A STUDY OF FACTORS INFLUENCING THE PRODUCTION OF TUBERS BY POTATO LEAF BUD CUTTINGS

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Sharon Arnold

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ABSTRACT

Arnold, Sharon, A. M.Sc., The University of Manitoba, February 1986.

A Study of Factors Influencing the Production of Tubers by Potato Leaf Bud Cuttings. Major Professor; Dr. L.J. LaCroix

Some factors influencing the production of tubers from potato leaf bud cuttings for the purpose of rapid clonal increase of selected stocks were studied. All cuttings used in this study were taken from plants grown from physiologically old seed tubers.

Axillary buds either developed directly into tubers, produced a stolon and then a tuber or produced a leafy shoot, a stolon and then a tuber. The latter two types of development generally resulted in larger tuber size.

With both Norland and Russet Burbank cuttings, the middle leaf positions were most desirable as a source of cuttings. Cuttings from younger plants tended to produce larger tubers than those from older plants. It was also found that the addition of a soluble fertilizer to the cuttings could promote high tuber yields.

With the cultivar Norland, exposure of the mother plants to inductive conditions (short days and low temperatures) for a period of two weeks before cuttings were taken generally favored leafy shoot and/or stolon development from axillary buds. Non-inductive culture conditions thereafter favored development of larger tubers.

The cultivar Russet Burbank appeared to be less responsive to an inductive environment both for the mother plants and for the cuttings. It would appear that the whole culture procedure may be most successful under non-inductive conditions.

INTRODUCTION

A potato breeder selects a promising cultivar that he wishes to introduce into his breeding program or a desirable cultivar that he wishes to release. A grower from an Elite Seed Potato Farm obtains a small amount of virus free material and must introduce it into commercial seed potato production. The next problem facing these individuals is to increase the selected stocks as rapidly as possible to meet demand, and, in the case of the virus free material, to keep re-infection rates at a minimum.

After the mother plants have been produced from tubers, plantlets, or whatever the original source, although tissue culture techniques may become important in the future, there are currently two main methods used to increase potato stocks.

The first method is the rooted cutting procedure. Mother plants are grown in the greenhouse in late winter. Stem cuttings consisting of several nodes are taken, rooted in the greenhouse, and then transplanted out in the field in May (Lauer, 1977). These cuttings produce fairly vigorous and uniform plants and yield tubers that do not differ markedly from control plants grown in the field from tuber seed pieces (Cole and Wright, 1967).

There are a number of problems associated with this method: 1) The handling of the cuttings, particularly the transplanting operation, is labor intensive (Rossnagel, 1980). 2) The cuttings may not be stored. 3) If transplanting is delayed, tubers may form on the cuttings, particularly on the early maturing cultivars such as Norland and Norchip. This severely inhibits further tuber

production in the field (Rossnagel, 1980). 4) The number of cuttings obtainable from each mother plant may be limited. Usually 5-8 rooted cuttings are secured from each plant during the spring (Lauer, 1977).

The second method is to obtain tubers directly from leaf bud cuttings. With this procedure, a potato leaf and short section of stem with a single subtended bud is taken, and the stem, axillary bud, and part of the petiole is placed in soil. Given the right conditions, the axillary bud will grow out and form one or more tubers. These tubers can be taken and planted out in the field. They can be handled like any other tuber in terms of being mechanically planted and harvested (Lauer 1977).

In one cycle of propagation, Lauer (1977) obtained well over 100 small tubers for each mother plant. Goodwin (1981) noted that a non dormant medium sized tuber yields 250 nodal cuttings within 2 months of planting and then up to 200 nodal cuttings every 2 weeks thereafter. This compares very favorably to the 5 to 8 rooted cuttings obtained by the first method.

Again, this procedure is not problem free: 1) There is difficulty sometimes in getting the cuttings to form tubers (Rossnagel, 1980). 2) The tubers obtained from leaf bud cuttings tend to be considerably smaller than conventional tubers produced from tuber seed pieces (Lauer, 1977). Some tubers are too small to be usable (Rossnagel, 1980). From the small tubers (i.e. up to 4 g) haulm development is slower than from the 42 g seed tuber used by the industry (Lauer, 1977). 3) The tubers tend to be quite dormant when first produced (Lauer, 1977) and so far none of the dormancy breaking mechanisms has been completely successful (Lauer, 1980). This necessitates the production of tubers by February 15, particularly in cultivars having long rest periods (Lauer, 1977) in order for them to be planted in the field in May.

Although there has been a great deal of research on tuberization in potatoes, very little has been reported on the process of obtaining tubers from leaf bud

cuttings for rapid clonal increase of selected stocks. The purpose of the research being reported in this thesis was to provide information on some of the factors that influence and/or enhance tuber production by leaf bud cuttings with the object of obtaining the largest numbers and sizes of tubers possible. Factors investigated include: 1) different cutting environments, 2) age of the mother plant, 3) pretreatment of mother plants with inductive and non-inductive temperatures and photoperiods, 4) leaf positions on the stem (leaf age), 5) leaf area of the cuttings, 6) addition of fertilizer to the cuttings, and 7) patterns of bud development. Under limited conditions, treatment of cuttings with ABA and CO_2 enrichment of the soil zone were also studied.

LITERATURE REVIEW

Tubers From Leaf Bud Cuttings

The leaf bud cutting consists of a leaf, petiole, and part of the stem with its subtended bud (Lauer, 1977). When the bud and piece of stem are placed in soil, the axillary buds can grow out and form a tuber. A tuber is a shortened thickened stem with leaves reduced to scales subtending the axillary buds known as eyes (Gregory, 1956). Frequently tubers are formed from rhizomes (modified shoots) which are often called stolons (Gregory, 1965).

The axillary bud will frequently enlarge and form a tuber, but tubers can arise from one or more stolon initials found at the sides of the axillary bud (Gregory, 1956). This phenomenon has long been recognized. Vochting (1887) first reported the formation of sessile tubers at covered nodes when cut pieces of stem were inserted in the soil. Bates (1943) observed the formation of small tubers in axils of leafless potato stem cuttings planted in the soil and in darkness. Gregory (1956) found that tubers could form in leaf axils without planting in soil or providing darkness. He concluded that where the tuberization stimulus is very strong, tubers may develop in leaf axils even in the light. Both Gregory (1956) and Chapman (1958) observed that every leaf axil of the stem of a potato plant can differentiate tubers when properly cultured.

With the exception of that of Lauer (1977), few studies have been published on obtaining tubers from leaf bud cuttings for the purpose of rapid clonal increase of selected stocks. However, leaf bud cuttings and other stem cuttings consisting of several nodes, have occasionally been used in studies as a screening tool for photoperiod and heat response (Ewing and Wareing, 1978; Ewing 1981). They have

also been used in studies to elucidate the nature of the tuberization stimulus and process (Kahn, 1982). Leaf bud cuttings have also been used to obtain rooted plants for transplant to the field (Goodwin, 1981).

Gregory (1965) recognized three phases of development in the formation of potato tubers: 1) tuber initiation characterized by the differentiation of the bud into tuber primordium but with no visible tuber present; 2) tuber enlargement characterized by high cell division activity accompanied by reserve food accumulation; 3) tuber maturation, when the tuber becomes dormant.

Tuber Initiation -- The Tuberization Stimulus

Early researchers suggested that tuberization was primarily controlled by levels of available photosynthate (Beaumont and Weaver, 1931; Werner, 1934). They thought that tuberization occurred whenever levels of carbohydrate produced by the plant exceeded that which was required for basic maintenance and growth. This would suggest the 'push' of carbohydrate from the leaves to a passive sink. It is now commonly accepted that a specific tuber forming substance is involved (Gregory, 1956; Chapman, 1958; Kumar and Wareing, 1973) which suggests the 'pull' of carbohydrate to an active sink.

The state of tuber initiation in the potato appears to be promoted by a graft transmissible stimulus arising in the plant under specific conditions of temperature and photoperiod (Gregory, 1956) and is influenced by nitrogen nutrition (Sattelmacher and Marschner, 1979) and the physiological age of mother tubers (Okazawa and Chapman, 1962; Montaldi and Claver, 1963).

Inductive conditions are said to be those environmental conditions under which tuberization takes place and non-inductive conditions those where no tubers form (Gregory, 1965). Similarly, the physiological condition in the plant that gives rise to tuberization is the inductive state of the plant and the state where no tubers form, the non-inductive state (Gregory, 1965). The inductive state in the plant is

attained by exposing the plant for a period of time to proper environmental conditions. As conditions for induction approach optimum levels, less time is required for the inductive state to be attained (Gregory, 1965).

The tuber inducing stimulus appears to be produced largely at active, above ground growing points (Chapman, 1958) and also in the mother tuber (Okazawa and Chapman, 1962; Lawrence and Barker, 1963). Hammes and Beyers (1973) found that the photoperiodic stimulus was perceived by old leaves as well as young leaves. It has been found to be present throughout the plant and not restricted to underground parts (Gregory, 1956; Chapman, 1958). A quantitative relationship seems to exist between the duration of induction (i.e., the length of time the plant is placed in conditions that promote tuberization) and the amount of the tuber producing factor produced (Gregory, 1956; Chapman, 1958).

This stimulus may remain active in the plant for a certain time even when the plant is moved to conditions unfavorable for tuber induction (Gregory, 1956). A reversion to a vegetative (i.e., non tuberizing) state may occur after the plant is moved to unfavorable conditions (Chapman, 1958). Gregory (1956) observed in one particular study that induction appeared to be irreversible in cuttings but not in the intact plant. Madec and Perennec (1962) and Ewing (1981) have since found cuttings in which induction is reversible. It has been concluded that there is a need for a continuous supply of the tuber inducing stimulus following tuber initiation for tuberization to continue (Chapman, 1958; Ewing, 1981).

The exact chemical or physical nature of the stimulus is still uncertain. Grafting experiments (Gregory, 1956; Chapman, 1958) and especially the demonstration that even a small piece of stem from an induced plant is capable of transmitting the tuberization stimulus (Kumar and Wareing, 1973) offer evidence that it is hormonal in nature.

Exogenously applied growth substances have been found to inhibit or promote

tuberization in intact plants or parts of plants. Gibberellins stimulate haulm growth and retard tuber formation (Lovell and Booth, 1967;Smith and Rappaport, 1969; Kumar and Wareing, 1974; Menzel, 1980). Low levels of gibberellins are associated with tuberization (Kumar and Wareing, 1974). An artificial antigibberellin, (2-chloroethyl) trimethylammonium chloride (CCC), has been shown to promote tuberization by overcoming effects of daylengths (Kumar and Wareing, 1974), temperatures (Menzel, 1980), and nitrogen levels (Krauss and Marschner, 1976) that normally have a negative effect on tuber initiation.

Applications of abscisic acid (ABA) have been found to promote tuberization in some studies (Krauss and Marschner, 1976; Menzel, 1980) and not in others (Smith and Rappaport, 1969; Claver, 1970). Cytokinins can promote tuberization in vitro (Forsline and Langille, 1976). They have also been found to increase in above ground parts of induced plants and in stolon tips and young tubers during tuberization (Okazawa, 1970; Forsline and Langille, 1975). Ethylene has also been found to have both positive (Catchpole and Hillman, 1969) and negative (Palmer and Barker, 1973; Mingo-Castel et al., 1974, 1976) effects on tuberization.

Yet none of these growth substances, alone, or in combination, have been able to perfectly mimic the tuberization stimulus. Although an increasing number of research workers are slowly reaching agreement on a number of important aspects, a generally accepted theory regarding the mechanism of tuber initiation does not exist. The contemporary school of thought is that tuber initiation is controlled by a balance of growth substances, generally phytohormones and inhibitors, rather than specific hormones (Hammes and Nel, 1975).

Tuber Enlargement

Tuber enlargement is recognized to be a distinct phase of development separate from tuber initiation (Gregory, 1965). Although carbohydrate accumulation is apparently not responsible for actual tuber initiation, it plays a key

role in tuber enlargement (Gregory, 1956; Gregory, 1965). It has been found that the size of a tuber is limited by photosynthate available (Gregory, 1965). The optimum conditions for tuber enlargement are those promoting the production and accumulation of photosynthate (e.g. high photosynthetic rate, arrested plant growth and slow respiration rate during the dark period). Factors affecting the rate of photosynthesis include temperature, light intensity, carbon dioxide supply, moisture supply, nutrient supply, chlorophyll content and age of the foliage (Smith, 1977). In intact plants, there is considerable evidence that the rate of tuber bulking controls the rate of photosynthesis (Nösberger and Humpheries, 1965; Moorby and Milthorpe, 1978). The rate of bulking appears to be determined around the time of tuber initiation and is probably some function of the number of growing tubers set and the amount of leaf area then supplying them with photosynthate (Moorby and Milthorpe, 1978). Once the rate of bulking is established, it is apparently relatively insensitive to fluctuating light supply and temperature, but not to water supply. Werner (1957) reported that the time and amount of tuber growth is determined by the interaction effect of temperature, photoperiod, soil fertility, moisture and cultivar.

In intact plants, it is estimated that 90% of the dry matter that moves into the tubers is dependent on current assimilation (Moorby, 1970). It is estimated that 10% of the dry weight of tubers comes from the transfer of previously assimilated material (Moorby and Milthorpe, 1978). Tuberization is accompanied by a growth shift away from vegetative growth to tuber growth which would indicate that the tubers are getting the largest supply of available photosynthate. After tuber initiation, there are decreases in shoot growth, stolon growth, and rooting (Hammes and Nel, 1975). There is a gradual increase in the rate of senescence in the older leaves, a net migration of N, P and K from the haulm to the tubers and a decrease in the dry weight of the haulm (Moorby and Milthorpe, 1978). The senescence of the

haulm accelerates as tuber growth increases (Moorby and Milthorpe, 1978) and when it becomes complete, tuber growth necessarily ceases.

Transition from the vegetative phase to the tuberization phase occurs progressively over a relatively short period of time (Madec and Perennec, 1962). This transition phase is one of unstable physiological equilibrium in the plant and it is during this time that a reversion to the vegetative phase of growth may occur if the environment ceases to be inductive (Madec and Perennec, 1962). In the intact plant, there are conditions where vegetative growth and tuberization may occur simultaneously if the mother tuber itself initiates and maintains tuberization (Madec and Perennec, 1962).

There are distinct differences between the development of the inductive state, the beginning of enlargement of tubers and the subsequent maintenance of the enlargement of the tubers (Madec, 1966). Enlargement is basically a form of growth that primarily requires carbohydrates and continues as long as the induction is sustained (Chapman, 1958; Madec, 1966; Ewing, 1981).

Ewing (1981), based on his work with cuttings with essentially a single locus for tuber initiation, states, "conditions that favor tuber initiation on a high percentage of cuttings invariably favor tuber enlargement as well. That is, in the cutting system, there is a strong positive correlation between the percentage of cuttings that initiate tubers and the average size of tubers." He concludes that the tuberization stimulus favors both tuber initiation and tuber enlargement in cuttings. This conclusion was reached while studying tuber production by cuttings which were selected for early tuberization .

Factors Influencing Tuber Initiation and Enlargement

The tuberization stimulus arises in the plant under specific temperature and photoperiod conditions (Gregory, 1956); the enlargement phase is dependent on available photosynthate (Gregory, 1965) and the continued presence of the

tuberization stimulus (Chapman, 1958; Madec, 1966; Ewing, 1981).

Each developmental phase possesses its own set of optimum conditions. As conditions for induction approach an optimum, less time is required for induction to occur (Gregory, 1965). Tuberization may also occur under conditions not thought to be favorable (Madec, 1966). The further conditions are from the optimum, the longer it will take for induction to occur, if it does occur at all (Gregory, 1965).

The range of conditions over which tuberization will occur may be either broadened or narrowed by interactions between conditions (Gregory, 1965). For example, in the cultivar Kennebec, tuberization takes place only under short photoperiods at high temperatures while tubers will form under continuous light at low temperatures (Gregory, 1965).

The genetic makeup of the plant establishes the optimum and range of acceptable conditions for tuberization. The environment, in turn, controls tuberization because a plant will not tuberize until its inherent requirements are met (Gregory, 1965). Variations in the response of plants to various conditions may be found within species, cultivars, and even clones (Gregory, 1965).

Photoperiod

It is generally agreed that the initiation of tuberization in the cultivated potato may be promoted by short photoperiods and delayed by long ones (Gregory, 1965). The tuberization stimulus is favored by photoperiods shorter than a critical photoperiod (Garner and Allard, 1923) and the length of this varies with genotype (Kopetz and Steineck, 1954). The critical photoperiod is defined as the longest photoperiod to which the plant may be exposed and still have tubers form (Ewing, 1978). This critical photoperiod is not absolute because it may be modified by other conditions such as temperature (Gregory, 1965).

Long days delay the onset of tuberization in many cultivars, but encourage vegetative growth, development and growth of stolons, and lengthen the period of

active tuber growth (Gregory, 1965). Therefore, greater yields may result under long days which favor the tuber enlargement phase.

Temperature

The potato plant has different optimum temperature requirements for different stages of growth (Smith, 1977). In general, high temperatures favor vegetative growth (Ewing, 1981) and low temperatures promote tuberization (Marinus and Bodlaender, 1969). Ku and Edwards (1976) estimated an optimum temperature range between 16 and 25°C for net photosynthesis in potatoes. The figure of 17°C has been widely accepted as the optimum mean temperature for good yields (Ewing, 1981). However, temperature effects are complex and subject to interactions and one cannot state a particular optimum temperature to give the highest yields under all conditions (Kahn, 1979).

High heat may result in reduced partitioning to the tubers (Ewing, 1981). Gregory (1965) found that Kennebec produced 12 times as much haulm growth at 30/23° C day/night temperatures as when temperatures were 17/10°C, yet tuber yields were 19 times greater at cooler temperatures.

Fertilizer

Continuous high levels of nitrogen delay or inhibit tuber initiation; discontinuous or low levels promote it (Sattelmacher and Marschner, 1979; Krauss and Marschner, 1976). The overall effect of supplying increasing amounts of nitrogen, phosporous and potassium to intact plants was to increase rate of haulm growth, time to tuber initiation, leaf area, leaf area duration and yield, with phosphorous and potassium having smaller and less persistent effects than nitrogen (Dyson and Watson, 1971; Moorby and Milthorpe, 1978). However, overfertilization with nitrogen results in decreased maturity and total solids and no further yield increase (Sawyer, 1959).

Dyson and Watson (1971) suggested that the availability of nitrogen (and possibly other nutrients) may influence sink capacity and tuber growth. When nitrogen uptake becomes inadequate, the consequent removal of nitrogen from the haulm accelerates leaf death, decreases leaf area and tuber growth rate.

Gunasena and Harris (1969) varied the rates and time of nitrogen application to intact plants and found that delaying nitrogen application until after tuber initiation increased yield and decreased tuber numbers.

It is apparent that short days, cool temperatures and low nitrogen fertilization favor tuber initiation. These factors, however, must not be present all together too early in the season, or the stimulus will be so strong that haulms will be dwarfed and tuber yield, though early, will be negligible (Ewing, 1981). It is essential for a plant to have the ability to support tuber growth after initiation. This ability is dependent on sufficient foliage to produce necessary assimilates and adequate supplies of water and mineral nutrients (Moorby, 1968).

Carbon Dioxide Enrichment of the Root Zone

Potato plants are especially well suited for CO_2 enrichment of the root zone because of their ability to tolerate high concentrations of CO_2 without the roots being damaged (Arteca et al., 1979.) Arteca et al. (1979) enriched the root zone of Russet Burbank plants with CO_2 and found increased carbon fixation, numbers of tubers per plant, tuber weights, stem lengths, and plant dry weights. Arteca and Poovaiah (1980) also found that short term CO_2 enrichment of the root zone of potato plants increased the photosynthetic rate and suppressed the rate of photorespiration.

Abscisic Acid

The natural growth inhibitor, abscisic acid, (ABA) has been found to promote tuberization in some studies (Krauss and Marschner, 1976; Menzel, 1980). In other

studies, it has failed to promote tuberization (Smith and Rappaport, 1969; Claver, 1970). ABA also plays a role in stomatal closure which suggests the possiblility that this action might have important regulatory functions in the water relationships of plants (Kriedemann et al., 1972).

Leaf Positions

From numerous comparisons of single leaf cuttings from both young and old nodes, Ewing (1978) found that all nodes on a given plant responded in approximately the same way, provided leaves had not become excessively senescent. Simmonds (1965) and Lauer (1977) reported that leaf bud cuttings from the apex of the plant tended to produce both roots and tubers. Leaf bud cuttings from the midsection of a plant produced only tubers, and those from the basal portion of the plant produced the smallest tubers.

After a certain point, the rate of photosynthesis of a leaf decreases with increasing leaf age. In intact plants, older leaves are generally less efficient and productive in terms of their photosynthetic capacity than younger ones (Chapman, 1958). This relationship also holds when the photosynthesis of detached potato leaves is measured (Meinl, 1965).

Leaf Area

Ewing (1978) found that cuttings with larger leaves seemed more likely to tuberize than cuttings with small leaves. Tuberization on stem cuttings was positively correlated with number of leaves and leaf area (Kahn, 1982). The rational suggested for these results is that plants with greater leaf areas have a potential for production of more tuberization stimulus under borderline inductive conditions than a smaller plant. As well, greater leaf area would indicate greater photosynthetic capability.

In the intact plant, with any one cultivar, the tendency is that the smaller the leaf area at the time of tuber initiation, then the slower is the rate of bulking and lower the final yield (Moorby and Milthorpe, 1978). In intact plants, attempts to correlate tuber growth with leaf area have been relatively unsuccessful (Moorby, 1968).

Mother Plant Age

As plants from which cuttings were taken grew older, the ability to root decreased. Tuber initiation showed the opposite trend, increasing with increasing mother plant age while rooting ability decreased (Simmonds, 1965).

Ewing (1978) found that the tuberization response in cuttings was less affected than tuber yield by the age of mother plants from which cuttings were taken. Although he did not find a greater percentage of cuttings from older mother plants tuberizing, he found that tubers from cuttings from older mother plants tended to be larger than those from younger plants. Again, he was selecting for early tuberization of cuttings.

Patterns of Bud Development in Leaf Bud Cuttings

Ewing and Wareing (1978) observed different responses at the buried bud of the cutting depending on the level of induction. A complete lack of induction was associated with no growth of the buried bud. Minimal induction led to the growth of long, thin shoots. Slightly stronger induction produced plagiotropic stolon like structures or else a leafy shoot with ancillary stolons. Stronger induction produced a thickened stolon and still stronger induction produced a stolon terminated by a tuber. Very strong induction produced a sessile tuber.

Madec and Perennec (1962) extensively studied the patterns of bud development on stem cuttings. The cuttings were classified into three types: 1) cuttings with early tuberization, 2) cuttings with retarded tuberization, and 3)

cuttings which showed early tuberization which was later reversed when cuttings came under non-inducing conditions.

The first type was the reaction of any cultivar under inductive conditions. It was characterized by reduced growth, little or no stem elongation after tuberization, little or no rooting, and tubers developing normally either on short stolons or flush in the leaf axil. The second type was the response of cultivars under long days or other conditions that were not inductive. Tuberization was inhibited until the critical photoperiod was past. Shoot growth and rooting were strong before tuberization and became weak or nil afterward. The cuttings formed long branched stolons, then tuberized with the tubers developing normally.

The third type occurred when cultivars were partially induced and then came under non-inductive conditions during which induction was lost. Cuttings showed early tuberization but then ceased enlarging and showed various types of regrowth, including new stolons that tuberized later. Shoot growth was weak before tuber initiation, strong when regrowth began, and continued until after the second tuberization.

MATERIALS AND METHODS

General Procedures

The experiments reported in this thesis were conducted from September 1980 through September 1982. They were all carried out in the controlled growth environment facilities at the University of Manitoba. One basic system was used throughout for the preparation and maintenance of mother plants and cuttings. Specific details varied from experiment to experiment.

Mother Plants

Mother plants were started from small whole tubers obtained from commercial seed potatoes grown the previous year. These tubers were visually selected for uniformity and ranged from 5 to 7 cm in diameter. Tubers were brought out of cold storage and green sprouted (22/17°C day/night; 18 hour day) for two weeks prior to planting. This was done to speed plant growth and development to compensate for limited growing space.

