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**VARIATION IN THE GRAIN PROPERTIES OF MAIZE  
HYBRIDS WITH DIFFERENT GRAIN HARDNESS  
CHARACTERISTICS AND THEIR RESPONSE TO NITROGEN  
FERTILIZER IN TERMS OF MILLING QUALITY**

A thesis presented in partial fulfilment of the requirements  
for the **Degree of Master of Agricultural Science**  
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## ABSTRACT

The proportion of grits and flour produced during the dry milling of maize (*Zea mays* L.) grain is related to the ratio of hard to soft endosperm. The quality standards required vary widely with different end uses, and for dry milling a hybrid with a 'hard' endosperm will usually yield the highest proportion of grits. The texture of the maize endosperm is variable and depends on the maize hybrid and agronomic conditions. In general the available literature showed protein concentration of the grain can be improved by nitrogen fertilizer application, and as the protein content increases, the amount of hard endosperm increases along with value to the miller.

A field trial to investigate the effect of nitrogen fertilizer on grain yield and quality, especially grain protein content and hardness, was carried out at the Frewens block, Massey University in the 1994/95 season. Three maize hybrids ( P3751, P3787 and A82-8 xNZ84) with three different endosperm textures ( soft, intermediate and hard) were grown at two sowings (October and November) with three different nitrogen levels (0, 250, 500kg urea/ha). Urea fertilizer was applied as a side dressing and split into three application times, i.e. at the three leaf stage, at canopy closure and at the 50% silking stage. Plant growth and development were measured by counting the leaf number and leaf appearance rate, formation of the black layer and grain moisture dry down for each hybrid. Grain yields and yield components were measured for different nitrogen treatments at both sowings. Grain protein content was measured from total nitrogen percentage as determined by the Macro Kjeldhal method. Grain hardness was measured by a Stenvert Hardness Tester, while bulk density and grain moisture content were measured by a grain analysis computer.

The total number of leaves per plant was greater in hybrid A82-8xNZ84 than hybrids P3787 and P3751 at both sowings, but rate of leaf appearance was faster for the November sowing than the October sowing. Formation of the 'black layer' (i.e physiological maturity) and moisture dry down rate was faster in hybrid P3787 than in hybrids P3751 and A82-8xNZ84 at both sowings.

Grain yield was significantly increased at both sowings by the application of 250kg/ha urea, but not by the 500kg urea/ha treatment. Hybrid A82-8xNZ84 gave the highest yield and P3787 gave the lowest. The main yield components which differed between hybrids were number of grains per cob and 100-grain weight.

Grain protein content increased progressively in response to the applied nitrogen fertilizer. Protein percentage increased from 8.81% in the control to 10.13% for 500kg urea/ha in the October sowing, and 8.72% in the control to 10.13% for 500kg urea/ha in the November sowing. At both sowings all three hybrids contained the highest amount of protein at the highest urea treatment i.e. 500kg urea/ha.

Increased nitrogen application improved grain hardness. For those grains grown under higher nitrogen levels grinding resistance time, energy required for grinding and milling duration time were higher than grains grown when no urea was applied. Grain bulk density (test weight) increased as nitrogen increased. Hybrids A82-8xNZ84 and P3787 had higher grain hardness under the high nitrogen treatment than hybrid P3751.

There was a strong, positive relationship between grain protein content and Stenvert Hardness Test parameters at both sowings. When nitrogen was applied grain contained a higher amount of protein ( which presumably made grain harder) than the no applied nitrogen treatment. Inherent endosperm texture was not changed by the increased protein percentage as the soft endosperm hybrid did not show an improved hardness, but the intermediate and hard endosperm hybrids showed an improvement in this regard. Results from both sowings indicated grain yield, protein and hardness quality can be improved by applying nitrogen fertilizer. This has implications for dry milling, where hard grain is a necessity for higher grits recovery.

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## CHAPTER I

### INTRODUCTION

Maize or corn (*Zea mays* L.) is a cereal crop grown for both grain and forage. It is the world's most widely distributed crop (CIMMYT, 1982) and is used as a staple food for humans, as feed for livestock and as the raw material for many industrial products. It is one of the most important cereals and on a world scale ranks third in production after wheat and rice (FAO, 1992). In New Zealand, it is the third ranked crop after wheat and barley, and has a diverse range of uses. Seventy percent of maize production is used by the feed industry and the remaining 30% is used by the wet and dry milling industries to produce starches, food oils, dextrose, flours and a range of grits (Chappell, 1985; Hardacre, et al., 1991).

Maize is milled by dry and wet processes. In the dry milling process, the primary products are isolated pieces of endosperm (i.e. grits), which are recovered by progressive grinding, sieving and aspiration. Grain hardness has always been the major concern of dry millers because it determines grinding times and energy requirements, as well as the performance of the final products. The quality standards required vary widely with different end uses, and to some extent, with the type of plant used to process the grain. For gritting, a hybrid with a 'hard' endosperm will usually yield the highest proportion of grits and the lowest proportion of flour (Watson, 1977; Wu et al., 1991; Hardacre, 1994). Processors pay more for grits than for flour.

In New Zealand, the corn-based snack food market is growing at about 25% per year due to the increasing consumption of convenience and snack foods (Hardacre, 1995). This has prompted food processors to look for more consistent, higher quality maize grits and flour from millers, and in turn millers are looking for more 'hard' grain from growers. Generally, maize hybrids containing soft endosperm are preferred by wet millers, because a softer grain requires less steeping time and gives better starch-protein separation (Wu et al., 1991). Therefore, maize hardness is of great significance to producers, millers and processors in the grain trade.

The maize seed is a single fruit called a kernel, which has three main morphological parts: the pericarp, endosperm and embryo or germ. Each part of the kernel has a different composition. The endosperm is the largest fraction of the kernel, composed largely of starch, and is usually 82-84% of the kernel dry weight (Watson, 1987). The endosperm is of two types: hard (horny) and soft (floury). In the hard endosperm, the protein matrix is thicker and remains intact on drying, so binding the starch granules tightly together in a strong structure with a translucent glassy appearance. In the floury endosperm, the starch granules shrink during drying, tearing the thin protein matrix, resulting in round loosely bound starch granules. The endosperm often contains voids and is structurally quite weak (Duvic, 1961). The outer region of the kernel tends to be comprised of hard endosperm while the inner region tends to be comprised of soft endosperm. The hard endosperm contains a high level of protein while soft endosperm contains a higher level of starch throughout the kernel (La'sztity, 1984).

The texture of the maize endosperm is variable according to the type of maize and the region of the kernel. There are six major types of kernels: dent, flint, flour, sweet, pop and pod corn. Major differences are largely based on the hard and soft endosperm ratio, quantity and pattern of endosperm composition. The endosperm of flour maize (eg. opaque-2) has a thin protein matrix throughout, while the endosperm of hard maize (eg. dent and flint) has a thicker protein matrix (Robutti et.al., 1974). The endosperm of the maize types differs not only in protein matrix thickness but also in hard to soft (H/S) ratios, thickness of the subaleurone layers, cell size and protein components (Christenson et al., 1969). In the hard endosperm, starch granules are tightly packed together, each held firmly in a protein matrix, while in soft endosperm, the starch granules are held together more loosely (Eckhoff, 1992). The protein content of the whole kernel varies from 6-18% depending on the hybrid and agronomic conditions (Pomeranz and Bechtel, 1978).

The composition of the maize kernel depends on the different constituents present and well documented evidence shows that the protein content is influenced by the available nitrogen and genetic makeup. Duvic (1961) has shown that the total protein



content of maize grain can vary from 4.4 to 26.6% without influencing grain yield. Changes in total protein content are primarily changes in endosperm protein content, mainly zein, and as the protein content increases, the amount of hard endosperm also increases (Hinton, 1953).

Kernel hardness is related to the physical and chemical properties of the endosperm. Physical properties of the grain depend on kernel density, breakage susceptibility, kernel hardness, water absorbity and average kernel weight (Weller et. al., 1988) and intrinsic quality characteristics depend on starch, oil and protein content which ultimately affect the value of the end product (Hurburgh, 1989). However, maize hardness is correlated with differences in the hard to soft (H/S) endosperm ratio. Although these quality differences mainly depend on the genetic make up of the hybrid, differences are also caused by environmental factors such as temperature, moisture and soil nitrogen supply (Watson, 1987).

Response of crops to nitrogen is highly variable, due to the complex interplay of soil factors affecting its availability and also environmental conditions. As nitrogen is the major constituent of plant proteins, enzymes, amino acids and ribonucleic acid which constitute the genetic code, it is important not only for the production but also for the quality of the grain. In general, the grain yield of maize and the concentration of protein in the kernel increases in response to nitrogen, while nitrogen stress generally decreases grain protein concentration, yield and kernel texture quality (Tsai et al., 1992).

Differences in kernel protein and yield responses to soil nitrogen levels may also be influenced by genetic differences in the capacity of genotypes to take up nitrogen from the soil and translocate it to the sink kernels (Pollner et al., 1979). In general, grain yields among maize hybrids are negatively correlated with protein concentration or positively correlated with C:N ratio. For a given hybrid, the grain yield induced by nitrogen fertility is highly correlated with its protein concentration and grain protein concentration may serve as a parameter for estimating the amounts of nitrogen fertilizer required for maximum yield of a given hybrid (Tsai et al., 1992).

It also seems (Hardacre unpublished data) that some hybrids show more variability in endosperm quality in a given range of environments than others. Clearly, for stability in a variable climate, hybrids with more uniform grain quality are preferred.

Environmental factors can influence the relationship between grain protein and yield. Temperatures over the grain-fill period in wheat have been shown to influence this relationship (Stevenson, 1987). The influence of temperature on grain growth is reflected in its effect on sink capacity, and the rate and duration of grain filling during the effective grain filling period (Jones et al., 1981). Mock and Pearce (1975) suggested that the grain filling period in maize should be as long as possible to allow maximum production and storage of dry matter.

In New Zealand there is little information on grain quality and nitrogen fertilizer relationships, and such data are urgently needed by growers and contractors. The rapidly growing snack food industry requires high quality grits and flour from the dry milling industries, which can be produced more effectively from hard grain. Therefore, this study was undertaken to examine the effects of three different rates of nitrogen fertilizer on three different hybrids sown at two different sowing times. The following objectives were set:

- To investigate the influence of different rates of nitrogen fertilizer on grain quality, especially grain protein and hardness of three different hybrids;
- To investigate the influence of different sowing times on grain yield and quality of three different hybrids.

## CHAPTER II

### LITERATURE REVIEW

#### 2.1 GROWTH AND DEVELOPMENT OF THE MAIZE PLANT

The growth pattern of the maize plant can be roughly divided into three periods, i.e. the vegetative, reproductive and grain filling periods. Seed germination to floral initiation is the vegetative period; floral initiation to silking is the reproductive period; and silking to formation of the 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).

Most of the growth during the vegetative period is concerned with leaves. Growth of leaves and the 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 a maximum at around tasselling and usually results in the peak rate of canopy photosynthesis (Waldren, 1983).

Growth of grass and cereal leaves is well described by Langer (1979) and Dale (1982) and is 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 close to the base of the stem and up to the production of the fourth or fifth leaf is below the soil surface.

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 out-growth 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 of the lower portion leads to growth of the leaf sheath.

Cell division and enlargement, largely in the basal region of the leaf sheath, cause the younger lamina to move up inside the folded sheaths of the 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 cell. 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 is moving inside the previous leaf sheath prior to exertion.

The number of leaves formed in maize is determined by the number of leaves present in the seed embryo, the rate of leaf initiation at the apical meristem and the duration of the vegetative phase (Hunter et al.,1974). These processes are in turn influenced by photoperiod and temperature (Warrington and Kanemasu, 1983b; Hardacre and Turnbull, 1986).

The pattern of increase in grain weight is a sigmoidal curve. The linear phase, or rapid dry matter accumulation in the kernel, occupies about half of the period in which more than 90% of the grain dry matter is accumulated. When the kernel is at its physiological maturity, a black layer is developed in the placental region of the kernel and probably cuts off movement of assimilate into the developing floret (Daynard and Duncan, 1969). After this stage, there is rapid loss of moisture from the grain. The rate of drying of the kernel in the field depends upon the hybrid, properties of the husk, grain and also the climatic conditions of the field (Larson and Hanway, 1977).

## **2.2 EFFECT OF NITROGEN ON CROP GROWTH AND DEVELOPMENT**

Just after emergence, seedlings rely on starch and fat reserves in the seed, but once chlorophyll is developed and seedlings are established, plants become independent of seed reserves. At this time, extrogeneous nitrogen supply begins to play an important role very early on in plant life. However, the requirements are minimal during this period compared to the whole life cycle (Arnon, 1975).

From the beginning of the linear phase, the rate of leaf expansion increases. This results in a higher demand for nitrogen compared with the previous phase. Starter fertiliser generally increases the rate of leaf appearance of maize (Eik and Hanway, 1966) because it probably increases the number of leaf cells resulting from the higher rates of cell division (Arnon, 1975).

Leaf growth is retarded in nitrogen depleted plants because of a reduction in chlorophyll content, resulting in a reduction in photosynthate supply and therefore negatively affecting growth of young leaves (Dale, 1982). The importance of the leaves for plant growth is that they are the major sites for the production of the carbohydrate and moreover, they also play an important role in amino acid synthesis and nitrogen storage (Novoa and Loomis, 1981).

During the early growth stages, leaves acts as a 'sink' for nitrogen, but later on they also start to play a role as a source of nitrogen through remobilisation; that is, mature leaves can provide nitrogen for the growth of the younger leaves and ears (Arnon, 1975). Therefore, there are two sources of nitrogen: from the soil and from the plant itself. This suggests that under short term, or mild nitrogen stress conditions, plants may not suffer in reduction in leaf area. It has been found in *Lolium* spp. that even when nitrogen stress approaches 100% (zero growth on a dry weight basis), leaf expansion still continues. This seems to be caused by a rapid recycling of nitrogen from the older leaves to the emerging leaves (Dale, 1978 and Greenwood, 1976).

Langer and Liew (1973) found that nitrogen did not affect 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. The number of leaves of cereal crops may not be affected by nitrogen supply because it is largely determined by genotype (Allen et al., 1973). However, under nitrogen stress leaf area and probably leaf function is severely affected.

The effect of nitrogen on leaf area is well established (Stoskopf, 1981) and apart

from increasing leaf area, nitrogen fertilizer can also increase chlorophyll content of the leaves, which has a direct relationship to flowering and grain yield ( Hurduc and Stefan, 1966).

From the onset of flowering to early grain formation, the nitrogen requirement of maize plants reaches a peak (Arnon, 1975). Ears start to act as strong 'sinks', and consequently nitrogen reserves from plant parts are gradually translocated to the ears. In this situation an adequate nitrogen supply from the soil can delay the senescence of leaves; otherwise a significant amount of nitrogen will be translocated from the leaves to overcome the nitrogen shortage leading to premature leaf death. Consequently, the photosynthetically active leaves and growth of ears will be affected.

Although nitrogen plays an important role in promoting dry matter production, excessive supply of nitrogen can cause lodging, which reduces grain yield, if lodging occur after flowering. Fisher and Smith (1960) reported that lodging in maize was significantly increased when high rates of nitrogen fertilizer were applied without balancing the potassium supply.

### **2.3 GENERAL ASPECTS OF NITROGEN**

Nitrogen is a major nutrient and it is essential to the growth and reproduction of plants. It plays a major role in the development and function of the cell, being an essential constituent of all proteins. Nitrogen is generally absorbed by the plant in the form of either ammonium or nitrate ions. Maize has a substantial need for nitrogen fertilizer for high yield and quality in intensive agriculture (Arnon, 1975).

