ion exchange reactions along negatively charged sites on the vessel walls

(Biddulph et al., 1961; Hanger, 1979). Movement can be promoted by the presence of divalent cations and by chelation of Ca.

Marschner (1974) maintained that the immobility of Ca in plants is due to its inability to move in the phloem. The high pH and high phosphate concentration in the phloem would precipitate Ca, thus interfering with phloem transport.

Calcium is deposited in idioblasts, mostly in the form of oxalate or carbonate (Arnott and Pautard, 1970). The steady accumulation of Ca oxalate, along with the immobility of crystals over a long period has resulted in the prevailing opinion that the salt serves as a mechanism by which excess Ca or oxalic acid is removed from the tissues.

Interactions of Calcium and Other Ions

Cations such as NH_4 , K, and Mg depress uptake of Ca (Millaway and Wiersholm, 1979), and there is ample evidence that high levels of these ions can cause severe Ca deficiency (Geraldson, 1957a, 1957b; Hanger, 1979; Kawaski and Moritsugu, 1979; Ludders, 1979; Mostafa and Ulrich, 1976; Shear, 1972, 1971; Wilcox et al., 1977, 1973). It is unlikely that these cations would be competing for the same carrier binding site because of differences in size and structure of the ions.

One explanation for the depressed uptake of Ca ions by NH_4 and K ions was proposed by Kirky (1979). Cells produce anion equivalents which attract cations nonspecifically. Those cations absorbed the fastest (such as K or NH_4), neutralize these anion equivalents first and reduce the electrostatic attraction for the uptake of other cation species (such as Ca).

Nitrogen can seriously affect the Ca balance in plants through its stimulation of rapid tissue growth which may create a Ca demand that cannot be met from the Ca available in the soil solution (Shear, 1975). Nitrogen also promotes shoot growth often at the expense of root growth, which could limit the extraction of soil or nutrient solution calcium.

Calcium has long been reported to interfere with the absorption of K, however Reitemeier (1951) showed evidence that Ca ions had the smallest effect on reducing K uptake while NH_4 had the greatest effect, Jacobson et al. (1960), in fact, found that Ca is required for the preferential uptake of K ions over Li or H when these competing ions are present in solution. There is evidence of a Ca-stimulated, membrane-bound ATPase which mediates K movement (Christiansen and Foy, 1979).

Magnesium and Ca are taken up and translocated along similar pathways, so it's not surprising that they interfere with each other. Mostafa and Ulrich (1976) showed that Mg interfered with Ca uptake and it was possible to induce Ca deficiency symptoms on sugarbeet with higher levels of magnesium. On the other hand, high concentrations of Ca interfered with Mg uptake but didn't induce Mg deficiency symptoms.

II. CALCIUM NUTRITION OF TARO (COLOCASIA ESCULENTA (L.) SCHOTT)

Introduction

Taro (Colocasia esculenta (L.) Schott) contains relatively high amounts of calcium (Plucknett and de la Pena, 1971). Miller (1927) calculated that the daily adult requirement of calcium would be more than adequately met when taro comprised the bulk of the diet. The strong skeletons and excellent teeth of the ancient Hawaiians, despite a diet lacking milk, provide additional evidence that taro can supply the required calcium for human needs (Potgieter, 1940).

The presence of calcium oxalate crystals in taro has long been recognized (Black, 1918), and the structure of the calcium oxalate raphides was described recently by Sakai and Hanson (1974). The function of calcium oxalate crystals is not known, but Arnott and Pautard (1970) suggested that storage of calcium in an inactive form allows certain plants to tolerate high levels of calcium.

Taro has a high calcium requirement and liming is beneficial in soils low in calcium (Plucknett and de la Pena, 1971); Kay, 1973), Chew (1971) reported increased tuber yields when acid peat soils were limed and he concluded that this response was partly due to the calcium and magnesium supplied by the lime.

Low levels of soil calcium have been found in taro growing areas in Hawaii by the Soil Testing Service of the University of Hawaii, Manoa. Although liming is recommended, little research has been done on the growth response of taro to calcium. This study was designed to characterize calcium deficiency symptoms and plant tissue levels of calcium in taro.

Materials and Methods

This experiment was conducted from July to August, 1978 in a greenhouse whitewashed to reduce light intensity to 70% of full sunlight. The taro plants were grown in glazed 10 liter ceramic crocks using a modified Hoagland's solution (Hoagland and Arnon, 1938). Micronutrients were one-fourth strength and iron was supplied from CIBA-Geigy Sequestrene 330 Fe.

The experimental design was a randomized complete block with 7 Ca levels and 3 replicates. Within each block, the taro 'hulis' (vegetative propagating material consisting of approximately the upper 0.5 cm of the corm plus 20 cm of the petioles) were selected for uniformity on the basis of fresh weight. The seven levels of Ca were 0.05, 0.10, 0.20, 0.50, 1.00, 4.00, and 20.00 parts per million (ppm) in the modified Hoagland's solution. In solutions where the concentration of Ca was less than 20 ppm, nitrogen was maintained by substituting NH_4NO_3 for $\text{Ca}(\text{NO}_3)_2$.

One-tenth strength modified Hoagland's solution was continuously dripped into the crocks from a 50 liter reservoir at a rate of 2.4 liters per day. A constant head device maintained a fairly constant flow rate of nutrient solution and flow rates were checked daily. Overflow tubes maintained a constant level as air was continuously bubbled through the solution.

The taro cultivar grown in this experiment was 'Lehua maoli', the primary commercial cultivar in Hawaii. 'Hulis' were supported on 2 crossed rubber strips that were suspended in the nutrient solution.

The plants were harvested after 7 weeks of growth at which time the shoot tips of several plants had died in the low calcium solutions. All blades on a plant were composited for tissue analysis. Petioles on a plant were separated on the basis of age, analyzed and the results composited. Roots and the corm on a plant were composited for tissue analysis.

Plant tissue analyses were carried out at the Plant Tissue Testing Lab at the University of Hawaii, Manoa. Tissue nitrogen was measured using the micro-Kjehldal method of Suehisa and Deputy (1979). Plant tissue levels of P, K, Ca, Mg, S, Si, Na, Cl, Mn, Fe, Cu, and Zn were determined with an Applied Research Laboratories vacuum x-ray fluorescence quantometer, model 72000.

Results

Calcium deficiency symptoms were observed on taro grown in solutions containing 1.0 ppm Ca or less. With mild Ca deficiency (1.0 ppm Ca), a slight interveinal chlorosis was evident on the leaf blades. As Ca concentrations in the solution decreased, plant tissue levels decreased (Figure 1) and visual deficiency symptoms increased in severity (Figure 2). The leaf blades of moderately Ca-deficient plants (0.50 ppm Ca) were extremely chlorotic in interveinal areas with some necrosis and the blades appeared cup-shaped (Figures 2A, 3A). With severe Ca deficiency (0.05 to 0.20 ppm Ca), the leaf blades failed to unfurl and the growing point died in several plants (Figure 3B).

Calcium deficiency symptoms of taro were also marked by severe stunting of the plant, and drooping of the petioles at the lower Ca

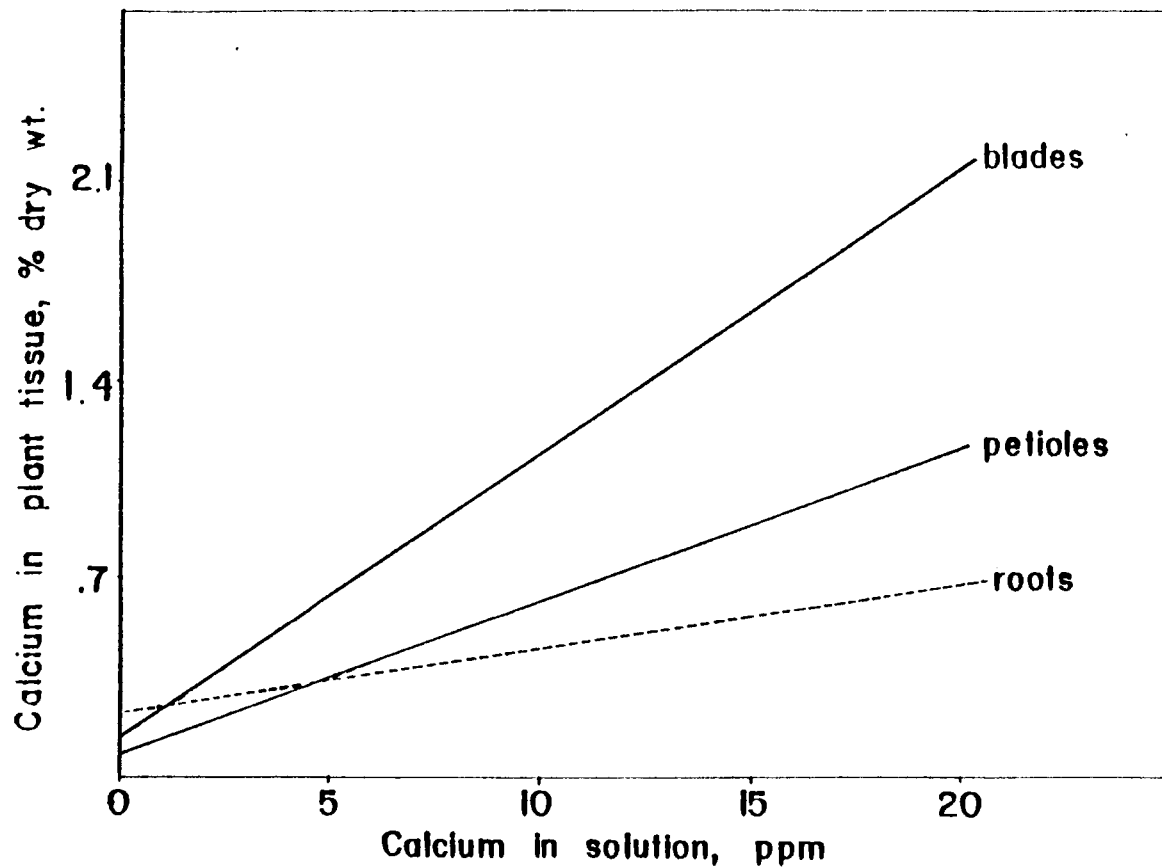


Figure 1. Relationship between Ca in solution and Ca concentration in taro plant tissues. Equations for the lines are, respectively, blades $y = 0.14 + 0.10(x)$, $r^2 = 0.99$; petioles, $y = 0.075 + 0.054(x)$, $r^2 = 0.97$; and roots, $y = 0.21 + 0.026(x)$, $r^2 = 0.95$.

Explanation of the Following Figures

Figure 2. Effect of Ca levels on the growth of taro. Note the stunting of taro plants grown at Ca levels of 0.05 to 0.50 ppm in solution (A) as compared with those grown at Ca levels of 1 ppm and above (B).



Figure 3. Symptoms of Ca deficiency. Marginal and interveinal chlorosis and necrosis can be seen on the leaf blades (A). Note the drooping petioles and failure of the leaf blades to unfurl (B).



levels (Figures 2A, 3B). Calcium deficiency also resulted in die-back of the roots and reduced mother corm size. The mean number of cormels increased significantly with Ca in solution from 2 at 0.05 ppm Ca to a maximum of 5 at 4.0 and 20.0 ppm Ca (Table 1). Total dry weight of the plant increased with Ca in solution to a maximum at 4.0 ppm, which corresponded to a Ca concentration of 0.57% dry weight in the leaf blades (Figure 4). Total plant dry weight decreased slightly at 20.0 ppm Ca in solution (Table 1).

Calcium concentration in plant tissues increased linearly with increasing Ca in solution (Figure 1). Leaf blades had the highest rate of increase in Ca concentration with increasing Ca in solution, petioles were intermediate, and roots the lowest. Calcium concentration in the petiole increased with age of the petiole (Table 2). Petiole 1 was from the youngest fully unfurled leaf, and petioles 2, 3 and 4 were from progressively older leaves.

The concentration of many nutrient elements was found to decrease with increasing Ca in solution. However, it was necessary to separate actual Ca interference with nutrient uptake or translocation from dilution effects due to increased growth. Comparisons of tissue nutrient levels were limited to plants grown with 4.0 and 20.0 ppm Ca in solution, because no significant difference was found in the growth of taro at these Ca levels (Table 1) and Ca concentration in the plant tissues was significantly higher in plants grown with the higher level of calcium (Table 3).

Magnesium was the only element that decreased significantly in both leaves and petioles of plants as Ca in solution increased from

Table 1. Effects of Ca in solution on total plant dry weight of taro and cormel number.

<u>Ca in solution, ppm</u>	<u>Plant dry weight, g</u>	<u>Number of cormels</u>
0.05	7.5a	2.0a
0.10	8.3a	3.0ab
0.20	8.6a	3.3ab
0.50	14.0ab	4.0ab
1.0	16.5ab	3.7ab
4.0	27.4b	5.0b
20.0	22.1ab	5.0b

¹ Means in a column followed by the same letter are not significantly different (95% probability level) as determined by the Studentized range test (Snedecor and Cochran, 1967).

Table 2. Calcium concentration in petioles of taro grown with
4.0 ppm calcium.

Petiole Number	1	2	3	4
Ca, % dry wt. ¹	0.20a	0.26a	0.35b	0.37b

Table 3. Effect of Ca in solution on tissue Ca concentration of taro.

Ca in solution, ppm	Ca, % Dry weight ¹		
	Blade	Petiole	Root
0.05	0.17a	0.06a	0.19a
0.10	0.14a	0.06a	0.23a
0.20	0.10a	0.10a	0.24a
0.50	0.18a	0.09a	0.23a
1.0	0.25a	0.20a	0.25a
4.0	0.57a	0.25a	0.29ab
20.0	2.09b	1.17b	0.74b

¹ Means in a column followed by the same letter are not significantly different (95% probability level) as determined by the Studentized range test.

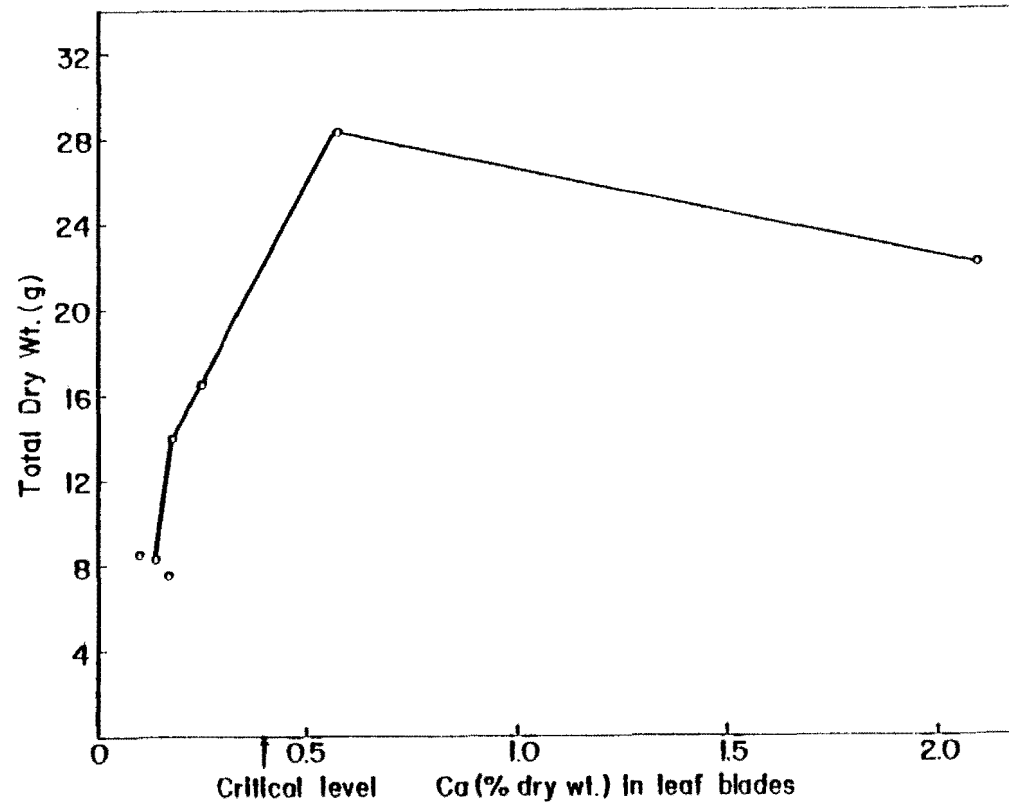


Figure 2. Changes in total dry weight of taro plant with changes in Ca concentration in leaf blades.

4.0 to 20.0 ppm (Table 4). Magnesium concentration in the roots increased significantly with increasing Ca in solution.

Discussion

Calcium deficiency symptoms of taro are typical of such symptoms found on other plants (Arnott and Pautard, 1970; Spense and Ahmad, 1967). The inability of leaf blades to unfurl, wrinkling of leaf blades due to bands of necrotic tissues, collapse of petioles, die-back of roots, and death of the growing point are commonly described Ca deficiency symptoms.

Luxury consumption of Ca was demonstrated with taro at 7 weeks. As Ca concentration in the leaf blades increased from 0.57% to 2.09% (dry weight), total dry weight of the plants decreased slightly (Figure 4). The high levels of Ca in the plant tissues seemed to have interfered with translocations of Mg, and this could account for the slightly decreased growth. Other experiments have shown that Ca interfered with Mg transport to the shoot, while Mg accumulation by root tissues was less affected (Jackson, 1967). The data for taro are in agreement with those from previously reported experiments.

An inadequate supply of soil calcium could reduce commercial yields of taro by reducing corm size and number of cormels. Tissue sampling could be used to diagnose this problem in the field. The leaf blades are probably the best index tissue because of their sensitivity to changes in Ca levels in the external solution (Figure 1). Since Ca concentration in the petiole increased with age of the petiole (Table 2), and since it is well known that Ca tends to

Table 4. Effect of calcium in solution on plant magnesium levels of taro.

Ca in solution, ppm	Mg, % dry wt.			Total, mg		
	Blade	Petiole	Root	Blade	Petiole	Root
4.0	0.51	0.43	0.29	50.58	36.85	22.62
20.0	0.41	0.34	0.74	21.94	17.37	42.70
	*	*	*	*	*	n.s.

* Means are significantly different (95% probability level) as determined by ANOVA.

n.s. Means are not significantly different.

accumulate in older tissues (Millaway and Wiersholm, 1979), it is likely that Ca concentration in the blade would also increase with age of the blade. Thus, blade 3 or 4 is probably the best tissue to sample for plant Ca levels because of higher Ca content.

