Effect of Harvest date on Seed quality and potential longevity of two Soyabeans (*Glycine max* (L.) Merr) Varieties.

By

Mutiti Alice (B.Sc. Agriculture.)

A dissertation submitted to the University of Zambia in partial fulfillment of the requirement of the degree of Master of Science in Agronomy

The University of Zambia, School of Agriculture

Department of Crop Science

2001

Declaration

I, Mutiti Alice, hereby declare that all the work presented in this dissertation is my own and has never been submitted for a degree at this or any other University.

APPROVAL PAGE

This dissertation of Ms. Mutiti Alice is approved as fulfilling part of the requirements for the award of the degree of Master of Agronomy by the University of Zambia.

Examiner	Signature	Date
S.S. MWALE		31-12-01
M. MAIAA		31.12.2001
O.A. YEROKUN		31.12.2001

Dedication

This dissertation is dedicated to my husband, Mweetwa Moses and my parents Mr. and Mrs. Kankwala.

Acknowledgements

I wish to extend my gratitude to my supervisors, Dr. D. N. Mbewe and Mr. S. S. Mwale for their support, guidance and patience during my research. I, therefore, acknowledge their great contribution through the comments and corrections during the research work and completion of this dissertation.

I wish to thank my classmates for all the knowledge shared during my short time with them. I also thank members of staff of the Crop Science Department for their contributions during the initial and final stages of the research work.

Special thanks go to the SADC Plant Genetic Resources Centre (SPGRC) for the use of their library and the laboratory facilities. I wish therefore to thank Mr. Floyd Mwanza, Mr. Kennedy Kaunda Hamudulu and Mr. Ben Luwiika for all the help they rendered, materially and otherwise.

My gratitude is also extended to Mr. Japhet Phiri and Mr. Numail Daka for all their assistance during the work at the Field Station, at the School of Agricultural Sciences.

I am also grateful to SACCAR/GTZ for awarding me the scholarship that made it possible for me to pursue my studies.

Finally, I wish to thank my husband for all the love, support and patience during my MSc studies.

Table of contents

List of Tables

List of Figures

List of Appendices

ABSTRACT

The need for preservation of crop species is being widely recognized. This requires that seeds posses good initial quality and potential to be stored for long periods. Time of harvest of soyabean seed has been identified as the main factor affecting quality and potential for long term storage. However, there exists no clear recommendation for time of harvest that ensures that these two attributes are achieved. This study was done to establish the optimum time of harvest for soyabean in order to produce seed with the best quality and highest possible potential for longevity. The experiment was done using a split-plot design with four replications in which the factor of variety was assigned to the main plots and harvest dates were randomly assigned to the subplots. Two soyabean varieties, Hernon-147 and Kaleya, and six times of harvest after seed podding (7, 14, 21, 28, 35 and 42 days) were used. Parameters measured at each harvest included seed dry matter content, initial seed moisture content, germination of fresh and dried seed, initial seed vigour and accelerated ageing test. Seed was artificially aged at each harvest for varying periods (1, 2, 3, 4, 5, 6 and 7 days) after which germination and vigour tests were done. The results showed that variety had significant effect on vigour of artificially aged seeds and dry matter content of the seeds. Time of harvest had significant effect on allthe parameters that were measured. Seeds of both varieties attained physiological maturity at 28 days after podding, but the amount of dry matter at physiological maturity differed significantly between the two varieties. Onset of germination in fresh seeds was

observed at 7 days after podding and in dried seeds at 14 days after podding for both varieties. Seed germination and vigour improved with time of harvest, being optimal at 28, 35 and 42 days after podding. Seed with high potential for long term storage was observed at 35 and 42 days after podding, when moisture contents are low enough (20% to 14%) for harvesting.

This stage coincided with the commercial soyabean harvest time (16% to 12% moisture), an indication that this recommendation could also be used for soyabean seed grown for long term storage.

CHAPTER 1

INTRODUCTION

Soyabean (*Glycine max* (L) Merr.) is the world's leading legume (Weiss, 1983) with oil and protein content estimated to be about 20% and 43%, respectively. Thus soyabean has twice as much protein as that of most pulses and its' protein has a high biological value. In addition, the seeds are rich in digestible nutrients and possess high calcium, iron and vitamin content (Scott and Aldrich, 1983). It is a primary source of edible oil and it is a protein rich meal for both livestock and human consumption. Many developing countries have recognized the potential of soyabean as a valuable source for supplementing the traditional staple food, such as maize, which has low protein content.

In Zambia, soyabean is a relatively new crop, having been introduced in the 1930's (Wellving, 1984). However, as a result of significant agronomic research and extension work, coupled with a vigorous public awareness campaign on its nutritive value, its cultivation has become a popular and reliable venture (Wellving, 1984). In addition, the country has very suitable climatic conditions for soyabean production. These two factors have led to the significant increase in soyabean production in the country. For example, between 1980 and1990, total production increased from 1,685 tons to 23,814 tons (MAFF, 1996).

The increased demand for soyabean for consumption in the form of flour, oil and fat cakes, has resulted in an increase in demand for soyabean planting seed. The required planting seed must be produced on an annual basis because the soyabean seed is too short lived (6 months) to carry over from one year to another in routine warehouse storage (TeKrony *et al.* , 1980; Mwale, 1993).

Moreover, the importance of preserving crop varieties is increasingly being recognised worldwide (Lungwangwa, 1993). Seed storage has since been on the increase and depending on the length of the storage period, three categories of storage have been recognised. Short term storage is less than 9 months, medium term storage lasts 2-3 seasons and long term storage is in excess of 3 years. As pointed by Bass (1995), the goal of gene banks is to maintain seed viability in a wide range of species for indefinite periods (long term storage, typically 10 to 100 years or more). But gene bank managers have recognised the fact that seeds eventually deteriorate whatever the conservation conditions. Even in long-term storage facilities, longevity of seed has been found to be affected by genotype and cultural practices (Ellis *et al*, 1993).

With respect to the storage of the soyabean seed, two problems have been identified. These problems are namely, whether adequate quality levels will be maintained for the time the seed is in storage and whether the seed vigour at the time of planting (after storage) will be high enough to achieve acceptable emergence under a wide range of field conditions (TeKrony *et al.*, 1979). These problems have mostly been associated with time of harvesting.

Delays in harvesting have been shown to lead to exposure of the seed to unfavourable environmental conditions and the acceleration of the deterioration process (Marcos - Filho *et al*, 1994). Unfavourable field environments may affect the length of seed storage by promoting rapid onset of the processes of seed deterioration. Extremes of temperature and moisture in the field may predispose the seed to rapid deterioration of when put in storage.

Harvesting soyabean seed too early has yielded seed of poor potential storability (Adetunji, 1991). Although it has been suggested that soyabean be harvested early (Tekrony *et al*, 1979), there are no specifications given as to which stage is early enough. Another suggestion has been that soyabean seed be harvested at a time as close as possible to physiological maturity. This is after the stabilisation of dry matter translocation to the seeds, at which stage the seeds are said to have reached the maximum germination and vigour (Mwale, 1993; Harrington, 1972).

The recommended time for harvest of soyabean seed crop in Zambia is when most of the leaves have turned yellow and fallen off, and when the pods feel brittle (MAFF, 1995). Gene bank managers follow the same recommendation when they are harvesting soyabean after rejuvenation or seed multiplication.It is not clear as to whether this recommended time of harvest which was originally developed for seed for immediate sowing is also suitable for soyabean meant for long term storage. There has already been an observation that this recommendation does not give a good guide and that it has been linked to the low quality of seed with respect to germination (SCCI, 1993).

Therefore, the objective of this study was to establish the most suitable harvest time for soyabean; a time that would provide both good seed quality and potential longevity with respect to long term seed storage in genebanks.

This study was designed to establish the following :

-The stage at which seeds acquire the ability to germinate.

-The changes in potential storability during seed development and maturation.

-The optimum harvest date that will give the highest seed quality in terms of vigor, potential longevity and germinability.

CHAPTER 2

LITERATURE REVIEW

2.1 Onset of Germination

Onset of germination can be determined through the germination of fresh seeds. This helps to determine at which moisture content the seed is able to germinate without dessication. Miles *et al* (1988) have shown that it is actually possible to germinate fresh immature seeds of soybeans. Other findings suggest that slow dessication is required for germination of immature soyabeean seeds (Adams and Rinnie, 1981). Germination levels as high as 93% have been attained in fresh seeds. Considering the actual moisture content at which seeds acquire the ability to germinate, Egli and Tekrony (1993) reported that soyabeans germinated at a moisture content of 74.1% without dessication. At this moisture content, a final germination of 91% was observed, even though the germination count had to be delayed until 13 days after planting. Miles *et al* (1988) also reported 93% germination in fresh soyabean seeds with 74% moisture content. This means that germination of fresh seeds with higher moisture content is possible except that their germination rate is low.

Miles *et al* (1988), have shown that seeds acquire the ability to germinate when they have acquired 35% of their maximum dry weight. This may suggest that ability to germinate is attained when sufficient dry matter is acquired.

It has been shown by Egli and Tekrony (1993), that when the seeds acquire the ability to germinate, even immature seeds absorbe water regardless of the initial water content. But the seed water content after imbibition tends to be higher than that associated with germination of the mature seed (Hunter and Erickson, 1952). There has been no report as to what effect onset of germination may have on the potential of the soyabeans for long term storage.

2.2 Initial Germination of Dry Seeds

Germination is defined as the resumption of development of the embryo of the seed when provided with conditions suitable for growth (Thompson, 1979). It has been said to be correlated with dry matter and seed water content at harvest. Lungwangwa (1993) reported that both seed size and weight affect germination. The large size of the seeds has been in many cases associated with a large quantity of stored food reserves, which is reflected in higher dry weight. High germination percentage of soyabeans seeds has been associated with large size of seeds (Powell, 1988). In soyabeans, stored food reserves are contained in cotyledons. This stored food is the principle source of energy during the early stages of seedling growth and development.

Therefore this implies that since the time of harvest has a bearing on the amount of dry matter, it should have an effect on the level of germination of seeds. In this regard it has generally been recognised that maximum quality of seed is realized at physiological maturity, defined as the stage at which seeds attain maximum dry weight and full germination capacity (Harrington, 1972). Other workers have shown that immature seeds are inferior to mature seeds in terms of viability (Dasgupta *et al,* 1982; Ellis *et al*, 1987). Work done on sunflower (*Helianthus annus*) has shown that sunflower harvested at 14 and 21 days after flowering produced seed of low germination. This finding was attributed to the high proportion of empty and immature seeds (Adetunji, 1991). Delayed harvesting produced seeds of higher germination.

It has been reported that there is either no effect or a negative relationship between germination and dry matter content (Marcos-Filho *et al*, 1994). There has been a report on soyabeans harvested at physiological maturity that did not posses the highest germination capacity (Tekrony *et al*, 1980). This may mean that dry matter content is not the sole determinant of the level of germination. Marcos-Filho *et al* (1994) also reported that the point of maximum soyabean seed weight did not coincide with maximum germination. Perez and Arguello (1995), also reported that groundnut (*Arachis hypogeae* L) did not exhibit maximum seed germination at physiological maturity, but prior to this stage.Such reports suggest that there maybe other factors affecting germination apart from dry matter content of the seed.

With regard to initial moisture content of the seed, it has been reported that as the seed water content declines, germination of seeds improves (Mayer, 1973). The loss of water is accompanied by structural changes at the microscopic and submicroscopic levels (Klein, 1970) leading to increased resistance of the seed to adverse external conditions, leading to enhanced viability.