The demand for nitrogen in a growing crop is dependent on growth rate and nitrogen content of new tissue, which in turn varies with nitrogen availability, moisture availability, temperature and plant competition. Maize production is limited by nitrogen deficiency more often than by that of any other nutrient (Nova and Loomis,

1981).

Grain yield in maize and yield responses to nitrogen fertilizer are related to the concentration of available nitrogen in the soil. Nitrogen is removed in the grain in larger amounts than any other nutrient supplied by soil or fertiliser, and the grain yield and composition of the maize plant is primarily determined by the nitrogen status of the soil within the confines imposed by climate and management (Steele, 1985).

Nitrogen use efficiency in the crop is the expression of nitrogen uptake efficiency and the utilization efficiency of the nitrogen absorbed in producing grain (Moll et al., 1982). Hybrid maize with limited nitrogen may be characterised by a low concentration of protein which produces kernels which are less translucent, have a lower portion of hard endosperm and are more susceptible to breakage (Tsai et al., 1983).

### **2.3.1 FUNCTION OF NITROGEN IN PLANT GROWTH**

The major functions of nitrogen in plant growth are (i) as a component of the chlorophyll molecule, (ii) as a component of amino acids (the building blocks of proteins), (iii) in carbohydrate utilization, (iv) components of enzymes and (v) to support the uptake of other nutrients ( Donald et al., 1963; Olson and Kurtz, 1982). Nitrogen is most commonly the key limiting factor for crop production. Thus, on average, considerably more nitrogen than any other elements is supplied to crops as fertilizer and is removed from agricultural lands in harvested crops (Olson and Kurtz, 1982)

### **2.3.2 SOURCES AND FORMS OF NITROGEN**

Even though the atmosphere is about 80% nitrogen, most higher plants (except some legumes) are unable to utilise this molecular nitrogen ( $N_2$ ) until it is chemically

bound with hydrogen, oxygen or carbon (Bray, 1983). Most of the soil nitrogen reserve is as organic matter, and hence relatively unavailable to higher plants unless broken down by soil microorganisms into organic ions (Thompson and Toreh, 1978).

In order to maximize crop production, nitrogen is often supplemented by the addition of inorganic, or organic nitrogen fertilizer. Most of the inorganic nitrogen fertilizers can provide either  $\text{NH}_4^+$  or  $\text{NO}_3^-$  or both. Although both ammonium and nitrate can be utilised by maize, most of the ammonium applied to or produced in soils is converted to nitrate by microbial action (Benson and Pearce, 1987). Therefore, most of the nitrogen taken up by maize is in the  $\text{NO}_3^-$  form, which is very mobile and can be lost by leaching or by volatilisation.

### 2.3.3 NITROGEN DEFICIENCY AND EXCESSIVE NITROGEN

Certain levels of nitrogen must be present in plant cells for optimum utilization of the carbohydrate produced during photosynthesis. Nitrogen deficiency affects the chlorophyll content and causes excessive deposition of carbohydrates in vegetative cells and consequent thickening of the cell wall, thereby limiting formation of protoplasm (Olson and Kurtz, 1982).

Nitrogen deficiency also limits the production of protein and absorption of other minerals essential for growth. This causes a decrease in cell size and a decrease in cell division (Devlin, 1975). Consequently plants appear spindly, stunted and their chlorophyll concentration is lower compared with healthy plants (Thompson and Troeh, 1978). In nitrogen deficient maize, a V-shaped pale symptom usually starts from the tip of the lower leaves with the sharp end progressing inwards along the midrib of the leaf. This can cause the premature senescence of these leaves (Arnon, 1975).

Plants can take up excessive nitrogen, especially if some other nutrients, eg. phosphorus, or potassium are inadequate (Thompson and Toreh, 1978). Under these



conditions, there is a tendency for leaf cell number and cell size to increase, resulting in an overall increase in leaf production and the plants generally produce dark and succulent vegetative growth. In some cases, vegetative growth may be at the expense of grain production.

#### 2.3.4 NITROGEN TRANSLOCATION AND ASSIMILATION

After ammonium and nitrate have been absorbed into the plant, they will be translocated to the plant parts, reduced (transformed) into organic forms and incorporated into proteins before being utilized by the plant. Assimilation of nitrate includes three reductive processes and one non-reductive process in converting nitrate to amino acid (Schrader and Thomas, 1981). The reaction for this pathway have been discussed in several reviews (Beevers and Hageman, 1980; Mifflin, 1980; Schrader and Thomas, 1981). The four enzymes involved are nitrate reductase, nitrite reductase, glutamine synthetase and glutamate synthase and the chain reactions are as follows (Schrader, 1981):



Amino acids can be transported circularly between roots and leaves. That is amino acids formed in the roots may be transported through the xylem and added with those in the leaves, being polymerised into protein there. Similarly amino acids in the leaves may be transported through the phloem to the roots and polymerised there (Novoa and Lomis, 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 storage protein (Kirkby, 1981).

#### 2.3.5 FACTORS AFFECTING NITROGEN ABSORPTION IN MAIZE

Plants obtain nitrogen either by roots approaching the source of nitrogen, or nitrogen

is translocated from the original site to plant roots. The mechanisms of absorption of the nutrients into the plant are of three types eg. root interception, mass flow and diffusion. Maize plants absorb virtually all of their total nitrogen by mass flow (Donhaue et al., 1977). Generally, there are three factors which affect nitrogen absorption in plants:

(i) **Ion factors:** There are some antagonistic reactions in the ion absorption. The most common effect is between nitrate and chloride ( $\text{Cl}^-$ ). Higher concentration of chloride in the nutrient medium lowers the nitrate uptake (Mangel and Kirkby, 1982).

(ii) **Soil factors:** Low soil water content can cause low nitrogen uptake, because lack of water closes stomata which reduces carbon dioxide intake and results in lower photosynthetic activity (Jackson et al., 1976). This can reduce the uptake of nitrogen (Jackson et al., 1976). Under water logged conditions oxygen content is reduced and this can change the structure, or morphology of the root system and influence the ion absorption (Drew, 1979). In severe cases accumulation of the products of anaerobic respiration will kill roots or severely inhibit root activity. Root temperatures of lower than  $10^{\circ}\text{C}$ , or greater than  $40^{\circ}\text{C}$  retard uptake of nitrogen (Shaw, 1976).

(iii) **Plant factors:** The proportion of ions transported across the cell membranes depends on the specific permeability of membranes to particular ion species. In the case of nitrogen both ammonium and nitrate are monovalent and selectivity of the ions is small (Salisbury and Ross, 1978).

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

### 2.3.6 NITROGEN FERTILIZER AND YIELD DEVELOPMENT

Generally crop yields depend on the yield components i.e. plants/m<sup>2</sup>, ears/plant, grain/ear and grain weight. A high degree of flexibility of these yield components allows adaptation to a wider range of environmental conditions (MacEwan, 1964). The first three components together determine the potential number of grains/m<sup>2</sup> and grain weight is determined after anthesis.

There are a large number of factors affecting crop yield and its quality eg. edaphic, climatic, genetic, chemical, phytosanitary and economic factors. Factors such as variety, climate, soil and agronomic techniques etc, may vary the quality of the crop within very large limits. However, it is the assimilation of carbon and its distribution within the plant that actually determines yield (Langer, 1967).

The demand for nitrogen in a growing crop is dependent on the growth rate and nitrogen content of new tissue, which in turn varies with nitrogen availability, moisture availability, temperature and plant competition (Novoa and Loomis, 1981).

The response of maize to nitrogen fertilizer is dependent on the availability of soil nitrogen, with soils having large reserves of nitrogen being very unlikely to produce yield responses to nitrogen fertilizer (Steele, 1985). In New Zealand, crop history is a strong indicator of soil nitrogen reserves, particularly the length of period since the ground was in permanent clover- based pasture or previous legumes crops eg. peas (Wright, 1967), due to the large reserves of organic nitrogen present under these conditions (Keeney and Gregg, 1982).

In general, grain yields among maize hybrids are negatively correlated with protein concentration, or positively correlated with C:N ratio. However, recent work by Tsai et al.,(1992) showed that the grain yield of maize hybrids increased in response to nitrogen, and also in the concentration of the protein in the kernels.

For a given hybrid, the grain yield induced by nitrogen fertility is highly correlated

with its protein concentration. Grain protein concentration may serve as a parameter for estimating the amount of N fertilizer required for maximum yield of a given hybrid ( Tsai et al., 1992).

## **2.4 EFFECTS OF THE ENVIRONMENT ON GROWTH AND DEVELOPMENT OF MAIZE**

### **2.4.1 TEMPERATURE**

Maize is a warm weather crop whose growth and developmental processes are strongly influenced by temperatures between 10 and 30°C (Duncan, 1975) and are optimal at temperatures between 21 and 27°C (Shaw, 1976). Hardacre and Turnbull (1986) reported that maize seeds germinate best at soil temperatures above 10°C, but at temperatures below this point, germination and growth rate decrease sharply, or cease. In New Zealand conditions soil temperatures of approximately 6 to 8°C and air temperatures of approximately 15°C are considered to be the minimum for maize growth (Hardacre et al., 1993).

The time at which tasselling and silking occur is also very temperature dependent. Cal and Obendorf (1972), Bonaparte (1975) and Warrington and Kanemasu (1983a) have all reported that increases in temperature reduce the time to anthesis. Bonaparte (1975) observed that an increase in temperature resulted in the acceleration of the development rate, as evidenced by a substantial reduction in days to anthesis.

Warrington and Kanemasu (1983a) studied temperature effects on tassel initiation and anthesis of two maize hybrids (XL45 and W346). They found that under warm temperature (>23° C) floral initiation was rapid, occurring in 17 days or less after sowing, whereas under continuous cool temperature (<15° C) plants took 40 days or more to reach that developmental stage.

Even though an increase in temperature results in the speed up of flowering, this may have an adverse effect on fertilisation if temperature rises above an optimum level.

Berbacel and Eftimescu (1973) found, maximum temperature above 32°C around tasselling and pollination sped up the differentiation process of the reproductive parts, but resulted in higher rates of kernel abortion.

#### **2.4.2 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 temporarily stored in the A horizon to be absorbed by plants later, and some may gradually percolate down to the lower horizons to be stored there.

Water is very much essential for any metabolic activity, although the total quantity of water required for growth in maize is relatively small. It has been estimated that less than 1% of water absorbed by maize during its growing stages is retained in the plant (Street and Opik, 1984). Most of the water entering a plant is lost by transpiration (Kramer, 1959).

Apart from the influence on metabolic activities, water also plays an important role in nutrient availability to plants. Since most of the fertilizers are in the forms of compounds, they need to be hydrolysed 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) as fertilizers tend to be applied on, or near, the soil surface. Therefore, soil water is needed to transport the dissolved nutrients to plant roots.

During the active vegetative stage, maize plants grow very rapidly and water use is greater than in the early stage of emergence. This is due to the higher leaf area index which increases transpiration. Water balance is very important at this period. Shaw (1983) 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.

### **2.4.3 EFFECT OF SOWING TIME ON GRAIN YIELD AND QUALITY OF MAIZE**

Planting time is one of the most important components in the maize cropping system which can influence the yield potential and quality of the grain. Delayed sowing generally depresses yield (Hatfield et al., 1965; Pendleton and Egli, 1969; Calvin et al., 1992) and kernel hardness and breakage susceptibility (Bauer and Carter, 1986; Kniep and Mason, 1989) because plants grown earlier get more time in the vegetative, reproductive, and also in grain filling periods. In New Zealand, delayed planting or later maturing hybrids delays dry down and increase the risk of cob-rots and thereby reduced yield and quality of the grain (Hardacre et al., 1993).

There is little information available on the effects of sowing date on maize grain quality. Helm et al., (1968) reported that delaying sowing caused a reduction in grain nitrogen concentration of several high amylose hybrids. Delaying sowing also caused a reduction in kernel hardness and a consequential increase in kernel breakage susceptibility (Bauer and Carter, 1986). Planting date may also affect the nitrogen concentration of the kernel. Ahmadi et al., (1992) found that when sowing was delayed, yield was reduced, but kernel nitrogen concentration increased.

## **2.5 THE MILLING OF MAIZE**

Maize or maize (*Zea mays* L.) has achieved a higher level of industrial utilization than any other cereal grain (Alexander, 1987). It is used by the milling industry in wet and dry milling processes to produce a wide range of products. The products from the wet milling process are oil, starch and dextrose syrups, and from dry milling a range of grits and flours (Eagles and Chappel, 1984 and Hardacre, 1995 ).

### **2.5.1 Wet Milling**

The wet milling process involves an initial water soak under carefully controlled conditions to soften kernels. The maize is then milled and its components separated by screening, centrifuging and washing to produce starch, oil, feed by products and

sweetener (May, 1987).

### **2.5.2 Dry Milling**

The dry milling process breaks down the maize kernel into its components. Two different systems are used in dry milling: non-degerming and degerming. The non degerming system grinds the whole maize grain into meal. The degerming system (tempering-degerming; i.e after cleaning by dry and wet process as the maize is tempered to equilibrate the grain moisture by water or steam) removes, essentially, all of the germ and hull and leaves the endosperm relatively free of oil and from pericarp, for the production of a range of grits, meal and flour (Alexander, 1987).

The objectives in dry milling are to obtain the maximum yield of grits and the minimum amount of flour (Kent, 1975). Dry milled products are used by the maize-based snacks industry to produce a wide range of snack foods which include maize flakes, maize chips, twisties, cheeseballs etc. In New Zealand, the market is growing rapidly at about 25% per year (Hardacre, 1995).

## **2.6 EFFECT OF HYBRIDS ON ENDOSPERM QUALITY**

Hybrid cultivars are produced by crossing inbred lines and the desired characteristics can be manipulated by different methods of breeding (Zuber and Darrah, 1987). Hybrids differ in grain yield potential, days to maturity, standability, grain drying rate and resistance to pests and diseases.

A range of maturity and endosperm types are available from maize hybrids appropriate to the New Zealand growing environment. Commonly grown hybrids tend to have soft endosperm. While hard endosperm hybrids are available, they tend to have lower yields or are of inappropriate maturity (Hardacre, 1994). However, differences in dry milling quality among commercial hybrids have been demonstrated with "hard" hybrids exhibiting more desirable milling characteristics (Pomeranz et al., 1987; Peplinski et al. 1989; Li et.al., 1995 unpublished).

In terms of kernel hardness characteristics there are five general classes of maize- flint maize, pop corn, sweet corn, dent maize and flour maize (Watson, 1987a and Zuber and Darrah, 1987).

-Flint maize (*Zea mays tunicata*): has the hardest kernels due to the presence of a large and continuous volume of hard endosperm, with a small soft endosperm in the centre;

-Pop corn (*Zea mays tunicata*): has a hard endosperm in the outer layer, but the inner portion of the grain contains soft starch. When the grain is heated the inner portion of the grain expands and 'pops' the kernel open;

-Sweet corn (*Zea mays sacchorata*): contain recessive genes which prevents the conversion of sugar to starch. The grains are translucent and very wrinkled because when sugars dry they occupy less room in the grain;

-Dent maize (*Zea mays endenta*): is a derivative of flint-floury crosses and has a harder endosperm around the side. The soft endosperm at the top shrinks when mature, which results in the formation of the dent shape;

-Floury maize (*Zea mays amylacea*): has mostly soft endosperm throughout the kernel.

Flour maize kernels are soft, with no hard endosperm, dent maize kernels are intermediate and flint and popcorn types are hard, with a high proportion of hard endosperm (Pomeranz et al.,1984). However, within each type, kernel hardness is related to differences in hard to floury ratios, pericarp thickness and cell structure (Szaniel et al., 1984).

