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EFFECT OF NITROGEN FERTILIZER PLACEMENT ON NITROGEN UPTAKE AND YIELD OF SWEET CORN (Zea mays L. saccharata)

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ABSTRACT

Five placements of nitrogen fertilizer applied to sweet corn (Zea mays L. saccharata) at the four fully expanded leaf stage, that is control (no nitrogen), a band of nitrogen placed on the soil surface near the row, on the soil surface between the rows, at 3 cm depth between the rows and at 10 cm depth between the rows were studied following three sowing times. Total plant nitrogen and sap nitrate were determined along with total plant dry weight at six growth stages. Leaf extension and leaf appearance were also followed in order to monitor the response of plants to nitrogen fertilizer applied.

Nitrogen fertilizer application resulted in significantly higher nitrogen uptake, plant dry weight and marketable ears under both dry and wet conditions. Nitrogen fertilizer applied at 10 cm depth between rows resulted in significantly higher nitrogen uptake, plant dry weight and marketable ears than that applied on the soil surface between rows under dry condition. Nitrogen fertilizer applied on the soil surface near the plants performed well under both dry and wet conditions. sap nitrate test was more sensitive than total nitrogen measurement in Sap nitrate levels were indicating the timing of nitrogen uptake. influenced by nitrogen fertilizer application and soil water content. The general critical value of sap nitrate over the vegetative growing period was about 1000 ppm. The sap nitrate test appeared to be a very useful monitoring tool for plant nitrogen status. Further studies in the uses of sap nitrate test, especially the critical value, are needed. Use of leaf extension to detect the response of plants to nitrogen fertilizer applied was not successful. Nitrogen fertilizer application tended to accelerate leaf appearance under the low soil nitrogen status.

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CHAPTER ONE

INTRODUCTION

Subsistence farming relying on rainfed monocropping has been traditionally practised by farmers in many cultivated areas in Thailand. In recent years, however, some cropping systems, for example, double cropping, have been developed and practised by some farmers. Unfortunately double cropping is often difficult as the pattern of rainfall is rather erratic, especially in most parts of the Northeastern Region of Thailand. In this region, the amount of rainfall drops sharply during the later period of the rainy season resulting in a declining soil moisture regime for the second crops. In order for double cropping to be successful, suitable second crops of short duration combined with good management are needed.

Among the promising second crops, sweet corn ($\underline{\text{Zea}}$ $\underline{\text{mays}}$ L. saccharata) seems to perform well in many areas. Apart from the relatively short duration required by this crop, the high demand for sweet corn locally and from processing plants in some areas, has made sweet corn an increasingly important crop in recent years.

Sweet corn needs a good supply of nitrogen fertilizer to achieve a high yield. At the present time nitrogen fertilizer is relatively expensive compared with the returns from produce sold. Moreover, the erratic rain causes the plants to respond variably to the applied nitrogen fertilizer. Consequently fertilization sometimes results in a very low profit to the farmers.

Rainfall pattern, especially the distribution, is rather erratic and unpredictable. This causes fluctuation and difference in soil water content between soil layers. Consequently this may affect the availability of nitrogen fertilizer applied. So that the proper placement of nitrogen fertilizer may be needed for maximum and

efficient nitrogen supply to plants.

It is often found that the residual fertility from the first crop is sufficient for the short duration second crop, or at least for the early growth (Jones, 1974; Rao et al., 1983; Sanmaneechai et al., 1984). Moreover, in practice, there is a problem of labour shortage occurring during the end of the first crop and the beginning of the second crop. Therefore this study examined the methods of nitrogen fertilizer application after crop establishment.

As the efficient use of nitrogen fetilizer is important for profitability, it was thought necessary to examine methods that could be used to monitor nitrogen supply to the plants and to detect the response of plants to nitrogen fertilizer applied. Among the many methods available, nitrate sap tests and measurement of leaf growth are some of the promising methods because they are quick and simple. Both these techniques were included in this study.

Although the distribution of rainfall in Palmerston North, New Zealand is relatively uniform throughout the year (N.Z.M.S., 1983) compared with that of Thailand, the latest 11 year rainfall data show that there were some dry spells during the summer months. Therefore the conditions at Palmerston North may be used to simulate those of Thailand through the appropriate experimental planning and design. This study wanted to simulate sweet corn as second crop in a double cropping system and under a declining rainfall situation. The objectives of this study were to study:

- 1. nitrogen uptake and dry matter yield of sweet corn as affected by nitrogen fertilizer application.
- 2. the effect of different nitrogen fertilizer placements on nitrogen uptake, dry matter and marketable sweet corn yield.
- 3. the nitrogen dynamics in the plants by using sap nitrate tests and the relationship between sap nitrate levels and nitrogen uptake.

4. the possibility of using leaf extension and leaf appearance to detect the response of plants to the nitrogen fertilizer applied.

CHAPTER TWO

LITERATURE REVIEW

Because of few references about sweet corn in the area of study, most of the literature reviewed is concerned with maize, as they both have the same morphological and physiological characteristics.

2.1 GENERAL ASPECTS OF NITROGEN

2.1.1 Functions of Nitrogen in Plants

The major functions of nitrogen in plant growth include (i) to act as a component of the chlorophyll molecule, (ii) a component of amino acids (the building blocks of proteins) (iii) carbohydrate utilization, (iv) components of enzymes and (v) support the uptake of other nutrients (Donald et al., 1963; Olson and Kurtz, 1982).

2.1.2 Nitrogen Deficiency and Excessive Nitrogen

A certain level of nitrogen must be present in plant cells for optimum utilization of carbohydrates produced during photosynthesis. Nitrogen deficiency affects the chlorophyll content (Thompson and Troeh, 1978) and causes excessive deposition of carbohydrates in vegetative cells with consequent thickening of the cell wall, thereby limiting formation of protoplasm (Olson and Kurtz, 1982). Nitrogen deficiency also limits the production of protein and other materials essential for the production of new cells. This causes a decrease in cell size and decreases cell division (Devlin, 1975). Consequently plants appear spindly, stunted and pale compared with healthy plants (Thompson and Troeh, 1978).

Paleness caused by nitrogen deficiency is usually most pronounced in the older leaves, especially along the veins. The symptom appears

last in the younger leaves, because of the high mobility of nitrogen in the plants (Devlin, 1975). That is, part of the nitrogen from the deficient areas is translocated and used in the other plant parts that are still growing (Arnon, 1975; Thompson and Troeh, 1978). In maize, the V-shaped pale symptom usually starts from the tip of the lower leaves with the sharp end progressing inward along the midrib of the leaf. This can cause the premature senescence of these leaves (Arnon, 1975; Donald et al., 1963).

Plants can take up excessive nitrogen (more than that required at the time), especially if some factors, for example, phosphorus, or potassium are inadequate (Thompson and Troeh, 1978). Under these conditions, there is a tendency for increased leaf cell number and cell size resulting in an overall increase in leaf production (Morton and Watson, 1948). The plants generally produce dark and succulent vegetative growth. In some cases, vegetative growth may be at the expense of seed production in grain crops. These effects of excess nitrogen on growing plants can be lessened if the phosphorus and potassium supplies are adequate for the rate of growth produced. High levels of these two elements help to avoid the succulent vegetative growth and the delay in maturity that can be caused by excess nitrogen (Thompson and Troeh, 1978).

2.1.3 Sources and Forms of Nitrogen

Even though the atmosphere is about 80% nitrogen, most higher plants (except some legumes) are unable to utilize this molecular nitrogen (N_2) until it is chemically combined with hydrogen, oxygen, or carbon (Bray, 1983; Thompson and Troeh, 1978). Most soil nitrogen reserve is in organic matter, but unfortunately most of it has complex molecules and is unavailable to higher plants unless broken down by soil microorganisms into inorganic ions (Thompson and Troeh, 1978). Consequently the amount of available nitrogen in the soil is usually only a small fraction of the total nitrogen and it may be insufficient to meet plants' requirement during a growing season (Scarsbrook, 1965). In order to maximize crop production, nitrogen is often supplemented to

the plants through inorganic, or organic nitrogen fertilizer application. Most of the inorganic nitrogen fertilizers can provide either ammonium $(\mathrm{NH_4^+})$, or nitrate $(\mathrm{NO_3^-})$, or both. Therefore, inorganic nitrogen is the principle form of nitrogen taken up by plants.

2.1.4 Nitrogen Translocation in Soil

Plants obtain nitrogen either by the root approaching the source of nitrogen, or nitrogen is translocated from the original site to plant roots. These mechanisms are as follows.

2.1.4.1 Root interception

As plant roots grow, they extend into new areas of soil where they meet, or intercept ions that are there in the soil solution. This root extension also decreases the distance needed for plant nutrients to move by mass flow, or diffusion to arrive at the root surface (Donahue et al., 1977). Generally the amount of nutrients which directly contact plant roots is small compared with the total nutrient demand. This is particularly true for nutrients required in high quantities (Mengel and Kirkby, 1982). Therefore this mechanism is less important for nitrogen supply. It was estimated that the supply of nitrogen to maize roots by this mechanism in soils was only 1.2% (Donahue et al., 1977).

2.1.4.2 Mass flow

The continual absorption and transpiration of water by plants mean that large volumes of water move through the soil. This causes dissolved nutrients, for example, nitrate which is highly mobile, to move along with water through the soil to plant roots (Donahue et al., 1977). Therefore this process is dominant when transpiration is high and is important for nutrients present in soil solution in high concentration, for example, nitrate (Mengel and Kirkby, 1982). It has been shown that mass flow accounted for 98.8% of the total nitrogen

absorbed by maize roots (Donahue et al., 1977).

2.1.4.3 Diffusion

Dissolved nutrients move toward the roots without water flow because of the laws of diffusion. When the solute is absorbed by plant roots at a relatively greater rate than water, then the ion concentration at the rhizosphere must fall. Consequently ions diffuse from a higher concentration towards the root surface which has a lower concentration by random thermal motion (Mengel and Kirkby, 1982). It has been reported that this mechanism played a negligible role in supplying nitrogen to maize plants (Donahue et al., 1977), but Mengel and Kirkby (1982) state that diffusion is the main mechanism for nutrients which are present in the soil solution in low concentrations, for example, ammonium.

2.1.4.4 Some factors affecting nitrogen translocation

(i) Soil water content. Generally transpiration and evaporation increase at high soil water content and optimum above ground conditions like low relative humidity. This causes rapid water movement from the soil through plant roots and increasing ion transport to plant roots by mass flow (Mengel and Kirkby, 1982).

Lower soil water content can affect the mode and rate of ion transportation. As the soil dries out, the solution becomes more concentrated. Some of its ion species may even reach concentrations higher than their solubility products (Mengel and Kirkby, 1982). For example, the concentration of an unabsorbed ion like nitrate will be increased (Nye and Tinker, 1977). Moreover, the decrease in soil water results in an increase in air filled pores. Water channels leading from the bulk soil to plant roots are affected. The cross sectional area of water allowing diffusion is reduced and also the pathways from the bulk soil to plant roots become less direct, or more tortuous. Consequently rate of diffusion is decreased by increased diffusive resistance (Mengel and Kirkby, 1982).

(ii) Soil type. In general, fine-textured soils would be expected to permit faster diffusion at the same bulk solution concentration than coarse-textured soils. This is because of their greater water holding capacities at equivalent soil water potentials (Corey, 1973).

Nutrient buffer capacity of soil also plays an important role. Soil with a higher nutrient buffer capacity can supply adequate nutrients to soil solutions. Consequently ion supply to plant roots by diffusion can be maintained at higher levels. This should be true for ammonium, but not in the case of nitrate supply (Mengel and Kirkby, 1982). This is because nitrate is weakly held on soil colloids. Therefore nitrate supply to plant roots by mass flow should be high under conditions of high soil water movement.

(iii) Ion concentration. The concentrations of the ion species in the soil solution can differ widely depending on soil properties. Generally the nitrogen concentration is rather high, as Larsen and Widdowson (1968) found that nitrate was the anion present in highest concentration. This makes nitrate the main form to be transported to plant roots by mass flow (Harmsen and Kolenbrander, 1965).

Nutrient transportation by diffusion depends largely on the concentration gradient in soil solution. The rate of diffusion will be higher with a greater concentration gradient, for example, in soils with higher nutrient level. If the concentration at the root surface is higher, the reverse is true (Mengel and Kirkby, 1982).

2.1.5 Nitrogen Absorption by Plant Roots

2.1.5.1 Nitrogen preferential absorption

Nitrogen is unlike other nutrients in that it is absorbed either as a cation (NH_4^+) , or an anion (NO_3^-) . Generally there is no preferential absorption in maize seedlings, but in some cases there can be a higher concentration of one ion resulting in lower absorption rate of the other ion (Warncke and Barber, 1973). Under favourable

conditions, both forms of nitrogen are effectively utilized by plants (DeKock, 1970; Zsoldos, 1971). Mills and McElhannon (1982) reported in sweet corn (grown in solution culture with a nitrogen ratio of 50% nitrate - 50% ammonium) that the absorption rates of both forms of nitrogen was similar up to the tasselling stage. Later in the maize development, uptake of nitrate was more than ammonium and nitrate may account for up to 90% of the total nitrogen uptake (Kurtz and Smith, 1966; Yoneyama et al., 1977).

2.1.5.2 Mechanisms

Among many theories proposed to explain ion absorption, the active (an ion moves to a site which has the same charge) and passive (an ion moves to a site which has the opposite charge) transport theory seems to be the most applicable. It suggests that ions in solution are subjected to two main 'forces'. One force arises from the chemical potential gradient and the other from the electrical gradient. Ions move down a chemical gradient, that is, from a higher concentration to a lower concentration zones. For ions acted upon by electrical gradient, cations are attracted to а negative electropotential, whereas anions are attracted to electropotential. Ion movement is thus dependent electro-chemical potential gradient (Mengel and Kirkby, 1982). the root surface is negatively charged (Yoneyama et al., 1975), this implies that nitrate which is an anion is more subjected to active transport than ammonium which is a cation. Ammonium is likely to be taken up by passive transport, that is it is attracted to negative electropotential in the cell (Mengel and Kirkby, 1982). Yoneyama et al. (1975) showed that ammonium was clearly adsorbed on the maize root surface, but nitrate was not.

The uptake of these ions tends to alter electro-chemical balance in the rhizosphere. In order to balance this, the plant excretes OH^- , HCO_3^- and H^+ ions in exchange for nitrate and ammonium ions taken up respectively (Donahue et al., 1977).

In addition to the absorption by roots, leaves also can absorb gaseous nitrogen through stomata (Hocking et al., 1984; Kramer, 1969), but this is probably negligible, except after foliar application of nitrogen fertilizer.

2.1.5.3 Some factors affecting nitrogen absorption

(i) Ion factors. There are some antagonistic reactions in ion absorption. The most common effect is between nitrate and chloride (Cl⁻). Higher concentration of chloride in the nutrient medium lowers the nitrate uptake (Mengel and Kirkby, 1982). This is probably because it strongly competes for the same carrier site with related ion species.

The rate at which an ion species is absorbed is also dependent on its concentration in the nutrient medium. That is, the higher the ion concentration, the higher the absorption rate (Mengel and Kirkby, 1982). As Warncke and Barber (1973) found that increased ammonium concentration lowered the uptake rate of nitrate, whereas increased nitrate concentration resulted in the lower uptake rate of ammonium. This relationship is not linear but follows an asymtotic curve (Mengel and Kirkby, 1982).

(ii) Soil factors. Low soil water content can cause low nitrogen uptake. That is, the closure of stomata due to lack of water supply can cause lower photosynthetic activity due to lower carbon dioxide supply. Since nitrate uptake is greatest when there is abundant photosynthate available to roots (Jackson et al., 1976). This can reduce the uptake of nitrogen.

Under waterlogging conditions, oxygen content in the soil will be reduced. It is found that when the oxygen concentration falls beneath the optimum level, the structure, or morphology of the root system is profoundly altered. This influences the ion absorption (Drew, 1979).

In general, the root temperatures of lower than 10°C, or greater

than 40°C retard uptake of nitrogen (Shaw, 1976). Yoneyama et al. (1977) found that the inhibitory effect of low temperature was more drastic on nitrate uptake than on ammonium uptake.

(iii) Plant factors. The permeability of the plant cell membrane has an influence on the absorption of different ion species. That is, the proportion of ions transported across the cell membrane depends on the specific permeability of membrane to particular ion species. This is related to the components which make up the membrane and the enzymes present in that membrane (Mengel and Kirkby, 1982). In the case of nitrogen, both ammonium and nitrate are the same ion species, that is monovalent ions (Salisbury and Ross, 1978). Therefore the selectivity of cell membrane to these ions should be small.