The critical level of Ca in the leaf blades was determined by plotting two perpendicular lines, one parallel with the x-axis and the other with the y-axis, so that a minimum number of observations were in the upper left and lower right quadrants (Gate and Nelson, 1965). The critical concentration of 0.4% Ca on a dry weight basis (Figure 4) was estimated by the point of intersection of the 2 lines. However, further work in the field needs to be done to correlate commercial yields with plant tissue analyses at different plant ages.

Summary

Calcium deficiency symptoms on taro, Colocasia esculenta (L.) Schott cv. 'Lehua maoli', grown in nutrient culture for 7 weeks, were leaf blade interveinal chlorosis and necrosis, failure of the leaf blades to unfurl, collapse of petioles, die-back of roots, and ultimately death of the growth point. The critical level of calcium in the leaf blades was 0.4% on a dry weight basis for 7 week-old plants. The best indicator tissue was the blade of the third or fourth leaf because of its greater sensitivity to changes in calcium levels.

III. CALCIUM NUTRITION OF TARO,
COLOCASIA ESCULENTA (L.) SCHOTT,
AND ITS POSSIBLE RELATIONSHIP TO GUAVA SEED DISEASE

Introduction

Taro (Colocasia esculenta (L.) Schott) produces edible corms and cormels which are rich in starch (Kay, 1973; Plucknett et al., 1970). Guava seed disease of taro is characterized by the development of corky tissues in the corm (Parris, 1941). No disease organism has been found to be associated with this disease, despite repeated isolation attempts and microscopic examinations of diseased tissues (Parris, 1941; Trujillo, 1979). This disease reduces the marketable yield of taro, because those portions of the corm affected by the disease are discarded.

Parris (1941) reported that liming at 2 T/A decreased guava seed disease incidence at 2 localities. However, he didn't recommend liming for disease control because of its inconsistent effects on disease at other localities.

Low levels of soil Ca have been found in taro growing areas by the Soil Testing Service of the University of Hawaii, Manoa. Although liming is recommended little research has been done on the Ca nutrition of taro. This study was carried out with three objectives: (1) to determine the growth response of taro to calcium; (2) to study the effects of excess nitrogen fertilization on Ca nutrition of taro; and (3) to determine whether guava seed disease could be induced by low levels of calcium, high levels of nitrogen, or a combination of the two.

Materials and Methods

This experiment was conducted from August 1978 to March 1979 in a greenhouse whitewashed to reduce light intensity to 70% of full sun-ling. The taro plants were grown in glazed 20 liter ceramic crocks using a modified Hoagland's solution (Hoagland and Arnon, 1938). Modifications were as follows: micronutrients were one-fourth strength and iron was supplied from CIBA-Geigy Sequestrene Fe 330.

Hoagland's solution was continuously dripped into the crocks from a 50 liter reservoir at a rate of 2.4 liters per day. A constant head device maintained a fairly constant flow of nutrient solution and flow rates were checked daily. Overflow tubes maintained a constant level as air was continuously bubbled through solution.

The taro cultivar grown in this experiment was 'Lehua maoli', the primary commercial cultivar in Hawaii. The plants were supported on sections of plastic "egg crate" light diffusers which were cut to the diameter of the crocks and suspended in the nutrient solution. The supporting frames were adjusted to keep the corms submerged.

The experimental design was a randomized complete block with 4 levels of calcium (Ca), 2 levels of nitrogen (N), and 6 replicates. Within each block, the taro 'hulis' (vegetative propagating material consisting of approximately the upper 0.5 cm of the corm plus 20 cm of the petioles) were selected for uniformity on the basis of fresh weight.

For the first 3 months of growth, one-tenth strength Hoagland's solution was adjusted to contain 1, 2, 4, and 20 parts per million (ppm) of Ca while the two levels of N were 21 and 100 ppm (see Table 1).

Table 1. Dates of the samplings, harvests, and changes in nutrient concentration used for the solution culture of taro.

Date	Month	Activity	Strength of Hoagland's Solution	Ca, ppm	N, ppm
Aug.10 - Sept.9	1	—	one-tenth	1,2,4,20	21,100
Sept.10 - Oct. 9	2	—	one-tenth	1,2,4,20	21,100
Oct.10 - Nov. 4	3	—	one-tenth	1,2,4,20	21,100
Nov. 4	3	Sample			
Nov. 5 - Dec. 4	4	—	one-fifth	2,4,8,40	42, 42
Dec. 5 - Jan. 6	5	—	one-fifth	2,4,8,40	42, 84
Jan. 6	5	Sample			
Jan. 7 - Feb 7	6	—	one-fifth	2,4,8,40	42, 84
Feb. 7	6	Harvest			
Feb. 8 -March 11	7	—	one-tenth	1,2,4,20	21, 42
March 12-April 12	8	—	one-tenth	1,2,4,20	0, 0
April 12	8	Harvest			

In solutions where the concentration of Ca was less than 20 ppm, N was maintained by substituting NH_4NO_3 for $\text{Ca}(\text{NO}_3)_2$. In solutions where the level of N was 100 ppm, NH_4NO_3 was added.

The lower N treatment had the same level of solution N as Hoagland's solution, and so was considered to be an adequate level. The higher N treatment had 5 times the solution N level in the lower N treatment, and so was considered to be an excess level.

The levels of K, Ca, and Mg in the crocks were monitored weekly. By the end of 3 months, the levels of solution K in the crocks with rapidly growing taro plants were extremely low (0.1-0.2 ppm) and so Hoagland's solution was increased at the beginning of the fifth month from one-tenth to one-fifth strength. Thus, the Ca levels in the stock solution increased to 2, 4, 8, and 40 ppm.

The N levels in both N treatments were maintained at 42 ppm for the duration of the fourth month, because Ca deficiency symptoms were so severe at the higher N treatment and two lower Ca treatments that the shoot tips were dying. The higher N treatment was increased to 84 ppm at the beginning of the fifth month, twice that of the lower N treatment.

Following the harvest of three replicates after 6 months, Hoagland's solution was decreased from one-fifth to one-tenth strength, to promote corm maturation. In commercial taro fields, all fertilizers are applied by 6 months after planting. Guava seed disease in the fields occurs particularly during the maturation phase after 6 months growth when taro leaf area is decreasing and the corm is expanding.

After 7 months, no N or K was added to the nutrient solution. The levels of Ca and P were maintained by substituting CaCl_2 for

$\text{Ca}(\text{NO}_3)_2$ and NaH_2PO_4 for KH_2PO_4 .

The third youngest fully expanded leaf was harvested for tissue analysis after 3 and 5 months of growth. Three of the 6 replicates were harvested after 6 months and the other 3 after 8 months. Leaves 1 to 4 were sampled at the time of harvest. Leaf 1 is the youngest fully unfurled leaf and leaves 2, 3, and 4 are consecutively older. The corms were examined for incidence of guava seed disease, and corm starch content was determined by the method of Allen et al. (1974). The standards for starch analysis were prepared using commercially available St. Vincent arrowroot (Maranta arundinacea) starch. Dry weights were obtained by placing the plant samples in drying ovens at 55°C for one week.

Plant tissue nutrient element analyses were carried out at the Plant Tissue Testing Lab at the University of Hawaii, Manoa. Tissue nitrogen was measured using the micro-Kjehldal method of Suehisa and Deputy (1979). Plant tissue levels of P, K, Ca, Mg, S, Si, Na, Cl, Mn, Fe, Cu, and Zn were determined with an Applied Research Laboratories vacuum x-ray fluorescence quantometer, model 72000.

The Mitscherlich growth equation (Tisdale and Nelson, 1975) was used to describe the response of taro to blade Ca levels. The equation, $y = B_0 (1 - \exp(-B_1 * x))$, was fitted to the data and the coefficients were calculated. The coefficient B_0 is the maximum yield and the coefficient B_1 is a measure of the slope of increasing yield with increasing blade Ca when y is yield and x is blade calcium.

Results and Discussion

Calcium deficiency symptoms on taro plants grown with 1 and 2 ppm

solution Ca were similar to those described previously in Chapter II. The plants were stunted, the roots were discolored, the leaf blades were interveinally chlorotic and necrotic, and the petioles drooped. The leaf blades tended to be cup-shaped and brittle, tearing readily under slight pressure.

Leaf Sample at 3 Months

From planting to sampling after 3 months of growth, one-tenth strength Hoagland's solution was used. The 4 solution Ca levels were 1, 2, 4, and 20 ppm while the 2 solution N levels were 21 and 100 ppm.

The higher N treatment and 2 lower Ca treatments resulted in extremely stunted and chlorotic plants. Due to inadequate blade and petiole dry weights at these treatments, replicates were composited for tissue analysis.

For statistical evaluation of N, Ca, and N by Ca interaction effects, only data from the two higher Ca treatments (4 and 20 ppm) and the two N treatments were analyzed (Table 2). The higher N treatment combined with the 2 lower Ca treatments had an insufficient number of replicates and so the 2 lower Ca treatments were not included in this analysis of variance (Table 2). Then for a more complete picture of the effect of all levels of Ca, data from the lower N treatments was analyzed separately (Table 3).

Increased solution N resulted in significant decreases in leaf (both blade and petiole) dry weight and leaf Ca concentration (Table 2). This depression in growth with high amounts of solution N was likely due to decreased Ca uptake caused by high levels of NH_4 ions present.

Table 2. Effects of solution N and Ca on the dry weight and nutrient content of the third blade of taro at 3 months.

Variable	Main Effects				Interaction Effect ^{1,2}			
	N, ppm		Ca, ppm		N, ppm			
	21		100		21		100	
	Ca, ppm		Ca, ppm		Ca, ppm		Ca, ppm	
	21	100	4	20	4	20	4	20
Dry wt., g								
Blade	3.4*	1.9	2.0*	3.2				
Petiole	2.2	1.1	1.3	2.0				
Ca, % dry wt.								
Blade	1.23*	0.78	0.50*	1.52	0.63c	1.83a	0.37d	1.20b
Petiole	0.76*	0.56	0.29*	1.02	0.31c	1.20a	0.27c	0.84b
N, % dry wt.								
Blade	4.4*	5.2	5.0*	4.6				
Petiole	2.8*	4.4	4.2*	3.0				
K, % dry wt.								
Blade	4.1*	4.5	4.5*	4.0				
Petiole	6.0*	7.4	7.4*	6.0				
Mg, % dry wt.								
Blade	0.38	0.33	0.42*	0.30				
Petiole	0.57	0.48	0.56*	0.49	0.76a	0.38c	0.35c	0.60b

* Means for the N and Ca main effects are significantly different at the 95% probability level as determined by ANOVA.

¹ Means in a row followed by the same letter are not significantly different at the 95% probability level as determined by Duncan's multiple range test.

² Means for all treatments are included only if the interaction effect is significant at the 95% probability level as determined by ANOVA.

Table 3. Effects of solution Ca on the dry weight and nutrient content of the third leaf at 3 months.

Variable	Ca, ppm ¹			
	1	2	4	20
Dry wt., g				
Blade	1.7c	2.1c	3.0b	3.8a
Petiole	0.9c	1.2c	1.8b	2.6a
Ca, % dry wt.				
Blade	0.15d	0.31c	0.63b	1.83a
Petiole	0.11c	0.20bc	0.31b	1.20a
Total Ca, mg				
Blade	2.4d	6.3c	18.9b	69.2a
Petiole	1.1c	2.5c	5.7b	29.8a
N, % dry wt.				
Blade	4.3bc	4.7a	4.6ab	4.2c
Petiole	4.2a	4.0a	3.4a	2.2b
Total N, mg				
Blade	72.0b	100.0b	138.0a	158.0a
Petiole	38.0a	48.0a	61.0a	57.0a
K, % dry wt.				
Blade	4.7ab	4.9a	4.4b	3.7c
Petiole	9.2a	8.6ab	7.1b	5.0c
Total K, mg				
Blade	78.0c	103.0bc	131.0ab	142.0a
Petiole	83.0a	103.0a	129.0a	130.0a
Mg, % dry wt.				
Blade	0.52a	0.59a	0.48a	0.29b
Petiole	0.51bc	0.65ab	0.76a	0.38c
Total Mg, mg				
Blade	8.3b	12.2ab	14.7a	10.9ab
Petiole	5.3b	8.6b	14.1a	9.7b

¹ Numbers in a row followed by the same letter are not significantly different at the 95% probability level as determined by Duncan's multiple range test.

Plants grown with the higher N level and low Ca levels appeared to have more severe Ca deficiency symptoms than those grown with the lower solution N levels and low Ca levels.

There is ample evidence in the literature that high levels of NH_4 salts can cause severe Ca deficiency (Hanger, 1979; Shear, 1972; Wilcox et al., 1977, 1973). The NH_4 ions can interfere with the absorption of Ca ions by neutralizing plant anion equivalents which attract solution cations (Hanger, 1979).

Increased solution N also resulted in significant increases in leaf N and K concentrations (Table 2). The increased leaf K concentration with increased solution N was probably due to luxury consumption of K at the higher N treatment where growth was retarded. Total K in the blade (blade K concentration times blade dry weight) decreased significantly from 137 milligrams (mg) at the lower N treatment to 84 mg at the higher N treatment.

As solution Ca increased, significant decreases in leaf (both blade and petiole) N and K concentrations occurred. These decreases were probably due to dilution effects caused by increased growth with increasing solution calcium. There was no significant difference in total N or K in the petiole, while total N or K in the blade increased significantly with increasing Ca in solution (Table 3).

Magnesium concentration in the leaf increased with increasing solution Ca up to 2 or 4 ppm and then decreased at 20 ppm (Table 3). Total Mg in the leaf showed a significant increase up to 4 ppm Ca in solution and a subsequent decrease at 20 ppm (Table 3). This decrease in total Mg was significant for the petioles but non-significant for the blades. It seemed that Mg uptake or translocation

was inhibited at very low and very high solution Ca levels.

As solution Ca increased, significant increases occurred in leaf dry weight and leaf Ca concentration (Table 3). A significant N by Ca interaction was found in leaf Ca concentration (Table 2). The rate of increase in leaf Ca with increasing levels of solution Ca was significantly less at the higher N treatment compared with the lower N treatment (Figures 1, 2).

Since the higher N treatment had a stunting effect on growth, which obscured the Mitscherlich type of growth response to solution Ca, only data from the lower N treatment was fitted to the growth curve equation to estimate the critical tissue Ca concentration (Figure 3). The critical Ca concentration of the third blade at 3 months, the Ca value at 95% of maximum blade dry weight, was estimated to be 0.8% on a dry weight basis.

This method of estimation of the critical tissue level for Ca is valid only if a high correlation exists between third blade dry weight and corm yield at harvest. Corm yield is determined by a series of blade dry weights over a fairly long period of time (6-12 months), however blades are much shorter-lived plant organs and reflect environmental conditions and nutrient status over only a month. Moreover, although the r^2 value for curve fitting using the Mitscherlich equation is significant at the 95% probability level, it is not sufficiently high to permit one to make predictions about critical levels of tissue Ca with confidence.

Leaf Sample at 5 Months

Rapid growth of the plants during the first 3 months resulted

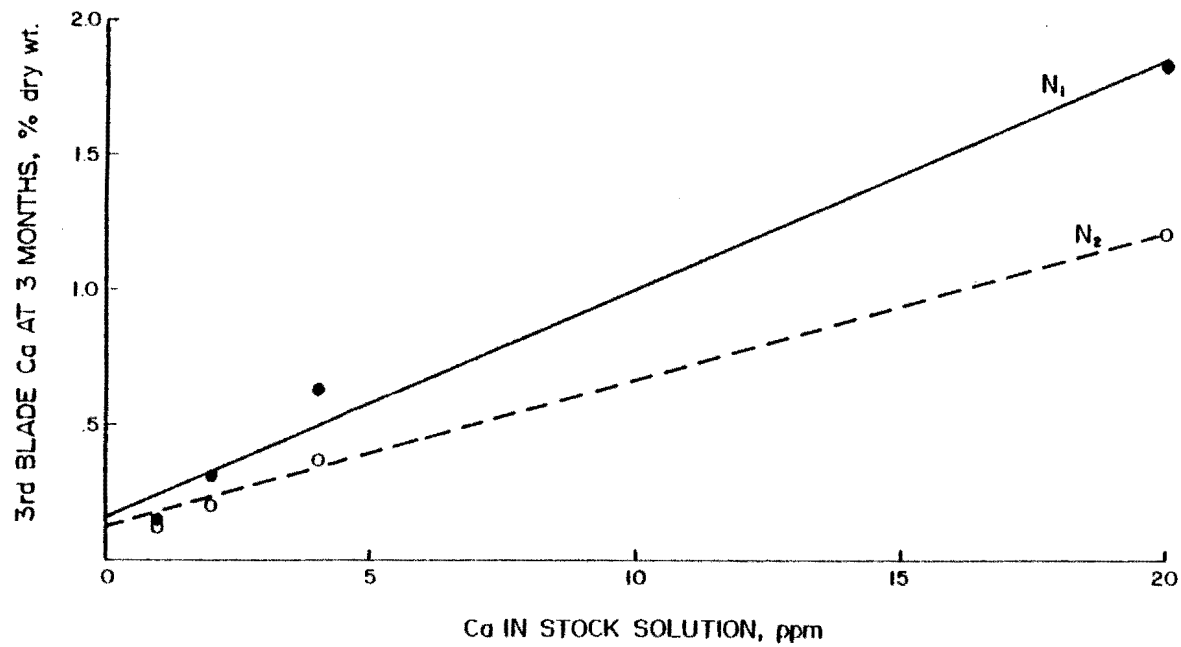


Figure 1. Relationship between Ca in solution and Ca concentration in the third blade of taro at 3 months. Equations for the lines are, N_1 , $y = 0.16 + 0.08(x)$, $r^2 = 0.96$; and N_2 , $y = 0.13 + 0.05(x)$, $r^2 = 0.91$.