## **2.7 EFFECTS OF ENDOSPERM ON DRY MILLING PRODUCTS**

Maize endosperm products are classified into three categories on the basis of particle size i.e. grits (coarse), meal (medium) and flour (fine). Dry-milled products in these categories can be further divided into different size fractions for various commercial uses. The largest grits (flaking grits) are used to make corn flakes for breakfast cereals, whereas smaller grits are used in other breakfast cereals, extruded maize



snacks and brewed alcoholic beverages (Eckhoff,1992).

### **2.7.1 GRITS PRODUCTION**

The amount of grits and meal produced depends upon the proportion of the hard to soft endosperm in the kernel (Brekke, 1970). The ratio among endosperm fractions varies with the quality of the maize and the milling conditions. Products are recovered from the dry milling process by progressive grinding, sieving and aspiration. The typical yield of products from dry milling are: hominy feed, 35%; maize oil,1%; grits, meal and flour, 60%; losses, 4% (Watson, 1977). All these products come from maize endosperm and are used for different purposes according to their particle size.

Hybrids may differ in the ratio of hard to soft endosperm, relative size of the germ, thickness of the pericarp and also the ratio of starch to protein which eventually affects the type of end product produced (Watson, 1977). The amount of 'hard' endosperm is an important factor in grits yield. For gritting, hybrids with hard flinty endosperm will usually yield the highest proportion of high quality grits (Hardacre, 1994) and hybrids with soft kernels will usually yield poor quality grits (Wu, 1992).

### **2.7.2 GRITS QUALITY**

Specific quality characteristics in the dry milling of maize are important because: (a) effective processing depends upon certain physical properties of kernels, large deviations from which may cause erratic mill separation; (b) the use of these primary products is in foods where purity is highly important and (c) dry milling has less ability to purify products (Watson, 1977).

Grits yield and specially fat content of the maize grits and flour are subsequently affected by the quality and age of the grain, as considerable migration of fat from the grain into the endosperm occurs with ageing (Wyss, 1965). High fat content in

grits can be caused by pieces of free germ in the grits attached to the large flaking grits causing quality deterioration of the product (Paulsen and Hill, 1985).

## **2.8 FACTORS AFFECTING ENDOSPERM QUALITY FOR DRY MILLING**

Wet and dry milling uses of maize depend on the kernel physical properties (Kniep and Mason, 1989). Kernel physical properties such as hardness, breakage susceptibility and density may depend on the protein concentration of the grain. Kernel density is one of the indicators of kernel hardness, based on the hard to soft endosperm ratios. Lack of hard endosperm results in lower grain density (Lambert et al., 1969 and Klein et al., 1980) and lower dry milling yield of flaking grits (Brekke, 1970).

The quality of unblemished kernels varies because of the influences of inheritance and environment on the structure and composition of the kernel components. The physical properties of the grain include bulk density (test weight), kernel density, breakage susceptibility, kernel hardness, water absorbity and average kernel weight (Weller et al., 1988) and intrinsic quality characteristics depend on starch, oil and protein content which ultimately affect the end products (Hurburgh, 1989).

The increase in grain yields resulting from the application of nitrogen fertiliser to nitrogen deficient soils, and the increased protein percentages resulting from excessive nitrogen fertiliser applications have long been recognized. The kernel protein percentage increases most rapidly in response to nitrogen supply, after the yield response to nitrogen levels off (Deckard et al. 1984).

### **2.8.1 NITROGEN FERTILISER AND GRAIN PROTEIN**

The relationship between yield and protein percentage across soil N levels can vary

from negative to positive, depending on environmental conditions (especially soil water). Smika and Greb (1973) showed that soil  $\text{NO}_3^-$ -N content was positively associated with grain protein, whereas the opposite relationship was noted between grain protein and available soil water by Deckard et al., (1984). However, recent reports suggest that grain yield and protein percentage are potentially compatible (Frey, 1977, Tsai et al., 1992).

Differences in kernel protein and yield response to soil nitrogen levels may also be influenced by genetic differences through the capacity of genotypes to take up nitrogen from the soil and translocate it to the sink kernels (Pollner et al., 1979). The efficiency of translocation of the total nitrogen absorbed by the crop has a major influence on the protein content of grain and with increasing nitrogen supply generally grain protein increases (Deckard et al., 1984).

Maize hybrids grown without nitrogen fertilizer may contain only 6% protein, but this increases to about 12% when grown under high N conditions (Pierre et al., 1977). For example the protein content of maize grain was significantly increased (from 6% to 10%) as the rate of nitrogen fertiliser applied increased from 0 to 166 kg/ha (Warren et al., 1980).

Developing grains are strong sinks for the nitrogen compounds used to manufacture protein. This nitrogen can originate from mobilisation of plant reserves or from uptake during the grain fill-period. Nitrogen supply depends on soil factors such as humus content and the balance between immobilisation and mineralisation (Novoa and Loomis, 1981).

The level of protein may represent saturation of the N sink and the point at which the movement of additional nutrients into the kernel is restricted (Tasi et al., 1992). When protein concentrations are high, protein can continue to increase but at the expense of starch accumulation (Singletary and Below, 1989) and this terminates further increases in kernel weight and grain yield (Tsai, 1983).

The major storage protein in maize endosperm is primarily zein. Zein functions as the primary N sink to regulate the movement of assimilates into the kernels. The accumulation of the starch in the kernels is substantially reduced when the deposition of amino acids in the N sink is terminated (Tsai et al., 1980, 1983; Singletary and Below, 1989). High levels of nitrogen fertiliser prolong and increase the rate of grain-fill period as compared with low nitrogen levels (Tsai et al., 1984).

The synthesis of large amounts of zein under high rates of nitrogen fertilizer may increase yield but reduce protein nutritional quality, because zein is deficient in lysine and tryptophan. Indeed, these two amino acids, as a percent of protein, decrease with increasing rates of nitrogen fertilizer applied ( Tsai et al., 1978, Randig and Broadbent, 1979).

The concentration of amino acids and the protein fraction in maize grains are also influenced when grown at different levels of nitrogen. The concentration in the grain protein of tryptophan, lysine, glycine, arginine and thionine decreased and the concentration of alanine, phenylalanine, tyrosine, glutamic acid and leucine was increased by the application of nitrogen (Rending and Jimenez,1978; Rendig and Broadbent, 1979).

## **2.8.2 GRAIN HARDNESS AND GRAIN PROTEIN.**

The kernel has both hard and soft endosperm. The hard portion is hard, vitreous and translucent, while the soft part is soft and opaque ( Dombrink-Kurtzman and Wilson, 1992). Many studies have described differences between these two types of endosperm (e.g. Wolf et al., 1952; Christenson, 1969; Subramanyam, 1980).

Differences in compactness of cellular components, cell size, and cell wall thickness within the endosperm play a part in hardness differences (Wolf et al, 1952). Hardness could be due, in the simplest case, to the presence of a specific 'hardness protein' or the absence of a 'softness protein' (Dombrink-Kurtzman and Wilson, 1992).

In the hard endosperm, the starch granules are very small and surrounded by a thick protein matrix, whereas in soft endosperm starch granules are big and surrounded by a thin protein matrix (Wolf et al., 1969). The protein composition and distribution directly influences endosperm texture and physical properties, thereby affecting milling products (Dombrink-Kurtzman and Wilson, 1992).

Hamilton et al (1951) found average hard to soft ratios of 0.4 to 1.4 dependant on different nitrogen fertility levels. These differences in protein composition may relate to composition of protein bodies and to the texture of the endosperm (Dombrink-Kurtzman and Wilson, 1992).

Duvic (1961) has shown that the total protein content of maize can vary from 4.4 to 26.6% without influencing grain yield. Changes in total protein content are primarily changes in endosperm protein content, mainly zein, and as the protein content increases, the amount of hard endosperm increases (Hinton, 1953).

In the hard endosperm, the protein matrix is thicker and remains intact on drying so binding the starch granules tightly together in a strong structure with a translucent glassy appearance. In the floury endosperm, the starch granules shrink during drying, tearing the thin protein matrix resulting in round loosely bound starch granules. The endosperm often contains voids and is structurally quite weak (Duvic, 1961). The outer region of the kernel tends to be comprised of hard endosperm while the inner region tends to be comprised of soft endosperm.

Differences in kernel protein and yield responses to soil nitrogen levels may also be influenced by genetic differences in the capacity of genotypes to take up nitrogen from the soil and translocate it to the sink kernels (Pollner et al., 1979). Zein appears to play an important role in determining the textural quality of grain in addition to its effect on grain yield and protein nutritional quality (Deckard et al., 1984).

### **2.8.3 GRAIN HARDNESS AND GRITS PRODUCTION**

Grain hardness is an index of the relative amount of vitreous to soft endosperm, and

is a major component of test weight, dry milling and food processing characteristics (Leford and Russel, 1985). Therefore, it is one of the important concerns to producers and processors in the grain trade. Hardness is related to kernel density, bulk density, breakage susceptibility, storability, handling and processing (Pomeranz et al., 1984).

The most important factor for determining suitability for dry milling is kernel hardness, which is influenced primarily by hybrid. The harder the kernel, the higher the yield of large grits (Good and Hill, 1992). However, these properties of maize kernels affect grinding power requirements, nutritive properties, dust formation, production of special foods, and yield of dry and wet milled products. Intrinsic hardness is difficult to measure because of the complexity of the kernel structure, but it is closely related to the ratio of hard to soft endosperm (Watson, 1977).

In maize, the most widely used measure of density is test weight (bulk density) and hybrids with low test weight often have a lower percentage of hard endosperm and therefore produce a lower prime yield of large grits when milled (Rutledge, 1978). Test weight, measured as weight of grain per unit of volume, is the simplest and most widely used criterion of milling quality. However, many factors may influence the relationship between test weight and milling quality (Watson, 1987b).

Wichser (1961) stated that grits can best be made from maize containing an average of 70% hard and 30% soft endosperm, whereas maximum yield of flour is obtained from soft maize averaging 20% hard and 80% soft endosperm. Hard endosperm has a lower fat content (about 0.04%) and fluctuates less, whereas soft endosperm has a higher fat content but can vary considerably, from 1.1 to 2.4% fat. As shown by Brekke (1966) grits yield generally increases as the proportion of hard endosperm in the kernel increases.

#### **2.8.4 GRAIN HARDNESS AND GRITS QUALITY**

As stated earlier, the success of the dry milling process depends largely on the physical properties of the grain. A study by Shove (1969) demonstrated that

selection of maize lots on the basis of the kernel density, breakage susceptibility or test weight could significantly improve grits yield. Large differences in these properties can cause erratic mill separation and serious problems in maintaining the purity of the products (Watson, 1977).

Previously the characteristic commonly used to assess quality was the colour of the grain i.e. yellow vs white (Watson, 1977). However, today, the value of freshly harvested 'hard' maize is considered much more important for the manufacture of high quality dry milling products (Alexander, 1987).

Different types and qualities of maize will produce grits and flour with widely differing fat contents, even though the maize is milled under identical conditions. Yield and fat content of the maize grits and flour are affected by the quality of the grain to a much a greater degree than in wheat milled for flour (Wyss, 1965).

Quality of the grits is judged on colour, odour, fat content, moisture, and freedom from dust, pericarp and insect fragments (Roberts, 1967). Gritting quality of the grain will decrease in sub-optimal growing conditions such as insufficient fertilizer nitrogen, late sowing, cold growing temperatures, drought and also when using a hybrid of inappropriate maturity (Hardacre, 1994). All these factors can influence the grain endosperm structure and composition during crop development.

## CHAPTER III

### MATERIALS AND METHODS

#### 3.1 PLANT MATERIAL

Three maize hybrids with different kernel characteristics were used in the experiment. Hybrid P3751 has a relatively soft endosperm texture while that of P3787 is intermediate and hybrid A82-8xNZ84 has a hard endosperm texture (Hardacre, personal communication). Pioneer brand hybrids P3751 and P3787 are adapted to the Manawatu region of New Zealand and are used for commercial grain production. Hybrid P3751 and P3751 have both shown exceptional yield performance in the Manawatu region with fast grain dry down and outstanding drought tolerance. However, Hybrid P3787 attains its physiological maturity, the formation of the 'black layer' in the placental region, slightly earlier than P3751 (Pioneer Brand Seeds, 1992-93) making a marginal sowing date or a cooler season somewhat more secure. In terms of disease resistance, P3751 is also recorded as having greater resistance to *Fusarium* ear rot than P3787, and is slightly superior in terms of resistance to eye spot and *Gibberella* ear rot (Pioneer Brand Products, 1993-94).

Hybrid A82-8xNZ84 was developed from four Corn Belt Dent inbred lines, A239, A658, and H99 and four partially inbred lines from the CIMMYT pool line 5 (Eagles and Hardacre, 1989). Lines from this pool were selected because of their high combining ability for grain yield with Corn Belt Dent testers (Eagles et al., 1983; Eagles and Hardacre, 1985) and the Corn Belt Dent lines were selected because of their ability to produce high yielding hybrids in New Zealand (Eagles and Hardacre, 1989).

#### 3.2 EXPERIMENTAL SITE

The experiment was conducted at Massey University (40° 23' S) at an altitude of 33m above sea level on a mottled, fine, sandy loam (N.Z.S.B. 1976) at the Frewens Block (Appendix-1). Temperature and rain fall data during the growing season were collected at the nearby AgResearch Grasslands, Crown Research Institute campus,





x=yield harvest, xx=destructive harvest

N<sub>0</sub>= no applied urea, N<sub>1</sub>= 250kg urea/ha and N<sub>2</sub>= 500kg urea/ha

Each plot consisted of six rows, two from each hybrid.

### 3.4 CULTURE AND NITROGEN TREATMENTS

Two sowings were made at the same site and adjacent to each other on 28th October and 21st November, 1994. The block had previously been in a ryegrass/white clover pasture (1990-93). The block was ploughed one month before sowing and was harrowed two days before sowing. Soil samples were collected before ploughing and analyzed in the Soil Science Laboratory (Appendix-3). As a result of the soil test report 150kg/ha of Nitro-phoshka (N:P:K : 12:10:10) was applied to provide a basal level of soil nutrients.

To ensure optimum plant establishment and therefore population, seeds were sown at approximately 5cm depth at each sowing station with two seeds per station. Each row was marked out by a furrow of approximately 5cm depth created by using a hand held grubber, and seeds were planted by hand at a distance of approximately 0.75cm apart between stations. Plants were thinned to the correct density at the 4th leaf stage of development.

Nitrogen fertilizer, as urea (46% N), was used in the experiment. The treatments were no extra nitrogen, 115kg N/ha and 230kg N/ha. To avoid the loss of nitrogen by leaching and volatilization, urea was applied at three times, one third after seedling emergence, one third after canopy closure and the remaining one third one week after anthesis, all as a side dressing. The first application of urea was applied for the October sowing on the 21st of November i.e. 24 days after sowing (DAS), while the second and third applications were applied at 87 DAS and 111 DAS respectively. For the November sowing urea was applied at 24, 70 and 86 DAS respectively.

All the seed lots were supplied by Mr. A.K.Hardacre, Crop & Food Research Ltd,

Palmerston North. Before sowing, seeds were treated with Promet 300Ew (a.i. 300g/l of Furthiocarb) at a rate of 40ml/kg of seed and an Absorbent (talcum powder like substance as a coating) at 25g/kg seed was added to prevent stickiness. The pre-emergence herbicides, Alachlor EC (a.i. 480g/l of alachlor) and Atrazine (a.i. atrazine 500g/l) were applied on 2nd of November for the October sowing and on 25th of November for the November sowing. The rate of application for Alachlor was 6.5l/ha and for Atrazine was 3l/ha respectively, and the herbicides were applied through a knapsack sprayer. Thereafter any weeding was done by hand. After signs of infestation by cutworm (*Agrotis ipsilon aneituma* (Walker)) an insecticide, Hallmark 5 EC (a.i.; 50g/litre esfenvalerate plus 741g/litre xylene) was applied on 25th of November for the October sowing and 7th of December for the November sowing at a rate of 15l/ha with 3-4ml of Rain-Guard (a.i. poly-1-p-menthene, non ionic) through a knapsack sprayer to control the cutworm.

