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**EFFECT OF PLANT NUTRITION, TIME AND METHOD OF
HARVESTING ON SEED YIELD AND QUALITY OF WRINKLED AND
SMOOTH-SEEDED PEA(*Pisum sativum* L.) VARIETIES.**

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ABSTRACT

Effects of nitrogen(N), phosphorus(P) and time and method of harvesting on seed yield and quality were examined for smooth(Maple) and wrinkled(Pania)-seeded pea cultivars. These cultivars were grown under field conditions, and at different rates of N(0, 100 and 200 kg N/ha) and P(0 and 250 kg superphosphate/ha). Seed was harvested at 3 different times(35%, 25% and 15%SMC), and shelled either by hand or by a combine-harvester(at 1,350rpm). For hand-shelling, 120 plants were sampled from each plot of which 40 plants were used to determine the number of pods/plant, seeds/pod and 1000-seed weight, and subsequently used to determine seed quality i.e. Standard Germination, Accelerated Aging, Conductivity and incidence of Hollow Heart. The remaining 80 plants from each plot were used for separation into bottom, middle and top pods and subsequently used to determine 1000-seed weight, seed germination, conductivity and the incidence of hollow heart.

For machine-threshing, 120 plants were sampled from each plot, threshed by combine-harvester and seed subsequently used to determine seed quality by 1000-seed weight, Standard Germination Test, Accelerated Aging, Conductivity Test and incidence of Hollow Heart. Samples from each plot, following machine-threshing, were also used to determine seed damage by visual observation and by the Ferric Chloride Test.

Pea cv. Pania produced higher seed yield than cv. Maple in both hand-shelling and machine-threshing due to a much higher numbers of seeds/pod(5.76 and 3.57 seeds/pod, respectively) and much lower mechanical damage(10.32% and 27.98%, respectively), but had a much lower capacity to produce seed of high vigour than cv. Maple.

Application of nitrogen increased seed yield of both hand-shelled and machine-threshed seeds due to increased numbers of pods/plant, whereas yield was not directly affected by phosphorus addition. However, interaction between 100kg N/ha and 250 kg superphosphate/ha decreased seed weight. Application of nitrogen also increased seed vigour as expressed by increased seed germination percentage after accelerated aging, decreased hollow heart incidence and decreased conductivity value, particularly in cv.Pania. Application of phosphorus had only a small effect on

seed vigour compared with that of nitrogen. Neither seed germination percentage nor mechanical damage was affected by application of nitrogen or phosphorus.

Hand-shelling at different seed moisture contents did not affect seed germination percentage or conductivity value of either cultivar, but delaying the harvest (at the lower seed moisture content) decreased seed vigour in cv. Pania, as expressed by decreased seed germination percentage after accelerated ageing and increased hollow heart incidence. Machine-threshing at different seed moisture contents resulted in different degrees of seed damage, and decreased seed vigour in both cultivars. The most severe damage in cv. Maple occurred when machine-threshed at 15%SMC, whereas in cv. Pania it occurred at 35%SMC. Least damage occurred at 35% and 25%SMC in cv. Maple and cv. Pania, respectively. Unlike hand-shelling, machine-threshing at lower seed moisture content resulted in higher seed vigour in both cultivars, suggesting that bruising which occurs mainly at the high seed moisture content is more harmful than splitting which mainly occurs at the low seed moisture content threshing, in terms of decreasing seed vigour.

The top pods on pea plants produced seeds with lower seed weight in both cultivars, with higher hollow heart incidence in cv. Pania and with higher conductivity value in cv. Maple, than middle and bottom pods. Application of 200 kg N/ha and 250 kg superphosphate/ha improved vigour of seed from different pod positions to a similar and high level.

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Effect of plant nutrition, time and method of harvesting on seed yield and quality of wrinkled and smooth - seeded pea (*Pisum sativum* L.) varieties.

CHAPTER 1.

INTRODUCTION

Peas (*Pisum sativum* L.) have been the most important grain legume in New Zealand for many years (Kelly, 1987). Most pea crops are spring sown, usually from mid October on, because earlier sowing into cold soils is considered to give rise to poor emergence, and a weak crop which has little chance of producing a high quality yield at harvest (Castillo et al., 1994).

During seed production, the environment during maturation can affect seed quality (Castillo et al., 1994). These authors showed that seed from a December sowing (late sowing) was of higher quality than seed from a November sowing (recommended sowing date), and suggested that the seeds from the November sowing encountered greater climatic extremes of temperature, rainfall, and relative humidity during seed maturation in January than seeds from the December sowing which matured in February. Climatic conditions during the post maturation pre-harvest period have a great influence on the quality of seed harvested (Delouche, 1980). High temperature during drying or drying too quickly or excessively, can also dramatically reduce seed viability (Bewley and Black, 1986) and vigour (Hampton, 1990), and can increase the incidence of hollow heart (Harrison, 1973).

Mineral deficiency predominantly affects the number of seeds produced, but unless the deficiency is severe, has relatively minor effects on seed composition. As with the vegetative tissues of plants, application of mineral fertilizer causes manifold changes in the element composition of seed (Austin, 1972). For example, Austin (1966a) found that phosphorus deficiency reduced the phosphorus concentration in pea seeds. Mean dry weight of the seed also decreased, but these

seeds contained slightly greater concentrations of nitrogen and potassium than seeds from plants receiving additional phosphorus. A negative correlation between the concentrations of nitrogen and phosphorus was also observed by Austin(1966b) in a radish(*Raphanus raphanistrum*) seed crop. Hadavizadeh and George(1989) reported that increasing plant nitrogen nutrition increased seed dry weight, and that the interaction of high nitrogen(i.e.1000 mg N/plant) with medium phosphorus supply(i.e.250 to 500 mg P/plant) produced an increase in vigour of pea seeds. Browning and George(1981) also found that the application of a medium nitrogen level(450 mg N/plant) with low level of phosphorus(88 mg P/plant)produced seed with a lower incidence of hollow heart compared with the combination of medium nitrogen levels and higher levels of phosphorus. However, high nitrogen nutrition levels tend to increase the number of pea plants with bleached seeds(Hawthorn and Pollard,1966; Browning and George,1981). The disruption of cellular membranes which is associated with seed bleaching normally produces high conductivity readings and often poor emergence in the field(Browning and George,1981).

Nitrogen and phosphorus are major constituents of proteins and phospholipids, which are the building blocks of cell membranes. The stability of membrane structure depends largely on the solubility properties of these molecules. Because of this, the interaction effect of nitrogen and phosphorus nutrition on seed vigour can be revealed by conductivity value during seed imbibition(Hadavizadeh and George,1989; Coolbear,1994).

The seed quality determination in this study will focus on seed vigour. Unlike germination, seed vigour refers to those properties of seed which influence the speed and uniformity of germination and the ability to germinate and emerge under a wide range of field conditions. High vigour seed lots will perform better under environmentally stressed seed bed conditions than low vigour seed lots, even though the laboratory germination of the lots are similar(Hampton,1995). Low seed vigour in high germinating seed lots may be seen as slow or uneven field emergence or failure to establish in the field. These effects may lead to reduced crop growth and yield(Hampton,1984). As shown by Hampton and Scott(1982), pea seeds with different vigour levels sown using equal numbers of seeds in the field, clearly show reduced emergence from the low vigour lines, resulting in lower plant populations

and a significant yield loss of green and seed peas. However, there are no differences in the vigour of seedlings which have established. In low vigour seed lots, when equal plant populations are obtained by sowing seeds at compensatory rates, no differences in yield are likely. Although individual pea plants can produce more pods/plant and more seeds/pod when plant population is low, this does not fully compensate in overall seed yield. Therefore, the optimum population is important for achieving high yields(Hampton and Scott,1982). Since accurate information is vital for determining optimum sowing rate, the vigour testing can be used to support this requirement. For peas, in particular, the hollow heart test, conductivity test and the multiple vigour test such as expected field emergence(EFE) which uses germination, conductivity and hollow heart data(Scott and Close,1976) can provide a more sensitive index of seed quality than the standard germination test for predicting field emergence (Hampton,1984).

Mechanical damage inflicted during harvesting can severely reduce seed viability, especially in large-seeded legumes. Injuries close to vital parts of the embryonic axis or near the point of attachment of the cotyledons to the axis usually bring about the most rapid loss of viability. It is therefore of the utmost importance to harvest and thresh seed carefully, ensuring that the drum speed is slow and seeds are harvested at the correct time(Matthews,1973; Gane et.al,1984). In peas(cv. Pania and cv. Princess), Castillo et al.(1992) reported that machine harvesting at 25% SMC produced higher vigour seeds than when the same cultivars were harvested at 15% SMC. Similarly, Gane et al.(1984) reported that harvesting pea seeds at around 25% SMC minimised harvest damage and produced higher quality seed than when harvested at 15% SMC. This is important since in New Zealand, peas are generally threshed at around 14% SMC.

Despite this, detailed studies on the effect of mechanical damage to pea seed are not widely reported in the literature, particularly information concerning the interactive effects of plant nutrition and varieties and the differential ability of different cultivars to withstand mechanical impact. Also there is little information available on the effects of time of harvest and pod position on seed quality. Further information on these effects would be helpful in providing a better understanding of pea seed production techniques.

The principle objectives of this study were to determine:

- the effects of applied nitrogen and phosphorus pea seed yield and quality, particularly seed vigour.
- the effects of time and method of harvesting on seed quality of smooth or wrinkled-seeded pea cultivars.
- the possible effects of threshing method and severity on levels of seed damage in smooth or wrinkled-seeded peas harvested at different seed moisture contents.
- the effects of plant pod position on subsequent seed quality.

CHAPTER 2.

LITERATURE REVIEW

2.1. SMOOTH-AND WRINKLED-SEEDED PEAS

Many workers (Peat *et al.*, 1948; Stickland and Wilson, 1983; Edwards and ap Rees, 1986; Smith, 1988; Bhattacharyya *et al.*, 1990; Wang and Hedley, 1991) have studied the different properties and characteristics of round and wrinkled-seeded pea cultivars. The production of alternative forms has been shown by Smith (1988) to be governed by a single genetic locus, *rugosus* or *r*. The visible effect of *r* on seed phenotype results from the profound effect of this locus on the composition of the developing pea seeds. Pea seeds that are homozygous recessive at the *r* locus (*rr*) are wrinkled when mature, whereas those that are heterozygous or homozygous dominant at this locus (*Rr*, *RR*) are round when mature (Hedley *et al.* 1986). There is also a marked difference in starch metabolism between round (*Rr* or *RR*) and wrinkled (*rr*) peas. In round-seeded peas, starch grains are large and simple, while in wrinkled-seeded peas they are small and compound. Round seeds contain larger amounts of starch than wrinkled seeds and have much higher ratios of amylopectin to amylose (Peat *et al.* 1948; Bhattachayya *et al.*, 1990). Starch is typically 60-70% amylopectin and represents 50% of the final dry weight in round seeds, but only 30% amylopectin and 30% of final dry weight in wrinkled seeds (Kellenbarger *et al.*, 1951; Greenwood and Thomson, 1962). Mature wrinkled seeds contain more sucrose and lipid than mature round seeds. Sucrose and lipid are typically 10% and 5% respectively of the dry weight of wrinkled seeds, but only 5-6% and 2-3%, respectively, of the dry weight of round seeds (Coxon and Davies, 1982).

In wrinkled seeds, the levels of free sucrose are higher than in round seeds, and this probably leads to the higher osmotic pressure and hence higher water content and greater cell size of developing wrinkled seeds (Hedley *et al.*, 1986; Ambrose *et al.*, 1987; Wang *et al.*, 1987). Wrinkled seeds lose a larger proportion of their volume at seed maturation, because the testa does not shrink at the same rate as the cotyledons therefore becomes distorted or wrinkled. Mature wrinkled seeds also

contain less storage protein, legumin, than round seeds (Davies, 1980; Coxon and Davies, 1982). Many of these effects of the *r* locus were deduced by Hedley *et al.* (1986) using different round and wrinkled pea cultivar, including the use of near-isogenic *RR* and *rr* lines.

The multiple differences observed between mature round and wrinkled peas could result from modification of a regulatory gene controlling all the affected processes or from a primary metabolic change, which in turn influences a number of other metabolic parameters (Bhattacharyya *et al.*, 1990). The relatively low starch content of wrinkled seeds indicates that the capacity of starch synthesis during development may be lower than in round seeds. Measurements of amounts of metabolites of the putative pathway of starch synthesis show that the levels (including the level of ADP-glucose, the substrate for starch synthesis) are higher in developing embryos of wrinkled-seeded peas than in round-seeded peas (Edwards and ap Rees, 1986). This indicates there may be a block very late in the pathway of starch synthesis in embryos of wrinkled peas, leading to an accumulation of metabolites of the pathway. A reduction in the activity of the starch-branching enzyme has been suggested as the cause of the block in pathway of starch synthesis in embryos of wrinkled peas. This enzyme catalyses the conversion of amylose to amylopectin (Smith, 1988). Matters and Boyer (1982) found that activity of the starch-branching enzyme in developing embryos of the wrinkled-seeded pea cv. Progress 9 was up to 12-fold lower than in the round-seeded pea cv. Alaska. Smith (1988) suggested that the difference in activity of the starch-branching enzyme between embryos of round and wrinkled peas is likely to be due to the absence from the embryos of wrinkled peas of one of the two isoforms occurring in embryos of round peas. This hypothesis has been confirmed by the work of Bhattacharyya *et al.* (1990), who described that the absence of this isoenzyme from the embryos of wrinkled peas was due to a lesion in the gene encoding the production of the starch-branching enzyme.

The level of sucrose in seeds is a reflection of the amount of translocation to seeds from parent plant which is mainly used for starch synthesis. Initially there is little such synthesis, and translocation leads to an accumulation of sucrose. The higher sucrose content in developing wrinkled peas is probably due to the much lower rate of conversion to starch than occurs in round peas (Stickland and

Wilson,1983; Smith,1988). The accumulation of sucrose increases osmotic pressure in developing embryos, a change which can affect protein biosynthesis(Wang and Hedley,1991). Turner *et al.*(1990) found that the increase in the internal osmotic pressure of the embryo in culture resulted in a decrease in mRNA levels for main storage proteins and the effect was greater for legumin than for vicilin. They suggested, therefore, that the difference in storage protein composition between wrinkled and round peas was the result of the differential stability of legumin and vicilin mRNAs. This links the low legumin phenotype with either higher sucrose concentration or higher osmotic pressure in wrinkled pea embryos(Wang and Hedley,1991).

The differences between round-and wrinkled-seeded peas are the result of complex changes in metabolic processes during seed development. The discovery of the *r* locus which encodes the starch-branching enzyme helps to clarify the complex changes to some extent.

2.2. SEED VIGOUR

Seed vigour is a concept which refers to those properties of seed which influence the speed and uniformity of germination and ability to germinate and emerge under a wide range of field conditions(AOSA,1983; ISTA,1987).

Seed vigour is an important factor in peas and seed should be of high vigour at planting so that the seeds are more likely to withstand adverse environmental conditions affecting germination and field establishment. Planting low vigour peas can lead to unsatisfactory results due to slow or uneven field emergence, poor establishment, reduced crop yield and may even lead to significant economic loss(Perry,1982; Hampton and Scott,1982; Hampton,1984; Matthews and Powell,1986). Perry(1982) has illustrated the reduced establishment and seedling growth associated with low vigour seed in peas grown in both protected environments and field conditions, and Hampton and Scott(1982) have shown that low vigour peas can reduce overall yield due to poor seedling emergence and reduced plant stand. However, they found no differences in the harvest yield of low and high vigour peas when sown at equal plant densities. Planting low vigour seeds

can also result in economic loss resulting from emergence failure necessitating resowing, and often causing a delay in harvest. This can lead to a fall in market prices, and lower profits from(Matthews and Powell,1986).

Maximum seed quality occurs at physiological maturity, the point when maximum dry seed weight is attained, after which vigour and viability decline during ageing on the plant(Delouche,1980), and during harvesting, processing and storage(Powell *et al.*1984). Seed ageing has come to be recognised as the major cause of reduced seed vigour and viability and involves the deterioration process,i.e. the accumulation of irreversible degenerative changes until eventually the ability to germinate is lost(Powell,1988). Seed deterioration is generally progressive and sequential, although it is very difficult to distinguish primary causes from secondary effects. Physiological and physical damage to cell membranes is likely to be the fundamental cause of seed deterioration. However, enzyme, respiration and hormonal changes, impaired protein and RNA synthesis, genetic change, and accumulation of toxic metabolites are also involved(Hampton,1994). It is possible that many or all of these forms of degradation can occur to some extent within a single seed, but no doubt certain deterioration processes will be more important than others, depending on species and the ageing environment(Priestley,1986). However, it is now generally accepted that causes of vigour differences in many species are physiological rather than directly physical(although the latter can influence the former), and that cell membrane integrity, as determined by deteriorative biochemical changes, and/or physical disruption, can therefore be considered to be a fundamental cause of differences in seed vigour(Hampton,1994).

Low vigour seed lots are physiological older(Hampton and Hill,1990), and can be influenced by a number of factors such as genetic constitution,nutrition of the parent plant, position on the plant, environment during seed development and maturation, mechanical damage during harvest and processing, and storage conditions. These factors can all be linked to the deteriorative changes of cell membranes during seed development, handling and storage.

2.2.1. Genetic Constitution

The inherent capacity of the seed is a fundamental factor which will determine the effects of both adverse environmental conditions and mechanical injuries on seed quality (Heydecker,1969). The location, shape, size and nature of individual seed structures account for wide differences in the frequency and seriousness of impact injuries. Large-seeded legumes, in particular, tend to be especially susceptible to such injuries(Moore,1972). The extent and intensity of mechanical damage can be affected by seed characteristics. For example, seeds of small-seeded crops tend to escape serious injuries during harvest. Also the difference in mass and nature of the vegetation harvested along with the seed can provide different protection against impact. Hardseedness in legumes, as well as the presence of a dry, firm endosperm can also provide extra protection(Moore,1972).