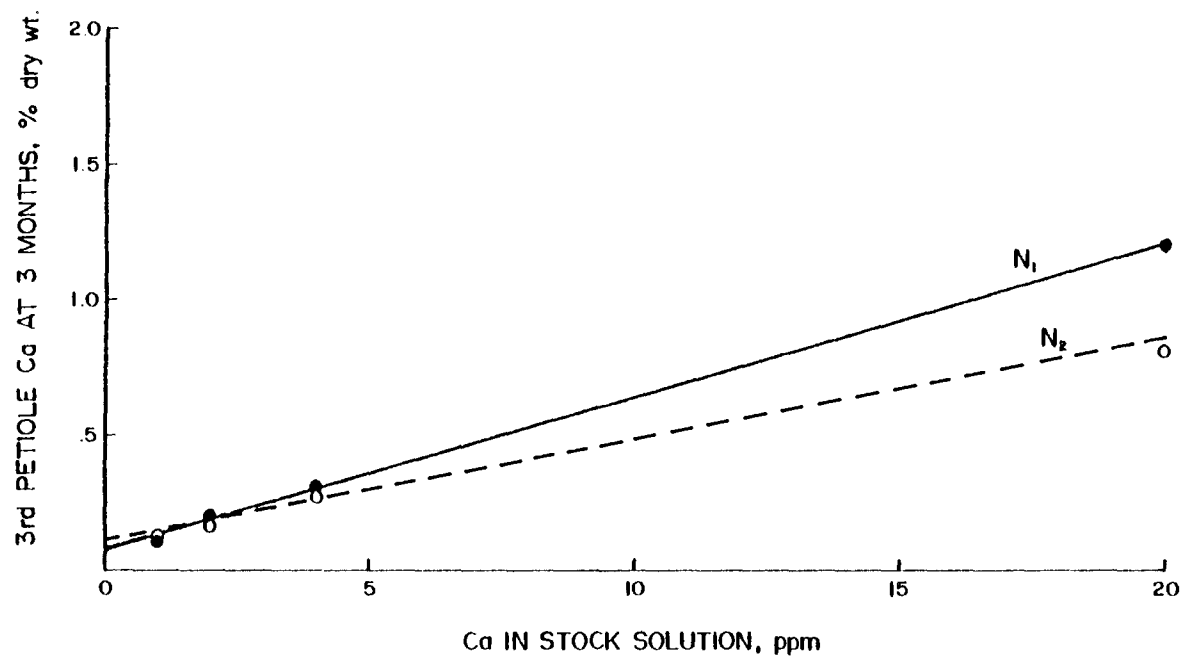


Figure 2. Relationship between Ca in solution and Ca concentration in the third petiole of taro at 3 months. Equations for the lines are, respectively, N_1 , $y = 0.08 + 0.06(x)$, $r^2 = 0.90$; and N_2 , $y = 0.11 + 0.04(x)$, $r^2 = 0.91$.

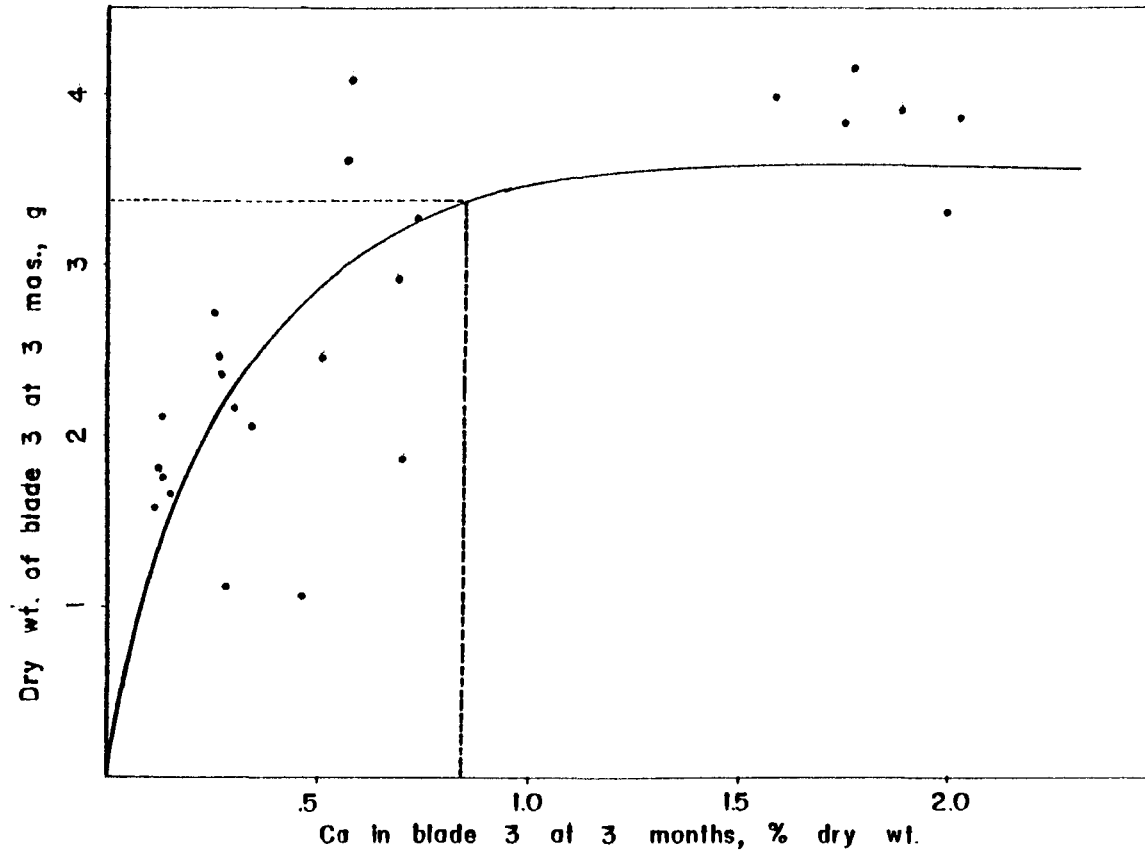


Figure 3. Relationship between the Ca concentration and dry weight of the third blade of taro at 3 months. Equation for the curve is $y = 3.59(1 - \exp(-3.30 * x))$, $r^2 = 0.50$. Critical level of the third blade Ca at 3 months is 0.8% Ca on a dry weight basis, which is the tissue Ca value found at 95% of maximum leaf dry matter.

in plants of such size that the nutrient in the crocks became depleted. To alleviate this problem, Hoagland's solution was raised to one-fifth strength after 3 months of growth. The 4 Ca levels were also increased to 2, 4, 8, and 40 ppm (Table 1). For the duration of the fourth month, both N treatments were 42 ppm, and then at the beginning of the fifth month, the higher N treatment was increased to 84 ppm N in solution (Table 1). Leaf samples were taken at the end of the fifth month. Due to changes in N treatments, the blade dry weights and nutrient concentrations at the higher N level were probably in a state of flux, making it difficult to draw conclusions about N effects.

Calcium deficiency symptoms were observed on plants grown at the 2 and 4 ppm levels of Ca even though the absolute amounts of solution Ca had doubled. This effect suggests that absolute amounts of solution Ca levels are not as important as the relative proportion of Ca ions to total cations, in determining solution Ca requirements.

The plants increased greatly in size between 3 and 5 months of growth. The blades doubled in dry weight while petioles tripled or even quadrupled in dry weight from 3 to 5 months (Tables 2, 4). Plants grown with the higher N levels no longer appeared greatly stunted relative to the lower N levels, and Ca deficiency symptoms appeared comparable at both N levels.

The higher N treatment compared to the lower N treatment resulted in significant increases in leaf Ca concentration and blade N concentration (Table 4). The decreased leaf Ca concentration with the lower N treatment might be partly due to dilution effects caused by the increased plant dry weights at the lower N treatment relative

Table 4. Effects of solution N and Ca on the dry weight and nutrient content of the third blade of taro at 5 months.

Variable	Main Effects						Interaction Effect ^{1,2}							
	N, ppm		Ca, ppm ¹				N, ppm							
	21	42	2	4	8	40	21		42					
							2	4	8	40	2	4	8	40
Dry wt., g														
Blade	6.1*	4.9	3.4y	3.8y	5.6x	8.8w	3.8d	4.9c	6.6b	9.2a	3.3d	2.7d	4.5cd	8.4a
Petiole	6.3*	5.1	2.8y	3.6y	5.7x	10.4w	2.6e	4.3d	7.1c	11.1a	3.0de	2.7e	4.4d	9.6b
Ca, % dry wt.														
Blade	0.67*	1.14	0.35y	0.42y	0.68x	2.04w								
Petiole	0.29*	0.43	0.19y	0.23xy	0.27x	0.70w								
N, % dry wt.														
Blade	4.3*	4.9	4.7w	4.8w	4.7w	4.1x								
Petiole	2.2	2.4	4.0w	2.7x	1.7y	0.9z	4.3a	2.5c	1.1d	0.7d	3.6ab	2.9bc	2.4c	1.0d
Total N, mg														
Blade	256.0	227.0	162.0c	182.0c	260.0b	362.0a	142.0d	223.0c	295.0b	363.0a	182.0cd	140.0d	225.0c	360.0a
Petiole	139.0	122.0	107.0w	89.0w	92.0w	88.0w								
K, % dry wt.														
Blade	3.9	3.8	4.3w	4.3w	3.8w	3.0x								
Petiole	4.4	4.1	7.2w	5.0x	3.4y	1.7z	8.0a	4.9c	2.8d	1.8de	6.4b	5.1c	4.0c	1.6e
Total K, mg														
Blade	232.0*	165.0	145.0c	176.0bc	212.0b	263.0a	134.0d	245.0ab	248.0ab	302.0a	156.0cd	106.0d	175.0bcd	224.0bc
Petiole	277.0*	209.0	196.0w	166.0w	186.0w	175.0w	206.0a	206.0a	196.0a	198.0a	186.0ab	126.0c	176.0ab	151.0bc
Mg, % dry wt.														
Blade	0.43	0.47	0.45x	0.60w	0.50wx	0.26y								
Petiole	0.24	0.32	0.30x	0.36w	0.30x	0.17y	0.23de	0.31bc	0.25cd	0.16e	0.37ab	0.43a	0.36ab	0.17e

* Means for the N main effect are significantly different at the 95% probability level as determined by ANOVA.

¹ Means in a row followed by the same letter are not significantly different at the 95% probability level as determined by Duncan's multiple range test.

² Means for all treatments are included only if the interaction effect is significant at the 95% probability level as determined by ANOVA.

to the higher N treatment.

As solution Ca increased leaf dry weight increased significantly in a curvilinear fashion (Figure 4). Increasing levels of solution Ca also resulted in significant increases in leaf Ca concentration, and significant decreases in leaf N and K concentrations (Table 4). The decreases in leaf N and K concentrations were probably due to dilution effects since total N and K in the petiole were not significantly different at the 4 solution Ca levels and total N and K in the blade increased significantly with increasing Ca in solution (Table 4).

Since the higher N treatment had depressed leaf growth, obscuring the Mitscherlich type of growth response to Ca, particularly at the lower Ca levels, only data from the lower N treatment were fitted by the growth equation (Figure 5). The critical Ca concentration in the third blade at 5 months, the blade Ca value at 95% of maximum blade dry weight, was estimated to be 0.7% on a dry weight basis (Figure 5). This estimate of the critical blade Ca at 5 months was very similar to the estimate at 3 months which was 0.8% Ca on a dry weight basis (Figure 3).

Harvest at 6 Months

During months 4 to 6, one-fifth strength Hoagland's solution was used and the four levels of solution Ca were 2, 4, 8, and 40 ppm (Table 1). The two levels of solution N during months 5 to 6 were 42 and 84 ppm (Table 1). Blade dry weights at the 6 month harvest were the same as at the 5 month sampling (Tables 4, 5). Petiole dry weights increased about 1.5 times over the same time period (Tables 4, 5), perhaps due to shading by neighboring plants.

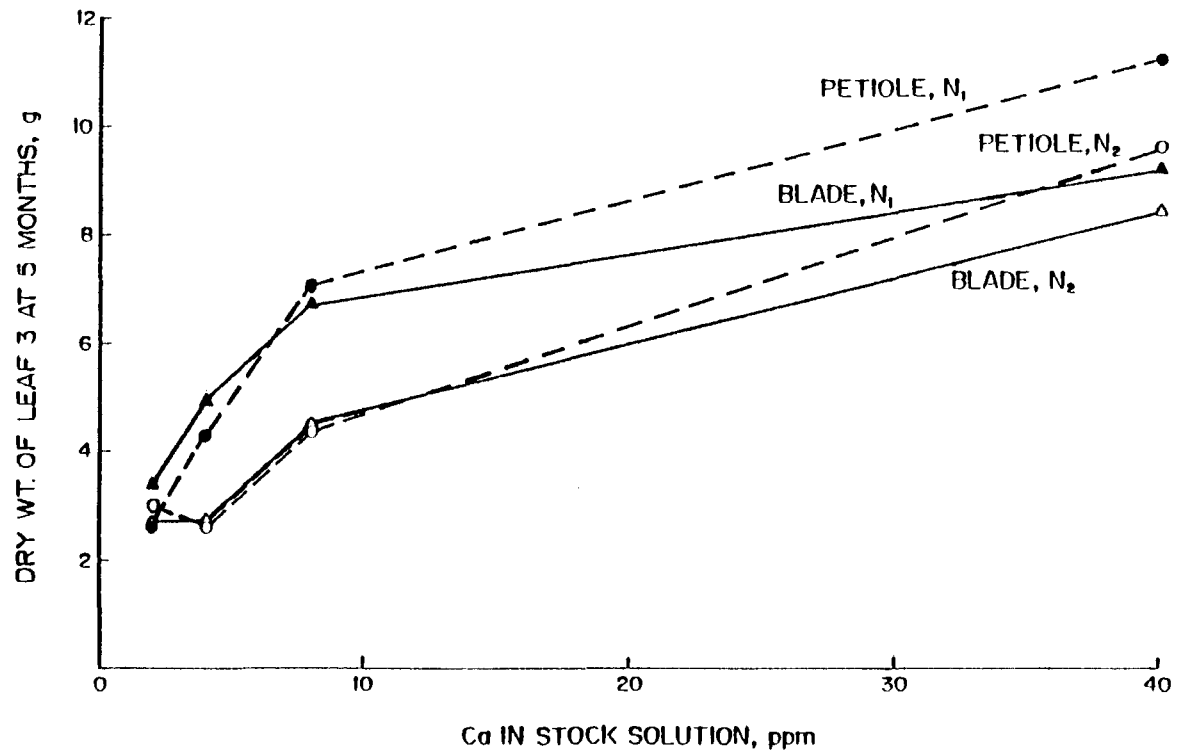


Figure 4. Changes in dry weight of the third leaf of taro at 5 months with changes in Ca levels in solution.

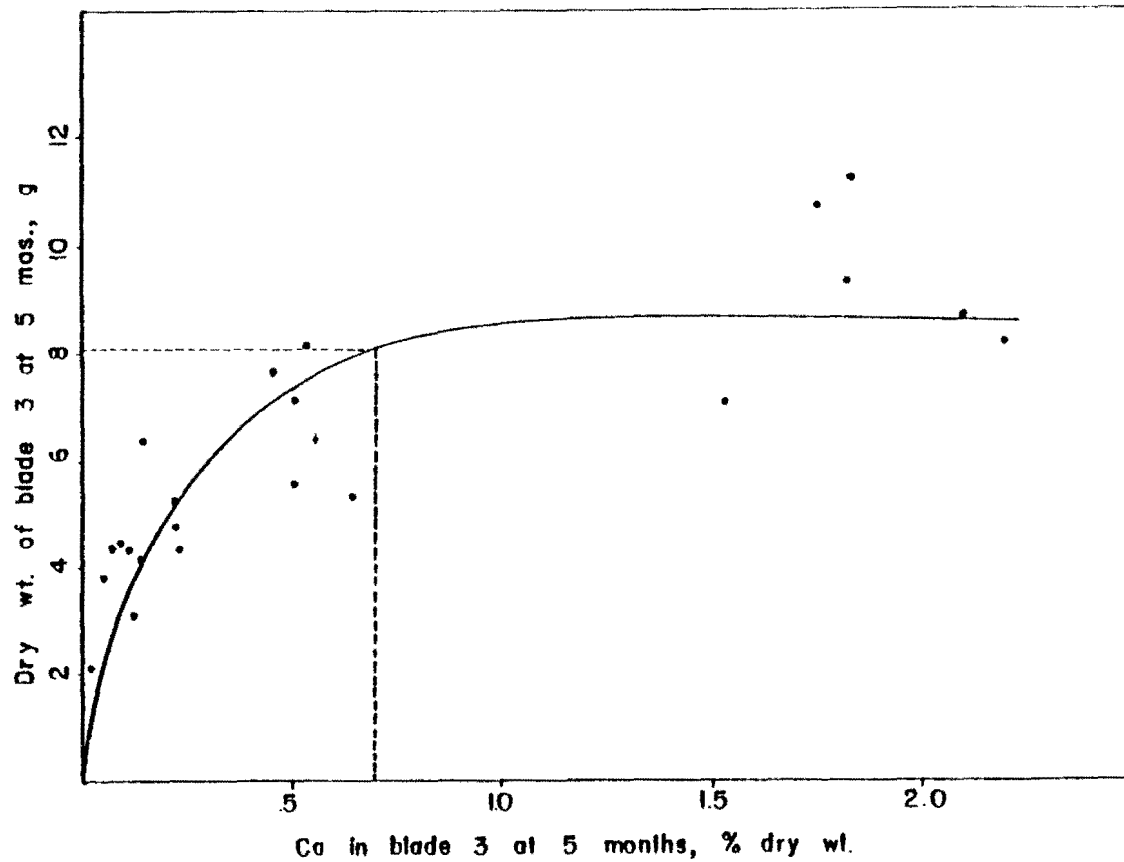


Figure 5. Relationship between the Ca concentration and the dry weight of the third blade of taro at 5 months. Equation for the curve is $y = 8.47(1 - \exp(-4.45 * x))$, $r^2 = 0.65$. The critical level of the third blade Ca at 5 months is 0.7% Ca on a dry weight basis, which is the tissue Ca level at 95% of maximum leaf dry weight.

6hr

Calcium deficiency symptoms were observed on plants grown with the 2 lower Ca treatments. The leaf blade was chlorotic or necrotic interveinally and the plants were stunted.

Root, Corm, Cormel and Leaf Dry Weights at 6 Months

No significant N effects were found on dry weight of roots, corm, or cormels after 6 months of growth (Table 5). Increasing solution Ca resulted in significant increases in the dry weights of roots, corms, and cormels (Table 5, Figure 6). Dry weight of the roots reached a maximum near 8 ppm Ca in solution with little increase in weight at 40 ppm calcium (Figure 6).