Uptake of nitrogen should vary at different growth stages, part of this is related to the growth of roots. During the seedling stage, nitrogen uptake is low (Arnon, 1975), probably because of the smaller root system and smaller demand. As plants grow, the root system also grows correspondingly. The area of root surface reaches a maximum at tasselling (Arnon, 1975) and the growth of roots ceases approximately at the beginning of grain development (Foth, 1962). This implies that nitrogen uptake should be greatest around tasselling because of the ageing of roots and the lack of new soil volume being explored, combined with increased depletion of nutrients within the soil volume being utilized.

2.1.6 Nitrogen Translocation and Assimilation in Plant

After ammonium and nitrate have been absorbed into plant roots, they will be translocated to plant parts, reduced (transformed) into organic forms and incorporated into proteins before being utilized by the plant. The steps are as follows.

2.1.6.1 Nitrate reduction

This process is to reduce nitrate into nitrite (NO_2^-) by enzyme nitrate reductase (NR) and takes place in either roots, or leaves. Nitrate absorbed, if it is not reduced in the roots, will be translocated through xylem to the shoot and reduced there (Novoa and Loomis, 1981). Ivanko and Ingversen (1971) found that nitrate was the major component of the xylem sap in 20 day old maize plants. The transport of nitrate ions from the roots to the shoot has a close relationship with light, or transpiration conditions (Martin, 1970).

Generally in young plants (non-legumes), nitrate is reduced mainly in the leaves, but progressively more is reduced in the roots as plants develop (Hocking et al., 1984). It has been found in 8-14 day old maize seedlings that the main accumulation of nitrate reductase enzyme is in the leaves and this suggests that the seedlings reduce most of the nitrate they acquire in the leaves (Shaner and Boyer, 1976; Wallace, 1973).

Nitrate reductase activity (NRA) is regulated by some factors. In maize, nitrate supply to leaves is a major determinant of the level of nitrate reductase activity (Shaner and Boyer, 1976). The activity may increase, or decrease sharply and irregularly during growth (Deckard et al., 1973) and may be variably increased by application of nitrogen fertilizer (Hocking et al., 1984). It is proposed that control of nitrate supply to the active site of the enzyme by membrane functions may be the important nitrate reductase activity regulatory step in leaves of all species (Steer, 1979). In the roots, high carbohydrate supply from the shoot usually favours nitrate reductase activity there (Fowden, 1979), probably due to the greater energy supply (Novoa and Loomis, 1981).

2.1.6.2 Nitrite reduction

The further step is the reduction of nitrite to ammonium (NH_4^+) . This process occurs in chloroplasts and is brought about by enzyme nitrite reductase. Both nitrate reductase and nitrite reductase function in series, so that no appreciable nitrite accumulation occurs. Eight electrons are required for the reduction, therefore the process requires a source of energy (Kirkby, 1981).

After this stage, ammonium derived from nitrite reduction, or taken up by roots will be metabolized to produce ammonia (NH₃) which is a neutral organic molecule (Kirkby and Hughes, 1970). Usually most ammonium is rapidly assimilated to amino acids and amides in the roots (Novoa and Loomis, 1981; Yoneyama and Kumazawa, 1974).

2.1.6.3 Assimilation of ammonia

Whatever the nitrogen sources to plants, all nitrogen absorbed must be reduced to ammonia which is the starting organic molecule for all nitrogen-containing organic compounds in the plants. That is, ammonia will be incorporated with derivatives of carbohydrate supplied from photosynthesis and form such low molecular organic fraction as amino acids, amides and amines (Donald et al., 1963; Gillet, 1983; Kirkby, 1981). After that, amino acids (not all of them in the plant) are then assembled in specific sequences to form different proteins which are the high molecular organic fractions (Donald et al., 1963; Kirkby, 1981).

Amino acids can be transported circularly between roots and leaves. That is, amino acids formed in the roots may be transported through xylem and added with those in the leaves and polymerized into proteins there. Similarly amino acids in the leaves may be transported through phloem to the roots and polymerized there (Novoa and Loomis, 1981). The assimilated nitrogen in proteins may be released as amino acids from protein hydrolysis and then reassimilated into protein several times within the plant before it is finally incorporated into seed protein (Kirkby, 1981).

2.1.7 <u>Distribution and Redistribution of Assimilated Nitrogen to Plant</u> Parts

The classical study of distribution of nitrogen to different parts of maize plant is probably the work done by Hanway (1962b). It shows that differences in soil fertility generally do not markedly change the pattern of uptake and distribution of nitrogen in the plant, but that the amounts distributed in each plant part are different, especially at various growth stages. This is due to the differences in nitrogen absorbed, photosynthates produced and the size of 'sources' and 'sinks' in the plant (see section 2.3.1).

During the period to silking, nitrogen in the vegetative parts increases and reaches maximum before silking, with leaves containing the highest amount of nitrogen (approximately 30% of the nitrogen accumulated by the plant at maturity - Hanway, 1962b). After silking, nitrogen in the vegetative parts, possibly including roots, also decreases due to a breakdown of the proteins by proteolysis. It is estimated that about 65% of the nitrogen present in the vegetative parts is translocated to the ears (Kissel and Ragland, 1967). Leaves contain only 13% of the total plant nitrogen at grain physiological maturity stage (Hanway, 1962b).

Nitrogen in the grains increases rapidly after grain formation and continues until they are mature. Approximately two-thirds of total nitrogen absorbed is accumulated in the grains (Arnon, 1975; Friedrich and Schrader, 1979; Hanway, 1962b). The increase of grain nitrogen is largely due to the translocation of nitrogen from other plant parts (Below et al., 1981; Hanway, 1963). It is estimated that about one-half of grain nitrogen is translocated from the above-ground parts of the plant (Arnon, 1975; Hanway, 1962b), mainly from the bottom leaves and stem (Kissel and Ragland, 1967). However, other plant parts, for example, cobs and husks also contribute nitrogen to the grains and the contribution from these parts (including stem) preceeds that from the leaves (Arnon, 1975; Hanway, 1962b). It is estimated that nitrogen contributed to grains from leaves, stem and other parts,

husks and cob are 59, 23, 12 and 7% respectively (Carlès et al., 1957; Hay et al., 1953).

Nitrogen translocation patterns vary for the various plant parts, for example, shank, husks and cob are initially nitrogen sinks and then function as nitrogen sources during grain filling (Crawford et al., 1982). The retranslocation often begins at anthesis (Hocking et al., 1984) and mostly occurs during the reproductive period (Arnon, 1975) enabling developing leaves and storage organs to receive additional nitrogen (Arnon, 1975; Hewitt, 1970) and therefore is of considerable importance for grain development (Hocking et al., 1984). Although the carbohydrate supply from photosynthesis is sufficient for ear demand, nitrogen taken up from the soil is inadequate for the initiation and filling of kernels without redistribution of vegetative nitrogen (Swank et al., 1982). Even when nitrate is supplied to maize throughout the growth period, the decline of nitrate nitrogen in the plant still occurs, suggesting that this is triggered by factors other than nitrogen availability (Hocking et al., 1984), or the presence of an ear, as the stem can act as an alternative sink for nitrogen redistribution from leaves (Christensen et al., 1981). Nitrogen redistribution is often increased in nitrogen stressed plants (Hocking et al., 1984) resulting in greater use of stored nitrate from all organs, especially roots and stems (Friedrich et al., 1979). High uptake of nitrogen in the pre-grain filling period followed by a period of low nitrogen uptake may also increase nitrogen redistribution from vegetative parts to grain (Pollmer et al., 1979).

2.2 PLANT NITROGEN MEASUREMENTS

In general, the purpose of plant analysis is to diagnose the nutrient status in the plants associated with deficiency, adequacy and toxicity, so that fertilizer responses can be evaluated by a comparison of analysis results with the critical nutrient values (Aldrich, 1973). Furthermore, the analysis is employed to indicate whether applied nutrients entered the plant and to indicate interactions, or antagonisms among nutrients as an aid to the understanding of the

internal functions of plants (Aldrich, 1973). Analytical results of plant analysis need to be calibrated with crop performance and response to fertilizer application. Standard, or optimal values as well as deficient, low and excess ranges for the nutrients must be established and can be used as guidelines for crop production (Ceesay, 1980).

There are several methods used for plant nitrogen measurement as follows.

2.2.1 Total Nitrogen Determination (Quantitative Methods)

These methods measure both the nitrogen that has already been incorporated into plant tissues and that which is still present as soluble constituents of the plant sap (Aldrich, 1973). The measurement can be made for total nitrogen in the whole plant, or selected plant In general, the percentage of total nitrogen parts as required. increases from the bottom to the top of maize plant (Hanway, Ear leaf at silking time is often selected for nitrogen measurement as it indicates well the nutrient status of the plant and is related grain yield (Arnon, 1975; Hanway, 1962a; Kurtz and Smith, 1966). is proposed that, for example, the optimum ear leaf concentration for New Zealand maize ranges from 2.3-3.3% (Cornforth and Steele, 1981). However, the results obtained are too late for any corrections in nitrogen fertilizer to be made to the current crop.

Numerous methods have been developed for these analyses. Most procedures involve ashing of tissue to destroy the organic component leaving various elements for analysis. Two methods have gained general acceptance for determination of total nitrogen, that is the Kjeldahl method which is essentially a wet-oxidation procedure and the Dumas method which is fundamentally a dry-oxidation (that is combustion) technique (Bremner, 1965). The Dumas procedure has not been widely adopted because of the time required per analysis and the investment required for equipment. Macro-Kjeldahl procedures have been widely used for total nitrogen analysis of plant materials, however, semi-micro modifications have been gaining favour in recent years

because of the increased speed of analysis, the decreased cost per sample and the reduced volumes of corrosive acid and alkali that are handled by the analyst (Nelson and Sommers, 1973).

The two-step Kjeldahl procedure described by Bremner (1965) has proved satisfactory for total nitrogen analysis of most of the nitrogenous compounds known to occur in plant material (Bremner, 1965). In recent years, a number of modifications have been suggested and a more rapid procedure devised by using Auto-Analysers. The results are expressed in terms of concentration, for example, per cent of total nitrogen in the plant samples.

2.2.2 Sap Nitrate Test (Rapid Tissue Test)

The principle of this method is that plants take up most of their nitrogen from soil as nitrate. In most species nitrate is translocated in this form to the leaves to be reduced and assimilated there. means nitrate is a raw material for leaves growing concentration in the plant is a very sensitive indication of the ability of the plants to find enough nitrogen to meet their immediate demands (Scaife and Stevens, 1977). If nitrogen supply exceeds demand, nitrate accumulates in the plants. When the reverse is true, nitrate rapidly disappears from plant tissues (Cornforth, 1980; Donald et al., 1963; Scaife, 1979). Therefore this method measures the unassimilated nitrate in plant sap. The results obtained are used to indicate nitrogen status of the plants. Some crops reduce most of their nitrogen in the roots, so very little nitrate is to be found in shoots. For example, it has been reported that about 50% of all absorbed nitrate by four weeks old maize was assimilated in the roots (Keltjens et al., 1986). In general, nitrate is higher in stems and petioles than in leaves (Viets, 1965). In maize, nitrate concentration usually decreases from the bottom to the top of plant (Hanway, Nelson, 1956). Nitrate concentration changes with time, that is the older the plant, the lower the nitrate concentration in the sap. Scaife (1979) suggested adequate levels of sap nitrate over time for nearly all fast-growing crops are as follows.

First quarter of life: 4000 ppm nitrate

Second quarter of life: 3000 ppm nitrate

Third quarter of life: 2000 ppm nitrate

Last quarter of life: 1000 ppm nitrate

Clarke <u>et al</u>. (1986) suggest the critical values of nitrate nitrogen (NO_3-N) in plant sap (in stem base) of 1100-1600 and 600-1000 ppm for sweet corn at 6 and 8 weeks from sowing respectively.

A simple method for measuring nitrate in all aqueous media has been devised enabling nitrate levels in plant sap to be measured and enabling deficiencies to be diagnosed before they become visible in the Scaife and plant (Cornforth, 1980; Scaife, 1979; Bray. Withers, 1982). They are test strips ('Merckoquant') consisting of a thin plastic strip to which is attached two zones of white filter impregnated with an aromatic N-(1-naphthyl)ethylenediamine. The filter paper at the further (end) zone contains a reducing agent and indicates both nitrate and nitrite, whilst the nearer zone only nitrite. This serves as a warning zone for interfering nitrite. When the further zone is briefly wetted with a nitrate solution, such as plant sap squeezed from leaf, or stem, then the nitrate is reduced to nitrite. It produces nitrous acid in the presence of an acid buffer, the nitrous acid then diazotizes an aromatic amine and coupling with N-(1-naphthyl)ethylenediamine produces a red-violet azo dye (Merck, 1986).

Two minutes are allowed for colour development, then the developed colour is read by comparing with the standard colours printed on the tube. The levels of concentration shown are 0, 10, 30, 60, 100, 250 and 500 mg/l (ppm) nitrate. In general, sap nitrate levels of most young crops are above the 500 ppm nitrate standard shown on the tube (Scaife and Bray, 1977). But by accurately recording the time in seconds for the colour to develop to that of the 500 ppm standard, the

nitrate concentration in the sap can be estimated with sufficient accuracy as this time is related to concentration (Scaife and Stevens, 1977; Scaife and Stevens, 1983).

Since sap nitrate depends to a large extent on the balance between uptake from the soil and the supply of carbohydrates for making proteins, this means it can be influenced by such factors as light intensity and soil water content (Conforth, 1980; Donald et al., 1963; Viets, 1965). It is desirable to avoid conditions when carbohydrate supply could be limiting and therefore the tests will be most reliable if done under sunny conditions and after mid-morning (Cornforth, 1980).

2.2.3 Plant Nitrate Measurement

Plant nitrate status can be used to reflect the cultural practices and also related to crop growth and dry matter production. It has been found that nitrate nitrogen concentration in some cereals is a function of rate of nitrogen fertilization (Baker and Tucker. Moreover, measurements can also Papastylianou and Puckridge, 1981). indicate the relative availability of nitrogen from (Papastylianou and Puckridge, 1981). In maize, it is reported that nitrate tests have a highly significant, positive correlation with total nitrogen in all the plant parts and nitrogen uptake (Boawn et al., 1963; Hanway, 1962c). To achieve high yield levels, it has been estimated that there should be about 1000 ppm of nitrate nitrogen in the midrib of the sixth leaf at the silking stage (Fuehring, This 1000 ppm nitrate level is associated with the 3.0% of total leaf nitrogen which is a critical level for maize (Hanway, 1962c).

Two laboratory methods are commonly used to measure plant nitrate. Plant material must be dried and ground and extracting solution added to the samples, then shaken and filtered. The nitrate nitrogen in the extracts is determined potentiometrically with a specific ion electrode and the results are calculated from the semilogarithmic paper previously prepared (Baker and Smith, 1969). In the other method, the aliquot of the filtrate is digested. Nitrate nitrogen in the samples

is then estimated by using a colorimeter (Johnson and Ulrich, 1950).

2.2.4 Nitrate Reductase Activity (NRA) Measurement

Nitrate reductase is sensitive to a number of environmental factors. When other factors remain constant, nitrate reductase activity appears to be inducible by nitrate supply. Therefore the activity of this enzyme is related to nitrate concentration in the soil solution (Beevers and Hageman, 1969) and rate of nitrate uptake by plants (Chantarotwong et al., 1976). It is concluded that there is a positive correlation between the nitrate reductase activity and the growth and protein content of the plants. (Hageman and Flesher, 1960).

It is found that top leaves of maize have a higher level of nitrate reductase activity (and protein content) than bottom leaves, as the activity is easily influenced by light. The activity in the leaves (and protein content) progressively decrease as shading is increased (Zieserl et al., 1963). This may be due to the supply of photosynthate to support respiration, which then drives the induction of this process (Aslam et al., 1973; Hageman and Flesher, 1960). Since the amount of enzyme that can be extracted from the tissue is very small and varies drastically with such factors as cultivars and plant age, the source material for the enzyme extraction is most important (Hageman and Hucklesby, 1971). In maize, it is found that good sources of nitrate reductase are leaves and scutellum (Beevers et al., 1964).

There are two methods used for measuring nitrate reductase activity as follows.

2.2.4.1 In vitro method

The principle of this method is that nitrate reductase is capable of utilizing reduced pyridine nucleotides, flavin, or benzyl viologen as electron donors for the reduction of nitrate to nitrite. These reductants can be added in the reduced form, or generated enzymatically in the reaction mixture. Since the NADH-dependent nitrate reductase is

most prevalent in plants, NADH has been the most commonly employed reductant. Enzyme activity is usually measured by the colorimetric determination of the nitrite formed during a timed incubation period at a fixed temperature, regardless of electron donor used (Hageman and Hucklesby, 1971).

2.2.4.2 In vivo method

The principle of this method is that plant tissue containing, or supplied with adequate amounts of carbohydrate when submerged in a solution containing nitrate and placed in the dark is capable of accumulating and exuding nitrite into the medium. The procedure is also well described by Hageman and Hucklesby (1971).