Plants were grown in a sand:peat:soil medium (1:1:1 volume) contained in 10-inch clay pots. They were watered as needed. One tablespoon (7-9 g) of commercially formulated 20-20-20 fertilizer was added to every 25 litres of medium when the tubers were initially planted. At weekly intervals thereafter, 500 mls of 10-52-10 fertilizer solution was added to each pot. (The 10-52-10 solution was made by dissolving 1 tablespoon of commercially formulated 10-52-10 in 4.5 litres of water.)

Mother plants were grown at 22/17°C day/night temperatures under 18 hours of

light provided by Gro-lux WS, VHO lamps. Photosynthetically active photon flux density (PPFD) at the top of the plant canopy averaged 260 $\mu E\ m^{-2}\ sec^{-1}$. All PPFD values given in this thesis were measured with a 'Li-Cor Li 185B' quantum meter.

Conditioning of Mother Plants

Cuttings were taken from two types of mother plants, conditioned and non-conditioned. All mother plants were started in the non-inducing conditions of long days (18h) and warm temperatures (22/17°C day/night). Conditioning treatments, when applied, consisted of exposure to short days (12h) and cool temperatures (17/11°C day/night) for two weeks prior to cuttings being taken. These plants were not fertilized during the conditioning period. Non-conditioned plants were grown under the long day, warm temperature regime and were fertilized as usual up until the cuttings were taken.

Preparation of Cuttings

Leaf bud cuttings, consisting of a leaf, petiole, stem section, and a single axillary bud, were taken from selected stems of mother plants. A typical leaf bud cutting is shown in Figure 1. For each treatment, mother plants were visually selected for uniformity and two or more similar stems were selected from each mother plant.

Each 'mother plant' in this thesis consists of a collection of stems all arising from the same mother tuber. Each stem arising from below ground is called a 'stem' which may actually be considered to be an individual plant (Moorby, 1967).

Leaf positions (nodes) were numbered from 1 to 8 starting from the top of the stem. Leaf position 1 was the youngest leaf on a stem with a leaf area greater than 30 square centimeters. This minimum leaf area was arbitrarily selected. Leaf position 8 was the oldest leaf used and was the eighth node below leaf position

Figure 1. A potato leaf bud cutting.
a - axillary bud. b - stem section.

one.

A razor blade was used to take cuttings. All cuts were made under water to ensure water continuity to the leaf. Cuttings were then placed in a sand:peat medium (1:1 volume) contained in wooden flats (inside dimensions 30cm x 45cm). The petiole was inserted into the soil so that the bud was well covered and the leaf was in an upright position. Twelve or sixteen cuttings were usually placed in each flat and were watered as required.

Leaf Area Measurements

In later experiments, leaf areas were determined for each leaf bud cutting at the time cuttings were taken. A leaf area meter, Model Li-3000, manufactured by the Lambda Instruments Corporation, was used for all measurements.

Overlapping leaflets on the cuttings introduced a certain amount of error into the measurement of leaf area. To obtain some idea of the magnitude of this error, fifty cuttings with varying amounts of overlap of the leaflets were run through the leaf area meter. Leaf areas were first determined on intact leaves, and then with the leaflets detached and separated so there was no overlap. Leaf area measurements in intact leaves were lower in all cases, with a range of 2% to 10.6% and an average of 4.9%. This error was not compensated for because there was no way of estimating error in measurement in any particular leaf without destroying the cutting. However, it may have made estimates of the effect of leaf area on tuber size less accurate, but not enough to invalidate them.

Harvesting

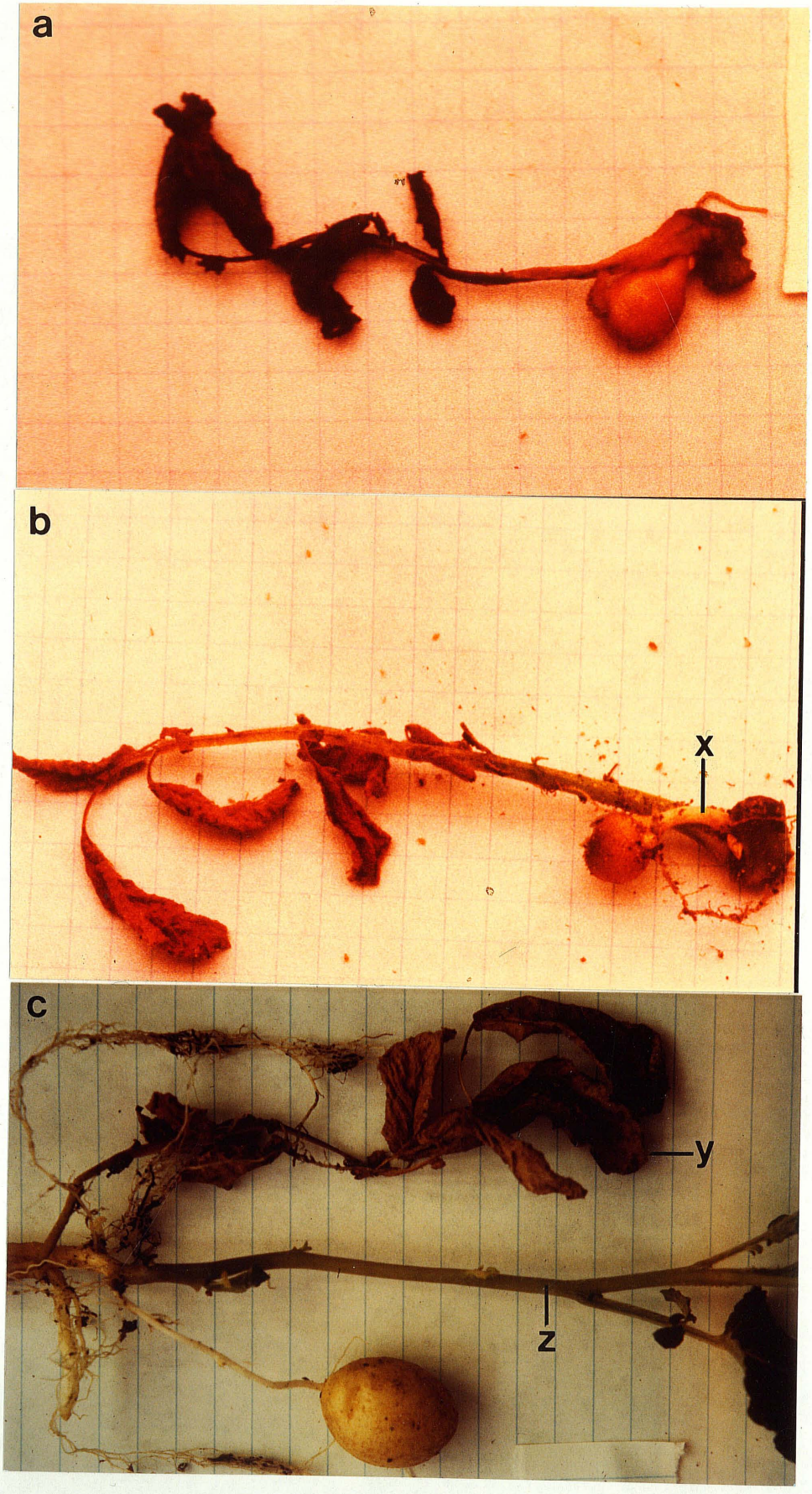
Cuttings were harvested when the majority of them had senesced. In a particular experiment, all cuttings were harvested on the same date unless otherwise noted. Fresh weights of the tubers were recorded. Types of bud development were noted and were grouped into four broad categories: Type 0 - no

Legend:

Figure 2a. Russet Burbank cutting showing typical Type 1 development.

Figure 2b. Russet Burbank cutting showing typical Type 2 development.
x - stolon.

Figure 2c. Russet Burbank cutting showing typical Type 3 development.
y - old leaf. z - new leafy shoot.


a
b
x
c
y
z

development of the axillary bud; Type 1 - tubers formed flush in the leaf axil (sessile tubers); Type 2 - the axillary bud formed a stolon, then a tuber, which was often accompanied by some rooting; Type 3 - the axillary bud formed a new leafy shoot and tubers were formed on stolons, which was usually accompanied by rooting. Types 1, 2 and 3 are shown in Figure 2.

Leaf Bud Cutting Environments

Cuttings in all the experiments were placed into one of four combinations of growing conditions. For each experiment, the cutting environments used were largely determined by space and conditions available at that particular time. These have been designated as Cutting Environments 1, 2, 3 and 4. Light in all cases was supplied by Gro-lux WS, VHO lights.

Cutting Environment 1 had a day/night temperature of 22/17°C, 18 hours of daylight, and a PPFD that approximately averaged 125 $\mu E\ m^{-2}\ sec^{-1}$. The rate of air movement across the plant canopy was not known, but it was low relative to that in Cutting Environments 2 and 3. Cutting Environment 2 had a day/night temperature of 17/11°C, 12 hours of daylight, and a PPFD that approximately averaged 350 $\mu E\ m^{-2}\ sec^{-1}$. The air speed across the leaf canopy was approximately 24 m/min unless otherwise noted. Cutting Environment 3 had a day/night temperature of 22/17°C, 18 hours of daylight and a PPFD that approximately averaged 350 $\mu E\ m^{-2}\ sec^{-1}$. Air speed across the leaf canopy was the same as in Cutting Environment 2. Cutting Environment 4 had a day/night temperature of 17/11°C, 12 hours of daylight, and a PPFD of approximately 250 $\mu E\ m^{-2}\ sec^{-1}$.

Supplemental Misting

An intermittant misting system was set up in Cutting Environments 2 and 3. Two misters, Model 707SM, manufactured by the Herrmidifier Co. Inc., were used in

each cutting environment. The mist applications were controlled by a time clock and were adjusted as needed. Relative humidity readings were taken by a Bacharach sling psychrometer, Model 12-7013, and, at 17°C, were found to average between 75 and 80 percent.

In initial experiments in Cutting Environment 2, there was severe wilting of the cuttings within 1 to 3 days after they were taken. There was limited tuber set (approximately 33% of cuttings set tubers) and the tubers produced were very small. The supplemental misting system minimized wilting and brought tuberization percentages up to nearly 100%. This supplemental misting system was in use in all experiments performed in Cutting Environments 2 and 3 reported in this thesis.

Details of Experiments

The following experiments have not been presented in chronological order, but in the order in which they will be discussed. For an overall presentation of the dates when experiments were performed, cutting environments, and cultivars used, refer to Table 9. Experiments 1 and 2 and Experiments 4 and 5 do not differ greatly in design or purpose, but they differ enough in other respects so that it is necessary to present them separately.

Experiment 1

The purpose of this experiment was to study the effects of leaf positions, mother plant age, conditioning of mother plants and cutting environments on bud development patterns and on sizes of tubers produced and to obtain some idea of possible interactions between these factors. An 8 x6 x2 x2 factorial arrangement was used in a completely randomized design. There were two replicates of each particular treatment. Observations were taken from 384 cuttings.

Cuttings were taken from 6 ages of Norland mother plants, ranging in age from

3 weeks to 8 weeks. Four plants of each age were selected. Two of these were conditioned and two were non-conditioned.

Two stems were selected from each plant. Eight leaf positions were used from each stem. Cuttings from one stem were randomized in flats in Cutting Environment 1 and cuttings from the other stem were randomized in Cutting Environment 2.

No leaf area measurements were taken in this experiment. When cuttings were harvested, types of bud development and fresh weights of the tubers were recorded.

Experiment 2

This experiment was very similar to Experiment 1. Norland cuttings were used to study the effects of leaf positions, mother plant age, conditioning of mother plants and cutting environments on bud development patterns and the sizes of tubers produced. An 8 x7 x2 x2 factorial arrangement was used in a completely randomized design. There were two replicates of each particular treatment. Observations were taken from 448 cuttings.

Cuttings were taken from 7 ages of mother plants, ranging from 4 weeks to 10 weeks. Four plants of each age were selected. Two of these were conditioned and two were non-conditioned.

Two stems were selected from each plant. Eight leaf positions were used from each stem. Cuttings from one stem were randomized in flats in Cutting Environment 1 and cuttings from the other stem were randomized in Cutting Environment 2.

Leaf area measurements were recorded at the time the cuttings were taken. Types of bud development and weights of the tubers were recorded when cuttings were harvested.

Experiment 3

Norland cuttings were used in this experiment to study the effects of leaf positions and three different cutting environments on bud development patterns and on the sizes of tubers produced. An 8 x3 factorial arrangement was used in a completely randomized design. There were four replicates of each particular treatment. Observations were taken from 96 cuttings.

Cuttings were taken from 5 week old non-conditioned mother plants. Four different plants were used with three stems being selected from each plant and randomly numbered. Eight leaf positions were used from each stem. Cuttings from stem 1 were randomized in flats in Cutting Environment 1, cuttings from stem 2 were randomized in flats in Cutting Environment 2, and cuttings from stem 3 were randomized in Cutting Environment 3. Leaf area measurements, types of bud development and weights of tubers were again recorded.

Experiment 4

The purpose of this experiment was to study the effects of leaf positions, mother plant age, conditioning of mother plants and cutting environments on bud development patterns and tuber size and to obtain some idea of possible interactions between these factors. An 8 x6 x2 x2 factorial arrangement was used in a completely randomized design. There were two replicates of each particular treatment. Observations were taken from 384 cuttings.

Cuttings were taken from 6 ages of Russet Burbank mother plants ranging from 5 weeks to 10 weeks. Four plants of each age were selected. Two of these were conditioned and two were non-conditioned.

Two stems were selected from each plant. Eight leaf positions were used from each stem. Cuttings from one stem were randomized in flats in Cutting Environment 1 and cuttings from the other stem were randomized in Cutting Environment 2.

No leaf area measurements were taken in this experiment. Types of bud development were recorded.

Experiment 5

The object of this experiment was to study the effects of leaf positions, mother plant age, conditioning of mother plants and cutting environments on bud development patterns and tuber sizes and to obtain some idea of the possible interactions between these factors. An 8 x6 x2 x2 factorial arrangement was used in a completely randomized design. There were two replicates of each particular treatment.

Cuttings were taken from 6 dates of Russet Burbank mother plants, ranging in age from 6 weeks to 11 weeks. Four plants of each age were selected. Two of these were conditioned and two were non-conditioned. Two stems were selected from each plant. Eight leaf positions from each stem were used. Cuttings from one stem were randomized in flats in Cutting Environment 2 and cuttings from the other stem were randomized in Cutting Environment 3.

Leaf area measurements were recorded at the time the cuttings were taken. Types of bud development and weights of the tubers were recorded when cuttings were harvested.

Experiment 6

In this experiment, cuttings were placed in Cutting Environment 3 with a day/night temperature of 22/17°C and 18 hours of daylight, but the rate of air flow across the leaf canopy and irradiances were modified. After 7 weeks, because of space problems, flats were transferred to Cutting Environment 1.

Cuttings were taken from 4 different 6 week old non-conditioned Russet Burbank mother plants. There were 96 cuttings in total. Three stems were

selected from each mother plant and 8 leaf positions were used from each stem. A completely randomized design was used.

The range of air flow rates across the leaf canopy that these cabinets are capable of producing is 10.5 - 24 m/min. In other experiments, the air flow rate was near the maximum or 24 m/min. In this experiment, the air flow rate would have been closer to the lower limit.

Light intensities were also modified. For the first week after the cuttings were taken, one-third of the lights were turned on, corresponding to a PPFD of approximately 130 μE m^{-2} sec^{-1}. At the beginning of the second week, two-thirds of the lights were turned on corresponding to a PPFD of approximately 230 μE m^{-2} sec^{-1}. At the beginning of the third week all the lights were turned on, corresponding to a PPFD of 320 μE m^{-2} sec^{-1}.

No leaf areas were determined for this experiment. Bud development types and tuber weights were recorded.

Experiment 7

Norland cuttings were used in this experiment to study the effect of adding fertilizer to the leaf bud cuttings. Age of the mother plants, conditioning of mother plants and leaf positions were also studied. An 8 x4 x2 x2 factorial arrangement was used in a completely randomized design. There were three replicates for each particular treatment. Observations were taken from 384 cuttings.

Cuttings were taken from 4 ages of mother plants, ranging from 4 weeks to 7 weeks. Six plants of each age were selected. Three of these were conditioned and three were non-conditioned.

Two stems were selected from each plant. Eight leaf positions were used from each stem. Cuttings from one stem were randomized in flats that were later fertilized. Cuttings from the other stem were randomized in flats as a control.

All cuttings were placed in Cutting Environment 1.

One week after cuttings were taken, 50 mls of 20-20-20 fertilizer solution (made by dissolving 1 tbsp of commercially formulated fertilizer in 4.5 litres of water) were added to the soil at the base of each cutting. Fifty mls of water were added to the soil at the base of the control cuttings. This treatment was repeated one week later, that is, two weeks after the cuttings were taken.

Leaf area measurements were recorded at the time the cuttings were taken. Types of bud development and tuber weights were recorded when the cuttings were harvested.

Experiment 8

The purpose of this experiment was to study the effect of adding fertilizer to the leaf bud cuttings. Leaf positions were also studied. An 8 x2 factorial arrangement was used in a completely randomized design. Two replicates per treatment were used.

Four stems were selected from a 4 week old non-conditioned Norland mother plant. Eight leaf positions were used from each stem. Two of the four stems were treated with fertilizer; the other two were designated as a control. Fertilizer treatments were applied exactly as described in Experiment 7. Cuttings were again placed in Cutting Environment 1.

Leaf areas were measured at the time the cuttings were taken. Types of bud development and tuber weights were recorded when cuttings were harvested.

Experiment 9

Three different experiments will be reported in this section - 9a, 9b and 9c. The purpose of these experiments was to assess the effect of enriching the soil zone of the cuttings with carbon dioxide. The methodology was nearly identical in all three experiments.

Two different types of flats were set up. Perforated polyethylene tubing was arranged in the bottom of the flats on top of a layer of gravel. A sand:soil:peat medium (1:1:1 volume) was placed on top of the tubing. Carbon dioxide was passed through the CO_2 treated flat at a rate of 10 mls/min. Air at 10 mls/min was passed through the tubing in the control flat.

Carbon dioxide readings were taken by a Series 225 Gas Analyzer, manufactured by the Analytical Development Co. Ltd. Readings were taken over several different days at different times of day and at different soil depths. Readings from the CO_2 treated flat ranged from 560 - over 1000 ppm. (The upper limit of detectability of the equipment was 1000 ppm.) Readings from the control flat ranged from 450 - 590 ppm.

In experiment 9a, cuttings were taken from 8 week old conditioned Russet Burbank mother plants. In experiment 9b cuttings were taken from 7 week old conditioned Russet Burbank mother plants. In experiment 9c, cuttings were taken from 7 week old non-conditioned Russet Burbank mother plants. All experiments were carried out in Cutting Environment 4.

The experiments were analyzed as completely randomized designs with 15 cuttings per treatment. Bud development types and tuber weights were recorded for all experiments.

Experiment 10

Three different experiments, 10a, 10b and 10c will be reported in this section. The purpose of these experiments was to study the effect of applying varying concentrations of ABA to the cuttings.

ABA solutions were prepared by dissolving ABA in 1 ml ethanol and diluting with distilled water to the desired concentration. The stems of the leaf bud cuttings were placed in the different ABA solutions and uptake was encouraged by placing them under high intensity lights for 4 hours. Cuttings were then placed in flats in

Cutting Environment 4.

In Experiment 10a, cuttings were taken from 6 week old conditioned Norland mother plants. Cuttings were treated with 0, 10, 20 and 50 mg/litre ABA solutions. There were 12 cuttings per treatment. A completely randomized design was used.

In experiments 10b and 10c, cuttings were taken from 6 week old conditioned Norland mother plants. Cuttings were treated with 0, 20, 50 and 100 mg/litre ABA solutions. There were 16 cuttings per treatment. A completely randomized block design was used in both experiments. Cuttings from each node were separated into statistical blocks.

Leaf areas were determined for Experiments 10b and 10c. Bud development types and tuber weights were noted for all experiments.

Statistical Analysis

All the data were analyzed following the standard procedure for analysis of variance. Square root transformations $x = \sqrt{x+.5}$ were done in most experiments on tuber weight (yield) data to reduce large variations observed within treatments. For purposes of analysis, where there was more than one tuber produced from a cutting, total tuber weight or yield was used. Where leaf area measurements were made, analysis of covariance was done with the leaf area data as the independent variable to assess the influence of leaf area on tuber yield. Duncan's multiple range test at the 5% level was used to test for differences between means. Comparisons have been presented with untransformed means.

RESULTS AND DISCUSSION

The experiments reported in this thesis do not lend themselves particularly well to groupings because there are many factors studied that are common to all experiments but no experiment is exactly identical to another. Because many factors studied are common to many experiments, the discussion of possible reasons for effects observed is very repetitious if done for each experiment. Therefore, except for Experiment 1, discussion in this section is limited and mostly covers points peculiar to a particular experiment. Common factors and the possible causes of some effects that are not immediately apparent are further discussed in the General Discussion section.

Norland

The experiments in this group were performed using the cultivar Norland, which is an early maturing variety. Effects of cutting environments, leaf positions,and bud development patterns were studied in all three experiments. Conditioning of mother plants and mother plant ages were studied in Experiments 1 and 2 but not in Experiment 3.

Experiment 1

The cuttings in this experiment were harvested after 7 weeks, when the majority of them had senesced. The average tuber size was 3.3 g and 80 percent of the cuttings produced tubers that were greater than 2 g, which is the minimum size of tuber that is able to be used for this method of rapid clonal propagation. (Rossnagel, 1980).

The purpose of this experiment was to study the effects of: 1)conditioning of

mother plants, 2)different mother plant ages, 3)different leaf positions and 4) different cutting environments on the sizes of tubers produced by cuttings. The interactions between these factors were of special interest. The effect of these factors on types of bud development were also noted.

The analysis of variance is shown in Table 10. It can be seen that the main effects of cutting environments , conditioning of mother plants, mother plant age and leaf positions were all significant at the .01 level. Some of the two way interactions were also significant. However, because the triple interaction between cutting environments, conditioning of mother plants and mother plant age (CE x C x A) is significant, the effects of these three factors can only be validly considered together. The main effects of these factors and the two way interactions in which these factors are involved are ignored and only the triple interaction is considered.

The effect of the different leaf positions was the only factor studied in this experiment that was not involved in interactions. It is therefore the only factor where the interpretation of the main effects is not modified by another factor.

A comparison of tuber sizes produced by cuttings from the different leaf positions is shown in Figure 3. Cuttings from leaf positions 1 and 2 produced the smallest tubers that differed from one another and from tubers produced by cuttings from the rest of the leaf positions. Tubers produced by cuttings from leaf positions 3 through 8 did not differ significantly in size. However, there was a tendency for cuttings from the middle leaf positions (4, 5 and 6) to produce larger tubers than cuttings from the older ones (7 and 8) and the younger ones (1, 2 and 3). No particular relationship was observed between bud development types and leaf position.