In most cases, judgement as regards seedlot storability is based on the results of a standard germination test. For example, a seedlot that has 70% germination will not generally store for as long a period as another one of the same cultivar that has germination level of 98% because low germination has been associated with seed deterioration, and therefore reduced ability to perform well (Mayer, 1973). It has been shown that germination of most lots is often quite similar (Delouche and Baskin, 1973). Even in cases where the initial germination is excellent and comparable, the actual storability of the individual lots could range from excellent to poor because they could have varying levels of vigour (Mayer, 1973). This means that the ordinary germination percent of the seedlots at the time they are placed in storage does not provide imformation on which to base a decision regarding their suitability for long term storage. Bewley and Black (1994), suggest that in terms of storage, the point of highest germination may not coincide with the point of maximum potential storability, because seeds have been shown to continue to mature after the completion of seed filling resulting in increased longevity in subsequent storage. But it has been shown by other workers that physiological maturity stage of soyabeans represents maximum seed viability (Wahab and Burris, 1971; Delouche, 1974). Soyabean seeds have been reported to attain maximum potential longevity about one to two weeks after physiological maturity (Rao and Jackson, 1996). This has also been reported in barley (Bewley and Black, 1994).

The results of Ellis and Pieta-Filho (1991) also agree that maximum ability to survive air dry storage is attained after mass maturity. But on the contrary, it has been stated that it is after mass maturity that germination and potential for storage is on the decline (Harrington, 1972).

2.3 Seed Vigour

International Seed Testing Association (ISTA, 1995) defines vigour as "the sum total of those properties of the seed, which determine the level of activity and performance of the seed or seed lot during germination and seedling emergence". Another definition of seed vigour is "Seed vigour comprises seed properties which determine the potential for rapid, uniform emergence and development of normal seedlings under a wide range of field conditions" (AOSA, 1983).

According to Hampton (1995), seed vigour is of great significance in the determination of seed storage potential. He stated that storage potential of high germinating seeds is related to their vigour status on entering storage. Even under controlled storage conditions (as in seed gene banks), performance after storage can still depend on the vigour of the seed (Helmer *et al*, 1962). Seed vigour, if properly evaluated, does not only serve as a measure of the seeds to survive and emerge under adverse field conditions but also as a measure of the storability of the seeds (Helmer *et al*, 1962). However in recent years, investigation on seed vigour has concentrated on the physiological causes of vigour differences, especially the role of seed ageing and membrane integrity.

Physiological seed ageing is now recognized as the major cause of reduced vigour in many species (Anderson, 1970; Powell, 1986).

Seed vigour is therefore determined by the stage of development of the seed. This, being concluded by fact that delays in harvesting contribute to weathering and loss in vigour (Rao and Jackson, 1996). It has been documented that delays beyond harvest maturity may render seeds sensitive to damage from dessication (Walters and Engels, 1998), leading to loss of membrane integrity and consequent loss in vigour. The fact that it is possible to have seeds with different storage potential occurring on the same indeterminate plant, is proof that vigour is greatly affected by degree of maturity (Copeland and McDonald, 1985).

It has been reported that soyabean seed vigour may continue to improve after physiological maturity (Rao and Jackson, 1996). This has been reported to be reflected in the increased longevity in subsequent storage (Bewley and Black, 1994). However, this report does not agree with Harrington's theory (1972), that vigour declines soon after physiological maturity. The theory further states that at physiological maturity, the seed achieves maximum seed weight and severs it's vascular connection with the parent plant and this leaves it with no opportunity for deterioration. At this stage, it has the maximum attainable seed quality (Wahab and Burris, 1971; Delouche, 1974; Tekrony*et al,* 1980). Therefore, for the purpose of long-term storage, the time of harvest becomes a critical issue and prompt harvest is imperative. On the other hand, immature seeds have been shown to have low vigour since the formation of the embryo, accumulation of food reserves and hormones, which are necessary for germination, has not yet been completed (Lungwangwa, 1993).

In soyabean seeds, time to harvest is particularly critical. Delays in harvesting may lead to the exposure of the seed to unfavourable environmental conditions and the acceleration of the deterioration process (Marcos- Filho *et al*, 1994). Unfavourable field environments have been reported to reduce the length of seed storage by prompting rapid onset of the processes of seed deterioration.

2.4 Seed Moisture and the Maturation Process

Though seed growth has been studied extensively (Hanson, 1986) the regulation of seed maturation, in particular, the role of seed water relations in growth ceasation is not understood. Soyabean seeds are highly hydrated during most of their development, but seed water content declines from about 80% early in seed development to about 55% at physiological maturity (Tekrony *et al*, 1979; Fraser *et al*, 1982). With the need to dry soyabeans seeds after harvesting to desired moisture levels (5%) for controlled storage in seed gene banks, dessication tolerance becomes an important aspect of seed quality. It has been reported that this aspect is greatly influenced by stage of development, being only attainable soon after attainment of maximum seed dry weight (Ellis *et al.*, 1987). Dessication tolerance has been reported to be absent during the early stages of seed development, until after the stage of physiological maturity (Ellis *et al.*, 1987).

Seed moisture content affects seed cleaning procedures. When the moisture is either too high or too low, seed vigour can be greatly reduced during processing, and this will consequently affect the storage potential of the seed (Clifford *et al.*, 1981).

2.5 Artificial and Natural Ageing of Seeds

According to Bewley and Black (1994), it is difficult to make comparisons among deteriorated seeds that show diverse symptoms of deterioration.
Among the many manifestation of seed deterioration are changes in seed colour, delayed germination, decreased tolerance to suboptimal environmental conditions, reduced growth of seedlings, reduced germinability and increased numbers of abnormal seedlings (Justice and Bass, 1978). Ellis *et al* (1993) state that soyabean deterioration starts in the field and that the rate at which seed deterioration will occur in the subsequent pre-harvest environment is quantitatively similar to that expected to occur under the same conditions in air dry post-harvest storage.

Deterioration of seeds in storage has been associated with the number of causes i.e., depletion of food reserves, alteration of chemical composition, membrane alteration, enzyme alteration and genetic damage (Abdul-Baki and Anderson, 1972). As regards the depletion of food reserves, examination of a few nonviable seeds has shown that there is still plenty of food reserves left in nonviable seeds, although mobilisation of these reserves may still be difficult (Perez and Arguello, 1995).

Membrane alteration has been shown to be a major cause of seed deterioration during ageing. It has been suggested the use of the electrical conductivity test as a better assay for determining the loss in the viability of the seeds in storage (Walters and Engels, 1998; Perez and Arguello, 1995). The electrical conductivity test shows the extent of substrate leakage from an imbibed seed. It has been proposed that solute leakage accompanying imbibition is the result of cell membrane damage allowing the free release of cytoplasmic solutes into water. According to Floris (1970), excess leakage from deteriorated seeds may represent loss of respirable substrate from some seed species, whereas others lose more amino acids than sugars. It is not known to what extent substrate loss alone affects germinability. Work done on soyabean seed has also shown a lack of significant correlation between conductivity of soak water and standard germination of seed harvested at harvest maturity and at seven days after harvest maturity (Moore, 1971).

Germination percent is not a sensitive assay in determining the deterioration of seeds. Walters and Engels (1998) showed that significant deterioration could be detected within 8 weeks of storage in Lettuce seeds stored at 7% moisture content and 35°C when root growth or germination time was monitored, and after 16 weeks when percent germination alone was monitored.

Work done on soyabean showed that after 44 months of ageing at 4°C and 8 to 10 % moisture content, decline in germination was only from 98 to 86 % when using the whole seed. In the same experiment, when only soyabean seed axes were used, the germination declined from 60 to 0 % (Walters and Engels, 1998). This is an indication that the deterioration observed in the soyabean seed is associated with damage to the axes rather than to the whole seed. It has also been suggested that rapid and non-uniform absorption of water results in considerable damage to the seed coat tissue and to the embryo in soyabean seed (Moore, 1971). Floris (1970) concluded that ageing is a progressive process accompanied by accumulation of toxic metabolites, which progressively depress germination and growth of seedlings with increased age.

CHAPTER 3

MATERIALS AND METHODS

3.1 Experimental Site

The experiment was conducted during the 1998/99 cropping season at the Field Station of the School of Agricultural Sciences, University of Zambia. The site is located in the agro-ecological zone II of Zambia, and located at 15° 23' South and 28° 20' East and is about 1140m above sea level (Chikuma *et al.*, 1985). The site receives annual rainfall of between 800 and 1000mm which is generally well distributed. The mean daily temperatures at this location range from 23° to 25° during the growing season and with a sandy loam soil type (MAFF, 1995).

The previous crop grown during the 1997/98 growing season was Bambara groundnuts (*Vigna subterranea* (L.) Verdc.) which was not fertilised.

3.2 Experimental Design and Treatments

Two factors (harvest date and variety) were studied in different combinations using a split-plot design with four replications. The factor of variety was assigned to the main plots and the harvest dates were randomly allocated to subplots. Each subplot had an area of 4m x 2.5m ($10.0m^2$) consisting five rows with an inter- row spacing of 60cm.

There were 12 treatments used in the study, i.e, 2 varieties of soyabean, (Hernon 147 and Kaleya) and 6 dates of seed harvest. The harvesting dates were at one week interval starting at 7 days after the date of 50% podding (DAP). Thus, harvesting was done at 7, 14, 21, 28, 35 and 42 DAP.

The two varieties used in this study were chosen based on similarities in their growth characteristics. They have determinate growth behaviour and semi-erect growth habits. Both are adapted to agro-ecological region II of Zambia where the experiment was conducted. The only difference however, is that Hernon147 is promiscuous while Kaleya requires inoculation with *Rhizobium* in order to nodulate.

3.3 Cultural Practices

3.3.1 Planting

The seed was sown by drilling on 23 rd December, 1998 at a depth of 4cm. Thinning of all plots was done one week after emergence of plants. Approximately 30 plants per meter were left within each row (Wellving, 1984). Before planting, three soil samples were collected from various points in the plot using an auger up to a depth of 25cm. These soil samples were then mixed to make a composite sample which was analysed for nitrogen, phosphorous, potassium and pH (CaCl). Standard procedures as outlined in the Practical Manual for Plant Nutrition (Songolo, 1993) were followed for these analyses.

3.3.2 Seed inoculation

Before planting, seed of Kaleya variety was inoculated with a peat –based inoculum, *Rhizobium japonicum,* obtained from Mount Makulu Research Station. Standard procedures were followed in treating the seed, that is, seed was mixed with sugar and water and then inoculum was added and mixed evenly with the seed (Wellving, 1984). Seed of variety Hernon 147 was planted directly without inoculation since it is a promiscuous variety, which performs well without inoculation..

3.3.3 Management of Experiment

The experimental management activities undertaken included weeding and fungicide spraying. Weeding was done by hand-hoeing at four and seven weeks after planting in all the plots.During the growth of the soyabean crop, there was an infestation of Frog eye leaf spot (*Cercospora spp.*) which was controlled by spraying the plants with Zoom(50% Carbendazim, wp) a broad spectrum fungicide, at a rate of 13.9g/l of distilled water.

3.3.4 Seed Harvesting

The seed was harvested manually by picking the pods from the appropriate plots (Tekrony *et al*, 1979). Harvesting was done only from the three middle rows leaving the two border rows. Starting at 7 days after podding, seed was harvested at 7-day intervals and approximately 1kg of seed was harvested at each time. All seeds were removed from pods and air dried at room temperature. The seeds were dried on top of the paper spread on benches (Tekrony *et al*, 1979).

3.4 Data Collection and Analysis

3.4.1. Days to 50% Flowering and 50% Podding

Regular checks were done in the field after emergence in order to monitor the flowering of the plants. The first date of flowering of the plants and the subsequent dates of flowering were observed and recorded. The date of 50% flowering was taken to be the date when 50% of the plants in a particular experimental plot had flowered. The same procedure was followed when determining the date of 50% podding.

3.4.2. Determination of Physiological Maturity

The stage of physiological maturity was determined by monitoring the dry matter content of seeds harvested at different stages. This was determined by monitoring the dry matter content of the seed at each harvest. A sample of 25 fresh seeds was weighed and then pre-dried in open air. After pre-drying, seeds were ground and then further dried in an oven at 104 °C to a constant weight in order to determine their dry matter.

