

GENOTYPE BY ENVIRONMENT INTERACTIONS IN SOYBEAN FOR AGRONOMIC TRAITS AND
NODULE FORMATION

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DECLARATION

I declare that the dissertation hereby submitted to the University of Limpopo for the degree of Master of Agricultural Management (Crop Science), has not previously been submitted by me for a degree at this or any other University; that it is my work in design and in execution, and that all material contained has been duly acknowledged.

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ABSTRACT

The nature and magnitude of the genotype by environment interactions is important to identify superior and stable genotypes under the target environments. This will assist to maximize specific adaptation and to speed up the transfer of new cultivars to growers. The objective of this study was to determine the stability of selected soybean genotypes with regards to the agronomic traits, high yield and nodule formation. Field experiments were conducted under dryland conditions during the 2007/2008 and 2008/2009 growing seasons at the University of Limpopo's experimental farm (Syferkuil) and at a farmer's field at Gabaza community, Mopani District near Tzaneen. Ten selected soybean cultivars were evaluated under a randomised complete block design with three replications. Stability was assessed via joint regression and superiority analyses. Significant differences were found for genotypes, environments and genotype by environment interactions. Stability analysis after Eberhart and Russell's model suggested that the genotypes showed marked differences to environmental changes. The cultivar superiority measure for seed yield indicated that variety Clark was the most stable genotype with an average yield of 5235 kg/ha, followed by L81-4858 and Barc-2 that provided average yield of 4839 kg/ha and 4582 kg/ha, respectively. In terms of number of nodules Magoye was observed to be stable with average of five nodules per plant. Cultivar Barc-2 was found stable for number of active nodules with an average of 3.17 active nodules per plant. Most of the genotypes performed better at Syferkuil than at Gabaza. In general Barc-2 was found stable for yield and other agronomic traits considered in this study. This variety could be suitable for large scale production in these or other similar environments in Limpopo Province.

CHAPTER 1

GENERAL INTRODUCTION

Soybean (*Glycine max* (L.) Merrill) is a leguminous crop with diverse nutritional and agricultural values such as a good source of high quality plant protein and vegetable oils and nitrogen fixing ability, respectively. Cereal crop production should be augmented in a more nutritive rich food like soybean. Soybean can play a vital role for substantial food production in South Africa. The demand for soybean food products is growing both in the national and international markets (Boerma and Mian, 1998). Soybean is viewed as a very attractive crop for the production of biodiesel. Selection of soybean genotype for soy-food offers potential for expanding the market. The increasing market for soy-foods and health benefits associated with them indicate the economic potential and emphasize the need for the identification and development of high yielding soybean cultivars suitable for food processing and human consumption (Kuhn, 1996). Because of the high probability of low yield and crop failures in unfavourable environments, the use of inputs such as fertilizer, pesticides and weed control is seen by farmers as risky. Therefore, the adoption of “improved agronomic practices” has been very limited, and the only economic solution to increase crop yield in unfavourable environments is through breeding (Blum, 1993).

Plant growth and development is the product of the interaction between the genotype (genetic potential) and the environment in which the plant grows (Acquaah, 2007). Many environmental changes are innocuous with respect to crop yield, production, and quality, but others are either beneficial or harmful to crops. Observational evidence indicates that recent regional changes in climate, particularly temperature increases, have already affected a diverse set of physical and biological systems in many parts of the world (Beggs, 2004).

Plants are exposed to different environmental factors such as soil type, soil fertility level, moisture level and temperature, and cultural practices. The soybean cultivars planted nowadays are results of intense genetic improvements that aimed mainly at higher grain yields. Such high yielding cultivars are specialised plants that require specific environmental conditions to express their yield potential. Changes in the relative performance of genotypes across different environments are referred to as genotype x environment interactions (Vijendra Das, 2005).

According to Simmonds (1979) the magnitude of genotype x environment interactions may be judged from an analysis of variance or from an analogous regression analysis of genotypes on a series of environments. Farmers and scientists want successful new soybean varieties that show high performance for yield and other essential agronomic traits. Their superiority should be reliable over a wide range of environmental conditions but also over years. The basic cause of differences between genotypes in their yield stability is the occurrence of genotype by environment interactions. Genotype by environment interactions reduce association between phenotypic and genotypic values, and may cause selection from one environment to perform poorly in another, forcing plant breeders to examine genotypic adaptations (Hayward *et al.*, 1993).