### 3.5. DATA COLLECTION

#### 3.5.1 GROWTH MEASUREMENTS

The number of seedling emerging every day was recorded from one row of each replicate until no further increase was noted. Total emergence was expressed as a percentage and calculations were made by using the following formula:

$$\% \text{ Emergence} = \frac{\text{Total emergence}}{\text{Total planted}} \times 100$$

Leaf numbers were counted from the two leaf stage of the plant for every sowing, when the formation of the 'ligule' was observed at the base of the leaf. The formation of the 'ligule' above the enclosing sheath of the preceding leaf indicated the end of elongation growth and that the foliar organ had reached its final length (Dale, 1982). The number of leaves per plant were recorded every 10 days by regular

inspection of 20 plants per replicate until tasselling, and the rate of leaf appearance subsequently calculated by recording the number of leaves over time for each hybrid. The mean number of days from sowing to 50 percent silk emergence was determined by visual inspection of 20 adjacent plants in each replicate.

Dry-down pattern of the grain was estimated by determination of the grain moisture content which was recorded weekly from physiological maturity, determined as the 'black layer' formation, to harvest for each hybrid. Three ears were selected at random every time from the 'destructive' half of the plot and grains were collected from two adjacent rows of the cob from the upper to the lower end. Percentage moisture content was determined by drying the grain at 130<sup>0</sup> C for 72 hours (Hardacre, personal communication ) and using the following formula:

$$\% \text{ Grain moisture} = \frac{\text{fresh weight} - \text{dry weight}}{\text{fresh weight}} \times 100$$

A Cumulative Growing Degree Days (GDD) system as proposed by Newton and Eagles (1991) was used to quantify durations between different developmental stages. The durations measured were seeding to 50 percent seedling emergence, seeding to 50 percent silking and seeding to physiological maturity as determined by the formation of the 'black layer' in the placental region of the kernel. Growing Degree Days (GDD) were calculated as:

$$\text{Growing Degree Days} = \frac{T_{\text{max}} + T_{\text{min}}}{2} - 6^{\circ}\text{C}$$

where T<sub>max</sub> and T<sub>min</sub> are daily maximum and minimum temperature. A base temperature of 6<sup>0</sup> C was used because it has provided the best fit for the temperature variation in New Zealand (Brooking and McPherson, 1989) and is also the base used for cool maritime environments in Europe ( Derieux, 1978).

To estimate the yield and yield components, ten plants were randomly selected from the non-destructive half and hand harvested from each plot. Yield and yield components were: number of plants per unit area, cobs/plant, grains/cob and 100-grain weight. Grain yield/ha was calculated by using the following formula:

Grain yield (t/ha)  $Y = P \times E \times N \times S$  where,

P= Plant population per hectare

N= Number of cobs per plant

E= Number of grains per cob

S= 100-grain weight

This calculated yield was then adjusted to that at a 14% grain moisture content. Harvesting occurred on the 6th of June, 1995 for both sowings. The grain moisture of the October sowing was 21.1% for P-3787, 23.9 % for P-3751 and 23.8% for A8-82XNZS-84, while for the November sowing grain moisture was 23.8 for P-3787, 24.9 for P-3751 and 25.3% for A8-82XNZS-84 respectively during harvesting.

Ten cobs were randomly selected from each replicate, dehusked and then spread in the glass house to dry under an ambient temperature of 25-30°C. After drying, all ears from each replicate were hand shelled and mixed. Mixed grains were then put in paper bags and kept at room temperature for moisture equilibration until the evaluation of grain quality and hardness characteristics.

## **3.5.2 LABORATORY METHODS**

### **3.5.2.1 PROTEIN ESTIMATION**

The protein content ( measured as nitrogen) in the maize grain sample is generally determined by the classical Kjeldhal analysis (Rasper, 1987). In this experiment the N content of the grain samples was determined by the macro Kjeldhal digestion method (AACC,1983). The nitrogen in protein and other compounds reacts with

boiling concentrated sulphuric acid in the presence of a selenium catalyst to form ammonium sulphate. The acid mixture is cooled, diluted with distilled water, and made strongly basic with sodium hydroxide. The ammonia is released and distilled into a boric acid solution. The ammonia in the boric acid solution is titrated with standardized hydrochloric acid using a Tecator Kjeltac auto 1030 analyzer. From the quantity of unrevealed acid determined by titration the quantity of released nitrogen is established and converted to protein for the sample.

From each treatment replicate about 10g of grain was taken randomly. All samples were then ground in a "Glen Creston" hammer mill until all grain was crushed and could pass through a 1mm sieve and be collected in the collection tube. Crushed samples were mixed and up to 0.2gm of samples weighed in duplicate and put in the digestion tube along with a 'Kjel tablet' which stimulated the digestion process. 10ml concentrated sulphuric acid was poured into the sample and mixed gently by swirling the tube by hand. The tube was then placed in the pre-heated digester (420°C). After completion of the digestion the sample became clear and colourless and was kept for cool down. Thereafter 30ml of distilled water was mixed into the sample for analysis. Before starting the analysis a blank sample of distilled water and a standard sample of Ammonium iron (II) sulfate hexahydrate GR containing 7.145% N was tested to set the analyzer correctly for each batch.

All nitrogen contents reported are on a dry matter basis. However grain nitrogen content was converted to protein content, using the conversion factor of 6.25 (AACC, 1983) and then adjusted to 14% moisture content.

### **3.5.2.2 STENVERT HARDNESS TEST**

Hardness of the grain constitutes a comparative measure of distinguishing between hard and soft kernels. In bulk tests hardness is estimated from the power or time required to grind a given quantity of grain, from the quantity of the abraded material, or from the particle size of the ground material (Rasper, 1987). The Stenvert



Figure-2. A Stenvert Hardness Tester



Figure-3. A Grain Analysis Computer



Hardness Test is based on the method described by Stenvert (1974) and Pomeranz et al. (1985).

A 20gm sample of grain was ground using a Glen-Creston micro hammer mill fitted with a 2 mm aperture particle screen. The mill speed was set to 7500rpm when empty and slowed substantially under load. This speed setting was considerably higher than recommended, but as the unit used was not fitted with a tachometer and as none was available at that time, a convenient setting was used. This method gave good estimation while estimating the hardness of 38 New Zealand grown maize hybrids (Li et. al., 1995 unpublished).

The mill used in this work was equipped with a computerized data logging system to log the instantaneous electric power consumption during the milling test. From these data the transient peak energy, total energy consumption and milling duration time for the 20 gm sample were determined.

Prior to collecting the data, the mill was switched on and allowed to warm up by milling a set of five dummy grain samples. The set of test samples was then milled and the data logged. Data acquisition began automatically, as soon as the power load increased above the unload power demand, and continued until power consumption decreased within 0.3 watts of the initial condition. Resistance time i.e. the time taken to mill 17 ml of meal, the energy required for milling and milling duration time was recorded. Samples from each replication were run several times until a 'good result obtained' message from the computer was received and then the data were averaged. A photographic view of the unit is presented in Figure 2.

### **3.5.2.3 BULK DENSITY AND GRAIN MOISTURE CONTENT**

Bulk density or Test Weight, measured as the weight of grain per unit volume, is the simplest and most widely used criterion of milling quality of grain (Rasper, 1987). To determine bulk density and moisture a Dicky-john GAC2000 grain analysis

computer was used at Crop and Food Research Limited, Palmerston North. The photographic view of the grain analysis computer is presented in Figure 3. The DICKY-john GAC2000 measures the capacitive reactance of a grain sample which is a measure of the dielectric constant and a predictor of grain moisture. As bulk density is highly dependant on grain moisture (Nelson, 1980), both parameters i.e, moisture and bulk density were determined simultaneously by the computer. Each sample was tested several times until a consistent result was obtained and then averaged for the final readings. Bulk density was recorded as kg/hectolitre and moisture as percent wet weight.

### 3.6 STATISTICAL ANALYSIS

Statistical Analysis System (SAS, 1992) was used to carry out statistical analyses. Analysis of variance was carried out on the data from the individual sowings. In the experimental design nitrogen and hybrids were fully replicated within each planting time but planting was not replicated. This is a limitation of the experimental design. Therefore effects of the different sowing times can not be separated from the block effects and it was not statistically possible to compare the effect of sowing on the response variables. Any comparison would have to assume that the block effect was negligible and that significant differences in response variables were due to sowing time not heterogeneity in the trial site. The model for the analysis was as follows:

$$X_{ijk} = \mu + B_k + U_i + \Sigma_{ik}^m + V_j + (UV)_{ij} + \Sigma_{ijk}^s$$

$\mu$  = Overall mean

$B_k$  = Block effect

$U_i$  = Nitrogen level

$\Sigma_{ik}^m$  = Main plot error

$V_j$  = Hybrid

$(UV)_{ij}$  = Hybrid and nitrogen interaction

$\Sigma_{ijk}^s$  = Split plot error

$i = 1$  to 3

$j = 1$  to 3

$k = 1$  to 3

Because the SAS programme does not give F-values for the interaction effect, Least Significant Differences (LSD's) for interaction effects for the analysis were calculated manually. Results have been presented with F test significance and LSD's. Where the F test indicates differences among treatment means, LSD's have been calculated using the one tailed t-test (unless otherwise stated) to separate treatment means. The closeness of the association in the series of experiments was measured by correlation analysis.

## **CHAPTER IV**

### **RESULTS**

#### **4.1 PLANT GROWTH AND DEVELOPMENT**

##### **4.1.1 SEEDLING EMERGENCE**

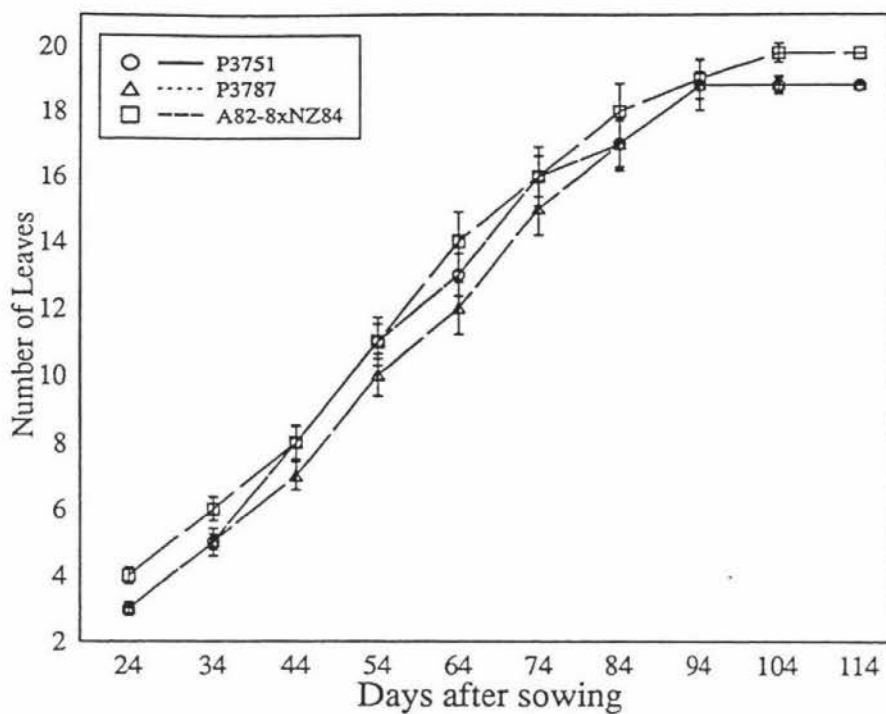
Field experiments were planted on the 28th October and 21st November, 1994. Hybrids P3751, P3787 and A82-8XNZS84 did not differ significantly in percent germination in the laboratory test or in field emergence for both sowings (Appendix 4). However, the time required for seedling emergence in the October sowing was greater (14 days) than in the November sowing (7 days). In terms of the growing degree days (GDD) the October sowing required 102.7 GDD and the November sowing 55.3 GDD respectively, from sowing to emergence.

##### **4.1.2 LEAF NUMBER AND LEAF APPEARANCE RATE**

For both sowings hybrid A82-8xNZ84 had a significantly greater number of leaves than hybrids P3751 and P3787. At the October sowing hybrids P3751, P3787 and A82-8X NZS-84 produced means of 18.8, 18.8 and 19.8 leaves per plant but in the November sowing they produced means of 18.9, 18.1 and 19.6 leaves per plant respectively ( Appendix 5).

The leaf appearance rate was significantly greater for the November sowing than for the October sowing as reflected in the fact that maximum leaf number recorded for the November sowing was reached in 81 days compared with 94 to 104 days for the October sowing (Figure 4). Leaf appearance rate indicated that the leaves of each hybrid in the November sowing appeared faster than those of the October sowing (Appendix 5).

### October Sowing



### November Sowing

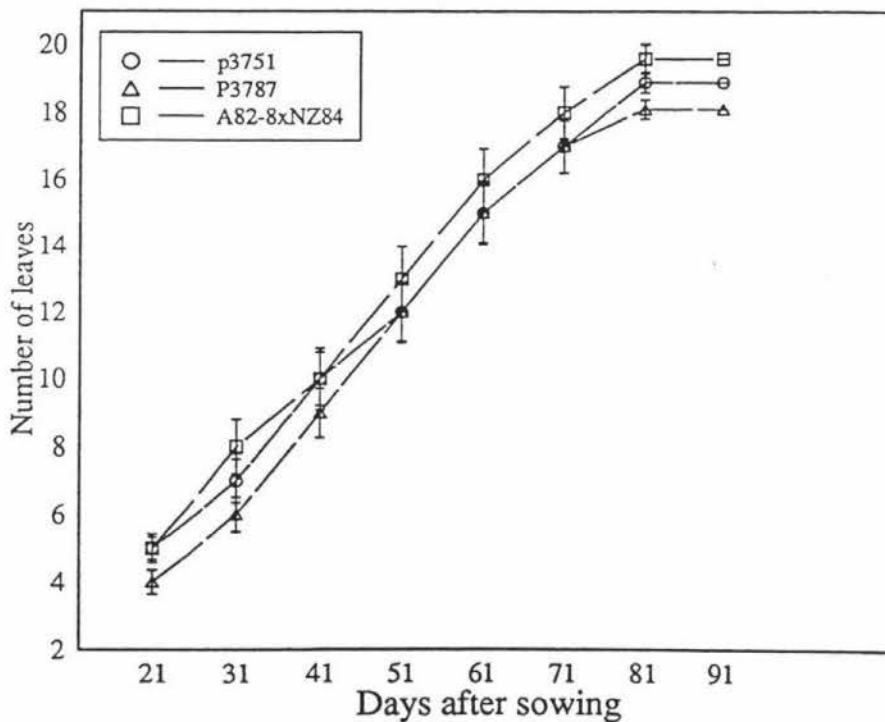


Figure-4. Leaf number and leaf appearance rate of P3787, P3751 and A82-8xNZ84 in both sowings

### 4.1.3 DAYS TO SILKING

Hybrid P3787 reached mid-silking significantly earlier than hybrids P3751 and A82-8XNZS84 in both sowings, while the latter two hybrids did not differ significantly at either sowing. In the October sowing P3787 reached mid-silking at 100.7 days after sowing (DAS) and P3751 and A82-8xNZ84 at 108.4 and 108.7 DAS respectively which was 13 days later than in the November sowing (Appendix 6).