The stage at which there was no further increase in the dry matter content of the seed was taken to be the point of physiological maturity.

3.4.3 Initial Moisture Content

This was determined at harvest using the low-constant temperature oven method (ISTA, 1985). Seven grams of seed was weighed from the harvested lot then pre-dried at 130 °C for 20 minutes and then further dried at 104 °C for 24 hours. Seed moisture content was then calculated as outlined in Appendix 7.A1 (ISTA, 1985).

3.4.4. Germination of fresh and dry seeds

Germination was conducted on fresh seeds(with initial moisture, not dried), for the determination of the stage of onset of germination and on dried seeds. Seeds were treated in four replicates which had 25 seeds each. The seeds were germinated on filter papers at 22 °C for 8 days (ISTA ,1985); Appendix 7.A2). For the germination of dry seeds, the same procedure was followed as that of moist seeds (Appendix 7.A2). In this case, the seeds were dried to 12-13% moisture before the test (ISTA, 1985).

3.4.5 Seed Vigour

This was done at each harvest by recording the electrical conductivity of a sample of 25 seeds soaked in 100ml of distilled water for 24 hours, following the procedure of Tekrony *et al* (1979). This procedure is outlined in Appendix 7.A3.

3.4.6 Potential Longevity

This was done using the accelerated ageing test. The 25 seeds in four replications were humidified to 21% moisture content between two pieces of mutton cloth. These seeds were then hermatically sealed in aluminium foil packets and stored at 42° C (Tekrony and Egli, 1977). Samples were then taken out at regular intervals of 1, 2, 3, 4, 5, 6 and 7 days and tested for germination and vigour.

3.4.7 Meteorological Data

Agro-meteorological data on rainfall, temperature and relative humidity for the 1998/99 growing season was obtained from the Geography Department of the University of Zambia. The data was used to determine if the year during which the experiment was conducted experienced normal seasonal weather patterns (Appendix 7.D).

3.4.8 Statistical Analysis

All the data collected for the parameters measured were subjected to Analysis of variance and the resulting means were separated at 5% probability levels using Duncan's multiple range test. For data that was expressed as percentages, a test for normality was done and then the data was transformed from percentages to degrees using Angular transformation.

CHAPTER 4

RESULTS

4.1 Soil Analysis Results

The results of the soil analysis (Table 1.) show that the soil had sufficient amounts of nitrogen, phosphorous and potassium for the growth of soyabeans. The pH of the soil was also within the recommended range for soyabean cultivation (MAFF,1995). The result of the soil test indicated that there was no need to make any amendments in terms of the nutrients that were analysed .

4.2 Time to 50% Flowering and 50% Podding

The time to 50% flowering and 50% podding are given in Table 2. The results for 50% flowering show that Hernon 147 and Kaleya had achieved 50% flowering at 60 and 62 days after sowing, respectively. The results show that Hernon-147 achieved 50% flowering significantly ($P \leq 0.05$) earlier than Kaleya. Time to 50% podding was achieved at 69 and 70 days after sowing for varieties Kaleya and Hernon-147, respectively. Although Hernon -147 achieved 50% flowering two days earlier than kaleya, it took longer ($P \leq 0.05$) (by one day) to achieve 50% podding as shown in Table 2.

Table 1. Chemical Analysis of Soil for Nitrogen, Potassium, Phosphorous and pH for the Site of the Experiment.

NUTRIENT	*AMOUNT IN SOIL (*	*OPTIMUM RANGE*(kg/ha)*	*COMMENT*
PH	6.69	5.8 to 7.0	Within Range
Phosphorous	22kg/ha	20- 40kg/ha	Sufficient
Potassium	30kg/ha	20 - 40kg/ha	Sufficient
Nitrogen	0.049%	0.03% to 0.4%	Sufficient

* MAFF, 1995.

Table 2. Field observations of the Time to 50% Flowering and 50% Podding of Hernon-147 and Kaleya Varieties of soyabeans.

Treatment	Time to 50% Flowering	Time to 50% Podding
V1H1	62a	69b
V2H1	60b	70a
V1H2	62a	69b
V2H2	60b	70a
V1H3	62a	69b
V2H3	60b	69b
V1H4	62a	69b
V2H4	60b	70a
V1H5	62a	69b
V2H5	60b	70a
V1H6	62a	69b
V2H6	60b	70a
CV%	0.23	0.026

Means in a column followed by the same letter are not significantly different ($p \leq 0.05$)

Where: V1 = Kaleya, **V2** = Hernon-147; **H1** = 7DAP, **H2** = 14DAP, **H3** = 21DAP,

H4 = 28DAP, **H5** = DAP and **H6** = 42DAP.

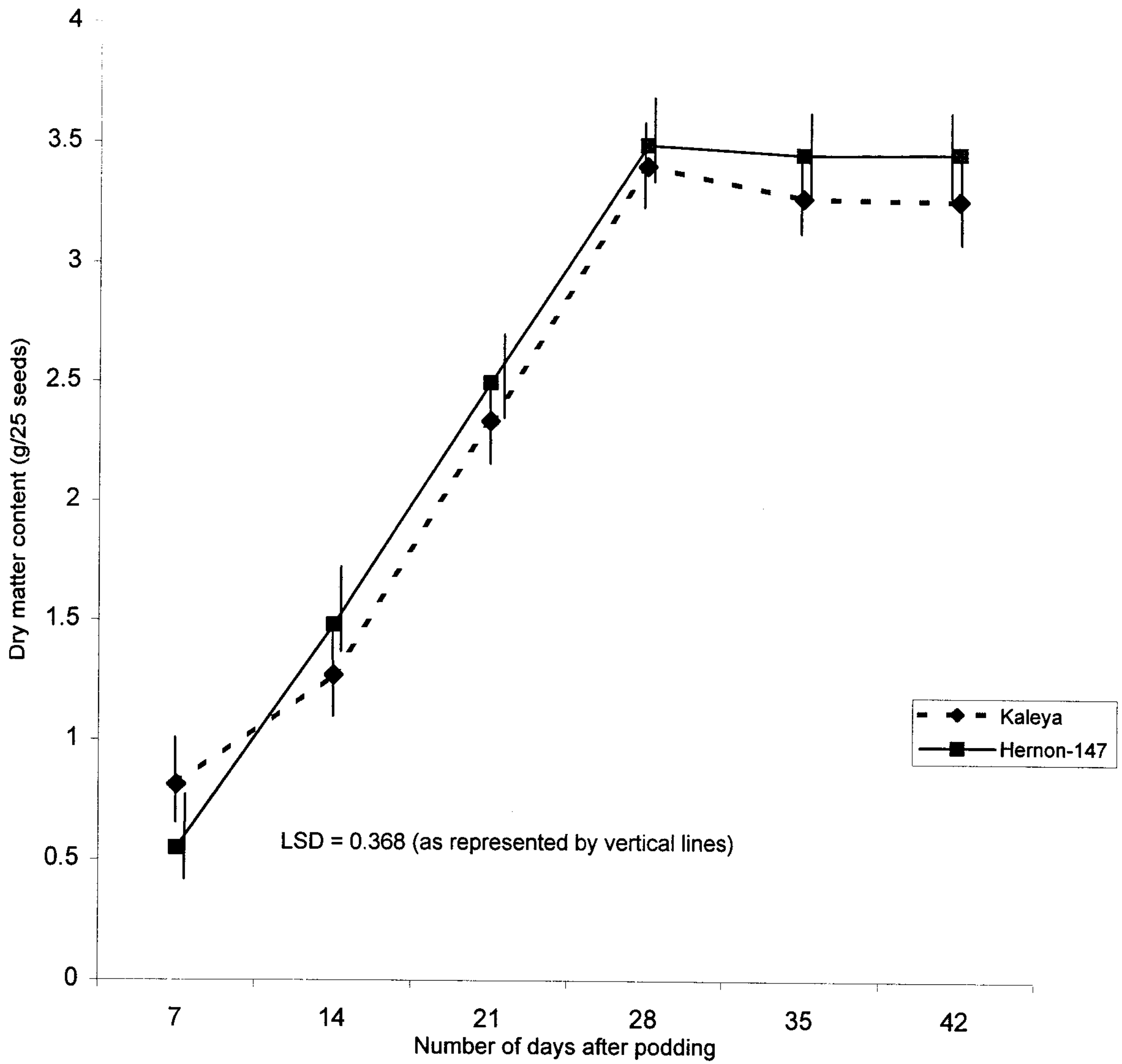
4.3 Seed Quality Parameters

4.3.1 Seed Dry Matter

Time of harvest of seed had a significant effect on the dry matter content of the seed ($P \leq 0.05$). The dry matter of the seeds increased with later harvest until the point of physiological maturity (Figure 1., Appendix B.2). There was a strong and positive relationship between time of harvest and dry matter content, giving coefficients of correlation (r) of $0.88^{\bullet}$ and $0.93^{\bullet}$ for Kaleya and Hernon-147, respectively. The dry matter content stabilised at 28 days after podding and no significant increase in dry matter contents were observed within each variety after this stage (Figure 1.). Therefore, the point of physiological maturity was found to be at 28 days after podding for both varieties in this study. Seed dry matter was significantly affected by variety (Appendix B.2).

$^{\bullet}$ $P \leq 0.05$

Figure 1. Accumulation of Dry matter in seeds of Kaleya and Hernon-147 during development.



The dry matter content of seeds of Hernon-147 was higher than that of the seeds of Kaleya from 14 DAP onwards. This higher dry matter content maintained in Hernon-147 accounts for the higher correlation coefficient in this variety (between seed dry matter and time of harvest) in comparison to that for Kaleya. Although this difference in dry matter for the two varieties was significant ($P \leq 0.05$), the time of attainment of physiological maturity was at 28 days after podding for both varieties. Despite the significant effect of variety on dry matter content of seeds, the interactive effect between variety and time of harvest on dry matter content was not significant (Appendix 7B.2).

4.3.2 Seed Moisture Content

Harvest time had a significant effect on the moisture content of the seeds at harvest. Seed moisture was high at 7, 14, 21 and 28 DAP. Generally, there was a progressive and significant decline in seed moisture content with time (Table 3). This gave a negative correlation between seed moisture content and time of harvesting with a correlation coefficient (r) of -0.961*.

Seeds of Hernon-147 generally maintained slightly higher moisture levels during the harvest period. The overall effect of variety on the initial moisture content of the seed at harvest, however, was non-significant. The interaction of variety with time of harvest was also non-significant (Appendix 7B.1).