When genotype by environment effects are present on yield or other characters, the breeder's objectives may, in a sense, evade them by defining a narrow range of environments to which adaptation is sought, or negate them by means of the stable, widely adapted genotypes (Simmonds, 1979). Genotype x environment interactions is a problem for plant breeders because it causes uncertainty when translating the performance of a cultivar relative to other cultivars in one environment to perform well in a different environment.

The phenomenon of global climatic change that is occurring over the years is partly responsible for modifying the crop production environment (Acquaah, 2007). Global warming will bring about heat stresses on plants that will even affect the type of crops grown. The challenges that global warming pose to agriculture require that agricultural scientists develop response mechanisms to mitigate the changes in the environment. Hence plant breeding programmers need to make necessary changes to adopt environment specific approaches to crop improvement (Reynolds *et al.*, 2001). South Africa with its very diverse climatic conditions and soil type escalates the problem of genotype by environment interactions even further. To overcome this problem, the universal practise when selecting genotypes, is to test them in yield trials over several environments and years to ensure that the selected genotypes have a high and stable performance over a wide range of environments (Lin and Binns, 1988). In fact, in order to close the gap between actual and potential yields it is also essential for the plant breeder to know all limiting factors (biotic and abiotic stresses), and their frequency and intensity (Ceccarelli, 1989). Some breeders resort to the tedious and expensive approach of screening very large populations. Some also use molecular markers to tag and select certain yield related quantitative trait loci to aid in the selection process (Acquaah, 2007).

Identification of yield contributing traits and knowledge of genotype by environment interactions and yield stability are important for breeding new cultivars with improved adaptation to the environmental constraints prevailing in the target environments. Measuring the genotype by environment interactions, the breeder will better be equipped to determine an optimum breeding strategy for releasing genotypes with adequate adaptation to a target environment (Hayward *et al.*, 1993). Yield trials usually consists of genotypes tested in environments with replications. The environments are normally regarded as location and year combinations. This type of testing helps breeders and agronomists to make estimates of a genotype's yield potential over environments and thus permit recommendations of cultivars for commercial use, while also assisting in selecting genotypes for breeding. When testing a diversity of genotypes in a number of dissimilar environments, it is often observed that the ranking of genotypes differs between environments. This can be described as an interactions between genotypes and environments. Selection of a suitable genotypes over environments may be possible by stratification of environments. Such technique has been used effectively to reduce genotype by environment interactions. Eberhart and Russell (1966) observed that even with this stratification technique, little interaction of genotype with location in a sub-region and with environments differed frequently in different years but still some progress has been achieved in reducing genotype by environment interactions. With the availability of improved statistical tools to analyze and understand genotype by environment interactions, it is now possible to develop improved cultivars for target environments by exploiting genotype by environment interactions and marker based selection integrated to traditional plant breeding.

Breeders look for a variety that has good mean trait performance over a wide array of environments and years and the concept of stability is overlooked. Such approach is reasonable if there are no genotype by environment interactions, but in most cases there are interactions. Some genotypes can have high yield in few environments and very low yield in other environments, showing better mean performance across environments. But few genotypes may have average yield that is stable over wider environments. Knowledge of the pattern and magnitude of genotype by environment interactions and stability analysis is important for understanding the response of different genotypes to varying environments and for identifying superior soybean genotypes under the target environment and agronomic conditions to maximize specific adaptation and to speed up the transfer of new cultivars to growers (Hayward *et al.*, 1993). The objective of this study was to determine genotype by environment interactions in yield, agronomic traits and nodule formation among selected soybean cultivars when tested across two locations in Limpopo Province.

Soybean cultivars that are stable for yield, nodule formation and other important agronomic traits may possibly be recommended for large scale production in the Province.