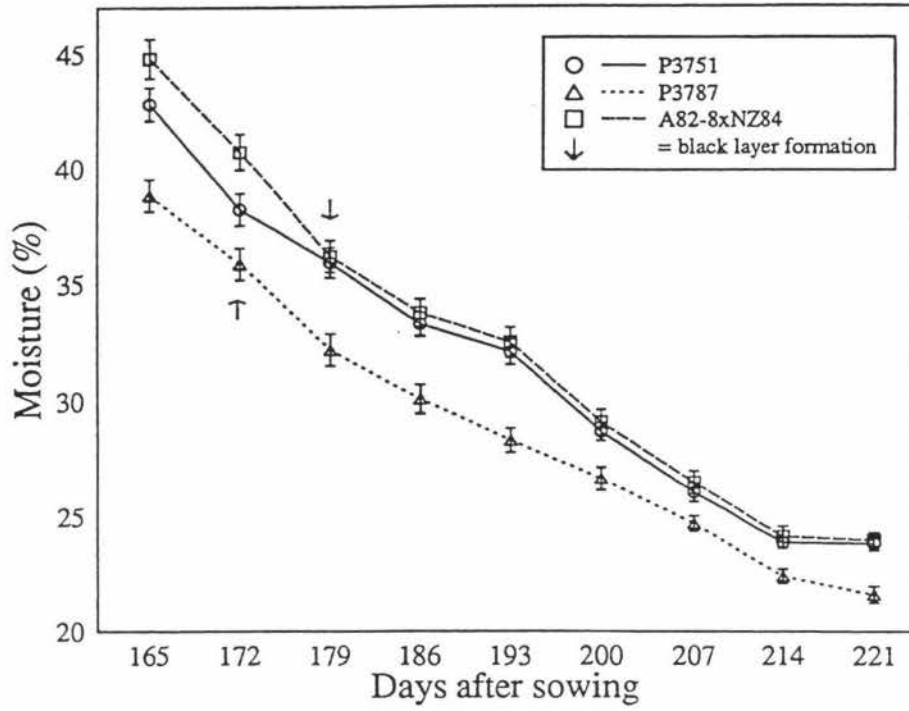
In terms of heat units required from sowing to mid-silking hybrid P3787 required significantly less than hybrids P3751 and A82-8xNZ84, but those two hybrids did not differ significantly. Hybrid P3787 required 961.3 GDD for the October sowing which was 13.0 GDD more than that for the November sowing. Hybrids P3751 and A82-8XNZS84 required 1047.4 and 1048.6 GDD for the October sowing which was 22.7 and 23.5 GDD more than for the November sowing (Appendix 6).

### 4.1.4 GRAIN MOISTURE DRY DOWN

Grain moisture loss from the time of 'black layer' formation to harvest was quicker in the November sowing (28-35 days) than in the October sowing (42-49 days) (Figure 5). At both sowings grain of hybrid P3787 dried significantly faster than that of the other two hybrids. Hybrid P3787 attained physiological maturity i.e. black layer formation, at 172 days after sowing (DAS) in the October sowing and 167 DAS in the November sowing, compared with the other two hybrids which took one week longer at 179 and 174 DAS in the October and November sowing respectively. While all the hybrids formed their 'black layer' at a similar grain moisture content (36% and 37%), this stage was reached one week earlier in hybrid P3787 (Figure 5).

The GDD required for the formation of the 'black layer' for hybrid P3787 was 1844.2 for the October sowing and 1779.2 for the November sowing, while hybrids P3751 and A82-8xNZ84 required 1991.3, 1831.4 and 1831.3, 1833.2 GDD respectively (Appendix 6).

### October Sowing



### November Sowing

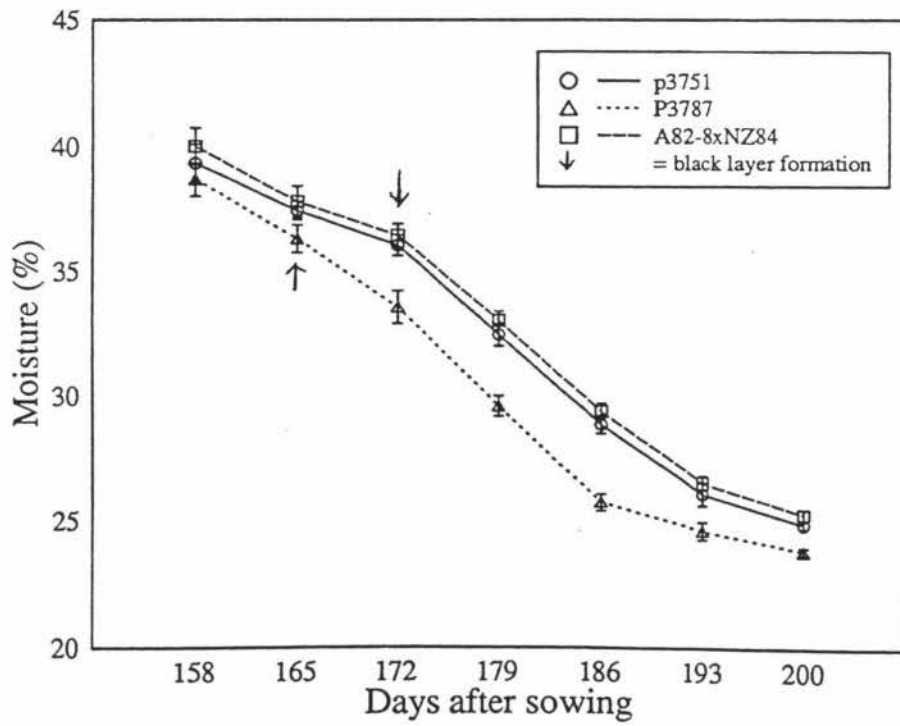


Figure-5. Grain moisture dry down rate of P3787, P3751 and A82-8xNZ84 in both sowings

## 4.2 YIELD AND YIELD COMPONENTS

The grain yield data presented in the following tables refer only to those effects that are statistically significant. The model used in the analysis of data was the same for each response variable and a full model of General Linear Models Procedure (GLM) for the analysis of variance is presented in Appendix 7. The statistical significance of the treatment effects for the October and the November sowing is presented separately. F-values at the 0.01 and 0.05 levels are shown as \*\* and \* respectively and ns denotes non-significant differences.

### 4.2.1 GRAIN YIELD

#### OCTOBER SOWING

Both nitrogen and hybrid significantly affected grain yield (Table 1). There was no interaction between nitrogen and hybrid.

**Table-1. Statistical significance of treatment effects for the October sowing for grain yield.**

Source	DF	F-Value	Pr>F
Replication	2	0.67	ns
Nitrogen	2	9.77	*
Rep x Nitro	4	1.03	ns
Hybrid	2	26.39	**
Nitro x Hybrid	4	0.77	ns

### NITROGEN EFFECT

The application of 250 kg/ha of urea increased grain yield significantly (Table 2). The application of 500 kg/ha resulted in a decline in yield from that recorded at 250 kg/ha, and this yield reduction did not differ from that of the control.



**Table-2. The effects of nitrogen fertilizer on grain yield**

	Urea (kg/ha)			Significance
	0kg	250kg	500kg	
Tonnes/ha	13.55	15.14	14.31	0.0287

LSD( $p < 0.05$ )= 1.02    CV%= 5.6

### **HYBRID EFFECT**

The grain yields of the hybrids compared were all significantly different (Table 3), with hybrid A82-8xNZ84 producing the highest yield, P3787 the lowest yield and P3751 being intermediate in yield.

**Table-3. The effects of hybrids on grain yield**

	Hybrid			Significance
	P3787	P3751	A82-8xNZ84	
Tonnes/ha	13.05	14.33	15.63	0.0001

LSD ( $p < 0.05$ )= 0.81    CV%= 5.3

### **NOVEMBER SOWING**

As for the November sowing, both nitrogen and hybrid affected grain yield, but there was no significant interaction (Table 4).

**Table-4. Statistical significance of treatment effects for the November sowing for grain yield.**

Source	DF	F-Value	Pr>F
Replication	2	1.82	ns
Nitrogen	2	13.20	*
Rep x Nitrogen	4	0.67	ns
Hybrid	2	39.23	**
Nitro x Hybrid	4	1.38	ns

### NITROGEN EFFECT

The application of 250 kg/ha of urea increased grain yield significantly (Table 5). However, the application of 500 kg/ha did not increase grain yield.

**Table-5. The effects of nitrogen fertilizer on grain yield**

	Urea (kg/ha)			Significance
	0kg	250kg	500kg	
Tonnes/ha	13.51	15.01	14.24	0.0227

LSD ( $p < 0.05$ ) = 0.81    CV% = 5.4

### HYBRID EFFECT

The grain yields of hybrid P3751 and A82-8xNZ84 were significantly greater than that of P3787, but the yields of the former two hybrids did not differ (Table 6).

**Table-6. The effects of hybrids on grain yield**

	Hybrid			Significance
	P3787	P3751	A82-8xNZ84	
Tonnes/ha	12.45	14.91	15.40	0.0001

LSD ( $p < 0.05$ ) = 0.78    CV% = 6.2

#### 4.2.2 NUMBER OF COBS PER PLANT

For both sowings, hybrid was the only factor to significantly affect cobs/plant (Table 7 and 9)

#### OCTOBER SOWING

**Table-7. Statistical significance of treatment effects for the October sowing for number of cobs per plant.**

Source	DF	F-Value	Pr>F
Replication	2	2.89	ns
Nitrogen	2	0.14	ns
Rep x Nitrogen	4	1.56	ns
Hybrid	2	4.22	*
Nitro x Hybrid	4	1.89	ns

#### **HYBRID EFFECT**

Hybrid P3787 had a significantly higher cob number /plant than hybrid A82-8XNZS84 but did not differ from hybrid P3751. However cob number did not differ significantly between hybrids P3751 and A82-8XNZS84 (Table 8).

**Table-8. The effect of hybrids on number of cobs per plant**

	Hybrid			Significance
	P3787	P3751	A82-8xNZ84	
Cob/plant	1.08	1.04	1.02	0.0409

LSD ( $p < 0.05$ ) = 0.04    CV% = 3.7

### NOVEMBER SOWING

**Table-9. Statistical significance of treatment effects for the November sowing for number of cobs per plant.**

Source	DF	F-Value	Pr>F
Replication	2	1.33	ns
Nitrogen	2	0.14	ns
Rep x Nitrogen	4	2.33	ns
Hybrid	2	4.33	**
Nitro x Hybrid	4	0.83	ns

### **HYBRID EFFECT**

Hybrid P3787 had a significantly higher cob number /plant than hybrid A82-8XNZS84 but did not differ from hybrid P3751. However cob number did not differ significantly between hybrids P3751 and A82-8xNZ84 (Table 10).

**Table-10. The effects of hybrids on number of cobs per plant**

	Hybrid			Significance
	P3787	P3751	A82-8xNZ84	
Cob/plant	1.04	1.01	1.00	0.0409

LSD ( $p < 0.05$ ) = 0.03    CV% = 3.9

#### 4.2.3 NUMBER OF GRAINS PER COB

##### OCTOBER SOWING

Hybrid was only the factor which significantly affected the number of grains per cob (Table 11)

**Table-11. Statistical significance of treatment effects for the October sowing for number of grains per cob.**

Source	DF	F-Value	Pr>F
Replication	2	1.23	ns
Nitrogen	2	0.81	ns
Rep x Nitrogen	4	0.98	ns
Hybrid	2	22.42	**
Nitro x Hybrid	4	0.26	ns

##### **HYBRID EFFECT**

There were significant difference in grains per cob among the hybrids (Table 12), with hybrid A82-8xNZ84 producing the highest number of grains, P3787 the lowest number and P3751 being intermediate in grains per cob.

**Table-12. The effects of hybrids on grain number per cob**

	Hybrid			Significance
	P3787	P3751	A82-8xNZ84	
Grains/cob	462.62	497.02	516.64	0.0001

LSD ( $p < 0.05$ ) = 16.12    CV% = 11.1

### NOVEMBER SOWING

Both nitrogen and hybrid significantly affected grains per cob (Table 13). There was no interaction between nitrogen and hybrid.

**Table-13. Statistical significance of treatment effects for the November sowing for number of grains per cob.**

Source	DF	F-Value	Pr>F
Replication	2	0.90	ns
Nitrogen	2	3.70	*
Rep x Nitrogen	4	1.12	ns
Hybrid	2	12.05	**
Nitro x Hybrid	4	0.48	ns

### **NITROGEN EFFECT**

At 500kg urea/ha the number of grains per cob was reduced significantly from the 250kg urea/ha treatment, but it was not significantly different from the no nitrogen treatment (Table 14).

**Table-14. The effects of nitrogen fertilizer on grain number per cob**

	Urea (kg/ha)			Significance
	0kg	250kg	500kg	
grains/cob	499.1	516.4	487.2	0.030

LSD ( $p < 0.05$ ) = 21.59    CV% = 13.6

### HYBRID EFFECT

There were significant differences in grains per cob between hybrid P3787 and the other two hybrids (Table 15). Hybrid A82-8xNZ84 produced the highest number of grains, P3787 the lowest number but there were no significant difference between hybrids P3751 and A82-8xNZ84 in number of grains per cob.

**Table-15. The effects of hybrids on grain number per cob**

	Hybrid			Significance
	P3787	P3751	A82-8xNZ84	
Grains/cob	472.6	501.5	519.6	0.0001

LSD ( $p < 0.05$ ) = 20.07    CV% = 12.9

### 4.2.4 100-GRAIN WEIGHT

#### OCTOBER SOWING

Both nitrogen and hybrid significantly affected grain yield (Table 16). There was no interaction between nitrogen and hybrid.

**Table-16. Statistical significance of treatment effects for the October sowing for 100-grain weight.**

Source	DF	F-Value	Pr>F
Replication	2	0.79	ns
Nitrogen	2	10.80	*
Rep x Nitrogen	4	0.70	ns
Hybrid	2	13.37	**
Nitro x Hybrid	4	0.59	ns

### NITROGEN EFFECT

An application of 250kg and 500kg urea/ha increased 100-grain weight significantly from the no nitrogen treatment (Table 17). While the application of 500kg urea/ha resulted in declining grain weight from that recorded at 250kg/ha, the difference between the 250kg and 500kg urea/ha treatments was not significant.

**Table-17. The effects of nitrogen fertilizer on 100-grain weight**

	Urea (kg/ha)			Significance
	0kg	250kg	500kg	
gm/100	29.70	32.66	31.75	0.0171

LSD ( $p < 0.05$ ) = 1.63    CV% = 5.4

### HYBRID EFFECT

As shown in the Table 18 the 100-grain weight of the three hybrids all differed significantly, with hybrid A82-8xNZ84 producing the highest weight, P-3787 the



lowest and P-3751 being intermediate in 100-grain weight.

**Table-18. The effects of hybrids on 100-grain weight**

	Hybrid			Significance
	P3787	P3751	A82-8xNZ84	
gm/100	29.37	31.34	33.41	0.0010

LSD ( $p < 0.05$ ) = 1.70    CV% = 5.3

### NOVEMBER SOWING

Both nitrogen and hybrid significantly affected 100-grain weight (Table 19). There was no interaction between nitrogen and hybrid.

**Table-19. Statistical significance of treatment effects for the November sowing for 100-grain weight.**

Source	DF	F-Value	Pr>F
Replication	2	0.74	ns
Nitrogen	2	15.99	**
Rep x Nitrogen	4	0.24	ns
Hybrid	2	9.09	**
Nitro x Hybrid	4	0.70	ns

### **NITROGEN EFFECT**

The application of 250 and 500 kg/ha of urea increased 100-grain weight significantly (Table 20). While the greatest response came from the 250 kg urea/ha application, it did not differ from that at the 500kg urea/ha rate.

**Table-20. The effects of nitrogen fertilizer on 100-grain weight**

	Urea (kg/ha)			Significance
	0kg	250kg	500kg	
gm/100	30.09	32.54	31.87	0.0065

LSD ( $p < 0.05$ ) = 1.29

CV% = 6.1

**HYBRID EFFECT**

Hybrid A82-8xNZ84 producing the highest 100-grain weight and this was significant when compared with the other two hybrids (Table 21). However there was no significant difference between hybrids P3787 and P3751 in 100-grain weight.