Seed quality is often reduced by imbibition damage caused by a faulty testa, but genotypic characters can also be involved(Hampton,1990). White-seeded cultivars of dwarf bean, and lighter-seeded types of chickpea have been found to have higher rates of water uptake and therefore a higher incidence of imbibition damage, leading to lowered vigour and poor field emergence than dark-seeded cultivars(Powell, *et al.*,1986). Moore(1972) also observed that white-seeded varieties of snap bean are generally highly susceptible to both mechanical damage and imbibition damage. The white coats are usually thin, adhere more loosely to the cotyledons, and are more permeable to water than dark-coloured coats.

Although breeders have been giving attention to the aims of improving yield, field character and disease resistance, they have often inadvertently selected for increased seed vigour(Copeland and McDonald,1985). In soybean, for example, studies have shown that viability is maintained longer in smaller than in larger seeds(Wein and Kueneman,1981), because small seeds appear to contain the hardseed character(Calero *et al.*1981; Minor and Paschal,1982;). Hardseeded cultivars resist weathering and also store longer in open conditions. However, hardseedness often causes staggered emergence over several weeks when planted in the field and therefore can create problems at harvest because of uneven ripening. Kuo(1989) found that a small number of soybean cultivars had seed-coats which exhibited

delayed permeability although seeds were not 'hard'. In these cultivars seed imbibition is initially delayed, making seeds less susceptible to cycles of wetting and drying prior to harvest. Also they absorb moisture more slowly from the ambient atmosphere during open storage. Selection for this character has the potential to allow the production of soybeans with high viability and vigour, and improved storage performance.

In peas, a higher incidence of hollow heart has been found in wrinkled than in smooth-seeded peas(Perry,1980). Rush(1987) found that this higher incidence in wrinkled peas was often correlated with the amount of seed exudate in 'steep' water detected by the electroconductivity test, thereby resulting in lower seed vigour than in smooth-seeded cultivars.

2.2.2. Nutrition of Parent Plant

The International Seed Testing Association(ISTA,1987) outlines a list of major factors which are associated with seed vigour. Environment and plant nutrition are considered to be vital factors. In terms of plant nutrition, crop fertilizer requirements have tended to be managed for optimising seed yield rather than seed quality. However, in order to obtain satisfactory yield and quality levels, there is need to also consider the nutrient requirements for seed quality rather than only for quantity(Delouche,1980; Hampton,1990).

Effect of plant nutrition on the quality of seed produced has been studied by a number of workers. Nitrogen, phosphorus and potassium have been the main nutrients examined. In peas, negative interactions between nitrogen and phosphorus were found by Austin(1966a), where phosphorus deficiency reduced both phosphorus concentration and mean dry weight of the seeds, but slightly increased nitrogen concentration compared with seeds from plants receiving additional phosphorus. This effect has also been observed by Austin(1966b) in a radish(*Raphanus raphanistrum*) seed crop. He found that irrespective of the time of nitrogen application, increases in nitrogen content in seeds was accompanied by a concomitant reduction in their phosphorus content. Hadavizadeh and George(1989) showed that the interaction of high nitrogen (i.e.1000 mg per plant) with medium phosphorus supply(i.e. 250 mg

and 500 mg per plant) produced an increase in seed vigour. However, potassium nutrition was found to have very little effect on seed yield and vigour in peas. Browning and George(1981) also reported that two treatments; 450 mg N with 255 mg P, and 450 mg N with 422 mg P per plant, produced pea seeds with a higher incidence of hollow heart compared with 450 mg N with a lower level of phosphorus(88 mg P per plant). High nitrogen nutrition levels(800 and 1,050 mg N per plant) also produced an increase in the number of plants with undesirable bleached seeds. Hawthorn and Pollard(1966) reported a similar finding for pea seed crops in Utah,USA, where several successive years of nitrogen fertilization increased seed scald or bleaching. Bleached seed reflects a disruption of cellular membranes, and results in an increase in membrane permeability with greater amounts of sugar and electrolytes being leached out during imbibition. This produces high electroconductivity readings and often leads to poor emergence in the field(Browning and George,1981).

2.2.3. Position on The Parent Plant

Position in the canopy can affect vigour in large-seeded legumes(Hampton,1991). For example Adam *et al.*(1989) demonstrated that soybean seed vigour(detected by germination, accelerated ageing test and seed weight) was higher in seeds obtained from the top of the plant. Soybean pods originating from early flowers start formation earlier than pods originating from late flowers. Consequently, early and late set soybean seeds encounter seed formation periods of differing durations and differing maturation environments(Dunphy *et al.*,1979). As a result bottom seeds mature on the plant in the field for a longer period than top seeds and are potentially exposed to more environmental stress factors such as high temperature and relative humidity which increase seed deterioration(Adam *et al.*,1989).

The longer exposure of bottom seeds to seed pathogens may also reduce seed quality. Some studies have shown that *Phomopsis*, which is primarily responsible for seed decay, is enhanced in seeds matured on the bottom of soybean plants(Thomison *et al.*1987; TeKrony *et al.*1983).

Assimilate mobilisation and sink response may also be involved in the differences in seed quality between seeds from different positions on the plant. Significant differences in the rate of dry matter accumulation of individual seeds among cultivars and planting dates have been reported by Egli *et al.*(1978), while Pascal and Crookstone(1978) concluded that late maturing soybean seeds consistently exhibited faster rates of dry matter accumulation than early maturing seeds. Also the greater rate of photosynthesis of the top leaves than the bottom leaves of soybean(Schow *et al.*,1978) may enhance the superior quality of the top seeds.

The micro-environment within the crop canopy has been linked to the reduction in quality of pea seeds. Halligan(1986) and Castillo(1992) have both reported that the maximum daily temperature measured within the canopy of a pea crop is 2-5°C higher than that above the crop canopy. Similarly, Perry and Harrison(1973) demonstrated that peas within a pod experience temperatures 4-5°C higher than ambient temperature. As reported by Halligan(1986) high ambient temperature during the seed filling stage can enhance hollow heart development in peas. Castillo(1992) also showed that relative humidity within the plant canopy was higher than that above the crop canopy, and suggested that high temperature and relative humidity within the crop canopy can reduce seed quality.

2.2.4. Environment During Seed Development and Maturation

The influence of the environment has been recognised as an important factor for seed production(Delouche,1980). Ideal conditions for seed production such as: good soils; suitable radiation, rainfall and temperature; low humidity and relatively calm weather during seed maturation and harvest period, have contributed to the selection of specialised seed production areas and manipulation of cropping systems(Delouche,1980; Hampton,1994). However, seed is not often produced in such conditions.

The advantages of producing seed in areas specially adapted to seed production are that seed set, seed yield, and recovery at harvest are higher and relatively stable; seed germination and seed vigour are consistently high; and seed borne diseases can often be avoided or are more easily controlled(Delouche,1980).

The environment plays a vital role in the development and maturation of the seeds. For example, acute deficiency in moisture supply resulting from temporary but severe drought can have disastrous effects. A drought during the seed development period usually interrupts seed development and results in light, shrivelled seed (Delouche, 1980). Dornbos and Mullen (1985) stated that a combination of drought and high temperature stress during seed fill of soybean crops reduces seed size, viability, and vigour more than drought stress alone. Such a combination may result in 100% shrivelled seeds in some soybean cultivars (Franca Neto *et al.*, 1993). Shrivelled seeds (92%) have been reported when plants of a mutant genotype (357-1) of soybean were exposed to high temperature (34/26°C, 8/16h) alone (Honeycutt *et al.*, 1989). In peas, the most critical time when high temperature adversely affects pea yield is from flowering through pod filling. The yield component primarily affected by temperature above the optimum is reduction in the number of pods per plant (Pumphrey and Ramig, 1990). A temperature range of 20 - 21°C is considered optimum for peas (Fletcher *et al.*, 1966). Information available on the heat:pea yield relationship in terms of optimum temperature is variable. Lambert and Linck (1958) and Nonnecke *et al.* (1971) have suggested 27°C as a critical level for maximum daily temperature. Wang (1962) gave 25-26°C as the upper optimum temperature during blooming, while Pumphrey and Ramig (1990) suggested an upper optimum temperature of 25.6°C. Apart from seed yield, high temperature stress can depress the quality of pea seeds produced. Perry and Harrison (1973) found evidence of an association between the predisposition of seed to hollow heart development during germination and the presence of high ambient temperatures during their maturation on the plant. Halligan (1986) showed that pea (cv. Pania) plants exposed to high temperature (35°C day/25°C night) at a seed moisture content of 70-80% produced seed with the highest incidence of hollow heart and at all stages of development this incidence increased with the length of exposure to high temperature. Hollow heart is a physiological disorder in pea seed, which can affect germination, seedling emergence and establishment and ultimately crop yield (Perry and Howell, 1965; Harrison and Perry, 1973; Scott and Close, 1976).

Climatic conditions during the preharvest period also have a great influence on the quality of seed harvested (Delouche, 1980). Maximum seed quality occurs at

physiological maturity after which vigour and viability decline because of the ageing process of the seed(Delouche,1980; Powell,1988). Adverse weather conditions, such as high ambient temperature and high relative humidity during the harvest period, cause seed quality problems(Delouche,1974). Seed moisture content and temperature are considered as major factors influencing the extent of seed ageing with an increase in either or both accelerating ageing. Seed ageing on the plant is frequently referred too as "weathering", reflecting the influence of these factors(Matthews and Powell, 1986). In a study on the effect of planting and maturity dates on soybean seed quality, Green *et al.*(1966) found that soybean plants from early planting dates which mature seed during hot, dry weather often produced seed of reduced quality. Seed from later dates of planting which reached maturity under cooler, moister weather conditions, were high in quality. A similar effect of planting date on pea seed quality was observed by Castillo(1992). Green *et al.*(1966) also found that delayed harvest of soybean seed results in a reduction in viability. In contrast, if the seed is harvested too early, immature seeds lower the percentage of viability of the lot and have greater mortality in the soil. Matthews(1973) found that if enhanced drying occurs following harvest, immature seeds are more vulnerable to membrane damage than mature seeds. Rainfall after seed maturation is one of the most dramatic adverse effect of weathering on seed quality. In peas, Matthews(1973) observed that heavy rainfall just prior to harvest reduces the percentage viability of dried seed and increases the leaching of solutes from the dried seed into steep water.

Weathering is a major problem in seed production. The severity of weathering and the limitation imposed on seed quality by weathering generally increases from cool to warm areas. The worst situation is in the humid subtropics and tropics, where the quality of seed produced is generally low and deterioration continues at a rapid rate during storage because of high temperatures and humidities(Delouche,1980).

2.2.5. Mechanical Damage

Irrespective of the inherited or original physiological potential of the seed, seed intactness is nevertheless thought, by many seed technologists, to be the key to the maintenance of the vigour of seeds(Heydecker,1972). Seeds deteriorate more

rapidly if they have been injured in some way(Roos,1980).

The nature of mechanical damage varies widely. The most intensive injuries reduce viability immediately, whereas small injuries often do not cause an immediate loss in viability, but become increasingly apparent as ageing occurs. In seeds that are extremely dry and brittle, fracturing is the predominant type of injury, while bruising tend to be prevalent in moist seeds(Moore,1972).

Damage may be caused during maturation even without any interference by man(Delouche,1980; Perry,1980; Matthews and Powell,1986). More frequently it is, however, due to faulty threshing or processing methods(Gane *et al.*,1984).

The degree of damage depends on the interaction of several effects such as seed moisture content, drum speed of the threshing machine and seed cultivar.

Threshing seed at too high or too low a seed moisture content may cause severely damaged seeds. Bartsch *et al.*(1986) demonstrated that soybean seeds at 13% and 18% moisture content exhibited similar impact damage resistance, but impact damage increased significantly as seed moisture content dropped from 13% to 8%. Castillo *et al.*(1994) found that pea seeds harvested at 15% and 40% moisture content gave lower seed vigour than seeds harvested at 25% moisture content due to higher mechanical damage levels. Jech *et al.*(1993) reported that even minimal height of free-fall may also cause damage and decrease germination in pea seeds. They suggested that the effect of seed moisture content was very significant, with over-dried peas(<9% seed moisture content) being very susceptible to mechanical damage.

Drum speed of the threshing machine is also closely associated with mechanical damage. Saini *et al.*(1982) examined the effect of drum speed on soybean damage levels and found that faster drum speed gave relatively lower quality results and proportionally lower quality after storage than lower drum speed. Similarly, Kausal *et al.*(1991) reported that soybean seeds threshed at 400 rpm gave the highest percentage of germination and vigour index compared with threshing at 600 and 800 rpm. The effect of threshing method on small-seeded legumes was investigated by Jech(1978), as cited by Boyd(1987), who reported that damage in threshing clover seed depended on drum speed, drum clearance, threshing concave and type of threshing bar.

Kind of seed is one of the significant factors influencing mechanical damage

in seeds. Weight and size of large-seeded legumes, in particular, tends to be especially susceptible to injuries that reduce viability(Moore,1972).

2.2.6. Storage Condition

The fundamental purpose of seed storage is to maintain the germination of seed at an acceptable level until the seed is planted in the field. The storage longevity of a seed lot is determined by the combination of initial seed quality and storage conditions(Hill,1995).

Good initial quality of seed prior to storage is fundamental to the success of storage system. In general, seeds with high initial quality store better than seeds with low initial quality and are more able to withstand adverse storage conditions(Arvier,1983;Hill,1995). The potential of different initial quality of soybean seed lots during storage has been investigated by Arulnandhy(1988), who found that soybean seedlots with high(>80% good seeds) and medium(60-80% good seeds) quality showed no loss of viability or vigour after 3 months storage under ambient storage(23.5-32.6°C) or 9 months in controlled storage conditions(20±1°C, RH 60%). Poor quality seed lots(<60% good seeds), however, deteriorated rapidly under both conditions. Marcos *et al.*(1988) stored soybean seeds under normal environmental conditions, in a dry chamber(RH 35%) and in cold chamber(10°C, 80% RH) for 6 months, and found that relative humidity and initial quality of seeds were the major factors determining satisfactory storage.

Problems occurring during storage are normally concerned with unfavourable environmental conditions such as high temperature and high relative humidity which are considered as the most important factors affecting seed quality during storage(Arvier,1983; Hill,1995). Many workers have investigated the relationship between storage conditions and the storage life of seed. Sripichitt *et al.*(1989) stored soybean seeds with moisture content of 6,8,10 or 12% under controlled conditions at 26°C and 80%RH. They found that germination of seeds stored at 10 and 12% moisture content declined more rapidly than seed stored at 6 and 8% moisture content. Charjan and Tarar(1992) stored five soybean cultivars at 9% moisture content at 32,63 and 92%RH in cloth bags for up to 18 months, and found that seed

moisture content decreased with storage at 32%RH and increased with storage at 63 and 92%RH. At 92%RH 3 of the 5 varieties lost their germinability within 3 months.

Problems of high temperature and high relative humidity conditions are very common in uncontrolled conditions(open storage), particularly in areas of high humidity and high average temperature, which occur in the humid tropics. These conditions are well known to shorten storage life. In ideal storage, both relative humidity and temperature should be kept low(Arvier,1983).

2.3. SEED VIGOUR MEASUREMENT IN PEAS

2.3.1. Hollow Heart

Hollow heart is a physiological disorder of a germinating pea seed which is apparent as cracked or sunken areas of white dead tissue in the centre of the adaxial surface of the cotyledons. Symptoms vary from a small shallow depression to a large cavity, often with cracks across the affected area, and with both cotyledons usually being similarly affected(Perry and Harrison,1973; Halligan,1986). The disorder develops during the maturation of the seed and becomes apparent only when the seed has germinated(Allen,1961; Perry and Howell,1965). The name 'hollow heart' was given and first described by Myers(1948) who found no pathogen associated with it and distinguished it from marsh spot caused by manganese deficiency(Perry and Harrison,1973). Hollow heart is a common physiological disorder of wrinkle-seeded garden pea and can be found in a high proportion of seeds from many countries(Perry and Howell,1965). It may affect germination, seedling emergence and establishment and ultimately crop yields. The effects appear to be greatest in cold, wet soils(Perry and Howell,1965; Harrison and Perry,1973; Scott and Close,1976; Gane and Biddle,1973).

Oxygen deficiency during seed imbibition, physical stress during maturation and drying seeds too rapidly have all been suggested as factors inducing the disorder(Allen,1961; Perry and Howell,1965; Moor,1964). Halligan(1986) showed that garden pea(cv. Pania) plants exposed to temperature(35°C day/25°C night) at seed moisture contents of 70% to 80%(about the onset of pod wrinkling) produced

seed with the highest incidence of hollow heart(exceeding 80% after 10 days exposure). At all stages of development the incidence increased with the length of exposure to high temperature. These results support the finding of Perry and Harrison(1973) who found evidence of an association between the predisposition of seeds to develop hollow heart symptoms and the presence of high ambient temperatures during their maturation on the parent plant. They also found that the disorder will develop during germination, and the proportion of seeds affected in any sample depends on the rate of water imbibition. Moore(1964) reported that more seeds develop hollow heart when water imbibition is rapid. Don *et al*(1984) suggested that too rapid imbibing of water during the germination process of a seed can cause dead cells on the adaxial surface and presents the potential for hollow heart induction. Conversely, Heydecker and Kohistani(1969)stated that slow water uptake and moisture stress enhanced symptom development. However,in their experiment, there was a positive causal correlation between incidence of hollow heart and microbial infection of the cotyledons under wet conditions.