Significant N by Ca interactions existed for dry weights of roots, corms, and cormels (Table 5, Figure 7). Dry weight of the roots increased 7 times from 2 to 8 ppm Ca in solution at the lower N treatment, whereas at the higher N treatment, root dry weight increased only 2 times over the same solution Ca range (Table 5). Similar trends can be found for dry weights of the corms and the cormels (Table 5).

The higher N treatment seemed to interfere with a Mitscherlich type of growth response to solution Ca levels. From planting to 3 months, the higher N treatment had 5 times the level of N as the lower N treatment. Leaf dry weight at 3 months significantly decreased with increased solution N probably as a result of the decreased leaf Ca content with increased solution nitrogen (Table 2).

At the 6 month harvest, no significant effect of N was found on dry weight or Ca concentration of leaves 1 to 3, except for blade 1 which had a significantly greater Ca concentration at the lower N

Table 5. Effects of solution Ca and N on taro dry weight and tissue N and Ca concentration at 6 months.

Variable	Main Effects						Interaction Effect ^{1,2}											
	N, ppm		Ca, ppm ¹				N, ppm											
	21	42	2	42			Ca, ppm											
				4	8	40	2	4	8	40	2	84						
4	8	40	4	8	40													
Dry wt., g																		
Blade 1	4.2	3.8	2.4y	3.3xy	3.9x	6.2w												
Petiole 1	9.1	10.0	5.3y	7.1y	8.8w	17.2w												
Blade 2	5.4	4.9	3.8x	3.7x	4.8x	8.1w												
Petiole 2	9.0	7.7	5.0y	5.5xy	8.0x	14.9w	3.3d	6.4cd	10.2b	16.0a	6.6c	4.6cd	5.8cd	13.8a				
Blade 3	5.4	4.8	3.8x	3.8x	4.9x	8.0w												
Petiole 3	8.1	7.6	4.5x	5.0x	7.4x	14.5w												
Blade 4	5.6	4.7	3.4x	3.6x	5.1x	8.4w												
Petiole 4	9.2*	7.3	4.5y	5.1y	8.2x	15.5w												
Roots	33.0	21.0	9.0y	23.0x	36.0w	39.0w	7.0e	28.0cd	52.0a	44.0ab	11.0e	18.0de	21.0cde	33.0bc				
Corm	95.0	62.0	21.0y	45.0y	88.0x	158.0w	10.0d	59.0c	123.0b	187.0a	33.0cd	31.0cd	54.0c	130.0b				
Cormels	63.0	65.0	15.0y	58.0x	68.0x	117.0w	5.0e	57.0cd	86.0bc	105.0ab	25.0de	58.0cd	50.0cd	130.0a				
Ca, % dry wt.																		
Blade 1	0.47*	0.35	0.11y	0.12y	0.26x	1.15w												
Petiole 1	0.22	0.19	0.06y	0.09xy	0.16x	0.51w												
Blade 2	0.58	0.58	0.13y	0.22xy	0.40x	1.55w												
Petiole 2	0.22	0.20	0.11y	0.12xy	0.16x	0.44w												
Blade 3	0.70	0.76	0.19y	0.34xy	0.60x	1.78w												
Petiole 3	0.26	0.25	0.12y	0.16y	0.22x	0.52w												
Blade 4	0.94*	1.18	0.38y	0.49y	0.80x	2.33w												
Petiole 4	0.28*	0.34	0.19y	0.21y	0.25x	0.60w	0.12e	0.19d	0.23c	0.59a	0.24c	0.24c	0.28b	0.62a				
Roots	0.48	0.49	0.24y	0.29y	0.40x	1.00w												
Corm	0.11	0.11	0.07xy	0.06y	0.08x	0.23w	0.10c	0.07de	0.08cd	0.21b	0.06e	0.07de	0.08cd	0.23a				
Cormels	0.13*	0.11	0.08x	0.06x	0.08x	0.27w												
N, % dry wt.																		
Blade 1	5.5	5.8	6.2w	5.6w	5.6w	5.2w												
Petiole 1	3.1	3.6	3.8w	3.7w	3.3w	2.5x												
Blade 2	4.7*	5.2	5.0wx	5.4w	4.9wx	4.7x												
Petiole 2	2.1*	2.6	3.9w	2.5x	1.8y	1.1z	4.4a	2.0de	1.1ef	0.8f	3.5ab	3.1bc	2.5cd	1.3ef				
Blade 3	4.1*	5.2	4.7w	4.9w	4.8w	4.2x	3.7b	4.4b	4.3b	4.0b	5.7a	5.4a	5.3a	4.5b				
Petiole 3	2.2*	2.7	4.2w	2.6x	2.0x	1.0y	4.8a	2.2cd	1.0e	0.7e	3.6b	3.0bc	3.0bc	1.2de				
Blade 4	4.1*	5.0	4.8w	4.8w	4.7w	4.0x												
Petiole 4	2.0*	2.7	4.3w	2.4x	1.8x	0.8y												
Roots	2.8*	3.6	3.9w	3.3x	3.1x	2.6y	3.9a	2.9b	2.3c	2.1c	3.9a	3.7a	3.9a	3.1b				
Corm	2.1	2.1	3.8w	2.0x	1.6y	1.0z	4.9a	1.7c	1.0d	0.8d	2.6b	2.4b	2.2b	1.3cd				
Cormels	2.5	2.2	4.5w	2.2x	1.7xy	1.1y	6.1a	1.9bcd	1.2d	0.9d	2.8b	2.6b	2.2bc	1.3cd				

* Means for the N main effect are significantly different at the 95% probability level as determined by ANOVA.

¹ Means in a row followed by the same letter are not significantly different at the 95% probability level as determined by Duncan's multiple range test.

² Means for all treatments are included only if the interaction effect is significant at the 95% probability level as determined by ANOVA.

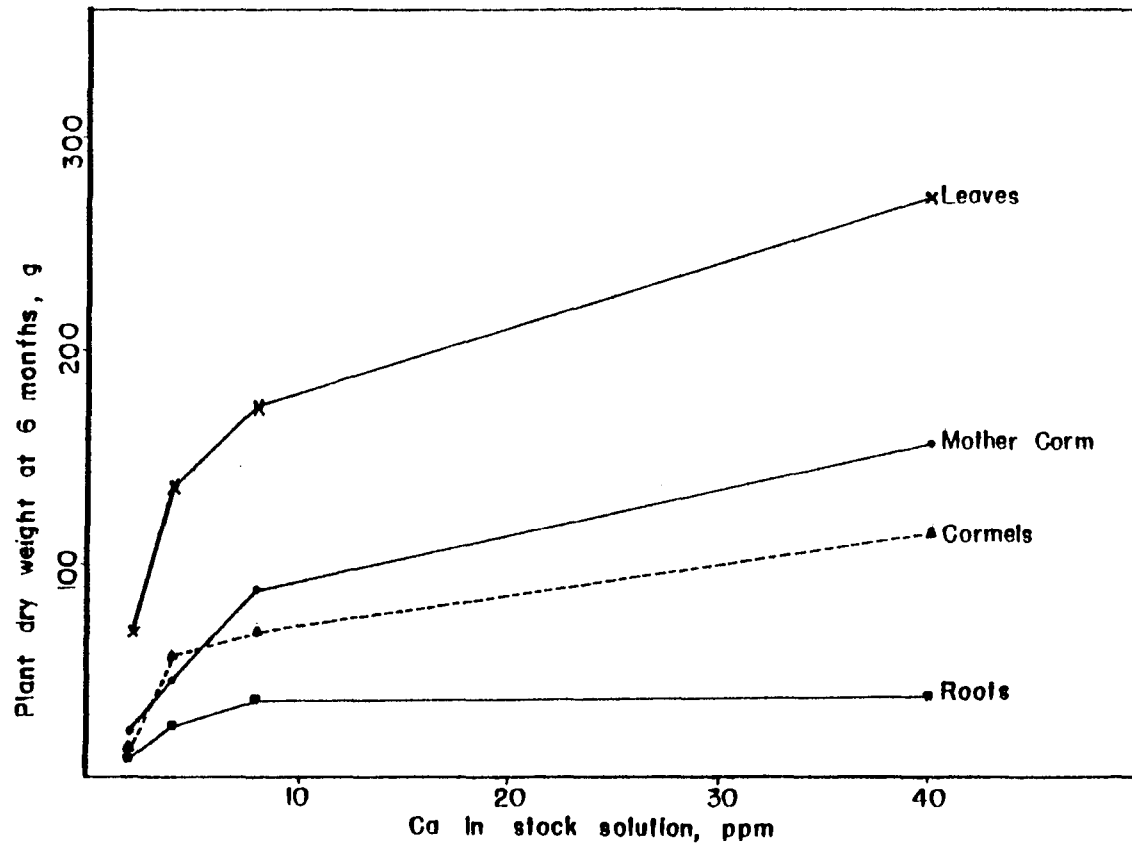


Figure 6. Changes in the dry weight of taro averaged over the two N treatments at 6 months with increasing Ca in solution.

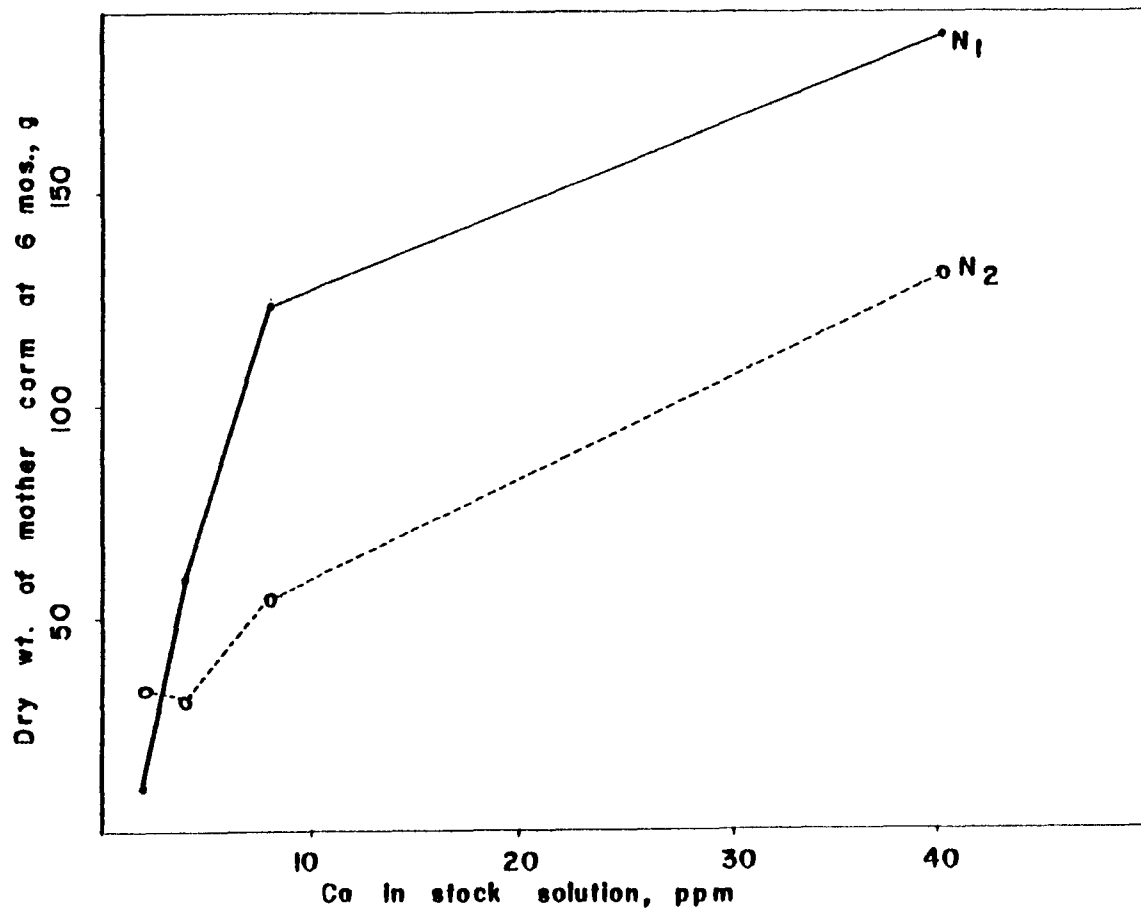


Figure 7. Changes in the dry weight of the taro corm at 6 months with increasing Ca in solution.

treatment relative to the higher N treatment (Table 5). However, the greater Ca concentration in blade 1 was due to one exceptionally high blade Ca value and so may be discounted.

The leaves of taro are short-lived plant organs which reflect environmental conditions and nutrient status over only about a month. The leaves produced during the fifth month did not show the effects of the early extremely high solution N levels and no significant N effect on dry weight and Ca concentration of leaves 1 to 3 was found (Table 5).

However, the corms and cormels are much longer-lived plant organs which reflect environmental conditions and nutrient status over the full 6 months as does the root mass. The cumulative effect of the higher N treatment over 6 months was to depress the growth response of taro roots, corms, and cormels to increased solution Ca at the 3 lowest Ca levels. One explanation for this is the high NH_4 ion levels particularly during the first 3 months of growth were toxic and a certain threshold level of solution Ca was necessary to overcome the toxic ion effects before growth response to increased solution Ca could occur. Calcium is known to stabilize membranes against such stress factors as toxic ion levels (Christiansen and Foy, 1979). Since roots, corms, and cormels are longer-lived plant organs, the effects of toxic NH_4 ion levels during the first 3 months of growth should still be evident at 6 months.

Total Yield, Corm Dry Matter, and Starch Content at 5 Months

There was no significant effect of N on corm starch content, corm and cormel dry matter, or on total yield (fresh weight of the

corm plus cormels) after 6 months of growth (Table 6). Corm starch content, corm dry matter and total yield increased with increasing Ca in solution, however a significant N by Ca interaction existed because the increase with increasing solution Ca differed at the two N treatments (Table 6). As with the dry weights of roots, corms, and cormels, the higher N treatment appeared to interfere with a Mitscherlich type of growth response of corm dry matter, corm starch and total yield to solution Ca levels.

Total yield was depressed at the two lower solution Ca levels because the available Ca was insufficient for growth. The N treatments had no significant effect on total yield at these solution Ca levels (Table 6), probably because yields were depressed by the inadequate Ca levels.

When solution Ca was increased from 2 to 8 ppm, total yield increased 6 times at the lower N treatment and about 70% of the maximum yield was reached (Table 6), indicating that the Ca level was almost adequate for optimum growth. However, at the higher N treatment, when the solution Ca was increased from 2 to 8 ppm, total yield increased 1.5 times and only 40% of maximum yield (Table 6) was attained, suggesting that the plant requirement for Ca in solution was greater at the higher N treatment compared to the lower N treatment.

Total yield of taro grown at the 8 ppm solution Ca level was severely reduced by the higher N treatment compared to the lower N treatment (Table 6). Total yield, corm dry matter, and corm starch content at the higher N treatment and 8 ppm Ca in solution were all significantly less than at the lower N treatment (Table 6).

At the highest level of Ca in solution (40 ppm), Ca was adequate

Table 6. Effects of solution Ca and N on taro corm starch, corm dry matter, total yield, and tissue K and Mg concentration at 6 months.

Variable	Main Effects						Interaction Effect ^{1,2}							
	N, ppm		Ca, ppm ¹				N, ppm							
	21	42	2	42			84							
				Ca, ppm			2		4		8		40	
21	42	2	4	8	40	2	4	8	40	2	4	8	40	
Starch														
Corm	40	41	30y	40x	44w	46w	23e	41cd	47ab	48a	38d	39cd	40cd	43bc
Fresh Wt.,g														
Corm + Cormels	669	572	225y	500x	650x	1105w	141e	551c	842b	1141a	310de	449cd	460bc	1069a
Dry Matter, %														
Corm	21	21	14y	20x	24w	25w	10e	21bcd	26a	26a	18d	20cd	22bc	24ab
Cormels	20	21	15y	19x	23w	24w								
K, % dry wt.														
Blade 1	4.4*	3.8	4.7w	3.9x	4.1x	3.8x								
Petiole 1	5.0	5.0	6.6w	5.4x	4.4y	3.5z	7.5a	5.1b	3.8c	3.6c	5.8b	5.7b	4.9b	3.4c
Blade 2	4.1	3.8	4.6w	4.1wx	3.9x	3.3y								
Petiole 2	4.1	4.0	6.9w	4.3x	3.3y	1.7z	8.3a	4.0b	2.4c	1.8c	5.5b	4.6b	4.1b	1.6c
Blade 3	3.8	4.0	4.5w	4.0x	4.0x	3.2y								
Petiole 3	4.2	4.1	7.2w	4.6x	3.4y	1.4z	8.7a	4.4b	2.2c	1.5c	5.7b	4.7b	4.5b	1.4c
Blade 4	4.0	3.9	5.1w	3.9x	3.9x	2.8y								
Petiole 4	4.0	4.1	7.1w	4.4x	3.2y	1.2z	8.5a	4.1c	2.1d	1.2e	6.2b	4.6c	4.3c	1.2e
Roots	2.9*	2.6	3.3w	3.2w	2.9x	1.5y								
Corms	1.0*	0.8	1.6w	0.9x	0.7xy	0.4y								
Cormels	1.1	0.9	1.5w	1.0x	0.9xy	0.6y	1.9a	1.0bc	0.8bc	0.7bc	1.1b	1.0b	0.9bc	0.5c
Mg, % dry wt.														
Blade 1	0.37	0.35	0.44w	0.42w	0.34x	0.24y								
Petiole 1	0.31	0.30	0.32x	0.38w	0.29x	0.23y								
Blade 2	0.43	0.42	0.43x	0.58w	0.41x	0.28y								
Petiole 2	0.26	0.22	0.26w	0.29w	0.22xy	0.19y	0.28ab	0.32a	0.22cd	0.21de	0.23bcd	0.26bc	0.22cd	0.17e
Blade 3	0.45	0.47	0.40x	0.66w	0.49x	0.28x								
Petiole 3	0.26	0.26	0.27w	0.30w	0.26w	0.20x								
Blade 4	0.55	0.63	0.55x	0.89w	0.58x	0.35y								
Petiole 4	0.25	0.32	0.30wx	0.34w	0.28xy	0.22y								
Roots	0.77	0.43	0.20z	0.42y	0.53x	1.24w	0.22e	0.58d	0.80c	1.47a	0.19e	0.25e	0.25e	1.02b
Corm	0.14	0.12	0.14w	0.13w	0.11x	0.13wx								
Cormels	0.15	0.13	0.16w	0.14x	0.13xy	0.12y	0.19a	0.15b	0.11c	0.14bc	0.14bc	0.13bc	0.12bc	0.12bc

* Means for the N main effect are significantly different at the 95% probability level as determined by ANOVA.