CHAPTER 2

LITERATURE REVIEW

2.1 History and Origin

Soybean (*Glycine max* (L) Merrill) is one of the world's most valuable crop, used as feed for livestock, as a source of dietary protein and oil by millions of people, and in manufacturing of various products (Nwokolo and Smart, 1996). Currently, there is an increasing interest to use soybean for biofuel and biodiesel production in addition to its traditional use in plant protein production and as animal feed (Acquaah, 2007). The crop is considered among the oldest cultivated crops. The first record of the crop is contained in a Chinese book dated 2838 BC in which Emperor Cheng-Nung described the plant as "Wu Ku", one of the sacred five grains (the others being rice, wheat, barley, and millet). The crop was considered essential for Chinese civilization. In 1903 the first soybean seeds were imported to South Africa on an organized basis (Nwokolo and Smart, 1996). Cultivated soybean is believed to have been derived from a wild progenitor, *Glycine ussuriensis*, which occurs in east Asia. Its first introduction into the USA is traced to a Samuel Bowen, an employee of East India Company, a seaman, who brought it to Savannah, Georgia, from China via England (Acquaah, 2007).

2.2 Classification and adaptation of soybean

Soybean is an important summer crop, as it needs a hot and humid climate to prosper (Norman, 1978). But the crop is currently grown all over the world, in climates ranging from temperate to subtropical (Kinloch, 1998). Soybeans require relatively high soil water content for germination which will occur in five to eight days at 20 °C (Norman *et al.*, 1984). Soybean is an herbaceous annual with determinate as well as indeterminate growth habit, belonging to the family Leguminosae (Carlson, 1973). The crop has an upright growth habit with trifoliate and alternate leaves, except one simple and opposite pair at the first node above the cotyledons (Martin, 1984). Soybean bears small, white or purple flowers on the stems or side branch nodes and these develop into pods containing one to four seeds. From three to as much as 350 pods may develop on a single plant depending on plant density, growth conditions and cultivar. The need for a long growing season and satisfactory soil moisture during flowering and pod filling are very important. Plant height can generally vary from 20-200 cm and the growth period from 70-170 days (Carlson, 1973). The

soybean plant usually needs five to six months to mature. Matching soybean maturity and genetics is important in stabilizing yields and economic returns. When the seeds start to mature, the leaves of the plant start senescence. This is the stage when the harvesting of this crop is done. It is highly dependent on rain and a change in the rainfall pattern affects the production of soybean (Nwokolo and Smartt, 1996). Soybean can be considered in rotations when irrigation water is available (Smit, 2000).

2.3 Soybean production in South Africa

The first soybean trials in South Africa were conducted in 1903 at the experiment at farm at Cedara, Natal (Biowatch, 2004). Soybean production in South Africa expanded greatly during the 1920s, but only in the past few years has it become a major cash crop. Mpumalanga is the main growing area for soybean, followed by KwaZulu-Natal and Free State. Farmers plant the crop in November and harvest it in April (Biowatch, 2004). Shurtleff and Akiko (2007) reported a crop of a remarkable 907194 metric tonnes in 1921-1922, increasing to 2177266 tonnes in 1925-1926, with a yield of 842 kg/ha in South Africa. The yield for 2004 was 180 000 tons in South Africa. South Africa is the largest producer of soybean in the African continent (Van Wyk and Smit, 1995) with approximately 163285 metric tons being produced during 2003 (Anon; 2004). Most soybean is used for animal feed and this is growing due to a shortage in fishmeal (Nwokolo and Smartt, 1996).

The soybean market is split into oil, whole soybean and soya protein meal. The oil has industrial and edible uses, while whole soybeans are used for human consumption and animal feed. Both the White Paper on Energy Policy as well as the Integrated Rural Development Strategy with regard to the supply and consumption of energy in South Africa, highlight the greater need for the development and implementation of renewable energy applications and their application to rural development to develop internal capacity for integrated and sustainable development (Biowatch, 2004), one of the identified sources of alternative energy is the development of biofuels. There is a great deal of interest in biodiesel worldwide because it is made from renewable resources and reduces air-polluting emissions by diesel engines. Typically, many commercially available biodiesel fuels are made from soy oil. Several technologies are being applied to the production of biodiesel from lower value less pure lipids such as soapstock and not only from highly refined oils as has been done thus far. According to USDA ARS (2002), soybean oil (soapstock) is a plentiful and relatively inexpensive resource and is seen as the ideal candidate for the application of this technology. According to Thomas (2003), the total amount of soybean production in Limpopo Province is small, but

registered a significant increase from 4607 to 11035 tons between 1980/1 and 1988/9. This was, however, accounted for the expansion in the area planted, particularly in the Waterberg district from 2116 ha to 5788 ha and yield increases resulting from the use of new high yielding cultivars. There is no report of production of the crop in the rest of the Limpopo Province.