Drying immature seeds at high temperature can also cause hollow heart. Perry and Harrison(1973) demonstrated that hollow heart incidence increased with increasing drying temperature and decreased as seed maturity advanced. They also suggested that drying temperature was less important as maturity progressed.

Hollow heart delays germination, causes poor emergence and reduces the growth of pea seedlings. Plants from seeds with the disorder are smaller and yield less than those from normal seeds(Harrison and perry,1973). Cells the area affected by hollow heart are dead(Perry and Howell,1965) and the reduction in the amount of food reserves available to the developing seedling axis may result in a decrease of seedling growth. However, immobilization of starch reserves does not wholly account for the reduced growth, suggesting the presence of a germination or growth inhibitor. The immobilization of starch within the dead tissue, coupled with the colonization and utilization of those reserves by soil borne micro-organisms may also cause a reduction in the growth of seedlings in the field(Harrison and Perry,1973). However, Heydecker and Kohistani(1969) stated that hollow heart does not directly affect germination and seedling growth but leads to rotting of the cotyledons, thus predisposing seedlings to fungal infection.

Although several observations are conflicting, it is evident that hollow heart can cause lower field emergence and therefore is an important factor to consider when planning for pea production (Scott and Close, 1976). Scott and Close (1976) and Hampton and Scott (1982) included a hollow heart value in a predictive equation for expected field emergence (EFE), which gave better prediction of field emergence than the standard laboratory germination test.

2.3.2. Conductivity Test

The electroconductivity test (ISTA, 1987) is a measurement of electrolytes leaking from plant tissues, and was first adapted for seed testing by Presley (1958) to measure cotton seed viability. It was later developed into a routine test and is now used extensively in the United Kingdom, Australia and New Zealand for the prediction of field emergence of wrinkled-seeded peas (Matthews and Bradnock, 1968; Hampton and Coolbear, 1990). The test is also useful for soybean (*Glycine max* L. (Merr.)) (Yaklieh, Kulik and Anderson; AOSA, 1983; Oliverira *et al.*, 1984), field bean (*Vicia faba* L.) (Hegarty, 1977) french bean (*Phaseolus vulgaris* L.) (Matthews and Bradnock, 1968) and other grain legumes (Matthews and Powell, 1987).

When seed desiccates during ripening the cell membranes lose their integrity as the moisture content falls, but during rehydration membrane integrity is re-established (Simon and Raja Harun, 1972; Short and Lacy, 1976; Bramlage *et al.*, 1978). When imbibition occurs even the healthiest seeds lose solutes, and this loss may be much more severe in less vigorous seeds (Coolbear, 1994). Vigorous seeds probably re-establish membranes at a faster rate with less leakage than less vigorous seeds. The integrity of membranes is important for many biochemical reactions in living cells. Subsequent physiological changes during ageing, decreased enzyme activity, respiration and synthesis are all indicative of reduced metabolic activity (Harman *et al.*, 1976), and could be the consequence of the initial deterioration of membranes (Powell, 1985). The deterioration of membrane ultrastructure and losses of permeability properties in ageing seeds have been detected by electron microscope and by electroconductivity (Harman and Granett, 1972; Powell and Matthews, 1977).

Membranes are constituted of lipids and proteins, with the most important

class of lipids being phospholipids such as phosphatidylcholine, phosphatidylethanolamine and phosphatidylinositol. The stability of membrane structure depends largely on the solubility properties of these molecules(Coolbear,1994), and ageing has been claimed as the fundamental cause of membrane deterioration(Powell,1985). As a result of the implication of membranes in the early stages of ageing, the phospholipid components of cell membranes have been examined by several workers, with the aim of possibly identifying the fundamental cause of seed ageing. Several grain legumes, including peas, have been used in this work in which two possible changes have been examined: the production of free radicals leading to peroxidation of the fatty acids of the phospholipids, and the hydrolysis of phospholipids. One approach to the search for evidence of peroxidation have been shown by Harman and Mattick(1976), where the decline in the unsaturated fatty acids linoleic and linolenic acid in pea seeds aged in 93% R.H. at 30°C for 15 weeks was said to result from their reaction with free radicals. This also correlated with a reduction of seedling growth.

The possible hydrolysis of phospholipids during the early stage of ageing has been indicated by decline in the total phospholipid content of aged pea seeds in which high levels of solute leakage from living cells suggest impaired membrane integrity(Powell and Matthews,1981). This loss of phospholipid is mainly attributable to a decline in phosphatidylcholine, the major phospholipid present. However, contrasting changes in phospholipids have been observed in pea seeds aged at different temperatures and relative humidities, in which increased, reduced and unchanged phospholipid contents were found after ageing. These results suggest that the method of rapid ageing may affect the membranal changes which occur. Thus, although changes in membranes are implicated as a fundamental cause of ageing leading to reduced seed quality, the biochemical events which initiate ageing are not yet clear(Powell,1985).

During rehydration of the seed, imbibition damage can also give rise to differences in seed vigour. Imbibition damage was first organised in pea embryos(removed testa seeds)(Powell and Matthews,1978) and subsequently shown in whole seeds(Powell and Matthews,1979), and was detected by electroconductivity and tetrazolium chloride staining. The damage has been found to be a cause of vigour

differences between seed lots(Powell and Matthews,1979,1980).

Differences in the extent of imbibition damage are often associated with the rate of water uptake due to the condition of the testa(Powell,1985). Large differences in imbibition damage have, however, been found in seeds with similar rates of water uptake both in water(Powell and Matthews,1979) and in soil(Powell and Matthews,1980), suggesting that the condition of the embryo is important in determining the incidence of imbibition damage. Differences in embryo condition may result from ageing and the observation of a greater increase in leakage from scarified compared with intact seeds after ageing than that seen in unaged seeds has suggested that aged seeds with weakened membranes would have greater sensitivity to imbibition damage (Matthews and Powell,1987).

Seed vigour in peas has been associated with electroconductivity of the seed steep water(Mullet and Wilkinson,1979; Perry and Harrison,1973; Bustamante *et al.*,1984). Low vigour seeds exude more electrolytes than high vigour seeds, and therefore give higher electroconductivity readings(Kraft,1986). Because peas exhibit hypogeal germination, increased exudation of electrolytes from seeds during germination may directly stimulate microorganism activity and secondary infection. Pea seed exudates have been shown to increase root rots caused by *Pythium ultimum* and *Fusarium solani* by providing available nutrients for the pathogen at the infection area(Kraft,1986).

Because of the extensive changes going on within the cell membranes the leakage of solutes during early imbibition can be a good indication of the health of seed tissue and thus, by inference, seed vigour. Essentially, leakage is a function of:

1. the level of seed coat cracking/permeability,
2. the rate of membrane reorganisation within the seed,
3. the amount of solute available to leak out(Coolbear,1994).

The conductivity test can assess this damage and discriminate between high and low vigour lots of pea seeds. Potentially, this method of testing may be extended routinely to other species(Coolbear,1994).

The high correlation between field emergence and electroconductivity reading in peas has been well documented(Matthews and Bradnock,1967,1968; Scott and Close,1976; Hampton and Scott,1982; Ladonne,1989; Bladon and Biddle,1991;

Castillo,1992), but Duczmal and Minicka(1989) reported that predicting emergence on the basis of seed evaluation with all laboratory methods failed, the least useful methods being electroconductivity and germination tests. However, their sowings were made under favourable environmental conditions, i.e. good rainfall and no temperature stress. Despite this, the large body of evidence is that the conductivity test is useful for predicting field performance of pea seed lots under suboptimal conditions.

CHAPTER 3.

EXPERIMENTAL DESIGN & MATERIALS AND METHODS

3.1. TREATMENTS

1. 2 cultivars, smooth(Maple) & wrinkled(Pania) - seeded peas
2. 3 nitrogen levels(0, 100, 200 kg N/ha)
3. 2 phosphorus levels(0, 250 kg superphosphate/ha)
4. 3 harvest times(35%, 25%, 15% SMC)
5. 2 harvest methods(hand & machine threshing)
6. 2 block replicates

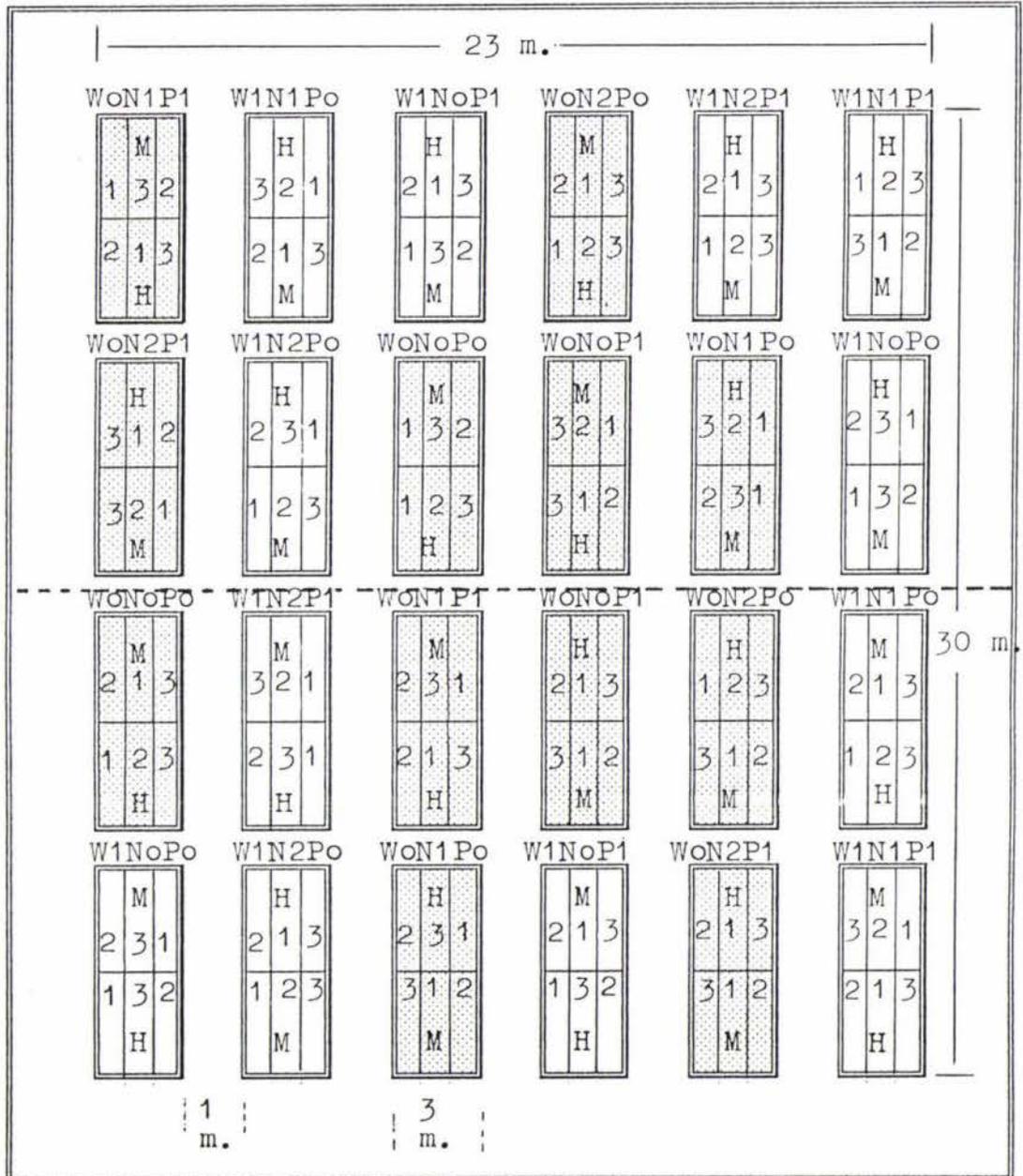
3.2. EXPERIMENTAL DESIGN

The experiment was a randomised , split plot design with 2 block replicates. The 3 main treatments - 2 cultivars, 3 nitrogen levels and 2 phosphorus levels - were in a randomised complete block design with 2 replicates and each main treatment was split into 6 sub-plots for 3 harvest times and 2 harvest methods.

Individual plot size was 3 m x 6 m giving a total of 144 plots.

All data collected was statistically analyzed by an analysis of variance.

3.3. THE FIELD LAY-OUT FOR EXPERIMENT



Wo=smooth-seeded pea(cv.Maple)
 W1=wrinkled-seeded pea(cv.Pania)
 No=0 kg N/ha.
 N1=100 kg N/ha.
 N2=200 kg N/ha.
 Po=0 kg superphosphate/ha.
 P1=250 kg superphosphate/ha.
 H= Hand-shelling
 M= Threshing machine(combine harvester)
 1= 35% SMC
 2= 25% SMC
 3= 15% SMC

3.4. MATERIALS AND METHODS

3.4.1. Experimental Field

The experiment was located on the Frewens Block of Massey University Palmerston North, New Zealand. The soil classification is a Manawatu sandy loam.

3.4.2. Land Preparation

The land was prepared by ploughing (mid-October, 1994) and then by harrowing (the first harrowing was done in late-October, 1994) twice to provide a good tilth. Before the last harrowing, 250 kg/ha superphosphate was applied (27 November 1994) to the appropriate plots. Because of the high mobilization and easy loss of nitrogen from the soil by volatilization and especially by leaching (Haynes, 1986), an adequate supply of nitrogen to the plant during flowering and pod set was ensured by applying 50% of nitrogen immediately prior to the last harrowing (27 November 1994), as for superphosphate, and the second 50% at the pre-flowering stage (12 February 1995).

3.4.3. Seed Lots

Seeds of two commercial cultivars, cv. Pania (wrinkled-seeded pea) and cv. Maple (smooth-seeded pea) were sown. The quality of these seeds in terms of germination and vigour (conductivity, accelerated ageing and hollow heart) was determined before sowing.

3.4.4. Time and Method of Sowing

Both cultivars were sown on 29 November 1994 with a cone seeder adjusted to a depth of 3 cm with the coulters 20 cm apart and with a row width of 5 cm.

3.4.5. Crop Management

Trifluralin herbicide was applied on 25 November 1994 with a knapsack sprayer at the rate of 2.5 kg a.i./ha (in 200 litres of water/ha.) to control weeds before sowing. Hand weeding was also carried out as required to remove weeds not controlled by the Trifluralin herbicide.

To control powdery mildew, nitrile (Bravo) fungicide was sprayed at the rate of 1 kg a.i./ha 40 days after sowing. Mancozeb (Dithane M -45) was also sprayed at the rate of 1.6 kg a.i./ha at 55 and 70 days after sowing, to further control powdery mildew. The crop was inspected regularly for other pest and disease problems.

At the pod filling stage, the peas were sprayed with Methiocarb at the rate of 0.075 kg a.i./ha (1 kg in 200 litres of water/ha) to repel birds.

3.4.6. Seed Moisture Content Determination

Seed moisture content was monitored after physiological maturity, approximately 35 days after anthesis (Flinn and Pate, 1968). All pods from five plants from each treatment were collected at random every 3 days up to the last harvest in order to obtain the required harvesting times of 35%, 25%, and 15% SMC. Pods were hand shelled and seeds then sampled to determine actual seed moisture content. When the required SMC was reached, the required number of whole plants were then harvested for measurements and processing.

3.4.7. Time of Harvest

Every treatment was hand-harvested according to seed moisture content as stated above. The first harvest was made at 35% SMC date; the second harvest at 25% SMC date and the third harvest at 15% SMC date. For each harvest, 120 plants were randomly collected from the plots (refer to the field lay-out), excluding border rows, by cutting at the base of plants and then placing in hessian bags.

3.4.8. Method of Harvest

For hand-shelling, all pods from 40 of the 120 plants (the rest were used for pod position determination) from each sub-plot were used to determine the number of pods/plant, seeds/pod and 1000-seed weight (adjusted to 12% SMC). Potential seed yield was calculated from the number of pods/plant, number of seeds/pod, the average seed weight and number of plants/unit area. The shelled seeds were then bulked for seed quality determination.

For machine-threshing, 120 plants were sampled per plot and threshed by a combine harvester (Seed Master Universal Hydrostatic small plot combine harvester) at a drum speed of 1,350 rpm. with a rhomboid threshing concave set at 8 cm.

After threshing, seeds were immediately dried to a safe moisture content (below 12%) at ambient air temperature, by laying the seeds in plastic trays in the glasshouse, and turning the seeds regularly. After drying (about 5-8 days, depends on the initial SMC and ambient air conditions), all seeds from each treatment were put into a plastic bag and placed in sealed plastic buckets at 5°C until the evaluation of seed quality.

Before placing at 5°C, seeds from the threshing machine were cleaned by hand.

3.4.9. Pod Position Determination

Eighty of the harvested 120 plants (forty plants had previously been used for hand-shelling), were used to study the effect of pod position on seed quality.

The harvested pods from each plant were divided into top, middle and bottom pods; pods at the distal end were regarded as top pods, pods at the first podding truss as bottom pods and remaining pods as middle pods. Seeds from the same pod position were bulked together and dried to 12% SMC at ambient air temperature as described above.

The incidence of hollow heart, germination percentage and conductivity measurement were determined for each plot and each pod position.

3.5. DETERMINATION OF SEED QUALITY

3.5.1. Seed Moisture Content Test

Using the constant temperature oven method (ISTA, 1993), two replicates of 10 gm seed were ground, placed in weighed containers and dried at 130°C for 1 hour. The moisture content of the seed was calculated using the formula:

$$\frac{(M2 - M3) \times 100}{M2 - M1}$$

where:

M1- is the weight in grams of the container and cover.

M2- is the weight in grams of the container, cover, and contents before drying.

M3- is the weight in grams of the container, cover, and contents after drying.