**Table-21. The effects of hybrids on 100-grain weight**

	Hybrid			Significance
	P3787	P3751	A82-8xNZ84	
gm/100	29.63	31.49	33.90	0.0036

LSD ( $p < 0.05$ ) = 2.08 CV% = 6.7**4.3 GRAIN PROTEIN**

The grain protein content data presented in the following Tables refer only to those effects that are statistically significant. The statistical significance of treatment effects for the October and the November sowing is presented separately.

## OCTOBER SOWING

Both nitrogen and hybrid significantly affected protein percentage in the grain (Table 22). There was no interaction between nitrogen and hybrid.

**Table-22. Statistical significance of treatment effects for the October sowing for grain protein percentage.**

Source	DF	F-Value	Pr>F
Replication	2	0.46	ns
Nitrogen	2	33.76	**
Rep x Nitro	4	1.20	ns
Hybrid	2	5.05	*
Nitro x Hybrid	4	1.26	ns

## **NITROGEN EFFECT**

The application of nitrogen fertilizer increased grain protein percentage significantly (Table 23). Treatments receiving 500 kg urea/ha synthesized the highest quantity of protein while treatments receiving no applied nitrogen synthesized the lowest and treatments receiving 250kg urea/ha synthesized an intermediate quantity in the grain.

**Table-23. The effects of nitrogen fertilizer on grain protein percentage**

	Urea (kg/ha)			Significance
	0	250	500	
Protein(%)	8.81	9.41	10.13	0.0001

LSD (p<0.05)= 0.39    CV%= 3.8

**HYBRID EFFECT**

There were significant differences in grain protein content among the hybrids, with hybrid P3751 synthesizing the lowest quantity of protein compared with the other two hybrids (Table 24). However, there was no significant difference in protein content between hybrid P3787 and A82-8xNZ84.

**Table-24 The effects of hybrids on grain protein percentage**

	Hybrid			Significance
	P3751	P3787	A82-8xNZ84	
Protein(%)	9.18	9.59	9.58	0.0001

LSD ( $p < 0.05$ ) = 0.31    CV% = 4.2

**NOVEMBER SOWING**

As for the November sowing, both nitrogen and hybrid affected protein percentage in the grain, but there was no significant interaction (Table 25).

**Table-25. Statistical significance of treatment effects for the November sowing for grain protein percentage.**

Source	DF	F-Value	Pr>F
Replication	2	0.42	ns
Nitrogen	2	80.50	**
Rep x Nitro	4	0.32	ns
Hybrid	2	3.06	*
Nitro x Hybrid	4	0.47	ns

## NITROGEN EFFECT

The application of nitrogen fertilizer increased grain protein percentage significantly (Table 26). Treatments receiving 500 kg urea/ha synthesized the highest quantity of protein while treatments receiving no applied nitrogen synthesized the lowest and treatments receiving 250kg urea/ha synthesized an intermediate quantity in the grain.

**Table-26. The effects of nitrogen fertilizer on grain protein percentage**

	Urea (kg/ha)			significance
	0	250	500	
Protein(%)	8.72	9.54	10.13	0.0001

LSD ( $p < 0.05$ ) = 0.27    CV% = 5.06

## HYBRID EFFECT

Hybrid A82-8xNZ84 synthesized the highest quantity and hybrid P3751 synthesized the lowest quantity of protein and hybrid P3787 synthesized an intermediate amount. However there was no significant difference between hybrids P3787 and P3751 (Table 27).

**Table-27 The effects of hybrids on grain protein percentage**

	Hybrid			Significance
	P3751	P3787	A82-8xNZ84	
Protein(%)	9.19	9.57	9.65	0.0001

LSD ( $p < 0.05$ ) = 0.41    CV% = 5.1

## 4.4 GRAIN QUALITY

In section 4.3 the effects of different treatments eg. nitrogen and hybrids on grain protein content were described. In this section data for three different parameters from the Stenvert Hardness Test (SHT) (i.e. grinding resistance time, required energy and milling duration time), bulk density and grain moisture are presented. The data presented in the following tables refer only to those effects that are statistically significant. The statistical significance of treatment effects are presented separately for both the October and the November sowing.

### 4.4.1 STENVERT HARDNESS TEST

#### 4.4.1.1 GRINDING RESISTANCE TIME

##### OCTOBER SOWING

Both nitrogen and hybrid significantly affected grinding resistance time of the grain (Table 28). There was no interaction between nitrogen and hybrid.

**Table-28. Statistical significance of treatment effects for the October sowing for grinding resistance time(sec).**

Source	DF	F-Value	Pr>F
Replication	2	1.09	ns
Nitrogen	2	103.97	**
Rep x Nitro	4	0.38	ns
Hybrid	2	13.63	**
Nitro x Hybrid	4	1.04	ns

##### **NITROGEN EFFECT**

Grinding resistance time of the grain increased progressively and significantly with

increasing levels of nitrogen application from 0 to 500kg/ha (Table 29).

**Table-29. The effects of nitrogen fertilizer on grinding resistance time (sec)**

	Nitrogen as Urea			Significance
	0	250	500	
Time(sec)	13.60	14.88	16.06	0.0001

LSD ( $p < 0.05$ ) = 0.47 CV% = 4.0

### **HYBRID EFFECT**

The grinding resistance time of the hybrids differed significantly, with hybrid A82-8xNZ84 showing the greatest resistance, P3751 the lowest resistance and P3787 being intermediate in resistance time (Table 30).

**Table-30. The effects of hybrids on grinding resistance time (sec)**

	Hybrid			Significance
	P-3751	P-3787	A82-8xNZ84	
Time(sec)	14.13	14.82	16.06	0.0001

LSD ( $p < 0.05$ ) = 0.60 CV% = 4.5

### **NOVEMBER SOWING**

Both nitrogen and hybrid significantly affected grinding resistance time of the grain (Table 31). There was also an interaction between nitrogen and hybrids.

**Table-31. Statistical significance of treatment effects for the November sowing for grinding resistance time (sec).**

Source	DF	F-Value	Pr>F
Replication	2	1.37	ns
Nitrogen	2	74.68	**
Rep x Nitro	4	0.62	ns
Hybrid	2	24.55	**
Nitro x Hybrid	4	4.05	*

### NITROGEN EFFECT

Grinding resistance time of the grain increased progressively and significantly with increasing levels of nitrogen application from 0 to 500kg urea/ha (Table 32).

**Table-32. The effects of nitrogen fertilizer on grinding resistance time (sec)**

		Urea (kg/ha)			
		0	250	500	Significance
Time(sec)	13.60	14.92	15.98	0.0001	

LSD ( $p < 0.05$ ) = 0.54    CV% = 3.6

### HYBRID EFFECT

The grinding resistance time of the hybrids differed significantly, with hybrid A82-8xNZ84 showing the greatest resistance and P3751 the lowest resistance time.



However there was no significant difference between hybrids P3787 and A82-8xNZ84 (Table 33).

**Table-33. The effects of hybrids on grinding resistance time (sec)**

	Hybrid			Significance
	P-3751	P-3787	A82-8xNZ84	
Time(sec)	13.84	15.20	15.47	0.0001

LSD ( $p < 0.05$ ) = 0.54, CV% = 6.3

## INTERACTION EFFECTS

### NITROGEN X HYBRID

Table 34 shows an interaction between nitrogen treatments and hybrids for grinding resistance time. Hybrid P3787 showed a significantly higher grinding resistance time in the higher nitrogen treatments than hybrid P3751, although those hybrids did not differ significantly for the no nitrogen treatment. However, hybrid P3787 did not differ from hybrid A82-8xNZ84.

**Table-34. The interactive effects of nitrogen and hybrid on grinding resistance time (sec)**

Urea(kg/ha)	Hybrid			Significance
	P3751	P3787	A82-8xNZ84	
0	13.37	13.50	13.93	0.0259
250	13.70	15.27	15.80	
500	14.47	16.83	16.67	

LSD ( $p < 0.05$ ) = 0.94 CV% = 7.2

#### 4.4.1.2 ENERGY REQUIRED FOR GRINDING

##### OCTOBER SOWING

Both nitrogen and hybrid significantly affected energy required for grinding of the grains (Table 35). There was no interaction between nitrogen and hybrid.

**Table-35. Statistical significance of treatment effects for the October sowing for energy required for grinding (J).**

Source	DF	F-Value	Pr>F
Replication	2	1.92	ns
Nitrogen	2	201.6	**
Rep x Nitro	4	0.15	ns
Hybrid	2	4.35	*
Nitro x Hybrid	4	0.57	ns

##### NITROGEN EFFECT

The energy required for grinding increased significantly with increasing urea application rates (Table 36).

**Table-36. The effects of nitrogen fertilizer on energy required for grinding (J).**

	Urea (kg/ha)			
	0	250	500	Significance
Energy(J)	5935.6	6412.3	6668.8	0.0001

LSD ( $p < 0.05$ ) = 102.9    CV% = 2.4

## HYBRID EFFECT

Different hybrids required significantly different amounts of energy to grind the grain (Table 37), with hybrid P3787 requiring the highest amount and P3751 requiring the least. However there was no significant difference between P3787 and A82-8xNZ84.

**Table-37. The effects of hybrids on energy required for grinding (J)**

	Hybrid			Significance
	P-3751	P-3787	A82-8xNZ84	
Energy(J)	6178.0	6446.8	6391.3	0.0001

LSD ( $p < 0.05$ ) = 209.52    CV% = 2.4

## NOVEMBER SOWING

Both nitrogen and hybrid significantly affected energy required for grinding of the grains (Table 38). There was no interaction between nitrogen and hybrid.

**Table-38. Statistical significance of treatment effects for the November sowing for energy required for grinding (J).**

Source	DF	F-Value	Pr>F
Replication	2	1.23	ns
Nitrogen	2	14.73	*
Rep x Nitro	4	0.91	ns
Hybrid	2	6.85	*
Nitro x Hybrid	4	0.72	ns

## NITROGEN EFFECT

The grain produced in the highest nitrogen treatment required the highest energy to

grind when compared with grain in the no nitrogen treatment (Table 39). However, there was no significant difference between grain produced from the intermediate and the highest levels of nitrogen application in this regard.

**Table-39. The effects of nitrogen fertilizer on energy required for grinding (J)**

	Urea (kg/ha)			significance
	0	250	500	
Energy(J)	6087.4	6605.8	6752.7	0.0143

LSD ( $p < 0.05$ ) = 357.47 CV% = 4.4

#### HYBRID EFFECT

Different hybrids required significantly different amounts of energy to grind the grain (Table 40), with hybrid P3787 requiring the highest amount and P3751 requiring the least. However there was no significant difference between P3787 and A82-8xNZ84.

**Table-40. The effects of hybrids on energy required for grinding (J).**

	Hybrid			Significance
	P-3751	P-3787	A82-8xNZ84	
Energy(J)	6205.9	6545.6	6694.4	0.0143

LSD ( $p < 0.05$ ) = 294.73 CV% = 3.9

#### 4.4.1.3 MILLING DURATION

#### OCTOBER SOWING

Both nitrogen and hybrid significantly affected milling duration time of the grains (Table 41). There was no interaction between nitrogen and hybrid.

**Table-41. Statistical significance of treatment effects for the October sowing for milling duration time (sec).**

Source	DF	F-Value	Pr>F
Replication	2	0.50	ns
Nitrogen	2	159.10	**
Rep x Nitro	4	0.55	ns
Hybrid	2	6.37	*
Nitro x Hybrid	4	0.84	ns

### NITROGEN EFFECT

The time taken to mill increased significantly with increasing levels of nitrogen application (Table 42).

**Table-42. The effects of nitrogen fertilizer on milling duration time (sec).**

	Urea (kg/ha)			significance
	0	250	500	
Time(sec)	33.78	36.22	37.08	0.0001

LSD ( $p < 0.05$ ) = 0.53    CV% = 2.1

### HYBRID EFFECT

As shown in Table 43 there was a significant difference in milling duration, with hybrid P3751 taking the shortest time compared with the other two hybrids.

However, there was no significant difference in milling duration time between hybrid P3787 and A82-8xNZ84.

**Table-43. The effects of hybrids on milling duration time (sec).**

	Hybrid			Significance
	P-3751	P-3787	A82-8xNZ84	
Time(sec)	35.1	36.08	35.90	0.0053

LSD ( $p < 0.05$ ) = 0.64    CV% = 1.6

### NOVEMBER SOWING

Both nitrogen and hybrid significantly affected milling duration time of the grains (Table 44). There was no interaction between nitrogen and hybrid.

**Table-44. Statistical significance of treatment effects for the November sowing for milling duration time(sec).**

Source	DF	F-Value	Pr>F
Replication	2	0.30	ns
Nitrogen	2	60.75	**
Rep x Nitro	4	2.19	ns
Hybrid	2	13.69	**
Nitro x Hybrid	4	2.31	ns

### **NITROGEN EFFECT**

The time taken to mill increased significantly with increasing levels of nitrogen

application (Table 45).

**Table-45. The effects of nitrogen fertilizer on milling duration time (sec).**

	Urea (kg/ha)			Significance
	0	250	500	
Time(sec)	33.81	36.05	37.59	0.0001

LSD(p<0.05)= 0.96    CV%= 1.4

### HYBRID EFFECT

As shown in Table 46 there was a significant difference in milling duration with hybrid P3751 taking the shortest time compared with other two hybrids. However, there was no significant difference in milling duration time between hybrid P3787 and A82-8XNZ84.

**Table-46. The effects of hybrids on milling duration time (sec).**

	Hybrid			Significance
	P-3751	P-3787	A82-8xNZ84	
Time(sec)	35.13	36.05	36.28	0.0008

LSD(p<0.05)= 0.51    CV%= 2.0

#### 4.4.1.4 BULK DENSITY

### OCTOBER SOWING

Both nitrogen and hybrid significantly affected grain bulk density (Table 44). There

was no interaction between nitrogen and hybrid.

**Table-47. Statistical significance of treatment effects for the October sowing for grain bulk density (kg/hl).**

Source	DF	F-Value	Pr>F
Replication	2	0.44	ns
Nitrogen	2	8.21	*
Rep x Nitro	4	0.90	ns
Hybrid	2	36.04	**
Nitro x Hybrid	4	0.26	ns

#### NITROGEN EFFECT

The bulk density of the grain increased with each addition of nitrogen fertilizer (Table 48). The highest bulk density were recorded at the highest level of nitrogen treatment and the lowest was recorded at the no nitrogen treatment. However there was no significant difference between the highest and intermediate level of nitrogen treatments.

**Table-48. The effects of nitrogen fertilizer on grain bulk density (kg/hl)**

	Urea (kg/ha)			Significance
	0kg	250kg	500kg	
BD(Kg/hl)	77.16	77.87	78.69	0.0384

LSD ( $p < 0.05$ ) = 1.04    CV% = 1.1

#### HYBRID EFFECT

The grain bulk density of the hybrids differed significantly, with hybrid A82-



8XNZS84 producing the highest grain bulk density, P3751 the lowest and P3787 being intermediate in bulk density (Table 49).

**Table-49. The effects of hybrids on grain bulk density (kg/hl)**

	Hybrid			Significance
	P-3751	P-3787	A82-8xNZ84	
BD(kg/hl)	76.19	77.99	79.54	0.0001

LSD (p<0.05)= 0.86 CV%= 1.2

### NOVEMBER SOWING

Both nitrogen and hybrid significantly affected grain bulk density (Table 50). There was no interaction between nitrogen and hybrid.