The reasons for the cuttings from the younger leaf positions producing significantly smaller tubers than the others may be partially due to leaf area effects

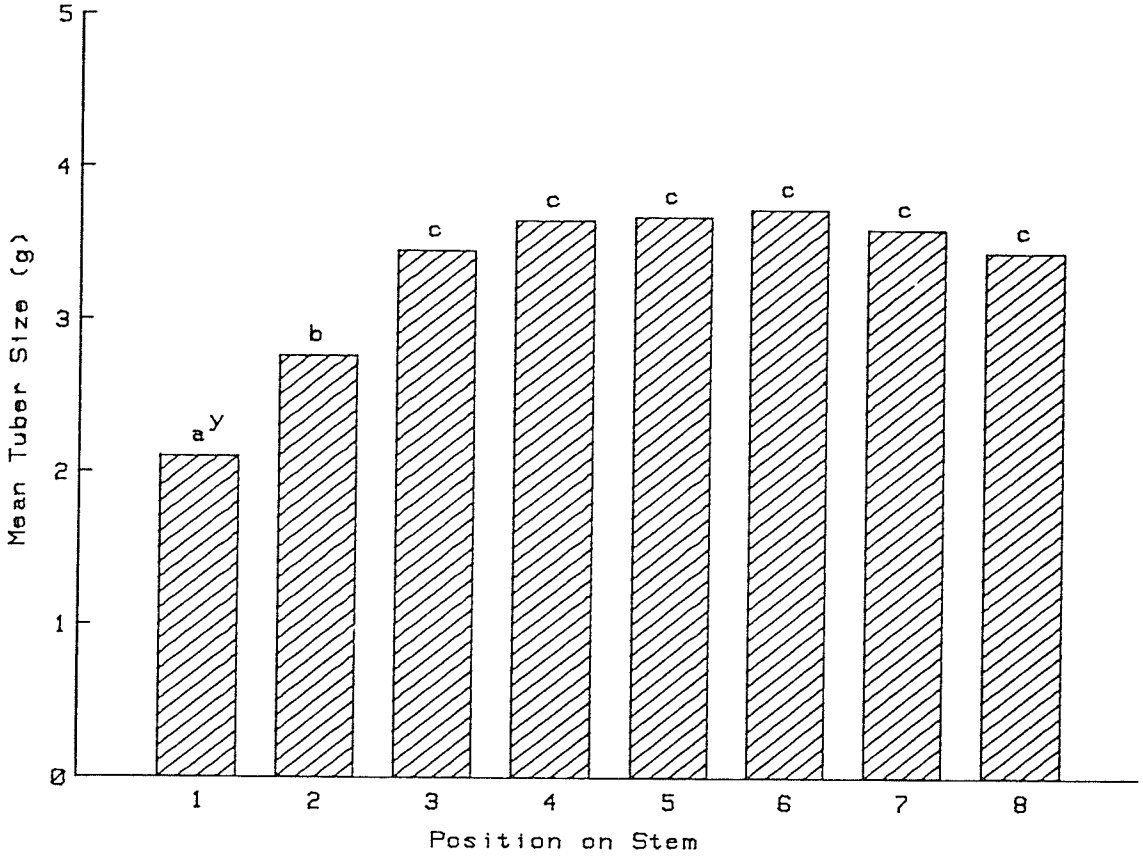


Figure 3. Comparison of tuber sizes produced by cuttings from leaf positions 1-8 in Experiment 1

[y]Values represented by bars with the same letter do not differ significantly by Duncan's Multiple Range test, .05 level. Values are means of 48 cuttings.

(not recorded in this experiment). Cuttings with physiologically young leaves may also have less ability to support tuberization. The tendency of cuttings from the older leaf positions to produce smaller tubers than cuttings from the middle leaf positions may be due to older leaves being less efficient and productive in terms of photosynthetic capacity.

The triple interaction between cutting environments, conditioning of mother plants and mother plant age is illustrated in Figures 4a and 4b.

The relationship between plant age and conditioning of mother plants for Cutting Environment 1 and Cutting Environment 2 is shown in Figure 4a. (Although both cutting environments are inductive in that tubers were formed in both, relatively speaking, Cutting Environment 2, in terms of temperature and photoperiod was more strongly inductive than Cutting Environment 1. Therefore, Cutting Environment 2 will be referred to as 'inductive' and Cutting Environment 1 as 'non-inductive'.) In the non-inductive Cutting Environment 1, tubers produced by cuttings from conditioned mother plants were significantly larger than those from non-conditioned mother plants for all plant ages except for the oldest (8 weeks). Differences in tuber size between conditioned and non-conditioned treatments decreased with increasing plant age. In the inductive Cutting Environment 2, tubers produced by cuttings from conditioned and non-conditoned mother plants, did not differ in size except for those from the 6 and 7 week old mother plant ages. These differences were not great and are perhaps a manifestation of leaf area effects or problems in the selection of mother plants or stems, which will be discussed later.

The relationship between mother plant age and cutting environments for conditioning and non-conditioning of mother plants is shown in Figure 4b. For both conditioned and non-conditioned mother plants, cuttings from the three youngest plant ages placed in Cutting Environment 1 produced significantly larger tubers than

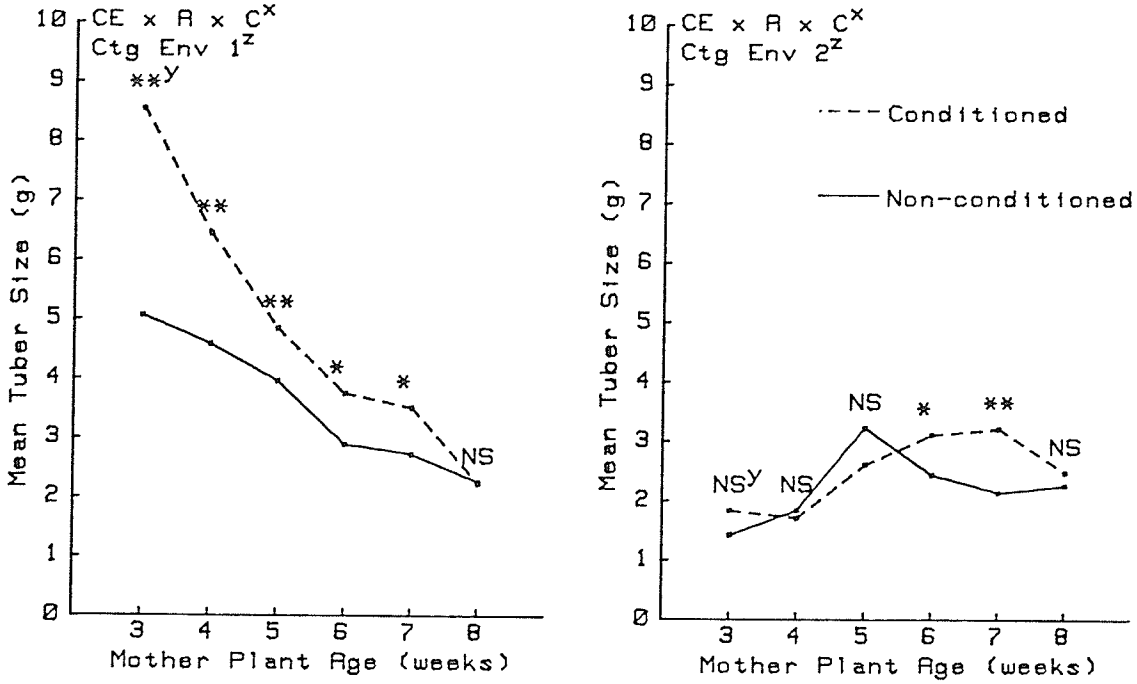


Figure 4a. Triple interaction between cutting environments, conditioning and mother plant age in Experiment 1. Relationship between plant age and conditioning for cutting environments 1 and 2.

[x]CE = cutting environments; A = mother plant age; C = conditioning of mother plants.

[y]Comparisons between sizes of tubers produced by cuttings (conditioned vs non-conditioned) from the same age of mother plants. * and ** indicate significance at the .05 and .01 levels respectively. NS indicates non-significance.

[z]Ctg Env 1 = Cutting Environment 1 (22/17°C; 18h; low PPFD; low air flow); Ctg Env 2 = Cutting Environment 2 (17/11°C; 12h; high PPFD; high air flow).

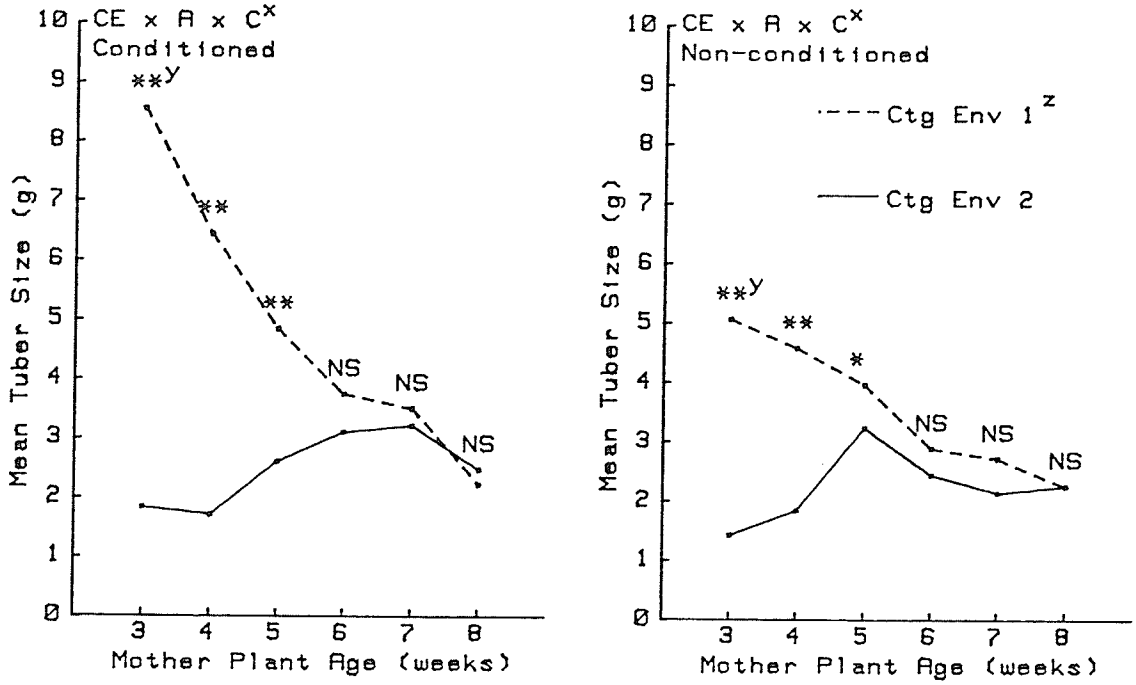


Figure 4b. Triple interaction between cutting environments, conditioning and mother plant age in Experiment 1. Relationship between plant age and cutting environments for conditioning and non-conditioning of mother plants.

[x]CE = cutting environments; A = mother plant age; C = conditioning of mother plants.

[y]Comparisons between sizes of tubers produced by cuttings (cutting environment 1 vs cutting environment 2) from the same age of mother plants. * and ** indicate significance at the .05 and .01 levels respectively. NS indicates non-significance.

[z]Ctg Env 1 = Cutting Environment 1 (22/17°C; 18h; low PPFD; low air flow); Ctg Env 2 = Cutting Environment 2 (17/11°C; 12h; high PPFD; high air flow).

cuttings placed in Cutting Environment 2. Differences decreased with increasing mother plant age with conditioned plants showing greater differences than non-conditioned. For the three oldest plant ages (6, 7 and 8 weeks), it didn't make any difference to the sizes of tubers produced, whether cuttings were placed in Cutting Environment 1 or Cutting Environment 2. Possible reasons for these effects will be discussed later, after bud development types are presented.

The influence of mother plant age, conditioning of mother plants and cutting environments on the numbers of cuttings exhibiting the different types of bud development are shown in Figure 5.

The response of cuttings taken from conditioned mother plants and placed in non-inductive Cutting Environment 1 is shown in Figure 5a. The cuttings from the youngest (3 weeks) mother plants all showed Type 3 development. The numbers of cuttings showing Type 2 plus Type 3 development tended to decrease with increasing plant age while the numbers of cuttings showing Type 1 development increased with increasing plant age. The cuttings from the oldest (8 weeks) plants all showed Type 1 development.

This effect of plant age is exactly what would be expected. Tubers formed flush in the leaf axil (sessile tubers) are thought to be associated with strong induction or a high level of the tuberization stimulus. With increasing mother plant age, one would expect increasing amounts of the tuberization stimulus to be present and therefore a greater amount of sessile tuberization.

The same trend for the effect of mother plant age on bud development types is also seen in Figures 5b and 5c. However, for the situations represented by these graphs, the transition of Type 3 to Type 1 is more abrupt and happens at a younger plant age than for conditioned plants in Cutting Environment 1. The reason for the responses of the cuttings to mother plant age from non-conditioned plants placed in Cutting Environment 2 (Figure 5d) is not known. It is likely that the effect of the 5

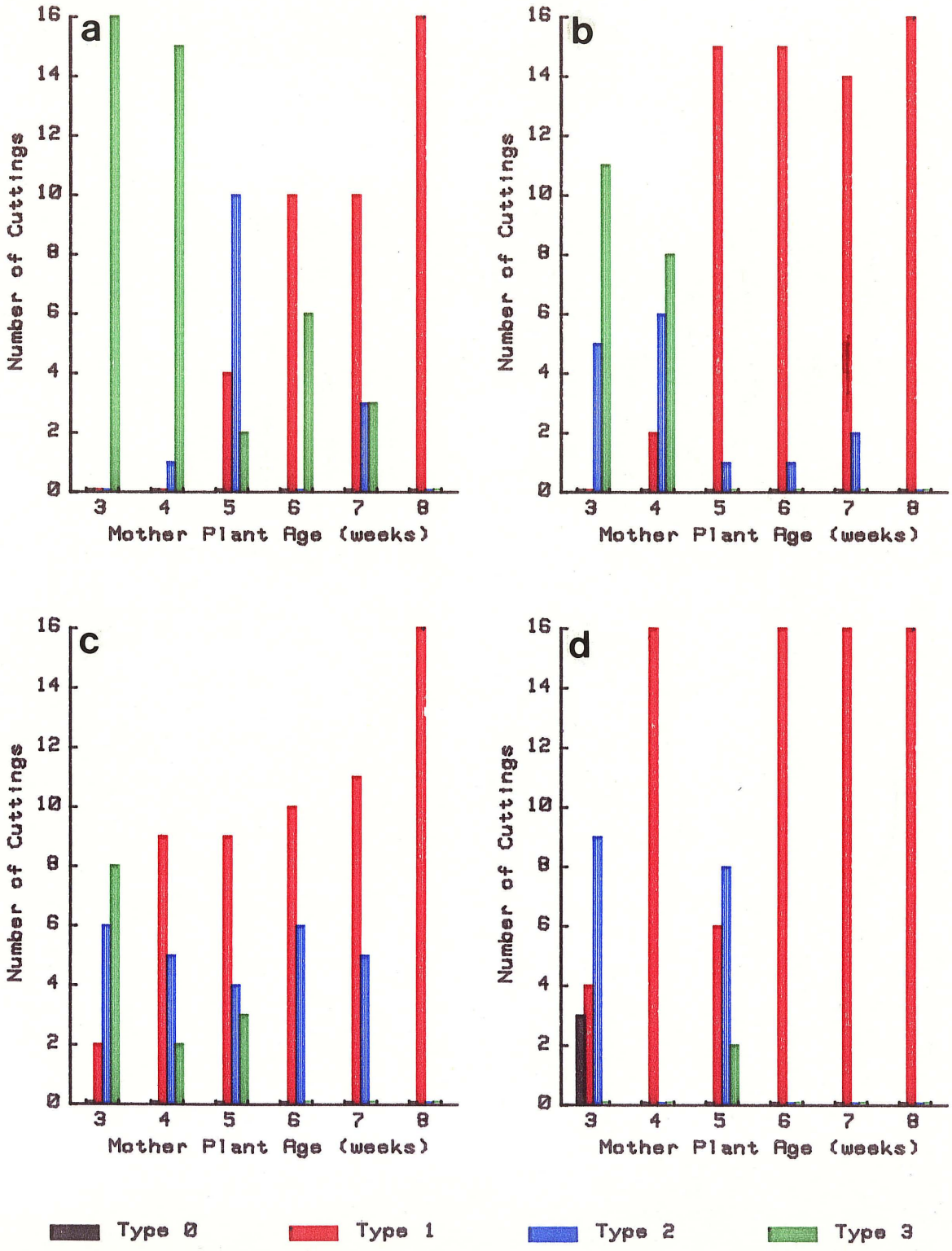
Legend:

Figure 5. The influence of mother plant age, conditioning of mother plants and cutting environments on the numbers of cuttings exhibiting the different types of bud development in Experiment 1.

Type 0 = no bud development
Type 1 = tuber formed from bud
Type 2 = tuber formed on stolon
Type 3 = tuber formed on stolon from new shoot

a - Cutting Environment 1; conditioned mother plants.
b - Cutting Environment 1; non-conditioned mother plants.
Cutting Environment 1 (22/17°C; 18h; low PPFD; low air flow)

c - Cutting Environment 2; conditioned mother plants.
d - Cutting Environment 2; non-conditioned mother plants.
Cutting Environment 2 (17/11°C; 12h; high PPFD; high air flow)


a
b
c
d
Number of Cuttings
Mother Plant Age (weeks)
0
2
4
6
8
10
12
14
16
3
4
5
6
7
8
Type 0
Type 1
Type 2
Type 3

week old mother plants is an anomaly.

The effect of Cutting Environments 1 and 2 on the numbers of cuttings showing the different types of bud development can be seen by comparing Figure 5a with 5c and 5b with 5d. The non-inductive conditions of Cutting Environment 1 appeared to favor Type 3 development and the inductive conditions of Cutting Environment 2 appeared to favor relatively more Type 1. One would expect that Cutting Environment 2, with its inductive temperature and photoperiod, would increase the level of induction in the cuttings which would result in more sessile tuberization.

The effect of conditioning mother plants on bud development patterns can be seen by comparing Figure 5a with 5b and 5c with 5d. Conditioning of mother plants appeared to favor more leafy shoot development and stolon elongation followed by tuber set (Type 3 development). This is not what would be expected with current theories of tuberization in cuttings because conditioning of mother plants corresponds with a higher level of induction and one would therefore expect an increase in the amount of Type 1 development. From these results, one might speculate that tuber formation may occur in two phases. Conditioning of mother plants induced stolon elongation, with the subsequent inductive cutting environment resulting in tuber set. In the absence of mother plant conditioning, tubers may be formed directly from axillary buds without intervening stolon development.

Table 1 shows mean tuber sizes produced by cuttings exhibiting the different types of bud development, regardless of what treatment or factor combinations were involved. The largest tubers were obtained from Type 3 development, that is, tubers formed on stolons with new leafy shoot development and, usually, extensive rooting. The next largest tubers were produced from Type 2 development, with tubers being formed on stolons, usually accompanied by some rooting. The smallest tubers were produced by Type 1 development, with tubers being formed flush in the leaf axil with little, if any, rooting. This is very logical because cuttings with new

Table 1. Mean tuber sizes produced by cuttings and percentage of cuttings exhibiting the different types of bud development in Experiment 1.

Type	Mean Tuber Size (g)	% of Cuttings
0	0.00	0.8
1	2.35	60.4
2	3.59	18.8
3	6.06	19.8

leafy shoots and extensive rooting would have a much greater potential for production of photosynthate and uptake of water and mineral nutrients needed for tuber enlargement than cuttings with no shoot development and no rooting. The tubers from cuttings with no new shoot and no rooting would rely on the original leaf for the production of photosynthate and would have a limited water and nutrient supply. The main source of nutrients for these tubers would be the original leaf itself. One would expect immediate migration of nitrogen, phosphorous and potassium from the leaf to the tuber which would speed leaf senescence and necessarily limit tuber enlargement.

Highest tuber yields in intact plants are achieved with a balance of factors favoring both vegetative growth and tuber initiation (Ewing, 1981). This provides enough vegetative growth to support tuber enlargement and enough tuberization stimulus present for tuber initiation and continued tuber enlargement. It seems likely from the relationship between types of bud development and tuber yields that the same situation holds for obtaining tubers from leaf bud cuttings.

The highest yields in this experiment were obtained from cuttings from young conditioned plants placed in the non-inductive Cutting Environment 1. It is likely that this gave the best balance of factors promoting vegetative growth and tuber initiation. Cutting Environment 1 would have favored vegetative growth and conditioning of mother plants would have ensured that there was a good level of the tuberization stimulus present. Young plants would have provided a greater potential for vegetative growth than old plants. The next highest yields were obtained from cuttings from young non-conditioned plants placed in Cutting Environment 1. The lower yields from the cuttings from young non-conditoned plants may be due to slightly lower levels of tuberization stimulus present.

In the inductive Cutting Environment 2, in general, tuber yields were low whether cuttings were from conditioned or non-conditioned plants. This may be

due to high levels of induction, resulting in rapid tuber set, with limited potential for vegetative growth, and consequently, tuber enlargement. In Cutting Environment 1, for cuttings from the oldest plant age (8 weeks), it didn't make any difference whether mother plants were conditioned or not. This may be due to the tuberization stimulus already being present to such an extent in the 8 week old plants that conditioning didn't make any further difference. Cuttings from both conditioned and non-conditioned plants from this plant age would have tuberized immediately, forming sessile tubers with little potential for tuber enlargement.

Other differences in environmental factors in Cutting Environment 1 and Cutting Environment 2 may have also influenced development of cuttings. In terms of temperature and photoperiod alone, Cutting Environment 1 was non-inductive and Cutting Environment 2 was inductive. However, in Cutting Environment 1, low irradiance was coupled with long days and a low rate of air flow, while Cutting Environment 2 had high irradiance, short days and a high rate of air flow. It is therefore possible that the difference between the sizes of tubers produced by Cutting Environment 1 and Cutting Environment 2 may be partly due to differences in cutting establishment although there was no visible differences in wilting between the two cutting environments. For the three oldest plant ages, it didn't make any difference if the cuttings were placed in Cutting Environment 1 or in Cutting Environment 2. This could be because the tuberization stimulus was present so strongly that it didn't matter if conditions favored vegetative growth or not. The cuttings would have tuberized immediately, forming sessile tubers with little potential for tuber enlargement. For the three youngest plant ages, where the tuberization stimulus was present less strongly, when the cuttings were placed in Cutting Environment 1, which favored vegetative growth, the cuttings underwent Type 3 development, which has good potential for tuber enlargement.

Experiment 2

The cuttings in this experiment were harvested after 7 weeks, as in Experiment 1. The average tuber size was 2.92 g and 71.2 percent of the cuttings produced tubers larger than 2 g.

As in Experiment 1, the purpose of this experiment was to study the effects of: 1)conditioning of mother plants, 2)different plant ages, 3)different leaf positions and 4)different cutting environments on the sizes of tuber produced by cuttings and on types of bud development. The interactions between these factors were again of special interest. The effect of leaf area of the cuttings on the sizes of tubers produced was also studied.

This experiment differs from Experiment 1 in that more mother plant ages and a different range of mother plant ages were used. There is also no guarantee that a 4 week old mother plant in this experiment is exactly the same physiological age as 4 week old plants in Experiment 1, because plants were grown from different ages of tubers and at different times.

The analysis of variance is shown in Table 11. It can be seen that the same effects and interactions were significant in this experiment as were in Experiment 1. Again, the triple interaction between cutting environments, conditioning of mother plants and mother plant ages (CE x C x A) is of special interest. The effect of the different leaf positions was again the only factor studied that was not involved in interactions.

A comparison of tuber sizes produced by cuttings from the different leaf positions is shown in Figure 6. The effect of leaf positions is fairly similar to that in Experiment 1. Leaf position 1 produced the smallest tubers that differed significantly from the tubers from the rest of the leaf positions. Cuttings from the youngest leaf positions (1, 2, and 3), and the oldest (7 and 8) again tended to produce smaller tubers than the middle leaf positions (4, 5 and 6). No relation was

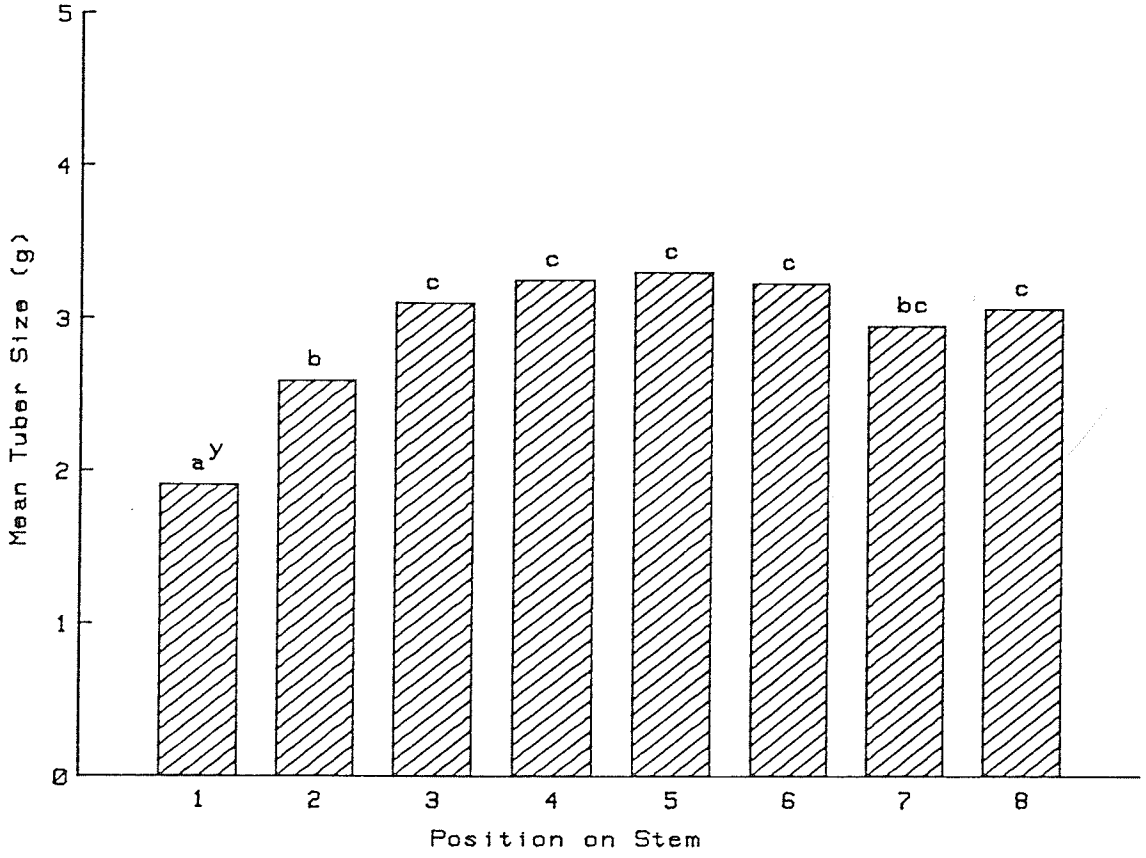


Figure 6. Comparison of tuber sizes produced by cuttings from leaf positions 1-8 in Experiment 2

[y] Values represented by bars with the same letter do not differ significantly by Duncan's Multiple Range test, .05 level. Values are means of 56 cuttings.

found between bud development types and leaf position.