---

*$P \leq 0.05$

Table 3. Effect of Harvest time on the seed moisture content

Harvest Time (DAP)	7	14	21	28	35	42
Kaleya	78.3a	69.14b	67.3b	53.9c	20.4d	14.2e
Hernon-147	80.18a	70.2b	68.7b	54.6c	26.2d	13.6e

Mean = **51.44**; CV% = **44.9**

** Means within a row followed by the same letter are not significantly different from each other.*

4.3.3 Germination of Fresh and Dried Seeds

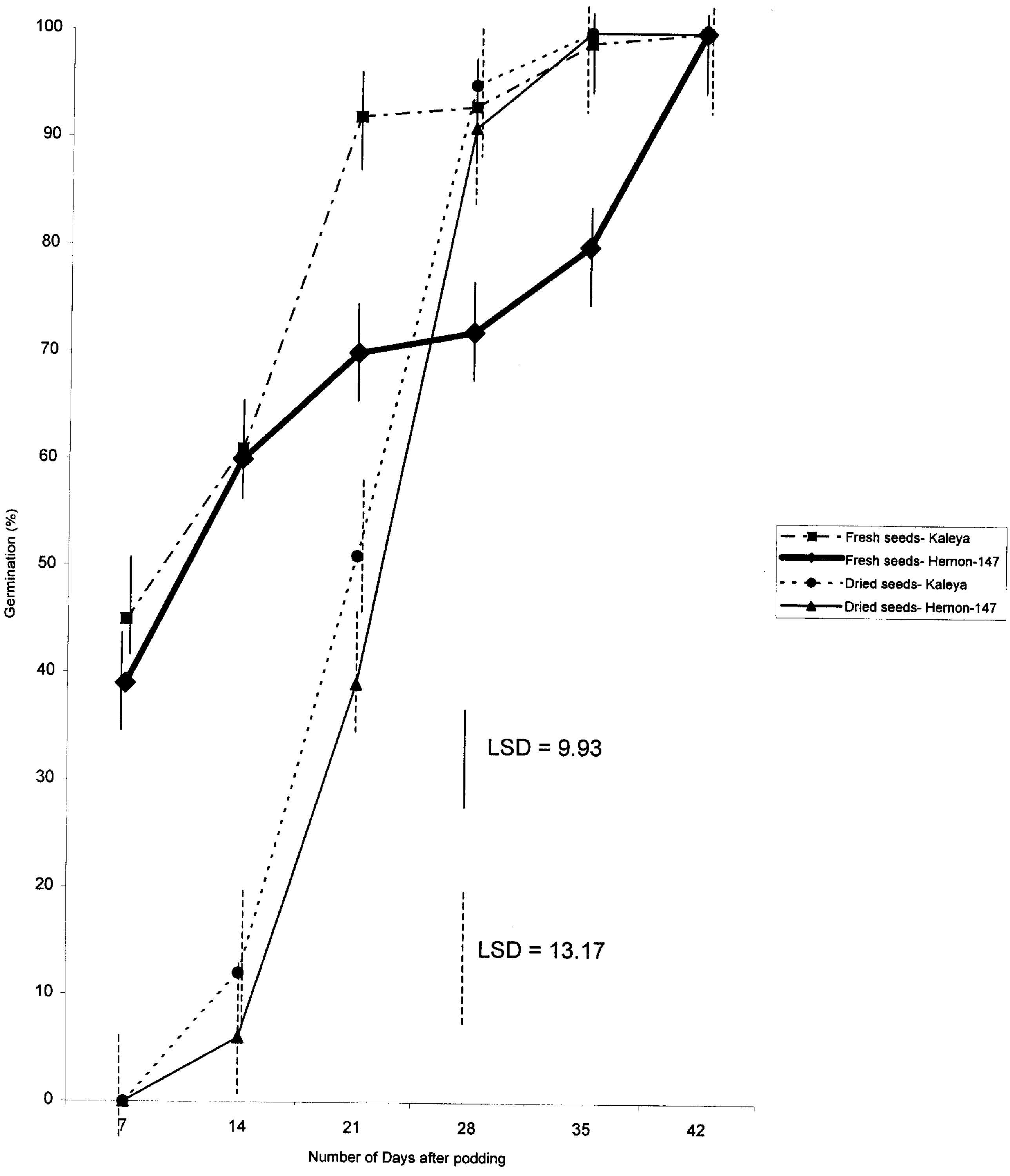
Following the germination of fresh and dried seeds results presented in Figure 2 were obtained. The time of harvest had a significant effect on the germination capacity of fresh seeds (Appendix B.3). In both varieties, fresh seeds were able to germinate as early as 7 days after podding (43% germination), when DMC of the seeds was 0.023g/ seed (Fig.2). At this stage, seeds had attained only 17% of their final dry weight. Variety had a significant effect on the number of fresh seeds that germinated. The germination level of fresh seeds of Hernon-147 was always significantly lower than that for Kaleya Variety (Figure 2), particularly between 21 and 35 DAP. Despite this difference, the germination levels of fresh seeds of the varieties became identical at 42 DAP. The harvest time by variety interaction also had a significant effect on the germination of fresh seeds(Appendix B.3)

.

The time of harvest of seeds had a significant effect on the germination level of dried seeds (Appendix B.4). There was an increase in the final germination of dried seeds with delay in harvesting of seed (Fig.2). This resulted in a positive relationship between time of harvest and level of germination of seeds of both varieties ($r=0.939^{\bullet}$ and $0.961^{\bullet}$ for Kaleya and Hernon-147, respectively). For example, at 7 days after podding, dried seeds of both varieties failed to germinate while at 14 DAP germination increased to 8.1% and continued to do so with time (Figure 2).

$^{\bullet}P \leq 0.05$

Figure 2. Germination of fresh and dried seeds of Hernon-147 and Kaleya varieties as affected by growth.



The germination of fresh seeds was generally higher than that of dried seeds until the attainment of physiological maturity (this applies only to Kaleya seeds), at which stage there was no significant difference inthe germination levels of the seeds. For example, at 14 and 21 DAP, the germination level of fresh Kaleya seeds was 60% and 92%, respectively, while that of dried Kaleya seeds was only 8.1% and 62%, respectively. However, Kaleya seeds generally maintained higher germination levels for both fresh and dried seeds than Hernon-147 throughout the period of harvest.

The results show that variety of seed significantly influence the germination level of fresh seeds at all stages of harvesting, but this was not true for dried seeds (Appendix B.4). The harvest time by variety interaction was not significant with respect to germination of dried seeds (Appendix B.4).

The point of physiological maturity (28 DAP) did not coincide with the point of highest germination for both varieties which fell at 35 and at 42 DAP (Figures 1. and 2). The results further show that germination levels of dried seeds were significantly different for each stage of seed harvesting except for 35 and 42 DAP at which stages the germination of dried seeds of both varieties were equal (Figure 2). Although germination in each of the two varieties was constant at 35 and 42 DAP, the average moisture content of the seeds at 35 DAP (23.3%) was significantly higher than that of seeds harvested at 42 DAP (13.9%).

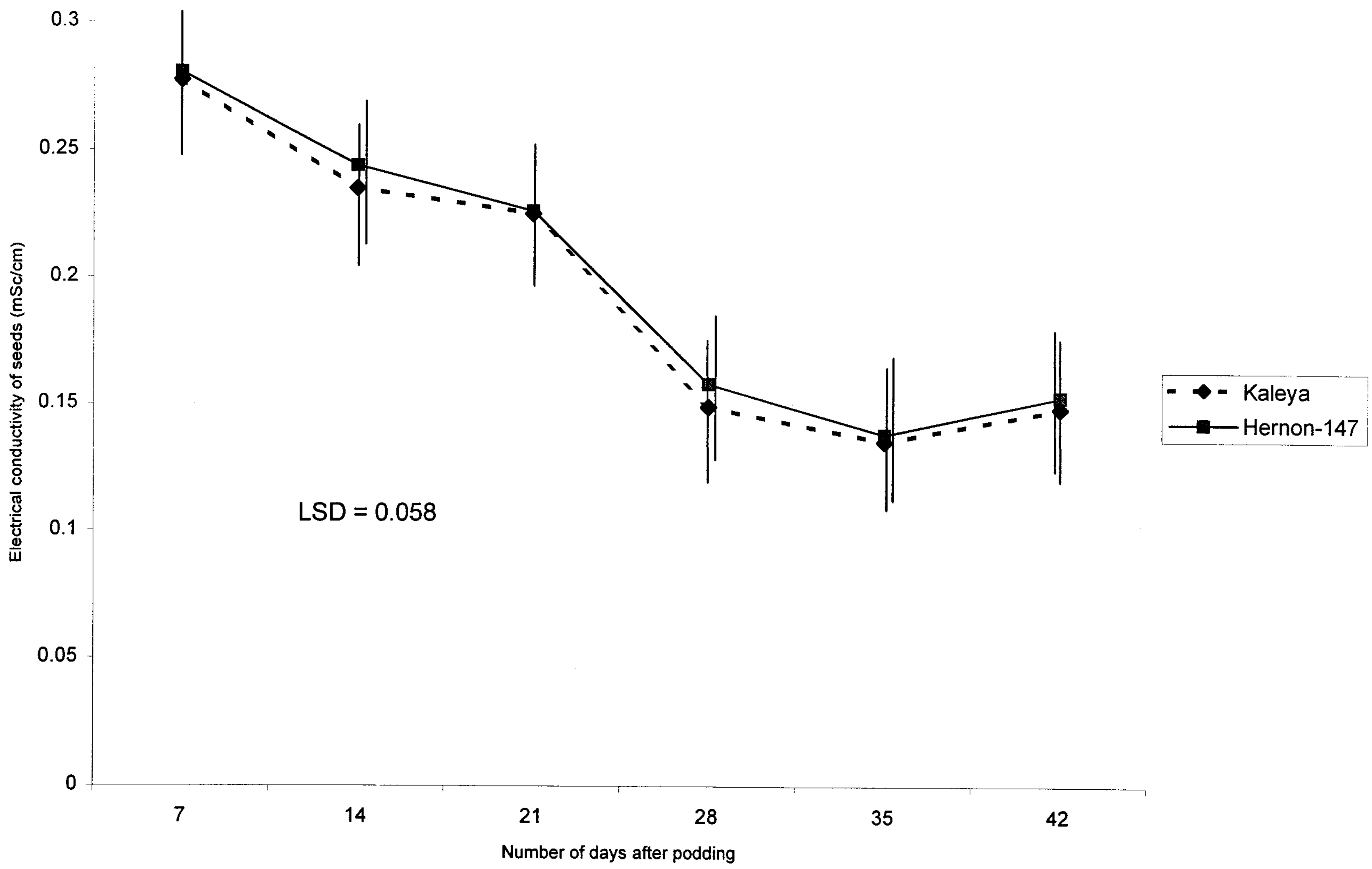
4.3.4 Vigour of fresh seed

This was measured using the electrical conductivity (EC) of the leachate. The results presented in Figure 3 show the trend in conductivity of the leachate from seeds harvested at different stages of growth. EC of seed leachate decreased with increase in DAP.

Time of harvest had significant effect on the conductivity of the leachate from the seeds ($P \leq 0.05$; Appendix B.5).Variety and variety by time of harvest interaction had no significant effect.

The conductivity of leachate from the seeds harvested at 7 (H1), 14 (H2) and 21(H3) DAP was not significantly different from each other. Similarly, the electrical conductivity values of seeds harvested at 28, 35 and 42 did not differ significantly either. Vigour remained very low (as indicated by high EC values) from 7 to 21 DAP. There was a distinct increase in vigour (low EC reading) at 28 DAP (physiological maturity stage) and remained high up to 42 DAP as shown in the Figure 3. Though the vigour of Hernon-147 seeds was generally lower than that of Kaleya seeds, this difference was non-significant ($P \leq 0.05$; Appendix 7B.5). The interaction between variety and time of harvest did not have a significant influence on the vigour of the seeds as measured by the EC.

Figure 3. Initial Vigour of Hernon-147 and Kaleya seeds harvested at different stages of growth.