2.3 GROWTH OF THE MAIZE PLANT

2.3.1 Growth Pattern and Distribution of Dry Matter to Plant Parts

The growth pattern of the maize plant can be roughly divided into three periods, that is vegetative, reproductive and grain filling periods. The beginning and the end of each period are distinguished by phenological stages. From seed germination to floral initiation is the vegetative period, floral initiation to silking is the reproductive period and silking to formation of a black layer at the base of the seed is the grain filling period. The major components related to each period are number of expanded leaves, number of spikelets and grain size respectively (Fischer and Palmer, 1984). Generally the pattern of dry matter distribution to each plant part is sigmoidal.

Most of the growth during the vegetative period is concerned with the leaves. This period ends at a few days after the four fully expanded leaf stage (Hanway, 1963) and the increase in total dry matter is in the lag phase (Hanway, 1962a).

Growth of leaves and stem is the major component during the early stage of the reproductive period. The number of expanded leaves

continues to increase and stem elongation increases rapidly early in this period. Leaf area reaches maximum at around tasselling and usually results in the peak rate of canopy photosynthesis (Waldren, 1983). The beginning of the linear phase for dry matter accumulation starts early in this period.

The number of spikelets is identifiable shortly after ear initiation. It increases rapidly and reaches a maximum shortly before tasselling. Spikelet formation ceases on the lowest ears first and then on successively higher ear shoots. Thus the uppermost ear has the longest duration of spikelet initiation and is usually bigger as a result (Fischer and Palmer, 1984).

The total dry matter at silking is about 50% of that at maturity (Fischer and Palmer, 1984; Hanway, 1962a). Thus most of the total dry matter, especially for ears, is accumulated and is redistributed from other vegetative parts, such as leaves and stem during the grain filling period (Hanway, 1962a; Thom, 1974; Waldren, 1983). This is similar to that for nitrogen distribution (see section 2.1.7).

For dry matter distribution to each plant part, root dry matter increases slowly, reaches a maximum around flowering then gradually decreases. Root dry matter is the least compared with other components (Fischer and Palmer, 1984).

Leaf dry matter reaches maximum shortly before silking. At this stage, leaves and stem are the major components of total dry matter. They are approximately equal, but the stem is usually a bigger component at maturity (Fischer and Palmer, 1984; Hanway, 1962a). This is probably due to the senescence of leaves.

The ears (including husks, shanks and grains) are a relatively unimportant component before silking because they are a very small portion of the total dry matter. However, after silking they increase rapidly to become the major component. At maturity, the ears contribute about 62% to the total dry matter (Hanway, 1962a; Thom,

1974).

The pattern of increase in grain weight is also a sigmoidal curve. The lag phase starts immediately after fertilization (silking). The linear phase, or rapid dry matter accumulation in the grains occupies about half of this period in which more than 90% of grain dry matter is accumulated. The final phase of slower dry matter growth reaches the maximum shortly before the black layer of seed occurs (Johnson and Tanner, 1972). At this stage, there is rapid loss of moisture from the grains. Grains contribute about 47% to the final total dry matter (Hanway, 1962a; Thom, 1974).

2.3.2 Growth of Leaves and Measurements of Growth

Growth of grass and cereal leaves are well described by Dale (1982) and Langer (1979) and are summarized as follows. During the seedling stage, the growing point occurs just above the highest node. Since the stem is very highly contracted, the growing point's actual position is at the base of the stem. At its inception, the whole leaf primordium is meristematic, but soon cell division activity becomes confined to an intercalary meristem at its base. This region becomes divided into two zones through the formation of a band of parenchyma cells. This coincides with the appearance of the ligule, originally an outgrowth of the epidermis. These events mark the beginning of separate development within the foliar organ, for the upper portion of the meristem is associated with growth of the leaf blade, while activity in the lower portion leads to growth of the leaf sheath. Cell division and enlargement, largely in the basal region of leaf sheath, cause the younger lamina to move up inside the folded sheaths of older leaves. Meristematic activity in the lamina comes to an end when the ligule is differentiated, but then the sheath elongates through division and enlargement of its cells, this continues until the ligule is exposed. This marks the end of elongation growth and the foliar organ has now reached its final length. Meanwhile the next leaf moving up inside the previous sheath.

The extension of leaves may be influenced by the growth of stem depending on the methods used for measurement. This is because growth of cereal stems is not apical, that is, it is not due to elongation from the tips. Instead, there is a meristematic region at the base of each internode. New cells are produced sequentially, starting at the first, or lowest internode, then at the second internode and so on. The pattern of stem growth is analogous to extending a telescope, or an aerial which is lengthened not by adding to the upper end, but by extension at each of the joints (Chapman and Carter, 1976). Therefore the elongation of the lower internodes can push, or add a length to the upper leaves, if for example, measured from ground level.

The importance of leaf growth in crop production has been of interest among the researchers for a long time. Apart from leaf area measurement which is the conventional method used in growth analysis, there are some other methods, such as leaf extension and appearance measurements which are relatively new. Many of the earlier studies about these two methods dealt with grasses (Anslow, 1966; Greenwood, 1976; Silsbury, 1970).

2.3.2.1 Leaf extension

In cereals, it refers to the length from the tip of the youngest leaf to the collar of the latest fully expanded leaf (Acevedo et al., 1971; Swan et al., 1981), or to a reference point, usually on the ground surface (Watts, 1972) depending on the purposes of measurement. The measurement is made on the youngest leaf because this leaf is most sensitive in responding to the changing environmental factors. For a detailed and accurate measurement, an auxanometer (1976) and Sharp et al. (1979) can be used. This Gallagher et al. method measures the changing extension rate of the leaf during a short period of time, but it needs sophisticated equipment and may not perform well under field conditions because of the interference from such factors as wind. For a less detailed, but more simple and practicable method, a ruler can be equally used in the field, or glasshouse. However, it may cause damage to the leaves if care is not taken. Consequently the results obtained may not be accurate and can lead to the misinterpretation.

The pattern of extension of a leaf is similar to the sigmoid pattern of other growth (Erickson and Michelini, 1957; Williams, 1975). Usually the lag phase is not expressed because in practice, it is not possible to measure since the leaf is still in the leaf whorl. Therefore leaf extension is usually expressed from the linear phase up to the final phase. During the linear phase, leaf extension increases rapidly and reaches the final phase within a few days. The beginning of the final phase is coincident with the appearance of a ligule which marks the end of leaf growth. In fact, leaf extension still slightly increases for a few days in this phase, due probably to the expansion of cells. When the extension of all leaves in a plant is plotted in series, the final phase of the first leaf is lowest and gradually increases in the succeeding leaves (Williams, 1975).

Since leaf extension is the most sensitive of plant processes in responding to environmental stresses, for example, water stress (Hsiao, 1973), it means that this measurement can be used to indicate the beginning, end, or levels of stresses imposed to plants relating to the plant growth and final grain yield.

2.3.2.2 Leaf appearance

In cereal crops, a leaf is fully expanded when the ligule, or collar is visible (Hanway, 1963; Tottman et al., 1979). Since the younger leaves extend from the whorl of fully expanded leaves, the number of fully expanded leaves is approximately one to three leaves lower than the actual number of leaves seen. In general, the rate of leaf appearance is expressed in terms of the number of days between the appearance of successive leaves in order to permit direct comparison between the results from different experiments (Anslow, 1966). It should be noted that this value is not the same as plastochron which is defined as the time interval between initiation of two successive leaves (Erickson and Michelini, 1957).

When the number of leaves is plotted against time under normal conditions, a nearly-linear pattern is obtained (Erickson and Michelini, 1957).

The appearance of leaves might be related to leaf extension and thus be influenced by environmental factors. Therefore this measurement should be used for monitoring the response of plant to stresses.

2.3.3 Growth of Roots

The growth of maize roots is well studied and reviewed by Foth (1962) and Waldren (1983) and is briefly described as follows. The radicle first emerges, then followed by seminal roots which may, or may not live to the maturity of the plant. Although the first roots to develop are seminal roots, the bulk of the plant's root system consists of secondary roots which includes crown roots and brace roots.

Root growth consists of a series of overlapping stages which are associated with stages of shoot growth. Early root growth occurs largely in a downward diagonal direction, followed by extensive lateral growth which is completed one or two weeks before tassel emergence. This lateral growth causes a marked uniformity in root density in the upper 30 cm of soil. At 23 days after planting (about 30.5 cm high), some root tips may laterally extend up to 38 cm from the stem base. At about the seven unrolled leaf stage (37 days after planting), maize roots extend about 53 cm from plant base.

Brace roots which provide support of the stem and as well as functioning in water and nutrient uptake, appear near the completion of lateral root development (shortly before tasselling). At about 10 unrolled leaf stage (54 days after planting), these roots are approximately 10.2-12.7 cm long, but at 18 days after beginning of tasselling, they are profusely branched. This is coincident with the completion of lateral growth of other roots.

In general, the size of maize root system increases rapidly until tasselling and then declines during grain filling period. During early growth stages, roots grow rapidly and few die, so the size of the root system increases exponentially. As the maize plant reaches flowering, old roots begin to die as fast as new roots are developing into new soil volume, so that the size of the root system remains constant. As the plant proceeds through later growth stages, the number of roots dying exceeds the number being produced and the overall size of the root system declines.

Since the development of most roots occurs prior to tasselling, so the length of time to tasselling affects the size of the root system. Earlier types of maize that tassel sooner will not have as large a root system as later maturing types which tassel later. Compared with a small type, a large type of maize may have a 50% greater spread, 10% deeper penetration and 311% greater root weight.

Although, in general, nitrogen is banded on one side of the crop row, root growth is similar on both sides of the row (Thom, 1974). Nitrogen fertilizer application also increases weight and penetration of roots, especially during early stages (Linscott et al., 1962).

2.3.4 Some Factors Affecting Leaf and Crop Growth of Maize Plants

2.3.4.1 Nitrogen

The functions of nitrogen have already been mentioned in section 2.1.1.

(i) Effect of nitrogen on leaf growth. Seedlings still mainly rely on food reserves in the seeds shortly after germination, but once the chlorophyll is developed and seedlings are established, plants are independent. This suggests that nitrogen supply starts to play its roles very early on in plant life. Since nitrogen reserves in the plants are very low at the seedling stage, most of nitrogen supplied to leaf growth must therfore come from the soil solution. However, the

seedlings are still in lag phase at this stage. Therefore the amount of nitrogen needed is relatively low compared with the later stages (Arnon, 1975).

From the beginning of the linear phase, the rate of leaf growth increases. This results in a higher demand of nitrogen in this phase compared with the previous phase. Starter fertilizer generally increases the rate of leaf appearance in maize (Eik and Hanway, 1965) because it probably increases the number of leaf cells resulting from the higher cell division activity (Arnon, 1975). Consequently this will result in a higher leaf area in plants with starter fertilizer than in nitrogen depleted plants. Leaf growth is retarded in the nitrogen depleted plants because of the reduction of chlorophyll content resulting in a reduction of photosynthate supply and therefore affecting growth of young leaves (Dale, 1982).

During the early to middle linear phase period, nitrogen fertilizer applied may cause increase in leaf size, or leaf area duration, especially if the seedlings are already well established. Nitrogen fertilizer applied to maize plants which have had complete starter fertilizer seems to delay the decrease of leaf area per plant caused by senescence. This results in a higher leaf area per plot of nitrogen topdressed plants during the later growth stages (Arnon, 1975). However, nitrogen fertilizer application at this stage does not seem to have any noticeable effect on the rate of leaf appearance (Eik and Hanway, 1965).

Death of lower (older) leaves may occur from the beginning of the linear phase and nitrogen (which is a mobile nutrient in the plant) can be translocated from the ageing leaves to the younger leaves for recycling (Dale, 1982). Therefore there are two sources of nitrogen for leaf growth from this stage, that is nitrogen from the soil and nitrogen reserve in the plant which is an alternative source when there is a lack of soil nitrogen supply. This suggests that the response of plants to nitrogen stress at later growth stages through the growth of leaves, for example, leaf extension, should be slower than that to such

other factors as water, or temperature stresses, especially if the plants are well supplied with nitrogen during early stages. Therefore under short term, or mild nitrogen stress conditions, plants may not respond to the stress through the growth of young leaves. It has been found in Lolium spp. that even when nitrogen stress approaches 100% (zero growth on a dry weight basis), leaf elongation still continues. This seems to be caused by a rapid recycling of nitrogen from the older leaves to the emerging leaves (Greenwood, 1976).

The number of leaves of cereal crops may not be affected by nitrogen supply because it is largely determined by genotype (Allen \underline{et} \underline{al} ., 1973). Langer and Liew (1973) found that there was no effect of nitrogen on the leaf number of the wheat main shoot. In maize, Eik and Hanway (1965) reported that the effect of increasing nitrogen early in the season on the number of leaves per plant was not as consistent as the increase in leaf size.

(ii) Effect of nitrogen on crop growth. The effect of nitrogen on crop growth can start from seed germination due to seed-fertilizer contact. Effect of nitrogen fertilizer salts that come in contact with the seeds depends on such factors as type, rate and placement of fertilizer and soil water level. The most injurious nitrogen fertilizers are anhydrous ammonia and urea (Fried and Broeshart, 1967). For example, Isensee et al. (1966) found that ammonium nitrate caused deformity and shortened seedling roots. The epidermal cells in the affected area were enlarged, become deformed and ruptured. Extensive cellular breakdown and necrosis also occurred in the meristemic region. Consequently this can cause failure, or delay of germination, or emergence.

Shortly after emergence, the seedlings start to be dependent on photosynthate production. This means nitrogen supply from the soil starts to play an important role. However, the requirements are minimal during this period compared with the whole life cycle (Arnon, 1975). Nitrogen supply at this stage is mainly for leaf growth, especially when compared with root growth. That is, shoot/root ratio

generally increases with higher nitrogen supply during seedling stage because a smaller quantity of carbohydrates is translocated to the roots (Arnon, 1975). Schreiber et al. (1962) found that nitrogen fertilizer applied before this stage could significantly increase the number of grains per ear, possibly a result of higher supply of photosynthate to the ear bud.

During the early growth stages, leaves act as 'sink' for nitrogen, but later on they start to play as 'source' as well. That is, leaves can provide nitrogen for the growth of younger leaves and ears (Arnon, 1975). The effect of nitrogen on leaf area is well established and can be found elsewhere (Arnon, 1975; Eik and Hanway, 1965; The importance of leaves for plant growth is that Stoskopf, 1981). they are the major sites for the production of carbohydrate, the major component of plant dry matter. Moreover, they also play an important role in amino acid synthesis and nitrogen storage (Novoa and Loomis, 1981). Since the production of dry matter and grain yield of maize are directy related to and highly correlated with the weight of leaves (Hanway, 1962a), or the leaf area index at silking time (Eik and Hanway, 1966), it is necessary to deal with leaf growth when discussing maize growth. Generally leaf area, or leaf area index increases rapidly during the linear phase and it falls after reaching the maximum (Arnon, 1975). It might be possible to shorten the period of very low leaf area index by nitrogen fertilizer application which promotes a high initial rate of leaf growth. Other objectives of nitrogen fertilization could be to maintain a large photosynthetically-active leaf through the period when assimilates are being stored in the ears by preventing premature death of leaves due to nutrient deficiencies (Arnon, 1975). Apart from the increase in leaf area, fertilizer applied can also increase chlorophyll content of the leaves. If the increased chlorophyll can be maintained up to after flowering, this is an advantage because there is a direct relationship between leaf chlorophyll content at flowering and grain yield (Hurduc and Stefan, 1966). To show the effect of nitrogen on maize growth between non-deficient and extremely nitrogen-deficient plants, Hanway (1962a) found that leaf weight and daily dry matter production of non-deficient

plants were 2048 and 245 kg/ha respectively compared with 957 and 82 kg/ha respectively for extremely nitrogen-deficient plants.

Although nitrogen plays an important role in promoting dry matter production, excessive supply of nitrogen can cause adverse effect by increasing lodging of maize plants resulting in the reduction of grain yield, especially if the lodging occurs after flowering. Fisher and Smith (1960) reported that lodging was significantly increased following application of high rates of nitrogen fertilizer because of the imbalance of nutrients, especially with potassium. It causes premature breakdown of parenchyma cells in the lower third of the stem due to the inhibition of protein synthesis (Arnon, 1975).