3.5.2. Germination Test

A standard laboratory germination test (ISTA, 1993) was conducted. Two hundred pure seeds (4x50) were germinated between paper at 20°C for 8 days. Tests were then evaluated and classified into normal or abnormal seedling and dead seeds using the descriptions prescribed by ISTA (1993). The data were averaged and expressed as a percentage.

3.5.3. Conductivity Test

Conductivity testing was carried out according to Hampton (1995), using four replicates of 50 seeds drawn from the pure seed fraction of each plot. Each replicate was weighed before being placed in a 500 ml conical flask and 250 ml distilled water at 20°C was then added to each flask. Four flasks containing 250 ml of distilled water alone were used as control. All flasks were covered with parafilm to reduce evaporation, and then placed in a controlled temperature room at a constant temperature of 20°C. After 24 hours of incubation the conductivity was measured by

a conductivity meter(CDM -83 Radiometer) without filtration. The reading from the distilled water was subtracted from the reading obtained from the solution containing the seeds. The conductivity results were expressed in microsiemens per cm per gram of seed($\mu\text{Scm}^{-1}\text{g}^{-1}$).

3.5.4. Hollow Heart Test

The incidence of hollow heart was determined using the normal seedlings from the germination test. Normal seedlings were dissected by separating the cotyledons by hand to expose the adaxial surface. The presence of a depression in either or both of the cotyledons was considered to be evidence of hollow heart. The data obtained were expressed as a percentage based on the total number of seeds evaluated in the germination test.

3.5.5. Accelerated Ageing Test

According to the method of Hampton(1995) 60 g of seed from each plot was used. Forty ml distilled water was placed in a 11x11x3.5cm plastic box. A 10x10x3cm wire-mesh screen was put into the box, ensuring that the screen was above water level. The seeds were then placed in a single layer on the wire screen. Lids were placed on the boxes, which were then placed on the upper and middle shelves of a germinator, at 40°C for 72h. After aging, 4 replicates of 50 aged seeds were germinated under the conditions recommended for the germination test. Seed moisture content after ageing was also measured. The results were compared with the results of the germination test of unaged seeds.

3.5.6. Thousand Seed Weight

The thousand seed weight(TSW) of each plot was determined by taking 8 replicates of 100 seeds at random and weighing. The mean weight of the eight replicates was then used to determine TSW, adjusted to 12% SMC (ISTA,1993).

3.5.7. Ferric Chloride Test

To determine the percentage of mechanically damaged seeds, the Ferric Chloride Test was carried out. Using the method described by Duangpathra(1986), two replicates of 100 seeds were soaked in 20% ferric chloride solution for 25 minutes. The damaged seeds were determined by counting the black stained seeds, and the results were expressed as a percentage.

CHAPTER 4.

RESULTS

4.1. SEED YIELD AND YIELD COMPONENTS

4.1.1. Seed Yield

In terms of main effects, overall seed yield was significantly affected by cultivar (Table 1., Pania > Maple); by seed moisture content (SMC) at threshing (Table 1., 25% SMC > 35% SMC > 15% SMC) and by threshing method (Table 2., hand-shelling > machine-threshing).

More importantly, however, cultivar responses interacted with both seed moisture content at threshing (Table 1.) and with threshing method (Table 2.) and was highly significant. For example, although the superiority of threshing at 25% SMC was significant as a main effect, when statistically separated according to cultivar the analyses showed that cv. Pania produced the highest and similar seed yields at both 15% and 25% SMC threshings with a significant decline at the 35% SMC threshing. In contrast, cv. Maple produced significantly lower seed yield at the 15% SMC threshing but a significant increase at the 25% and 35% SMC threshings.

Table 1 Interaction effects of cultivar and seed moisture content on seed yield.

Cultivar	Mean yield (kg/ha) at different seed moisture			Means
	15	25	35	
Maple	3,852 ^{d**}	5,036 ^c	5,105 ^c	4,664 ^b
Pania	5,665 ^a	5,690 ^a	5,425 ^b	5,593 ^a
LSD (P < 0.05) = 90				
Means	4,758 ^c	5,363 ^a	5,264 ^b	
LSD (P < 0.05) = 64				LSD (P < 0.05) = 40

* Results are means of 24 observations adjusted to 12% SMC.

** Results followed by the same letter are not significantly different.

Similarly, the effect of hand-shelling or machine-threshing on seed yield was also greatly influenced by cultivar. As shown in Table 2, cv. Pania showed a less severe effect from machine-threshing than cv. Maple when compared with hand-shelling(10% and 28%, respectively).

Table 2. Interaction effects of cultivar and threshing method on seed yield.

Cultivar	Mean yield(kg/ha) at different threshing methods*	
	Hand	Machine
Maple	5,395b**	3,933c
Pania	5,888a	5,299b
LSD(P < 0.05) = 104		
Means	5,642a	4,616b
LSD(P < 0.05) = 73		

* Results are means of 36 observations adjusted to 12% SMC.

** Results followed by the same letter are not significantly different.

Interaction between threshing method and seed moisture content at threshing was highly significant. As shown in Table 3., seed yield from hand-shelling at different seed moisture contents showed no significant differences, while machine-threshing had highly significant effects on seed yield with machine- threshing at 25%SMC giving the highest seed yield(5,106 kg/ha), and threshing at 15%SMC resulting in the lowest seed yield(3,866 kg/ha).

Table 3. Interaction effects of seed moisture content at threshing and threshing method on seed yield.

Seed moisture content (%)	Mean yield(kg/ha) at different threshing methods*	
	Hand	Machine
15	5,651 ^a **	3,866 ^d
25	5,621 ^a	5,106 ^b
35	5,653 ^a	4,876 ^c
LSD(P < 0.05)= 127		

* Results are means of 24 observations adjusted to 12%SMC.

** Results followed by the same letter are not significantly different.

Nitrogen application rate also had a highly significant interactive effect with threshing method on seed yield (Table 4.). Although, in both hand-shelling and machine-threshing, seed yield increased with increasing rates of nitrogen application, it is suggested that the positive interaction recorded is possibly due to differences in the percentage increase in yield from 100 to 200 kg N/ha, which was approximately 10% in hand-shelling versus 5% in machine-threshing, reflecting the relatively high level of damage in the 200 kg N/ha treatment.

Table 4. Interaction effects of nitrogen application and threshing method on seed yield.

N-level (kg N/ha)	Mean yield(kg/ha) at different threshing methods*	
	Hand	Machine
0	5,324 ^{c**}	4,363 ^f
100	5,578 ^b	4,631 ^e
200	6,024 ^a	4,854 ^d
LSD(P < 0.05) = 127		

* Results are means of 24 observations adjusted to 12%SMC.

** Results followed by the same letter are not significantly different.

Although nitrogen had no significant effect on overall seed yield, the rate of nitrogen applied had a highly significant effect on seed yield when combined with interactions between cultivar and seed moisture content at threshing. As shown in Table 5., in both cultivars, seed yield tended to increase with increasing nitrogen supply at all seed moisture contents. In cv. Maple, the highest seed yield(5,416 kg/ha) was obtained when seed was threshed at 35%SMC, combined with 200 kg N/ha. Threshing at 15%SMC combined with no nitrogen gave the lowest seed yield(3,558 kg/ha). The highest seed yields in cv. Pania(6,091 and 6,119 kg/ha) were obtained from the 200 kg N/ha application when threshed at 15% or 25% SMC, while the lowest yield(5,188kg/ha) was obtained from threshing at 35%SMC with no nitrogen application.

Table 5. Interaction effects of cultivar, addition of nitrogen and seed moisture content on seed yield.

Cultivar	N-level (kg/ha)	Mean yield(kg/ha) at different SMC*		
		15%	25%	35%
Maple	0	3,558 ⁱ **	4,694 ^g	4,769 ^g
	100	3,961 ^h	5,185 ^f	5,128 ^f
	200	4,036 ^h	5,230 ^{ef}	5,416 ^{cd}
Pania	0	5,368 ^{de}	5,483 ^{cd}	5,188 ^f
	100	5,537 ^c	5,469 ^{cd}	5,346 ^{de}
	200	6,091 ^a	6,119 ^a	5,742 ^b
LSD(P < 0.05) = 156				

* Results are means of 8 observations adjusted to 12%SMC.

** Results followed by the same letter are not significantly different.

Table 6 shows a highly significant interactive effect of seed moisture content at threshing on seed yield when combined with cultivar and threshing method. In

hand-shelling, neither cv. Maple nor cv. Pania seed yield was affected by seed moisture content at threshing, while in machine-threshing the effect of seed moisture content at threshing on seed yield was highly significant in both cultivars. The highest seed yield of cv. Pania(5,581kg/ha) threshed by machine was obtained from 25%SMC at threshing, while the lowest seed yield(4,907kg/ha) was obtained from the 35%SMC threshing. In cv. Maple, machine-threshing at 35%SMC provided the highest seed yield(4,845 kg/ha), whereas the lowest seed yield(2,323 kg/ha) was obtained from threshing at 15%SMC.

Table 6. Interaction effects of cultivars, seed moisture content at threshing and threshing method on seed yield.

Cultivar	SMC(%)	Mean yield(kg/ha) at different threshing methods*	
		Hand	Machine
Maple	15	5,380c**	2,323f
	25	5,442bc	4,631e
	35	5,364c	4,845d
Pania	15	5,922a	5,408bc
	25	5,800a	5,581b
	35	5,943a	4,907d
LSD(P < 0.05) = 180			

* Results are means of 12 observations adjusted to 12%SMC.

** Results followed by the same letter are not significantly different.

4.1.2. Yield Components

4.1.2.1. Number of pods/plant and seeds/pod

Seed cultivar had a significant effect on the number of pods/plant and seeds/pod, with cv. Pania producing the greater number of seeds/pod, but fewer pods/plant than cv. Maple (Table 7).

Nitrogen application only affected number of pods/plant, increasing nitrogen-levels tending to result in greater pod production (Table 7.).

Table 7. Effect of cultivar and nitrogen-level on number of pods/plant and seeds/pod.

Factors		Pods/plant	Seeds/pod
Cultivar	Pania	5.19 ^{b*}	5.76 ^a
	Maple	6.60 ^a	3.57 ^b
	LSD(P<0.05)	0.45	0.36
N-level	0	5.51 ^b	4.54
	100	6.03 ^{ab}	4.78
	200	6.15 ^a	4.68
	LSD(P<0.05)	0.55	NS

* Results followed by the same letter are not significantly different.

4.1.2.2. Seed weight(1000-seed weight)

Overall seed weight was significantly affected by cultivar (Table 8., Maple > Pania); by nitrogen fertilizer rate (Table 9., 100 kg N/ha < 0 or 200 kg N/ha); and by threshing method (Table 10, hand-shelling > machine-threshing).

Although seed moisture content at threshing alone had no main effect on seed weight, when considered separately for each cultivar the effect was highly significant. In Table 8., for example, cv. Maple seeds threshed at 35% and 25% SMC had a similar seed weight and higher than those threshed at 15% SMC, whereas in cv. Pania the lowest seed weight was obtained from threshing at 25% SMC and the higher seed weight from threshing at 15% and 35% SMC. The latter two threshing times showed no significant difference in seed weight.

Table 8 also shows that cv. Maple generally produced seeds with a higher individual seed weight than cv. Pania, particularly when threshed at 25% and 35% SMC. Nevertheless, within each cultivar the effect of time of harvest(15%, 25%, 35%SMC) on thousand-seed weight was relatively small(1.8-3.1% in Pania and Maple, respectively).

Table 8. Interaction effects of cultivar and seed moisture content at threshing on seed weight.

Cultivar	1000-seed weight(gm) at different seed moisture contents.			Means
	15%	25%	35%	
Maple	248.99 ^{b..}	257.82 ^a	255.58 ^a	254.13 ^a
Pania	245.03 ^c	240.84 ^d	242.98 ^{cd}	242.95 ^b
LSD(P < 0.05) = 3.89				
Means	247.01	249.33	249.28	LSD(<0.05)=3.75
NS				

* Results are means of 24 observations adjusted to 12%SMC.

** Results followed by the same letter are not significantly different.

While machine-threshing decreased seed weight in both cultivars compared with hand-shelling (Table 9.), the degree was again noticeably greater in cv. Maple (5%) than in cv. Pania (2%). It is interesting to note that the highest seed weight was recorded in the hand-shelled Maple, while the lowest seed weight was recorded in machine-threshed Pania.

Table 9. Interaction effects of cultivar and threshing method on seed weight.

Cultivar	1000-seed weight(gm) in different threshing methods.	
	Hand..	Machine
Maple	259.94 a	248.32 b
Pania	245.37 b	240.52 c
LSD(P < 0.05) = 4.45		
Means	252.66 a	244.43 b
LSD(P < 0.05) = 3.14		

* Results are means of 36 observations adjusted to 12% seed moisture content.

**Results followed by the same letter are not significantly different.

With hand-shelling there was no significant effect of different seed moisture contents at shelling on the seed weight of either cv. Maple or cv. Pania. However, with machine-threshing the seed weight of Maple harvested at 15%SMC was significantly lower than that obtained following seed threshings at 25% or 35%SMC. Pania, however, showed no significant effect of seed moisture content on seed weight(Table 10.).

Table 10. Interaction effects of cultivar, seed moisture content and threshing method on seed weight.

Cultivar	Seed moisture content(%)	1000-seed weight(gm) in different threshing methods.	
		Hand	Machine
Maple	15	259.13ab..	238.86e
	25	262.31a	253.32bc
	35	258.37ab	252.78bc
Pania	15	246.89cd	243.16de
	25	241.70de	239.98de
	35	247.52cd	238.44e
LSD(0.05) = 7.70			

* Results are means of 12 observations adjusted to 12%SMC.

** Results followed by the same letter are not significantly different.

As shown in Table 11., when nitrogen was applied without added phosphorus there was little difference in seed weight between the different rates of nitrogen application. However, when applied with the addition of phosphorus there was a significant drop in seed weight at the 100 kg N/ha level followed by an increase in seed weight at the 200 kg N/ha level which was not significantly different from the no nitrogen treatment.

Table 11. Interaction effects of nitrogen and phosphorus applications on seed weight.

Nitrogen level (kg /ha)	1000-seed weight(gm) at different phosphorus levels (kg superphosphate/ha).		Means
	0	250	
0	247.44 ^{bc..}	255.21 ^a	251.33 ^a
100	246.25 ^c	239.55 ^d	242.90 ^b
200	253.36 ^{ab}	249.43 ^{abc}	251.40 ^a
LSD(0.05) = 6.49			LSD(P<0.05)=4.59

* Results are means of 24 observations adjusted to 12%SMC.

** Results followed by the same letter are not significantly different.

4.2. HARVESTING AND MECHANICAL DAMAGE

4.2.1. Seed Damage

The seed damage results presented in this section refer to physical damage which could be visually detected, and is expressed on a weight basis(at 12%SMC). The degree of damage ranged from a small cracks or bruises on the seed coat to severely shattered or bruised seed. The characteristic extent of seed damage depended on the damage factors involved, as shown in Appendix I.

Seed damage was significantly affected by cultivar(Table 12., Maple>Pania); by seed moisture content at threshing(Table 12., 15%SMC > 35%SMC > 25%SMC) and by threshing method(Table 13., machine-threshing > hand-shelling).

However, the effect of seed moisture content on seed damage significantly interacted with cultivar. As shown in Table 12., Maple seed was most severely damaged(29.84%) when threshed at 15%SMC, but showed much less and similar damage when threshed at 25% or 35%SMC. In contrast, cv. Pania generally showed less damage than cv. Maple at the different seed moisture contents except when threshed at 35% SMC where damage was significantly greater than in cv. Maple.

Table 12. Interaction effects of cultivar and seed moisture content at threshing on seed damage.

Cultivar	Seed damage(%) following threshing at			Means
	15%SMC	25%SMC	35%SMC	
Maple	29.84 ^{**}	7.15 ^c	5.37 ^{cd}	14.12 ^a
Pania	6.56 ^c	3.52 ^d	10.81 ^b	6.96 ^b
LSD(P< 0.05) = 2.06				
Means	18.2 ^a	5.33 ^c	8.08 ^b	LSD(P<0.05)=1.90
LSD(P < 0.05)= 1.45				

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

The effect of threshing method on seed damage was also significantly influenced by cultivar. As shown in Table 13., machine-threshing had a much greater damaging effect on cv. Maple than on cv. Pania(27.98% and 10.32%, respectively). In contrast, cv. Pania was damaged to a greater extent than cv. Maple when hand-shelled. However, this difference in seed damage recorded between cultivars during hand-shelling -as reflected mainly in the cleavage of the seed coat- is likely to have occurred prior to hand-shelling since such damage would be unlikely to occur during a careful hand- shelling process.

Table 13. Interaction effects of cultivar and threshing method on seed damage.

Cultivar	Seed damage(%) following different threshing methods.*	
	Hand	Machine
Maple	0.27 <i>d</i> **	27.98 <i>a</i>
Pania	3.60 <i>c</i>	10.32 <i>b</i>
	LSD(P < 0.05) = 1.84	
Means	1.93 <i>b</i>	19.15 <i>a</i>
	LSD(P < 0.05) = 1.30	

* Results are means of 36 observations.

** Results followed by the same letter are not significantly different.

The effect of threshing method had a significant interactive effect with seed moisture content at threshing. There were no differences in seed damage when the seed was shelled by hand at different seed moisture contents, but when machine threshed seed damage was highly affected by seed moisture content. Machine threshing at 15%SMC showed the greatest seed damage(34.24%), whereas at 25%SMC the lowest degree of seed damage(9.11%) was recorded, with the seed threshed at 35%SMC being intermediate in terms of seed damage(Table 14.).

Table 14. Interaction effects of seed moisture content at threshing and threshing method on seed damage.