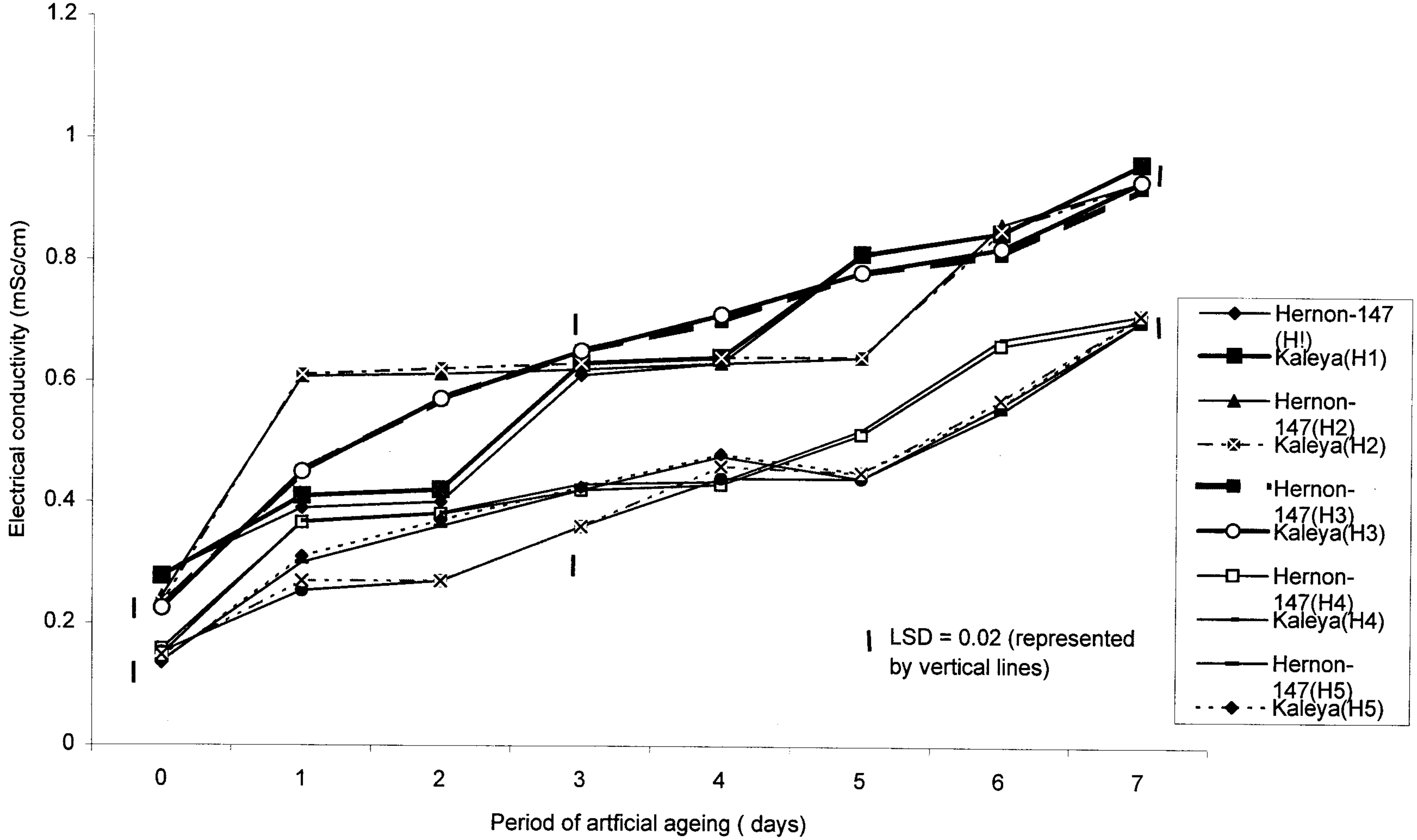
4.4. Accelerated Ageing Test

4.4.1 Vigour of artificially aged seeds

The results of the vigour test of artificially aged seeds are presented in Figure 4. Variety did not have a significant effect on vigour of aged seeds, however, harvest time, days of artificial ageing, and variety by harvest time, variety by harvest time by days of artificial ageing, variety by days of artificial ageing and harvest time by days of artificial ageing interactions were all not significant (Appendix B.6)

The initial seed vigour (measured by EC) was significantly higher than that of the aged seeds harvested at different stages The average electrical conductivity value of the leachate from the seeds that were not aged was 0.185mSc/cm. For the artificially aged seeds, the electrical conductivity values of their leachate ranged from 0.377 to 0.78mSc/cm for the 1 to 7 days of ageing. The results show a rapid decline in vigour (i.e. increase in EC) with artificial ageing of seeds. One day of artificially ageing, resulted in a rise in conductivity from 0.185 to 0.378 mSc/cm. Seed vigour at and beyond the stage of physiological maturity (28, 35 and 42 DAP) was higher than that of the seeds harvested before physiological maturity (Figure 4). In terms of the EC of seed leachate, seeds harvested before physiological maturity showed significantly higher EC levels as compared to those harvested at and after physiological maturity after being aged for the same number of days.

Figure 4 Electrical conductivity of artificially aged seeds harvested at 7 (H1), 14(H2), 21(H3), 28(H4), 35(H5) and 42 (H6) days after podding. The stage of physiological maturity of both varieties is 28 DAP



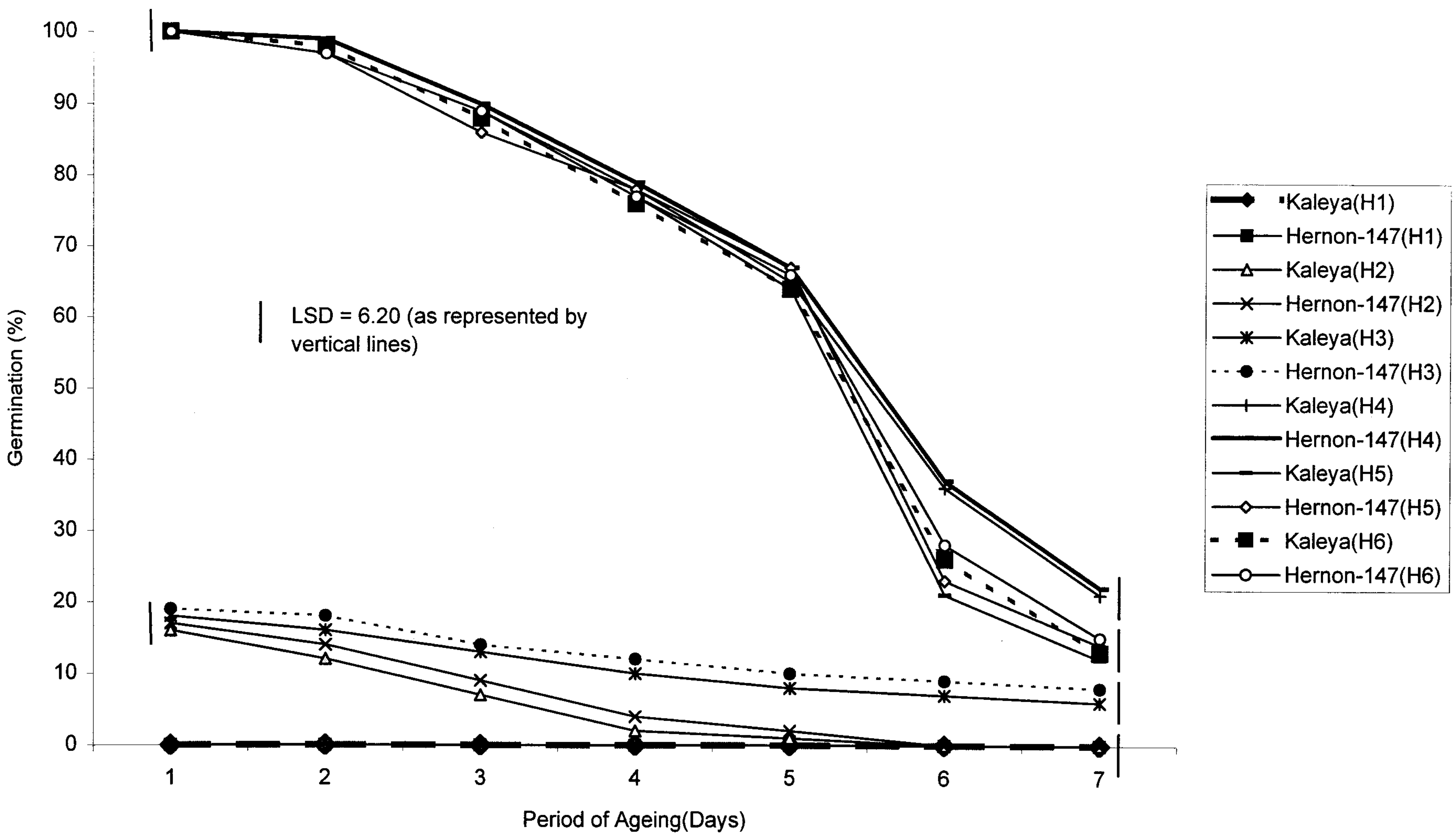
4.4.2 Germination of Artificially aged seeds

Figure 5 shows the results obtained from the germination test of the artificially aged seeds of both soyabean varieties. Germination of artificially aged seeds depended on the time of harvest i.e., harvest time had significant effect on germination of artificially aged seeds(Appendix B.7).Days of artificial ageing and days of artificial ageing by harvest time interaction was also significant (Appendix B.7)

Seeds harvested at and after the attainment of physiological maturity (28, 35 and 42 DAP) gave relatively higher germination after ageing than those harvested before this stage (7, 14 and 21 DAP).

When aged for the same number of days, seeds harvested at and after the attainment of physiological maturity had higher germination values than those harvested earlier. Seeds harvested at 28(H4), 35(H5) and 42(H6) days after podding gave high germination after being aged for 4 and 5 days. On the other extreme, seeds harvested 7 days after podding did not germinate at all after ageing (figure 5). Poor germination (10% and 5%), was observed when seeds were aged for 6-7 days, although the germination of seeds harvested at 28 DAP still gave 36 and 21 % after 6 and 7 days of ageing (Figure 5), respectively. There was no significant effect of variety on the germination of aged seeds, neither was there significant interaction between harvest time, period of ageing and variety or variety by days of ageing (Appendix B.7).

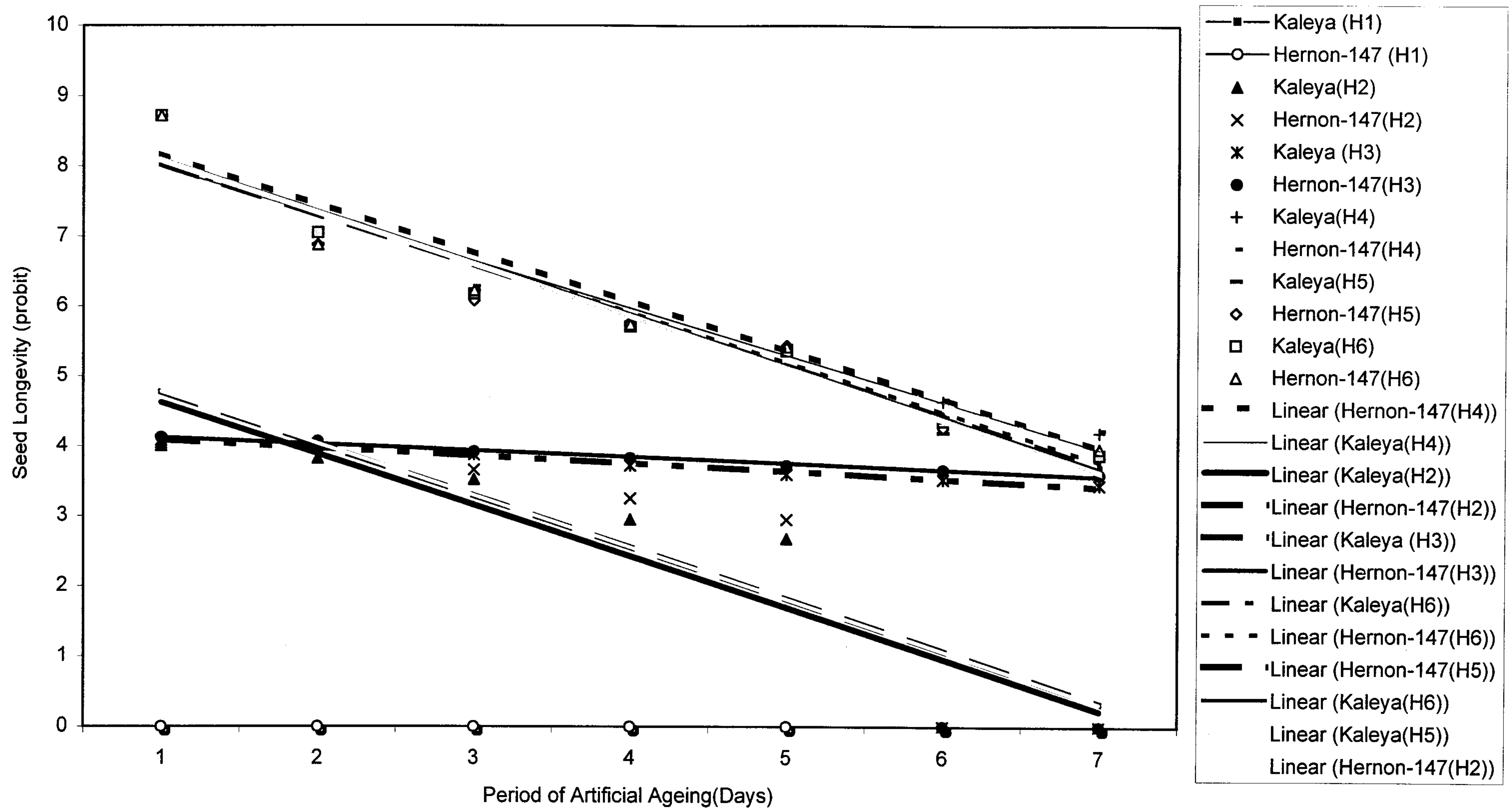
Figure 5. Germination of Artificially Aged seeds of Hernon and Kaleya harvested at different stages of growth.