From the onset of flowering to early grain formation, the nitrogen requirements of maize plants reach a peak (Arnon, 1975). Ears start to act as strong 'sinks', consequently nitrogen reserves from plant parts are gradually retranslocated to the ears. In this situation nitrogen supply from the soil can delay the senescence of leaves, otherwise a significant amount of nitrogen will be retranslocated from the leaves in case there is a nitrogen supply shortage which would cause the premature death of leaves (Arnon, 1975). Consequently the photosynthetically-active leaves and growth of ears (especially ear and grain sizes) will be affected. However, nitrogen fertilizer applied during this period should not influence the number of seeds because seed number have already been determined earlier (Schreiber et al., 1962). But nitrogen fertilizer applied at this time increases percent nitrogen in the grain and stem tissue (Jung et al., 1972). agreed that for a good grain yield, nitrate should be present in the stem below the ear (Kurtz and Smith, 1966).

(iii) Effect of different placement of nitrogen fertilizer at topdressing. Proper placement is important for two reasons: (a) Prevention of injury to the plant. (b) Efficient utilization by the plant (Nelson and Stanford, 1958). In general, placement of solid nitrogen fertilizer at topdressing may be classified as surface and deep, near and far from row and the combination of these. The

effectiveness of each method varies with the depth and spread of plant roots, soil types, climatic conditions, equipment, nitrogen carrier and the amount to be applied (Oertli, 1979; Olson and Kurtz, 1982; Robertson and Ohlrogge, 1952).

Surface application is relatively easy to employ. Rainfall, both pattern and amount, should play an important role in the availability of nitrogen by transporting nitrogen down to plant roots. of nitrogen leached down below the root zone should be minimal compared with the deep placement, because it will be intercepted by the root mass, provided nitrogen fertilizer is placed near crop rows and soil is not excessively well drained. However, an amount of nitrogen can be lost by erosion and volatilization. For example, urea and some of the ammonium salts can lose significant quantities of nitrogen as ammonia by volatilization, especially under drying conditions and high soils (Ernst and Massey, 1960; Fenn and Kissel, 1973). Consequently this may result in a smaller amount of nitrogen being Results from a long term experiment show that available to plants. urea applied on the soil surface resulted in the substantially lower maize grain yield compared with that placed at a depth of 15-20 cm (Soubiès and Lenain, 1967). Since the nitrogen fertilizer is placed on the soil surface, relatively earlier than normal application may be needed for nitrogen fertilizer applied to infiltrate from soil surface into the root zone. Plants should respond differently to nitrogen fertilizer placed at different distances from the row. Results of Robertson and Ohlrogge (1952) show that nitrogen fertilizer applied at 25.4 cm from the maize row tended to produce higher yield than that placed at the center between the rows (spacing not reported), though there was little difference between treatments. The near-row placement may supply nitrogen to plant roots quicker than center-between rows placement because of the shorter distance of nitrogen transportation.

During later growth stages, certain amounts of nutrients in the surface layer of soil are depleted. Most of the roots are in the lower soil layers exploring for nutrients. This means nitrogen fertilizer

applied at some depth can easily be transported to plant roots compared with surface, or shallow placement, especially under dry conditions with different soil water content of soil layers. Robertson and Ohlrogge (1952) found in a silt loam soil that under relatively uneven distribution of rainfall, the deeper placed nitrogen bands (10.2-12.7 cm) when maize plants were knee-high, produced significantly more yield than the shallow bands (2.5-5.1 cm). This is because in deeper placement, nitrogen fertilizer was placed in the absorbing root zone and readily available to plant roots, whereas the shallow placed nitrogen fertilizer remained near the soil surface and out of the major nutrient absorption zone. In well drained soils with adequate and well distributed rainfall, deep nitrogen fertilizer placement was generally not significantly superior to the shallow, or surface placement (Robertson and Ohlrogge, 1952; Trierweiler and Omar, 1983). Therefore deep and near the row placement may minimize the loss of nitrogen fertilizer applied, but the high salt concentration of the nitrogen fertilizer used may cause fertilizer injury to the roots under dry conditions (Aldrich et al., 1975). Moreover, the implements used also cause root damage, therefore affecting the growth of plants.

2.3.4.2 Rainfall

Rainfall generally has very little direct effect on maize growth, but it has a great influence on soil water which plays a significant role on growth of plants. When rain falls, an amount of water will usually infiltrate into the soil. Some may be immediately absorbed by plant roots, some may be temporary stored in the A horizon to be absorbed by plant later, or some may gradually percolate down to the lower horizons to be stored there.

The importance of water, in general, is that it comprises over 90% of organisms and participates, either directly, or indirectly in all metabolic reactions (Devlin, 1975). However, the total quantity of water required for growth directly is relatively small. It has been estimated that less than 1% of the water absorbed by maize during its growing season is retained in the plant (Street and Öpik, 1984). Most

of the water entering a plant is lost in transpiration (Kramer, 1959).

Apart from the influence on metabolic activities, water also plays an important role in nutrient availability to plants. Since most fertilizers are in the forms of compounds, they need to be hydrolyzed into ions before they are available to plants. Furthermore, in general, the available plant nutrients are more concentrated in the A horizon than in the lower horizons of soils (Thompson and Troeh, 1978) fertilizers tend to be applied on, or near soil surface. Consequently soil fertility is usually highest at soil surface and gradually decreases following soil depth. Therefore soil water is needed to transport the dissolved nutrients to plant roots (see section 2.1.4). Rain water also helps to release some non-mobile ions (such as ammonium) from soil colloids which makes them more available to plants (Salisbury and Ross, 1978).

Generally most of the problems involed with crop growth is an insufficient supply of water. A slight decrease in turgidity is nearly always accompanied by a decrease in stomatal aperture (Kramer, 1959) which thus affects transpiration, which consequently affects water and nutrient absorption. Lack of water supply can cause dehydration of protoplasm which reduces photosynthetic capacity (Ensgraber, 1954). It has been shown in maize that the relative net photosynthesis is near zero when the relative turgidity of leaves are near 70% (Downey, 1971). Moreover, the closure of stomata also impedes carbon dioxide diffusion into the leaves. This affects photosynthate production because carbon dioxide is one of the raw materials needed.

The influence of rainfall on crop growth can obviously start before planting. That is, it is still the major factor determining the time of planting in most areas. Water stored in the soil before planting can affect seed germination and seedling emergence. Optimum soil water content also aids the preparation of a suitable seedbed. This improves the seed-soil contact, so that water can easily transfer to the seeds.

When the seedlings are established, the rapid development of roots corresponds to the growth of the above ground parts. Turgidity of root cells which affects the absorption of nutrients is influenced by soil water supply and lower plant water status decreases physiological activity (Kramer, 1959). This can decrease starch and chlorophyll content of seedlings (Maranville and Paulsen, 1970). Consequently photosynthate supply will be reduced and growth of seedlings can be affected.

Plants can also be affected by excess water supply. Ritter and Beer (1969) found that, at a low nitrogen fertilizer level, flooding when maize was 15 cm in height for 24-72 hours reduced yields by 18-32%. But at a high nitrogen fertilizer level, these reductions ranged from 14-19%. Mittra and Stickler (1961) also found that flooding at the five-leaf stage reduced dry matter (harvested 21 days after flooding) by 7.5% if flooded 7 days, 34% if flooded 14 days and 43% if flooded 21 days.

During the active vegetative stage, maize plants grow very rapidly and water use is greater than during the previous period. This is due to the higher leaf area index which increases transpiration. Water balance becomes very important at this period under low rainfall conditions, since the atmospheric demand for water is high and the plants require much more water to meet their needs (Shaw, 1976). Water stress during this period can also affect grain yield production. Shaw (1976) reported that when maize plants at this stage were subjected to a rather severe stress for four to six days, grain yield was reduced about 2.8% per day. Some of the results suggest that the yield reduction was due in part to the decrease of leaf area below the optimum.

Flooding during this period results in different plant response compared with that flooded during seedling stage. Ritter and Beer (1969) had showed that at a low level of nitrogen, 24 hours of flooding during the vegetative period reduced yields 14% and this yield reduction increased to 30% with 96 hours of flooding. But with a high

level of nitrogen, very little yield reduction occurred even with 96 hours of flooding.

The flowering (tasselling and silking) stage is a very critical time in maize production, because the number of ovules that will be fertilized is being determined during this stage (Shaw, 1983). maize is an often-cross-pollinated plant, any factors which affect the viability of pollen and silk will influence fertilization consequently grain yield. Smith (1920) determined a correlation between the rainfall over 10 day periods at different stages of growth and the yield of maize over a 30 year period. It was found that the highest correlation coefficients which were statistically significant, were obtained for rainfall in the periods just before, during and just after flowering. Barnes and Woolley (1969) found a 6-8% reduction yield when water stress was imposed for a period of a few days at tassel emergence. For water stress at silking, Claassen and Shaw (1970) found that water stress imposed at 6% silking reduced yield only 3% per day, but at 75% silking the yield reduction was 7% per day. Results from other researchers also confirm that a 6-8% reduction per day was common (Shaw, 1976). Moreover, stress imposed at 75% silking and combined with a fertility stress gave a yield reduction of 13% per day with a large reduction in the number of developed grains per ear (Claassen and Shaw, 1970). Some researchers also found that water stress during this stage caused the time between pollen shedding and silking to be delayed. With severe stress, silking may be delayed until after much, or all of the pollen has been shed, increasing the number of poorly filled ears (Shaw, 1976).

When flooding near silking occurred, Ritter and Beer (1969) found that yield reductions up to 16% occurred with 96 hours of flooding at the low level of nitrogen. However, no reduction in yield occurred at a high nitrogen level.

Rose (1936) found that rainfall early in the grain filling period was positively correlated with yield in most areas of the Corn Belt, U.S.A. Increased rainfall was associated with higher yields (Thompson,

1962; 1963). Moreover, it has been found that rainfall had a more significant effect on yield than temperature at the beginning of this period (Bondavalli et al., 1970). When maize is subjected to water stress starting at 10-40 days after silking. It shows that four days of stress causes an average yield reduction of approximately 4.2% per day of stress (Mallett, 1972).

2.3.4.3 Temperature

Temperature, either soil, or air temperature affects plant growth by influencing plant processes, especially the enzyme-catalyzed reactions.

Nutrient absorption is influenced by temperature changes. It has been reported that the maximum rate of nitrate uptake in maize is between 30-40°C (Ezeta and Jackson, 1975). In general, an increase in temperature results in an acceleration of salt absorption. But this influence is confined to a relatively narrow range, if temperature increases past a maximum point, it will inhibit and eventually terminate the process. When temperature decreases approaching 0°C (less than 5°C), the uptake of nitrogen is retarded (Shaw, 1976) due probably to the decline of stomatal opening lowering the rate of transpiration, therefore restricting water absorption.

As in all life processes, photosynthesis is restricted to a temperature range that roughly corresponds to that tolerated by protein compounds (Devlin, 1975). Although the photochemical part of photosynthesis is independent of temperature, the biological part which is controlled by enzyme activity, is strictly temperature dependent. Cold temperatures inhibit the rate of photosynthesis by lowering the activity of enzymes involved in the dark reactions of photosynthesis. In general, increase in temperature results in an acceleration of photosynthesis when other factors are not limiting. This increase is linear at the lower temperatures, starts to drop off when higher temperatures are reached and finally reaches an optimum above which photosynthesis is inhibited. Under field conditions, Thomas and Hill

(1949) found that the influence of temperature on the rate of photosynthesis is practically nonexistent in a range from 16-29°C. This is because of the limiting effect of light, or carbon dioxide concentration.

The effect of temperature on translocation rates is complicated by the influence of temperature on other plant processes that may directly, or indirectly affect the movement of solutes. In general, the translocation of solutes is influenced by temperature in much the same manner as other physiological processes. That is, the rate of translocation increases with temperature to a maximum and then decreases due to the detrimental effects of high temperature (Swanson and Böhning, 1951).

The influence of temperature on crop growth can start from sowing. Maize seeds germinate best at soil temperatures above 10°C, but at temperature below this point, germination sharply decreases, or fails (Cummins and Parks, 1961). This is probably due to the retardation of water supply for seed imbibition combined with the reduction of enzyme activity in the seed.

During seedling emergence, the rates of radicle and shoot elongation of maize plants are greatest at soil temperature of about 30°C, but cease at constant temperatures of 9 and 40°C (Blacklow, 1972). Later on, during the early active growth stage, the optimum soil temperature for growth is influenced by moisture conditions. Allmaras and Nelson (1971) found that when soils were dry, a mulch between the rows aided root and shoot growth even with temperatures below 26°C. But when soils were moist, treatments with temperatures below 26°C consistently reduced growth and yield of dry matter.

In practice, shoot and root growth are inhibited at 5°C, however, photosynthesis still continues, but at a slower rate (Shaw, 1976). This prolongs the life of the plants. Temperatures below this level can injure the leaves and consequently affect on growth, as Sellschop and Salmon (1928) found in 6-week-old plants chilled at temperatures of

0.5-5°C for different lengths of time. If less than 50% of the leaf area showed injury soon after chilling, plants occasionally recovered and were capable of seed production if less than 25% of the leaf area injured. Whereas those with more than 50% injury seldom recovered. As the length of chilling increased, the amount of injury also increased and since leaf area is the major source of assimilate production, this should therefore affect crop growth and dry matter production.

The growth of maize during the vegetative period has been found to be related to air temperature. The best correlation between growth and air temperatures occurred when remainder indices above 10°C were used. They are now widely used and known as 'Growing Degree Day - GDD'. Loomis (1934) found that the growth rate decreased rapidly as the temperature decreased toward 10°C and most rapid growth was made in the late afternoon, or early evening and morning.

The optimum temperature for growth during the late vegetative period varies with the amount of rainfall. For example, the optimum average Iowa temperature is near 21°C with only 25 mm of rainfall, but is about 28°C with 150 mm of rainfall (Thompson, 1962; 1963; 1966). It has been found that temperatures above normal reduce the yield sharply. One of the causes is probably because maize plants grow very rapidly during this stage and water use is greater. Therefore the higher temperature might increase the transpiration and evaporation rates, consequently water deficits develop (Shaw, 1976).

The time at which tasselling and silking occur are very temperature dependent. Wallace and Bressman (1937) found that a so-called 115-day cultivar took 74 days from planting to tasselling with an average temperature of 20°C, but only 54 days with an average temperature near 23°C. As an average for the 60-day period after planting, they found that for each degree the temperature averaged above 21.1°C, tasselling was hastened by two to three days. Even though increase in temperature results in the speed up of flowering, this may have an adverse effect on fertilization if temperature rises above an optimum level. As Berbecel and Eftimescu (1973) found that

maximum temperatures above 32°C around tasselling and pollination sped up the differentiation process of the reproductive parts, but resulted in the higher rates of kernel abortion. If too many kernels are aborted, this can lower grain yield.

For the period from flowering to maturity, Peters et al. (1971) found a night air temperature of 29.4°C reduced maize yield almost 40% compared with a cool temperature of 16.6°C . The high temperature produced earlier senescence and maturity and may have induced a water stress on the plants. Whereas cooler temperatures increased the length of the period to some extent. During the middle of grain filling period, Bondavalli et al. (1970) found that temperature had a more significant effect on maize yield than the rainfall in the same period.

CHAPTER THREE

MATERIALS AND METHODS

3.1 EXPERIMENTAL DESIGN AND LAYOUT

In order to obtain a declining rainfall situation, different growing periods were spread over time. A split-plot design with four replications was used by allocating three growing periods as main-plots and five nitrogen fertilizer placements as sub-plots. Nitrogen fertilizer placements were as follows.

- 1. Control (no nitrogen application):
- 2. One band of nitrogen on soil surface, 10 cm from the row: SN
- 3. One band of nitrogen on soil surface, between the rows: SB
- 4. One band of nitrogen at 3 cm depth, between the rows: D1
- 5. One band of nitrogen at 10 cm depth, between the rows: D2

A spacing of 75 cm between rows was used to suit the spacing of the tractor tyres. Plant spacing within row was 20 cm, giving a population of 66666 plants/ha which is the density generally recommended to farmers.

Five harvests of plant samples were planned initially, but in growing periods 2 and 3, one more harvest (harvest 3 at G.S. 2 - Hanway, 1963) was added later to obtain more details about plant growth.

The number of sampled plants needed per harvest was four plants per plot. These were harvested from the two-middle rows. Four buffer

plants at the ends of each plot, two buffer plants between harvests and three buffer rows on each side of the plot were provided to ensure a uniform competitive effect and prevent external effects on sampled plants. Therefore this led to a desired plot size of an eight-row plot of 6 m \times 6 m with 1.5 m space between plots.

3.2 EXPERIMENTAL SITE

3.2.1 Site Description

The experiment was conducted at Massey University (40° 23′ S) at an altitude about 33 m above sea level on a Karapoti brown sandy loam soil (N.Z.S.B., 1976). This soil is a recent soil from alluvium of river flats and it is non-accumulating. Profiles show 20 cm of very dark greyish brown friable sandy loam, or fine sandy loam overlying light olive brown slightly firm sandy loam, or loamy sand with few faint yellowish brown mottles. At 30 cm from the surface, this passes down to olive slightly firm sand with thin horizontal dark brown bands and with depth the sand becomes greyer in colour. The topsoil has a moderately developed medium nut structure and the subsoil has a weakly developed nut structure breaking to single grains. Drainage is good to somewhat excessive (Cowie, 1978).