¹ Means in a row followed by the same letter are not significantly different at the 95% probability level as determined by Duncan's multiple range test.

² Means for all treatments are included only if the interaction effect is significant at the 95% probability level as determined by ANOVA.

for plant growth at both N treatments. No significant difference in total yield for the 2 N treatments was found (Table 6).

Thus, excessively high N fertilization may reduce taro yields particularly in taro plants with incipient Ca deficiency. Since taro farmers in Hawaii are reported to use high amount of N, P, and K fertilizers (Plucknett and de la Pena, 1971), it would perhaps be desirable to apply fertilizers that contain Ca as a filler or to lime fields where soil Ca levels are known to be on the borderline of being adequate, to prevent excess N from reducing total yield.

Dry weights of the third blade at 3 and 5 months sampling were significantly correlated with total yield at the 6 month harvest, and Pearson's correlation coefficients for 3 and 5 months were, respectively, 0.88 and 0.93. Thus, blade dry weights at 3 and 5 months sampling were a reasonably accurate measure of potential corm plus cormel yield at 6 months under the controlled conditions of this experiment. It is possible that sampling of the third blade at 3 or 5 months in the field may be used to predict total yield at 12 to 14 months harvest, provided no serious disruption in growth occurred between the time of sampling and harvest.

The corms produced at 6 months with solution culture were not mature. The maximum corm dry matter at 6 months in this study was only 25% for plants grown at the highest level of solution Ca, while Ezumah (1972) reported a range of 35 to 40% corm dry matter for mature taro harvested after 10 to 13 months of growth under flooded field conditions.

Plant Nutrient Status at 6 Months

Leaf Ca concentration increased significantly with increasing Ca in solution (Table 5) and with age of the leaf blades (Figure 8). Calcium concentration was averaged across the two N treatments for blades 1 to 4 because N by Ca interactions were not significant and the N effects on blades 1 and 4 were discounted. The significance of the N effect on blade 1 was probably an artifact due to one exceptionally high value, while the significance of the N effect on blade 4 was probably due to the change in treatment levels of solution N during months 4 and 5 (Table 1).

Leaf blade Ca increased linearly with increasing solution Ca and blade 4 had the greatest increase, blade 1 the least, and blades 2 and 3 were intermediate (Figure 8). The slopes calculated for blades 2 and 3 were not significantly different from each other (Figure 8). All other slopes were significantly different from each other.

Critical Ca concentration ranges were calculated for blades 1 to 4 at 6 months by fitting the Mitscherlich growth equation to corm yield and blade Ca data. The critical ranges of Ca on a dry weight basis were, respectively from blade 1 to blade 4, 0.70-0.85%, 0.85-1.00%, 0.90-1.05%, and 0.90-1.05% Ca (Figures 9-12). The critical Ca ranges were taken to be the blade Ca values at 85-90% of maximum yield. This range of 85 to 90% of maximum yield was chosen rather than 95% of maximum yield, because a large increase in blade Ca was associated with a small yield increase from 90 to 95% of maximum.

The critical third blade Ca values on a dry weight basis were

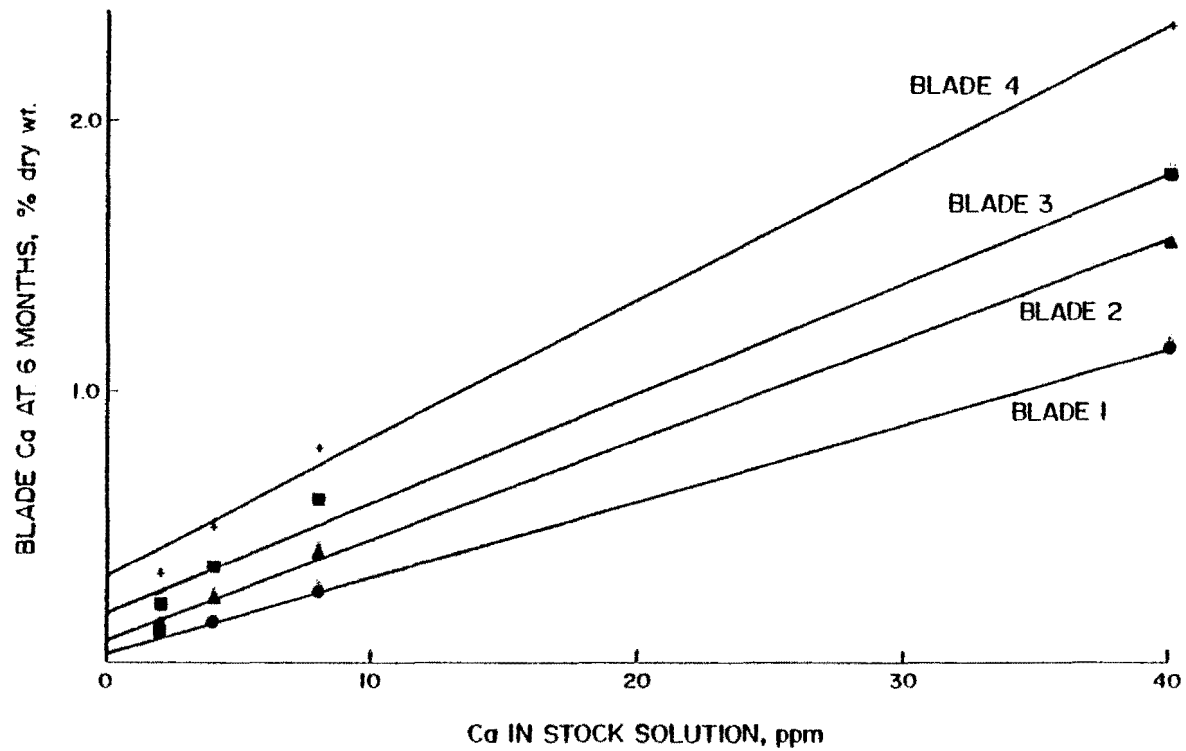


Figure 8. Relationship between tissue Ca concentration of taro leaf blades, averaged over the two N treatments at 6 months, and solution Ca concentration. Equations for the lines are, respectively, blade 1, $y = 0.033 + 0.28(x)$, $r^2 = 0.91$; blade 2 $y = 0.19 + 0.37(x)$, $r^2 = 0.94$; blade 3, $y = 0.32 + 0.40(x)$, $r^2 = 0.86$; and blade 4, $y = 0.78 + 0.51(x)$, $r^2 = 0.89$.

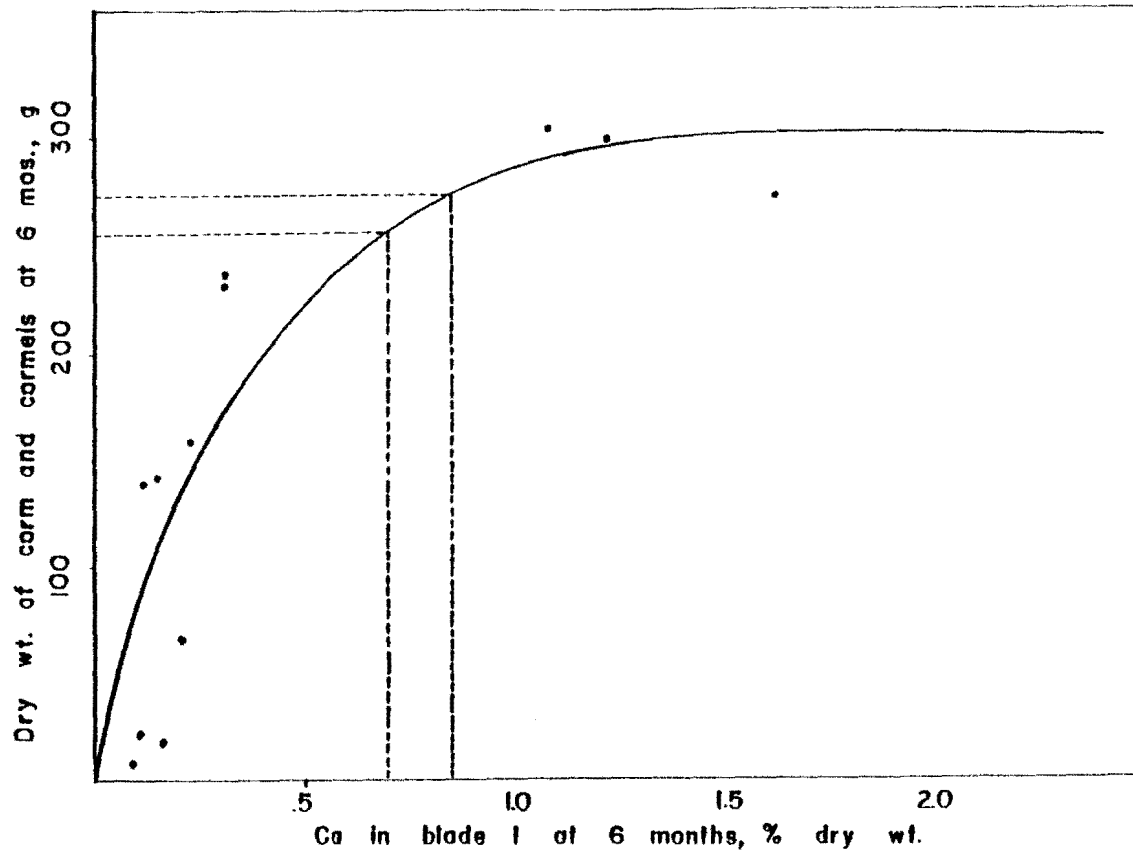


Figure 9. Relationship between Ca concentration in the first blade of taro at 6 months and dry weight of the corm plus cormels. The equation for the curve is $y = 307(1 - \exp(-2.72 * x))$, $r^2 = 0.76$. The critical range of Ca is 0.70-0.85% on a dry weight basis and this is the tissue Ca range at 85-90% of maximum yield.

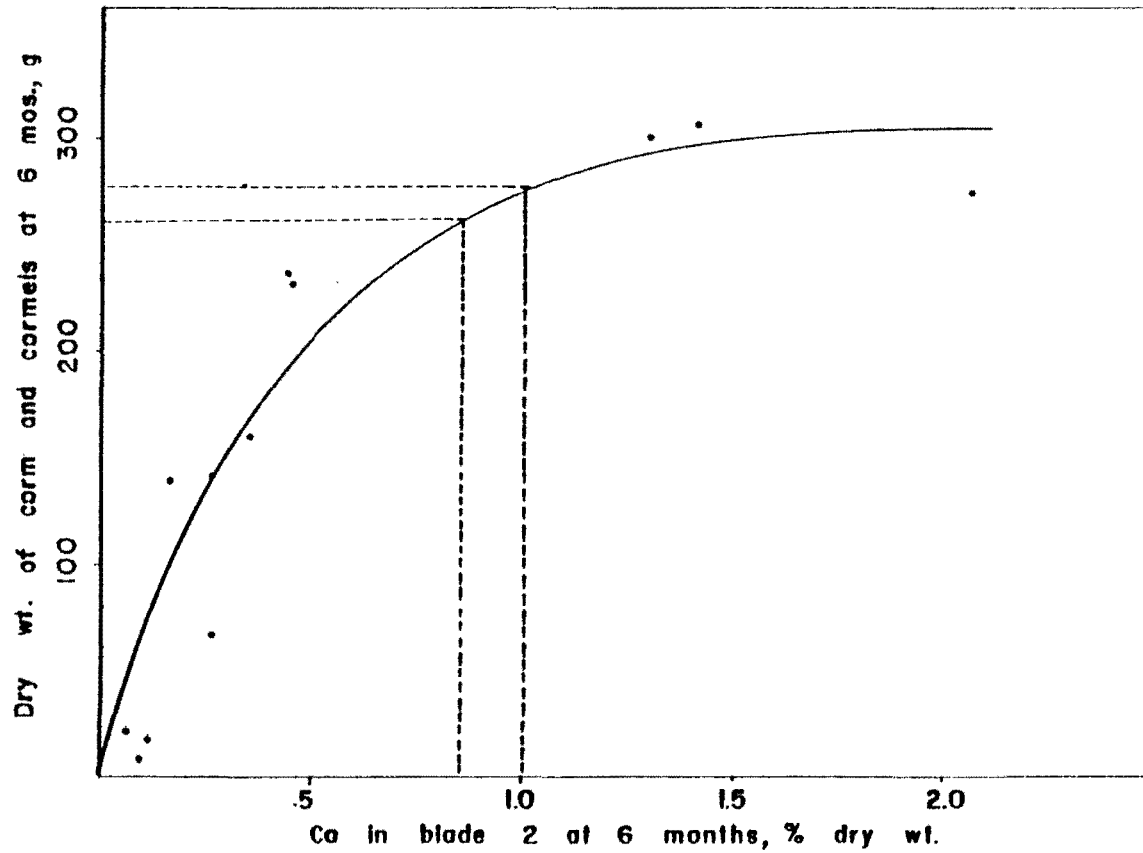


Figure 10. Relationship between Ca concentration in the second blade at 6 months and the dry weight of the corm plus cormels. The equation for the curve is $y = 307(1 - \exp(-2.24 * x))$, $r^2 = 0.88$. The critical range of Ca is 0.85-1.00% on a dry weight basis and this is the tissue Ca range at 85-90% of maximum yield.

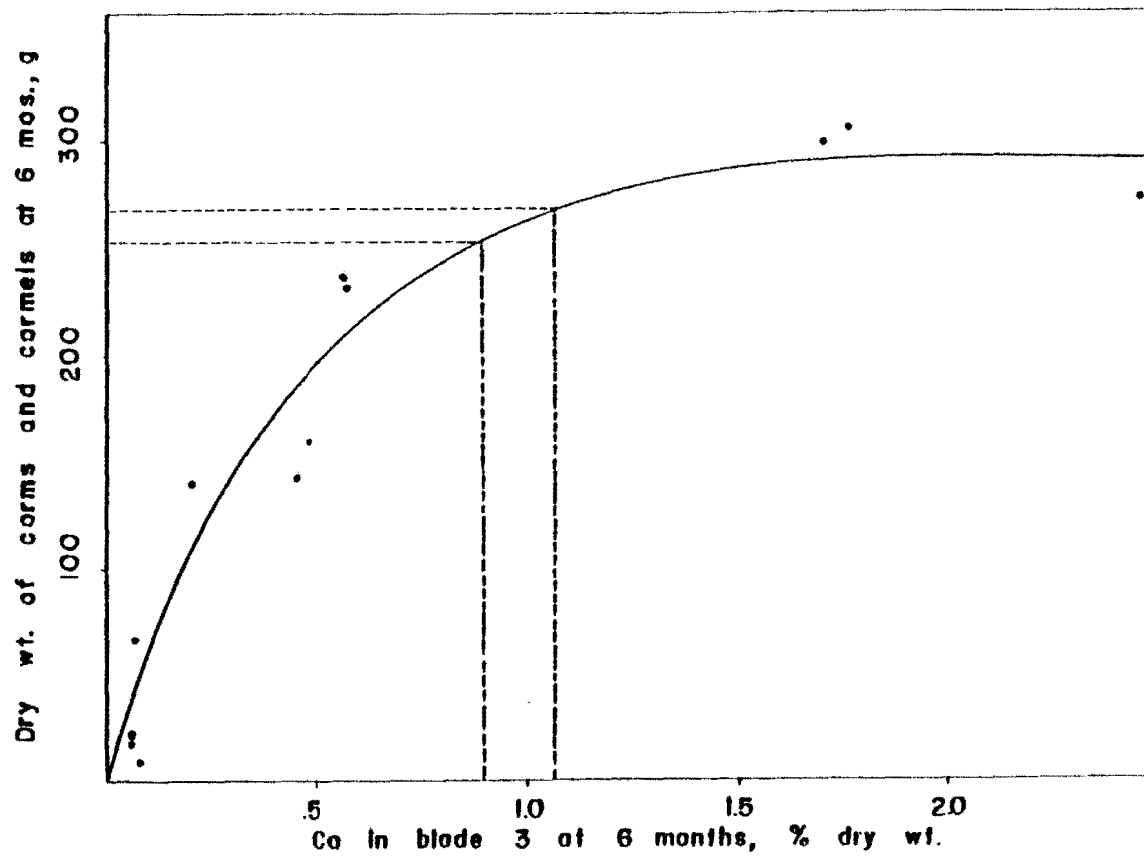


Figure 11. Relationship between Ca concentration in the third blade of taro at 6 months and dry weight of corm plus cormels. The equation for the curve is $y = 298(1 - \exp(-2.10 * x))$, $r^2 = 0.94$. The critical range of Ca is 0.90-1.05% a on a dry weight basis and this is the tissue Ca range at 85-90% of maximum yield.