4.4.3. Seed Longevity

The results in Figure 6 show that seeds harvested at 7DAP were not viable after ageing (1-7 days) and had no potential for storage (both Kaleya and Hernon-147). The longevity values of both varieties were zero. After ageing the seeds for 1-7 days, the seeds harvested at 14 DAP had potential longevity (probit) values declining from 4.00 to 0 (4 units), whilst the longevity for those harvested at 21 DAP declined from 4.12 to only 3.60 (0.6 units). Potential longevity of seeds harvested at and after physiology maturity (28, 35 and 42 DAP) was higher than that of seeds harvested before this stage in both varieties. The probit values of seeds harvested at 28, 35 and 42 DAP ranged from 8.72 to about 4.0 after ageing for 1-7 days. These values are higher than the 4.12 to 3.6, 4.0 to 0 and 0 probits observed at 21 ,14 and 7 DAP respectively.

Figure 6. Longevity of Kaleya and Hernon-147 seeds harvested at different stages of growth.



CHAPTER 5

DISCUSSION

5.1 INITIAL SEED QUALITY

Dry matter content, onset of germination, physiological maturity and germination

In this study, physiological maturity of soyabean seed was attained at 28 days after podding (Figure 1). With respect to flowering, physiological maturity was attained at 41 days post-flowering. The mean dry matter content of the seeds for both varieties at this stage was 3.348g / 25 seeds (0.139 g/ seed). Both Kaleya and Hernon-147 attained physiological maturity at the same stage (28 DAP). This may suggest that at this stage the seed had gone through all the physiological changes (such as cell production and organisation, and embryo growth) that are characteristic of developing seeds. The dry weight of the seeds of the two varieties at physiological maturity when compared to those observed by Tekrony *et al* (1980) (4.25g/25 seeds i.e., 0.17g/seed) and Marcos - Filho *et al* (1994) (4.08g/25 seeds i.e., 0.16g/seed), were relatively low. The differences in the maximum seed dry weights have generally been said to be related to environmental conditions and genotype which dictate the final seed size (Teaser, 1984). The lower dry weights of the seeds could therefore be attributed to varietal differences.

Although the results of Tekrony *et al* (1980) and Marcos - Filho *et al* (1994) show higher seed weights, they seem to agree with the results from this study that the maximum weights could be attained at 41 days after flowering.

Other reports by Kato *et al.* (1954) and Fraser *et al.,* (1982) show that soyabeans pods may attain maximum size within 20 days of flowering, but maximum seed weight is not reached until 30 to 40 days after flowering. This finding agrees with the results from this study in which maximum weight was attained at 41 days after flowering.

The accumulation of dry matter in the seeds from 7 to 28 days after podding was gradual in Hernon-147 variety. Dry matter content related positively with time of harvest giving correlation coefficients of 0.88 and 0.93 for Hernon-147 and Kaleya, respectively. The observed increase in germination of seeds for both varieties could therefore be associated with the increasing dry matter content with time of harvest. This may mean that the accumulated dry matter was acting as a principle source of food during germination and the early stage of seedling development. Furthermore, it may suggest that Hernon 147 and Kaleya varieties harvested at physiological maturity stage and 14 days later may give seeds of higher germination since the dry matter contents are maximum during this period. It follows, therefore, that a period of 21 days (i.e. from 28 to 42 DAP) may exist during which seeds of good germination could be harvested as long as there are no other limiting factors during this period. This may imply that the recommended time of harvest for commercial (which in this study may correspond to 35 DAP) is suitable for the harvest of seed meant for germplasm.

The progressive improvement in the germination of seed during development observed in this study may be attributed to a number of changes that occur in the seed. Among these changes is the accumulation of large amounts of storage material such as fats, carbohydrates, protein and phytin, which has been reported to change the overall metabolism of the seed (Mayer, 1973). It has been reported that seed embryo mass is the major determinant of seed viability and vigour for soyabeans (Adams and Rinnie, 1981). The large size of the seed indicates a large storage capacity, showing that seed size and seed weight have a role in germination (Lungwangwa, 1993; Powell, 1998).

However, it has also been observed that other factors play a major part in germination and that dry matter content is not the sole determinant of the level and rate of germination. Uncertain relationships have been shown to exist between seed size (and weight) and germination (Marcos - Filho *et al.,* 1994).

In this study it was observed that germination of fresh seeds was possible at 7 days after podding (43%) when the dry matter of the seeds was 0.023g/seed. At this stage the seeds had accumulated only 17% of the maximum dry weight observed at physiological maturity. It may appear that lower dry weights may allow some level of germination though this may imply lower levels of germination. This may suggest therefore that germination could commence earlier than 7 days after podding, though this has not been documented before and therefore, may need experimental verification. It has beeen observed, however, that germination is possible at 12 days after podding (Clifford*et al.*, 1981). Dry matter accumulation of 25% of the maximum dry weight is has been suddested to be sufficient for germination of soyabean seed (Egli and TeKrony, 1993). The results suggest that germination is possible at lower dry matter weights than those observed by the earlier workers. However, even if the seeds were able to germinate at this stage, harvesting at such an early stage would not be practical. The seeds at this stage were too small to be harvested and they could not go through dessication without losing viability.

Seed moisture content and germination

Germination began when the moisture content was about 79%. After drying the seed to 12-13 % moisture content, it was observed that germination dropped to 0%. This failure of dried seeds to germinate could be an indication of dessication intolerance. Immature soyabean seeds which are shelled and air dried have been reported to have poor viability (Adams *et al.*, 1983). The harvested immature seeds could have gone through rapid drying, which did not allow for maturation to occur. Maturation of seeds has been shown to be essential for viability (Adams *et al*, 1983).

The inability of these seeds to germinate after drying may be said to be because the seeds had not yet attained the dessication tolerance stage. Loss of water from the seed while maintaining viability is a characteristic feature of seed maturation, and is rapid especially during the later stages of development (Mayer, 1973). In this study, the moisture content at which the seeds attained tolerance to dessication was around 70%. This was after some natural seed drying from the initial 79%. The observed moisture level at which seeds could further be dried seems to be slightly higher than the earlier observed 55-65% for soyabeans (Ellis *et al.*, 1987).

The improvement in germination with time of harvest that was observed could be explained by the structural changes at microscopic and submicroscopic levels that accompany the loss of moisture (Mayer, 1973). This loss of water has been shown to be accompanied by the increase in resistance of the seed to adverse external conditions and consequently, increased germination (Klein, 1970). It has also been shown that seed dessication in nature is meant to reduce metabolic activities within the seed and preserve the viability. It has otherwise been observed that this maturation process may not be accompanied by any dramatic changes in the activities of various enzymes (Adams and Rinnie, 1981). At 14 days after podding, germination of fresh seeds improved in both varieties (to about 52.5%). However, at 21, 28 and 35 days after podding, the germination of fresh Hernon-147 seeds was significantly lower than those of Kaleya seeds and of the dried seeds of both varieties (Figure 2). It appears that germination of Hernon-147 seeds was improved by drying and this also seems to be a varietal characteristic. This behaviour was observed especially before the attainment of physiological maturity. Gbikpi and Crookston (1981) observed that during natural seed development and drying on the plant, pod to pod (seed to seed) variations may occur with respect to germination especially before physiological maturity leading to reduced germination levels, and this characteristic is more pronounced in certain genotypes than in others(Thompson, 1979).

This seed to seed variation may account for the low germination observed especially considering that the seeds that were subjected to the germination test was a random sample. This suggests that artificial drying of the seed could have had an effect on the seeds which improved the overall germination or perhaps reduced the seed to seed variation existing in the fresh seeds. Despite the lower germination of fresh Hernon-147 seeds at the three stages (12, 28 and 35 DAP), good germination of fresh seeds has been reported (Miles*et al,* 1988) at similar growth stages.

Seed vigour

The results obtained show that the electrical conductivity decreased as the days after podding increased. This could be because at the early stages of development e.g., 7, 14 and 21 days after podding, the organisation of the cellular membrane integrity was still in the initial stages. Consequently, more exudates are expected to leak out of these seeds at these stages than at any later stage of seed development since it has been suggested (Marcos- Filho *et al,* 1994) that the organisation of the membrane structure in seeds is completed just before physiological maturity, during seed maturation. This explains the lower conductivity obtained at 28 days after podding, which is the point of physiological maturity. The fact that there were no significant differences in the electrical conductivity of the exudates after the stage of physiological maturity could be an indication that deterioration of the cell membrane structure had not begun even at 35 and 42 days after podding. This is with the view that the first deterioration event is the loss in membrane intergrity. The maintenance of soyabean seed membrane structure beyond the stage physiological maturity (28 DAP) reported here is at variance with idea advanced by Harrington (1927) that seed vigour declines soon after physiological maturity.

5.2 SEED LONGEVITY

The seeds with high initial viability gave rise to higher germination after artificial ageing and the opposite was true (Figure 5). The results, therefore, mean that harvesting seeds close to or at physiological maturity could yield seeds of higher potential for storability in seed gene banks. From the germination results, it is therefore possible to judge the potential storability from the initial seed viability. The results further show that harvesting seeds at physiological maturity and within 14 days after this stage yields seeds that can be stored for longer periods than those harvested earlier. This observation was further confirmed by the actual longevity values (probit) which were observed. The probit results shown in Figure 6 suggest that seeds aged for 1-7 days were superior in terms of potential longevity if harvested at and after the attainment of physiological maturity. The longevity values for seeds harvested at 28, 35 and 42 days after podding ranged from 8-4 probits as opposed to that for the seeds harvested prior to physiological maturity which ranged from 0- 4.12 probits.

The results of the vigour test show that seeds harvested at or close to physiological maturity gave rise to high initial vigour and to high final vigour after artificial ageing. There was increased electrical conductivity observed after ageing of seed regardless of the harvest time. This could be attributed to the fact that during the ageing period, when seeds are subjected to high temperature and high humidity , there was either some development of dead tissue on the cotyledons, or the cell membrane integrity of the living tissue had deteriorated (Powell, 1988).

Generally, the integrity of cell membranes, as determined by deteriorative biochemical changes or physical disruption, can therefore be considered to be the fundamental cause of differences in seed vigour (Abdul-Baki and Anderson, 1972).

These results are consistent with those obtained by Perez and Arguello (1995), who observed increased leakage of solutes after artificially ageing soybean seeds. The results suggest a direct and positive relationship between initial seed vigour and potential storability. The results mean therefore that seeds harvested at physiological maturity and 14 days after this period could be stored for longer periods than those harvested earlier. Unlike reports by Walters and Engels (1998)and Moore (1971), the results suggest that both the germination test and the electrical conductivity test are good assays in predicting the potential storability of the seed. However, the results show that at physiological maturity the seed moisture content was as high as 54%, which may not be practical for harvesting seed on a large scale. When seed is to be stored as germplasm, it necessary to pay more attention to the quality and the storability of seed than to the actual quantity. This means that if the seed at this stage, has the highest ability to be stored, small amounts of seed, though high in moisture could be harvested, dried and stored.