2.4 Uses and economic importance of soybean

Soybean became a valuable source of human food and animal feed. The crop is a major food crop worldwide in terms of gross production value, averaging 198,233,748 metric tons during 2003 (Hancock, 2004). The USA is the world's leading producer of soybean, accounting for nearly 50% of the world's total production followed by Argentina, Brazil and China (Acquaah, 2007). Soybean consists of about 35-40% protein, oil content of 20%, carbohydrate of 30% and 10% fiber. Soybean has the highest protein content of all food crops, and second only to groundnut in terms of oil content among food legumes (Mpepereki, 2001). Compared to other protein rich foods such as meat, fish and eggs, soybean is by far the cheapest. Besides being an important source of vegetable oil and protein meal, the immature bean and pod of soybean can be eaten as a green vegetable, and the dried bean is consumed whole, split, sprouted, or in various processed forms. The crop remains the most valuable legume crop, with numerous nutritional and industrial uses due to its unique chemical composition (Smit, 2000).

Another advantage of soybean is that it improves soil fertility by adding nitrogen from the atmosphere (Mpepereki *et al.*, 2000). Except for its importance as food crop, the soybean crop has also a range of commercial applications, i.e. pharmaceutical applications (lecithin and vitamin E) and industrial applications (printing ink, polyethelene, etc.) (Carlson, 1973). Soyfoods have been reported to provide protection against heart disease, cancer, menopausal symptoms, and other diseases (Carter and Wilson, 1998; Messina and Messina, 1991). The crop will increase in importance as the world becomes more aware of the need for renewable resources.

A major constraint to agricultural productivity in most developing countries, in particular southern African countries, is N-deficient soils. Biological nitrogen fixation by soybean contributes significant quantities of nitrogen to both natural and managed ecosystems and offers a relatively cheap alternative source of nitrogen to resource poor farmers. Musiyiwa (2001) indicated that the multiple benefits of soybean include

soil fertility improvement, humans and livestock protein nutrition and cash income from grain and processed products. Small holder farmers only need access to the seeds to be able to grow soybean, which brings multiple benefits. Other potential benefits of soybean include low susceptibility to pests and diseases and better grain storage quality (Mpepereki *et al.*, 1996).

Soybean is viewed as a very attractive crop for the production of biodiesel. Patzek and Pimentel (2005) conducted a net energy analysis for deriving biodiesel from soybeans. According to Patzek and Pimentel (2005) soybean yield is as high as 2.668 tones/hectare translating into 480 kg of oil per hectare, as compared to sunflower seeds which will produce only 390 kg of oil. Soybeans can fix atmospheric nitrogen and require minimal nitrogen fertilizer inputs, which often can account for the single largest energy input in agriculture.

The high energy cost, however, lies in processing with about 11.9 million kcal required to produce 1000 kg of soy oil which will have an energy content of 9 million kcal. This translates to a net energy loss of 32%. The contribution of the soy meal produced as a useful by-product, when added to the energy calculation, reduces this net energy loss to 8%. Still, at 2002 figures soy oil was found to be 2.8 times more expensive than petroleum diesel to produce (Nwokolo and Smartt, 1996). The use of a genetically modified crop as in soybean for the production of biodiesel would only add to the cost because of the economics associated with the growth of transgenic plants. Patent protection of genetically modified crops ensures that there is a fixed cost associated with their planting which would have to be taken into consideration in the energy calculation (Patzek and Pimentel, 2005).

2.5 Growth habits in soybean

Soybean is also classified by its growth and floral initiation. Determinate and indeterminate growth habits exist for different soybean cultivars. A distinction is drawn between cultivars with a determinate habit, where vegetative development stops during flowering and indeterminate growth habit soybean types begin to flower and form pods early in the growing season while continuing to grow vegetatively, that is adding new nodes that grow when floral buds are initiated (Smith, 1995). The vegetative period, usually six to eight weeks long, lasts from when the plant emerges until it begins to flower. Soybean plants are photoperiodically sensitive, and flowering depends on daylength and temperature. Both flowering and pod

formation are more uniform in cultivars with a determinate habit, although variation does occur among individual plants.