3.2.2 Pre-trial Preparation

Since the site had been used for an experiment in the winter before this experiment was started, soil heterogeneity problems from the residual fertilizer and crop residue could have been caused. Moreover, it had been reported that maize grown in this area did not respond to nitrogen fertilizer (Thom, 1974). In order to improve soil homogeneity and decrease the soil fertility level, the paddock was cultivated and a high density of barley was sown without fertilization two months before the sweet corn was sown. It was hoped that this should adjust the soil into the desired conditions for this experiment. Moreover, barley also served as the first crop in a simulated double cropping system.

Before this experiment was started, two soil samples were taken for soil tests (Appendix 1). Each sample composed of 22 sub-samples collected peripherally and diagonally within the barley sown area. The depth of sampling was 15 cm. One sample was for phosphorus and potassium tests. The other for nitrogen test was frozen shortly after collection to stop nitrogen transformation processes in the soil sample. It was expected that the results of the soil tests would be used to determine the rates of fertilizer application. But unfortunately the results were not available until the date of sowing. Therefore the rate of fertilizers used was determined from general recommendations.

3.3 EXPERIMENTAL PROCEDURES AND TECHNIQUES

3.3.1 Stand Establishment

Before sowing at each growing period, barley plants in the four allocated strips were moved and taken out of the strips. Growth stages of barley plants before moving in each growing period were 22, 51-53 and 75 respectively (Zadoks et al., 1974). The height of barley stubble left in the strips was around 6.5 cm.

Minimum cultivation technique was used for land preparation to reduce nitrogen transformation processes in the soil. This minimum cultivation was done by using times and rotary hoes to shallow depth.

A relatively high rate of starter fertilizer was used to ensure good stand establishment. Fifteen percent potassic superphosphate (N-P-K-S = 0-7-7-8) at the rate of 500 kg/ha was mixed with granular systemic insecticide (Thimet*20G, a.i. 200g/kg phorate) at the rate of 7.7 kg/ha. Then the mixture was manually broadcast and incorporated into the soil by using a rotary hoe. The insecticide used was to control stem weevil and greasy cutworm. Apart from this insecticide, no other chemicals were used to control insects, or diseases throughout the experimental season.

'Royal Crest Medium' which is a short season sweet corn ($\underline{\text{Zea}}$ $\underline{\text{mays}}$ L. saccharata) variety was sown in this experiment. The seeds were treated with Captan 75.

A precision drilling machine (Nodet Gougis Pneumasem II) was set to sow at 5 cm depth, 20 cm apart within a row and 75 cm between rows. Two rows could be sown per pass, therefore four passes were made to obtain eight rows of sweet corn. The row direction was approximately North-South. Since the sowing plates of the machine placed only one seed per position in a row, this resulted in some non-germinated positions because of dead seeds. In this case resowing was done manually shortly after 70-80% of seedlings' tips emerged above ground by using a hand trowel. However, the drilling machine was generally precise and the germination percentage of the seeds was very high, so little resowing was required.

Growing periods 1, 2 and 3 were sown on 22 November, 19 December 1984 and 8 January 1985 and emergence dates were 29 November, 24 December 1984 and 14 January 1985 respectively.

Actazine*5A (a.i. 50% atrazine) was sprayed immediately at the rate of 3 1/ha after the seeds had been sown in growing period 1 to control broadleaf weeds. Spraying was done manually by using a sprayer. Unfortunately this could not control knapsack efficiently as expected. Patches of summer grass (Digitaria sanguinalis (L.) Scop.) were noticed in some plots of replications 1 and 2 around two weeks after sowing. Therefore paraquat (a.i. 200 paraquat as dichloride salt) at a rate of 5 paraquat/litre of water was sprayed selectively to destroy this grass between sweet corn rows. In subsequent growing periods, lasso (a.i. 50% alachlor (2-chloro-2, 6-diethyl-N-methoxymethyl acetanilide)) was mixed with actazine at the rate of 6.5 1/ha.

After the seedlings had been established, irrigation was applied during dry spells to maintain seedling stands. The applied water was adjusted and restricted within the six-middle rows of the crop strips.

Only one irrigation was applied in each growing period. The amounts of water applied were 13, 31 and 75 mm for growing periods 1, 2 and 3 respectively.

3.3.2 Nitrogen Fertilizer Application

Granular calcium ammonium nitrate (CAN, $\mathrm{NH_4NO_3}$ + $\mathrm{CaCO_3}$, 26% N) was applied for topdressing at the four fully expanded leaf stage. This was around 22-28 days after the sowing date. The precision drilling machine used for sowing seeds was adjusted to deliver nitrogen fertilizer at the rate of 300 kg CAN/ha (78 kg N/ha) for the D1 and D2 treatments.

The six-middle rows received CAN with a row on each side of a plot left as a buffer row.

The depth of CAN applied was relatively uneven because of the uneven soil surface. Where CAN was left on the soil surface (for D1 treatment), it was covered by using a hand rake.

CAN was weighed before being loaded into the hopper and reweighed after D1 and D2 treatments were fertilized. The amount of CAN applied to these two treatments was calculated for re-checking. Then calculation was done to determine the amount of CAN to be applied by hand to the SN and SB treatments. This was to ensure that all treatments received the same amount of nitrgen fertilizer. The amount of CAN to be applied was calculated to the rate per row. A measuring can was made to measure the amount of CAN needed per row. Then CAN was applied to these two treatments (SN and SB) manually.

3.3.3 Data Collection and Measurements

3.3.3.1 Plant measurements

(i) Leaf extension and appearance. Three to four plants per plot were randomly selected around one week before topdressing. An inverted

L-wire was driven into the ground close to each plant base as a permanent reference point for measuring leaf extension. These plants were monitored for leaf extension every day until one day before topdressing. Finally one plant was selected as representative in each plot.

Leaf extension measurement was done at around 8-9 a.m. on the youngest leaf. A ruler was vertically placed on the reference point (inverted L-wire). The stem and youngest leaf were placed alongside the ruler and a reading was recorded. The measurement at each leaf was stopped when another younger leaf tip was measureable and then that new leaf was measured. Leaf extension measurement was stopped when the collar of the flag leaf was visible.

Leaf appearance was recorded simultaneously with leaf extension measurement. This was done by placing a plastic clip near the collar of the newest fully-expanded leaf. Leaves from below this reference leaf were counted as fully expanded leaves.

Data were calculated every day to detect the beginning of differences between treatments. It was hoped that this could be used to indicate the response of plants to nitrogen fertilizer applied. Whenever the differences occurred, plant samples would be taken.

(ii) Plant samples. The first samples (above ground parts of four plants per plot) were taken one day before topdressing, except in growing period 1 which was three days in advance. Since the differences between treatments could not be detected by using leaf extension and leaf appearance, the subsequent samples were taken at the following growth stages (Hanway, 1963):

Harvest	Growth stage	Plant characteristics
1	1	Collar of fourth leaf visible.
2	1.5	Collar of sixth leaf visible.
3	2	Collar of eighth leaf visible.
4	4	Tassel visible.
5	5	75% of plants have silks visible.
6	7	Roasting ear stage.

Usually sampled plants in replications 1 and 2 were taken in the morning and replications 3 and 4 were taken in the afternoon. Secateurs was used to cut at the base of stems of predetermined plants, that is, four plants from the two-middle rows. The four sampled plants in each plot were put into a labelled paper bag. Then the samples were carried to a nearby shed for sap nitrate test.

(iii) Sap nitrate test. Two plants that were representative of the rows were randomly selected from the four sampled plants. A 1-1.5cm cross section of stem (leaf sheaths in a young plant) was cut around 8-10 cm above the stem base for a young plant in the early harvests, or around 1.5-2 cm under the uppermost ear node in the later harvests. This was to have the cutting from the same position in a plant and in every harvest. This section was put into a small metallic cup closed with a plug. A C-clamp was used to squeeze the cutting until plant sap oozed around the plug. The sensitive paper of a 'Merckoquant' strip was thoroughly, but briefly dipped into the sap. Time was taken for the paper square to change to '500 ppm zone' violet. After that, the time recorded was converted to ppm of nitrate by using a standard curve applied from a standard table (Appendix 2). If the paper square did not turn violet after two minutes, the developed colour was compared with standard colours printed on the strip container concentration (ppm) of nitrate was recorded. After that the squeezed

cutting was returned to its bag to be included with the stem component.

When the test had been finished for the first two replications, it was followed by the other two replications. Then all of the plant samples were transported to the laboratory for plant dissection. If the work for all of the replications could not be finished within a day, especially during later growth stages, only the first two replications would be harvested and processed until finished. The other two replications would be processed on the following day.

(iv) Plant dissection. The definitions of plant parts are presented in Appendix 3 for clarification. When plant samples arrived at the laboratory, leaves and dead leaves were separated first by using razor blade and scissors. Then leaf area was measured by passing the leaves through a 'LI-COR' area meter (model 3100). After that, the samples were dried in the oven at 80°C for one to four days depending on the amount of leaves.

Stems, tassels and ears were separated, chopped into smaller pieces and dried the same as leaf samples.

When these samples had been dried, each plant part was weighed by using a 'Sartorius' scale (model 1303 MP).

At the final harvest some plant characteristics were recorded. These were number of marketable and non-marketable ears (ear length, excluding shank, of 20 cm was used as a criterion for separation), number of live and dead tillers, length of uppermost cobs and length of fertilized kernels.

(v) Total nitrogen determination. Dried plant samples of harvests 1 and 2 were finely ground by using a 'Culatti' grinder (type DFH 48). Samples of harvests 3 to 6, except tassels, were rough ground before fine grinding. Samples were sub-sampled from the bags during fine grinding. It should be noted that dead leaves were excluded from this total nitrogen analysis. Therefore leaf nitrogen was slightly

underestimated.

Nitrogen content in the samples was determined by using the Micro-Kjeldahl method (Bremner and Mulvaney, 1982). 'Kjeltec Auto 1030' analyzer was used for the analyses, the procedures of operation followed that described by A.G. Robertson, Massey University (pers. comm.).

3.3.3.2 Soil measurement

Soil water content was measured periodically by using gravimetric method (Sopher and Baird, 1978) and calculated on a dry weight basis. Soil samples were collected per replication at the depth of 0-4, 8-12 and 16-20 cm by using a soil corer. The sub-samples were taken in a zigzag pattern from between the four middle rows and bulked in plastic bags before transporting to the laboratory. Soil samples were dried in the oven at 105°C for 24 hours.

3.3.3.3 Weather measurements

A cylinder type, 'Marquis' rains gauge (model MS 102-2) was installed at the site for rainfall measurement. It was close to the experimental plot, but was clear of any obstracles which could interfere with the rainfall. The height of the top of the rain guage was 50 cm above ground level. The measurement was done in the morning at around 9 a.m.

Other data, such as temperatures were obtained from Glasslands Division, Department of Scientific and Industrial Research which is around 0.5 km south of the site.

3.3.4 Statistical Methods

3.3.4.1 Analysis of variance

The analysis was carried out by using SPSS-X programme (Norman,

1983). Data were analyzed separately, that is, randomized complete block analysis of variance for within a growing period (harvest) and split-plot analysis of variance for between growing periods. When the F test in the analysis of variance was significant, least significant differences at 0.05 level of probability were calculated for comparisons between treatment means (Little and Hills, 1978).

3.3.4.2 Regression analysis and curve fitting

A more direct analysis was made of the growth curves by fitting curves and testing the treatments for significant differences. Scattergrams were done and then several types of equations, for example, second and third degree polynomials, logistic and quadratic logistic were tried to fit the data. Eventually the quadratic logistic equation showed the most appropriate fit. The linear form of this equation is

$$ln(Y/(y^0-Y)) = a + bX + cX^2$$

where Y is the dependent variable; y^0 is the upper asymtote; a,b and c are the regression coefficients and X is the independent variable.

The SPSS-X programme was also employed for the regression analysis. In fitting a curve, firstly the upper asymote was estimated from the data. Then the iterative procedures were used to find the best upper asymtote by choosing the highest value of coefficient of determination (R²). After that, the curve was tried by superimposing on the scattergram to find the best fit. In some cases, the slightly lower values of coefficient of determination were selected because of the better eye fit. When the appropriate upper asymtotes were obtained, the REGCOM programme (I.L. Gordon, Massey University, pers. comm.) which compares curves in pairs, was used to compare the equations.

CHAPTER FOUR

RESULTS

4.1 CLIMATE

4.1.1 Rainfall

Rainfall over the experimental season was higher than the 53 year average (Table 4.1), it was not uniformly distributed throughout the season (Figure 4.1). Approximately 61% of the total rainfall occurred in the first half of the season which covered growing period 1 and growing period 2 up to harvest 3. There were some dry and low rainfall spells in the second half of this experimental season from harvest 1 to 6 in growing period 3.

Table 4.1: Amount of rainfall and grass minimum temperature means during harvest 1-6.

Growing	<u>R</u> a	Temperature		
period	53 year measu		Deviation ^{2/}	(°C)
	average ^{1/}	at site	(%)	
1	141.3	175.3	+24.1	11.4
2	134.5	160.3	+19.2	11.0
3	127.7	89.5	-29.9	9.8
1-3	319.0	371.1	+16.3	

^{1/} N.Z.M.S. (1983), at Department of Scientific and Industrial Research, Palmerston North.

^{2/} from 53 year average.

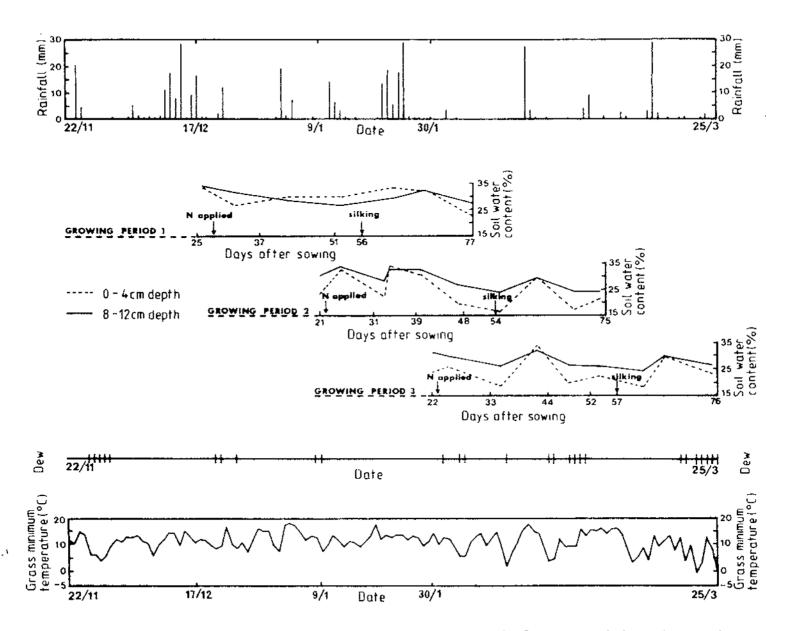


Figure 4.1: Daily rainfall, soil water content, dew days and daily grass minimum temperature.

4.1.2 Dew Days

It was observed that sometimes dew contributed significant amount of water around the base of plants. There were many dew days throughout growing period 3 (Figure 4.1).

4.1.3 Temperature

The pattern of temperatures (grass minimum, maximum and minimum air temperatures) was similar. It was relatively stable from harvest 1 in growing period 1 to the silking stage in growing period 2 (Figure 4.1). After that it tended to fluctuate, particularly in growing period 3 and resulted in a lower mean temperature for this period compared with the slightly higher and similar temperatures for growing periods 1 and 2 (Table 4.1).

4.2 SOIL WATER CONTENT

Soil water at 16-20 cm depth, in general, was similar to and only slightly lower than that at 8-12 cm depth. Figure 4.1 shows soil water content at 0-4 and 8-12 cm depths, the same depths as nitrogen fertilizer placement. The high rainfall during the first half of the experimental season resulted in the high and similar soil water content at both depths throughout growing period 1 and until around harvest 3 in growing period 2. In growing period 3, the long dry spell resulted in the low soil water content of the surface soil (0-4 cm) from the first harvest.

4.3 NITROGEN UPTAKE, MARKETABLE EARS AND LEAF AREA

Generally in this study, the nitrogen concentration in plant parts for each harvest was not significantly different, so that the trends of nitrogen uptake and that of plant dry weight was the same, or similar. In order to avoid repetition in presenting the results and discussion, nitrogen uptake of plants was chosen for presentation and results for plant dry weight are in Appendix 4.