The results also show that the seed still has the high initial viability and vigour, and potential storability at 35 and 42 days after podding. This means that in cases where artificial drying is not possible, the seed can still be left in the field for 14 days beyond physiological maturity and then harvested and stored for equally long periods. According to MAFF (1995), recommendation, the commercial soyabean crop should be harvested when the pods feel brittle and moisture has fallen to around 16 to 12 %. The results may suggest that this recommended period falls between 35 and 42 DAP, and within a few days after 42 DAP. This implies that the current recommendation for harvest of soyabeans seeds grown commercially is also useful for harvest of soyabean seed grown for germplasm conservation. The results of both initial seed quality and seed longevity seem to suggest a turning point at physiological maturity. The seed quality and potential longevity seems to be best at and after the attainment of physiological maturity. It appears that a number of physiological changes take place at physiological maturity which render the seed more superior. It has been observed that by the stage of physiological maturity all the lipid, protein and other biosynthetic activity in the cotyledon is complete and certain marked changes in cell organisation and structure are also evident at this stage. Apart from these changes, the embryo of the seed is fully grown by then (Teaser, 1984). The full growth of the embryo could be the main reason for the difference between physiologically immature and mature seed.

CHAPTER 6

CONCLUSION

Seeds of Hernon 147 and Kaleya soyabean varieties acquire the ability to germinate at 7 days after podding when the dry matter content is only 17% of the final dry matter content. The ability to germinate after drying was attained at 14 days after podding was observed.

Potential storability of Hernon 147 and Kaleya seed is affected by time of harvest since it affects the initial seed quality. Longevity of seeds of both varieties improved as harvesting was delayed.

It can also be concluded that harvesting at physiological maturity or 14 days after this stage can yield best seed in terms of initial seed viability and vigour and high potential storability for Hernon 147 and Kaleya. This means that the current recommendation for harvest of soyabean seed crop in Zambia may serve as a guide as to when to harvest Hernon 147 and Kaleya seed meant for long term storage.

REFERENCES

AOSA. (1983), Seed vigour testing handbook. Springfield.

Anderson, J.D. (1970). Physiological and biochemical differences in deteriorating barley seed. *Crop Science*, 10: 36-39.

Adetunji, A.I. (1991). Effect of harvest date on seed quality and viability of sunflower in semi-arid tropics. *Seed Science. and Technology*, 19:571-580.

Adams, C.A. and Rinnie, R.W. (1981). Seed maturation in Soyabeans is independent of seed mass and of the parent yet necessary for production of viable seeds. *Journal of Experimental Botany*, 32: 650-657.

Adams, C.A., Fjerstad, M.C. and Rinnie, R.W. (1983). Characteristics of soyabean seed maturation: Necessity for slow dehydration. *Crop Science*, 23:265-267.

Abdul- Baki, A.A. and Anderson, J.D. (1972). Physiological and Biochemical deterioration of seeds. In: Kozlowski, T.T. (Ed). Seed Biology 11: 283-313. Alless. New York.

Bass, L.N. (1995). Seed viability during long-term storage. *Horticultural reviews.* 117-141.

Bewley, J.D. and Black, M. (1994). Seed physiology of development and germination (2nd Edn). New York. Plenum Press.

Buchheim, J.A., Colburn, S.M. and Ranch, J.P. (1989). Maturation of soyabeans somatic embryos and the transition to plantlet growth. *Plant Physiology*, 89: 768-775.

Chikuma, G. V., Chileshe, W.M.A. and Pungwa, L. (1985). Resource Atlas for Zambia. National Education Company of Zambia. 12-95.

Clifford, A., Adams, A.and Rinnie, W. (1981). Seed maturation in soyabeans is Independent of seed mass and of the parent plant, yet is necessary for production of viable seed. *Journal of Experimental Botany*, 32(128): 615-620.

Copeland, L.O.and McDonald, M.B. (1985). Principles of Seed Science and Technology. 2nd ed. Macmillan Publishing Company, New York. 152-153.

Dasgupta, J., Bewley, J. D. and Yeung, E. C. (1982). Dessication- tolerant stages during the development and germination of *Phaseolus vulgaris* seeds. *Journal of experimental Botany*, 33: 59-68.

Delouche, J.C. (1974). Maintaining soybean seed quality. Bull Y - 69. National Fertility Development Centre, TVA. Muscle Shoals, Ala. 46-62.

Delouche, J.C. and Baskin, C.C. (1973). Accelerated ageing techniques for predicting the relative storability of seed lots. *Seed Science and Technology*, 1:427-452.

Egli, D. B. and Tekrony, D. M. (1993). Germination and water relations of immature soyabean seed. *Seed Science and Technology*, 21: 139-148.

Egli, D. B., White, G. M. and Tekrony, D. M. (1978). Relationship between seed vigour and the storability of soybean seed. *Journal of Seed Technology*, 3 (2):1-11.

Ellis, R. H. and Filho Pieta, C. (1991). The development of seed quality in spring and winter cultivars of barley and wheat. *Seed Science Research*, 2: 9-15.

Ellis, R. H., Hong, T. D. and Jackson, M. T. (1993). Seed production environment, time of harvest and the potential longevity of seeds of three cultivars of rice (*Oryza sativa* L.). *Annals of Botany*, 72 :583-590.

Gbikpi, P. and Crookston, R. K. (1981). A whole-plant indicator of soyabean physiological maturity. *Crop Science*, 21: 469-472.

Ellis, R. H., Hong, T. D. and Roberts, E. H. (1987). The development of desiccation-tolerance and maximum seed quality during seed maturation in six grain legumes. *Annals of Botany*, 59: 23-29.

Filho, C. P. and Ellis, R. H. (1991). The development of seed quality in spring barley in four environments. Germination and longevity. *Seed Science Research*, 1: 163-177.

Floris, C. (1970). Ageing in *Triticum durum* seeds: behaviour of embryos and endosperms from aged seeds as revealed by the embryo transplantation techinique. *Journal of Experimental Botany,* 21:462-468.

Fraser, J., Egli, D.B. and Leggett, J.E. (1982). Pod and seed development in soyabean cultivars with differences in seed size. *Agronomy Journal*, 7481-85.

Hanson,W.D. (1986). Rates of sucrose release from opened seed coats of soyabean as affected by genotypes: Relation with rate of seed development. *Crop Science,* 26: 301-307.

Hampton, M. (1995). Seed vigour testing Lecture notes, Seed technology Centre, Massey University.

Harrington, J.F. (1972). Seed Storage Longevity. *In*: Kozlowski, T.T. (ed). Seed Biology, Volume III. New York: Academic Press.142-245.

Helmer, J. D., Lienhard, M. and Delouche, J.C. (1962). Some Indices ofvigour and determination in seed of crimson clover. Proceedings of the Association of Official Seed Analysts. 52: 154-161.

Hunter, J. R. and Erickson, A. E. (1952). Relation of seed germination to soil moisture tension. *Agronomy Journal*, 44: 107-109.

International Seed Testing Association (1985a). International rules for seed testing. Rules 1985. *Seed Science and technology* 13: 299-355.

International Seed Testing Association (1985b). International rules for seed testing. Annexes 1985. *Seed Science and technology* 13: 356-513.

Judd, R.H. (1980). Effect of freezing temperatures during soyabean seed maturation on seed quality. Msc. Thesis. University of Kentucky.

Justice, O. L. and Bass, L. N. (1978). Principles and practices of seed storage. Agricultural Hand Book, 506: 289.

Kato, K. Sakaguchi, S. and Naito, Y. (1954). Development of flower parts and seed in soyabean plants. *Takai-Kinki Natl Agricultural Experimental Station Division of Plant Breeding,* 1:96-114.

Klein, S. (1970). Submicroscopical structural changes in mitch and other cellular structures during seed development, dessication, rehydration and very early germination. US Deparment of Agriculture. Grant: A-10-CR-28, Final report.

Lungwangwa, R. A. (1993). A Study of vigour test methodology variables and the relationship between the germination test, vigour tests and field emergence in Birdsfoot Tresfoil (*Lotus corniculatus* L.) and Maku Lotus (*Lotus uligosus* Sckr). Dip. Thesis.

MAFF (1996). Agricultural Statistics Bulletin. Policy and Planning Division. Zambia. 15,16.

MAFF (1995). Zambia Seed Technology Handbook, MAFF. Zambia.195-199.

Marcos-Filho, J., Chamma, H. M. C. P., Casagrande, J. R. R., Marcos, E. A and Regitano-d'arce, A. B. (1994). Effect of harvesting time on seed physiological quality, chemical composition and storability of soybean. *Sciantia Agric. Piracicaba,* 5(2): 298-304.

Mayer, A.M. (1973). The control of the initial stages of germination: Some biochemical investigations. *Seed Science and Technology,* 1: 51-72.

Miles, D.F., Tekrony, D.M. and Egli, D.B. (1988). Changes in viability, germination, and respiration of freshly harvested soybean seed during development. *Crop Science,* 28: 700-704.

Moore, R. P. (1971). Mechanism of water damage to mature soyabean seed. Proceedings of the AOSA, 61: 112-118.

Mwale, S. S. (1993). Factors affecting quality of Seed in storage. MAFF Seed Training Programme. Module IV: Seed Marketing and Storage, June - July 1993. Zambia. 38 - 43.

Perez, M.A. and Arguello, J.A. (1995). Deterioration in peanut seeds under natural and accelerated ageing. *Seed Science and Technology,* 23: 439-445.

Powell, A.A. (1988) Seed vigour and field establishment. *Advances in Research and Technology of seeds* 2: 29-61.

Powell, A.A. (1986). Cell membranes and seed leachate conductivity in relation to the quality of seed for sowing. *Journal of Seed Technology*, 10 (2): 233-238.

Rao, K. N and M. T. Jackson (1996). Seed production environment and storage longevity of japonica rices (*Oryza sativa* L.) Seed Science Research, pp. 17-21.

Roberts, E.H., (1973). Predicting the viability of seeds. *Seed Science and Technology* 1: 515-527.

Rosenberg, L.A. and Rinnie, R.W. (1986). Moisture loss as a prerequisite for seedling growth in soyabeans seed. *Journal of experimental Botany*. 37:1663-1674.

SCCI. (1993). Descriptive list of Cultivars - SOYABEAN. Variety release Committee, Chilanga. 3-16.

Scott, O.W. and Aldrich, R.S. (1983). Modern soyabean production. 2nd Ed. S and A Publications, Inc. USA. 91 - 92.

Simon, E.W. (1984). Early events in germination. In: Murray, D.R. (ed) Seed Physiology Germination and reserve mobilisation, 77-115.

Songolo, H.L. (1993). Practical Manual for plant nutrition. School of Agricultural Sciences, University of Zambia. 9-54.

Teaser, M.B (1984). Physiological basis of crop growth and development. *American Society of Agronomy*. USA. 13-52.

TeKrony, D.M. and Egli, D.B. (1977). Relationship between laboratory indices of soya bean seed vigor and field emergence. *Crop Science* 17:573-577.

TeKrony, D.M., Egli, D.B. and Balles. J. (1979). The effects of field production environment on soyabean seed quality. In P:D Hebblethwaite, (ed). Seed Production, Proc. of the 28th Easter school of Agricultural Science. U. of Nottingham.

TeKrony, D.M., Egli, D.B., Balles. J., Pfeiffer, T. and Fellows, J.R. (1979). Physiological maturity of soya beans. *Agronomy Journal*, Madison, 71:771-775.

TeKrony, D.M., Egli, D.B. and Phillips, A.D. (1980). Effects of field weathering on the viability and vigour of soyabean seed. *Agronomy Journal*, 72:749-753.

Thompson, J.R. (1979). An Introduction to seed technology. Thompson Litho Ltd. Scotland. 28-43.

Walters, C. and Engels, J. (1998). The effects of storing seeds under extremely dry conditions. *Seed Science Research*, .8 (1): 3 –8.