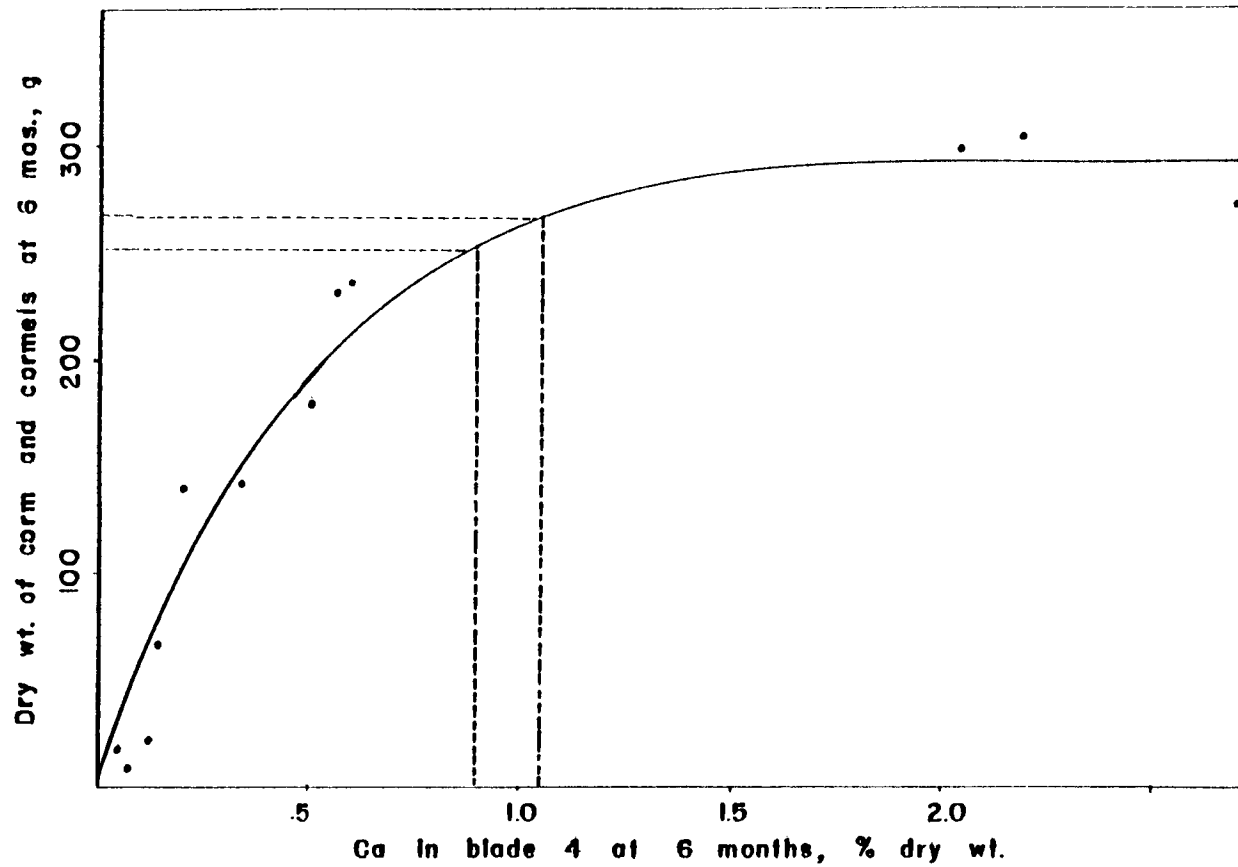


Figure 12. Relationship between Ca concentration in the fourth blade of taro at 6 months and dry weight of the corm plus cormels. The equation for the curve is $y = 297(1 - \exp(-2.10 * x))$, $r^2 = 0.94$. The critical range of Ca is 0.90-1.05% on a dry weight basis and this is the tissue Ca range at 85-90% of maximum yield.

estimated to be 0.8% and 0.7% Ca at 3 and 5 months, respectively. These critical third blade Ca values were not greatly different from the values estimated at 6 months. The relatively constant critical third blade Ca range of 0.7-1.0% from 3 to 6 months results in increased confidence for using the values found in this study to define the Ca nutrient status of taro. However, field work should be carried out to confirm whether the findings of this study can be extrapolated to the field.

Third blade Ca values for field-grown taro have been reported in studies not directly dealing with Ca nutrition. de la Pena and Plucknett (1972) reported a range of third blade Ca concentration on a dry weight basis from 1.2 to 1.5% during 3 to 6 months of growth for the same 'Lehua' cultivar grown during this experiment. Ezumah (1972) found third blade Ca concentrations on a dry weight basis which ranged from 0.6 to 0.8% at 2 months for the same cultivar. A third blade critical Ca range on a dry weight basis of 0.7-1.0% would fall between the range of values reported for the 'Lehua' cultivar in the literature.

The best tissue to sample for plant Ca levels is the third blade, for the following reasons. First, the third blade had a rate of increase in tissue Ca concentration with increasing solution Ca that was second only to the fourth blade (Figure 8). Also, considerable research has been carried out on tissue levels for N, P, and K in the third blade (de la Pena and Plucknett, 1969; Ezumah, 1972). Finally, sampling of the third blade from 3 to 6 months of growth in this experiment resulted in a fairly consistent estimate of the critical Ca concentration.

Blade N concentrations for blades 2 to 4 were significantly greater at the higher N treatment relative to the lower N treatment (Table 5). No significant effect of N was found on tissue K concentrations in blades 2 to 4 (Table 6). Blade N and K concentration decreased significantly with increasing solution Ca (Tables 5, 6) and this decrease was probably due to dilution effects, as discussed earlier for the leaf samples at 3 and 5 months.

No significant effect of N was found on tissue Mg concentrations in blades 1 to 4 (Table 6). Magnesium concentrations in blades 2 to 4 increased significantly with increasing solution Ca from 2 to 4 ppm and subsequently decreased at Ca levels of 8 and 40 ppm (Table 6). This increase in blade Mg concentrations with increasing solution Ca levels to 4 ppm might be explained by the fact that translocation of Ca and Mg is very similar (Ferguson and Clarkson, 1976). Movement of Ca in the xylem is thought to occur by a series of ion exchange reactions along negatively charged sites on the vessel walls and is reported to be promoted by the presence of other divalent cations (Hanger, 1979). It is therefore possible that the movement of Mg in the xylem is promoted by the presence of increased Ca ions up to a certain level.

Guava Seed Disease of Taro

Guava seed disease of taro is characterized by abundant, scattered corky tissues in the corm (Parris, 19741), which reduce yield because affected portions of the corm are discarded. The corky tissues are rings of periderm surrounding dead tissues (Hawaii Agr. Exp. Sta., 1938). There are no foliar diagnostic symptoms of this disease.

No guava seed disease was found with any solution level of Ca or N at 6 months. Since guava seed disease is thought to occur particularly during the maturation phase of taro which begins after 5 to 6 months growth, half of the 6 replicates were grown for 8 months to see if the incidence of guava seed disease increased with increased maturity.

Harvest at 8 Months

Following the harvest of 3 replicates at 6 months, Hoagland's solution was decreased from one-fifth to one-tenth strength to promote corm maturation. After 7 months, no N or K was added to the nutrient solution.

Dry weights of blades 2 and 3 decreased about 1.5 times from 6 to 8 months, while petiole dry weights remained fairly constant (Tables 5, 7). Fresh weight of the corm plus cormels almost doubled between 6 and 8 months (Tables 6, 7).

The quality of the corms harvested at 8 months was good. Dry matter had increased in the corms grown with the highest level of solution Ca from a maximum of 25% at 6 months to a maximum of 38% at 8 months (Tables 6, 7), probably due to an increase in starch content from 46% to 51% over the same time span (Tables 6, 7). Ezumah (1972) reported that corms grown under field conditions had a corm dry matter percentage of 35 at 7 months. The maximum dry matter of corms at 8 months in this study was comparable to values for field grown taro at the same age.

Ching (1969) determined starch contents for 'Lehua maoli' corms grown under dryland conditions and he obtained values of starch content

Table 7. Effects of solution N and Ca on taro corm specific gravity, corm starch, corm dry matter, total yield, plant dry weights and nutrient content at 8 months.

Variable	Main Effects						Interaction Effect ^{1,2}							
	N, ppm		Ca, ppm ¹				N, ppm							
	21	42	1	2			21				42			
				2	4	20	Ca, ppm		Ca, ppm		Ca, ppm			
1	2	4	20	1	2	4	20	1	2	4	20			
Starch, %														
Corm	45	45	38y	44x	48w	51w	34c	44b	52a	51a	43b	43b	44b	51a
Fresh Wt., g														
Corm + Cormels	1266	1071	408z	800y	1289x	2184w	343d	892c	1673b	2157a	505cd	662cd	904c	2212a
Dry matter, %														
Corm	28	29	21x	27x	28x	38w								
Specific gravity														
Corm	1.01	1.02	0.95w	1.01w	1.04w	1.05w								
Dry wt., g														
Corm	193.0*	130.0	40.0y	94.0xy	148.0x	370.0w								
Blade 2	4.3*	2.9	3.0x	3.2x	3.9x	5.1w								
Petiole 2	8.4	6.2	4.2y	4.8y	8.7x	12.0w								
Blade 3	4.3	2.9	2.2y	3.1xy	4.2wx	5.3w								
Petiole 3	9.0*	5.9	2.7y	5.2y	9.0x	13.6w								
Ca, % dry wt.														
Blade 2	0.33	0.25	0.06y	0.10y	0.19x	0.82w	0.06d	0.10d	0.21c	0.94a	0.05d	0.08d	0.18c	0.70b
Petiole 2	0.14	0.14	0.06z	0.11y	0.14x	0.25w								
Blade 3	0.40	0.46	0.35x	0.13x	0.22x	0.97w								
Petiole 3	0.20	0.24	0.26w	0.15x	0.16x	0.31w								
N, % dry wt.														
Blade 2	3.6*	4.2	3.4x	3.8x	3.8x	4.4w								
Petiole 2	1.1	1.1	2.5w	1.3x	0.9y	0.6z								
Blade 3	3.8	3.9	4.2w	3.8wx	3.7xy	3.5y								
Petiole 3	1.2	1.3	2.5w	1.4x	1.0xy	0.6y								
K, % dry wt.														
Blade 2	3.3	3.3	2.7w	3.2x	3.5xy	3.7y								
Petiole 2	3.2	3.2	5.5w	3.8x	2.2y	1.3z								
Blade 3	3.8*	3.4	4.2w	3.4x	3.4x	3.4x	4.2a	4.0ab	3.6bc	3.4bc	4.2a	2.7d	3.2cd	3.4c
Petiole 3	3.1*	3.8	6.9w	3.6x	2.1y	1.1z	5.9b	3.4d	2.0e	1.0f	8.3a	3.8c	2.1e	1.1f
Mg, % dry wt.														
Blade 2	0.36	0.35	0.36w	0.45w	0.36w	0.23x								
Petiole 2	0.21	0.20	0.31w	0.26w	0.18x	0.14x								
Blade 3	0.44	0.46	0.60w	0.52x	0.41x	0.19y								
Petiole 3	0.23	0.28	0.41w	0.26x	0.18y	0.15y								

* Means for the N main effect are significantly different at the 95% probability level as determined by ANOVA.

¹ Means in a row followed by the same letter are not significantly different at the 95% probability level as determined by Duncan's multiple range test.

² Means for all treatments are included only if the interaction effect is significant at the 95% probability level as determined by ANOVA.

as high as 62 to 73% between 5 to 11 months. Since the values of starch content are dependent on the method used, his results are probably not comparable with the values reported here.

Specific gravity for corms grown with the highest solution Ca level was determined to be 1.05 at 8 months (Table 7). Bowers et al. (1964) reported that a corm specific gravity of 1.05 to 1.15 was considered to be mature, and so the corms grown with the highest solution Ca level at 8 months were mature.

Total yield (fresh weight of the corm plus cormels) increased with increasing solution Ca, however a significant N by Ca interaction existed (Table 7). Total yield increased almost 5 times between the lowest and third lowest solution Ca levels at the lower N treatment, while at the higher N treatment, fresh weight only doubled over the same solution Ca range (Table 7). A similar N by Ca interaction was found for total yield at 6 months.

The important effect of N treatments on yield at 8 months was the significant decrease of the fresh weight of corm plus cormels at 4 ppm Ca at the higher N level compared to the lower N level. Fertilization with excess N in taro fields might seriously reduce total yield if fairly high amounts of soil solution Ca are not present. Calcium might prevent yield reductions through its stabilizing effect on cell membranes which would minimize the effects of such stress factors as toxic ion levels (Christiansen and Foy, 1979).

Starch content in the corms increased with increasing levels of solution Ca, however a significant N by Ca interaction existed (Table 7). Again, the higher level of N resulted in significantly reduced starch content at 4 ppm Ca in solution compared to the lower

N treatment.

Guava seed disease was not found to be induced by any of the solution levels of N or calcium. Thus, the disease is not directly caused by Ca deficiency, NH_4 toxicity, or N-induced Ca deficiency under the relatively sterile conditions of a solution culture experiment.

While no evidence of a direct relationship between guava seed disease and Ca was found in this study, an inadequate level of Ca or a short-term Ca deficiency caused by high levels of total soluble salts due to excess fertilization, could result in die-back of the taro roots. This root die-back could permit normally non-pathogenic organisms to enter the roots and cause the corking response in the corm that is typical of guava seed disease of taro. Thus, Ca may be an indirect cause of the disease.

Summary

Calcium deficiency symptoms on taro plants grown in nutrient solution culture were leaf blade interveinal chlorosis and necrosis, die-back of roots, drooping of the petioles, and overall stunting of the plants. Leaf blades tended to be cup-shaped and brittle, tearing readily under slight pressure.

For the first 3 months of growth, one-tenth strength Hoagland's solution was adjusted to 4 levels of Ca (1, 2, 4 and 20 ppm) and 2 levels of N (21 and 100 ppm). The increased level of solution N resulted in significant decreases in leaf dry weight and leaf Ca concentration at 3 months. Increasing levels of solution Ca resulted in significant increases in leaf dry weight and leaf Ca concentration. The rate of increase in leaf Ca with increasing levels of solution Ca

was significantly less at the higher N treatment compared with the lower N treatment at 3 months.

From the beginning of the fourth month, one-fifth strength Hoagland's solution was adjusted to 4 levels of Ca (2, 4, 8, and 40 ppm). The N treatments were both 42 ppm during the fourth month, and then at the beginning of the fifth month, the higher N treatment was increased to 84 ppm while the lower N treatment remained at 42 ppm until harvest at 6 months. The corms harvested after 6 months of growth were immature, containing a maximum of 25% dry matter as compared to 35% for mature field grown taro (Ezumah, 1972).

Increasing levels of solution Ca resulted in significant increases in total yield, dry matter content of the corm, and starch content of the corm at 6 months, however significant N by Ca interaction existed. The higher N treatment seemed to interfere with a Mitscherlich type of growth response of taro at 6 months to solution Ca levels. Total yield, dry matter content of the corm and starch content of the corm at 6 months was significantly less at the higher N treatment and 8 ppm Ca compared to the lower N treatment and 8 ppm calcium, and this effect was possibly due to NH_4 toxicity at the higher N treatment. At the other levels of solution Ca, no significant difference in total yield, corm dry matter content, and corm starch content was found between N treatments.

The best tissue to sample for plant Ca levels is the third blade. The critical Ca concentration range for the third blade between 3 to 6 months was estimated to be 0.7 to 1.0% Ca on a dry weight basis.

Corms that were harvested at 8 months were considered to be mature since the maximum corm specific gravity was 1.05 and corm dry matter was 38%. These corms grown in solution culture for 8 months were comparable to field grown taro.

Guava seed disease of taro was not induced by any solution level of Ca at either level of nitrogen. The disease was not directly caused by Ca deficiency, NH_4 toxicity, or N-induced Ca deficiency under the relatively sterile conditions of this solution culture experiment.

IV. STUDY OF THE EFFECTS OF CALCIUM DEFICIENCY
ON THE ULTRASTRUCTURE OF TARO (COLOCASIA ESCULENTA (L.) SCHOTT)
LEAF BLADES

Introduction

Symptoms of calcium deficiency on taro (Colocasia esculenta (L.) Schott) are interveinal chlorosis and necrosis of the leaf blade and uneven growth of the lamina resulting in an upward cupping of the leaf blade. The petioles lose their strength and droop with the weight of the leaf blades. Calcium deficiency also results in physiological die-back of the roots, and reduced corm size. Under extreme deficiency conditions, the growing point usually dies.

The purpose of this study was to examine the ultrastructural changes in taro leaf blade parenchyma cells grown under increasing levels of calcium stress. Experiments on tomato fruit tissue, vacuolated barley root cells, and green algae have shown that Ca deficiency results in a general degeneration of membranes in the cytoplasm (Bangerth, 1973; Marschner and Gunther, 1964; Nilshammar et al., 1972).

Marinos (1962) worked with shoot apices of barley and found that the breakdown of the nuclear envelope and the plasma and vacuolar membranes was the first sign of structural abnormalities due to Ca deficiency. What he termed "structure-less areas" appeared in the cell, followed by the disorganization of other membrane-bound structures such as mitochondria and Golgi apparatus. Plastids were more persistent, but eventually they also disintegrated.

Materials and Methods

Taro cv. 'Lehua maoli' was grown in continuously flowing nutrient solution culture as described previously in Chapter III for 5 months. Leaf blades were sampled from plants grown in the highest and lowest solution Ca levels and the lower N treatment.

The procedures for fixation, dehydration, and embedding were essentially those of Sakai and Hanson (1974) with the following modifications: (1) fixation was carried out at room temperature because the floating leaf sections in fixative were repeatedly pumped down in the vacuum chamber to enhance infiltration of the fixative; (2) bulk staining with 2% uranyl acetate was done after fixation for one minute; and (3) after embedding, sectioning was done with a glass knife. Hitachi Electron microscope, model HS-8-1, located at the St. John Plant Sciences Laboratory, was used for examination of the sectioned plant materials.

Results

Under calcium sufficient conditions, the plasma membrane and membrane-bound organelles, such as mitochondria and chloroplasts with well-defined grana and starch grains were clearly present in the cell (Figures 1, 2A). A slight plasmolysis of the cells was evident from the wrinkled appearance of the plasma membranes.

Under Ca deficient conditions, the cells were dramatically plasmolyzed with extreme separation of the plasma membrane and cytoplasm from the cell wall (Figures 3-5). In Figure 4, the chloroplasts appeared intact with starch grains and the grana were

distinguishable but less so than in Figures 1 and 2. The plasma membrane was present although severely plasmolyzed. Mitochondria were present in the cells although they were difficult to see since they stained lightly.

In other cells, the chloroplasts appeared to be in the process of disintegrating and starch granules were absent (Figures 2B, 4). No tonoplast was present and it appeared that the vacuolar contents had mixed with the cytoplasmic material (Figure 5A). Neither was it possible to distinguish the plasmalemma or mitochondria in these cells.

Finally, there appeared to be cells that were totally disorganized. The compartments of the cytoplasm had completely degenerated (Figure 5B).

Discussion

Sakai and Hanson's (1974) method of fixation resulted in slightly plasmolyzed cells when the taro plants were grown under Ca sufficient conditions and severely plasmolyzed cells under Ca deficient conditions, probably due to the fact that the fixative was hypertonic to the cells. Marinos (1962) noted that plasma membranes in barley cells under calcium deficient conditions were plasmolyzed although plasma membranes under calcium sufficient conditions were smooth in outline and normal.

Calcium deficiency resulted in some cells that were more readily plasmolyzed. One explanation is that the plasma membranes were more fragile under Ca deficiency since Morre and Bracker (1976) observed that Ca-treated plasma membranes were thicker than controls treated with water. A second explanation is that Ca deficient plasma

membranes were more permeable to water flow since Gary-bobo (1970) showed that the osmotic water flow of water across artificial phospholipid membranes decreased in the presence of calcium.