The triple interaction between cutting environments, conditioning of mother plants and mother plant age is illustrated in Figures 7a and 7b.

The relationship between plant age and conditioning of mother plants for Cutting Environment 1 and Cutting Environment 2 is shown in Figure 7a. In the non-inductive Cutting Environment 1, tubers produced by cuttings from conditioned mother plants were significantly larger than those from non- conditioned plants for all ages except for 6 weeks, 7 weeks and 10 weeks. The largest differences in tuber size between conditioned and non-conditioned treatments were found at the youngest plant age (4 weeks). In the inductive Cutting Environment 2, tuber yields produced by cuttings from conditioned mother plants were not significantly different from those from non-conditioned plants, except for the 5 and 8 week old mother plant ages. The differences between tuber yields from cuttings from conditioned and non-conditioned plants for the 5 and 8 week old plant ages were not great.

The relationship between mother plant age and cutting environments for conditioned and non-conditioned mother plants is shown in Figure 7b. For the conditioned mother plants, cuttings placed in the non-inductive Cutting Environment 1 produced significantly larger tubers than cuttings placed in the inductive Cutting Environment 2 for all plant ages except for the 7 and 10 week old mother plant ages. The largest differences in tuber yield from cuttings placed in Cutting Environment 1 as compared to Cutting Environment 2 was for the youngest plant age (4 weeks). For the non-conditioned mother plants, it didn't make any difference to tuber yield whether cuttings were placed in Cutting Environment 1 or 2, except for cuttings from the youngest mother plant age.

The influence of mother plant age, conditioning of mother plants and cutting environments on the numbers of cuttings exhibiting the different types of bud

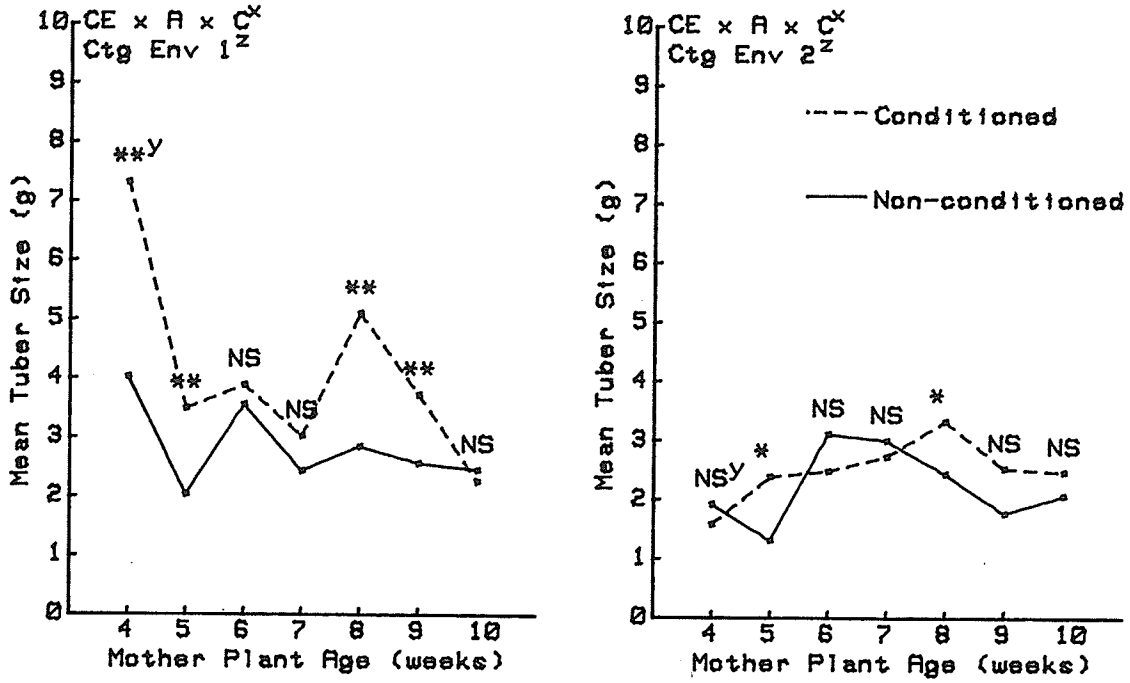


Figure 7a. Triple interaction between cutting environments, conditioning and mother plant age in Experiment 2. Relationship between plant age and conditioning for cutting environments 1 and 2.

[x]CE = cutting environments; A = mother plant age; C = conditioning of mother plants.

[y]Comparisons between sizes of tubers produced by cuttings (conditioned vs non-conditioned) from the same age of mother plants. * and ** indicate significance at the .05 and .01 levels respectively. NS indicates non-significance.

[z]Ctg Env 1 = Cutting Environment 1 (22/17 C; 18h; low PPFD; low air flow); Ctg Env 2 = Cutting Environment 2 (17/11 C; 12h; high PPFD; high air flow.)

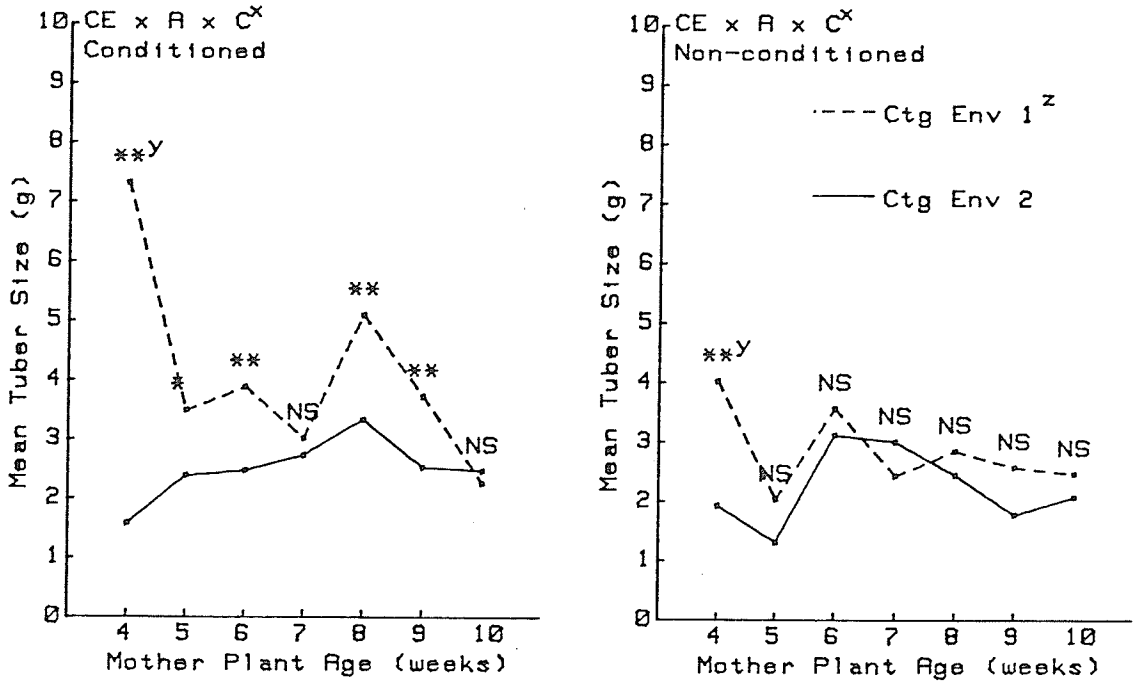


Figure 7b. Triple interaction between cutting environments, conditioning and mother plant age in Experiment 2. Relationship between plant age and cutting environments for conditioning and non-conditioning of mother plants.

[x]CE = cutting environments; A = mother plant age; C = conditioning of mother plants.

[y]Comparisons between sizes of tubers produced by cuttings (cutting environment 1 vs cutting environment 2) from the same age of mother plants. * and ** indicate significance at the .05 and .01 levels respectively. NS indicates non-significance.

[z]Ctg Env 1 = Cutting Environment 1 (22/17°C; 18h; low PPFD; low air flow); Ctg Env 2 = Cutting Environment 2 (17/11°C; 12h; high PPFD; high air flow).

development are shown in Figure 8. The response by the cuttings for bud development types is very similar to that seen in Experiment 1. The major difference is that there is more Type 1 in this experiment which is exactly what would be expected because it involves an older range of mother plants. Again, a transition from Type 3 development to Type 1 can be seen with increasing mother plant age.

The effect of Cutting Environments 1 and 2 on the numbers of cuttings exhibiting the different types of bud development types can be seen by comparing Figure 8a with 8c and 8b with 8d. The non-inductive Cutting Environment 1 would appear to favor Type 3 development.

The effect of conditioning mother plants on bud development types can be seen by comparing Figure 8a with 8b and 8c with 8d. Conditioning of mother plants would appear to favor more Type 2 and Type 3 development and less Type 1. Conditioning also prolonged the transition of Type 3 and Type 2 to Type 1 across the plant ages. This, again, is not exactly what would be expected and the reason for it is not known.

Table 2 shows mean tuber sizes produced by cuttings exhibiting the different types of bud development, regardless of what treatment or factor combinations were involved. As in Experiment 1, the largest tubers were produced by the cuttings showing Type 3 development. Cuttings exhibiting Type 2 development produced an intermediate size of tubers. Type 1 development produced the smallest tubers.

The effects of cutting environments, conditioning of mother plants and mother plant age on sizes of tubers produced are much more irregular than in Experiment 1. However, many of the overall effects are similar. One would expect that there were some factors exerting an influence here that were not exerting such an effect in Experiment 1. This experiment does deal with a range of older plant ages. The

Legend:

Figure 8. The influence of mother plant age, conditioning of mother plants and cutting environments on the numbers of cuttings exhibiting the different types of bud development in Experiment 2.

Type 0 = no bud development
Type 1 = tuber formed from bud
Type 2 = tuber formed on stolon
Type 3 = tuber formed on stolon from new shoot

a - Cutting Environment 1; conditioned mother plants.
b - Cutting Environment 1; non-conditioned mother plants.
Cutting Environment 1 (22/17°C; 18h; low PPFD; low air flow)

c - Cutting Environment 2; conditioned mother plants.
d - Cutting Environment 2; non-conditioned mother plants.
Cutting Environment 2 (17/11°C; 12h; high PPFD; high air flow)

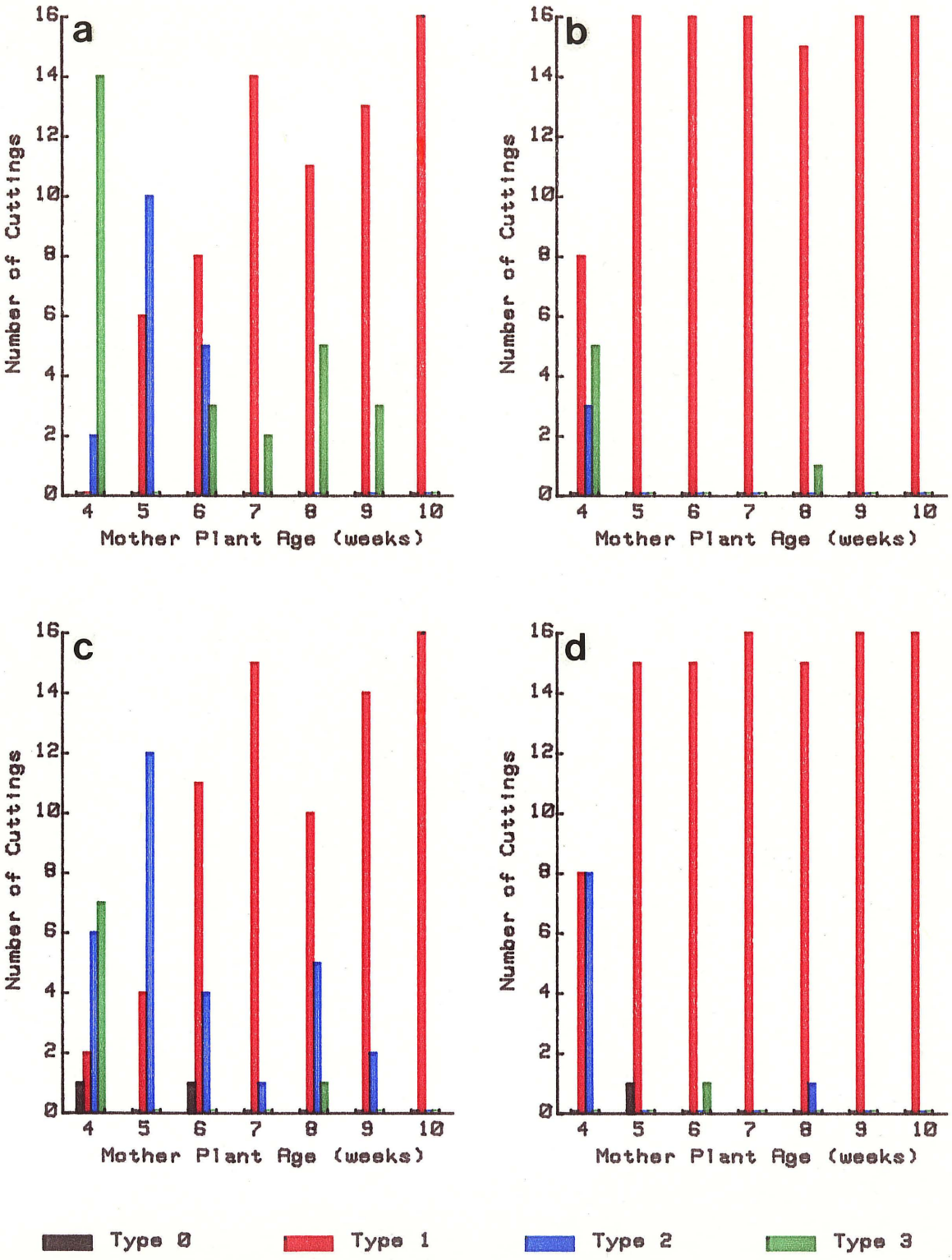

a
b
c
d
Number of Cuttings
Mother Plant Age (weeks)
0
2
4
6
8
10
12
14
16
4
5
6
7
8
9
10
Type 0
Type 1
Type 2
Type 3

Table 2. Mean tuber sizes produced by cuttings and percentage of cuttings exhibiting the different types of bud development in Experiment 2.

Type	Mean Tuber Size (g)	% of Cuttings
0	0.00	0.7
1	2.57	76.8
2	3.27	13.2
3	5.52	9.4

irregularities in the response of cuttings may be due to leaf area effects or to problems in the selection of similar mother plants and stems. (See General Discussion.)

Leaf areas were recorded in this experiment. Analysis of covariance was performed with the leaf area data. The 'r' value for the error term was significant at the .01 level ($r=+0.52$) which indicates that there is an underlying relationship between tuber size and leaf area. This means that leaf area was exerting an effect on tuber size no matter what 'treatment' was being applied or if any 'treatment' was applied. Differences in leaf area can account for approximately 27 percent of the unexplained variation in this experiment.

Because of the design of the experiment, 'r' values could not be determined for all factors but they could be determined for dates and positions and were found to be significant for both. Regression coefficients were computed, then tested for significance on tuber size (yield) data after adjustment for leaf area. For both dates and positions, 'F' values were smaller than for the analysis of original data which indicates that the differences in leaf areas were actually the cause of part of the differences between mother plant age and leaf position effects on tuber yields. When differences between yields of cuttings from different plant ages and leaf positions were tested as if all leaf areas were equalized, the 'F' values were still significant which means that differences in yield between dates and leaf positions were not solely due to differences in leaf areas.

It was decided not to correct tuber size data for leaf area, but leaf areas may account for some of the irregularities observed in the plant age data. For example, cuttings from the 5 week old plants unexpectedly supported the least amount of tuberization. Leaf areas for the 5 week old plants were also much smaller than for any of the other plant ages.

Experiment 3

The purpose of this experiment was to study the effects of leaf positions and three different cutting environments on bud development types and on the sizes of tubers produced. Only one age of mother plant (5 weeks old) was used and all plants were non-conditioned. Cuttings from Cutting Environments 2 and 3 were harvested after 5 weeks when the majority of them had senesced. Cuttings from Cutting Environment 1 were harvested after 9 weeks.

The analysis of variance is shown in Table 12. The differences between the three different cutting environments were highly significant. Differences between plants were not significant, which was expected because the plants were all the same age and were all non-conditoned. The differences between leaf positions were also not significant.

The percentage of cuttings exhibiting the different patterns of bud development and mean tuber sizes in each of the three cutting environments are shown in Table 3. In Cutting Environment 1, 100 percent of the cuttings tuberized, 97 percent tuberized in Cutting Environment 2, and only 50 percent of the cuttings formed tubers in Cutting Environment 3. The cuttings in Cutting Environment 1 produced the largest tubers, while the cuttings in Cutting Environment 2 produced smaller tubers and the ones in Cutting Environment 3 produced only very small tubers.

The three different Cutting Environments represent different combinations of conditions. Cutting Environment 1 had relatively low irradiance, high temperature, long photoperiod and a low rate of air movement across the leaf canopy. Relative humidity was slightly lower than in Cutting Environments 2 and 3. Cutting Environment 2 had a relatively high irradiance, low temperature, short photoperiod, high humidity and high rate of air flow across the leaf canopy. Cutting Environment 3 had relatively high irradiance, high temperature, long photoperiod,

Table 3. Mean tuber sizes produced by cuttings and percentage of cuttings exhibiting the different types of bud development in the different cutting environments in Experiment 3.

Type		Mean Tuber Size (g)	% of Cuttings
0	CE 1[y]	--	0.0
	CE 2	0.00	3.1
	CE 3	0.00	50.0
1	CE 1	2.43	25.0
	CE 2	1.42	43.8
	CE 3	0.52	12.5
2	CE 1	6.09	65.6
	CE 2	2.73	37.5
	CE 3	0.52	12.5
3	CE 1	8.12	9.4
	CE 2	2.06	15.6
	CE 3	0.79	25.0

[y]CE = Cutting Environment.

[z]% of Cuttings = percentage of cuttings wihin each cutting environment having a specific type of bud development.

high humidity and high air flow across the leaf canopy. These factors are completely confounded. It is impossible to tell exactly what the different effects of the Cutting Environments would have been on cutting establishment, vegetative growth, tuber initiation and tuber enlargement, because there are factors favoring each process in each Cutting Environment.

It is likely that the differences between rooms is partially due to differences in cutting establishment. There didn't appear to be any visible differences in the degree of wilting between Cutting Environments 1 and 2, but there was more wilting in Cutting Environment 3.

However, the effect of Cutting Environment 3 would not seem to be just in being unfavorable for cutting establishment. The highest percentage of Type 3 development was in Cutting Environment 3. Often new leafy shoots were formed with no tubers or with very tiny tubers. This would indicate a lack of the tuberization stimulus (Ewing, 1981), which may also partly account for the high percentage of cuttings with no bud development. The cuttings were taken from fairly young, non-conditioned plants. These plants may have been reversibly induced and Cutting Environment 3 may have caused the induction to be reversed. If this was the case, one would suppose that conditions in Cutting Environments 1 and 2 did not cause the induction to be reversed.

In Experiments 1 and 2, the cuttings exhibiting Type 3 bud development produced the largest tubers. In this experiment, this was the case in Cutting Environments 1. In Cutting Environment 2, Type 2 development produced the largest tubers. This reflects the problem with Type 3 development sometimes producing only very tiny tubers or no tubers at all which would indicate a lack of tuberization stimulus.

Cutting Environment 1 seemed to favor vegetative growth in comparison to Cutting Environment 2. Cutting Environment 2 had a much higher percentage of

sessile tuberization. Cutting Environment 1 would appear to have the best combination of conditions for the production of large tubers for this particular age of mother plant. It should be remembered that these cuttings were taken from relatively young non-conditioned plants and that cuttings from older, conditioned plants may react quite differently to these cutting environments. It is apparent that different cutting environments can drastically affect the responses of cuttings in terms of bud development patterns and sizes of tubers produced.

Analysis of covariance was done on leaf area data to see if leaf areas of the cuttings exerted any effect on the sizes of tubers produced. The 'r' value for the error term was small ($r=+0.23$) and was not significant. This means that, in this experiment, there was no underlying relationship between leaf area and size of tubers produced. This is perhaps not surprising, especially because of the large numbers of tubers that exhibited no bud development at all.

Russet Burbank

Russet Burbank, a late maturing cultivar, was used in Experiments 4, 5 and 6. Effects of leaf position and bud development patterns were studied in all three experiments. Cutting environments, conditioning of mother plants, and mother plant age were studied in Experiments 4 and 5 but not in Experiment 6. In Experiment 6, only one age of mother plant was used and the cutting environment was modified with respect to irradiance and air flow. Experiments 4 and 5 differ in mother plant ages and cutting environments used.

Experiment 4

The cuttings were harvested after 5 weeks, when the majority of them had senesced. The average tuber size was 1.78 g and 32 percent of the cuttings produced tubers that were greater than 2 g. In terms of usable tubers produced, this experiment was less successful than Experiments 1 and 2.

The purpose of this experiment was to study the effects of: 1)mother plant age, 2)conditioning of mother plants, 3)leaf positions and 4)cutting environments on the sizes of tubers produced and on bud development patterns. The interactions between these factors were of special interest.

The analysis of variance is shown in Table 13. It can be seen that the main effects of cutting environments, conditioning of mother plants, mother plant age and leaf positions were all significant at the .01 level. The interactions between cutting environments and mother plant age, between cutting environments and leaf positions, and between conditioning of mother plants and mother plant age were also significant at the .01 level. It is therefore these interactions that are primarily of interest. In this experiment all factors studied were involved in interactions.

The interaction between conditioning of mother plants and mother plant age is shown in Figure 9. For the oldest mother plant ages, cuttings from conditioned mother plants produced the largest tubers. For the two youngest plant ages, cuttings from the non-conditioned mother plants produced significantly larger tubers than cuttings from conditioned plants. This is unexpected in terms of what is known of the tuberization stimulus and the reason for this effect is not known.