Wahab, A. H. and Burris, J. S. (1971). Physiological and chemical differences in low and high quality soyabean seed. Proc. Assoc. Off. Seed Analysts . 61: 58-67.

Weiss, E.A. (1983). Oilseed Crop. Tropical Agricultural Series, Longman. London.

Wellving, A.H.A. (1984). Seed Production Handbook of Zambia, MAFF, Zambia. 234-238.

APPENDICES

APPENDIX 7.A

APPENDIX A1

Procedure for Determination of Moisture content (ISTA Rules for Seed Testing, 1993)

For the determination of moisture content of the seeds, the Low Constant Temperature Oven Method was followed. The working sample of all the harvested soyabean seeds was 7 grams. This was an adjustment from the recommended 4 to 5 grams for seeds less than 8cm diameter.

1. Pre-drying

   Pre-drying is obligatory for soyabeans seeds. Soyabean seeds were weighed to 7g, these were then pre-dried in a constant temperature oven at 130° C for 20 minutes. The partly dried seeds were left exposed for 2hours in the plant reception room.

2 Moisture Determination

   The soyabean seeds were evenly distributed over the petri-dishes. These were then placed in an oven maintained at a temperature of 104° C for 24 hours. After this period, the seeds were weighed.

2. Calculation of Results

   For each stage of the procedure moisture was calculated as follows:

   (M2 – M3) * 100 / M2– M1

Where

M1- is the weight in grams of the container and it's cover,

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

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

After this calculation for each stage (Pre-drying and final drying), the following formula was employed to find the water content:

$$S1 + S2 - S1 * S2 / 100$$

Where

S1- is the moisture lost in the first stage, and

S2- is the moisture lost in the second stage.

APPENDIX 7.A2

Procedure for Standard Germination Test (ISTA Rules for Seed Testing, 1993)

1. Materials for the test

   i. filter paper

   ii. Glass petri-dishes (9cm)

   iii. Wash bottles

   iv. Distilled Water

2. Procedure

   One hundred seeds in replicates of 25 were taken at random from the harvested sample. These were spaced evenly on the moistened filter papers placed in the petri-dishes. The seeds were harvested within an environment with the average temperature of about 22^{0} C. The test was conducted for 8 days, this when the germination count was done.

   The seeds that had developed protruding radicles were counted as having had germinated. No count of abnormal and dead seeds was taken.

   During the test period distilled water was added to the substrate if dry.

3. Results

   The results were recorded from each replicate and percentage germination was made up of the sum of counts from each replicate.

APPENDIX 7.A3

Procedure for Seed Vigor Test (Tekrony *et al*, 1996)

Sample

Two replicates of 25 seeds each were used.

1. Materials

150ml flasks

EC meter and probe

Distilled water

2. Procedure

The seeds were put in 100ml of distilled water, the bottles were partially covered to prevent foreign particles dropping into the bottle. The seeds were then left in the distilled water for 24 hours. After this period, the electrical conductivity of the solution was measured using a probe and the results recorded in millisiemiens per centimeter.

APPENDIX 7B

ANALYSIS OF VARIANCE TABLES

APPENDIX B.1

Analysis of Variance Table for **Initial Moisture Content** of Soyabeans seeds harvested at different times.

Source of variation	Degrees of freedom	Sums of squares	Mean Square	f- value	Probability
Replication	3	24.55	8.184	1.15	
Variety	1	1.20	1.203	0.17	0.454
Error	3	21.29	7.095		
Harvest Time	5	11513.06	2302.612	276.24	0.00*
Var * H-time	5	2.67	0.534	0.06	0.07
Error	30	250.07	8.336		

CV = 6.40%

APPENDIX B.2

Analysis of Variance Table for **Dry Matter Content** of Soyabeans seeds harvested at different times.

Source of Variation	Degrees of freedom	Sums of squares	Mean square	F- value	Probability
Replication	3	0.14	0.047	0.27	
Variety	1	0.34	0.342	1.98	0.0254*
Error	3	0.52	0.173		
Harvest Time	5	55.60	11.121	213.46	0.000*
Var * H-time	5	0.30	0.061	1.16	0.34
Error	30	1.56	0.052		

CV = 9.5%

APPENDIX B.3

Analysis of Variance Table for **Germination of fresh** Soyabean seeds harvested different. Data transformed to degrees using angular transformation.

Source of Variation	Degrees of Freedom	Sums of	Mean Square	F- value	Probability
Replication	3	128.79	42.931	0.81	
Variety	1	2017.61	2017.613	37.86	0.008*
Error	3	159.89	53.296		
Harvest Time	5	13123.24	2624.6	55.20	0.000*
Var * H-time	5	3006.62	601.323	12.65	0.000*
Error	30	1426.36			

CV = 11.13%

APPENDIX B.4

Analysis of Variance Table for **Germination of Dry** soyabean seeds harvested at different times. Data transformed to degrees using angular transformation.

Source of Variation	Degrees of freedom	Sums of Squares	Mean square	F- value	Probability
Replication	3	82.14	27.381	0.38	
Variety	1	244.80	244.803	3.39	0.162
Error	3	216.64	72.213		
Harvest Time	5	55515.520	11103.104	111.16	0.000*
Var * H-time	5	923.81	184.761	1.85	0.133
Error	30	2996.48	99.883		

CV = 19.96%

APPENDIX B.5

Analysis of Variance Table for **Initial Seed Vigour** Measured as **Electrical Conductivity** of Leachate in distilled water

Source of Variation	Degrees of freedom	Sums of squares	Mean squares	F- value	Probability
Replication	3	0.02	0.006	7.18	
Variety	1	0.00	0.000	0.45	0.069
Error	3	0.00	0.001		
Harvest Time	5	0.17	0.033	24.22	0.000*
Var * H-time	5	0.00	0.000	0.14	0.99
Error	30	0.04	0.001		

CV = 20.28%

APPENDIX B.6

Analysis of Variance Table for **Vigour** of Artificially Aged seeds measured as **Electrical Conductivity** for seeds harvested at different times.

Source of Variation	Degrees of	Sums of squares	Mean square	F- Value	Probability
Replication	3	0.01	0.004	5.72	
Variety	1	0.04	0.0036	49.57	0.05*
Error	3	0.00	0.001		
Harvest Time	5	3.75	0.751	1259.79	0.00*
Var * H-time	5	0.03	0.007	11.06	0.00*
Error	30	0.02	0.001		
Days of Aging	6	7.19	1.198	2515.85	0.00*
Var * Days of Aging	6	0.02	0.003	5.66	0.00*
H-time* D. of Aging	30	0.82	0.027	57.50	0.00*
Var*H-time*D.o.Ag	30	0.13	0.004	9.13	0.00*
Error	216	0.10	0.000		

CV = 3.78%

APPENDIX B.7

Analysis of Variance Table for **Germination** of Artificially aged soyabean seed harvested at different times.

Source of Variation	Degrees of	Sums of squares	Mean square	F- Value	Probability
Replication	3	34.52	11.508	2.63	
Variety	1	33.44	33.44	7.63	0.70
Error	3	13.15	4.382		
Harvest Time	5	216083.18	43216.637	4540.67	0.00*
Var * H-time	5	43.27	8.655	0.91	
Error	30	285.53	9.518		
Days of Aging	6	62381.09	10396.848	1331.46	0.00*
Var * Days of Aging	6	17.03	2.839	0.36	
H-time* D. of Aging	30	33141.04	1104.701	141.47	0.00*
Var*H-time*D.o.Ag	30	197.53	6.584	0.84	
Error	216	1686.65	7.809		

CV = 7.99%

APPENDIX B.8

. Analysis of variance for **time to 50% flowering**

Source of Variation	Degrees of	Sums of squares	Mean square	F- Value
Harv.	5	0.435	0.087	0.3425
Replication	3	1.726	0.575	-
Error (Harv)	15	3.81	0.254	-
Variety	1	15.19	15.19	106.33
Harv. * Variety	5	0.44	0.088	0.616
Error	47	6.714	0.142	-

APPENDIX B. 9

Analysis of variance for **time to 50% podding.**

Source of Variation	Degrees of	Sums of squares	Mean square	F- Value
Harvest time	5	0.685	0.137	0.0731
Replications	3	0.2267	0.0755	-
Error	15	2.81	0.187	-
Variety	1	1.685	1.685	93.92
Harv. * Variety	5	3.06	0.612	34.11
Error	47	0.843	0.0179	-

APPENDIX 7.C

Transformation Table for Germination Data (Angular transformation of percentages to degrees.

%	**0**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**
0	0	5.7	8.1	10.0	11.5	12.9	14.2	15.3	16.4	17.5
10	18.4	19.4	20.3	21.1	22.0	22.8	23.6	24.4	25.1	25.8
20	26.6	27.3	28.0	28.7	29.3	30.0	30.7	31.3	31.9	32.6
30	33.2	33.8	34.4	35.1	35.7	36.3	36.9	37.5	38.1	38.6
40	39.2	39.8	40.4	41.0	41.6	42.1	42.7	43.3	43.9	44.4
50	45.0	45.6	46.1	46.7	47.3	47.9	48.4	49.0	49.6	50.2
60	50.8	51.4	51.9	52.5	53.1	53.7	54.3	54.9	55.6	56.2
70	56.8	57.4	58.1	58.7	59.3	60.0	60.7	61.3	62.0	62.7
80	63.4	64.2	64.9	65.6	66.4	67.2	68.0	68.9	69.7	70.6
90	71.6	72.5	73.6	74.7	75.8	77.1	78.5	80.0	81.9	84.3
100	90.0	-	-	-	-	-	-	-	-	-

Adopted from Little and Hills, Agricultural Experimentation (Abridged from Fisher and Yates: Statistical Tables for Biological Agricultural, and Medical Research, published by Longman Group, Ltd, London (previously published by Oliver and Boyd, Edingburgh)).

APPENDIX 7.D

Agrometeorological Data for the 1998/99 Growing Season. Obtained from The University of Zambia, Department of Geography.

MONTH	DEKAD (10 Days)	RAINFALL DATA (mm		TEMERATURE DATA °C		HUMIDITY %
		Mean	Total	Maximum	Minimum	
December	1	0	0	-	-	-
	2	0	0	29.12	19.4	21.2
	3	10.23	102.3	26.9	19.85	18.84
January	1	0.84	8.4	26.0	18.05	16.0
	2	17.52	175.2	25.4	17.9	19.75
	3	2.39	23.9	30.2	16.8	21.43
February	1	13.75	137.5	27.3	13.7	22.44
	2	1.69	16.9	29.0	17.1	12.31
	3	5.5	55	27.6	17.2	13.24
March	1	1.69	16.9	27.83	17.0	13.82
	2	0.31	3.1	27.83	16.76	17.2
	3	0	0	25.9	11.78	27.8
April	1	0	0	29.0	8.57	31.0
	2	0	0	23.06	12.1	27.6
	3	0	0	27.4	16.7	27.3
May	1	0	0	24.68	13.21	27.06
	2	0	0	26.44	11.68	41.8
	3					

APPENDIX 7.E FIELD PLAN

FIELD LAYOUT

REP.1

4m

H-147

H4	H6	H2	H5	H1	H3

1m

KALEYA

H6	H3	H4	H5	H2	H1

1m 20m

REP.2

KALEYA

H1	H4	H3	H2	H6	H5

H-147

H5	H4	H1	H6	H3	H2

REP.3

H-147

H2	H3	H6	H1	H4	H5

KALEYA

H5	H6	H2	H3	H4	H1

REP.4

KALEYA

H1	H2	H5	H4	H3	H6

H-147

H3	H1	H5	H2	H6	H4

N.B : Diagram not to scale.

KEY

H1 = First Harvest date ; 7 days after podding

H2 = Second Harvest ; 14 days

H3 = Third Harvest ; 21 days

H4 = Fourth Harvest ; 28 days

H5 = Fifth Harvest ; 35 days

H6 =sixth Harvest; 42 days