Threshing methods	Seed damage(%) at different seed moisture contents *		
	15%SMC	25%SMC	35%SMC
Hand	2.17 <i>d</i> **	1.56 <i>d</i>	2.07 <i>d</i>
Machine	34.24 <i>a</i>	9.11 <i>c</i>	14.10 <i>b</i>
LSD(P < 0.05) = 2.26			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

The interaction effect between cultivar and seed moisture content on seed damage was also influenced by nitrogen application level(Table 15.).

In cv. Maple, irrespective of nitrogen application levels, the greatest degree of seed damage occurred during threshing at 15%SMC, but no significant differences in seed damage were recorded in seeds threshed at 25% or 35%SMC.

However, in cv. Pania, the effect of seed moisture content at harvest on seed damage was highly affected by additional nitrogen levels. In treatments with no applied nitrogen, the highest degree of seed damage(10.95%) occurred from threshing at 35%SMC, but seed damage was not significantly different when seed was threshed at 25% or 15%SMC. In the 100 kg N/ha treatments, the different seed moisture content at threshing had no significant effect on seed damage, but in the 200 kg N/ha

treatments degree of seed damage was significantly influenced by seed moisture content, i.e. threshing at 35%SMC resulted in the highest degree of seed damage, whereas threshing at 25%SMC provided the least seed damage.

Table 15. Interaction effects of cultivar, seed moisture content at threshing and addition of nitrogen on seed damage.

Cultivar	SMC at threshing (%)	Seed damage(%) at different N-levels(kg/ha)*		
		0	100	200
Maple				
	15	28.54a**	29.70a	31.31a
	25	7.19de	6.13defg	8.15cd
	35	4.11efg	6.83def	5.16defg
Pania				
	15	6.72def	5.56defg	7.41cde
	25	3.48fg	3.98efg	3.08fg
	35	10.95bc	7.42cde	14.05b
LSD(P < 0.05) = 3.56				

* Results are means of 8 observations.

** Results followed by the same letter are not significantly different.

The interaction effect between cultivar and seed moisture content on seed damage was also influenced by threshing method, as shown in Table 16. When hand shelled, seed moisture content at threshing had no significant effect on seed damage in either cultivar, whereas following machine-threshing the interaction effect between cultivar and seed moisture content was significant. In cv. Maple, machine threshing at 15%SMC produced seeds with a very high degree of seed damage(59.17%), while the least seed damage(10.52%) occurred at 35%SMC. In contrast, machine threshing

of cv. Pania at 35%SMC resulted in the highest level of seed damage(17.69%), whereas the lowest level of seed damage(3.97%) was obtained from seeds threshed at 25%SMC.

Table 16. Interaction effects of cultivar, seed moisture content at threshing and threshing method on seed damage.

Cultivar	SMC (%)	Seed damage(%) following different threshing methods*	
		Hand	Machine
Maple	15	0.53 ^{f*}	59.17 ^a
	25	0.06 ^f	14.25 ^c
	35	0.22 ^f	10.52 ^d
Pania	15	3.82 ^e	9.3 ^d
	25	3.06 ^{ef}	3.97 ^e
	35	3.92 ^e	17.69 ^b
LSD(P < 0.05) = 3.19			

* Results are means of 12 observations.

** Results followed by the same letter are not significantly different.

4.2.2. Ferric Chloride Test

The purpose of the Ferric Chloride Test is to detect physical damage on seed, in particular, minor cracks in the seed coat which normally cannot be obviously detected visually. Results presented in this section relate to the percentage of pure seed showing damage, following soaking in 20% Ferric Chloride for 25 minutes at room temperature.

Overall seed damage as detected by the Ferric Chloride technique was significantly influenced by seed cultivar(Table 17., Pania > Maple); by seed moisture

content at threshing (Table 17., 15%SMC > 35%SMC > 25%SMC) and by threshing method (Table 18., machine-threshing > hand-shelling).

When the cultivars are considered separately according to seed moisture content at threshing, the level of seed damage recorded is quite different. In cv. Pania, the highest percentage of seed damage (22.75%) occurred following threshing at 35%SMC, but at 15%SMC in cv. Maple (17.08%). However, the lowest percentage of seed damage in cv. Pania was recorded at 25%SMC, but was equally low at 35% and 25%SMC in cv. Maple (Table 17.). Again, however, the results clearly show the greater threshing damage susceptibility of cv. Pania compared with cv. Maple.

Table 17. Interaction effects of cultivar and seed moisture content at threshing on seed damage.

Cultivar	% Seed damage at different seed moisture contents*			Means
	15%	25%	35%	
Pania	19.54b**	9.13c	22.75a	17.14a
Maple	17.08b	5.58d	6.37cd	9.68b
LSD(P < 0.05) = 3.05				
Means	18.31a	7.35c	14.56b	LSD(P < 0.05) = 4.41
LSD(P < 0.05) = 2.15				

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

Although machine-threshing in both cultivars produced a higher percentage of damaged seed than hand-shelling, the degree of damage in each cultivar was markedly different. For example, machine-threshing Pania increased the percentage of seed damage to a much greater extent than cv. Maple (Table 18).

Table 18. Interaction effects of cultivar and threshing method on seed damage.

Cultivar	% Seed damage following different threshing methods*	
	Hand	Machine
Pania	3.64c**	30.69a
Maple	0.17c	19.19b
	LSD(P < 0.05) = 3.62	
Means	1.90b	24.92a
	LSD(P < 0.05) = 2.56	

* Results are means of 36 observations.

** Results followed by the same letter are not significantly different.

When seeds were threshed at different seed moisture contents (Table 19.), there were no significant differences in seed damage for the hand-shelled seeds, but the damage was significantly different in machine-threshed seeds. Machine-threshing at 15%SMC resulted in the highest percentage of seed damage (34.58%), the lowest at 25%SMC (13.20%) and intermediate at 35%SMC (26.96%).

Table 19. Interaction effects of threshing method and seed moisture content at threshing on seed damage.

Threshing method	% Seed damage at different seed moisture contents*		
	15%	25%	35%
Hand	2.04 ^{d**}	1.5 ^d	2.17 ^d
Machine	34.58 ^a	13.20 ^c	26.96 ^b
LSD(P < 0.05) = 4.44			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

Interaction effects between seed moisture content at threshing and threshing methods on seed damage was influenced by cultivar(table 20.). Hand-shelling at different seed moisture contents did not affect seed damage significantly in either cultivar, but the percentage of damaged seeds in machine-threshing was much greater and significantly influenced by seed moisture content.

In cv. Pania, there was no significant difference in seed damage between seeds machine-threshed at 35% and 15%SMC, whereas in cv. Maple, machine-threshing at 15%SMC resulted in a much higher percentage of seed damage than at 25% or 35%SMC(34%, 11% and 12.58%, respectively). However, in both cultivars, machine-threshing at 25%SMC produced seeds with similarly lower levels of damage.

Table 20. Interaction effects of cultivar, threshing method and seed moisture contents at threshing on seed damage.

Cultivar	Threshing method	% Damage at different SMC*		
		35%	25%	15%
Pania	Hand	4.17 ^{d**}	2.83 ^d	3.92 ^d
	Machine	41.33 ^a	15.42 ^c	35.17 ^{ab}
Maple	Hand	0.17 ^d	0.17 ^d	0.17 ^d
	Machine	12.58 ^c	11.00 ^c	34.00 ^b
LSD(P < 0.05) = 6.27				

* Results are means of 12 observations.

** Results followed by the same letter are not significantly different.

4.3. SEED QUALITY

4.3.1. Seed Germination

Seed quality as measured by germination percentage was significantly influenced by cultivar (Table 21., Maple > Pania); by seed moisture content at threshing (Table 22., 15% > 35% > 25%SMC) and by threshing method (Table 23.).

Although, as shown in Table 21., there was a significant interaction between cultivar and nitrogen level, with Pania receiving 100 kg N/ha showing a significant depression in germination percentage, this difference was small and of little practical significance.

Table 21. Interaction effect of cultivar and addition of nitrogen on seed germination.

Cultivars	Seed germination(%) at different N levels(kg/ha)*			Means
	0	100	200	
Maple	89a**	92a	91a	91a
Pania	83b	80c	85b	83b
LSD(P <0.05) = 3.3				
Means	86	86	88	
NS				LSD(P<0.05)=1.9

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

It must be noted that for cv. Pania only, the intended harvesting time of 25%SMC was not achieved owing to heavy rain during harvesting, resulting in actual seed moisture content being about 35%SMC. The results of seed quality

measurements of this treatment must be considered with caution particularly for the machine-threshed seed.

The interactive effects between cultivar and seed moisture content at threshing on seed germination percentage are shown in Table 22. Germination of Maple seeds threshed at 15% and 25%SMC were not significantly different(94% and 93%, respectively), but both were significantly higher than the seed threshed at 35%SMC(85%). In cv. Pania, germination percentage of the seeds threshed at 15% and 35%SMC were the same(90%), whereas threshing at 25%SMC seed germination was markedly decreased(70%)- see note above.

Table 22. Interaction effect of cultivar and seed moisture content at threshing on seed germination.

Cultivar	Seed germination(%) at different seed moisture contents*		
	15%	25%	35%
Maple	94a**	93a	85c
Pania	90b	70d	90b
	LSD(P < 0.05) = 2.6		
Means	92a	81c	87b
	LSD(P < 0.05) = 1.9		

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

The interactive effects between cultivar and threshing methods on seed germination were highly significant. As shown in Table 23, there were no significant differences in germination of hand-shelled seeds between the two cultivars, while in machine-threshing the germination of cv. Pania suffered to a much greater extent than cv. Maple(69% and 82%, respectively).

Table 23. Interaction effect between cultivar and threshing method on seed germination.

Cultivar	Seed germination(%) in different threshing methods.*	
	Hand	Machine
Maple	99a**	82b
Pania	97a	69c
	LSD(P < 0.05) = 2.1	
Means	98a	75b
	LSD(P < 0.05) = 1.2	

* Results are means of 36 observations.

** Results followed by the same letter are not significantly different.

Data in Table 24. shows no significant difference in germination percentage when seeds were hand-shelled at different seed moisture contents. However, following machine-threshing the effect of seed moisture content at harvest on seed germination was highly significant. Machine-threshing at 15% produced seeds with the highest germination(86%), whereas the lowest germination percentage(64%) was recorded at the questionable level of 25%SMC.

Table 24. Interaction effects of threshing method and seed moisture content at threshing on seed germination.

Harvest methods	Seed germination(%) at different seed moisture contents*		
	15%	25%	35%
Hand	97a**	98a	99a
Machine	86b	64d	76c
LSD(P < 0.05) = 2.6			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

Interaction effects between cultivar and threshing method on seed germination, as previously described, were also significantly influenced by nitrogen application in the field. As shown in Table 25, hand-shelling had no significant effect on germination percentage of either cv. Maple or cv. Pania at all nitrogen levels, but with machine-threshing germination percentage of cv. Maple was depressed to the greatest extent in the no nitrogen treatment, while cv. Pania showed the greatest depression at 100 kg N/ha.

Table 25. Interaction effects of cultivar, threshing method and seed moisture content at threshing on seed germination.

Cultivar	N-levels(kg/ha)	Seed germination(%) in different threshing methods*	
		Hand	Machine
Maple	0	99a**	79c
	100	99a	84b
	200	99a	83b
Pania	0	97a	71d
	100	97a	64e
	200	98a	71d
LSD(P < 0.05) = 3.7			

* Results are means of 12 observations.

** Results followed by the same letter are not significantly different.

As shown in Table 26, interaction effects between cultivar and harvest method on seed germination were also significantly affected by seed moisture content at threshing.

Germination percentage of machine-threshed Maple seeds increased when seed moisture content at threshing decreased(germination percentage of seeds

harvested at 35%, 25% and 15%SMC were 71%, 85% and 90%, respectively). However, in cv. Pania, machine-threshing at 15% or 35%SMC showed no significant difference in seed germination(82% and 81%, respectively), although machine threshing at the questionable of 25%SMC resulted in a significant drop in germination percentage(42%).

Table 26. Interaction effects of cultivar, threshing method and seed moisture content at threshing on seed germination.

Cultivars	Seed moisture contents(%)	Seed germination(%) in different harvest methods*	
		Hand	Machine
Maple	15	98a**	90b
	25	100a	85c
	35	99a	71e
Pania	15	97a	82cd
	25	97a	42f
	35	98a	81d
LSD(P < 0.05) = 3.7			

* Results are means of 12 observations.

** Results followed by the same letter are not significantly different.

The interactive effects between cultivar, threshing method, nitrogen application rate and seed moisture content at threshing on seed germination were highly significant. The following explanations refer to Table 27.

Germination of hand-shelled seeds was not significantly affected by the interactive effects between cultivar, additional nitrogen and seed moisture content at shelling, and germination of both seed cultivars was not significantly different (95-100%).

However, for machine-threshed seeds of both cultivars, seed germination was highly influenced by the interactive effects between seed moisture content at threshing and nitrogen application rates. For example in cv. Maple, for the machine-threshing at 15%SMC, different rates of nitrogen application did not affect seed germination, but machine-threshing at 25% or 35%SMC, tended to increase seed germination as nitrogen application rates increased.

In machine-threshed Pania seeds, different rates of nitrogen applied did not affect germination of seed threshed at 35%SMC, but nitrogen application rates significantly affected seed germination when the seeds were threshed at 15% or 25% SMC. For example, when threshing seed at 15%SMC the application of 100 kg N/ha significantly depressed germination percentage when compared with the application of 200 kg N/ha, but not when compared with the no-nitrogen treatment. However, when threshing seed at so-called 25%SMC the depression in germination percentage from applying 100kg N/ha did reach significance compared with both the control and 200kg N/ha treatments.

Table 27. Interaction effects between cultivar, threshing method, nitrogen application rate and seed moisture content at threshing on seed germination.

Cultivar	Threshing methods	N-levels (kg/ha)	Seed germination(%) at different seed moisture contents.*		
			15%	25%	35%
Maple	Hand	0	98a**	100a	99a
		100	98a	100a	100a
		200	99a	100a	100a
	Machine	0	91bc	81de	66h
		100	90bc	90bc	72gh
		200	90bc	86cd	74fg
Pania	Hand	0	96ab	95ab	99a
		100	96ab	98a	98a
		200	98a	99a	98a
	Machine	0	83de	49i	81de
		100	79ef	32j	81de
		200	86cd	47i	82de
LSD(P < 0.05) = 6.4					

* Results are means of 4 observations.

** Results followed by the same letter are not significantly different.

4.3.2. Accelerated Ageing Test

The effect of accelerated ageing(AA) on seed germination is considered to be important in ranking seed samples in order of 'vigour'. In this study the use of the

AA-test method resulted in the detection of seed vigour differences between cultivars, level of N&P, seed moisture content at threshing and following threshing. In terms of main effects, seed vigour was significantly influenced by cultivar (Table 28., Maple > Pania); by addition of nitrogen (Table 28.), where seed vigour increased with increasing nitrogen application rates; by seed moisture content at threshing (Table 29.), where seed threshed at 15%SMC showed the highest level of seed vigour, while seed threshed at 25%SMC or 35%SMC produced seed with similar vigour; by threshing method (Table 30., hand-harvesting > machine-harvesting) and by the addition of phosphorus (Table 32) where application of phosphorus increased seed vigour.

Although overall, seed vigour was positively correlated with the rate of nitrogen application, when individually considered for each cultivar, results were significantly different. Data presented in Table 28. shows that the addition of nitrogen did not affect seed vigour in cv. Maple, but significantly affected seed vigour in Pania, where seed vigour of cv. Pania increased as the nitrogen application rate increased.

Table 28. Interaction effects of cultivar and addition of nitrogen on seed germination after accelerated ageing.

Cultivars	% Germination after AA of seed samples			Means
	0	100	200	
Maple	83a**	85a	85a	84a
Pania	57d	61c	67b	62b
LSD(P < 0.05) = 3.5				
Means	70c	73b	77a	
LSD(P < 0.05) = 2.5				LSD(P<0.05)=2.0

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

The response of seed vigour to seed moisture content at harvest was significantly influenced by cultivar (Table 29.). In cv. Maple, seed vigour increased as seed moisture content at threshing decreased, i.e. seed with the highest vigour (91%) were recorded at the 15% SMC threshing, while threshing at 35% SMC provided seeds with the lowest seed vigour (75%). In contrast, cv. Pania recorded highest seed vigour in samples at the highest seed moisture content of 35% and decreased at the lowest seed moisture content of 15% and to an even greater extent at the intermediate seed moisture content of 25%. However, the very low level of seed vigour recorded at 25% SMC (54%) must again be regarded with caution owing to the practical problems experienced, as described previously.

Table 29. Interaction effects of cultivar and seed moisture content at threshing on seed germination after accelerated ageing.

Cultivar	% Germination after AA of seed samples threshed at different SMC*		
	15%	25%	35%
Maple	91a**	87b	75c
Pania	63e	54f	69d
LSD(P < 0.05) = 2.9			
Means	77a	71b	72b
LSD(P < 0.05) = 2.0			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

Interaction effect of cultivar and threshing method on seed vigour was highly significant. It is rather difficult to explain this statistically significant interaction when comparing the data obtained. However, one possible explanation is that the drop in seed vigour from machine- threshing compared with hand-shelling occurred to a much greater extent in cv. Pania than in cv. Maple(Table 30.).

Table 30. Interaction effects of cultivar and threshing method on seed germination after accelerated ageing.

Cultivar	% Germination after AA in different threshing methods*	
	Hand	Machine
Maple	97a**	72c
Pania	87b	37d
LSD(P < 0.05) = 3.6		
Means	92a	54b
LSD(P < 0.05) = 2.6		

* Results are means of 36 observations.

** Results followed by the same letter are not significantly different.

Threshing method and seed moisture content at threshing had a significantly interactive effect on seed vigour. As shown in Table 31, vigour of hand-shelled seed tended to decrease as seed moisture content decreased, but following machine-threshing (and ignoring the doubtful value at 25%SMC), seed vigour tended to increase with decreasing seed moisture content at threshing.