The interaction between cutting environments and mother plant age is shown in Figure 10. Cuttings from all plant ages in the non-inductive Cutting Environment 1 produced significantly larger tubers than those in the inductive Cutting Environment 2. In Cutting Environment 1, cuttings from the seven week old mother plants produced the largest tubers and tuber size decreased with increasing plant age and also with decreasing plant age. This may be due to the fact that the 7 week old plants had the optimum balance of factors favoring both tuberization and vegetative growth. The younger plants may lack a good level of the tuberization stimulus; the older plants may have the tuberization stimulus present so strongly that vegetative growth is reduced and tuber size is necessarily limited. In Cutting

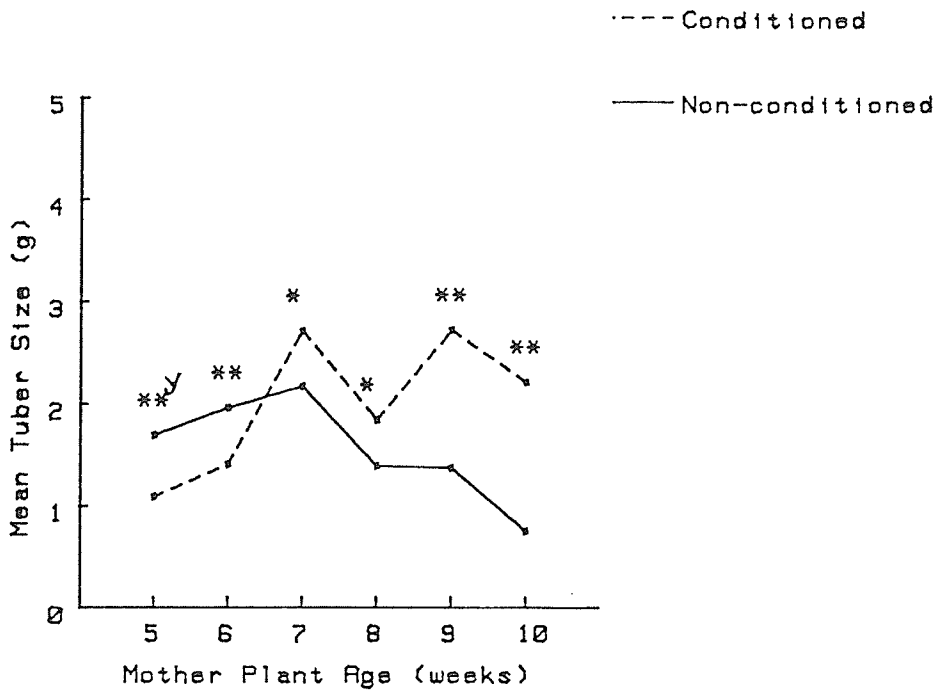


Figure 9. Interaction between conditioning of mother plants and mother plant ages in Experiment 4.

[y] Comparisons between sizes of tubers produced by cuttings (conditioned vs non-conditioned) from the same age of mother plants; * and ** indicate significance at the .05 and .01 levels respectively.

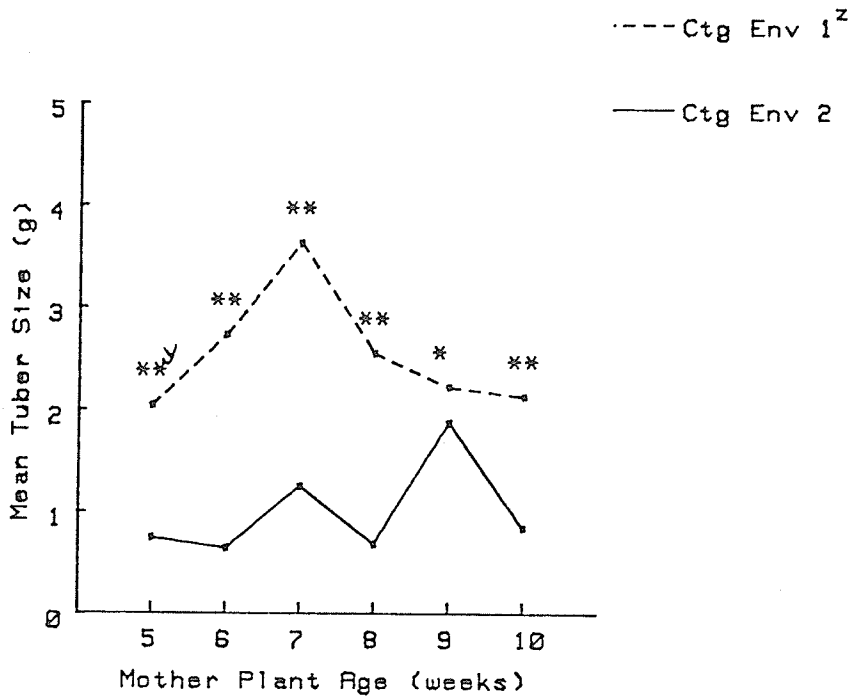


Figure 10. Interaction between cutting environments and mother plant ages in Experiment 4.

[y] Comparisons between sizes of tubers produced by cuttings (Cutting Environment 1 vs Cutting Environment 2) from the same age of mother plants. * and ** indicate significance at the .05 and .01 levels respectively.

[z] Ctg Env 1 = Cutting Environment 1 (22/17 °C; 18h; low PPFD; low air flow); Ctg Env 2 = Cutting Environment 2 (17/11 °C; 12h; high PPFD; high air flow).

Environment 2 the optimum plant age was 9 weeks and the response of cuttings was much more irregular. Reasons for the irregularity may be leaf area effects or problems with the selection of mother plants or stems.

The interaction between cutting environments and leaf positions is shown in Figure 11. Cuttings from all leaf positions in the non-inductive Cutting Environment 1 produced significantly larger tubers than the corresponding leaf positions in Cutting Environment 2. In Cutting Environment 1, cuttings from leaf positions 5, 6 and 7 produced the largest tubers while cuttings from the youngest leaf positions (1, 2 and 3) produced the smallest tubers. In the inductive Cutting Environment 2, there were no significant differences in the sizes of tubers produced from the different leaf positions. There was still a tendency for cuttings from leaf positions 1 and 2 to produce smaller tubers than the rest, but these differences were not significant by Duncan's Multiple Range test .05 level.

The influence of mother plant age, conditioning of mother plants and cutting environments on the number of cuttings exhibiting the different types of bud development are shown in Figure 12. The majority of cuttings showed Type 3 development, which might be the effect of using a late maturing cultivar, as compared to Norland in Experiments 1 and 2, where the majority of cuttings showed Type 1 development.

The tendency of the numbers of cuttings showing Type 3 development to decrease, and for Type 1 to increase with increasing plant age is not seen here, except for cuttings from non-conditioned mother plants placed in Cutting Environment 1 (Figure 12b). For the remainder of the cuttings (Figures 12a, 12c and 12d), changing mother plant age did not seem to have much of an effect on bud development types.

The effect of Cutting Environments 1 and 2 on the numbers of cuttings showing the different types of bud development can be seen by comparing Figure 12a with

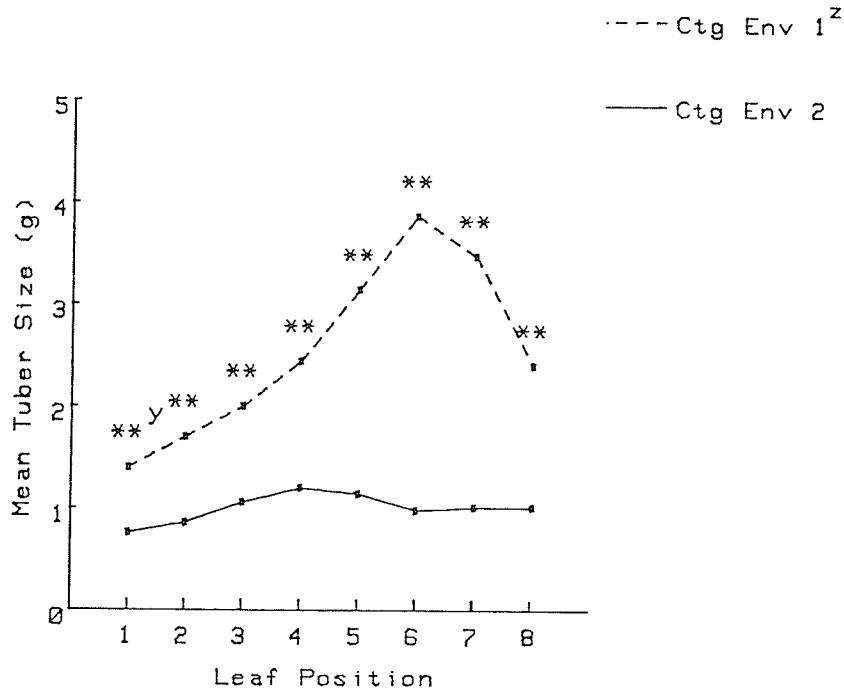


Figure 11. Interaction between cutting environments and leaf positions in Experiment 4.

[y] Comparisons between sizes of tubers produced by cuttings (Cutting Environment 1 vs Cutting Environment 2) from the same leaf position. ** indicates significance at the .01 level.

[z] Ctg Env 1 = Cutting Environment 1 (22/17°C; 18h; low PPFD; low air flow); Ctg Env 2 = Cutting Environment 2 (17/11°C; 12h; high PPFD; high air flow).

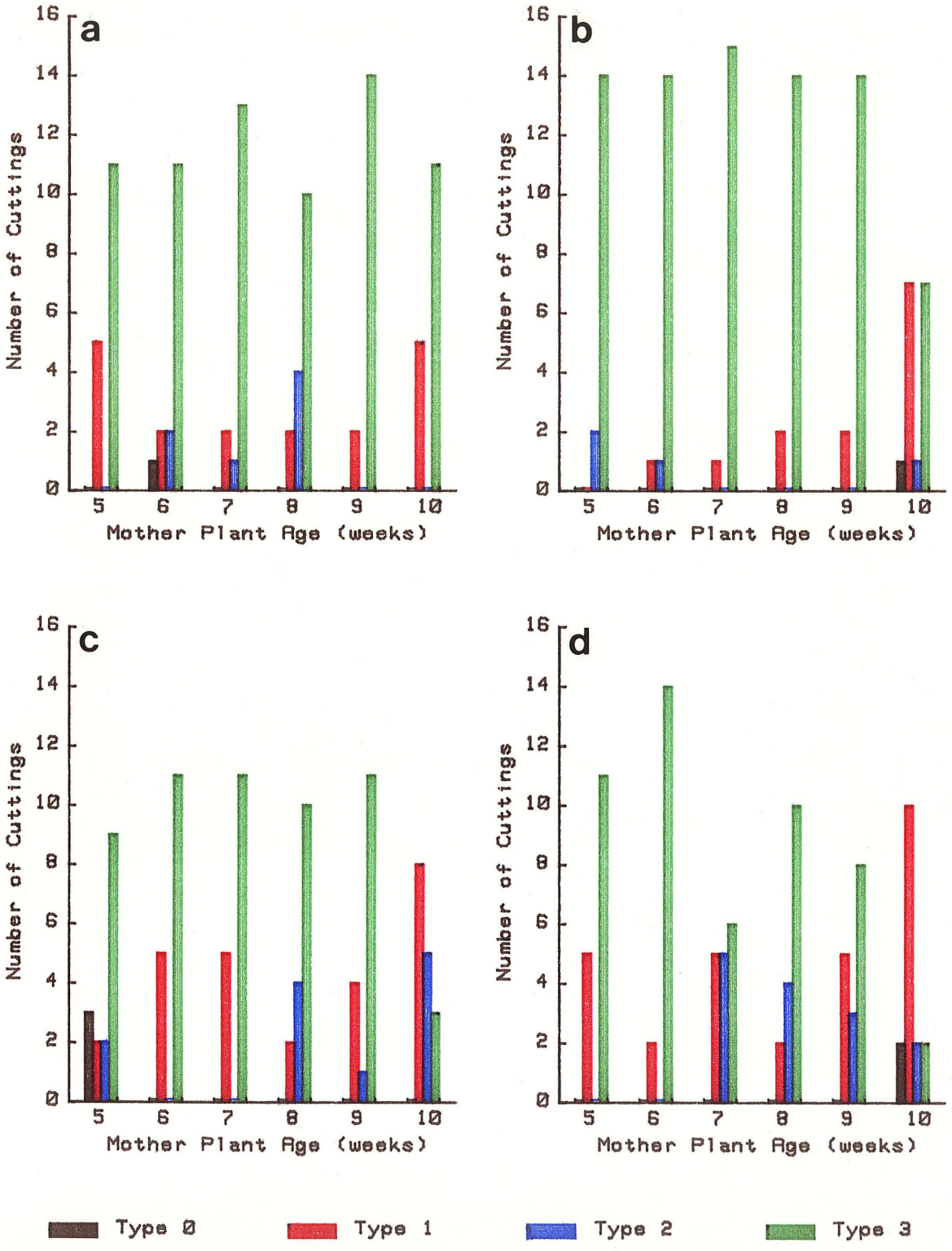
Legend:

Figure 12. The influence of mother plant age, conditioning of mother plants and cutting environments on the numbers of cuttings exhibiting the different types of bud development in Experiment 4.

Type 0 = no bud development
Type 1 = tuber formed from bud
Type 2 = tuber formed on stolon
Type 3 = tuber formed on stolon from new shoot

a - Cutting Environment 1; conditioned mother plants.
b - Cutting Environment 1; non-conditioned mother plants.
Cutting Environment 1 (22/17°C; 18h; low PPFD; low air flow)

c - Cutting Environment 2; conditioned mother plants.
d - Cutting Environment 2; non-conditioned mother plants.
Cutting Environment 2 (17/11°C; 12h; high PPFD; high air flow)


a
b
c
d
Number of Cuttings
Mother Plant Age (weeks)
0
2
4
6
8
10
12
14
16
5
6
7
8
9
10
Type 0
Type 1
Type 2
Type 3

12c and 12b with 12d. There would appear to be more Type 1 development in Cutting Environment 2 than in Cutting Environment 1, which is exactly what would be expected.

Figure 12a is compared with 12b and 12c with 12d to ascertain the effect of conditioning mother plants on bud development patterns. In this experiment, conditioning of mother plants would appear to have had little impact on bud development patterns.

Table 4 shows the mean tuber sizes produced by cuttings exhibiting the different types of bud development, regardless of what treatment or factor combinations were involved. Cuttings showing Type 3 development again produced the largest tubers while those showing Type 1 development produced the smallest tubers.

Experiment 5

The cuttings were harvested after 6 weeks, when the majority of them had senesced. The average tuber size was 2.41 g and 41.9 percent of the cuttings produced tubers that were greater than 2 g.

The purpose of this experiment was to study the effects of: 1)mother plant age, 2)conditioning of mother plants, 3)leaf positions and 4)cutting environments on the sizes of tubers produced and on bud development patterns. The interactions between these factors were of special interest.

The analysis of variance is shown in Table 14. It can be seen that the main effects of cutting environments, conditioning of mother plants, mother plant age and leaf positions were all significant at the .01 level. The interaction between cutting environments and mother plant age was significant at the .01 level. The interaction between conditioning of mother plants and mother plant age was significant at the .05 level. Leaf position was the only factor studied in this experiment that was not involved in an interaction.

Table 4. Mean tuber sizes produced by cuttings and percentage of cuttings exhibiting the different types of bud development in Experiment 4.

Type	Mean Tuber Size (g)	% of Cuttings
0	0.00	1.8
1	0.97	22.4
2	1.05	9.6
3	2.20	66.2

A comparison of tuber sizes produced by cuttings from the different leaf positions is shown in Figure 13. There are many overlapping categories, but the overall trend was for the youngest leaf positions (1, 2 and 3) to produce the smallest tubers. This time, cuttings from the oldest leaf positions did not tend to produce smaller tubers than the middle leaf positions.

The interaction between conditioning of mother plants and mother plant ages is shown in Figure 14. For the three oldest plant ages (9, 10 and 11 weeks), it didn't make any difference if cuttings were from conditioned or non-conditioned mother plants. For the three youngest mother plant ages, cuttings from conditioned plants produced significantly larger tubers than those from non-conditioned mother plants. This is exactly what would be expected in terms of what is known of the tuberization stimulus.

For cuttings from the conditioned plants, the cuttings from the 7 week old plants produced the largest tubers. Tuber size decreased for both younger and older plants. There were no significant differences in tuber sizes produced by cuttings from the 9, 10 and 11 week old conditioned plants. For cuttings from the non-conditioned plants, the effects of plant age were more irregular, with there being no great differences between any of the plant ages.

The interaction between cutting environments and mother plant ages is shown in Figure 15. Cuttings from all plant ages in the inductive Cutting Environment 2 produced significantly larger tubers than those in the non-inductive, high light Cutting Environment 3. In Cutting Environment 2, the cuttings from the 7 week old plants produced by far the largest tubers while the sizes of tubers produced by cuttings from the other plant ages did not differ greatly. The effect of plant age in Cutting Environment 3 was not as great as in Cutting Environment 2, with there being very little difference between the tuber sizes produced by the different plant ages. This may be due to the range of ages of mother plants used. In Cutting

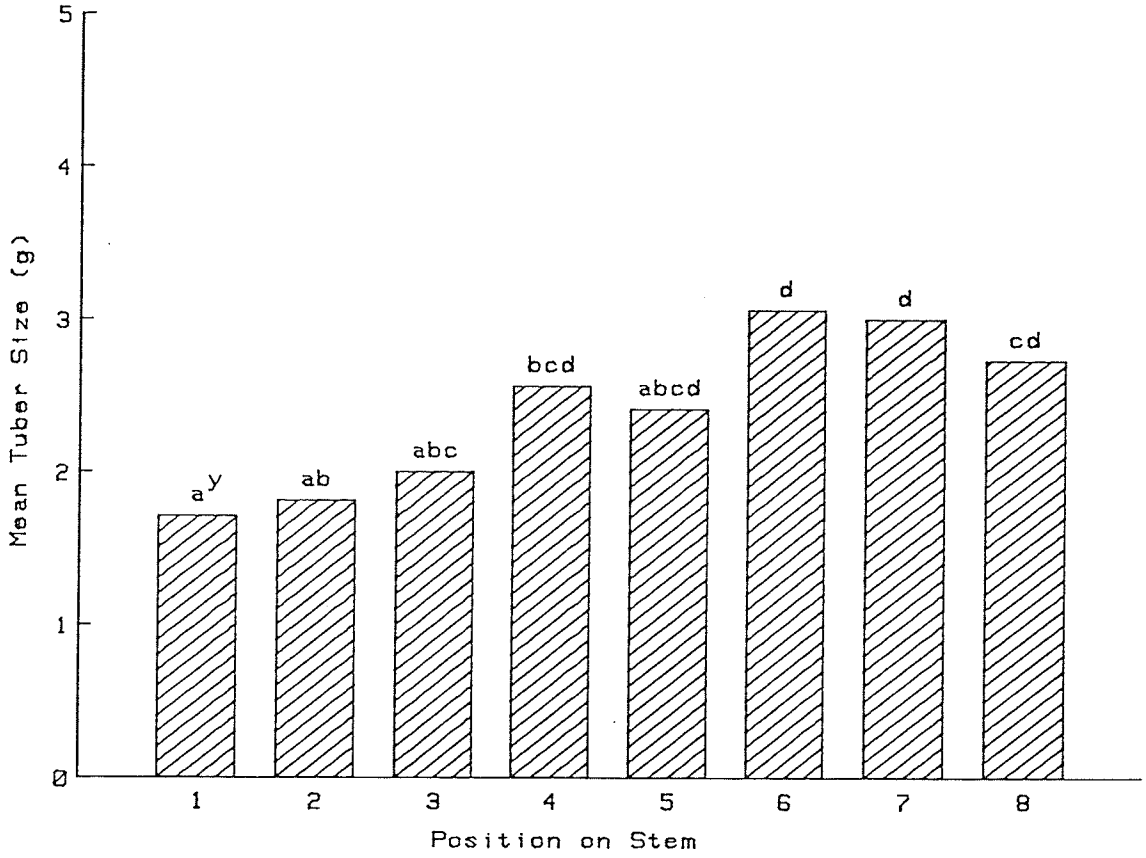


Figure 13. Comparison of tuber sizes produced by cuttings from leaf positions 1-8 in Experiment 5

[y]Values represented by bars with the same letter do not differ significantly by Duncan's Multiple Range test, .05 level. Values are means of 48 cuttings.

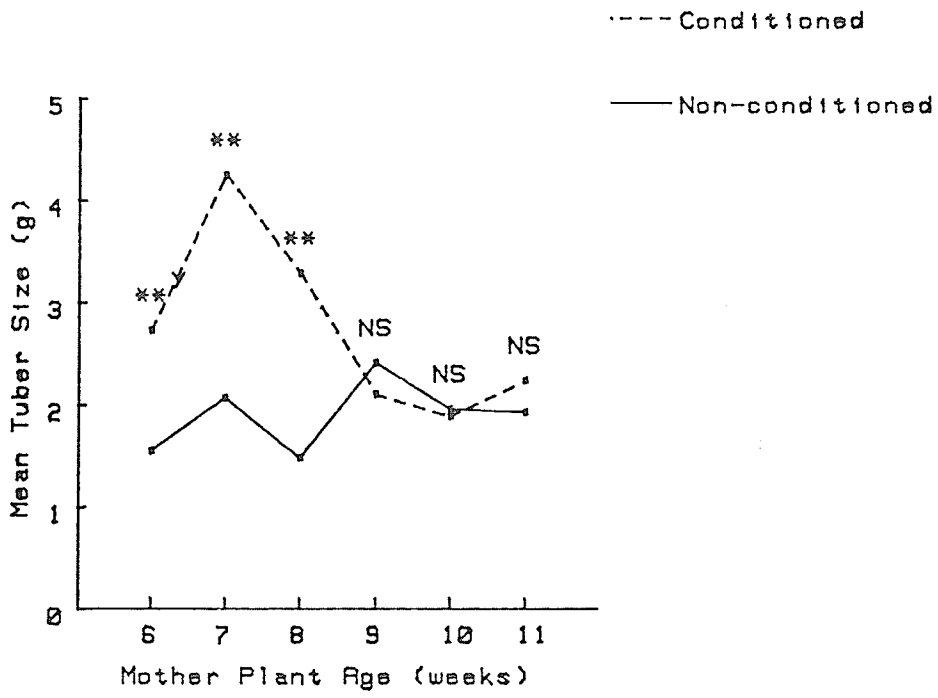


Figure 14. Interaction between conditioning of mother plants and mother plant ages in Experiment 5.

[y] Comparisons between sizes of tubers produced by cuttings (conditioned vs non-conditioned) from the same age of mother plants. * and ** indicate significance at the .05 and .01 levels respectively. NS indicates non-significance.

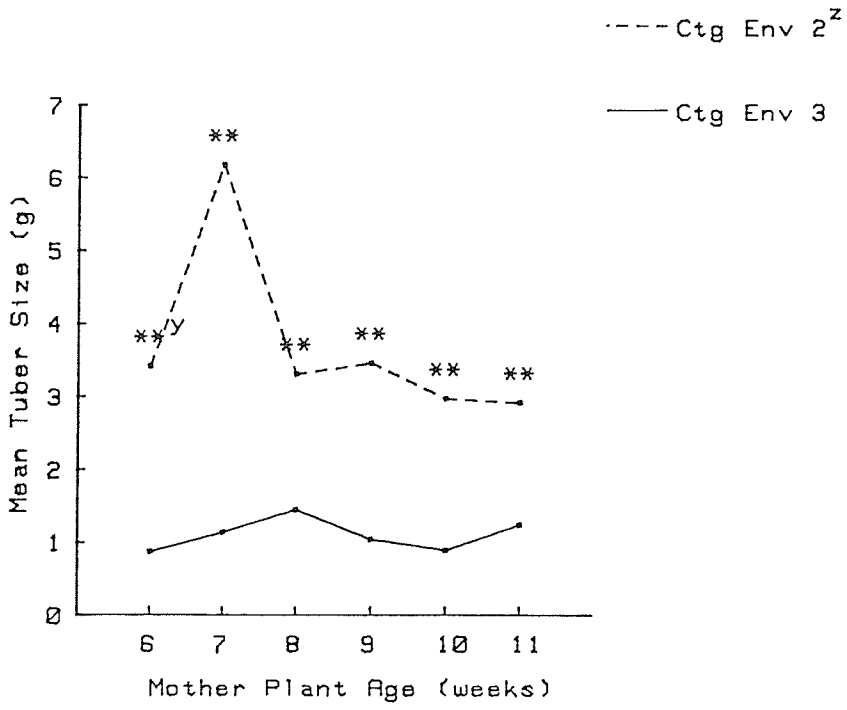


Figure 15. Interaction between cutting environments and mother plant ages in Experiment 5.

[y]Comparisons between sizes of tubers produced by cuttings (Cutting Environment 2 vs Cutting Environment 3) from the same age of mother plants. * and ** indicate significance at the .05 and .01 levels respectively.

[z]Ctg Env 2 = Cutting Environment 2 (17/11°C; 12h; high PPFD; high air flow); Ctg Env 3 = Cutting Environment 3 (22/17°C; 18h; high PPFD; high air flow).

Environment 3, this may reflect the difficulties in cutting establishment.