**Table-50. Statistical significance of treatment effects for the November sowing for grain bulk density (kg/hl).**

Source	DF	F-Value	Pr>F
Replication	2	0.71	ns
Nitrogen	2	14.66	*
Rep x Nitro	4	0.37	ns
Hybrid	2	48.19	**
Nitro x Hybrid	4	0.56	ns

### **NITROGEN EFFECT**

The bulk density of the grain increased with each addition of nitrogen fertilizer (Table 51). The highest bulk density was recorded at the highest level of nitrogen

treatment and the lowest was recorded at the no nitrogen treatment. However there was no significant difference between the highest and intermediate level of nitrogen treatments.

**Table-51. The effects of nitrogen fertilizer on grain bulk density (kg/hl)**

	Urea (kg/ha)			significance
	0kg	250kg	500kg	
MD(sec)	77.12	77.77	78.27	0.0144

LSD( $p < 0.05$ ) = 0.59 CV% = 0.96

#### **HYBRID EFFECT**

The grain bulk density of the hybrids differed significantly, with hybrid A82-8xNZ84 producing the highest grain bulk density, P3751 the lowest and P3787 being intermediate in bulk density (Table 52).

**Table-52. The effects of hybrids on grain bulk density (kg/hl)**

	Hybrid			Significance
	P-3751	P-3787	A82-8xNZ84	
BD(kg/hl)	75.94	77.88	79.33	0.0001

LSD ( $p < 0.05$ ) = 0.75 CV% = 1.1

#### **4.4.4.4 GRAIN MOISTURE**

##### **OCTOBER SOWING**

Only hybrid significantly affected grain moisture content (Table 53). There was no

interaction between nitrogen and hybrid.

**Table-53. Statistical significance of treatment effects for the October sowing for grain moisture content (%).**

Source	DF	F-Value	Pr>F
Replication	2	0.46	ns
Nitrogen	2	1.42	ns
Rep x Nitro	4	0.78	ns
Hybrid	2	12.87	**
Nitro x Hybrid	4	2.49	ns

### HYBRID EFFECT

The grain moisture percentage of the hybrids differed significantly (Table 54), with hybrid A82-8xNZ84 containing the highest grain moisture, P3751 intermediate and P3787 the lowest grain moisture content.

**Table-54. The effects of hybrids on grain moisture content (%)**

	Hybrid			Significance
	P3751	P3787	A82-8xNZ84	
MC(%)	12.56	12.13	12.72	0.0010

LSD (p<0.05)= 0.26 CV%= 2.1

### NOVEMBER SOWING

Both nitrogen and hybrid significantly affected grain moisture content (Table 55). There was also an interaction between nitrogen and hybrid.

**Table-55. Statistical significance of treatment effects for the November sowing for grain moisture content (%).**

Source	DF	F-Value	Pr>F
Replication	2	1.03	ns
Nitrogen	2	14.96	*
Rep x Nitro	4	0.62	ns
Hybrid	2	23.57	**
Nitro x Hybrid	4	8.32	**

### NITROGEN EFFECT

Grains sample taken from the 500kg urea/ha plots retained the highest amount of grain moisture which was significantly greater than for the other two nitrogen treatments (Table 56). However there was no significant difference between grains taken from the 250kg and no nitrogen treatments.

**Table-56. The effects of nitrogen fertilizer on the grain moisture content (%)**

	Urea (kg/ha)			Significance
	0kg	250kg	500kg	
MC(%)	12.58	12.53	12.84	0.0036

LSD(p<0.05)= 0.17    CV%= 1.3

### HYBRID EFFECT

The grain moisture percentages of the hybrids were all significantly different (Table 57), with hybrid A82-8xNZ84 containing the highest grain moisture, P3751 intermediate and P3787 the lowest grain moisture content.

**Table-57. The effects of hybrids on grain moisture content (%)**

	Hybrid			Significance
	P3751	P3787	A82-8xNZ84	
MC(%)	12.62	12.40	12.93	0.0001

LSD ( $p < 0.05$ ) = 0.17    CV% = 1.3

## INTERACTION EFFECTS

### NITROGEN X HYBRID

Table 58 shows an interesting interaction between nitrogen treatments and hybrids for grain moisture content. Hybrid P3787 showed a significantly lower grain moisture content than the other two hybrids in treatments receiving no nitrogen and 250 kg urea/ha. However, at the highest levels of nitrogen application, hybrid A82-8XNZS84 maintained a significantly high grain moisture content, while hybrids P3787 and P3751 were not significantly different.

**Table-58. The interactive effects of nitrogen and hybrids on grain moisture content (%)**

Urea(kg/ha)	Hybrid			Significance
	P3751	P3787	A82-8xNZ84	
0	12.53	12.20	13.00	0.0019
250	12.80	12.20	12.60	
500	12.53	12.80	13.20	

LSD ( $p < 0.05$ ) = 0.29    CV% = 3.78

#### 4.5 RELATIONSHIP BETWEEN GRAIN QUALITY PARAMETERS

##### OCTOBER SOWING

The association between grain protein and Stenvert hardness test parameters was analyzed by using simple correlation, and correlation coefficient are presented in Table 59. From the data it is evident that all measured variables show at least one correlation coefficient greater than 0.5. The correlations between grain protein and SHT parameters were strong e.g., with grinding resistance time (0.80), required energy (0.74) and milling duration time (0.65). Grain protein was also correlated with bulk density of the grain. Grain moisture was negatively but not significantly correlated with required energy for grinding and also with the milling duration time.

**Table:59** Correlation coefficient of grain protein, SHT parameters, bulk density and grain moisture content in the October sowing

	Protein	Resist	Energy	Mill	Bulk	Moist
Protein	1.000					
Resist	0.798**	1.000				
Energy	0.742**	0.868**	1.000			
Mill	0.772**	0.848**	0.864**	1.000		
bulk	0.646*	0.783**	0.597*	0.579**	1.000	
moist	0.166ns	0.183ns	-0.355ns	-0.182ns	0.057	1.000

**Note:**

Protein= Grain protein percentage

Resist= Resistance time for grinding

Energy= Required energy for grinding

Mill= Milling duration time for energy

Bulk= Bulk density of the grain

Moist= Moisture content of the grain

### NOVEMBER SOWING

The association between grain protein and Stenvert hardness test parameters, bulk density and grain moisture content was analyzed by using a simple correlation, and correlation coefficient are presented in Table 60. From the data it is evident that all measured variables show at least one correlation coefficient greater than 0.5. The correlations between protein and SHT parameters were strong e.g., with grinding resistance time (0.82), required energy (0.75) and milling duration time (0.86). Grain protein was also correlated with bulk density of the grain. Grain moisture was only correlated significantly with the bulk density.

**Table:60**      **Correlation coefficient of grain protein, SHT parameters, bulk density and grain moisture content in the November sowing**

	Protein	Resist	Energy	Mill	Bulk	Moist
Protein	1.000					
Resist	0.819**	1.000				
Energy	0.750**	0.868**	1.000			
Mill	0.865**	0.868**	0.784**	1.000		
bulk	0.443*	0.739**	0.619*	0.579**	1.000	
moist	0.076ns	0.329ns	0.146ns	0.262ns	0.431*	1.000

**Note:**

Protein= Grain protein percentage

Resist= Resistance time for grinding

Energy= Required energy for grinding

Mill= Milling duration time for energy

Bulk= Bulk density of the grain

Moist= Moisture content of the grain

## CHAPTER V

### DISCUSSION AND CONCLUSIONS

#### DISCUSSION

##### 5.1 PLANT GROWTH AND DEVELOPMENT

Neither percent germination in the laboratory or field emergence at both sowings differed significantly among the hybrids, suggesting that the seed was of similar quality. However, the time required for seedling emergence in the October sowing was greater than in the November sowing. Similar results were also reported by Groot (1976), Warrington and Kanemasu (1983b) and Hardacre and Eagles (1989). This was because of the day and night temperature differences i.e., cooler diurnal temperatures in October (14/6° C) compared with November (16/10°C). This demonstrates the importance of temperature for seedling emergence. At the time of sowing, low soil and air temperature can delay and reduce seedling emergence, and as a result seedlings are more prone to damage by soil and seed borne fungi (Groot, 1976; Krager, 1989). In New Zealand conditions soil temperatures of approximately 6 to 8° C and air temperatures of approximately 15° C are considered to be the minimum for maize growth (Hardacre et al., 1993). This implies that warm temperatures allow germination/ emergence to start early and proceed faster, and indeed, optimum germination temperatures for maize are 20 - 25° C (ISTA, 1995).

In this study, A82-8xNZ84 produced a significantly greater number of leaves than P3751 and P3787 at both sowings. Differences in leaf number between A82-8xNZ84 and the other two hybrids were presumably due to the genetic traits and the response of the hybrids to the environment. Under field conditions all hybrids experienced similar environmental conditions, so differences in leaf number are mainly genetic attributes, and final leaf number depends on the number of leaves present in the seed embryo, the rate of leaf initiation at the apical meristem and the duration of the



vegetative phase (Hunter et.al., 1977; Warrington and Kanemasu, 1983a,b).

Leaf appearance rate was faster in the November sowing than in the October sowing in all hybrids, due to the higher temperature and longer photoperiod. Chase and Nanda (1967), Bonaparte (1975) and Warrington and Kanemassu (1983 b,c) all showed that leaf appearance rate increased with increasing photoperiod and mean daily temperatures from a range of 12-16h and 16-26°C respectively. Differences in rates were for the time taken from seeding to tassel initiation by the hybrids. In the October sowing initial leaf appearance rate was slower due to lower temperature and shorter photoperiod than the November sowing.

In this work there was no effect of nitrogen on leaf number, and it is therefore suggested that the absence of a leaf number response to applied nitrogen was due to presumably high levels of previous soil nitrogen available from the three years of the previous white clover based pasture, and following a basal dressing of N:P:K (12:10:10). However, no soil nitrogen measurements were made.

Nitrogen requirements by maize plants differ between growing periods. In the lag phase (during seedling establishment) the requirement is less than in the linear phase (up to tasselling). In this phase nitrogen requirement is higher, because at this stage the number of leaf cells are increasing as a results of the higher cell division activity (Arnon, 1975). Leaf growth is retarded in nitrogen depleted plants because the reduction of chlorophyll content results in a reduction of photosynthate supply and therefore affects the growth of the young leaf (Dale, 1982). Leaf area and leaf dry weight probably could have been used to determine the effect of different nitrogen treatments, but these were not measured in this study. Eik and Hanway (1965) reported that nitrogen availability had no effect on leaf number or leaf appearance rate. This is in contrast to Bonaparte (1975) who obtained higher leaf number when nitrogen was applied to a lower fertility soil. In this study leaf numbers varied among

hybrids but not among the nitrogen treatments. This is in agreement with the work of Allen et al., (1973) who reported that the number of leaves of cereal crops may not be affected by nitrogen supply because it is largely determined by genotype.

The hybrids differed in time to 50% silking as P3787 flowered earlier than P3751 and A82-8xNZ84. However, the November sowing took fewer days to 50% silking than the October sowing. This was due to the higher temperature received during the growing period by the November sown plants. This is in agreement with Cal and Obendorf (1972), and Warrington and Kanemasu (1983a) who reported that an increase in temperature resulted in the acceleration of developmental rates, as evidenced by substantial reduction in days to silking.

In this study hybrid P3787 formed the 'black layer' earlier than the other two hybrids, P3751 and A82-8xNZ84. Formation of the black layer is an indicator of physiological maturity (Daynard and Duncan, 1969) when grains on the ear have achieved maximum dry matter accumulation. Grain moisture content at physiological maturity (mass maturity) of P3787 was lower than P3751 and A82-8xNS84. Grain moisture at black layer development or physiological maturity varies with hybrid (Shaw,1977). After attainment of mass maturity grains begin to dry. Thus the drying began earlier in P3787, and this hybrid was significantly lower in seed moisture content at harvest for both the October and the November sowings. This was due to the lower leaf number and responses of ontogeny to the environment.

Before physiological maturity (mass maturity) grain moisture content is primarily a physiological process, but after the attainment of the maturity it depends not only the hybrids' genetic trait but also on environmental factors such as air temperatures and relative humidity. Schmidt and Hallauer (1966), Larson and Hanway (1977), and Newton and Eagles (1991) who considered that the rate of drying of the kernel in the field depended on ear drying rate after physiological maturity of the hybrid, the

cultivar characteristics and also on environmental conditions. In this study the grain moisture dry down was faster in P3787 than P3751 and A82-8xNZ84 after formation of the black layer, presumably because of genotypic and/or environmental effects. However, nitrogen treatments had no effects in this regard.

## 5.2. GRAIN YIELD AND YIELD COMPONENTS

Yield response to the applied nitrogen was similar at both sowings. The addition of 250kg urea/ha increased yield about 12% in the October sowing and about 10% in the November sowing over the control treatment. Maize is a warm weather crop whose growth and developmental processes are strongly influenced by environmental factors (Shaw, 1977). Yields are associated with day length and temperature during the growing season. Early sowing of maize allows grain formation in mid summer, when day length are greater and more radiant energy is available for photosynthates than for late sowings. Highest maize yields have been recorded with day time maximum temperatures of 24-30° C with an optimum rainfall at all times (Aldrich et al., 1975). However in this study the yield difference between sowing times was not significant, probably because the sowing time difference was not critical for day length and temperature.

The addition of a further 250kg urea/ha (i.e a total of 500kg urea/ha) caused a depression in yield of about 5% at both sowings over the highest yield recorded at 250kg/ha. However, this reduction was not statistically significant. Yield depression at high levels of applied nitrogen is well known ( Olson, R.A. 1984; Martin et al., 1989). Yield increases and decreases following the application of fertilizer nitrogen depend on the initial soil fertility level. Benzian and Lane (1981) reported that if the soil nitrogen status is low, the initial increase in nitrogen supply will increase yield, while with higher rates of nitrogen, yields reach a plateau and may start to decline.

Hybrids showed different yield performance at both sowings, and it was clearly demonstrated that A82-8xNZ84 was superior for grain production compared to P3787, but did not differ significantly from P3751. Hybrid P3787 formed the 'black layer' which cut off the movement of solutes into grains earlier, than A82-8xNZ84 and P3751 which formed the 'black layer' two weeks later and thereby matured later. It is expected that late maturing hybrids will give higher yield than early maturing hybrids, provided the climatic conditions are favourable (Cumberland et al., 1970; Hardacre et al., 1992). Both hybrids (i.e. A82-8xNZ84 and P3751) were therefore later maturing than P3787, and thereby produced higher grain yields.

Yield components were also influenced by applied urea treatments in this study. 100-grain weight and grains per cob were higher when nitrogen was applied, but cobs per plant were not affected. From the onset of flowering to early grain formation, the nitrogen requirements of maize plants reach a peak (Arnon, 1975) and ears become the strongest sink. At this stage nitrogen unavailability can cause a yield limitation. Increased nitrogen availability usually increases the leaf area and the potential photosynthetic capacity of the crop increases (Pearman et al., 1977). The overall effects of nitrogen are therefore to increase the source capacity of the plant and provide more assimilate for the sink. Thereby, yield components (100-grain weight and grains per cob) were increased over from the no nitrogen treatment in this study. This is in agreement with the work of Eck (1982) who reported that the major effect of nitrogen in increasing yields was through increasing seed numbers and seed weight.

In both sowings cobs/plant were higher in hybrid P3787 than the other two hybrids (i.e. P3751 and A82-8xNZ84) and this was due to a higher frequency of two eared plants. However, P3787 did not produced higher yields when compared with P3751 and A82-8xNZ84, This was due the lower number of grains in the cob and 100-grain weight of P3787 than other hybrids. As well, P3787 is a smaller plant and may have required a greater plant density to give higher yield. For hybrid P3787 the

recommended plant density per hectare is 100,000 (Pioneer Brand Products, 1993-94).