Table 31. Interaction effects of threshing method and seed moisture content at threshing on seed germination after accelerated ageing.

Harvest method	% Germination after AA at different seed moisture contents.*		
	15%	25%	35%
Hand	89b**	93ab	94a
Machine	65c	48d	50d
LSD(P < 0.05) = 4.4			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

Data presented in Table 32. shows the significant influences of cultivar on the interactive effect between additional phosphorus and seed moisture content on seed vigour.

In cv. Maple, application of phosphorus had no significant effect on vigour of seeds threshed at 15% or 25%SMC, but did increase seed vigour when harvested at 35%. However, phosphorus application did increase the vigour of cv. Pania seeds threshed at 15% and 25%SMC, but not at 35%SMC.

Table 32. Interaction effects of cultivar, addition of phosphorus and seed moisture content at threshing on seed germination after accelerated ageing.

Cultivar	Seed moisture content	%Germination after AA at different P-levels(kg superphosphate/ha)*	
		0	250
Maple	15%	91a**	91a
	25%	87b	87b
	35%	72d	78c
Pania	15%	61f	66e
	25%	51g	57f
	35%	68de	70de
LSD(P < 0.05) = 4.0			
Means		72b	75a
LSD(P < 0.05) = 2.0			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

The interactive effect between seed moisture content and threshing method on seed vigour was also significantly influenced by cultivar (Table 33.).

In cv. Maple, shelling at different seed moisture contents had no significant effect on seed vigour of hand-shelled seeds, while seed vigour declined as seed was machine-threshed at higher moisture contents .

In cv. Pania, seed moisture content significantly affected seed vigour following both hand-shelling or machine-threshing. For example in hand-shelled samples, seed vigour decreased as seed moisture content decreased at shelling, in other words delaying harvesting decreased the vigour of hand-shelled seeds. Machine-threshing at 35% or 15%SMC produced seeds with similar vigour, whereas the lowest seed vigour was recorded following AA-test on Pania seed machine-threshed at 25%SMC.

Table 33. Interaction effects of cultivar, threshing method and seed moisture content at threshing on seed germination after accelerated ageing.

Cultivar	Seed moisture contents(%)	% Germination after AA in different threshing methods.*	
		Hand	Machine
Maple	15	98a**	84cd
	25	97a	77e
	35	96a	55f
Pania	15	81de	46g
	25	89bc	19h
	35	92ab	46g
LSD(P < 0.05) = 6.3			

* Results are means of 12 observations.

** Results followed by the same letter are not significantly different.

Besides the individual effects of nitrogen, phosphorus and seed moisture content at threshing on seed vigour, the interactive effects of all these factors on seed vigour were also highly significant (Table 34.).

In the treatments with no nitrogen applied, application of phosphorus had no significant effect on seed vigour at all seed moisture contents. In the 100 kg N/ha treatment, however, phosphorus application increased seed vigour at the harvesting times of 15% and 25%SMC, but not at 35%SMC.

Application of phosphorus in the 200 kg N/ha treatments had no effect on seed vigour at 15% or 25%SMC, but significantly increased seed vigour at 35%SMC.

Table 34. Interaction effects of addition of nitrogen and phosphorus, and seed moisture content at threshing on seed germination after accelerated ageing.

N-levels (kg/ha)	P-levels (kg superphosphate/ha)	% Germination after AA of seed threshed at different seed moisture contents.*		
		15%	25%	35%
0	0	72cde**	66g	70defg
	250	73cde	67fg	71cdef
100	0	74bcd	67fg	72cde
	250	80a	74bcd	72cde
200	0	82a	73cde	69efg
	250	81a	75bc	78ab
LSD(P < 0.05) = 4.9				

* Results are means of 8 observations.

** Results followed by the same letter are not significantly different.

4.3.3. Electroconductivity

The purpose of the electroconductivity test is to rank seed samples in terms of seed vigour by measuring the membrane integrity of the seed, expressed as the amount of leachate in steep water after seeds had been soaked for 24 hours at 20°C. The greater the amount of exudate detected in steep water the greater the conductivity value, reflecting a weaker cell membrane integrity, and hence lower seed vigour. A healthy, stronger membrane will release less electrolyte and hence reflect seed of greater vigour. The conductivity values recorded in different treatments are presented as follows:

Seed vigour as revealed in the conductivity value was significantly influenced by cultivar (Table 35, Maple > Pania); by seed moisture content at threshing (Table 35., threshing at 15%SMC > at 35%SMC > 25%SMC); by threshing method (Table 36., hand-shelling > machine-threshing) and by addition of nitrogen (Table 39) with the addition of nitrogen at 200 kg/ha providing the highest seed vigour. Seed vigour of seed grown from plants receiving 100 kg N/ha or the no-nitrogen was not significantly different.

Although cultivar and seed moisture content showed significant influences on the conductivity value of the seeds in terms of main effects, the clearer view can be considered in the interaction of these effects. As shown in Table 35., the trend in conductivity values of the two cultivars at different seed moisture contents was quite similar, i.e. the lower the seed moisture content at threshing the lower the conductivity value. However, threshing of Pania at 25%SMC showed the highest conductivity value ($33.02 \mu\text{Scm}^{-1}\text{gm}^{-1}$), but this is possibly due to the practical problem previously described.

Table 35. Interaction effects of cultivar and seed moisture content at threshing on the conductivity of pea seeds.

Cultivar	Conductivity value of seed threshed at different seed moisture contents* ($\mu\text{Scm}^{-1}\text{gm}^{-1}$)			Means
	15%	25%	35%	
Maple	9.55 <i>d</i> **	10.27 <i>d</i>	22.99 <i>c</i>	11.28 <i>b</i>
Pania	22.99 <i>c</i>	33.02 <i>a</i>	25.90 <i>b</i>	27.31 <i>a</i>
LSD(P < 0.05) = 1.62				
Means	16.27 <i>c</i>	21.64 <i>a</i>	19.96 <i>b</i>	
LSD(P < 0.05) = 1.14				LSD(P<0.05)=1.38

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

Although machine-threshing of both cultivars significantly increased conductivity values of the seed, this effect was much greater in cv. Pania than in cv. Maple. For example, when compared with hand-shelling, machine-threshed seeds of cv. Maple increased conductivity value by only 11.21 $\mu\text{S cm}^{-1}\text{gm}^{-1}$ (from 5.67 $\mu\text{Scm}^{-1}\text{gm}^{-1}$ to 16.88 $\mu\text{Scm}^{-1}\text{gm}^{-1}$), whereas in cv. Pania the corresponding increase was 24.53 $\mu\text{Scm}^{-1}\text{gm}^{-1}$ (from 15.04 $\mu\text{Scm}^{-1}\text{gm}^{-1}$ to 39.57 $\mu\text{Scm}^{-1}\text{gm}^{-1}$, Table 36).

Table 36. Interaction effects of cultivar and threshing method on the conductivity of pea seeds.

Cultivar	Conductivity value at different threshing methods*	
	Hand	Machine
Maple	5.67 <i>d</i> **	16.88 <i>b</i>
Pania	15.04 <i>c</i>	39.57 <i>a</i>
LSD(P < 0.05) = 1.28		
Means	10.36 <i>b</i>	28.23 <i>a</i>
LSD(P < 0.05) = 0.9		

* Results are means of 36 observations.

** Results followed by the same letter are not significantly different.

Hand-shelling at different seed moisture contents did not affect conductivity values, but the conductivity value of the machine-threshed seed was significantly affected by seed moisture content at threshing, with conductivity increasing at higher seed moisture content (Ignoring the doubtful value at 25% SMC, Table 37.).

Table 37. Interaction effects of threshing method and seed moisture content at threshing on the conductivity of pea seeds.

Harvest methods	Conductivity value of seed samples threshed at different SMCs* ($\mu\text{Scm}^{-1}\text{gm}^{-1}$)		
	15%	25%	35%
Hand	10.62 <i>d</i> **	10.12 <i>d</i>	10.32 <i>d</i>
Machine	21.93 <i>c</i>	33.17 <i>a</i>	29.58 <i>b</i>
LSD(P < 0.05) = 1.56			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

Once again, the probable error associated with the Pania treatment machine-threshed at so-called 25%SMC may have lead to the significant 3-factor interaction presented in Table 38. If one excludes this treatment, the results indicate that hand-shelled seed of both cultivars showed no significant difference as a result of differences in seed moisture content at threshing, but machine-threshed seed showed a significant increase in conductivity as seed moisture content increased in both cultivars.

Table 38. Interaction effects of cultivar, threshing method and seed moisture content at threshing on the conductivity of pea seeds.

Cultivar	%SMC	Conductivity value in different threshing methods($\mu\text{Scm}^{-1}\text{gm}^{-1}$) [*]	
		Hand	Machine
Maple	15	6.39g ^{**}	12.72j
	25	5.21g	15.33e
	35	5.41g	22.61d
Pania	15	14.85ef	31.14c
	25	15.03e	51.01a
	35	15.24e	36.56b
LSD(P < 0.05) = 2.21			

* Results are means of 12 observations.

** Results followed by the same letter are not significantly different.

Table 39 shows that the conductivity of hand-shelled Maple seed was not influenced by nitrogen fertilizer application, but hand-shelled Pania seed and machine-threshed seed of both cv. Maple and cv. Pania showed a significant negative response to added nitrogen particularly at the highest level(200 kg N/ha).

Table 39. Interaction effects of cultivar, addition of nitrogen and threshing method on the conductivity of pea seeds.

Cultivars	Threshing methods	Conductivity value($\mu\text{Scm}^{-1}\text{gm}^{-1}$) at different N-levels (kg/ha)*		
		0	100	200
Maple	Hand	5.52 ^{f**}	5.88 ^f	5.62 ^f
	Machine	18.50 ^c	16.21 ^d	15.94 ^d
Pania	Hand	16.56 ^{cd}	15.39 ^d	13.17 ^e
	Machine	40.02 ^a	41.87 ^a	36.82 ^b
LSD(P < 0.05) = 2.21				
Means		20.15 ^a	19.83 ^a	17.89 ^b
LSD(P < 0.05) = 1.69				

* Results are means of 12 observations.

** Results followed by the same letter are not significantly different.

4.3.4. HOLLOW HEART

In terms of main effects, the incidence of hollow heart was significantly influenced by threshing method (Table 40.) with machine-threshing significantly increasing hollow heart incidence; by cultivar (Table 41., Pania > Maple); by addition of nitrogen (Table 41.) with the addition of nitrogen significantly decreasing hollow heart incidence and by seed moisture content at harvest (Table 42.) where delayed harvest tended to increase the incidence of hollow heart.

Table 40. Effect of threshing methods on the incidence of hollow heart.

Threshing method	Hollow heart (%) [*]
Hand	4.64 ^{b**}
Machine	6.02 ^a
LSD(P < 0.05) = 0.83	

* Results are means of 48 observations.

** Results followed by the same letter are not significantly different.

As shown in Table 41, the significant main effect of nitrogen on the incidence of hollow heart was due entirely to the effect on cv. Pania only i.e. the percentage of hollow heart decreased as nitrogen level increased. In contrast, cv. Maple showed a much lower incidence(mean 0.47%) and was unaffected by nitrogen level.

Table 41. Interaction effects of cultivar and nitrogen application rate on the incidence of hollow heart.

Cultivars	Hollow heart(%) at different N-rates (kg N/ha)*			Means
	0	100	200	
Maple	0.42 <i>d</i> **	0.58 <i>d</i>	0.42 <i>d</i>	0.47 <i>b</i>
Pania	17.25 <i>a</i>	9.25 <i>b</i>	4.08 <i>c</i>	10.19 <i>a</i>
LSD(P < 0.05) = 3.91				
Means	8.83 <i>a</i>	4.92 <i>b</i>	2.25 <i>b</i>	
LSD(P < 0.05) = 2.76				LSD(P<0.05)=2.26

* Results are mean of 24 observations.

** Results followed by the same letter are not significantly different.

Although the overall hollow heart incidence was not significantly affected by phosphorus fertilizer (Table 42.), hand-shelled seed showed a significant decline in hollow heart incidence in the 250 kg superphosphate/ha treatment compared with the 0 phosphorus treatment, whereas this significance did not occur in machine-threshed seed.

Table 42. Interaction effect of the addition of phosphorus and threshing method on the incidence of hollow heart.

Threshing method	Hollow heart(%) at different P-levels* (kg superphosphate/ha)	
	0	250
Hand	5.83a**	3.44b
Machine	6.33a	5.72a
LSD(P < 0.05) = 1.18		
Means	6.08	4.58
	NS	

* Results are mean of 36 observations.

** Results followed by the same letter are not significantly different.

Although plant nutrition(N&P) and subsequent post harvest management(e.g. threshing method) had a significant effect on the incidence of hollow heart, only cv. Pania was affected by seed moisture content at threshing(Table 43.).

Table 43. Interaction effects of cultivar and seed moisture content at threshing on the incidence of hollow heart.

Cultivar	Hollow heart(%) in seed samples threshed at different SMCs*		
	15%	25%	35%
Maple	0c**	0.42c	1c
Pania	11.25a	11.08a	8.25b
LSD(P < 0.05) = 1.25			
Means	5.62a	5.75a	4.62b
LSD(P < 0.05) = 0.88			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

4.4. EFFECT OF POD POSITION ON SEED QUALITY

The effect of pod position on seed quality, presented in this section, shows the overall influence of pod position and the interactive effects with cultivar, nitrogen, phosphorus and seed moisture content on seed weight, seed germination, hollow heart and on electroconductivity.

4.4.1. Seed Weight

The effect of pod position on seed weight was highly significant, as shown in Table 44, with pods borne on top of plants showing the lowest seed weight, and

Pods from the bottom of plants having the highest seed weight and the middle position intermediate.

Pod position had a significant interaction with the addition of nitrogen (Table 44.). Although seeds from all positions showed the lowest seed weight in the 100 kg N/ha treatment, only seed from bottom and top pods showed similar seed weight in the no-nitrogen and 200 kg N/ha treatments. Seed from middle pods in the 200 kg N/ha treatment showed a small but significantly higher seed weight than that of the no-nitrogen treatment.

Table 44. Interaction effects of nitrogen application rate and pod position on seed weight.

N-rates (kg/ha)	1000-seed weight(gm) at different pod positions*		
	Bottom	Middle	Top
0	256.73a**	251.10c	240.69e
100	250.70c	247.57d	232.69f
200	255.86a	253.54b	240.43e
LSD(P < 0.05) = 2.26			
Means	254.43a	250.74b	237.94c
LSD(P < 0.05) = 1.31			

* Results are means of 24 observations adjusted to 12%SMC.

**Results followed by the same letter are not significantly different.

Seed weight was also affected by the interactive effect of pod position and application of phosphorus. As shown in Table 45, only seed weight of seeds from middle pods was affected by additional phosphorus, with application of superphosphate at 250 kg/ha significantly decreasing seed weight compared with the no-phosphorus treatment.

Table 45. Interaction effects of phosphorus application rate and pod position on seed weight.

P-rates(kg/ha of superphosphate)	1000-seed weight(gm) at different pod positions*		
	Bottom	Middle	Top
0	254.21a**	252.14b	237.95d
250	254.65a	249.33c	237.93d
LSD(P < 0.05) =1.85			

* Results are means of 36 observations adjusted to 12%SMC.

**Results followed by the same letter are not significantly different.

4.4.2. Seed Germination

As shown in Table 46, pod position significantly affected seed germination, with seed from top pods showing the highest germination percentage, despite the fact that there was no difference in germination percentage between the seeds from middle and bottom pods on the plant. However, such minor differences, although significantly different, have little practical relevance, with all germination results being over 95%.

The germination percentage of seeds from different positions on the plant also varied according to the seed moisture content at harvest(Table 46.). For example, at the harvesting time of 15%SMC, seeds from middle pods showed the lowest germination percentage, but no difference was found between seeds from bottom and top pods on the plant. In contrast, the germination percentage of the seeds harvested

at 25% and 35%SMC tended to increase from the bottom to the top position on the plant. Again, however, such differences were small, seed in all treatments producing a germination result of over 95%.

Table 46. Interaction effects of seed moisture content at harvest and pod position on seed germination.

Seed moisture content(%)	Seed germination(%) at different pod positions.*		
	Bottom	Middle	Top
15	98.67a**	97.25b	98.67a
25	95.58d	95.75cd	96.67bc
35	96.46bcd	97.08b	98.29a
LSD(P < 0.05) = 0.98			
Means	97b	97b	98a
LSD(P < 0.05) = 0.56			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

The interactive effect of pod position and nitrogen application on seed germination was significant (Table 46). For example, seed from the bottom pods showed the highest germination percentage at the 100 kg/ha of nitrogen application rate, whereas the highest germination percentage of the seed from the top pods occurred at the 200 kg N/ha treatment. In contrast, nitrogen application rates did not affect germination of the seed from the middle pods. Again, these differences although statistically significant were very small with the biggest differences being less than 2%.

Table 47. Interaction effects of nitrogen application rate and pod position on seed germination.

N-rates(kg/ha)	Seed germination(%) at different pod positions.*		
	Bottom	Middle	Top
0	96.71cde**	96.58de	97.58bc
100	97.71ab	96.29e	97.54bc
200	96.29e	97.21bcde	98.50a
LSD(P < 0.05) = 0.98			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

4.4.3. Hollow Heart

The occurrence of hollow heart was only found in cv. Pania (pod position study used hand-shelled seed, and hollow heart incidence did not occur in hand-shelled Maple seed), and hence the statistical analyses of treatment effects was confined to this cultivar only.

The proportion of seed with hollow heart was significantly affected by pod position on the plant. As shown in Table 48, seeds from top pods showed the highest

percentage of hollow heart, while the percentage of hollow heart in the seeds from middle and bottom pods was lower and similar.