The influence of mother plant age, conditioning of mother plants and cutting environments on the numbers of cuttings exhibiting the different types of bud development are shown in Figure 16. Type 2 has the most common bud development type in this experiment. There was also as significant amount of Type 0 development.

As in Experiment 4, mother plant ages would appear to have little effect on bud development patterns. The effect of conditioning of mother plants was not great, but non-conditioning appeared to favor Type 0 and Type 3 development, while conditioning favored Types 1 and 2.

The effect of Cutting Environments 2 and 3 on bud development types can be seen by comparing Figure 16a with 16c and 16b with 16d. There would appear to be more Type 1 development in Cutting Environment 2 than in Cutting Environment 3, which is exactly what would be expected. There is more Type 0 development in Cutting Environment 3 than in Cutting Environment 2, which may be due to there being more wilting or to a lack of the tuberization stimulus in Cutting Environment 3.

Table 5 shows the mean tuber sizes produced by cuttings exhibiting the different types of bud development, regardless of what treatment or factor combinations were involved. The largest tubers in this experiment were produced by cuttings showing Type 2 development, not Type 3. This reflects the failure of some Type 3 cutting to set tubers. Some of the tubers harvested from Type 3 cuttings were very small. However the leaves on the new shoots were still green, and there is a possibility that if given more time, they may have produced considerably larger tubers

Analysis of covariance was carried out on the leaf area data to see if leaf area was exerting an effect on tuber size. The 'r' value for the error term was very

Legend:

Figure 16. The influence of mother plant age, conditioning of mother plants and cutting environments on the numbers of cuttings exhibiting the different types of bud development in Experiment 5.

Type 0 = no bud development
Type 1 = tuber formed from bud
Type 2 = tuber formed on stolon
Type 3 = tuber formed on stolon from new shoot

a - Cutting Environment 3; conditioned mother plants.
b - Cutting Environment 3; non-conditioned mother plants.
Cutting Environment 1 (22/17°C; 18h; high PPFD; high air flow)

c - Cutting Environment 2; conditioned mother plants.
d - Cutting Environment 2; non-conditioned mother plants.
Cutting Environment 2 (17/11°C; 12h; high PPFD; high air flow)

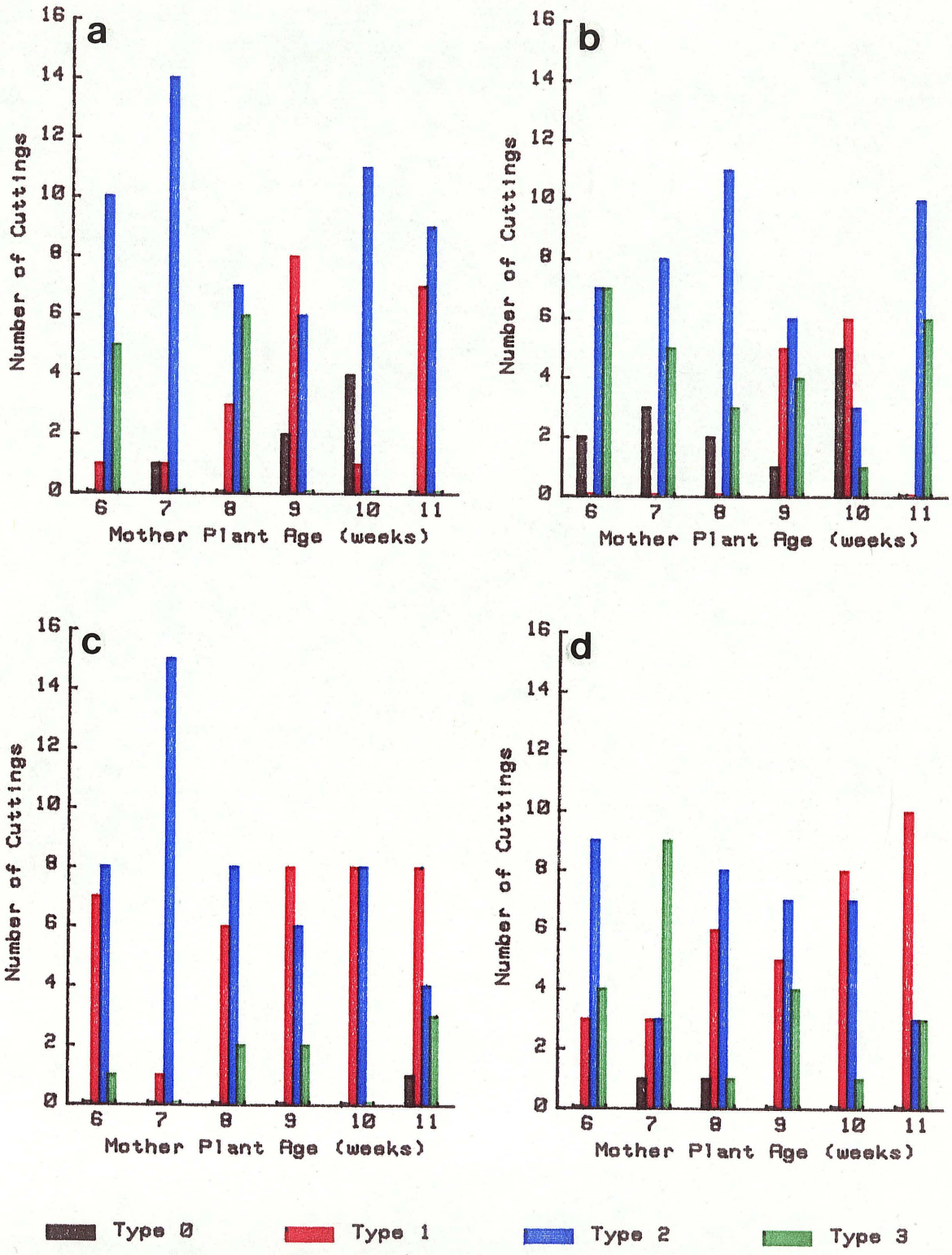

a
b
c
d
Number of Cuttings
Mother Plant Age (weeks)
0
2
4
6
8
10
12
14
16
6
7
8
9
10
11
Type 0
Type 1
Type 2
Type 3

Table 5. Mean tuber sizes produced by cuttings and percentage of cuttings exhibiting the different types of bud development in Experiment 5.

Type	Mean Tuber Size (g)	% of Cuttings
0	0.00	6.0
1	2.03	27.6
2	2.83	49.0
3	2.66	17.4

small (r=+0.03) which means that there was no underlying relationship between leaf area and tuber size in this experiment.

Experiment 6

The purpose of this experiment was to study tuber sizes and bud development patterns under modified conditions of irradiance and speed of air movement across the leaf canopy. Irradiances were reduced for the two weeks after cuttings were taken and the air flow rate was reduced for the duration of the experiment. One would expect these modifications to reduce wilting in cuttings and enhance cutting establishment.

The cuttings were harvested after 11 weeks. The original leaf on all the cuttings had senesced. In cuttings exhibiting Type 3 development, the new leafy shoots were still green, and, although exhibiting some symptoms of nutrient deficiency, were still fairly vigorous. The range of tuber sizes produced was 0.32 g. - 48.2 g. The average tuber yield per cutting was 10.02 g, which in terms of tuber production in comparison to previous experiments, is very successful.

Wilting was virtually non-existent. Under standard conditons in Cutting Environments 1 and 2 as described in the Materials and Methods, wilting was slight and, at the time, was thought to be well within acceptable limits. This is brought into question by the results of this experiment.

The analysis of variance is shown in Table 15. There were no significant differences between sizes of tubers from cuttings from stems selected from the same plants. There were no significant differences between tuber sizes from cuttings from the individual plants which were selected plants of Russet Burbank, 6 weeks old and non-conditioned. There were no significant interactions.

The differences between tubers produced from cuttings from the different leaf positions were significant at the .05 level. Leaf positions 1 (youngest) and 8 (oldest) produced significantly smaller tubers than the rest of the leaf positions.

Table 6 shows the percentages of cuttings exhibiting the different bud development patterns, the numbers of tubers produced and the average tuber weights. The reason why these results are presented differently than previous experiments, is that in previous experiments, it was relatively rare for a cutting to produce more than one tuber. In this experiment, some of the cuttings exhibiting Type 2 development and many of the cuttings exhibiting Type 3 development produced more than one fairly large tuber. As well, some cuttings that did form new leafy shoots did not produce tubers.

The largest tubers were produced from cuttings showing Type 3 development, which was the most common in this experiment. An intermediate size of tuber was produced by Type 2 development and the smallest tubers were from Type 1 development. There was also a significant percentage of cuttings (5.2%) showing no bud development at all.

Fertilizer

The purpose of Experiments 7 and 8 was to study the effects of adding fertilizer to the cuttings on tuber size and bud development patterns. In Experiment 7, a small range of mother plant ages were used. In Experiment 8, only one plant age was used.

Experiment 7

In this experiment, the effects of adding fertilizer to cuttings from a small range of mother plants ages, some conditioned, some non-conditioned, were observed. Leaf positions and leaf areas were also noted.

Light banks were mistakenly lowered on the cuttings two days after being taken, and extensive wilting resulted. The problem was caught quickly and steps were taken to minimize the effects. There is no doubt that this wilting had an impact on tuberization in terms of the sizes of tubers produced, the percentage of

Table 6. Mean tuber sizes produced by cuttings and percentage of cuttings exhibiting the different types of bud development in Experiment 6.

Type	Mean Tuber Size (g)	Percent of Cuttings	# of Tubers Produced	# of Tubers >2g
0	0.00	5.2	0	0
1	1.94	20.8	22	10
2	5.71	27.1	27	21
3	12.76	43.8	60	49

cuttings that did not tuberize, and on the longevity of the cuttings. This must be kept in mind while interpreting the results. However, useful observations can still be made about the addition of fertilizer in relation to plant ages, conditioning of mother plants and leaf positions.

The cuttings were harvested after 5 weeks. The average tuber size was 2.03 g and 39.6 percent of all the cuttings formed tubers greater than 2 g. The analysis of variance is shown in Table 16. The triple interaction of fertilizer, conditioning of mother plants and mother plant age was significant at the .01 level. The effect of leaf positions was the only factor studied not in the interaction.

A comparison of tuber sizes produced by cuttings from the different leaf postions is shown in Figure 17. Although the categories overlap a fair bit, again the trend is for cuttings from the youngest leaf positions (1 and 2) and the oldest (8) to produce smaller tubers than cuttings from the middle leaf positions.

The triple interaction between cutting environments, conditioning of mother plants and mother plant age is illustrated in Figures 18a and 18b.

The relationship between mother plant age and conditioning of mother plants for fertilized and non-fertilized cuttings is shown in Figure 18a. For fertilized cuttings, it didn't make any difference whether the cuttings were taken from conditioned or non-conditioned mother plants for plant ages 5, 6 and 7 weeks. For the cuttings from the youngest (4 weeks) mother plants, cuttings from non-conditioned plants produced much larger tubers than conditioned plants.

For the non-fertilized cuttings, differences between tuber sizes produced by cuttings from conditioned and non-conditioned plants were not significant for the 5 and 6 week plant ages. For the 4 week mother plant age cuttings from non-conditioned mother plants produced significantly larger tubers than those from conditioned plants. For the 7 week mother plant age cuttings from conditioned mother plants produced significantly larger tubers. This effect of conditioning on

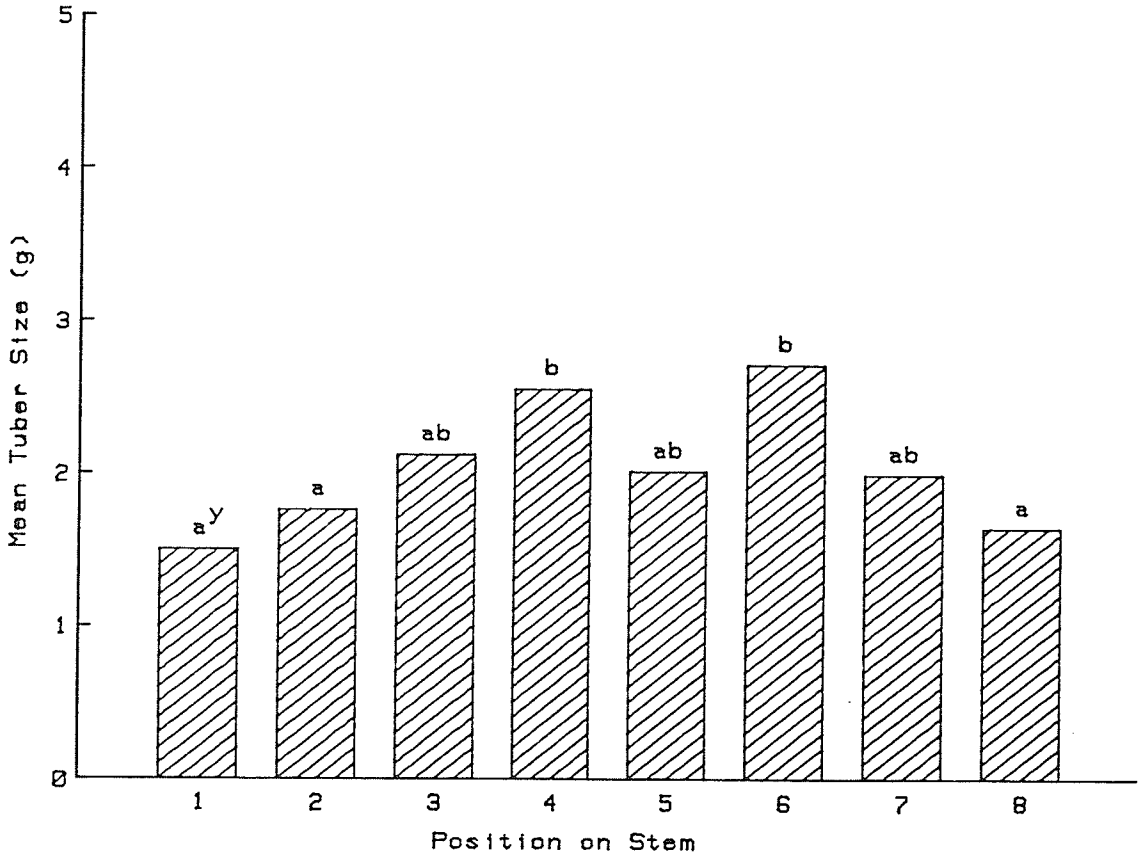


Figure 17. Comparison of tuber sizes produced by cuttings from leaf positions 1-8 in Experiment 7

[y]Values represented by bars with the same letter do not differ significantly by Duncan's Multiple Range test, .05 level. Values are means of 48 cuttings.

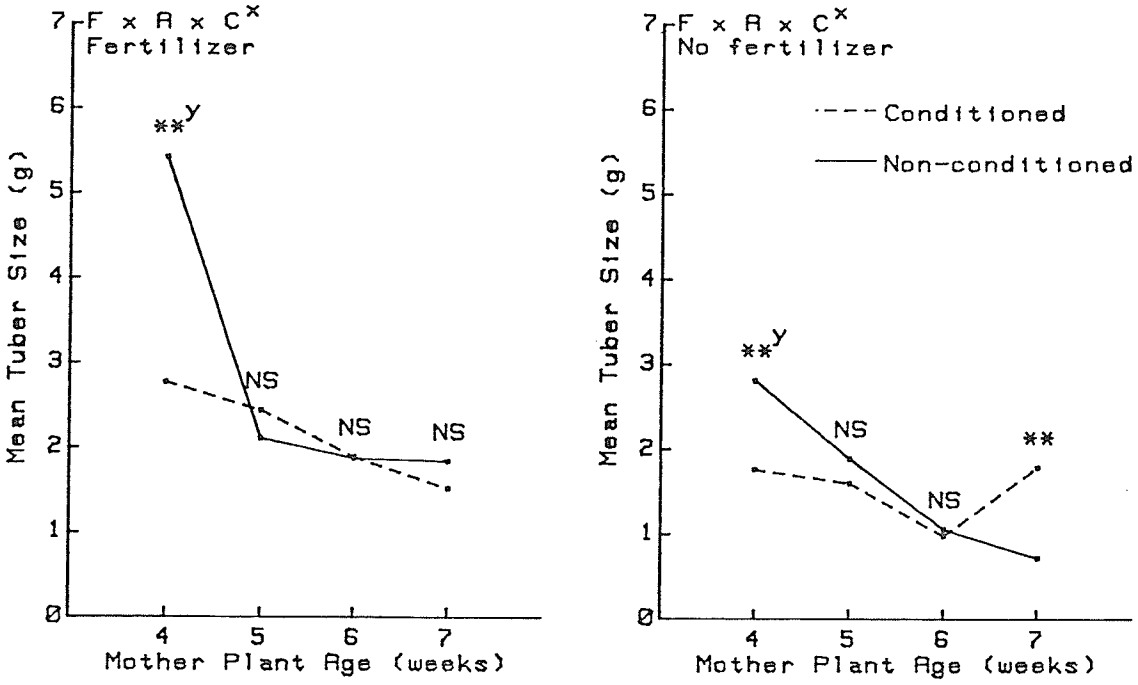


Figure 18a. Triple interaction between fertilizer, conditioning and mother plant age in Experiment 7. Relationship between plant age and conditioning for fertilized and non-fertilized cuttings.

[x]F = fertilizer; A = mother plant age; C = conditioning of mother plants.

[y]Comparisons between sizes of tubers produced by cuttings (conditioned vs non-conditioned) from the same age of mother plants. * and ** indicate significance at the .05 and .01 levels respectively. NS indicates non-significance.

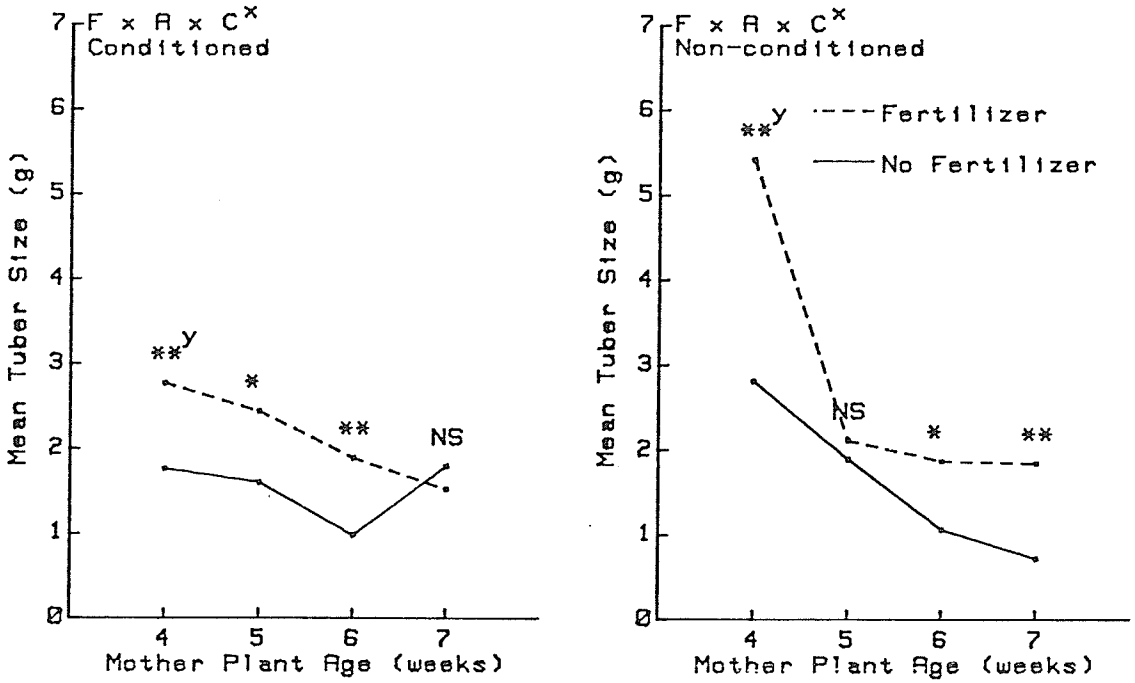


Figure 18b. Triple interaction between fertilizer, conditioning and mother plant age in Experiment 7. Relationship between plant age and fertilizer for conditioning and non-conditioning of mother plants.

[x]F = fertilizer; A = mother plant age; C = conditioning of mother plants.

[y]Comparisons between sizes of tubers produced by cuttings (fertilizer vs no fertilizer) from the same age of mother plants. * and ** indicate significance at the .05 and .01 levels respectively. NS indicates non-significance.

the cuttings from the 4 week plants is likely due to bud development patterns as will be seen later. It is difficult to know why tubers produced by the cuttings from the 7 week old conditioned plants were significantly larger, unless it is due to effects of plant or stem selection.

The relationship between mother plant age and fertilizer for conditioning and non-conditioning of mother plants is shown in Figure 18b. For cuttings from conditioned plants, fertilized cuttings produced significantly larger tubers, except for the oldest plant age (7 weeks). For cuttings from non-conditioned plants, fertilized cuttings produced significantly larger tubers except for the 5 week old plants. In general, fertilized cuttings produced larger tubers than non-fertilized cuttings.

The influence of mother plant age, conditioning of mother plants and cutting environments on bud development types is shown in Figure 19. The numbers of cuttings displaying Type 1 development are greater than would normally be expected for Norland cuttings that are this young. This may be an effect of wilting.

Almost all the cuttings showed all Type 1 development with the exception of the 4 week old, non-conditioned, non-fertilized cuttings, which showed mostly Type 2 development, and the 4 week old, non-conditioned, fertilized cuttings, which showed more Type 3 development.

Although it should be remembered that wilting was a factor here, the effect of conditioning of mother plants would appear to be to encourage Type 1 development over Type 2 or Type 3. The effect of adding fertilizer, in addition to increasing tuber size in Type 1 development, would appear to be to encourage the cuttings showing delayed tuberization to produce Type 3 development over Type 2.

Table 7 shows the mean tuber sizes produced by cuttings exhibiting the different types of bud development, regardless of what treatment or factor

Legend:

Figure 19. The influence of mother plant age, conditioning of mother plants and cutting environments on the numbers of cuttings exhibiting the different types of bud development in Experiment 7.

Type 0 = no bud development
Type 1 = tuber formed from bud
Type 2 = tuber formed on stolon
Type 3 = tuber formed on stolon from new shoot

a - fertilized cuttings; conditioned mother plants.
b - fertilized cuttings; non-conditioned mother plants.

c - non-fertilized cuttings; conditioned mother plants.
d - non-fertilized cuttings; non-conditioned mother plants.