The most significant nitrogen responses were produced in growing period 3 and so all results are presented. This growing period was the most pertinent to the situation in Thailand. In growing periods 1 and 2, plants generally did not respond to different nitrogen fertilizer placements and the significant differences occurred between control and all nitrogen treatments. In order to simplify presentation, the results of growing periods 1 and 2 are presented as comparisons between control and means of all nitrogen treatments. Full details of all treatments for marketable ears are presented in Appendix 5.

4.3.1 Total Plant Nitrogen

Plants did not significantly respond to both nitrogen fertilizer application and different nitrogen fertilizer placements at the final harvest in growing period 1, but they responded to nitrogen fertilizer application in growing period 2 (Table 4.2). Similar conclusions were made when curves were fitted to the data over all harvests (Figure 4.2) and comparison between curves for control and pooled-nitrogen treatments was made (Table 4.3). In general, nitrogen fertilizer placed on soil surface near the row (SN) tended to produce greatest total plant nitrogen in both growing periods.

Table 4.2: Total plant nitrogen (g/m^2) for each harvest and growing period.

Growing	Treatment	Harvest					
period		1	2	3	4	5	6
	C	0.23	1.91a	1/_	5.65a	6.12a	8.80a
	SN	0.23	2.70a	-	7.42a	8.58a	12.93a
1	SB	0.23	2.27a	-	7.07a	8.48a	12.12a
	D1	0.23	2.55a	_	6.72a	8.37a	12.47a
	D2	0.23	2.18a	-	6.78a	7.07a	10.42a
	Mean	0.23	2.33	-	6.73	7.73	11.32
	LSD (.05)	_		-	-	-	-
	CV (%)	-	22.97	_	25.11	21.89	16.83
	С	0.20	1.25a	3.88a	5.92a	5.68Ь	8.27b
	SN	0.20	1.52a	4.78a	8.45a	10.10a	17.57a
2	SB	0.20	1.43a	4.18a	7.02a	9.47a	15.40a
	D1	0.20	1.47a	3.83a	6.92a	8.47a	16.50a
	D2	0.20	1.17a	3.83a	8.43a	10.23a	16.38a
	Mean	0.20	1.37	4.10	7.35	8.78	15.17
	LSD (.05)	-	-		_	2.10	2.75
	CV (%)	-	24.60	21.25	17.51	15.46	11.67
	С	0.11	0.93a	2.48	c 4.25	c 3.90b	4.77 c
	SN	0.11	1.02a	3.63ab	6.63ab	7.13a	12.00ab
3	SB	0.11	0.92a	3.05 в	c 5.15 b	c 7.13a	9.93 b
	D1	0.11	1.17a	3.55ab	5.97ab	c 6.63a	11.30ab
	D2	0.11	0.93a	4. 15 a	7.33a	7.83a	13.88a
	Mean	0.11	1.00	3.38	5.87	6.53	10.38
	LSD (.05)	-	_	1.00	1.97	2.05	3.63
	CV (%)		19.13	19.31	21.77	20.37	22.68

^{1/} Means followed by a common letter are not significantly different at the 0.05 level of probability. Letters apply only within harvests and growing periods.

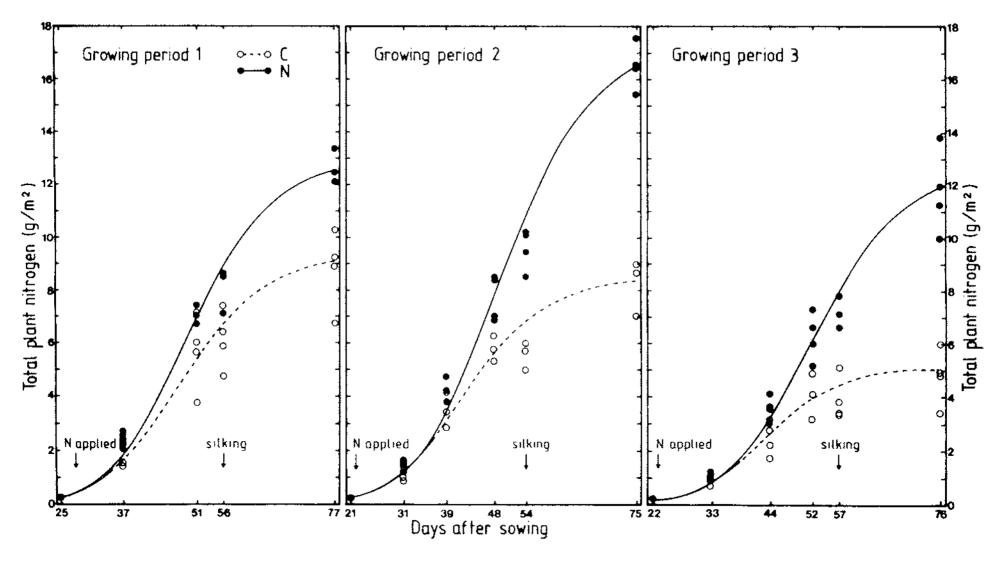


Figure 4.2: Growth in total plant nitrogen for control (C) and pooled-nitrogen (N) treatments (see Tables 4.3 and 4.4). (Equations and coefficient of determination are presented in Appendix 6).

Table 4.3: Coefficients of quadratic logistic equations for total plant nitrogen for each growing period (see Figure 4.2).

Growing	Treatment		Coefficien	t
period		β0	β1	β 2
1	Control	-3.936	0.188	-0.002
	Nitrogen	-4.321	0.197	-0.002
	t-test	1.21	0.31	0.38
	SGNF	NS1/	NS	NS
2	Control	-3.935	0.200	-0.002
	Nitrogen	-4.726	0.191	-0.002
	t-test	3.30	0.44	0.70
	SGNF	***	NS	NS
3	Control	-4.190	0.216	-0.002
	Nitrogen	-5.172	0.199	-0.002
	t-test	2.91	0.62	0.86
	SGNF	***	NS	NS

1/ Level of significance.

NS - Not significant.

(NS) - At 0.1 level of probability.

* - At 0.05 level of probability.

** - At 0.01 level of probability.

*** - At 0.005 level of probability.

**** - At 0.001 level of probability.

In growing period 3 which had different climatic conditions, nitrogen fertilizer application significantly increased nitrogen uptake of plants at the final harvest (Table 4.2). The significant difference started earlier than in the first two growing periods. Figure 4.2 shows that the significant difference in this growing period was a function of a reduced uptake of nitrogen in control treatment rather than the increase in the pooled-nitrogen treatments compared with that in growing periods 1 and 2. Plants also responded to different nitrogen fertilizer placements in this growing period. fertilizer placed at 10 cm depth between rows (D2) resulted significantly higher nitrogen uptake than that placed on soil surface between rows (SB) at the final harvest and this trend was consistent for all harvests after harvest 2. However, when the curves were fitted to these two extreme nitrogen treatments (D2 and SB), they were not When the data of nitrogen treatments were significantly different. pooled as in the first two growing periods, similar results to growing period 2 were obtained (Figure 4.2 and Table 4.3).

Although the significant differences of nitrogen content in plant parts in growing period 3 occurred at earlier harvests than those in growing periods 1 and 2, the trend of these components was similar to that of total plant nitrogen. In order to condense the results, only D2 and SB treatments were selected to be compared with the control treatment for growing period 3 (Figure 4.3). In general, nitrogen fertilizer application increased nitrogen content in every plant part compared with control plants. Leaf and stem nitrogen for plants from D2 treatment increased from harvests 1 to 6, those for plants from SB treatment tended to be stable at the final two harvests, whereas those of control plants tended to decline after harvests 4 and 5. Ear nitrogen tended to be the biggest component for total plant nitrogen at the final harvest, especially for plants in D2 treatment. This resulted in the significant increase of total nitrogen uptake at the final harvest.

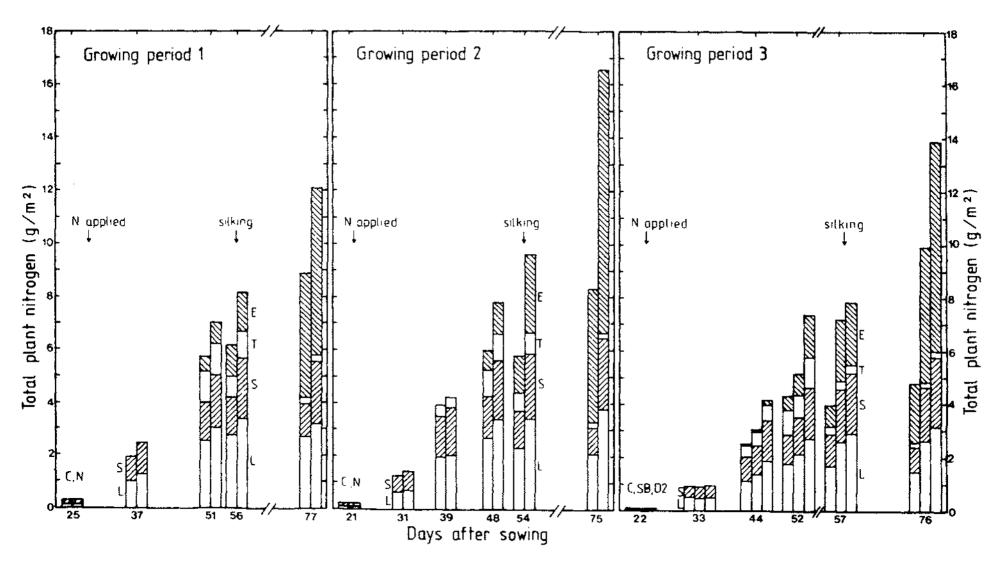


Figure 4.3: Total plant nitrogen and distribution in plant parts for control (C), pooled-nitrogen (N), surface between rows (SB) and 10 cm depth (D2) treatments.

E, T, S and L are ear, tassel, stem and leaf respectively.

Amounts of total nitrogen taken up by control plants at the final harvest in growing periods 1 and 2 were similar, but was significantly lower in growing period 3 (Table 4.2). However, the comparison between growth curves fitted over the growing period for the control treatments shows that there was no significant difference between growing periods (Table 4.4).

Table 4.4: Coefficients of quadratic logistic equations for total plant nitrogen for each growing period (see Figure 4.2).

Treatment	Growing		Coefficient						
	period		β0		ß1		β2		
		t-test	SGNF	t-test	SGNF	t-test	SGNF		
	1 vs 2	0.00	NS	0.39	NS	0.36	NS		
Control	1 vs 3	0.60	NS	0.79	NS	0.82	NS		
	2 vs 3	0.66	NS	0.50	NS	0.56	NS		
	1 vs 2	2.31	*	0.37	NS	0.89	NS		
Nitrogen	1 vs 3	4.47	***	0.15	NS	0.29	NS		
	2 vs 3	3.12	***	0.68	NS	0.77	NS		

Table 4.4 shows that there was significant difference between growth curves fitted over the growing period for plants receiving nitrogen fertilizer. Plants in growing period 2 took up largest amount of nitrogen followed by those in growing periods 1 and 3 (Figure 4.2).

4.3.2 Marketable Ears at Harvest 6

The trend in yield of marketable ears, in general, was similar to that of total nitrogen uptake and total plant dry weight. Plants receiving nitrogen fertilizer in growing periods 1 and 2 tended to produce higher marketable ear weight with a higher number of marketable ears than control plants and these were higher than those in growing period 3 (Table 4.5).

Table 4.5: Marketable ear dry weight, marketable and total ear number at harvest 6.

Growing	Treatment	Ear weight	Ear number	Ear number (/m²)			
period		(g/m²)	Marketable	Total			
1	Control	314.0	5.5	16.3			
	Nitrogen	359.1	6.0	16.8			
	Mean	350.1	5.9	16.7			
	SGNF	NS	NS	*			
2	Control	316.7	5.0	19.2			
	Nitrogen	506.7	7.8	24.4			
	Mean	468.7	7.3	23.3			
	SGNF	(NS)	NS	*			
	С	15.0 c	0.4 c	11.3b			
	SN	297.7ab	5.8ab	20.0a			
3	SB	225.5 b	5.0 Ь	19.2a			
	D1	244.3 b	4.7 b	22.9a			
	D2	349.7a	6.7a	22.1a			
	Mean	226.4	4.5	19.1			
	LSD (0.05)	96.9	1.3	7.5			
	CV (%)	27.8	18.9	25.7			

The dry condition in growing period 3 also caused the plants to respond differently to nitrogen fertilizer application and placement in terms of marketable ears produced. Plants in D2 treatment produced significantly higher marketable ear weight and number than those in SB and D1 treatments (Table 4.5).

4.3.3 Leaf Area

The trend of leaf area at the final harvest was generally similar to that of total nitrogen uptake, total plant dry weight and marketable ears. Plants in D2 treatment for growing period 3 produced significantly higher leaf area than those in SB treatment (Table 4.6).

Table 4.6: Leaf area (m^2/m^2) for each harvest and growing period.

Growing	Treatment			est			
period		1	2	3	4	5	6
1	Control	0.08	0.68	_	1.61	1.64	1.57
	Nitrogen	0.08	0.77	_	1.69	1.68	1.54
	Mean	0.08	0.75	_	1.65	1.66	1.64
	SGNF	-	พร	-	NS	NS	NS
2	Control	0.06	0.39	1.07	1.57	1.66	1.50
	Nitrogen	0.06	0.40	1.10	1.75	1.93	1.85
	Mean	0.06	0.39	1.08	1.70	1.88	1.76
	SGNF	-	NS	NS	NS	NS	NS
	С	0.04	0.34a	0.79a	1.15a	1.01a	0.77 c
	SN	0.04	0.37a	1.04a	1.36a	1.35a	1.28ab
3	SB	0.04	0.32a	0.92a	1.25a	1.34a	1.05 b
	D1	0.04	0.39a	1.03a	1.30a	1.33a	1.15ab
	D2	0.04	0.32a	1.04a	1.38a	1.47a	1.31a
	Mean	0.04	0.35	0.96	1.29	1.30	1.11

4.4 SAP NITRATE TESTS

In general, the trends of sap nitrate levels were similar between nitrogen treatments, except for plants from SN treatment in growing period 2 (Figure 4.4). Sap nitrate levels, in general, sharply dropped from the first to the second harvest. After that levels gradually decreased to the final harvest for control plants, whereas those of plants receiving nitrogen fertilizer started to increase from harvest 2. Sap nitrate levels increased sharply during harvests 2 to 3 in growing period 2, but increased slowly in growing period 3. They tended to be different between surface and deep placement treatments during harvests 3 to 5 and 5 to 6 in growing periods 2 and 3 respectively.

The highest sap nitrate levels over every harvest of plants from SN and D2 treatments in growing periods 2 and 3 respectively corresponded with the highest nitrogen uptake (Table 4.2). The lowest sap nitrate levels of control plants (except at harvest 2 in growing period 3) corresponded with the lowest nitrogen uptake. Sap nitrate levels at each harvest generally tended to reflect nitrogen supply from different placements at that harvest, especially at harvests 3 and 4.

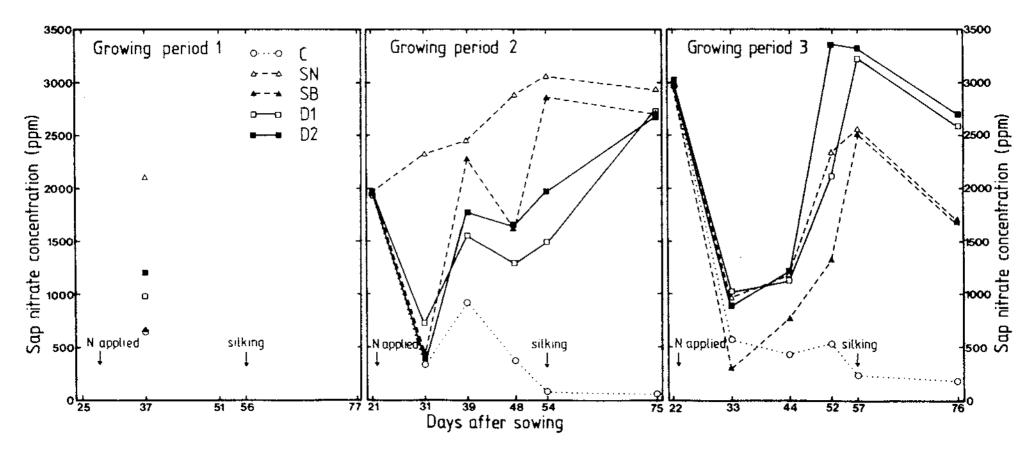


Figure 4.4: Patterns of sap nitrate concentration.

4.5 RESPONSE OF LEAF GROWTH TO APPLIED NITROGEN FERTILIZER

4.5.1 Leaf Extension

Leaf extension and rate of extension during the period shortly (a few days) after leaf emergence (beginning of linear phase) did not show any response to the nitrogen fertilizer applied under these experimental conditions (Figure 4.5).