According to Mpeperekwi *et al.* (2000), a major advantage to smallholder farmers of the naturally nodulating soybean varieties is their indeterminate growth habit and relatively low grain and nitrogen harvest index. These varieties produce a significantly greater biomass which can provide both fodder for livestock and an organic amendment for soil fertility improvement when the biomass is ploughed under. Determinate types add much less additional biomass after flowering begins (Smith, 1995).

2.6 Nodule formation and nitrogen fixation in soybean

A distinguishing feature of soybean from cereals is its ability to utilize atmospheric nitrogen and make it available to the host plant. The *Rhizobia spp.* of bacteria which are normally free living in the soil, enter the legume roots from the seedling stage onwards (Duffus and Slaughter, 1980). Nodulation in soybean can be defined as a soil bacteria's (*Rhizobium*) ability to penetrate and fix nitrogen with the soybean roots. All legumes have the ability to do this. However the strain of bacteria is quite different for each crop. For nitrogen fixation to occur, nitrogen-fixing bacteria in the species of *Rhizobium* need to be present in the soil. If soils do not already contain a high population of *Rhizobium*, these bacteria can be added either as liquid or granular peat inoculants or as a peat-based powder. No-till and earlier planting situations, soil compaction and cool soil temperatures put stress on the legume seedling, which in turn can reduce nodulation (Vance, 2001). *Rhizobia* are usually defined as nitrogen-fixing soil bacteria capable of inducing the formation of root nodules on leguminous plants in which atmospheric nitrogen is reduced to ammonia for the benefit of the plant. There are a number of strains of *Rhizobium* each with a broad range of specificity (Duffus and Slaughter, 1980).

2.7 Environmental factors in soybean production

Environmental factors are particularly important in the cultivation of legumes. Water deficits, extreme temperature, soil acidity and light intensity all have profound effects on the growth, development and yield of soybean (Ramdance *et al.*, 2002). The environment represents conditions under which plants grow including location, years, and management practices. Any stress (biotic or abiotic) in the environment will

adversely impact growth and development (Kang, 2005). There are macro-environments and micro-environments for growth. Micro-environment includes soil and meteorological factors, and biotic factors in a limited space, intimately associated with the organism. Macro-environment refers to the abiotic and biotic factors on a large scale at a particular period of time (Acquaah, 2007). Environment variation itself may impede accurate genotyping by causing the phenotype of different genotypes to overlap. This is particularly a problem with polygenic traits. Some phenotype such as resistance to particular abiotic factors like drought or salinity, may only appear under particular conditions which are difficult to define or control (Callow *et al.*, 1997). The nature and effect of the environment has implications in the design and conduct of field trials.

2.8 Components of the environment in soybean production

2.8.1 Soil

Soybeans grow best under good soil conditions. Soybeans may be planted on any soil suitable for maize production, but do not do as well as maize on sandy soils with a low organic matter content. Generally they are adapted to heavier soils than most other crops. Legumes are normally grown on soils with a higher pH than used for maize production. They are sensitive to waterlogging, drought stress, eelworm, crusting, pH and atrazine or related herbicides. Soybean, however, are better adapted to soils with a lower pH than lucerne, for instance. Soybean planted in soils with a pH greater than 7.8 with high salinity or in high lime soils may have leaf yellowing due to iron chlorosis or other nutrient problems. Some varieties of soybean are more tolerant than others to high lime soils. The hypocotyl of the soybean seedling breaks easily during emergence if under pressure. Soils that compact easily and form a crust should, therefore, be avoided, or alternatively these conditions must be prevented (Anon; 2006).

2.8.2 Carbon dioxide

The Oxygen is required for respiration and carbon dioxide for photosynthesis. Higher concentrations of carbon dioxide in the rooting zone of crops are harmful to all crops; it is only the green, aerial parts that can benefit (Youdeowei *et al.*, 1986).