### 5.3 GRAIN PROTEIN

Grain protein percentage increased by about 15% in the October and about 16% in the November sowing at the highest urea treatment (500 kg/ha) over the no urea treatment. The total nitrogen absorbed by the crop has a major influence on the protein content of grain and increasing nitrogen supply generally increases grain protein (Deckard et al., 1984). In this study protein percentage increased progressively with the increase in rate of urea fertilizer. Maximum protein yield requires significantly higher nitrogen rate than is needed for maximum grain yield. This finding is well established, and similar results have been reported by many workers (Pierre et al., 1974; Olson et al., 1976; Tsai et al., 1980, 1983; Deckard et al., 1984; Sander et al., 1987).

In both sowings P3751 contained a lower amount of protein compared to P3787 and A82-8xNZ84. On the other hand P3787 produced a lower grain yield than P3751 and A82-8xNZ84. This might be due to the different genetic potential of the hybrids for uptake capacity and metabolism. Hybrids A82-8xNZ84 and P3787 might have had a higher capacity to uptake or translocate nitrogen, and thereby synthesized higher amounts of protein than hybrid P3751, although this was not determined. Developing grains are strong sinks for the nitrogen compounds used to manufacture protein. So, differences in grain protein and yield responses to available soil nitrogen may be influenced by genetic differences in the capacity of genotypes to take up nitrogen from the soil and translocate it to the sink grain (Pollner et al., 1979).

#### 5.4 GRAIN HARDNESS

Grain hardness is an index of the relative amount of hard to soft endosperm. Grain hardness is related to the protein content and grain physical properties including kernel density and the ratio of hard and soft endosperm (Watson, 1987). In this study kernel hardness was measured by a Stenvert Hardness Tester (SHT), because SHT has been reported as a useful measurement of kernel hardness (Pomeranz et al., 1985, 1986a, 1986b; Li et al., 1995).

Grains grown under applied nitrogen treatments and therefore with higher protein content showed greater hardness than grains from the control treatment in both sowings. Grinding resistance time, required energy and milling duration times were higher with these grains. Similar results were also reported by Li et al., (1995). These authors found that total energy, resistance time, bulk density and hard to soft endosperm were all highly correlated and that the Stenvert hardness test is a quick and simple method of comparing the endosperm hardness.

When nitrogen fertilizer was applied, grain contained a higher amount of protein which changed the endosperm texture. Changes in total protein content are primarily changes in endosperm protein content. As the hard endosperm ratio increases the amount of protein content increases because the hard endosperm has a thicker protein matrix than soft endosperm. This is also in agreement with the work of Dombink-Kurtzman and Wilson (1992) who reported that the protein concentration and distribution directly influence endosperm texture and physical properties.

Grinding resistance time, required energy for grinding and milling duration times were greater in A82-8xNZ84 and P3787, than P3751. This was presumably due to the presence of more hard endosperm in the grains of A82-8xNZ84 and P3787 and more soft endosperm in P3751. The grain protein content of the hybrids increased as

applied nitrogen increased, and as the protein content increases, the amount of hard endosperm increases (Hinton, 1953).

Another interesting finding was the protein and hardness relationship within genotypes. Generally, maize grain comprises 8.1-11.5% protein (Watson, 1987) and in this study P3751 contained a relatively similar amount of protein (9.18% and 9.19%) when compared with P3787 (9.59% and 9.54%) and A82-8xNZ84 (9.58% and 9.65%) in the October and the November sowing respectively, but P3751 did not show relatively similar hardness. This is probably due to the lack of a particular protein which governs the hardness. This factor was emphasized in work with Opaque and Quality Protein Maize (QPM) by Dombrink-Kurtzman and Willson (1992) who reported that hardness could be due, in the simplest case, to the presence of a specific 'hardness protein' or the absence of a 'softness protein'. While comparing the composition and distribution between QPM and normal maize they found that soft endosperm contained an increased percentage of gamma-zein but hard endosperm contained higher total amounts of alcohol soluble proteins and alpha-zein. These differences in protein composition may relate to composition of protein bodies and to texture of endosperm, although this was not measured in this study.

Grain bulk density (test weight) was increased with increasing nitrogen treatments at both sowings, and this was presumably due to the increase of hard endosperm in the grains. This is based on the fact that hard endosperm is very dense, whereas soft endosperm is full of microfissures or void spaces and therefore less dense. High soil nitrogen has a significant influence in producing a higher hard to soft endosperm ratio (Hamilton et al., 1951) and hard endosperm is denser than soft endosperm (Wichser, 1961). The bulk density (test weight) differed significantly among hybrids at both sowings and this difference is probably due to the different hard and soft endosperm ratios and/or genetic attribute of the hybrids.

An effort was made to equilibrate the moisture content to a similar level before the

estimation of grain hardness by the Stenvert Hardness Tester and bulk density, because moisture content has an inverse relationship on hardness measurements (Tran et al., 1981), and grain moisture and bulk density are linearly and highly correlated (Nelson, 1980). Equilibrium moisture content depends on sorption isotherms of grain and also on starch, protein, oil or fibre content of the grain (Johnson, 1988). In this study it is likely that the composition of the endosperm of the hybrids varied with different moisture equilibria. However, it was found that moisture content varied with sowings and nitrogen treatments as well as with hybrids. This was presumably due to the variation of the harvest moisture, or changes of the endosperm properties of the hybrids between nitrogen treatments at both sowings, or differences in the equilibrium moisture content of the hybrids. However in this study there was no inverse relationship when hardness was measured or no positive linear relationship with bulk density (test weight). This might be due to the fact that the differences found in grain moisture content during testing were probably too small to have a significant effect on hardness and bulk density determination.

## **5.5 THE RELATIONSHIP BETWEEN GRAIN QUALITY PARAMETERS**

Grain protein and measured hardness parameters (i.e. SHT parameters and bulk density) were highly correlated at both sowings. This indicates that grain protein has a significant effect on grain hardness. Resistance time, required energy for grinding, milling duration time and bulk density were all highly correlated and are therefore all likely to be good estimators of grain hardness. A similar finding was reported by Pomeranz et al., (1985) who found significant correlations between protein contents and hardness parameters while determining corn hardness by a Stenvert Hardness Tester.

However, in this study resistance time for grinding had a higher correlation coefficient with total energy, milling duration and bulk density at both sowings. This indicated that resistance time could predict the grain hardness more precisely than



other parameters. Similar results were also reported by Pomeranz et al., ( 1986a,b) who measured the resistance time of grains during milling but not the energy required for milling.

In this study grain yields achieved due to the application of urea were above the mean yield recorded for the Manawatu region (Hardacre et al., 1991). This confirms the high yield potential of these hybrids. The one month difference in sowing time did not affect the grain yield and quality. The application of nitrogen improved grain protein and hardness. Grain hardness was the highest for the highest urea treatment (i.e. 500kg/ha) although yield was slightly reduced. However, the response to nitrogen application for grain yield and quality was not compared in a cost/benefit analysis, because at present in the New Zealand maize trade market, no premium values are considered for the higher quality maize grain (Robert Coulson personal communication). This issue must be addressed, as growers want to get premium values for the quality grain produced, because they must pay higher costs, a situation which is very common for wheat growers in New Zealand (Millner, 1993).

In spite of the complexity of the yield and quality relationship, a good understanding of a few factors can serve as the basis for nitrogen fertilizer use. Target end uses of the grains should be one of the deciding factors as to how much nitrogen fertilizer should be used. As corn based snack foods are becoming increasingly popular, it is likely that the dry milling industry will be interested in getting more hard grain to produce a higher recovery of grits, and provided premiums are paid, ultimately farmers will benefit.

In New Zealand commercially available hybrids have been shown to be capable of high yield. However, the grain tends to be soft and not particularly suitable for dry milling for grits production (Hardacre, 1995). However it is concluded from this study that higher nitrogen treatment could improve both yield and quality, especially grain protein and hardness. However, genetic trait of the hybrid should be taken into consideration before selection of the hybrid to produce hard endosperm grains.

## CONCLUSIONS

1. Nitrogen fertilizer ( 250 kg/ha as urea) application increased grain yield, grain protein content, grain hardness and bulk density at both sowing dates.
2. Urea application at the rate of 250kg/ha produced the highest yield. On the other hand better quality (i.e. grain protein, bulk density and hardness) was recorded at 500kg/ha than 250kg/ha, although yield was lower.
3. Yield components which differed between hybrids for the treatments were total number of grains per cob and 100-grain weight.
4. Vegetative and reproductive development were not influenced by the nitrogen treatments.
5. Grain protein percentage increased with the increased rate of applied nitrogen fertilizer; the response was linear among all hybrids.
6. Grain protein content was strongly related to grain hardness and hardness increased with increasing protein content in hybrids P3787 and A82-8xNZ84 but not in hybrid P3751.
7. Kernel bulk density (test weight) was increased at the higher nitrogen application rate.
8. Hybrids P3787 and A82-8xNZ84 showed improvement of hardness at higher nitrogen application rate and are more suitable for dry milling than hybrid P3751.

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**APPENDICES**

**Appendix 1.** Soil description for the trial area at Frewens Block, Massey University, Palmerston North.

**Soil name:** Manawatu mottled fine sandy loam

**Parent material:** Medium textured alluvium

**Slope topographic position:** flat, river-levee and flats

**Description of representative soil profile:**

**0 - 23cm:** dark greyish brown fine sandy loam to silt loam; friable, strong nut structure

**23- 53cm:** olive brown fine sandy loam, friable, weak nut structure

**53-277cm:** olive medium to fine sand becoming coarser with depth, loose, structureless

**277-318cm:** olive grey coarse sand with stone and gravels

**Distinguishing features of soil and environment:**

Occurs on occasionally flooded river levels and flats, well to moderately drained with fine sandy loam textures. Differs from Manawatu silt loam in being slightly sandier throughout with fine sandy loam subsoil, passing down to sand.

**Nature of the underlying subsurface:**

Underlain by loose sand with silty bands and with weakly packed gravels and stone, within about 3m of the surface.

**Drainage:** Well drained



**Appendix 2.** Weekly mean minimum and maximum temperature ( $^{\circ}\text{C}$ ), and rainfall (mm) for the 1994/95 season recorded at AgResearch, Grasslands.

Month	Week 1			Week 2			Week 3			Week 4		
	Tmin ( $^{\circ}\text{C}$ )	Tmax ( $^{\circ}\text{C}$ )	Rainfall (mm)	Tmin ( $^{\circ}\text{C}$ )	Tmax ( $^{\circ}\text{C}$ )	Rainfall (mm)	Tmin ( $^{\circ}\text{C}$ )	Tmax ( $^{\circ}\text{C}$ )	Rainfall (mm)	Tmin ( $^{\circ}\text{C}$ )	Tmax ( $^{\circ}\text{C}$ )	Rainfall (mm)
October	4.6	14.6	20.8	8.1	15.7	31.7	6.4	14.4	4.5	9.2	18.5	3.9
November	9.0	18.8	35.7	11.2	16.8	73.6	9.9	15.5	70.2	10.0	17.8	0.2
December	10.5	19.9	0.0	12.3	20.1	4.0	11.0	21.9	10.4	19.0	16.0	14.7
January	9.2	19.3	1.8	11.2	24.9	16.5	14.1	25.2	0.6	15.4	24.2	34.4
February	14.2	22.9	6.0	15.5	22.7	0.0	13.0	20.9	20.5	14.8	23.8	31.4
March	12.1	21.0	6.2	13.5	22.3	32.5	11.1	18.9	66.2	14.6	21.3	10.2
April	15.1	21.4	10.0	12.8	20.0	54.4	10.3	19.5	8.6	13.1	19.0	30.2
May	11.6	17.9	34.7	5.7	15.6	15.7	7.0	15.3	13.0	4.1	14.9	34.0
June	3.6	12.3	9.5	7.0	14.1	23.8	5.8	12.5	15.7	5.8	12.4	34.7

Appendix-3. Soil Test Data

Sample	pH	Olsen P	SO <sub>4</sub>	Exch K	Exch Ca	Exch Mg	CEC
Before ploughing	5.6	9	3.5	0.43	7.5	1.35	21
After application of 150kg/ha Nitrophoska	5.3	13	5.5	0.49	7.2	1.39	21

**Note:** Phosphate and sulphate values are expressed as  $\mu\text{g/g}$  (air-dry). Exchangeable cations and CEC values are expressed as meq/100g (air-dry).

**Appendix-4.** Mean laboratory germination (%) and field emergence (%) for hybrids P3751, P3787 and A82-8xNZ84 at two sowing dates.

Hybrid	Laboratory germination (%)	Field emergence (%) <sup>1</sup>	
		October sowing	November sowing
P3751	96	78.6	80.1
P3787	96	81.1	81.9
A82-8xNZ84	98	82.1	83.2
	ns	ns	ns
LSD	6.35	9.20	7.3
CV%	0.9	5.3	4.5

<sup>1</sup> recorded at 14 (October) and 7 (November) days after sowing.

Appendix-5. Leaf number and leaf appearance rate of hybrids P3787, P3751 and A82-8xNZ84 in both the October and November sowings

Hybrid	October sowing		November sowing	
	Leaf Number	Leaf appearance rate	Leaf Number	Leaf appearance rate
P3787	18.8	0.18 (±0.01)	18.1	0.22 (±0.01)
P3751	18.8	0.18 (±0.01)	18.9	0.23 (±0.01)
A82-8xNZ84	19.8	0.19 (±0.01)	19.6	0.23 (±0.01)
LSD(p<0.05)	0.82		0.56	

Values within brackets are standard errors of the mean

**Appendix-6.** Required days and heat units to mid-silking and physiological maturity from the days after sowing (DAS) in terms of days and in terms of Growing degree days (GDD).

Hybrid	October sowing				November sowing			
	Days to mid-silking DAS	Required GDD	Days to Physiological Maturity	Required GDD	Days to mid-silking DAS	Required GDD	Days to Physiological Maturity	Required GDD
P3787	100.7	961.3	172.1	1844.2	88.1	948.3	167.2	1779.2
P3751	108.4	1047.4	179.9	1991.3	95.6	1024.6	174.5	1831.4
A82-8xNZ84	108.7	1048.6	180.2	1993.1	95.8	1025.1	174.3	1833.2
LSD(p<0.05)	0.82	1.86	0.74	2.40	0.54	1.42	0.64	2.96

**Appendix-7.** Analysis of variance (ANOVA) model for the analysis of response variables in GLM procedure

General Linear Models Procedure

Dependent Variable: Cobs per Plant

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	14	0.04740741	0.00338624	2.03	0.1127
Error	12	0.02000000	0.00166667		
Corrected Total	26	0.06740741			

R-Square	C.V.	Root MSE	COB_P Mean
0.703297	3.894948	0.040825	1.048148

Source	DF	Type I SS	Mean Square	F Value	Pr > F
REP	2	0.00962963	0.00481481	2.89	0.0946
NITRO	2	0.00074074	0.00037037	0.22	0.8040
REP*NITRO	4	0.01037037	0.00259259	1.56	0.2487
HYBRID	2	0.01407407	0.00703704	4.22	0.0409
NITRO*HYBRID	4	0.01259259	0.00314815	1.89	0.1772

Source	DF	Type II SS	Mean Square	F Value	Pr > F
REP	2	0.00962963	0.00481481	2.89	0.0946
NITRO	2	0.00074074	0.00037037	0.22	0.8040
REP*NITRO	4	0.01037037	0.00259259	1.56	0.2487
HYBRID	2	0.01407407	0.00703704	4.22	0.0409
NITRO*HYBRID	4	0.01259259	0.00314815	1.89	0.1772

Tests of Hypotheses using the Type III MS for REP\*NITRO as an error term

Source	DF	Type III SS	Mean Square	F Value	Pr > F
NITRO	2	0.00074074	0.00037037	0.14	0.8711