In other Ca deficient cells, no plasmalemma or tonoplast was found and membrane-bound organelles in the cytoplasm were in the process of degenerating (Figures 4, 5). Arnott (1970) hypothesized that membrane failure, caused by an increasing lack of available Ca was the major cause of cell disruption and death.

Unquestionably, Ca appears to have a role in maintaining membrane structure. The result of a lack of calcium in the taro leaf blade parenchyma cells was total breakdown of membrane systems in the cytoplasm.

Summary

Taro (Colocasia esculenta (L.) Schott) was grown in nutrient solution culture with varying levels of calcium for five months. Sections of the leaf blades were sampled to study the long-term effects of calcium deficiency on the ultrastructure of taro parenchyma cells. The disintegration of the plasmalemma, tonoplast, and all other membranous systems in the cytoplasm were observed in calcium deficient cells.

Explanation of Figures

Figure 1. Calcium sufficient taro leaf blade parenchyma cells.
Note the starch grains (S) and distinct grana (G) in the chloroplasts (C) and the well-defined mitochondria (M). The cells are slightly plasmolyzed as shown by the wrinkled appearance of the plasma membrane (PM). The nucleus (N) with the nuclear membrane (NM) can be seen in the lower cell as well as the endoplasmic reticulum (ER). (x 5600)

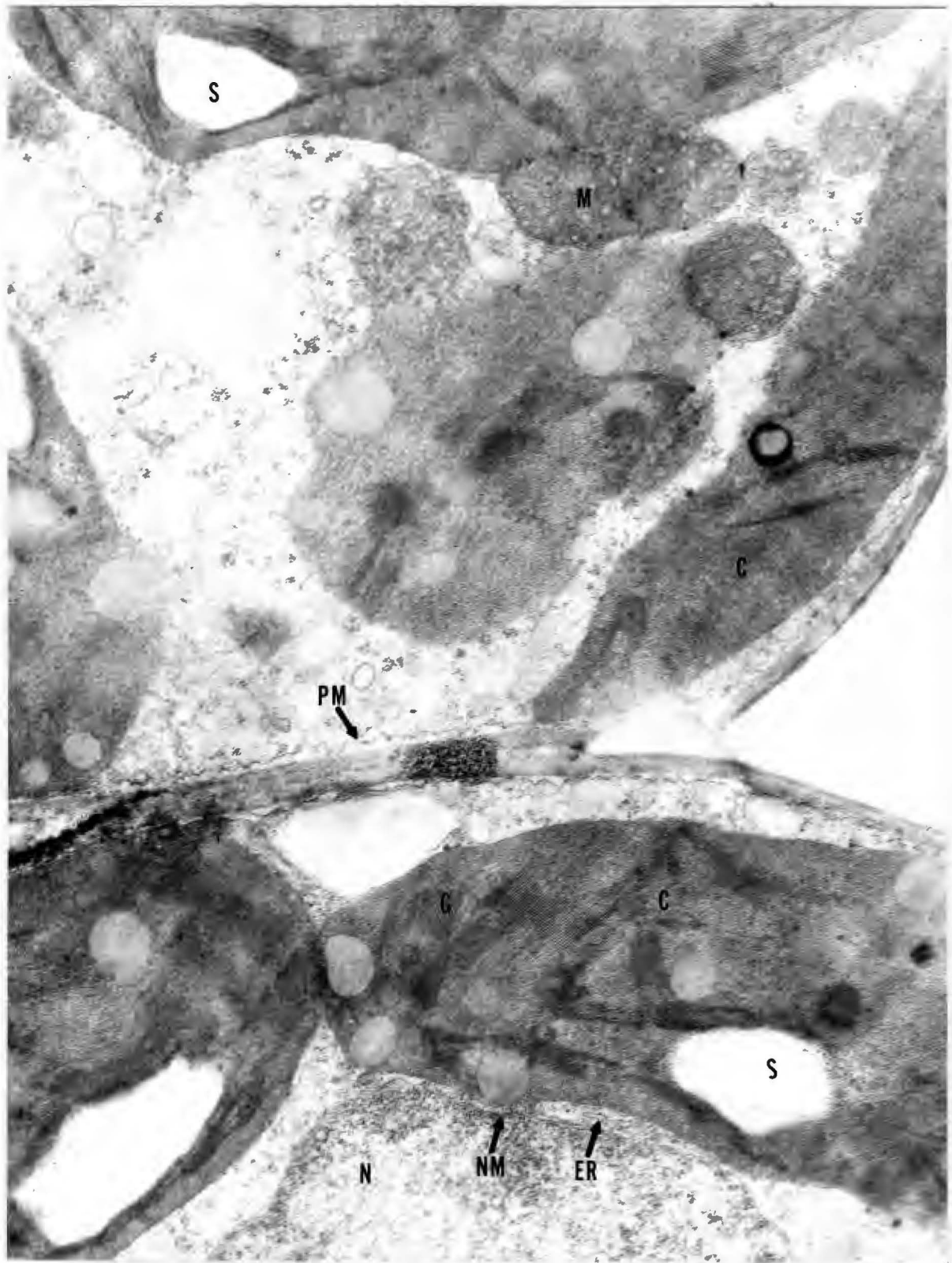


Figure 2A. Calcium sufficient taro leaf blade parenchyma cell. Note The vacuole (V), plasma membrane (PM), and distinct grana (G) and starch grains (S) in the chloroplasts (C). (x 5600)

Figure 2B. Calcium deficient taro leaf blade parenchyma cell. Arrows point to the extreme separation of the cytoplasm from the cell wall. The chloroplasts (C) appear to be disorganized. (x 7100)

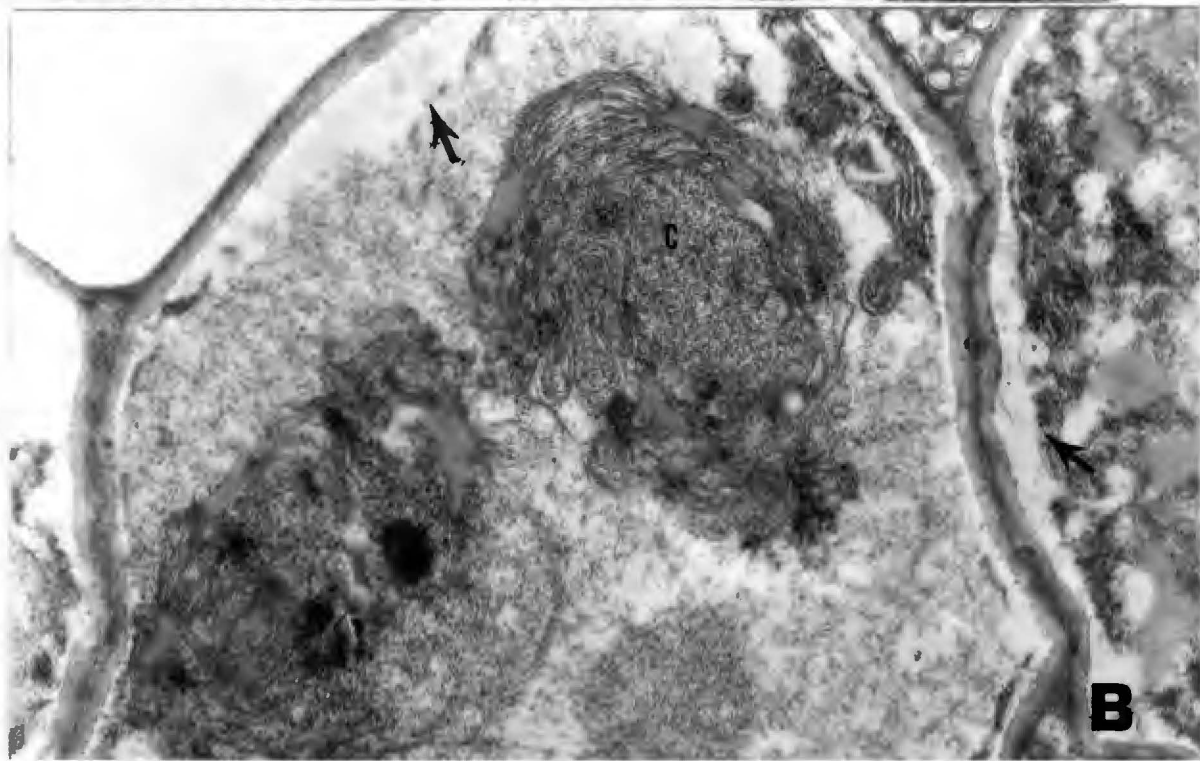
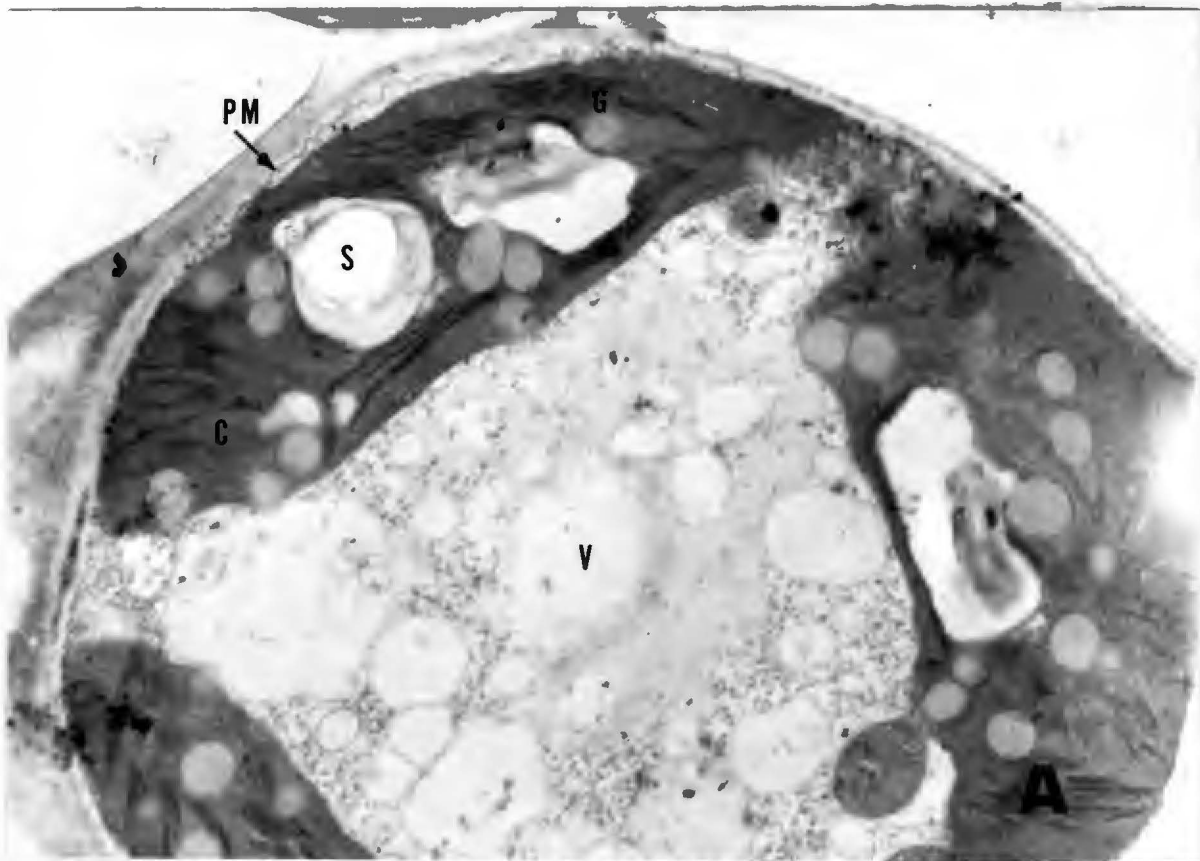


Figure 3. Calcium deficient taro leaf blade parenchyma cell. The cell is extremely plasmolyzed as shown by the severe separation of the plasma membrane (PM) from the cell wall. The tonoplast (T) is present, along with chloroplasts (C), starch grains (S), and grana (G). Indistinct mitochondria (M) can be seen in the lower and upper extreme cells.
(x 4000)



Figure 4. Calcium deficient taro leaf blade parenchyma cell. The chloroplasts (C) appear to be disintegrating and no starch grains are found in the chloroplasts. Arrows point to the extreme separation of the cytoplasm from the cell wall, and no plasma membrane or tonoplast is distinguishable. (x 4000).

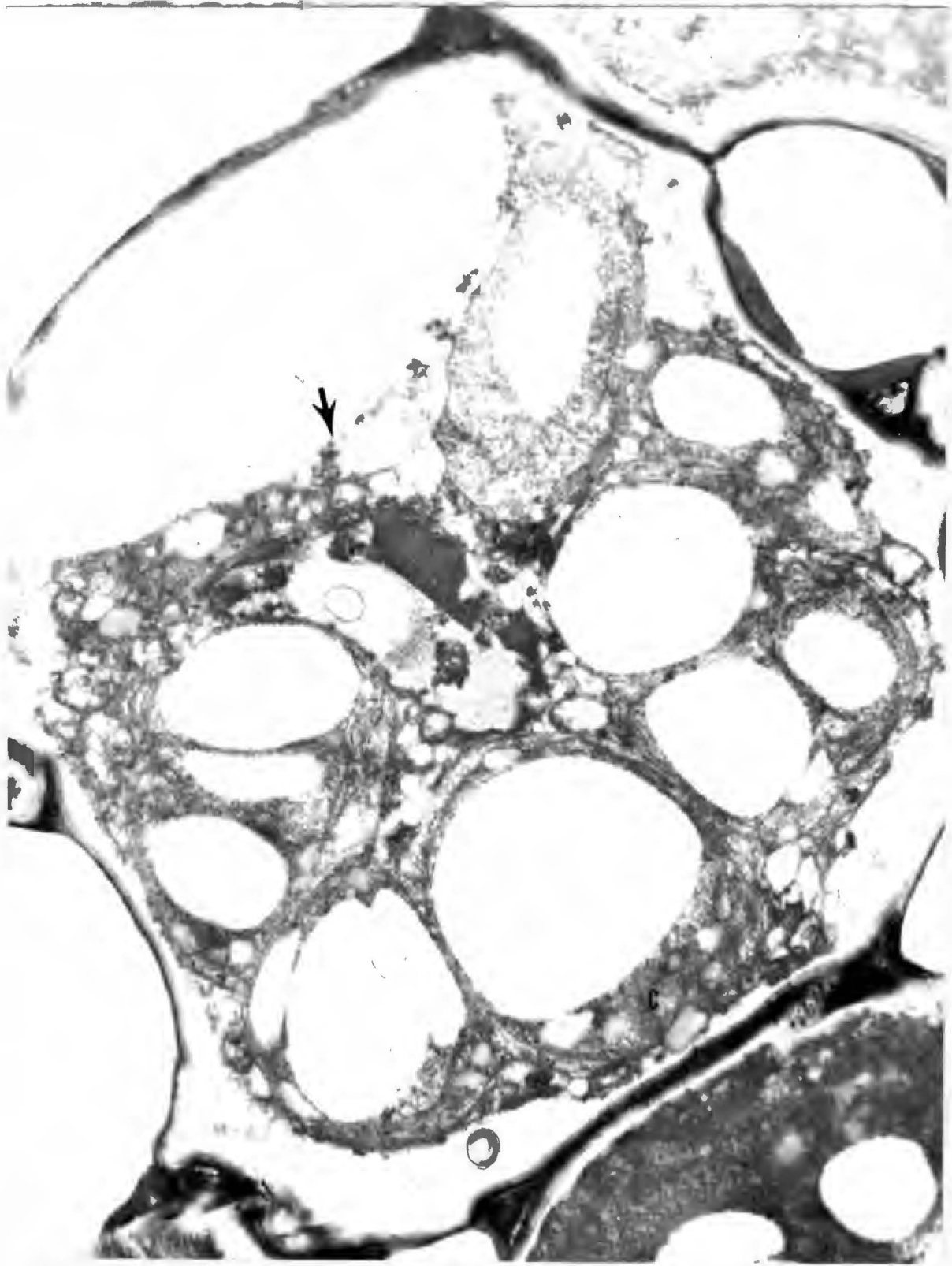
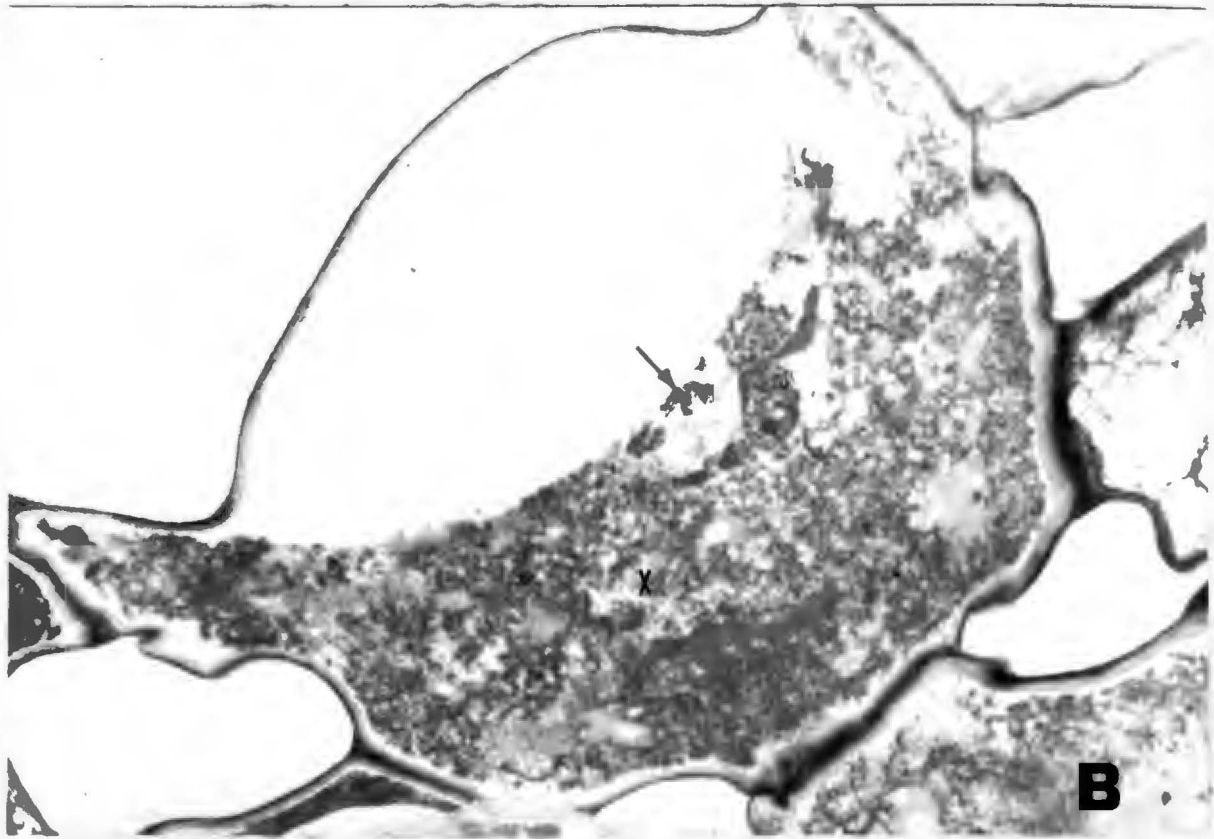
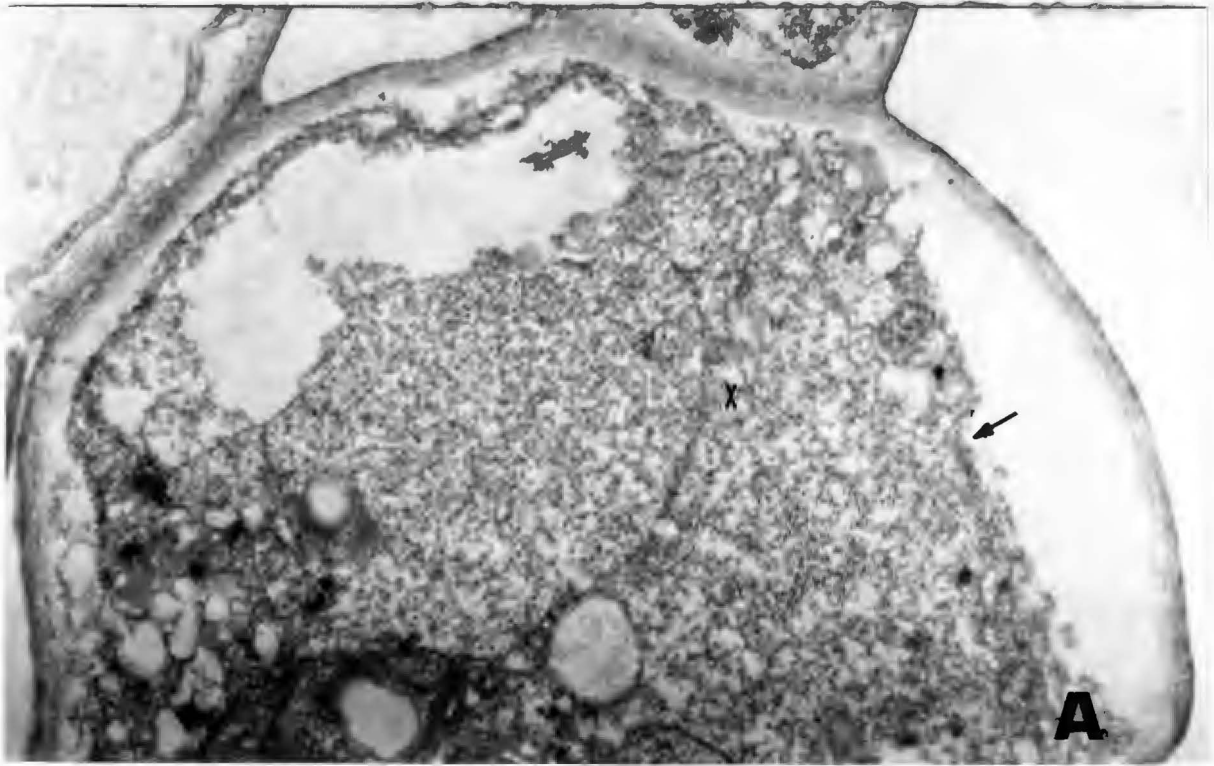