2.8.3 Sunlight

The sun is the primary source of energy in the earth's atmosphere. Total solar energy varies from region to region over the earth's surface. The techniques used to obtain maximum utilization of sunlight in crop production include the choice of location, type of plant, distribution and density of plants through spacing, pruning or training that ensures maximum utilization of sunlight. Because of the specific photoperiodic (daylight length) sensitivity of soybeans and the available genetic variation of the characteristic, it is possible to cultivate soybeans in a variety of climatic conditions. One of the best ways of utilizing sunlight in crop production is to adjust the time of planting so that the crop grows through a period when sunlight is brightest and longest in duration (Youdeowei *et al.*, 1986).

2.8.4 Water

Water plays a crucial role in the survival of all organisms. In plants in particular, aside from fulfilling the role of solvent, transport medium and evaporative coolant, water provides the energy necessary to drive photosynthesis, the natural plant process which synthesizes organic food. Photoautotrophs are organisms that possess their own chlorophyll and are thus able to harness the energy associated with sunlight in photosynthesis. Under drought conditions the loss of water in the plant protoplasm may result in the concentration of ions in the protoplasm to toxic levels resulting in possible protein denaturation and membrane fusion and negatively impacting plant metabolism (Nwokolo and Smartt, 1996).

Warm, moist conditions, with a rainfall of 400-800 mm, evenly distributed over the growing season are ideal for soybean production (Nwokolo and Smartt, 1996). However, soybeans also do well in warm, dry areas under irrigation. Water is required for the process of photosynthesis and for all metabolic reactions. The intensity and duration of rainfall, varies considerably from the equator to the higher latitudes. Humidity also affects crop production by influencing evapotranspiration. Most tropical crops are adapted to intermediate moisture supply conditions and their growth and yield are severely affected by excess or reduced moisture availability. Certain stages of reproductive growth, e.g. flowering and pod setting, are very sensitive to moisture stress (Youdeowei *et al.*, 1986).

2.8.5 Temperature

Solar radiation is a major determinant of ambient temperature. Soybean seed requires a minimum soil temperature of 12 °C for germination. The minimum temperature for growth is 10 °C. Optimum temperatures for soybean lie between 5 °C and 34 °C (Youdeowei *et al.*, 1986). Flowering is hampered at temperatures below 13 °C. Different parts of the plants, however, respond differently to the same temperature conditions. Temperature fluctuation is only important for crop growth and yield when moisture supply is limiting (Liebenberg, 2002).

2.9 Selection of soybean variety

Soybean varietal selection should be based on maturity, yield, and disease reaction (Ceccaralli, 1996). The selection and use of high quality seed and matching disease resistance to location are basic keys to optimize soybean yield. Stress development and maturation may reduce seed size, increase seed injury, and contribute to lowered germination. In breeding evaluation programs, selection of cultivars under high input conditions may be favoured compared to those selected in low input conditions (Ceccaralli, 1996). This could be why research on crop improvement has not had as much an impact on the small-scale farms compared to commercial farms. Falconer (1990) supported the idea of breeding for specific adaptation rather than broad adaptation. Ceccarelli (1996) found that breeding programs conducted under high input and uniform conditions may favour selection of cultivars adapted to good management and eliminate individuals adapted to poor conditions. In many cases, one or more factors limit production and prevent the full yield potential from being realized. Adaptation of a cultivar is affected by factors that vary from one location to another and from year to year. The effects of these factors are usually reflected in their yields. Therefore adaptation is an important factor that may increase productivity of a crop. It is better to replicate trials over years than over localities within years for effective comparison of cultivars, because cultivar x year interaction is greater than that of locality and locality x year (Patterson *et al.*, 1983). When breeding varieties that are adapted to different environments, a breeder has a choice of either breeding for similar ecological conditions or more variable conditions that include various environments (Finlay and Wilkinson, 1963). Scientists should aim to produce cultivars that are able to withstand unpredictable environmental variations (Allard and Bradshaw, 1964). In the dry land agriculture of Africa, abiotic and biotic stresses limit potential grain yields (Kenga *et al.*, 2003). The demand for legumes in Africa calls for an increase in

production of bambara groundnut, which is one of the legumes grown in African countries. Poor grain yields may be associated with low yield stability (Fisher and Maurer, 1978; Sinha *et al.*, 1986).