Hollow heart incidence was significantly influenced by the interactive effect between nitrogen application rate and pod position (Table 48). For example, for the no-nitrogen treatment the highest percentage of seed with hollow heart occurred in top pods on the plant, while no effect of pod position on the incidence of hollow heart was recorded in the 200 kg N/ha treatment, and only a small increase was found in top pods in the 100 kg N/ha treatment. The results, however, also strongly suggest that nitrogen application has a major effect in reducing hollow heart levels in Pania irrespective of pod position (0 N treatment, mean hollow heart = 17.94%; 200 kg N/ha treatment, mean hollow heart = 2.44%).

Table 48. Interaction effects of nitrogen application rate and pod position on the incidence of hollow heart in Pania.

N-rates(kg/ha)	% Hollow heart at different pod positions*		
	Bottom	Middle	Top
0	13.83bc**	15.67b	24.33a
100	6.75e	8.75de	11.25cd
200	2.50f	3.00f	1.83f
LSD(P < 0.05) = 3.10			
Means	7.69b	9.14b	12.47a
LSD(P < 0.05) = 1.79			

* Results are means of 12 observations.

** Results followed by the same letter are not significantly different.

The effect of pod position on hollow heart incidence was also significantly influenced by additional phosphorus. As shown in Table 49, the application of phosphorus reduced the incidence of hollow heart significantly at all pod positions. However, in the no-phosphorus treatment the highest percentage of hollow heart occurred in the seeds from top pods, whereas pod position had no effect on the incidence of hollow heart in the 250 kg/ha superphosphate treatment. Again, however, the overall effect of phosphorus application was to reduce hollow heart levels (0 superphosphate treatment, mean hollow heart = 12.16%; 250 kg superphosphate/ha treatment, mean hollow heart = 7.37%)

Table 49. Interaction effects of phosphorus application rate and pod position on hollow heart incidence in Pania.

P-rates (kg/ha of superphosphate)	% Hollow heart at different pod positions*		
	Bottom	Middle	Top
0	9.00bc**	11.44b	16.06a
250	6.39d	6.83cd	8.89cd
LSD(P < 0.05) = 2.53			

* Results are means of 18 observations.

** Results followed by the same letter are not significantly different.

4.4.4. Electroconductivity

Data in Table 50, shows the significant effects of pod position on conductivity values, with seeds from bottom pods showing the lowest conductivity value, particularly in Maple. Despite this there were no significant differences in conductivity values between the seeds from middle or top pods. Obviously, such small but statistically significant differences are of little practical relevance in terms of seed vigour.

The effect of pod position on conductivity values was significantly influenced by cultivar. As shown in Table 50, pod position affected conductivity value only in Maple seed, where electroconductivity values tended to increase from the bottom to the top pods on the plant.

Table 50. Interaction effects between cultivar and pod position on electroconductivity.

Cultivar	Conductivity value($\mu\text{Scm}^{-1}\text{gm}^{-1}$) at different pod positions*		
	Bottom	Middle	Top
Maple	6.88c**	7.12bc	7.33b
Pania	15.94a	16.12a	16.21a
LSD(P < 0.05) = 0.29			
Means	11.41b	11.62a	11.77a
LSD(P < 0.05) = 0.20			

* Results are means of 36 observations.

** Results followed by the same letter are not significantly different.

The effect of pod position on conductivity values was also highly influenced by seed moisture content at harvest (Table 51). Only conductivity of the seed harvested at 35%SMC was affected by pod position, increasing from the bottom to the top pods on the plant. Nevertheless, conductivity values were low in all cases (11.15 - 12.18 $\mu\text{Scm}^{-1}\text{gm}^{-1}$), suggesting that pod position and seed moisture content at harvest did not result in practical differences in seed vigour.

Table 51. Interaction effects of pod position and seed moisture content at harvest on electroconductivity.

Seed moisture content(%)	Conductivity value($\mu\text{Scm}^{-1}\text{gm}^{-1}$) at different pod positions*		
	Bottom	Middle	Top
15	11.75b**	11.77b	12.02ab
25	11.15c	11.37c	11.11c
35	11.33c	11.71b	12.18a
LSD(P < 0.05) = 0.35			

* Results are means of 24 observations.

** Results followed by the same letter are not significantly different.

The interactive effect between cultivar and pod position on conductivity was significantly influenced by nitrogen application rate. As shown in Table 52, in the Maple cultivar, pod position affected conductivity value only in the 100 kg N/ha treatment, being significantly higher at the top position than the bottom position. However, in cv. Pania this effect occurred only in the no-nitrogen treatment. At the same time, the influence of pod position in both these treatments was similar, i.e. the conductivity value tended to increase as the pod position on the plant increased. Again, however, conductivity values were more a reflection of cultivar differences (i.e. Maple range 6.79- 7.37 $\mu\text{Scm}^{-1}\text{gm}^{-1}$, Pania range 14.17- 18.46 $\mu\text{Scm}^{-1}\text{gm}^{-1}$), with nitrogen application within each cultivar having only a relatively minor influence.

Table 52. Interaction effects of cultivar, nitrogen application rate and pod position on electroconductivity.

Cultivar	N-rates (kg/ha)	Conductivity value($\mu\text{Scm}^{-1}\text{gm}^{-1}$) at different pod positions*		
		Bottom	Middle	Top
Maple	0	7.05fg**	7.12fg	7.33f
	100	6.80g	6.96fg	7.37f
	200	6.79g	7.27fg	7.28fg
Pania	0	17.22c	17.90b	18.46a
	100	16.08d	16.28d	15.92d
	200	14.51e	14.17e	14.25e
LSD(P < 0.05) = 0.50				

* Results are means of 12 observations.

** Results followed by the same letter are not significantly different.

CHAPTER 5

DISCUSSION

5.1. SEED YIELD AND YIELD COMPONENTS

Seed yield in this study is calculated from yield components converted to 12% seed moisture content.

Genetically, cv. Pania produced a higher seed yield than cv. Maple, as measured in the hand-shelled seed, with cv. Pania being 9% higher than cv. Maple (Table 2.). This superiority of cv. Pania was due to its much higher number of seeds/pod which more than compensated for fewer pods/plant and lower seed weight compared with cv. Maple (Table 7., 9.).

Harvesting at different seed moisture contents did not affect yield of hand-shelled seeds, but did affect yield of machine-threshed seeds. For example, machine-threshing at 15%SMC resulted in the lowest overall seed yield, whereas the highest seed yield was obtained from the threshing at 25%SMC (Table 3.). This effect was due to differences in the degree of damage when threshing seeds at different seed moisture contents (see seed damage section). This damage led not only to a decrease in seed weight (Table 9), but also to a major decrease in the number of pure seeds. The reduction in seed weight from machine-threshing can be attributed to the loss of cell contents through damaged membranes, cracked seed coats and loss of seed tissues due to splitting or bruising. A similar finding was reported by Castillo (1992), who found a decrease in seed weight in machine-threshed pea seed.

The yield of machine-threshed seed at different seed moisture contents also differed significantly between the two cultivars. For example, the greatest reduction (57%) in seed yield occurred at the 15%SMC threshing in cv. Maple, but at the 35%SMC threshing in cv. Pania but to a much lesser extent (17%). Such reductions in seed yield were also reflected by reduced seed weight, particularly of cv. Maple threshed at 15%SMC (Table 9.).

Nitrogen application significantly affected seed yield. As shown in Table 4., yield of both hand-shelled and machine-threshed seed increased as the rate of

nitrogen application increased, due mainly to a significant increase in the number of pods/plant as nitrogen-levels increased (Table 7.). This supports the work of Hadavizadeh and George (1988) who found that seed yield of pea increased with increased nitrogen supply. This was perhaps a surprising effect since pea plants of both cultivars were well nodulated and would have been expected to have fixed significant plant supporting levels of nitrogen. Apparently, however, additional nitrogen was required by plants to achieve yield increases. Although, internationally, N fertilizer in peas have been variable with many negative results, there have also been some positive responses, the reasons for these being unclear (McLeod, 1987). In New Zealand, fertilizer trials on peas have also given variable results and suggest that N&P stimulate vine development without a significant increase in yield (McLeod, 1987).

Although the application of phosphorus did not affect seed yield, it did have a significant effect on seed weight in both the presence and absence of nitrogen fertilizer. For example, the application of 250 kg superphosphate/ha without added nitrogen resulted in a significant increase in seed weight, but when applied along with added nitrogen, particularly at the intermediate level of 100 kg N/ha, seed weight was reduced. These findings tend to agree with those of Hadavizadeh and George (1988) who recorded an increase in seed weight of pea grown in pots from the addition of phosphorus in the presence of low nitrogen (probably equivalent to soil N levels in the present trial), and a decrease in seed weight when phosphorus was applied along with high rates of nitrogen fertilizer.

5.2. SEED DAMAGE

The fundamental factor determining the effects of both adverse environmental conditions and mechanical damage on seed quality is the inherent capacity of the seed (Heydecker, 1969). Differences in seed location, shape, size, and the nature of individual seed structure account for the wide differences in the frequency and seriousness of impact damage. Large-seeded legumes are especially susceptible to such damage (Moore, 1972). In the present study, cv. Maple showed much greater damage than cv. Pania (14.12% and 6.96%, respectively, Table 12.), but only in

machine-threshing, whereas in hand-shelling cv. Pania showed greater damage than cv. Maple (3.6% and 0.27%, respectively, Table 13.).

Seed damage in hand-shelled seeds in both cultivars, as revealed in the cleavage of the seed-coat (Plate I.), is likely to have occurred during seed development rather than reflecting lack of care in hand-shelling. In fact, during seed development, both cultivars encountered a serious drought followed by heavy rain, which may have resulted in the cotyledons suddenly absorbing a large amount of water and expanding more rapidly than the seed-coat, thereby causing seed-coat cleavage. Generally, wrinkled seed contains a higher percentage of sucrose than smooth seed (Hedley *et al*, 1986), resulting in higher osmotic pressure in wrinkled seed (Ambrose *et al*, 1987; Wang *et al*, 1987). Thus, Pania seed may have absorbed a greater rate and amount of water than Maple seed following heavy rain after drought stress, as mentioned above, resulting in a greater degree of seed-coat damage in Pania than in Maple. It can be said therefore that seeds of Pania are probably more sensitive to this type of field weathering than seeds of Maple.

Table 12. shows a significant interaction between cultivar and moisture content at harvest affecting seed damage. Pea cv. Maple is most susceptible to mechanical damage when threshed at 15%SMC, whereas this occurred at 35%SMC for cv. Pania. However, although the type of seed damage was expressed in a similar way in both cultivars, the level of damage between cultivars was very different. Although seed fracturing was predominant in seed threshed at low moisture content, seed bruising predominated when seed was threshed at high moisture content. The higher degree of damage in cv. Maple at 15%SMC suggests that it is more brittle than cv. Pania, but has a higher resistance to bruising than cv. Pania when threshed at high seed moisture content.

When examining the significant interactive effects between cultivar, seed moisture content and nitrogen levels on seed damage, it appears that the greater susceptibility of cv. Pania to bruising damage at high seed moisture content tends to be greater at higher nitrogen levels than at lower nitrogen levels (Table 15.).

The apparent advantage of threshing Pania seed at 25%SMC after exposure to a long period of heavy rain should be explained further. When Pania plants were harvested under wet conditions and kept longer than 24 hours in the glasshouse

PLATE 1

Type of Seed Damage



- A. Cracked seed coat of cv. Pania which occurred during seed development.
- B. Fractured seed following threshing at low seed moisture content and high speed machine-threshing.
- C. Bruised seed following threshing at high seed moisture content and high speed machine-threshing.

before threshing, most seeds became high in seed moisture content, and some seeds even experienced full rehydration after gaining water from the wet straws and pods. The seeds were then dried down rapidly due to high glasshouse temperatures. However, during threshing, the internal tissues of the seeds were still high in moisture content, while the outer tissues were relatively dry. Thus, although the threshed seeds showed low external damage, in fact they probably suffered severe internal damage during threshing due to the high speed of the combine-harvester. This may well explain the rapid loss in seed vigour in this treatment.

5.3. FERRIC CHLORIDE TEST

Although cv. Maple showed much higher overall seed damage than cv. Pania(see above), results from the ferric chloride test, using pure seeds only, showed a higher level of damage in Pania seed than in Maple seed(Table 17.). Much of the damaged Maple seeds was under-sized(<50% of normal size) due to seed fracturing and was not classified as pure seed, whereas cracking of the seed-coat and bruising was the main problem in cv. Pania. In other words, brittleness appears to be a major problem in cv. Maple when machine-threshed at low seed moisture content, whereas the major problem of seed-coat cracking and bruising in cv. Pania was probably due to the wrinkled shape of the seed-coat and its relatively higher susceptible to such injuries during machine-threshing i.e. to seed-coat cracking at low seed moisture content and bruising at high seed moisture content.

Although machine-threshing at 35%SMC showed a much higher level of seed damage in cv.Pania(41.33%) than in cv. Maple(12.58%), machine-threshed at 15%SMC damage in both cultivars was still high but similar(35.2% in Pania and 34% in Maple). This suggests that in threshing seeds in both cultivars highest resistance to mechanical damage can be obtained by threshing seed at 25%SMC, where damage in cv. Pania(15.42) and cv. Maple(11%)was comparatively low and similar.

Obviously, if one ignores the questionable harvest figure for cv. Pania at 25%SMC, it is clear that in order to minimise seed damage, machine-threshing of cv. Maple must be carried out at a seed moisture content of 25% to 35%, and not

allowed to dry to 15% prior to threshing. In contrast cv. Pania is very prone to mechanical damage at all threshing times(seed moisture contents) and hence demands much greater care at harvest if damage is to be minimised. As expected, hand-shelled seed of both cultivars showed similar and negligible damage at all seed moisture contents.

5.4. SEED GERMINATION

The results from the Standard Germination Test show the superiority of cv. Maple over cv. Pania(Table 21.), and neither cultivar was affected by phosphorus fertilizer nor by nitrogen fertilizer(if one ignores the doubtful Pania result at 25% SMC.). Machine-threshing decreased seed germination of cv. Maple when harvested earlier at the higher seed moisture contents but seed germination of cv. Pania was unaffected by seed moisture content at threshing(excluding the Pania 25%SMC treatment). This result supports the previous finding by Castillo(1992) that peas machine-threshed at a higher seed moisture content(40%) resulted in a lower germination percentage than at lower seed moisture contents(25% and 15%). In contrast, hand-shelled seeds of both cultivars were unaffected by seed moisture content(Table 24.).

The types and extent of seed damage has been categorized in Appendix I., and shows that split seeds predominate when threshed at low seed moisture content. Damage due to bruising was even more prevalent when threshing occurred at high seed moisture content, indicating that bruising was relatively more detrimental to seed germination than splitting.

Although the effect of pod position on seed germination was significant(Table 46), the differences were relatively minor and are therefore thought to be of little practical significance.

5.5. HOLLOW HEART

The general consensus from previous studies is that the incidence of hollow heart can be influenced by high ambient temperature during maturation(Perry and

Harrison, 1973; Halligan, 1986), by rapid drying of seed(Allen, 1961; Perry and Howell, 1965) and by the rate and quantity of water imbibed by dry seeds(Perry and Harrison, 1973). The results of this experiment demonstrate that hollow heart incidence in peas can be influenced by variety, parent plant nutrition, seed moisture content at harvest, harvest method and pod position on the plant.

The most striking difference recorded in the incidence of hollow heart was between cultivars with the wrinkled-seeded Pania being much more susceptible than the smooth-seeded Maple.

As reported by Hedley *et al.*(1986) wrinkled seed contains a higher percentage of sucrose than smooth seed, but a lower percentage of storage protein(Davies,1980). Therefore it is possible that the wrinkled Pania seed with its higher sucrose content leading to a higher osmotic pressure and hence higher moisture content than smooth-seeded Maple, was more susceptible to adverse weather conditions. High rainfall followed by rapid drying, may have been responsible for the greater incidence of hollow heart in this cultivar. Furthermore, the lower percentage of storage protein, an important constituent influencing the strength of the cell membranes, in cv. Pania may also have contributed to its susceptibility to hollow heart, compared with the higher storage protein content of Maple seed.

This role of nitrogen, and hence storage protein in affecting the strength of the cell membranes and its possible associated resistance to hollow heart is further supported by the incidence of hollow heart recorded in cv.Pania at the different nitrogen treatments. For example, the level of hollow heart recorded in the no-nitrogen treatment was 2-fold higher(17.25%) than in the medium nitrogen treatment(9.25%), and 4-fold higher than in the high nitrogen treatment(4.08%). This effect of high nitrogen in reducing the incidence of hollow heart in peas was also recorded by Browning and George(1981). Halligan(1986) also suggested that any environmental effect, especially high ambient temperature, might interfere with seed protein syntheses during maturation and predispose the seed to hollow heart.

The role of phosphorus in this experiment in influencing the incidence of hollow heart, was negligible. A significant interaction has been recorded by Browning and George(1981) between nitrogen and phosphorus on the incidence of hollow heart. The addition of very high levels of phosphorus at low nitrogen input

increased hollow heart greatly, but had no effect at medium and high nitrogen inputs. This work, however, has little relevance to the present field experiment as the rates of phosphorus application used by Browning and George(1981)were far above normal practice, e.g. up to 2.5 tonnes of superphosphate/ha.