4.5.2 Leaf Appearance

In general, the appearance of leaves and rate of leaf appearance also did not respond to nitrogen fertilizer application (Table 4.7), leaf appearance of leaves 8-10 (from harvest 3) of plants receiving nitrogen fertilizer in growing period 3 tended to be quicker than that of control plants.

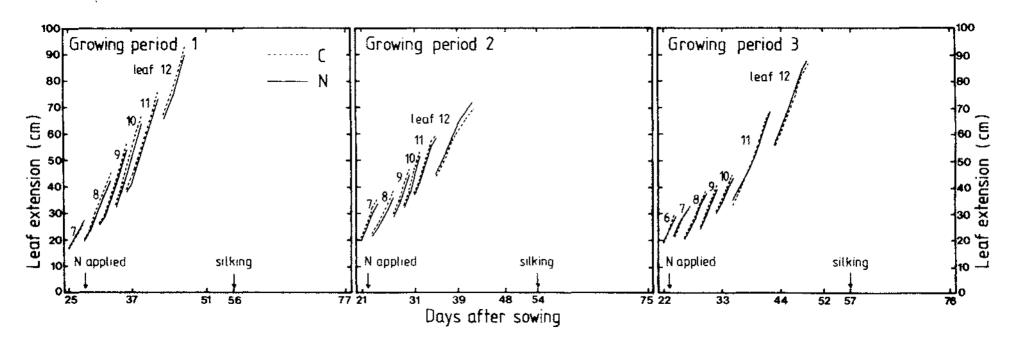


Figure 4.5: Leaf extension for control (C) and pooled-nitrogen (N) treatments.

Table 4.7: Number of fully expanded leaves per plant at each harvest.

Growin period	ng Treatmen I	t			Ī	Days a	after	sowing	•		
<u></u>		25	31	37	41	43	47	49	51		
		(HV1)		(HV2)				_	(HV4)		
1	Control	4.0	5.0	6.0	7.0	8.0	9.3	10.5	12.0		
	Nitrogen	4.0	5.0	6.1	7.2	8.0	9.3	10.2	11.8		
	Mean	4.0	5.0	6.1	7.1	8.0	9.3	10.4	11.9		
	SGNF	NS	NS	NS	NS	NS	NS	NS	NS		
		21	25	31	35	39	43	45	47	48	
		(HV1)		(HV2)		(HV3))			(HV4)	
2	Control	4.0	5.0	6.0	7.0	8.0	9.8	10.8	11.7	12.1	
	Nitrogen	4.0	5.0	6.4	7.1	8.0	9.7	10.8	11.7	11.9	
	Mean	4.0	5.0	6.2	7.1	8.0	9.8	10.8	11.7	12.0	
	SGNF	NS	NS	NS	NS	NS	NS	NS	NS	NS	
		22	28	33	38	40	44	46	48	50	52
		(HV1)		(HV2)		_	(HV3)	.	<u> </u>		(HV4)
3	Control	4.0	5.0	6.0	6.5	7.3	8.0	9.0	10.0	11.3	12.3
	Nitrogen	4.0	5.0	6.0	6.9	7.6	8.7	9.5	10.6	11.6	12.0
	Mean	4.0	5.0	6.0	6.7	7.5	8.4	9.3	10.3	11.5	12.2
	SGNF	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

CHAPTER FIVE

DISCUSSION

5.1 EFFECT OF NITROGEN FERTILIZER APPLICATION

Nitrogen fertilizer application resulted in significantly greater nitrogen uptake (Tables 4.2, 4.3 and Figure 4.2), dry matter yield (Appendix 4), marketable ears (Table 4.5) and leaf area (Table 4.6) at the final harvest for growing period 3 and for growing period 2 except for marketable ears and leaf area. This is similar to the results of maize at the equivalent growth stage obtained by Thom (1974), that is, nitrogen uptake and total dry matter of plants receiving nitrogen fertilizer were about 21-29 and 1744-1967 g/m² respectively compared with those of 19 and 1802 g/m² respectively for control plants. The higher yields in that experiment were probably due to the higher population used (96900 plants/ha) and the use of irrigation.

Fitted curves for nitrogen uptake of control plants were similar for all growing periods (Table 4.4). However, analysis of variance at the final harvest shows that nitrogen uptake of control plants in growing period 3 was significantly lower than those in growing periods 1 and 2. This was probably due to the lower rainfall and temperature in this growing period (Table 4.1). Moreover, soil nitrogen in this growing period might have been lower than that in growing periods 1 and 2 because the barley plants were very mature (growth stage 75 - Zadoks et al., 1974) before this growing period started and so probably took more nitrogen out of the soil. All curves of nitrogen uptake for pooled-nitrogen treatments were significantly different (Table 4.4) with that of growing period 2 having the greatest maximum nitrogen uptake (Figure 4.2) which also was associated with highest number of marketable ears (Table 4.5). Early in growth, nitrogen uptake of plants receiving nitrogen fertilizer in growing period 1 tended to be the highest and that in growing period 3 was significantly the lowest

(Table 4.2). Later, however, rate of increase in nitrogen uptake for plants receiving nitrogen fertilizer from growing period 1 tended to be lower than that in growing period 2. Consequently nitrogen uptake for plants from growing period 2 caught up to and passed that in growing period 1. The poorer performance of plants receiving fertilizer in growing period 1 compared with those in growing period 2 could have resulted from the high rainfall (115.1 mm) early in the growing period (during harvest 1 and nitrogen fertilizer application) (Figure 4.1) which could have brought nitrogen fertilizer applied down to lower soil layers as suggested by Steele (1981). Later on, the rain from after harvest 2 which maintained the high soil water content at lower depth might have increased leaching losses οf Wetselaar (1962) reported that large amounts of nitrate could be moved down to 60-90 cm depth by 60 mm rainfall. Although there was high rainfall (86.4 mm) during harvests 2-3 in growing period 2, nitrate might have moved upward with soil water during the long dry spell (from harvest 3) and been available again to plants (Krantz et al., 1943). This dry spell occurred relatively late in growing period 1 (12 days before the final harvest), therefore the nitrate which was available again to plants might not play any important role. The lower nitrogen uptake of plants receiving nitrogen fertilizer for growing period 3 could have been caused by the reduction in rainfall and temperatures (Figure 4.1 and Table 4.1). Root and air temperatures below 10°C retard uptake of nitrogen (Shaw, 1976) and retard maize growth (Waldren, 1983). The highest yields obtained in growing period 2 support the recommendation by Ministry of Agriculture and Fisheries that the main planting period for sweet corn is mid-October to mid-December (Wood, 1983).

Plants grown in different growing periods responded differently to nitrogen fertilizer applied (Figure 4.2 and Table 4.3). The non-response to nitrogen fertilizer application of plants in growing period 1 could have been due to the high and good distribution of rainfall over the growing period (Figure 4.1) and the relatively high soil nitrogen status (Appendix 1) which might be able to produce about 5.6 t/ha of wheat grain yield without nitrogen fertilization (Quin et

<u>al.</u>, 1982). This is similar to earlier results from irrigated maize (total dry matter at physiological maturity) grown in the nearby paddock which did not respond to nitrogen fertilizer application (Thom, 1974). Therefore nitrogen fertilizer application may not be needed under conditions in growing period 1. Whereas, the significant differences in growing periods 2 and 3 were probably due to the dry spells early in the growing period, possibly combined with the lower soil nitrogen status depleted by the older barley plants, nitrogen fertilizer was therefore needed for plants grown under these conditions.

5.2 EFFECT OF NITROGEN FERTILIZER PLACEMENT

Although rainfall in this experimental season was higher than the 53 year average (Table 4.1), the distribution was not uniform (Figure 4.1). About 61% of the rain occurred in the first half of the season causing wet conditions for growing periods 1 and 2, but drier condition for growing period 3. This made growing period 3 typical of a double cropping situation in Thailand, as the sweet corn was grown after nearly-mature barley and under reducing rainfall. Therefore the discussion in this section is centered around growing period 3 because it was the main period of interest and because there was no yield response to nitrogen fertilizer placement in growing periods 1 and 2.

Because of the low and irregular rainfall pattern during growing period 3 (Figure 4.1 and Table 4.1), nitrogen fertilizer placed at 10 cm depth in the soil (D2) resulted in significantly higher nitrogen uptake than that applied on the soil surface between rows (SB) (Table 4.2). Plants from D2 treatment also produced significantly higher dry matter yield (Appendix 4), higher weight and number of marketable ears (Table 4.5) and higher leaf area (Table 4.6) than those from SB treatment at the final harvest. This is similar to the results of maize obtained by Robertson and Ohlrogge (1952) conducted under similar conditions (non-uniform distribution of rainfall), where bands of nitrogen fertilizer placed at 10.2-12.7 cm depth produced significantly more yield than that placed at 2.5-5.1 cm depth.

Differences between plants from D2 and SB treatments were apparent at harvest 2 for the sap nitrate test (Figure 4.4), but differences in nitrogen uptake were not apparent until harvest 3 (Figure 4.3 and Table 4.2). This would indicate a lag phase between availability of nitrogen to the plants as indicated by the sap test and its utilization as measured by total nitrogen uptake. Therefore the sap nitrate test appears to be a more sensitive measure of the timing of nitrogen uptake.

The higher soil water content at 8-12 cm depth at the early stage (Figure 4.1) which could have caused higher nitrogen uptake for plants from D2 treatment could have two advantages: (i) The deeply placed nitrogen fertilizer would be readily available in the moist zone (Mengel and Kirkby, 1982). Plant roots might not reach the nitrogen fertilizer bands at this growth stage as Foth (1962) found that maize roots extended to about 30 cm from the base at the growth stage equivalent to harvest 2. (ii) Roots would be better able to penetrate into that zone compared with the drier surface zone enabling roots to reach the deeply placed nitrogen fertilizer more quickly.

Nitrogen uptake of plants receiving nitrogen fertilizer applied on the soil surface near the plants (SN) and at a shallow depth (D1) was intermediate between that from D2 and SB treatments at this early stage (Table 4.2). Presumably therefore, plants from SN and D1 treatments were able to obtain some advantages from these placements (that is, close distance between nitrogen fertilizer applied and plant roots for SN treatment and higher soil water content at depth for D1 treatment) compared with those from SB treatment, but not to the extent of those from D2 treatment.

From the sap nitrate test data (Figure 4.4), nitrogen availability for plants from SN, D1 and D2 treatments was similar until harvest 3. This could have been the result of the favourable rainfall between harvests 2 and 3 (Figure 4.1). The favourable water status of surface

soil near plants' base would have favoured plants in the SN treatment as the nitrogen fertilizer was placed near the plants and therefore was better able to be utilized. In addition, it was noted that the structure of the leaf whorl tended to intercept precipitation (rainfall and dew) and concentrated it at the base of the plants. This probably assisted in making nitrogen fertilizer applied in SN treatment readily available to the plants compared with that in SB treatment early in growth (as indicated in sap nitrate levels) before full dispersal of the nitrogen fertilizer applied became available to the whole root system. Utilization of the nitrogen fertilizer applied by plants in D1 treatment would also have been assisted by the sufficient soil water content in the O-4 cm layer over this period.

After harvest 3, however, the relative levels of sap nitrate between the various nitrogen fertilizer placements changed (Figure 4.4). Except for control plants, sap nitrate levels generally increased rapidly over the period from harvest 3 to 5. Levels in plants from the D2 treatment increased more rapidly than those from the other treatments which could have assisted in the superior nitrogen uptake from this placement. Plants from SN and D1 treatments were again intermediate in the rate of increase in sap nitrate levels during this stage. However, plants from D1 treatment reached a higher peak of sap nitrate than those from SN treatment at harvest 5, possibly because nitrogen fertilizer from this placement was in a more favourable position for utilization than that from SN placement during this stage. This should be due to the drying out of surface soil between harvests 3 and 5 (Figure 4.1). The drying of the surface soil during this stage is also probably the reason why surface applied nitrogen fertilizer (SN and SB treatments) resulted in the lower sap nitrate levels over the later stages of growth (low soil water content reduces diffusion rate of nutrients - Mengel and Kirkby, 1982), despite some significant rainfall at 64 days after sowing. This supports the view that nitrate level in plants is easily influenced by soil water conditions (Viets, 1965).

It should also be noted that sap nitrate levels of plants from SB treatment steadily improved from harvest 2 and eventually reached similar levels to those from SN treatment at the final two harvests (Figure 4.4). Thus, the substantial rainfall between harvests 2 and 3 (Figure 4.1) appeared to be sufficient to increase the availability of the nitrogen fertilizer placed in this treatment, although not to the extent of that placed at depth as noted in the nitrogen uptake data (Table 4.2).

At the final harvest, the total nitrogen uptake for plants from SN, D1 and D2 treatments were similar (Table 4.2) and that for plants from SB treatment was lower, even though it was not significantly different from that from SN and D1 treatments. This was the general relationship between the treatments for all harvests after the nitrogen fertilizer was applied, except for harvest 5. Therefore this would indicate that patterns of nitrogen uptake and dry matter production which are established early are important in determining the final yield.

Rainfall which influenced early nitrogen availability seemed to have a significant effect on later growth. It should be noted that, for growing periods 1 and 2, 14.0 and 24.9 mm rain respectively fell immediately after the nitrogen fertilizer was applied (Figure 4.1). It is likely that this would have caused similar nitrogen availability and early growth in every treatment resulting in the lack of response to nitrogen fertilizer placement. (Although surface soil in growing period 1 seemed to be drier during the stage shortly after nitrogen fertilizer application, this was due to the delay in sample taking, that is two days after rainfall). This contrasts to that in growing period 3 which had negligible amount of rain at the same stage. All growing periods had significant rainfall between harvests 2 and 3.

Plants from the SN treatment from both growing periods 1 and 2 had relatively higher levels of sap nitrate at harvest 2 compared with those from other nitrogen fertilizer placements (Figure 4.4). This is in contrast to that in growing period 3 at the same stage and supports

the view that the ability of the plants to capture limited rainfall and concentrate it around the base of the plants is important in situations where limited rainfall can be expected. Presumably this effect was not so significant for growing period 3 because of the dry spell between nitrogen fertilizer application and harvest 2.

When water continued to be available during the growing season, as in growing period 2, the SN treatment continued to supply adequate nitrogen as indicated by the higher sap nitrate levels (Figure 4.4). Although not statistically different, plants from the SN treatment for both growing periods 1 and 2 had greater nitrogen uptake for all except one harvest for growing period 2 (Table 4.2). However, when water became limiting as in growing period 3, the surface treatments (SN and SB treatments) may not be so efficient as also indicated by the lower sap nitrate levels during the later stage.

Differences in the weight and number of marketable ears occurred only for plants from growing period 3 (Table 4.5). Trends were similar to those for nitrogen uptake (Table 4.2), dry matter yield (Appendix 4) and leaf area (Table 4.6) at the final harvest. The relationship between marketable ear weight and leaf area is similar to that in maize obtained by Eik and Hanway (1966), that is, grain yields tended to be linearly related to the leaf area index at silking time. The number of marketable ears per square metre was the main component contributing to the higher marketable yield (Feigin et al., 1980) in this experiment which is also similar to that in nitrogen uptake (Figure 4.3). similar number of total ears per square metre of plants from all nitrogen treatments suggests that the reduced growth caused by poor nitrogen fertilizer placement had an effect on reducing ear development rather than the number of ears. This presumably was because of greater internal competition for a reduced amount of assimilate during the early stage of ear development (Aldrich et al., 1975).

In conclusion, it would appear from these results that, as would be expected, there was an interaction between nitrogen fertilizer placement and water availability. It was unfortunate that no extensive

period of dry weather occurred which might have separated the various nitrogen fertilizer placements more clearly. Moreover, the relatively high coefficient of variation (CV) values (partly due to the relatively small sample size taken) over the growing period compared with those obtained in maize (Edmeades, 1972; Thom, 1974) might also reduce the sensitivity. However, these results indicated that conditions like during the end of rainy season in Thailand, placing nitrogen fertilizer at depth would be an advantage. But to do would require special equipment and power which are not normally available in that country. Moreover, the speed of operation would be relatively slow compared with the surface placements. The application of nitrogen fertilizer on the soil surface close to the plants would utilize the plants' ability to trap limited amounts of rainfall (provided that some rainfall occurs soon after nitrogen fertilizer appears to be a practical alternative to nitrogen application) fertilizer application at depth. The other alternative is that sweet corn should be sown in furrows which help to concentrate the limited water near plants. At topdressing time, nitrogen fertilizer can be applied in the furrows near plants and covered with soil from the ridges, this results in the near plants and at depth nitrogen fertilizer placement. However, the availability of equipment, power and time must be taken into consideration. Under conditions of adequate rainfall, no differences between the methods of placement would be expected. Therefore nitrogen fertilizer applied on soil surface near the plants would appear to be more practical than the other methods because of the quicker and simpler operation.