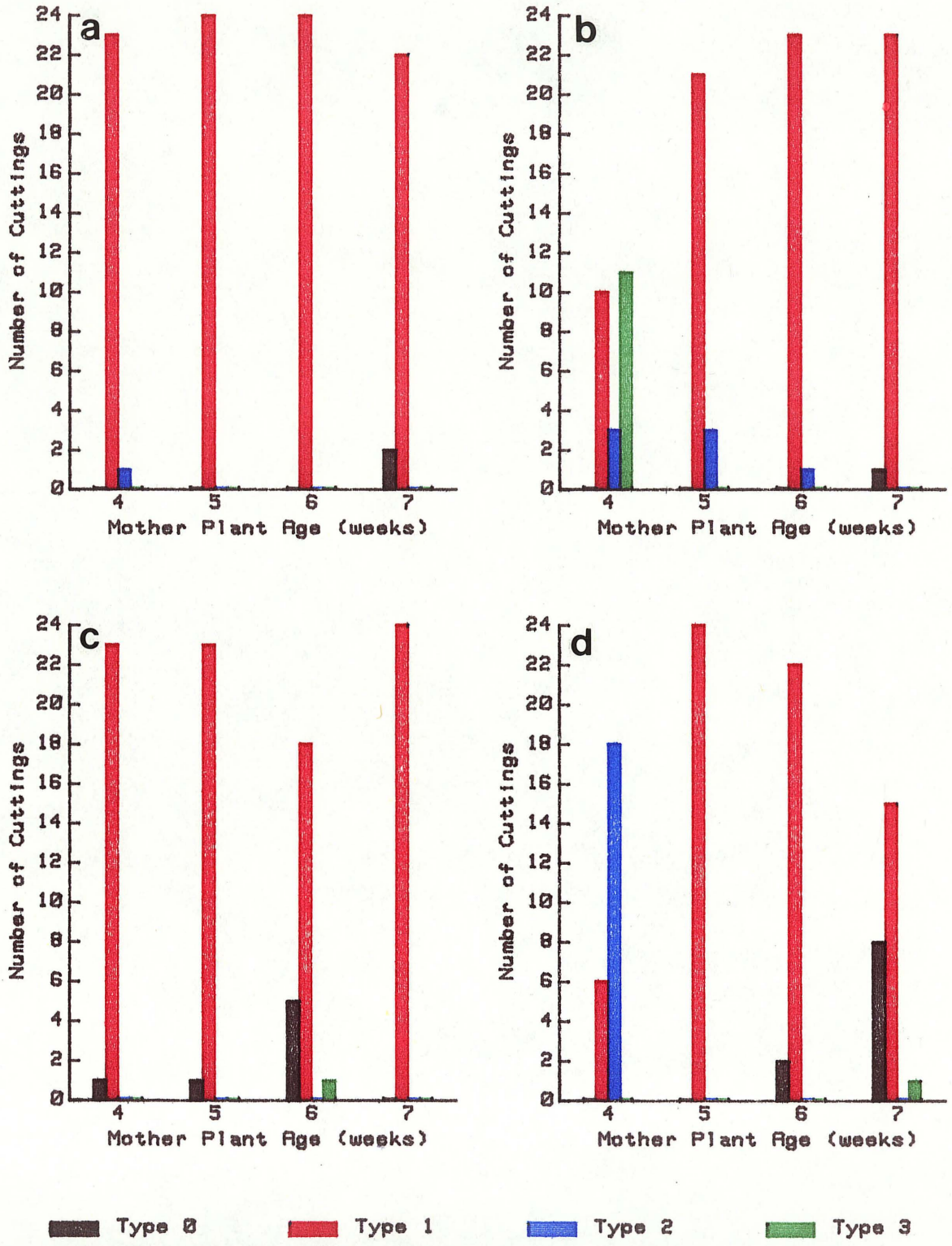

a
b
c
d
Number of Cuttings
Mother Plant Age (weeks)
0
2
4
6
8
10
12
14
16
18
20
22
24
4
5
6
7
Type 0
Type 1
Type 2
Type 3

Table 7. Mean tuber sizes produced by cuttings and percentage of cuttings exhibiting the different types of bud development in Experiment 7.

Type	Mean Tuber Size (g)	% of Cuttings
0	0.00	5.2
1	1.84	84.6
2	3.27	6.8
3	7.76	3.4

combinations were involved. Again, Type 3 cuttings produced the largest tubers, Type 2 cuttings produced tubers of an intermediate size and Type 1 cuttings produced the smallest tubers.

It is difficult to assess the impact of wilting on tuberization of the cuttings in this experiment, other than it would appear to lead to earlier senescence and smaller tubers being produced. In this experiment, non-conditioned cuttings produced larger tubers than conditioned cuttings from the same age of mother plant. Part of this effect may be due to the fertilizer treatment, but even the non-fertilized, non-conditioned 4 week old cuttings produced larger tubers than the rest of the non-fertilized cuttings. This effect, as well as the numbers of young Norland cuttings showing Type 1 development, could be explained if wilting caused an increase in the level of the tuberization stimulus.

Analysis of covariance was done on the leaf area data. The 'r' value for the error term was very small ($r=+0.05$) which would indicate that there was no underlying relationship between leaf area and the sizes of tuber produced. This would be particularly expected in this experiment because of the severe wilting. All the cuttings recovered quite well, but the larger cuttings did not recover as well as cuttings with smaller leaf areas.

Experiment 8

The purpose of this experiment was to study the effect of adding fertilizer to cuttings from a young non-conditioned plant. The effects of leaf areas and leaf positions were also noted.

The cuttings were harvested after 9 weeks. The cuttings showing Type 1 development had mostly senesced. The original leaves from the cuttings showing Type 3 development had senesced, but the new green leafy shoots were still very green and vigorous. The skin on tubers produced from these cuttings had not yet set. It is very likely that if these cuttings were not harvested so soon, they may

have produced even larger tubers.

The analysis of variance is shown in Table 17. Differences between fertilized cuttings and control cuttings were highly significant. Differences between leaf positions were not significant. The interaction between fertilizer and leaf positions was not significant.

The effect of adding fertilizer to cuttings is shown in Figures 20a and 20b. Flat B in Figure 20a had fertilizer added to the cuttings; flat A was a control flat. Figure 20b shows the tuber yield from the above flats. Cuttings with added fertilizer produced much larger tubers and all cuttings produced tubers, which was not the case for the control cuttings. Figure 20c shows a fertilizer treated cutting with extensive leafy shoot and root development. any leafy shoots in other experiments where fertilizer was not added tended to be much more spindly and to have smaller, lighter green leaves.

Table 8 shows tuberization by the cuttings, with patterns of bud development, numbers of tubers produced and mean tuber sizes. The largest tubers by far were produced by Type 3 development. Only cuttings with added fertilizer showed Type 3 development. Cuttings with no added fertilizer showed Type 0 and Type 1 development, with a small amount of Type 2.

It is interesting to note that an equal number of cuttings (31%) showing no bud development in the no fertilizer flat formed extensive green leafy shoots in the fertilizer flat. The effect of fertilizer would appear to be to encourage leafy shoot development over no bud development and to encourage Type 2 development over Type 1

One would not expect the effect of the addition of fertilizer to be to increase the level of tuberization stimulus in a cutting. There would appear to be no lack of the tuberization stimulus in the cuttings in the no fertilizer flat that did form tubers because they were mostly sessile, which is associated with high levels of

Legend:

Figure 20a. A - cuttings with no added fertilizer; B - cuttings with added fertilizer.

Figure 20b. A - tuber yield from flat A (above); B - tuber yield from flat B.

Figure 20c. A cutting with added fertilizer showing extensive Type 3 development.

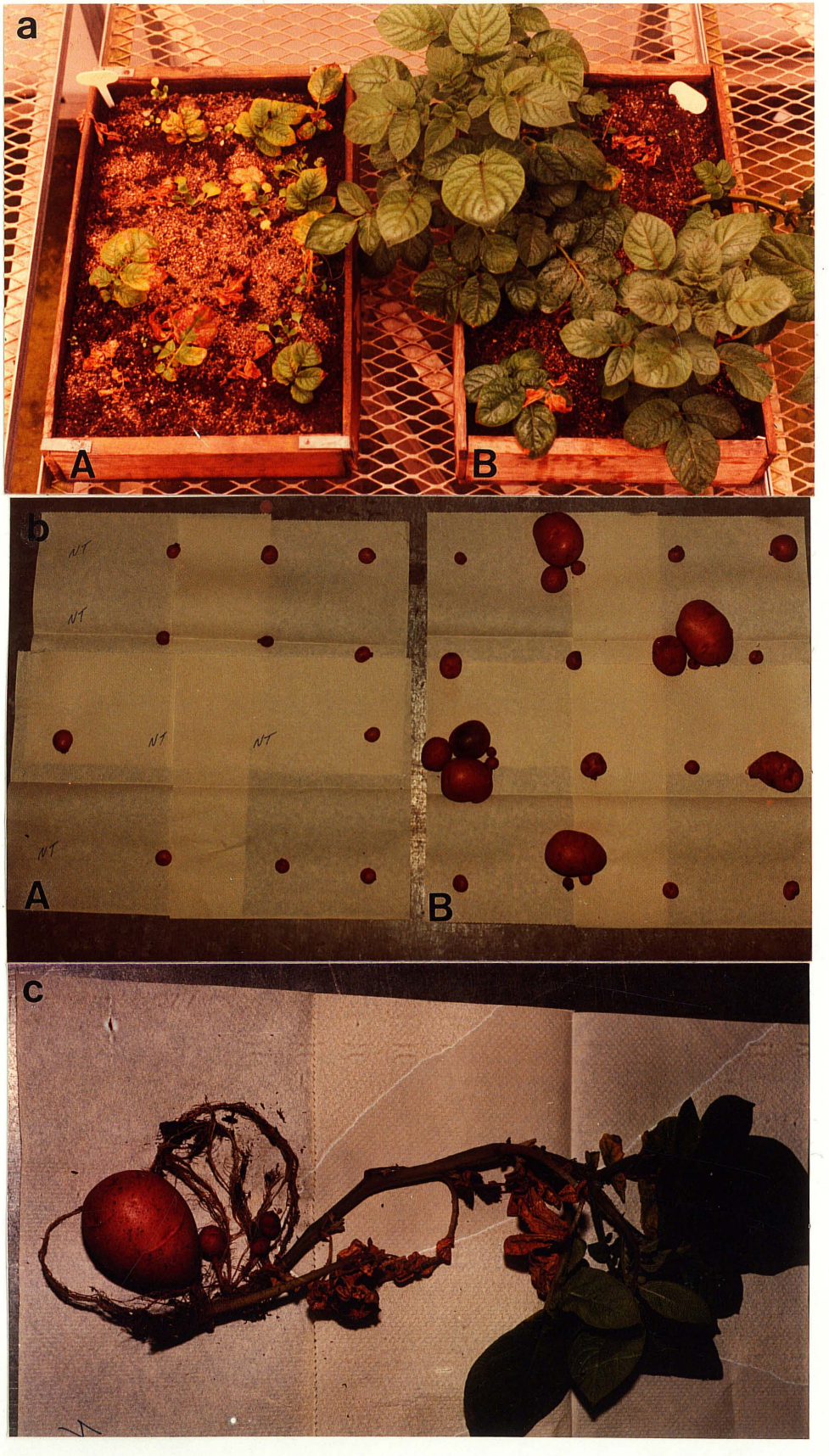

a
A
B
b
NT
NT
NT
NT
NT
A
B
c

Table 8. Mean tuber sizes produced by cuttings and percentage of cuttings exhibiting the different types of bud development in Experiment 8.

Type	Mean Tuber Size (g)	# of Tubers Produced	# of Tubers >2g	Percent of Cuttings
0	0.00	0	0	NF[y] 31.0 F 0.0
1	1.83	17	6	NF 62.5 F 44.0
2	5.79	5	3	NF 6.3 F 25.0
3	29.29	13	8	NF -- F 31.0

[y]NF- cuttings with no added fertilizer; F- cuttings with added fertilizer.

Legend:

Figure 8. The influence of mother plant age, conditioning of mother plants and cutting environments on the numbers of cuttings exhibiting the different types of bud development in Experiment 2.

Type 0 = no bud development
Type 1 = tuber formed from bud
Type 2 = tuber formed on stolon
Type 3 = tuber formed on stolon from new shoot

a - Cutting Environment 1; conditioned mother plants.
b - Cutting Environment 1; non-conditioned mother plants.
Cutting Environment 1 (22/17°C; 18h; low PPFD; low air flow)

c - Cutting Environment 2; conditioned mother plants.
d - Cutting Environment 2; non-conditioned mother plants.
Cutting Environment 2 (17/11°C; 12h; high PPFD; high air flow)

tuberization stimulus (Ewing and Wareing, 1978). It doesn't make sense that certain leaves on a stem would have very high levels of tuberization stimulus and others adjacent on the same stem to have so little that there is no bud development. One might therefore question if lack of bud development is always associated with a complete lack of induction as suggested by Ewing and Wareing (1978). Lack of bud development is sometimes associated with wilting, but wilting was not a factor in this experiment.

Analysis of covariance was done on the leaf area data. The 'r' value for the error term was +0.27, which is not significant. This means that there was no underlying relationship between leaf area and size of tubers produced.

Carbon Dioxide Enrichment

The purpose of experiments 9a, 9b and 9c was to study the effect of enriching the soil zone of the cuttings with carbon dioxide to see if it would increase sizes of tubers produced or modify bud development patterns.

The analysis of variance is shown in Table 18. There were no significant differences between tubers produced by cuttings treated with carbon dioxide and control cuttings.

There was a slight tendency for the tubers from carbon dioxide treated cuttings to be larger than the others. All cuttings formed sessile tubers with virtually no roots. Tubers from cuttings in this experiment tended to be smaller than from comparable types grown in Cutting Environments 1 and 2 which may be due to a tendency for more wilting in Cutting Environment 4.

The equipment used here was relatively simple, but according to measurements, it did succeed in raising carbon dioxide levels in the CO_2 treated flat, although levels tended to be quite variable when taken at different locations across the flat and at different soil levels.

The success at increasing plant yields by soil enrichment with CO_2 depends on

CO_2 uptake by the roots of a plant. It is possible that the lack of success here was because the cuttings had almost no roots. This type of development (lacking roots) was possibly due to the age of plants used and to the cutting environment. There may be potential for success with this method if one was working with a combination of factors that would produce rooted cuttings.

In these experiments, a continual exposure to elevated CO_2 levels was used. It may in fact be sufficient to use a single exposure for a short period of time as done by Arteca et al. (1979) with intact plants.

Abscisic Acid

Varying concentrations of ABA were applied to cuttings in Experiments 10a, 10b, and 10c. Although there is controversy over whether ABA does promote tuberization (Menzel, 1980; Claver, 1970), it has been found to play a role in reducing transpiration and this is apparently due to stomatal closure. It was thought that ABA might help prevent initial wilting of cuttings, after they are first taken.

After cuttings were taken, they were placed in Cutting Environment 4, where there was a moderate amount of wilting. No visible differences were observed in the wilting of the cuttings, whether they were control, or treated with ABA.

The analysis of variance is shown in Table 19. There were no differences in tuber sizes produced by control cuttings or any of the ABA treated cuttings in any of the experiments. There were no effects on bud development patterns by any of the treatments. All tubers were formed flush in the leaf axil.

For experiments 10b and 10c, leaf areas were determined. Analysis of covariance was done on the leaf area data to assess the effect of leaf area on the sizes of tubers produced. The 'r' values for the error terms were high for both experiments ($r=+0.89$ and $r=+0.83$) and were highly significant. This would mean that for these experiments, there was a real relationship between leaf area and

sizes of tubers produced. For Experiment 10b, 79 percent of the unexplained variation could be accounted for by differences in leaf area and for Experiment 10c, 69 percent of the unexplained variation could be accounted for by differences in leaf area.

There were no positive effects of ABA on wilting or on tuber production in these experiments, but this does not mean the use of ABA does not have possibilities with this propagation method. Among the factors that might be considered is using a different cutting environment, where stress would be less , with only very slight wilting. The concentrations of ABA and methods of application could be changed. We have no idea of how long the ABA may have persisted in the cuttings. Perhaps more than one application over the first several days until cuttings have had an opportunity to get established would be more effective. A range of plant ages could also be used. ABA could be applied to the mother plants before cuttings are taken.

GENERAL DISCUSSION

Tubers From Leaf Bud Cuttings - the Ideal Situation

It is apparent that in obtaining tubers from leaf bud cuttings, different processes are involved. These are: 1)cutting establishment, 2)vegetative growth of the cuttings and 3)tuberization. Tuberization involves i)tuber initiation involving the tuberization stimulus and ii)tuber enlargement. Each of these processes is favored by different combinations of conditions.

Cutting establishment is favored by a low transpiration rate which is favored by relatively low temperatures, low irradiance, high humidity and low rate of air movement across the leaf canopy (Raven et al.). Vegetative growth in the potato is favored by relatively high temperatures, high irradiance, long photoperiods, young plants and plants grown from physiologically young mother tubers. The formation of the tuberization stimulus is favored by low temperatures, short photoperiods, old plants and plants grown from physiologically old mother tubers. Tuber enlargement is favored by the factors favoring vegetative growth as well as the continued presence of the tuberization stimulus at high levels.

Highest yields in intact potato plants are achieved with a balance of the above factors (Ewing, 1981) in order to have enough vegetative growth to support tuber enlargement and enough tuberization stimulus for tuber initiation and continued tuber enlargement.

Ewing (1981) describes hypothetically ideal conditions for getting the highest yields in intact potato plants. These include physiologically young seed tubers, high rates of nitrogen fertilization early in the season, a cultivar with a moderately critical photoperiod, long days and warm temperatures early in the growing season

and short days and cool temperatures late in the season when large haulms have developed.

In intact plants, early yields are realized by conditions favoring the early development of the tuberization stimulus. These include moderate to low rates of nitrogen fertilization, short days and cool temperatures (Ewing, 1981). However, if all of these factors are present too early in the season, the tuberization stimulus will be present so strongly that haulms will be dwarfed and tuber yields, though early, will be negligible (Ewing, 1981). It is essential for a plant to have the ability to support tuber growth after initiation. At the other extreme, there can be a preponderance of conditions favoring vegetative growth so that the production of the tuberization stimulus is so delayed that tuber yields are limited by the growing season. It is apparent, then, that a balance of factors favoring vegetative growth and tuber enlargement and ones that promote the formation of the tuberization stimulus, is desirable.

Many of the factors involved in intact plants regarding tuberization and high tuber yields would appear to be applicable for obtaining tubers from leaf bud cuttings. The concern in obtaining tubers from leaf bud cuttings is not just to get cuttings to tuberize, but to get them to form sizable tubers. It may also be possible to describe hypothetically ideal conditions for highest yields from leaf bud cuttings in the same manner as it has been done for intact plants.

In cuttings where sessile tubers were formed, the level of induction was thought to be very strong (Ewing and Wareing, 1978). With this type of development, there was no shoot development, little, if any, rooting and tubers tended to be very small. There would appear to be a limited source for tuber enlargement. There would be limited production of photosynthate, and, with little or no rooting, there would be relatively little water and nutrient uptake. It is therefore entirely logical that tuber size was necessarily limited.

Cuttings showing Type 1 development also tended to senesce earlier than cuttings showing other types of development. This is perhaps due to the migration of nitrogen, phosphorous and potassium from the leaf to the developing tuber and to more moisture stress because of the lack of roots. Tuber production may have been earlier than for the other patterns of bud development, but this is not known for sure because once cuttings were placed in the flats, they were not disturbed until the harvest date.

Tubers produced on stolons usually had some rooting, and, sometimes, quite extensive rooting. Tubers produced from cuttings showing this type of development tended to be larger sessile tubers. This is likely due to these cuttings having a greater capacity for water and nutrient uptake and a longer duration of the tuber bulking phase. These types of cuttings did not tend to senesce as early as leaf bud cuttings with sessile tubers.

Leaf bud cuttings with new shoot development, which was usually accompanied by fairly extensive rooting, tended to produce the largest tubers with few exceptions. Cuttings with this type of development would have a greater potential for the production of photosynthate and for the uptake of water and nutrients. They also have a longer duration of growth. Often when they were harvested, the original leaf had senesced, but the new leafy shoot was still green and vigorous.

There were times, however, when cuttings showing Type 3 development produced only very small tubers or none at all. This happened with some young Russet Burbank cuttings and sometimes with young non-conditioned Norland. This may be due to a lack of induction. It is possible, that with this type of development, tuber formation was merely delayed, and given enough time, the very small tubers would have enlarged, and cuttings without tubers may have formed tubers.

It would appear then, that contrary to Ewing's (1981) observations, greater

intensity of the induction does not necessarily have a positive effect on the sizes of tubers formed by cuttings. His observations, however were made by studying cuttings in a situation where they were being selected for early tuberization. This was not the case in the experiments reported in this thesis.

The largest tuber yields in this study were realized in experiments where the cuttings had the greatest longevity, and, therefore, the longest duration of tuber growth. For example, yields were much greater from experiments where cuttings were harvested after 9 - 11 weeks, rather than the usual 4 - 7 weeks. It is true that the object of obtaining tubers from leaf bud cuttings is to quickly increase selected plant stocks. It would therefore be desirable to have early yields. However, if these early yields are almost negligible, as is often the case, there is no value in having early yields over somewhat later yields.

General Considerations for Experimental Planning and Design

Experimental Design

One of the biggest problems with studying tuberization in leaf bud cuttings is to get similar experimental units on which to perform different treatments. This is a formidable task in dealing with a situation where cuttings with different leaf areas are being taken from different leaf positions, stems, and even plants, if the experiment is of any size. It has been recognized that, in the potato plant, because of differences in the genetic makeup of various species, cultivated varieties and even clones, reactions to the environment may differ from plant to plant (Gregory, 1965).

One thing that can be done, is to completely randomize cuttings and randomize leaf positions within blocks or treatments as done in Experiments 9 and 10. It is likely that any differences due to leaf positions and plants will all even out and one can get some idea of whether a particular treatment works or not. However, a lot

of valuable information can be lost this way. It is well known that tuberization in the potato is affected by many interactions (eg. temperature and photoperiod) and the relationship between these tends to be quite complex (Gregory, 1965).

The above method gives no indication of interactions and no indication if a particular 'treatment' would work especially well with a certain age of mother plant, a particular set of prior treatment conditions on the mother plants or certain leaf positions. It would therefore be desirable to keep track of these factors and to have some idea of possible interactions. It was for this reason that most experiments in this study were performed within a factorial arrangement.

If this had not been done, less information would have been obtained in this study. For example, if in Experiment 1, conditioning of mother plants was studied and plant age 4 weeks was used, and the cuttings had been placed only in Cutting Environment 1, one would have concluded that conditioning of mother plants made a difference. If cuttings had been placed only in Cutting Environment 2, one would have concluded that conditioning of mother plants didn't make any difference.

Selection of Mother Plants and Stems

In an attempt to obtain similar experimental units on which to test the effect of different factors, the assumption was made that similar stems on a given plant will react in the same way. Two or three stems were visually selected for uniformity from one mother plant. In most large experiments, four mother plants of the same age were selected out of a possible 6 or 8. These were selected on the basis of uniformity, and each had to possess at least two very similar stems. Mother plants themselves were started from tubers selected for uniformity and any plant that did not emerge within 2-3 days of the others was culled.

In initial investigations, two experiments were carried out to study the effectiveness of the selection procedure. In the first experiment, 4 mother plants were selected out of a possible 8. In the second experiment, 6 mother plants were

selected out of a possible 10. Selected stems from the same mother plants behaved in approximately the same way and tuber yields from stems from the same plant did not differ significantly. This was also observed in Experiment 6 when stems were tested against one another.

However, selected mother plants did not always behave in the same way. One out of the six selected mother plants in the second experiment differed significantly from the others in terms of tuber yield. In designing the experiments, some protection against this was built in by having two replicates of each mother plant age per treatment.

The preliminary experiments mentioned above would help justify the assumption that similar stems from a given plant will react in the same way. But there is no guarantee that this will always be the case. Moorby (1978), in studying the growth of the potato plant, states that he believes that the simplest and most logical definition of an individual plant is to consider each stem which arises below ground, to be an individual.

Some of the seemingly unexplainable effects of some of the factors studied in these experiments may be attributable to problems in the selection of stems and mother plants, ie., similar ones did not behave in a similar way.

Bud Development Patterns

Four different categories were developed for the purpose of classifying the types of growth shown by the axillary bud. These were not chosen to correspond to any theoretical induction levels or levels of the tuberization stimulus. They were chosen, instead, for ease and uniformity of classification from experiment to experiment.

With Type 0 development, there was no development of the axillary bud at all. This could have been due to lack of the tuberization stimulus (Ewing and Wareing, 1978), wilting, or perhaps some other cause as suggested by the results in

Experiment 8. With Type 1 development, sessile tubers were formed. With Type 2 development, tubers developed on stolons. Stolons may have been extremely shortened and thickened, or may have been long and thin. These types of stolons really corresponded to different levels of induction. A cutting with a short thick stolon may have been more closely related (in terms of the level of tuber-inducing stimulus) to a Type 1 cutting than to another Type 2 cutting with long, slender, multiple branched stolons.

A cutting showing Type 3 development formed a new leafy shoot, stolons, and tuber(s). Any negatively geotropic structure with a bit of green was designated as a leafy shoot. It could have been a very reduced fleshy shoot (stolon-like) or a very large leafy shoot as shown in Figure 20c.

Classification of cuttings into these categories was, at times, rather subjective. For example, sometimes there was difficulty in judging if a cutting had a stolon or if the tuber was sessile with an elongated base. If the structure was very short and tapering, it was judged to be an elongated base and the tuber to be sessile. If the structure was of fairly uniform thickness and white in color, it was judged to be a stolon. There was in fact some variations in these patterns of formation and there were some intermediate types of development which were placed in the category to which they most closely corresponded. It is possible that with a bit of experience, finer classification categories could be developed that would more closely correspond to graduated theoretical levels of induction.

Other Considerations

Although not specifically reported in this thesis or elsewhere, there were basically two types of tubers formed by leaf bud cuttings. The first type was perfectly formed miniature tubers with dormant buds or 'eyes'. These were generally the type formed on stolons. The other type tended to be be odd shaped with small green leaflets. This was generally the type formed flush in the leaf axil

or on very fleshy stolons.