Figure 5A, 5B. Calcium deficient taro leaf blade parenchyma cells. No tonoplast or plasma membrane is evident. The vacuolar contents appear to have mixed with the cytoplasm (X). Arrows point to the extreme separation of the cytoplasm from the cell walls. (x 4000)



LITERATURE CITED

- Allen, S. E., H. M. Grimshaw, J. A. Parkinson and C. Quarmby. 1974. Chemical Analyses of Ecological Materials. Blackwell Scientific Publications. Oxford. pp. 260-261.
- Arnott, H. J. and F. G. E. Pautard. 1970. Calcification in plants. In Biological Calcifications: Cellular and Molecular Aspects. (ed. H. Schraer). Appleton-Century-Crofts. pp. 375-446.
- Bangerth, F. 1973. Investigations upon Ca related physiological disorders. Phytopath. Z. 7:20-37.
- Bibbulph, O., F. S. Nakayama and R. Cory. 1961. Transpiration stream and ascension of calcium. Plant Physiol. 36:429-426.
- Black, O. F. 1918. Calcium oxalate in the dasheen. Am. J. Bot. Sci. 5:447-451.
- Bowers, F. A. I., D. L. Plucknett and O. R. Younge. 1964. Specific gravity evaluation of corm quality in taro. Hawaii Agric. Exp. Sta. Circ. 61. 19 pgs.
- Brumagan, D. M. and A. J. Hiatt. 1966. The relationship of oxalic acid to the translocation and utilization of calcium in Nicotiana tabacum. Plant and Soil. 24:239-249.
- Cable, W. J. 1973. Potassium requirement of taro (Colocasia esculenta (L.) Schott) in solution culture. M.S. Thesis, University of Hawaii.
- Cate, R. B. and L. A. Nelson. 1965. A rapid method for correlation of soil test analyses with plant response data. Inter. Soil Testing Ser. Tech. Bull. 1. N. Carolina State Univ. Agric. Exp. Sta., Raleigh.
- Chang, S. Y., R. H. Lowe and A. J. Hiatt. Relationship of temperature to the development of calcium deficiency symptoms in Nicotiana tabacum. Agron. J. 60:435-436.
- Chew, W. Y. 1971. Yield and growth responses of some leguminous and root crops grown on acid peat to magnesium lime. Malaysian Agric. J. 48:142-157.
- Ching, K. W. 1969. Some aspects of the physiological ontogeny of starch, total sugar, protein, leaf and corm of Colocasia esculenta. M.S. Thesis, University of Hawaii.
- Christiansen, M. N. and C. D. Foy. 1979. Fate and function of calcium in tissues. Comm. Soil Sci. and Plant Analysis. 10: 427-442.

- Christiansen, M. N., H. R. Carns and D. J. Slyter. 1970. Stimulation of solute loss from radicles of Gossypium hirsutum L. by chilling, anaerobiosis, and low pH. *Plant Physiol.* 46:53-56.
- Coil, B. J. and J. Bonner. 1956. The nature of growth inhibition by calcium in Avena coleoptile. *Planta.* 48:696-723.
- de la Pena, R. S. and D. L. Plucknett. 1969. The response of taro (Colocasia esculenta (L.) Schott) to N, P, and K fertilization under upland and lowland conditions in Hawaii. *Inter. Symp. Trop. Root Crops, Trinidad, 1967, Proc. 1st. Vol. 1, Sect. 2:* 270-284.
- Dickinson, D. B. and J. P. McCollum. 1964. Effect of calcium on cracking in tomato fruits. *Am. Soc. Hort. Sci. Proc.* 84:485-490.
- Epstein, E. 1972. Mineral Nutrition of Plants: Principles and Perspectives. Wiley. pgs. 117-120.
- Evans, E. C. 1964. Polar transport of calcium in the primary root of Zea mays. *Science.* 144:174-177.
- Essau, K. 1977. Anatomy of Seed Plants. John Wiley & Sons. pgs. 183-185, 192-193.
- Ezumah, H. C. 1972. The growth and development of taro, Colocasia esculenta (L.) Schott, in relation to selected cultural management practices. Ph.D. Dissertation, University of Hawaii.
- Ferguson, I. B. and D. T. Clarkson. 1976. Simultaneous uptake and translocation of magnesium and calcium in barley (Hordeum vulgare (L.) roots. *Planta (Berl.)* 128:267-269.
- Garrard, L. A. and T. E. Humphreys. 1967. The effect of divalent cations on the leakage of sucrose from corn scutellum slices. *Phytochem.* 6:1085-1095.
- Gary-bobo, C. M. 1970. Effect of Ca^{++} on water and non-electrolyte permeability of phospholipid membranes. *Nature.* 228:1101-1102.
- Geraldson, C. M. 1957a. Factors affecting calcium nutrition of celery, tomato and pepper. *SSAP.* 21:621-625.
- _____. 1957b. Control of blossom end rot of tomatoes. *Am. Soc. Hort. Sci.* 69:309-317.
- Gerard, C. J. 1971. Influence of osmotic potential, temperature, and calcium on growth of plant roots. *Agron. J.* 63:555-558.
- _____ and E. Hinojosa. 1973. Cell wall properties of cotton roots as influenced by calcium and salinity. *Agron. J.* 65: 556-560.

- Hanger, B. C. 1979. Movement of calcium in plants. *Comm. Soil Sci. Plant Analysis.* 10:171-193.
- Hawaii Agr. Exp. Sta. 1938. Diseases of taro. *Ann. Rep. Hawaii Agr. Exp. Sta.* 1938:36-39.
- _____. 1937. Diseases of taro. *Ann. Rep. Hawaii Agr. Exp. Sta.* 1936:35-38.
- _____. 1936. Diseases of taro. *Ann. Rep. Hawaii Agr. Exp. Sta.* 1936:33-35.
- Hecht-Buchholz, Ch. 1979. Calcium deficiency and plant ultra-structure. *Comm. Soil Sci. Plant Analysis.* 10:67-82.
- Hoagland, D. R. and L. A. Nelson. 1965. The water culture method for growing plants without soil. *Calif. Agric. Exp. Sta. Circ.* 347.
- Jacobson, L., D. P. Moore and R. J. Hannapel. 1960. Role of calcium in absorption of monovalent cations. *Plant Physiol.* 35:352-358.
- Jackson, W. A. 1967. Physiological effects of soil acidity. In *Soil Acidity and Liming.* (ed. R. W. Pearson and F. Adams). *Amer. Soc. Agron., No. 12 Ser. Agron. Madison, Wisconsin.* pp. 43-123.
- Jones, R. G. W. and O. R. Lunt. 1967. Function of calcium in plants. *Bot. Rev.* 33:407-426.
- Kawaski, T. and M. Moritsugu. 1979. A characteristic symptom of calcium deficiency in maize and sorghum. *Comm. Soil Sci. Plant Analysis.* 10:41-56.
- Kay, D. E. 1973. *Root Crops.* Trop. Prod. Institute. pp. 71-75, 169-179.
- Kirkby, E. A. 1979. Maximizing calcium uptake by plants. *Comm. Soil Sci. Plant Analysis.* 10:89-113.
- Loneragan, J. F. and K. Snowball. 1969. Rate of calcium absorption by plant roots and its relation to growth. *Austr. J. Agric. Res.* 20:479-490.
- Ludders, P. 1979. Effect of nitrogen nutrition on Bitter Pit in apples. *Comm. Soil Sci. Plant Analysis.* 10:401-415.
- Marinos, N. G. 1962. Studies on submicroscopic aspects of mineral deficiencies. I. Calcium deficiency in the shoot apex of barley. *Am. J. Bot.* 49:834-841.

- Marschner, H. 1974. Calcium nutrition of higher plants. Neth. J. Agric. Sci. 22:275-282.
- _____ and I. Gunther. 1964. Ionenaufnahme und zellstruktur bei gerstenwurzeln in abhankigkeit von der calcium-versorgung. Zeitschrift fur Pflanzenernahrung Dungung Bodenkunde. 107:118-137.
- Maynard, D. M. and A. V. Barker. 1972. Internal Browning of brussel sprouts: a calcium deficiency disorder. J. Am. Soc. Hort. Sci. 97:789-792.
- Millaway, R. M. and L. Wiersholm. 1979. Calcium and metabolic disorders. Comm. Soil Sci. Plant Analysis. 10:1-28.
- Morre, D. J. and C. E. Bracker. 1976. Ultrastructural alteration of plant plasma membranes induced by auxin and calcium ions. Plant Physiol. 58:544-547.
- Mostafa, M. A. E. and A. Ulrich. 1976. Interaction of calcium and magnesium in nutrition of intact sugarbeets. Soil Sci. 121: 16-20.
- Nada, I. A. A. and M. A. Moursi. 1958. Distribution of nitrogen, phosphorus, potassium and calcium in the corms of dasheen. Ann. Agric. Sci. Fac. Agric., Ain Shams, Univ. Cairo. 3(2): 113-119.
- Nilshammar, M., B. Walles and A. Kylin. 1972. The effect of calcium deficiency on the ultrastructure of the green alga Scenedesmus Z. Pflanzenphysiol. 66:197-205.
- Paliwal, G. S. and A. K. Kavathekar. 1972. Anatomy of vegetative food storage organs. II. Stems. Acta Agron. Acad. Sci. Hungaricae. 21(3/4):313-318.
- Parris, G. K. 1941. Diseases of taro in Hawaii and their control with notes on field production. Hawaii Agric. Exp. Sta. Circ. No. 18. 29 pgs.
- Paul, D. C. and Ch. W. Goff. 1973. Comparative effects of caffeine, its analogues and calcium deficiency on cytokinesis. Exptl. Cell Res. 78:399-413.
- Pearson, R. W. 1975. Soil acidity and liming in the humid tropics. Cornell Internatl. Agric. Bull. No. 30. 66 pgs.
- Plucknett, D. L. and R. S. de la Pena. 1971. Taro production in Hawaii. World Crops. 23(5):244-249.
- _____, R. S. de la Pena and F. Obrero. 1970. Taro (Colocasia esculenta). Field Crop Abstracts. 23(4):413-426.

- Poovaiah, B. W. 1979a. Effects of inorganic cations on Ethephon-induced increases in membrane permeability. *J. Am. Soc. Hort. Sci.* 104:164-166.
- _____. 1979b. Roles of calcium in ripening and senescence. *Comm. Soil Sci. Plant Analysis.* 10:83-88.
- _____ and A. C. Leopold. 1976. Effects of inorganic salts on the binding of auxin. *Plant Physiol.* 58:783-785.
- Potgieter, M. 1940. Taro (*Colocasia esculenta*) as a food. *J. Am. Diet. Assoc.* 16:536-540.
- Reitemeir, R. F. 1951. Soil potassium. *Adv. in Agron.* 3:143-151.
- Rotar, P. P., D. L. Plucknett and B. K. Bird. 1978. Bibliography of Taro and Other Edible Aroids. University of Hawaii. 3420 pgs.
- Sakai, W. S. and M. Hanson. 1974. Mature raphid and raphid idioblast structure in plants of the edible aroid genera *Colocasia*, *Alocasia*, and *Xanthosoma*. *Ann. Bot.* 38:739-748.
- _____, M. Hanson and R. C. Jones. 1972. Raphides with barbs and grooves in *Xanthosoma sagittifolium* (Araceae). *Science.* 178:314-315.
- Shear, C. B. 1975. Calcium-related disorders of fruits and vegetables. *Hort Sci.* 10:361-365.
- _____. 1972. Incidence of cork spot as related to calcium in the leaves and fruit of 'York Imperial' apples and fruit of 'York Imperial' apples. *J. Am. Soc. Hort. Sci.* 97:61-64.
- _____. 1971. Symptoms of calcium deficiency on leaves and fruit of 'York Imperial' apple. *J. Am. Soc. Hort. Sci.* 96:415-417.
- Sickey, B. 1973. Diseases of taro. *Science in New Guinea.* 1(3/4): 45-50.
- Snedecor, G. W. and W. G. Cochran. 1967. Statistical Methods, 6th ed., pgs. 272-273. Iowa State Univ. Press. Ames, Iowa.
- Sorokin, H. and A. L. Sommer. 1940. Effects of calcium deficiency upon the roots of *Pisum sativum*. *Am. J. Bot.* 27:308-318.
- Srivastava, S. K. and P. S. Krishnan. 1959a. Role of oxalic acid in higher plants. *J. Sci. and Indus. Res.* 18C:220-224.
- _____ and _____. 1959b. Oxalate content of plant tissues. *J. Sci. and Indus. Res.* 18C:220-224.
- Steffensen, D. 1958. Chromosome Aberrations in calcium-deficient *Tradescantia* produced by irradiation. *Nature.* 182:1750-1751.

- Suehisa, R. A. and J. C. Deputy. 1979. In Sugarcane Crop Logging and Crop Control, Principles and Practices. Ed. H. F. Clements. University of Hawaii Press. Honolulu, Hawaii.
- Takahashi, M. 1953. Report of Taro Diseases in Hawaii. Unpublished manuscript. 61 pgs.
- Tisdale, S. L. and W. L. Nelson. 1975. Soil Fertility and Fertilizers. MacMillan Publishing Co., New York.
- Trujillo, E. E. 1967. Diseases of the genus Colocasia in the Pacific area and their control. Proc. Int. Symp. Trop. Root Crops, Trinidad. 2(4):13-18.
- _____. 1979. Personal communication.
- Walker, J. M. 1969. One-degree increments in soil temperatures affect maize seedling behavior. SSSAP. 33:729-736.
- Wallace, A. and E. Frohlich. 1966. Calcium requirements of higher plants. Nature. 209:634.
- Watson, L. J. 1970. The legal importance of the water requirements of taro (Colocasia esculenta) in Hawaii. Trop. Root and Tuber Crops Tomorrow. 1:149-151.
- Wiersum, L. K. 1966. Calcium content of fruits and storage tissues in relation to the mode of water supply. Acta Botanica Neerlandica. 15:406-418.
- Wilcox, G. E., C. A. Mitchell and J. E. Hoff. 1977. Influence of nitrogen form on exudation rate, and ammonium, amide, and cation composition of xylem exudate in tomato. J. Am. Soc. Hort. Sci. 102:192-196.
- Wilcox, G. E., J. E. Hoff and C. M. Jones. 1973. Ammonium reduction of calcium and magnesium content of tomato and sweet corn leaf tissue and influence on incidence of blossom end rot of tomato fruit. J. Am. Soc. Hort. Sci. 98:86-89.
- Woodbridge, C. G. 1971. Calcium levels of pear tissue affected with Cork and Black End. Hort Sci. 6:451-453.