5.3 VALUE OF SAP NITRATE TESTS

The suggestion that sap nitrate levels are good indicators of nitrogen status of crops has been made for maize (Cornforth, 1980), barley (Ismail and Withers, 1984) and wheat (Withers and Palenski, 1984). The results in this experiment generally support this view. Sap nitrate levels of control plants and plants receiving nitrogen fertilizer at a particular harvest for growing periods 2 and 3 corresponded with the total nitrogen uptake at that harvest, especially

after harvest 2 (Figure 4.4 and Table 4.2).

Figure 4.4 shows that sap nitrate levels in control plants for both growing periods 2 and 3 generally decreased from early harvest to later harvest. This might indicate a shortfall in soil nitrogen supply relative to increasing demand of plants during the later stages of growth and reflected in the slow increase of total nitrogen uptake at the same stage (Figure 4.2). Sap nitrate levels of plants receiving nitrogen fertilizer increased following nitrogen fertilizer application. This is similar to the pattern in barley (Ismail and Withers, 1984) and wheat (Withers and Palenski, 1984) and shows that nitrogen fertilizer application at an early stage could increase sap nitrate levels. The increase of sap nitrate levels was also reflected in later increases in total nitrogen uptake.

The higher sensitivity of the sap nitrate test compared with total nitrogen analysis has already been discussed in section 5.2. Sap nitrate levels over the growing period also tended to reflect nitrogen supply from different nitrogen fertilizer placements. For example, the highest and lowest sap nitrate levels of plants from D2 and SB treatments respectively over growing period 3 (Figure 4.4) corresponded with nitrogen uptake at every harvest (Table 4.2). This was also true for the relationship between sap nitrate levels and nitrogen uptake for plants from SN treatment in growing period 2. The closest relationship occurred at harvests 3 and 4 for growing periods 2 and 3 respectively. This indicates that sap nitrate test is a sensitive and seemingly accurate indicator of plant nitrogen status.

It is desirable to know the critical levels of plant nitrogen status at an early stage, so that if additional nitrogen fertilizer is needed, correction through nitrogen fertilizer application can be made. Sap nitrate levels of plants from every treatment at harvest 2 for every growing period were generally similar at the 1000 ppm level (Figure 4.4). Later on, the levels of control plants were always below the 1000 ppm level, whereas those of plants receiving nitrogen were always above this level, except that of plants from SB treatment at

harvest 3 in growing period 3. This suggests that the general critical level over the growing period (from harvest 2, or six fully expanded leaf stage) would be about 1000 ppm. This value is in the critical range of 900-2000 ppm levels suggested by Cornforth (1980) for 42 day old maize plants which is around the 5 fully expanded leaf stage (between harvests 1 and 2) for sweet corn in this experiment (McCormick, 1974; Table 4.7). Therefore it is suggested that as a result of this work, sap nitrate levels of sweet corn should not be lower than the 1000 ppm level from the 6 fully expanded leaf stage (harvest 2) onwards.

Sap nitrate levels can easily be influenced by soil (Cornforth, 1980; Viets, 1965). Figure 4.4 shows that even though every growing period received the same amount of nitrogen fertilizer at the same growth stage, sap nitrate levels for plants receiving nitrogen fertilizer in growing period 2 increased rapidly from harvests 2 to 3. This was associated with the high rainfall at this stage (Figure 4.1), whereas the slower increases of sap nitrate levels at the same stage in growing period 3 coincided with the lower rainfall. The distribution of rainfall over the whole growing period was also not regular, except for growing period 1. These factors (amount and distribution of rain) caused some limitations in using this test as a predictive tool, that is, whether or not sap nitrate levels at an early stage could indicate the final nitrogen uptake. For example, sap nitrate levels of plants receiving nitrogen (except SN treatment) at harvest 2 for growing period 2, was lower than the 1000 ppm level, whereas those from growing periods 1 (except SN treatment) and 3 (except SB treatment) at the same harvest were around the 1000 ppm level. However, finally plants from these treatments (SB, D1 and D2) for growing period 2 took up higher nitrogen than those for growing periods 1 and 3 (Table 4.2). It shows that the test should be used as a monitoring tool for plant nitrogen status as suggested by Withers and Palenski (1984) rather than a predictive tool due to the difficulties in predicting the amount of rainfall. This is, however, merely reflecting the difficulty in predicting nitrogen availability. It is unlikely that simple nitrogen tests will completely solve this problem.

In practice, the relatively high cost of test strips may prevent the average Thai farmer in using this test at regular intervals over the growing period in the present situation (one packet of test strips costs about \$25 and the per capita income for a Thai farmer is about \$1414). However, this cost would be cheap relative to the possible saving if nitrogen fertilizer can be used more efficiently by farmers. The complicated underlying process of the test may also prevent farmers from using this test, but this can be overcome by transferring the knowledge through extension workers (both government departments and private companies). The strategy suggested by Aldrich et al. and Withers (1982) can be adopted whereby farmers could apply a small rate of nitrogen fertilizer at sowing and with monitoring of the crop during 5 to 7 fully expanded leaf stages, then make decision on whether to apply additional nitrogen fertilizer. The critical level of 1000 ppm may be used in considering whether or not additional nitrogen fertilizer is needed and such factors as rainfall (soil water content), method of nitrogen fertilizer placement and field history must be taken into consideration (Donahue et al., 1983). If nitrogen fertilizer is applied, the test should be used to check the availability of nitrogen fertilizer applied a few days after application and sap nitrate levels should increase higher than the previous stage, provided that water is It may be useless to check sap nitrate levels at later not limiting. stage, for example, after silking stage because it is too late to correct any deficiency by nitrogen fertilizer application (Kurtz and Smith, 1966), unless to increase percent nitrogen in the grains (Jung et al., 1972), or to gain more understanding about the plant nitrogen status for use in the next season. To reduce the cost involved, Withers (1982) suggested for wheat and barley that by careful placing of the sap at the edges, or corners of the paper, in this way upto four tests can be obtained from one strip. This must be done very carefully with sweet corn, such as by using a capillary tube (or plastic rod) to place sap on the paper because of the larger amount of sap produced compared with other cereals. Another alternative is, a strip should be split longitudinally into two halves. So that two tests can be made and without fearing about sap oozed over the whole paper.

5.4 USE OF LEAF EXTENSION AND LEAF APPEARANCE

The successful use of leaf extension to show the response of maize plants to water stress has been reported by Acevedo et al. (1971). It has also been reported that the rate of leaf extension in grasses is affected by nitrogen fertilizer application (Greenwood, 1976; Wilman and Mohamed, 1981). Therefore there might be a potential to use this technique for monitoring nitrogen response in sweet corn plants. However, it was not successful in this experiment (Figure 4.5).

The results in grasses obtained by J.F. Power (Greenwood, 1976) show that nitrogen fertilizer application seemed to retard the rate of leaf extension at the stage shortly after nitrogen fertilizer application. The non response of leaf extension to nitrogen fertilizer applied in this experiment could be the result of factors as follows:

- (i) Since nitrogen is highly mobile within the plant (Devlin, 1975), new leaves may be supplied from older leaves under nitrogen deficient conditions (Dale and Milthorpe, 1983), or be supplied preferentially with limited amounts of nitrogen. Therefore the extension of young leaves may have been buffered from the effects of the nitrogen treatments. Greenwood (1976) found in Lolium sp. that even when nitrogen stress approaches 100% (zero growth on a dry weight basis), leaf extension still continues.
- (ii) The measurement period at each leaf might have been too short (three to seven days) to detect the differences. The retranslocation of nitrogen reserves to the young leaves might have been occurring as discussed in (i). Moreover, the overall duration of measurement was also relatively short (upto shortly before harvest 4 Figure 4.5). At the time leaf extension measurements were taken, leaf area development and nitrogen uptake (except for harvest 3 in growing period 3) for all treatments were similar (Table 4.6 and Figures 4.2, 4.3). Differences in leaf area and nitrogen uptake developed after the period of leaf extension measurements. Although plants receiving nitrogen fertilizer for growing period 3 produced significantly higher leaf area than

control plants at the final harvest, this should be largely due to the contribution from the higher number of tillers as reported for other cereals (Spiertz et al., 1984).

Therefore these results suggest that the measurement of leaf extension is not an appropriate technique for monitoring nitrogen response, partly because the period of response to nitrogen fertilizer applied occurs when measurements are unable to be made.

Table 4.7 shows that leaf appearance generally did not respond to nitrogen fertilizer application, especially in growing periods 1 and 2, although the differences at the later leaves (from harvest 3) in growing period 3 were relatively large. Eik and Hanway (1965) reported that nitrogen fertilizer application to maize as a topdressing did not seem to have any noticeable effect on the rate of leaf appearance. However, Dale and Wilson (1978) reported for barley grown in washed sand that the high nitrogen treatment (applied as basal dressing) resulted in the quicker rate of leaf appearance than the low nitrogen treatment. The difference in the number of fully expanded leaves at harvest 3 tended to correspond with the difference in total nitrogen uptake at the same stage (Figures 4.2, 4.3 and Table 4.2). It is likely that leaf appearance might not be able to detect the response of plants to nitrogen fertilizer applied as a topdressing under high soil nitrogen status, but might be possible under low soil nitrogen status (Dale and Wilson, 1978). Therefore the measurement of leaf appearance seemed to be more appropriate than that of leaf extension for detecting the response of plants to nitrogen fertilizer applied under these experimental conditions and further studies should be conducted to confirm these results.

CHAPTER SIX

CONCLUSIONS

- 1. Nitrogen fertilizer application at topdressing time resulted in significantly higher nitrogen uptake, dry matter yield and marketable ear weight and number under low soil nitrogen status, but not under high soil nitrogen status.
- 2. Nitrogen fertilizer placement at topdressing time played an important role under dry conditions, but not under wet conditions. Whether or not rain fell soon after nitrogen fertilizer application seemed to have a significant effect on nitrogen availability to plants and therefore influenced plants response to different nitrogen fertilizer placements. Nitrogen fertilizer applied at 10 cm depth between rows resulted in significantly higher nitrogen uptake, dry matter yield and marketable ear weight and number compared with that placed on the soil surface between rows under dry conditions. However, nitrogen fertilizer applied on the soil surface near the plants performed well under both dry and wet conditions.
- 3. Leaf structure of sweet corn helped in concentrating limited precipitation (rainfall and dew) around plants' base and resulted in the more rapid and efficient use of nitrogen fertilizer applied on the soil surface near plants compared with that placed on the soil surface between rows.
- 4. Sap nitrate levels were good indicator of plant nitrogen status. Sap nitrate test was more sensitive than the total plant nitrogen measurement in indicating the timing of nitrogen uptake by plants. Sap nitrate test appeared to be a good monitoring tool to be used to understand what was happening to nitrogen fertilizer applied and plant nitrogen status.

- 5. The general critical value of sap nitrate over the growing period (from six fully expanded leaf stage) for sweet corn was about 1000 ppm.
- 6. To detect the response of plants to nitrogen fertilizer applied, the measurement of leaf extension was not appropriate, but that of leaf appearance seemed to be possible under low soil nitrogen status.

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APPENDICES

Appendix 1: Results of soil tests.

1. Nitrogen (Quin <u>et al</u>., 1982).

	Initial levels	After incubation
	(ppm)	(mqq)
Nitrate	22.5	60.3
Ammonium	15.8	14.0
total N	38.3	74.3
ΔN	(N difference)	36.0 ppm

2. Ministry of Agriculture and Fisheries soil test data.

	P	K	Mg	рН
Test values	54	12	26	6.1
Target values	45-55	12-15	10-12	6.0-6.5

Appendix 2: Standard conversion from the time taken to reach the 500 ppm level to nitrate concentration (Cornforth, 1980; Scaife, 1979; N.J. Withers, pers. comm.).

Time (seconds)	Nitrate concentration (ppm)
5	20000
6	16000
7	14000
8	12000
9	10000
10	7000
11	5600
12	5000
13	4700
14	4350
15	4000
16	3800
17	3550
18	3300
19	3150
20	3000
22	2750
24	2500
26	2300
28	2100
30	2000
35	1650
40	1400
45	1250
50	1100
60	900
70	750
80	620
90	600
100	590
120	500

Appendix 3: Definitions for plant parts.

- 1. Leaf: Fully expanded part in a young leaf, or photosyntheticable (green) areas of leaf blade in a mature leaf (dead parts were excluded). This part was for leaf area measurement.
- 2. Dead leaf: Non-photosyntheticable areas (excluded parts) of a leaf blade.
- 3. Stem: The part remaining after leaves had been separated in a young plant. In a mature plant, this referred to the part from ground level up to the lowest spikelet of tassel including leaf sheaths.
- 4. Tassel: In a young plant (when the tassel tip was not visible), it was included in the stem. When its tip was visible, this referred to the part from the tip to the lowest spikelet.
- 5. Ear: It was included in stem when it was not visible in a young plant. When its tip was visible, an ear was separated at the joining of stem and shank. This included cob, husks and shank.

Appendix 4: Total plant dry weight (g/m^2) for each harvest and growing period.

Growing	Treatment			Har	vest		
period		1	2	3	4	5	6
	С	4.6	68.2a ¹	_	276.7a	357.8a	785.2a
	SN	4.6	81.2a	-	279.8a	373.2a	823.3a
1	SB	4.6	78.0a	_	285.8a	372.0a	815.7a
	D1	4.6	85.5a	-	288.0a	400.3a	830.7a
	D2	4.6	76.3a	-	283.3a	357.7a	757.5a
	Mean	4.6	77.8	-	282.7	372.2	803.8
	LSD (.05)	_	_		-	_	-
	CV (%)	-	21.7	-	20.3	15.7	8.8
	С	5.3	38.8a	125.3a	283.5a	407.2 b	758.8b
	SN	5.3	40.0a	134.0a	342.2a	534.3a	1082.2a
2	SB	5.3	43.5a	121.7a	292.7a	498.3a	1019.3a
	D1	5.3	43.3a	118.7a	302.8a	466.2ab	1053.7a
	D2	5.3	36.8a	120.0a	355.8a	538.2a	1015.8a
	Mean	5.3	40.5	124.0	315.5	488.8	998.0
	LSD (.05)	-	-	-		89.3	159.8
	CV (%)	-	21.0	24.2	13.6	11.9	10.3
	С	2.3	32.5a	100.8a	217.5a	228.0b	374.0 c
	SN	2.3	33 .8 a	135.0a	293.8a	348.8a	700.0ab
3	SB	2.3	33.5a	125.2a	241.8a	354.8a	591.2 b
	D1	2.3	36.3a	127.0a	274.0a	346.7a	676.7ab
	D2	2.3	31.5a	140.3a	303.7a	384.3a	755.3a
	Mean	2.3	33.5	125.7	266.2	332.5	619.3
	LSD (.05)		-	_	-	100.3	151.8
	CV (%)		13.3	13.7	16.7	19.6	15.9

^{1/} Means followed by a common letter are not significantly
 different at the 0.05 level of probability.
 Letters apply only within harvests and growing periods.

Appendix 5: Marketable ear dry weight, marketable and total ear number at harvest 6.

	Ear dr	y weight		Ear nu	mber (/m	ı²)	
Gr	Growing (g/m²)			etable	<u>To</u> tal		
pe	riod 1	2	1	2	1	2	
Treatment							
С	314.0a	316.7a	5.5a	5.0a	16.3 bc	19.2 b	
SN	409.2a	523.0a	6.7a	8.3a	20.0a	26.3a	
SB	392.3a	494.2a	6.7a	7.5a	16.3 bc	22.5ab	
D1	369.7a	538.0a	6.3a	8.3a	18.0ab	25.8a	
D2	265.3a	471.5a	4.5a	7.2a	12.8 c	23.0ab	
Mean	350.1	468.7	5.9	7.3	16.7	23.3	
LSD (0.0	5) -	-	_	_	3.5	4.8	
CV (%)	21.1	17.0	21.4	20.6	13.1	13.4	

Appendix 6: Quadratic logistic equations and coefficient of determination (R^2) of control and pooled-nitrogen treatments for total plant nitrogen (N).

Growing	Treatment	Equation	R ²
period			
1	Control		
	ln(N/(10.8-N))=-	3.936+0.188(Days)-0.002(Days ²)	0.93
	Nitrogen		
	ln(N/(16.3-N))=-	4.321+0.197(Days)-0.002(Days ²)	0.91
2	Control		
	ln(N/(9.7-N)) = -	3.935+0.200(Days)-0.002(Days ²)	0.94
	Nitrogen		
	ln(N/(21.3-N))=-	4.726+0.191(Days)-0.002(Days ²)	0.97
3	Control		
	ln(N/(6.2-N)) = -	4.190+0.216(Days)-0.002(Days ²)	0.90
	Nitrogen		
	ln(N/(17.5-N))=-	5.172+0.199(Days)-0.002(Days ²)	0.94