Evaluation studies should be done on the performance of these types of tubers. One type may not outperform the other, but if the first type did outperform the second type, it would provide even more reason to attempt to get cuttings to form stolons, then tubers.

Evaluation studies should also be done to establish target tuber sizes for the leaf bud cutting procedure. Clearly defined desirable and permissable tuber sizes are needed. Rossnagel (1980) designated any tuber greater than 2 g as 'usable'. Lauer (1977) mentions that haulm development tends to be slow from tubers smaller than 4 g.

Only rough records were kept on the longevity of cuttings because of the difficulty in uniformly judging when a cutting had senesced. With experience, this could possibly be done with a fair degree of accuracy. In this study, cuttings with Type 0 or Type 1 development tended to senesce sooner than ones showing Type 2 or Type 3 development. With type 3 development, the original leaf tended to be long senesced while the new leafy shoot was still green and vigorous.

One thing that was not recorded in this study was the different sizes of the axillary buds of the cuttings. Ewing (1978) recommends that, for the sake of uniformity, it is desirable to choose nodes that have buds less than 1 cm long at the time of cutting. In this study, sometimes the buds were very small and sometimes they were fairly large. This may have contributed to the unexplained variability in the experiments.

Specific Factors Studied

One thing that is clear from this study is that many factors influence tuberization in terms of bud development patterns and sizes of tubers produced. Many of the factors tend to influence one another and therefore are subject to interactions. Because of these interactions, it is unlikely that one could find an

optimum condition for a factor that will work under all conditions in all situations. The best one can hope for is an indication of conditions of factors that will increase the probability of obtaining the largest tubers within a given range of situations.

Cutting Environments

The greatest differences in tuberization in leaf bud cuttings were found between the different cutting environments. Cutting Environments can drastically affect the responses of cuttings in terms of bud development types and tuber yield.

Cutting Environment 1 tended to favor Type 3 development and Cutting Environment 2 tended to favor Type 1. By alterations in bud development patterns, it is likely that differences between Cutting Environments 1 and 2 are due to Cutting Environment 1 having a non-inductive temperature and photoperiod, and Cutting Environment 2 having an inductive temperature and photoperiod. But this is not likely in comparisons of Cutting Environments 1 and 2 to Cutting Environment 3. The temperature and photoperiod in Cutting Environment 3 were identical to that in Cutting Environment 1 and yet there were very great differences in the percentage of cuttings tuberizing, and in sizes of tubers produced. It would appear that differences in Cutting Environment 3 are more related to wilting or cutting establishment due to high irradiance and high rate of air movementcoupled with long days and high temperatures. Differences between Cutting Environments 2 and 3 are likely due to both inductive and non-inductive conditions, and to higher daily irradiance in Cutting Environment 3.

Wilting was present in some of the experiments. There was only very slight wilting in Cutting Environments 1 and 2. This was thought to be well within acceptable limits because nearly all cuttings tuberized and a considerable number of fairly large tubers were formed. However, in Cutting Environment 3, when irradiance and air flow rates were reduced, and the supplemental misting system

was used, larger tubers, along with many sizable multiple tubers, were formed. Cuttings also did not senesce as soon as in other experiments. Was this due to a particularly favorable combination of plant ages and conditions, or to a total lack of wilting?

Wilting of potato cuttings is not specifically mentioned in the literature, although it does seem likely that it was a factor with some of the irradiances and temperatures used. What influence does wilting have on bud development patterns, sizes of tubers formed and longevity of cuttings?

In preliminary experiments in Cutting Environment 2, with no supplemental mist, there was a very low percentage of cuttings that tuberized, tubers were very small and all cuttings senesced within 3 weeks. With just some wilting, although effects are not as severe, and a high percentage of cuttings tuberize, it would still follow that some of the effects would still be operative (eg. smaller tubers and shorter lived cuttings). There is some evidence (for example, Experiment 7) that wilting may cause a greater than expected amount of Type 1 development but this is not proven.

In any further research, the primary issue to be addressed would be to eliminate wilting. This likely can be done with reduced irradiance, supplemental misting and a growth facility with no forced air flow.

The irradiances in Cutting Environments 2 and 3 were greater than in Cutting Environment 1. Goodwin (1981), in rooting leaf bud cuttings for transplant to the field, found that the optimum irradiance for the establishment of cuttings was under 80% shade and that cuttings grew best under 30-50% shade. (Units for irradiance were not given.) It is likely that there are optimum irradiances for obtaining tubers from leaf bud cuttings as well.

Conditioning of Mother Plants

Rossnagel (1980) found that cuttings did not tuberize well unless the mother

plants were subjected to 10 to 14 days of inducing conditions. One would expect that the effect of conditioning mother plants would be to increase the level of induction in the plant. From current theories of tuberization in cuttings, one would expect two main effects of conditioning of mother plants: 1) If the level of the tuberization stimulus is limiting, there would be a greater percentage of cuttings from conditioned mother plants tuberizing than from non-conditioned mother plants. 2) There would be more Type 1 development and less Type 3 development in cuttings from conditioned mother plants.

In this study, except for some young Russet Burbank cuttings, it is doubtful that the level of the tuberization stimulus was ever limiting as far as tuber initiation and enlargement is concerned. This is likely due to all plants being grown from physiologically old mother tubers.

Only for Russet Burbank cuttings in Experiment 5, did the cuttings behave exactly as expected. For the three oldest plant ages, which had a relatively high level of induction, it didn't make any further difference if mother plants were conditioned or not. For the three youngest plant ages, cuttings from conditioned mother plants produced significantly larger tubers than cuttings from non-conditioned mother plants. Conditioning also decreased the numbers of cuttings showing Type 0 and Type 3 development and increased the numbers showing Types 1 and 2.

Results of conditioning mother plants in Experiment 4 were unexpected. In Experiment 4, there appeared to be no shifts in bud development types in the Russet Burbank cuttings with conditioning of mother plants. The effect of conditioning of mother plants on tuber size was positive for the four oldest plant ages, but negative for the two youngest plant ages. The effect of conditioning on the two youngest plant ages may be due to the cuttings from these plants producing exceptionally large leafy shoots which later became sufficiently induced to produce sizable

tubers.

In Experiments 1 and 2, with Norland cuttings, conditioning of mother plants increased the numbers of cuttings showing Type 3 development and decreased the numbers of cuttings showing Type 1 development. In Cutting Environment 1 (non-inductive), conditioning of mother plants generally had positive effects on tuber size. In Cutting Environment 2 (inductive), it generally didn't make any difference if cuttings were from conditioned mother plants or from non-conditioned mother plants. The observed effects on tuber size is likely due to bud development types. However, the reason for conditioned mother plants favoring Type 3 development in cuttings cannot be explained with current theories of levels of induction and bud development patterns

Leaf Positions

Leaf position designations gave an indication of the positions of leaves on a stem and a general indication of leaf age. Leaf position 1 from one plant age was not necessarily the same age as leaf position 1 from another plant. For this to be the case, there would have to be constant growth rates in all plants, regardless of age.

There were some limits placed on the experimental material used. For example, leaf position 1 was arbitrarily designated as the first leaf on a stem with an area greater than $30cm^2$. Also, for the range of plant dates used, it necessitated the use of only 8 leaf positions because some of the younger plants only had 8 leaf positions on a stem and some of the older plants only had 8 leaf positions that were not senescent. This removed some of the extreme effects that might be observed with very young (small leaf area) or very old (senescent) leaves.

There were some variations from experiment to experiment but the youngest leaf areas supported significantly less tuberization than the others. This may be partly due to leaf area effects, with young leaves having smaller leaf areas than

older leaves. It may be partly due to the rate of photosynthesis per unit leaf area of young leaves being low (Moorby 1978). While leaves are expanding, they tend to be net importers of photosynthate. The tendency of older leaf positions to produce smaller tubers than the middle leaf positions may be due to older leaves being less efficient and productive in terms of photosynthetic capacity.

There appeared to be no relationship between leaf position and the type of bud development. The observations of Simmonds (1965) and Lauer (1977) that leaf bud cuttings from the apex of the plant tended to produce roots and tubers, those from the midsection produced only tubers, and those from the basal portion produced the smallest tubers was not borne out by experiments in this study. An occasional stem could be found that showed these tendencies, but it was not common. This may be due to their observations being made under limited conditions or to the cuttings in this study being taken from plants that were started from physiologically old seed tubers.

Types of bud development in this study were most often influenced by the age of mother plants and cutting environments. Observations agreed more closely with Ewing's (1978) that all nodes on a given plant responded in approximately the same way.

Leaf Areas

Kahn (1982) positively correlated tuberization on stem cuttings with leaf area. Ewing (1978) found that cuttings with larger leaves seemed more likely to tuberize than cuttings with small leaves.

In the analysis of covariance, the 'r' values for the error term were significant for some experiments and not for others. Leaf area determinations were not done for all experiments and observations are being taken from a smaller base than would be preferred. Because plants were grown from tubers, the leaf areas of the cuttings in these experiments may be greater than with leaf bud cuttings taken from

cuttings.

However, it would appear that leaf areas had a significant effect on tuber size in experiments where all tubers were formed sessile in the leaf axil (= high 'r' values) or where an overwhelming majority of cuttings formed tubers sessile in the leaf axil (= lower 'r' values, but still significant). The exception to this is Experiment 7. However, there was initially severe wilting of the cuttings in this experiment, and some of the larger leaves did not recover as well as some of the smaller ones.

It is logical to think that where tuber enlargement is dependent on photosynthate produced by the original leaf that photosynthetic area would exert an influence. When a new leafy shoot forms, if formed before tubers are initiated, there is the possibility of correlating tuber size with the leaf area of the new leafy shoot. But if tuberization and leafy shoot growth occur much at the same time (due to the influence of the mother tuber of the original plant), or if rate of tuber growth (sink strength) exerts a significant influence (Nösberger and Humpheries, 1965), one would expect no correlation at all.

Mother Plant Age

Simmonds (1965) found that the ability of cuttings to root decreased with increasing mother plant age, but that tuber initiation increased with increasing mother plant age. Ewing (1978) found that there wasn't a great deal of difference between old and young mother plants in percent tuberization, but the older the mother plants, the larger the tubers formed on cuttings. Again, he was selecting for early tuberization of cuttings.

The effects of plant age in this study tended to be somewhat variable which may, in part, be due to leaf area effects and problems in the selection of mother plants. Plant ages in these experiments are not an absolute indication of the physiological age of a plant, but rather a relative ranking which indicates

relationships between plants within a given experiment.

Generally, for Norland plants, cuttings from younger plants produced larger tubers than cuttings from older plants. Younger plants showed more Type 3 development than older plants which almost all showed Type 1 development. This would reflect the different levels of induction in the plants.

For Russet Burbank plants, cuttings from the youngest and oldest plants produced the smallest tubers. The shift in Type 3 development in younger plants to Type 1 in older plants was not quite as clear cut as in Norland, but it still existed. The reason for the youngest plants producing smaller tubers than the slightly older ones would likely be due to lack of the tuberization stimulus or to time limitataions.

There were limits placed on the range of mother plant ages used by studying leaf positions at the same time. A particular mother plant age could only be used if it had at least 8 leaf positions. Different results may be obtained by using more extreme mother plant ages (eg. very old or very young).

All mother plants used in this study were grown from physiologically old mother tubers. It is likely that the mother tuber contributed some of the tuberization stimulus that was obviously present in the cuttings. Perhaps in mother plants grown from physiologically young tubers or from plants grown from cuttings where the mother tuber's influence is almost entirely non-existent as far as contributing tuberization stimulus is concerned, the situation might have been completely different. The plant would be entirely dependent on tuberization stimulus produced in the leaves. There may be difficulty in getting cuttings from young mother plants to tuberize at all. Cuttings from older plants may tuberize better.

Tubers From Leaf Bud Cuttings - the Future

Tissue culture techniques offer rates of multiplication that equal or exceed rates of multiplication from leaf bud cuttings (Goodwin 1981). With tissue culture techniques, in pathogen tested stocks, the chance of undetected re-introduction of

pathogens is minimal, and propagation is done in the reliable environment of the laboratory. It is possible that the use of leaf bud cuttings as a method of rapid propagation of selected stocks may become obsolete because of the advantages of using tissue culture techniques. However, for tissue culture, a relatively sophisticated level of technology is involved. The advantage of obtaining tubers from leaf bud cuttings is that it doesn't require sophisticated equipment or a high level of expertise, which may ensure its continued use.

In considering further research, one must remember that this method is now being successfully used and it is questionable whether it is worthwhile to expend the research time and energy needed to sort out the most complex effects. Some of the things that might be considered for further research are as follows:

1) The effects of different amounts of wilting on the tuberization of leaf bud cuttings could be studied to see if wilting does alter bud development patterns. To what extent does even a slight amount of wilting affect tuber size and cutting longevity?

2) Optimum irradiances for cutting establishment and then for maximum tuberization of cuttings could be established. Do cuttings establish better under relatively low irradiances and, once established, tuberize better under higher irradiances?

3) The performance of tubers from leaf bud cuttings could be evaluated. An optimum range of tuber sizes could be established. Do the perfectly formed types of tubers outperform the odd-shaped types with green leaflets?

4) Carbon dioxide enrichment of the soil zone could be studied with rooted cuttings.

5) Different fertilizer rates and timings could be investigated. In this study, fertilizer rates were found that enhanced tuber production. It would be expected that there would be inhibitory rates of fertilizer application as well. Optimum

rates and timings of fertilizer application could be established.

6) Tuberization in cuttings taken from plants grown from physiologically young seed tubers or from other cuttings could be studied. Effects of mother plant age, conditioning of mother plants and even leaf positions could be different than effects observed in this study.

7) The confounded factors that caused such great differences in cutting response to the different cutting environments in this study could be separated out. How much of the effects observed are due to differences in wilting and to differences due to inductive and non-inductive temperatures and photoperiods?

8) Experiments could be designed to specifically study the effects of leaf areas on sizes of tubers produced. Particular attention should be paid to patterns of bud development. Is there a good correlation between leaf area and tuber size only when sessile tubers develop?

9) The use of different soil media could be studied to see if soil media has any effect on tuber size and patterns of bud development.

10) Axillary buds could be measured when cuttings are first taken to see if different sizes of buds have an effect on tuber sizes and bud development patterns and therefore contribute to variability in an experiment.

11) Longevity of cuttings could be studied with particular emphasis on the relationship between cutting longevity and tuber size and the relationship between longevity and and bud development patterns, particularly the amount of rooting.

12) For studying plant age, stage of tuber development in the mother plant should be noted when cuttings are taken.

CONCLUSIONS

All plants used in this study were grown from physiologically old seed tubers. Cutting response, in terms of the sizes of tubers produced and bud development patterns, may be quite different for cuttings taken from plants grown from physiologically young seed tubers or from cuttings.

1) Fairly sizable tubers (up to 89 g in this study) may be produced by this method within a reasonable time limit.

2) A number of factors, such as leaf position, mother plant age, conditioning of mother plants and cutting environments, influence the sizes of tubers produced and these factors tend to interact with one another.

3) Generally, the cuttings from the middle leaf positions of a stem produced larger tubers than those from the youngest and oldest leaf positions.

4) For the range of plant ages used in this study, cuttings from the younger plants generally produced larger tubers than cuttings from older plants.

5) Mother plant age and cutting environments influenced the patterns of bud development more than the other factors. No relationship was found between leaf positions and bud development patterns.

6) With the different patterns of bud development, Types 2 and 3 appear to have a greater potential for the production of large tubers than Type 1.

7) Cuttings from young plants tended to produce more Type 3 development than cuttings from older plants which showed more Type 1 development.

8) Addition of fertilizer to the cuttings from young Norland plants can have a very positive effect on tuber size. The largest tubers produced in this study

were from cuttings with added fertilizer.

9) In experiments where there were large amounts of Type 1 development, leaf area of the original leaf appeared to be positively correlated with tuber size.

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APPENDIX

Table 9. Comparison of experiments - harvest dates, cultivars and cutting environments.

Experiment	Start Date	Harvest Date	Ctg Env[x]	Cultivar[y]
1	01/10/81	18/11/81	1,2	NOR
2	07/04/82	25/05/82	1,2	NOR
3	19/08/82	25/09/82 and 25/10/82	1,2,3	NOR
4	27/11/81	02/01/82	1,2	RB
5	08/07/82	17/07/82	2,3	RB
6	02/02/82	24/04/82	3/1	RB
7	05/06/82	07/07/82	1	NOR
8	19/08/82	25/10/82	1	NOR
9a	06/02/81	06/03/81	4	RB
9b	13/03/81	14/04/81	4	RB
9c	24/04/81	23/04/81	4	RB
10a	14/12/81	05/01/82	4	NOR
10b	29/01/82	30/02/82	4	NOR
10c	24/04/82	25/04/82	4	NOR

[x]See Materials and Methods pp.19 and 22 for descriptions of cutting environments.

[y]NOR=Norland; RB=Russet Burbank

Table 10. The analysis of variance for Experiment 1.[y]

Source of Variation	DF	F	
Cutting Environment (CE)	1	180.67**[z]	
Conditioning (C)	1	32.44**	
Mother Plant Age (A)	5	14.92**	
Leaf Positions (Po)	7	8.33**	
CE x C	1	14.04**	
CE x A	5	38.06**	
CE x Po	7	1.25ns	
C x A	5	3.84**	
C x Po	7	1.90ns	
A x Po	35	1.39ns	
CE x C x A	5	8.50**	
CE x C x Po	7	1.40ns	
CE x A x Po	35	1.04ns	
C x A x Po	35	0.90ns	s=1.37
			x=3.30g
CE x C x A x Po	35	0.27ns	cv=41.5%
Error	192		

[y]Analysis done on untransformed data.

[z]** indicates significance at the .01 level; ns indicates non-significance.

Table 11. The analysis of variance for Experiment 2.[y]

Source of Variation	DF	F	
Cutting Environment (CE)	1	85.07**[z]	
Conditioning (C)	1	35.38**	
Mother Plant Age (A)	6	9.22**	
Leaf Positions (Po)	7	8.97**	
CE x C	1	11.95**	
CE x A	6	15.12**	
CE x Po	7	0.35ns	
C x A	6	5.38**	
C x Po	7	0.99ns	raw data
A x Po	42	0.71ns	s=1.22
			x=2.92g
CE x C x A	6	3.29**	cv=41.9%
CE x C x Po	7	0.16ns	
CE x A x Po	42	0.99ns	$\sqrt{x+.5}$ data
C x A x Po	42	0.80ns	s=0.33
			x=1.80g
CE x C xA xPo	42	0.96ns	cv=18.3%
Error	224		

[y]Analysis done on transformed data $\sqrt{x+.5}$.

[z] ** indicates significance at the .01 level; ns indicates non-significance.

Table 12. The analysis of variance for Experiment 3.[y]

Source of Variation	DF	F	
			raw data
Cutting Environment (CE)	2	62.41**[z]	s=2.09
Plants (Pl)	3	1.35ns	x=2.64g
Leaf Positions (Po)	7	2.19ns	cv=79.1%
CE x Pl	6	2.13ns	$\sqrt{x+.5}$ data
CE x Po	14	0.88ns	s=0.49
Pl x Po	21	1.05ns	x=1.59g
			cv=31.3%
Error	42		

[y]Analysis done on transformed data $\sqrt{x+.5}$.

[z] ** indicates significance at the .01 level; ns indicates non-significance.

Table 13. The analysis of variance for Experiment 4.[y]

Source of Variation	DF	F	
Cutting Environment (CE)	1	189.71**[z]	
Conditioning (C)	1	12.70**	
Mother Plant Age (A)	5	7.73**	
Leaf Positions (Po)	7	6.79**	
CE x C	1	1.24ns	
CE x A	5	6.12**	
CE x Po	7	4.09**	
C x A	5	11.42**	
C x Po	7	0.85ns	raw data
A x Po	35	0.81ns	s=1.19
			x=1.78g
CE x C x A	5	1.82ns	cv=66.8%
CE x C x Po	7	0.62ns	
CE x A x Po	35	1.24ns	$\sqrt{x+.5}$ data
C x A x Po	35	1.19ns	s=0.35
			x=1.43g
CE xC xA xPo	35	0.98ns	cv=24.3%
Error	192		

[y]Analysis done on transformed data $\sqrt{x+.5}$.

[z]** indicates significance at the .01 level; ns indicates non-significance.

Table 14. The analysis of variance for Experiment 5.[y]

Source of Variation	DF	F	
Cutting Environment (CE)	1	206.91**[z]	
Conditioning (C)	1	17.30**	
Mother Plant Age (A)	5	4.08**	
Leaf Position (Po)	7	2.29*	
CE x C	1	1.62ns	
CE x A	5	4.01**	
CE x Po	7	1.16ns	
C x A	5	3.19*	
C x Po	7	0.70ns	raw data
A x Po	35	1.41ns	s=1.99
			x=2.41g
CE x C x A	5	0.26ns	cv=82.6%
CE x C x Po	7	1.93ns	
CE x A x Po	35	0.99ns	$\sqrt{x+.5}$ data
C x A x Po	35	0.73ns	s=0.51
			x=1.57g
CE xC xA xPo	35	0.60ns	cv=31.2%
Error	192		

[y]Analysis done on transformed data $\sqrt{x=.5}$.

[z]*indicates significance at the .05 level; ** indicates significance at the .01 level; ns indicates non-significance.

Table 15. The analysis of variance for Experiment 6.[y]

Source of Variation	DF	F	
Stems (St)	2	2.04ns[z]	
Plants (Pl)	3	2.98ns	
Leaf Positions (Po)	7	2.31*	
Pl x St	6	2.19ns	
St x Po	14	1.53ns	s=7.72
Pl x Po	21	1.60ns	x=10.02g
			cv=77.1%
Error	42		

[y]Analysis done on untransformed data.

[z] * indicates significance at the .05 level; ns indicates non-significance.

Table 16. The analysis of variance for Experiment 7.[y]

Source of Variation	DF	F	
Fertilizer (F)	1	41.93**[z]	
Conditioning (C)	1	1.13ns	
Mother Plant Age (A)	3	30.20**	
Leaf Position (Po)	7	3.67**	
F x C	1	1.93ns	
F x A	3	2.00ns	
F x Po	7	1.87ns	
C x A	3	7.67**	
C x Po	7	0.60ns	raw data
A x Po	21	1.33ns	s=1.61
			x=2.03g
F x C x A	3	4.33**	cv=79.3%
F x C x Po	7	0.67ns	
F x A x Po	21	0.73ns	$\sqrt{x+.5}$ data
C x A x Po	21	1.00ns	s=0.39
			x=1.52g
F xC xA xPo	21	0.53ns	cv=25.6%
Error	256		

[y]Analysis done on transformed data $\sqrt{x+.5}$.

[z] ** indicates significance at the .01 level; ns indicates non-significance.

Table 17. The analysis of variance for Experiment 8.[y]

Source of Variation	DF	F	
Fertilizer (F)	1	8.12**[z]	
Leaf Position (Po)	7	0.44ns	
F x Po	7	0.41ns	s=7.72g
			x=2.26g
Error	16		cv=96.9%

[y]Analysis done on transformed data $\sqrt{x+.5}$.

[z] ** indicates significance at the .01 level; ns indicates non-significance.

Table 18. The analysis of variance for Experiment 9.[y]

Experiment	Source of Variation	DF	F	
9a	Treatment	1	2.95ns	s=0.45 x=1.22g
	Error	28		cv=36.8% Tab F= 4.18
9b	Treatment	1	2.11ns	s=0.44 x=1.56g
	Error	28		cv=28.2% Tab F = 4.18
9c	Treatment	1	3.17ns	s=0.54 x=1.91g
	Error	28		cv=28.3% Tab F = 4.18

[y]Analysis done on untransformed data.

[z] ns indicates non-significance.

Table 19. The analysis of variance for Experiment 10.[y]

Experiment	Source of Variation	DF	F	
10a	Treatments	3	0.54ns	s=0.58
				x=0.95g
	Error	60		cv=61.1%
10b	Blocks	3	0.82ns	
	Treatments	3	0.12ns	s=1.11
				x=1.67g
	Error	57		cv=66.0%
10c	Blocks	3	0.56ns	
	Treatments	3	0.31ns	s=1.05
				x=1.71g
	Error	57		cv=61.4%

[y]Analysis done on untransformed data.

[z] ns indicates non-significance.