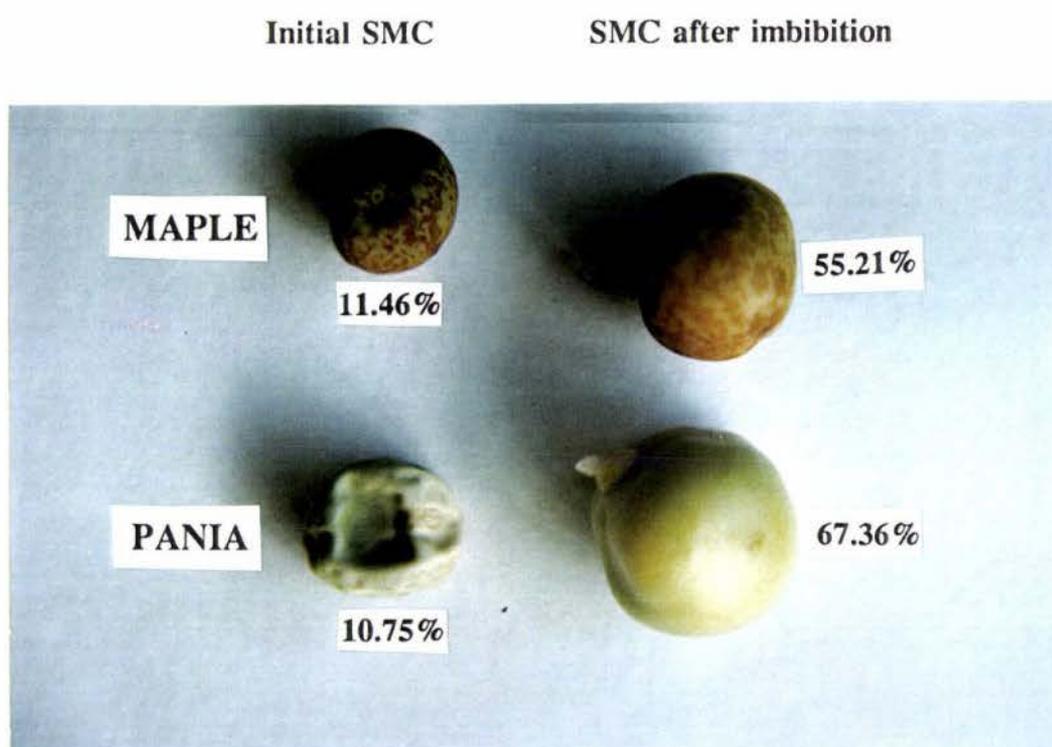
It is again important to highlight the fact that cv. Maple was highly resistant to the development of hollow heart whatever the moisture content of the seed at harvest. By comparison, cv. Pania was highly susceptible at all harvest times and particularly if harvested late i.e. at low seed moisture content. Such late harvested seed would obviously have been longer exposed to environmental stress conditions and therefore more prone to develop hollow heart due to high temperature and excessive drying rate. These results support the findings of Perry and Harrison(1973) who found that there was more hollow heart in pea seeds bleached by sunlight when they were left attached to the parent plant in the glasshouse for two months after they reached maturity, than unbleached seeds or seeds harvested at maturity. They suggested that rapid drying of seeds of high moisture content causes predisposition to hollow heart because the outer cells lose water more rapidly than those in the centre of the seed. At high temperature the internal tissue remains moist after the outer layers are dry and further loss of water may be restricted by a fall in the hydraulic conductivity of the dry outer cells. Deterioration of moist seeds is rapid when they are subjected to high temperature.

Perry and Harrison(1973) also found that the incidence of hollow heart can be induced by the rate of water imbibed during germination. Cracks in the seed-coat resulting from mechanical damage can increase the rate of water uptake(Perry and Harrison,1973; Powell and Matthews,1979). This may account for the significant increase in the incidence of hollow heart recorded in machine-threshed seed(Table 40.).

Seeds from top pods on the plant showed a higher incidence of hollow heart than seeds from middle and bottom pods(Table 48). This finding supports previous results by Castillo(1992). It also appears that the beneficial effects of phosphorus and particularly nitrogen in reducing the incidence of hollow heart were largely expressed in pods from the top position and suggests that the associated but unexplainable build-up and balance of proteins and phospholipids in the cell membranes of the

PLATE 2.

Imbibition characteristics of Maple and Pania seeds.



Due to higher osmotic pressure, after 24-hour soaking in distilled water at room temperature, Pania seed contain greater amount of water than Maple seed.

PLATE 3.

PLATE 3.**Hollow Heart in Garden Pea****PLATE 4.****Normal Seed**

developing seed, particularly in the late developing pods at the top of the plant, had a significant influence on the susceptibility of seed to hollow heart. Obviously, top pods originate later than middle and bottom pods. As a result they may suffer a deficiency of nutrient supply during development, due to environmental stress, and also suffer further stress due to the priority sink in earlier originating pods resulting in a weaker membrane and hence greater susceptibility to hollow heart (Flinn and Pate, 1968).

Early reports on hollow heart in peas by Allen (1961) indicated that it cannot be detected in fresh peas, only becoming visible following drying. More recently, it has been suggested that a low molecular weight, heat labile growth inhibitor is present within hollow heart tissue which is capable of delaying pea seed germination and that it may be formed from products of autolysis following death of cells during water imbibition (Harrison and Perry, 1973).

5.6. SEED VIGOUR

Seed vigour as measured by the accelerated ageing and conductivity tests, show the potential for cv. Maple to produce seeds with much higher vigour than cv. Pania (Table 28, 35.). Genetic constituents likely to be responsible for this phenomenon, as revealed in different seed morphology and seed compositions between the two cultivars, have been described in the hollow heart section. These differences may also result in different degrees of resistance to environmental stress and/or mechanical impact during harvesting or processing.

The results show a significant effect of nitrogen on seed vigour, with the application of 200 kg N/ha providing the highest seed vigour compared with the 100 kg N/ha and no nitrogen treatments (Table 28, 39). This expression of vigour was largely confined to cv. Pania and was more evident in the Accelerated Aging Test than in the Conductivity Test. The response of seed vigour to nitrogen levels supports the finding of Browning (1980), as cited by Hadavizadeh and George, 1988, who observed that seed with a higher nitrogen content produced leachates with a lower conductivity. Thus, the lower seed protein content concentration and the reduced amount of protein per seed is associated with a higher level of leaching. Hadavizadeh

and George(1988) also claimed that this increase in seed vigour, due to added nitrogen, only occurred in the presence of medium phosphorus supply(250-500 mg P/plant). They suggested that the low vigour of seed was associated with the impairment of membrane functioning due to chemical and physical structural defects. In the present study, however, seed vigour was not significantly influenced by the interaction effect of nitrogen and phosphorus. In fact, the addition of phosphorus only had a minor beneficial effect on germination following accelerated aging, and suggests that soil phosphorus levels of the control(no-phosphorus) plots were adequate for growth and development.

The mechanism of seed leaching is also linked to the physical condition of the testa. Powell and Matthews(1979) showed that seeds with cracked testae produced a higher conductivity reading than intact seeds. Biddle(1980) also showed that seed coat cracks were associated with high conductivity and were mainly caused by mechanical harvesting and threshing operations. These findings are relevant to the present study, where machine-threshed seeds showed a much lower germination percentage after accelerated ageing(Table 30.) and a much higher conductivity reading(Table 36.) than hand-shelled seeds.

Seed moisture content at threshing also had a significant effect on seed vigour(Table 29, 35), presumably reflecting differing resistance of pea seeds to mechanical injury when threshed at different seed moisture contents. Excluding the Pania seeds harvested at 25%SMC, seed vigour generally improved with decreasing seed moisture content at threshing, particularly in cv. Maple. Seeds threshed at 35%SMC suffered high levels of bruising damage, while those threshed at 15%SMC suffered mainly seed splitting(see Appendix I). Although threshing at 35%SMC showed a lower percentage of seeds with seed-coat damage, as revealed by ferric chloride staining, than threshing at 15%SMC, it did result in a greater loss in seed vigour suggesting that bruising is more damaging than splitting, in terms of decreasing seed vigour. Moore(1972) found that cleanly cut surfaces are much less harmful than bruised tissues. This agrees with the work of Castillo(1992) who found that seeds threshed at 40%SMC produced a higher conductivity reading and lower germination after controlled deterioration than seeds threshed at 25%SMC or 15%SMC. However, the present data shows that this effect was confined to machine-

threshed seed, particularly in Maple. This reinforces the claim that bruising (at high seed moisture content) is more detrimental to seed vigour than splitting (at low seed moisture content). In hand-shelled Pania seed (Table 33.), delayed harvesting resulted, however, in a decline in seed vigour, which is likely to be related to seed deterioration due to adverse weather conditions (Hampton, 1994). Green *et al.* (1966) working with soybean and Castillo (1992) working with peas, also found a reduction in seed vigour when harvest timing was delayed. The present results also show clearly that Pania seed is much more susceptible to field weathering than Maple.

Results of the pod position study clearly demonstrate the significance of environmental effects, as seeds were all hand-harvested. Although conductivity readings of seed from different pod positions were similar, seeds from top pods showed a significantly higher conductivity reading than those from bottom pods, and that this effect only occurred in cv. Maple (Table 50). The effect of pod position on seed vigour in this respect was significant only in pea seeds harvested at 35% SMC (Table 51), reflecting the immaturity of the seed. Matthews (1973) found that the readiness with which electrolytes are leached from dried seeds in steep water decreases with increasing time after fertilization, i.e. more mature seed. This result is also in keeping with the work of Castillo (1992), who found that seed of 40% SMC harvested from top pods were still immature, and provided higher conductivity readings than seeds harvested at 25% or 15% SMC.

Again, cv. Pania showed a higher sensitivity to nitrogen in improving seed vigour than cv. Maple (Table 52.). Application of nitrogen at both 100 and 200 kg/ha in cv. Pania, but only at 200 kg/ha in cv. Maple, improved vigour of the seed from top pods to a level similar to the seeds from middle and bottom pods. The seed vigour results, in terms of conductivity and even hollow heart incidence leads to the suggestion that application of nitrogen at 200 kg/ha together with careful harvesting at a seed moisture content of 15% SMC can produce seed with homogeneously high vigour.

CONCLUSION

Pea cv. Pania has a higher inherent yield capacity than cv. Maple due mainly to higher numbers of seeds/pod, and despite the production of fewer pods/plant and lower seed weight. However, cv. Pania has a lower capacity to produce seed with high quality, in particular seed vigour, as determined by the Accelerated Aging Test, the Conductivity Test and hollow heart incidence.

Application of nitrogen increased seed yield solely BY increasing number of pods/plant. There was no direct effect of phosphorus addition on seed yield, although a significant interaction with nitrogen, especially when combined with the medium rate of nitrogen (250 kg superphosphate/ha + 100 kg N/ha), resulted in depressed seed weight. Addition of nitrogen also increased seed vigour by increasing seed germination percentage after accelerated aging, decreasing conductivity values and lowering hollow heart incidence, particularly in cv. Pania. The effect of added phosphorus on seed vigour was smaller than that of nitrogen, and occurred only in the Accelerated Aging Test results. Again, a bigger increase was found in cv. Pania than in cv. Maple.

Machine-threshing decreased both seed yield and seed weight, due to seed damage. This occurred to a greater extent in cv. Maple than in cv. Pania at the 15%SMC harvest, but to a greater extent in cv. Pania than in cv. Maple at the 35%SMC harvest. However, in terms of seed vigour, machine-threshed Pania seed suffered to a greater extent than Maple.

Time of harvest (seed moisture content) did not affect either seed yield or seed weight in hand-shelling, but did affect machine-threshed seed due to altering degrees of seed damage. Time of harvest also affected seed vigour, with delayed harvest decreasing seed vigour of hand-shelled seed due to field weathering, as determined by the Accelerated Aging Test and by the Hollow Heart Test. Only Pania seed was affected. In contrast, delayed harvest increased seed vigour in machine-threshed seed samples of both cultivars, as determined by the Accelerated Aging Test, for Maple ;and by the Conductivity Test, for both cultivars. This reflected the more harmful effect of bruising at the higher SMC harvest than splitting at the lower SMC harvest.

Seeds from top pods on pea plants showed a lower seed weight and lower

seed vigour as shown by the higher incidence of hollow heart in Pania and higher conductivity reading in Maple, than seed from middle and bottom pods. Although germination test results were significantly influenced by pod position, these differences were only minor and are considered to be of only negligible practical importance. Application of nitrogen and phosphorus also resulted in improved seed vigour in cv. Pania, especially when they were applied at 200 Kg N/ha and 250 kg superphosphate/ha. Seeds from all pod positions had similarly high vigour, as shown by low conductivity value and especially by low hollow heart incidence.

The results of this experiment clearly show that hand-shelling of both cultivars and machine-threshing at 25 to 35%SMC for cv. Maple and at 15% to 25%SMC for cv. Pania, are appropriate methods and times of seed harvesting in order to minimise seed damage. Application of 200 kg N/ha and 250 kg super phosphate can be beneficial in improving seed vigour in garden peas(Pania). However, under these fertilizer inputs cv. Pania has a lower capacity to produce seed of high vigour than field peas(Maple).

This study has clearly shown differences in cultivar susceptibility to machine threshing damage in peas which are further affected by time of harvest(SMC), and to a lesser extent, pod position. Nevertheless, further work is needed to clarify the role of nitrogen and phosphorus, the apparently different effects of seed cracking and bruising on seed quality, in particular seed vigour and the incidence of hollow heart.

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APPENDIX 1.

Proportion of split and bruised seed following machine-threshing.

Cultivar	SMC at harvest	Damage proportion(%)		% whole damaged seed
		splitting*	bruising	
Pania	15%	90.55	9.45	9.28
	25%	24.02	75.98	3.96
	35%	72.30	27.70	17.69
Maple	15%	100	0	59.17
	25%	80.33	19.17	14.24
	35%	42.97	57.03	10.52

* Splitting includes cracked seed coat, broken seed, half seed and fractured seed.

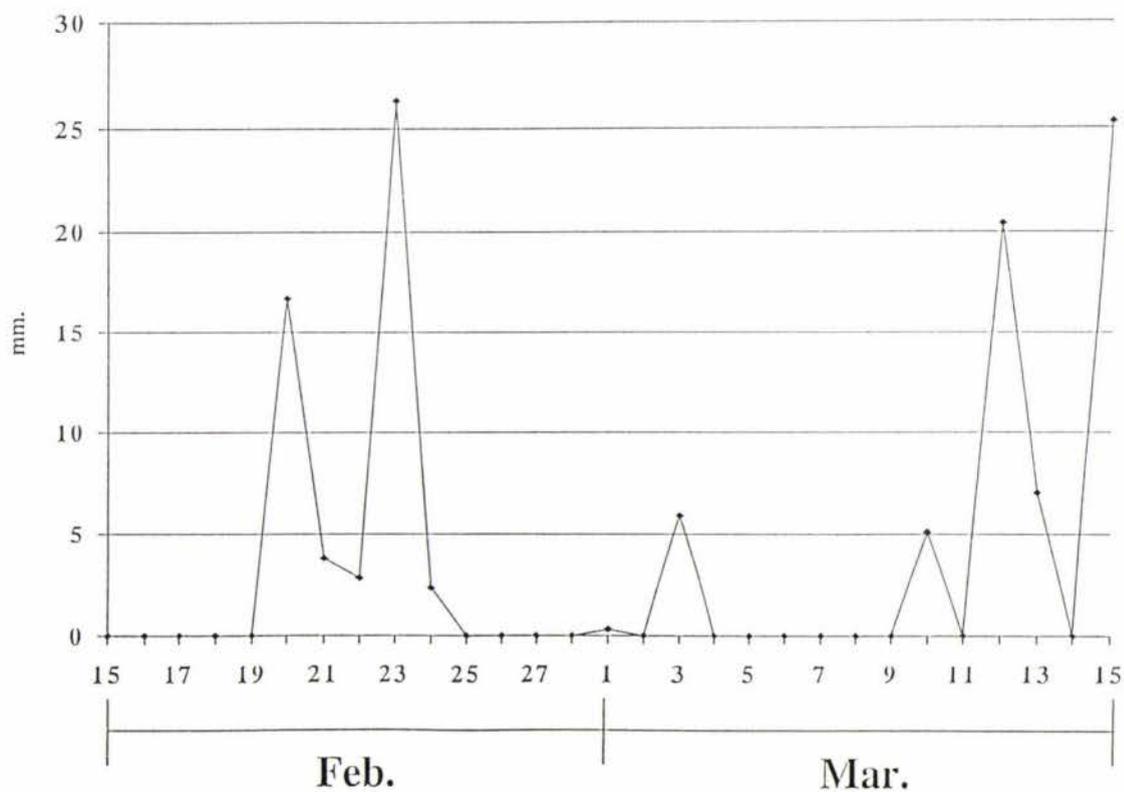
APPENDIX 2.

Harvesting Dates and Rainfall Data.

Harvesting Date

	1 st harvest	2 nd harvest	3 rd harvest
Pania	19 Feb.1995	23 Feb.1995	26 Feb.1995
Maple	28 Feb.1995	2 Feb.1995	7 Mar.1995

Rainfall Data During 15 Feb.-15 Mar.1995



* Data from Grasslands AgResearch Meteorological Station, Palmerston North, NZ.

APPENDIX 3.

Soil Fertility Data

pH	Olsen P	SO ₄	Exch. K	Exch. Ca	Exch. Mg	CEC	Soil volume correction factor
5.3	13	5.5	0.49	7.2	1.39	21	1.00

Comment: Phosphate and sulphate values are expressed as $\mu\text{g/g}$ (air-dry). Exchangeable cations and CEC values are expressed as $\text{meq}/100\text{g}$ (air-dry). The soil volume correction factor is a measure of the weight of air-dry soil(g) per volume(ml) and can be used to convert results to a volume basis(eg. $\mu\text{g/g} \times \text{soil volume correction factor} = \mu/\text{mg}$)

* Data from analysis carried out by the Fertilizer & Lime Research Centre, Massey University, Palmerston North, NZ.