Making selections in the presence of genotype by environment interactions is a major problem facing many scientists. The process to develop genotypes that are stable and high yielding across different environments is an ongoing process all over the world. In every plant breeding program breeders have to plant materials for a number of years in various locations in order to test stability of materials over a range of environments (Yan and Hunt, 1998). Yates and Cochran (1938) stated that agricultural experiments on the same, or group of factors, are usually carried out at a number of places and repeated over years, because the effect of most factors (varieties, fertilizers etc.) varies considerably between places and from year to year, due to differences in soil, agronomic practices, climatic conditions and other variations in the environment. There are cultivars that are less influenced by the productivity level of the environment, and then others whose performance is directly related to the productivity of the environment. According to Joppa *et al.* (1971), the sets of varieties will not rank the same for several given trials. Experimental error and genotype by environment interactions lead to differences expressed by changes in the rankings. To select for the best experimental lines, the yield trials should also be replicated. Therefore results from one year in the same place are of limited use even though they are accurate. According to Eberhart and Russell (1966), to obtain useful information for stability parameters, cultivars must be grown in various localities. Assessing a cultivar's suitability for a given environment is based on its yield stability at the environment, yielding ability, days to maturity, etc. There are a number of measures that are used for studying the stability of genotypes in the presence of genotype by environment interactions.

2.10 Genotype by environment (G x E) interactions

Genotype by environment interactions (G x E) in several crops has been studied widely (Alberts, 2004; Annicchiarico, 2002; Annicchiarico *et al.*, 2005; De La Vega and Chapman, 2006). G x E occurs when two or more genotypes are compared across different environments and their relative performances (responses to the environment) are found to differ. That is, one cultivar may have the highest performance in one environment but perform poorly in others (Vijendra Das, 2005), which results from differential response of the cultivar, to various edaphic, climatic and biotic factors. The effect of these interactions is that the association between phenotype and genotype is reduced. This raises the important issue of adaptation

because a breeder's selection in one environment of superior performers may not hold true in another environment. An understanding of environmental and genotypic causes of G x E is important at all stages of plant breeding, including ideotype design, parent selection, selection based on traits, and selection based on yield (Yan and Hunt, 1998). Understanding of the cause of G x E can be used to establish breeding objectives, identify ideal test conditions, and formulate recommendations for areas of optimal cultivar adaptation (Yan and Hunt, 2001). The basic cause of differences between genotypes in their stability is the wide occurrence of G x E.

Genotype by environment interactions occur in two ways. Firstly the difference between genotypes vary without alteration in their rank i.e. G x E is present because one cultivar yields more than another cultivar in all the environments, and secondly the ranking between cultivars changes across environment, while the other cultivar is more productive in another (Dixon *et al.*, 1994). According to Misra and Panda (1990), inconsistent yield performance of cultivars in different environments may be a contributing factor to productivity due to large G x E. Breeding materials can be selected and assessed on the basis of their different responses to the environments. The G x E poses a serious problem in breeding programs because it can influence any stage of the breeding program, like identifying appropriate sources or parent material. But it can also play a role in the expression of quantitative traits. Studying of G x E is very important to plant breeders because interactions can limit the progress in the selection process and since it is a basic cause of differences between genotypes for yield stability. Understanding the cause of G x E is important to help in selecting varieties with the best adaptation and that can give stable yields. Varieties that show low G x E and have high stable yields are desirable for crop breeders and farmers, because that indicates that the environment has less effect on them and their higher yields are largely due to their genetic composition. It is important to understand crop development in relation to biophysical conditions and changes in season when selecting well adapted genotypes and correct planting date (Linnemann *et al.*, 1995).

Yield stability is defined in many different ways using various stability measures. Blum (1993) defined yield stability as a measure of variation between potential and actual yield of genotypes across different environments. Fehr (1987) stated that yield stability of a cultivar is influenced by the genotype of individual plants and the genetic relationship between plants. It can be measured through analysis of variance procedures and regression analysis. Domitruk *et al.* (2001) indicated that the analysis of variance procedure is a useful tool for estimating the existence and magnitude of G x E. However, the components of variances

do not provide satisfactory explanation of the interaction. There are a number of suggested or proposed methods that can be used for stability measurement. Yates and Cochran (1938) proposed a purely statistical analysis, which was later used by Finlay and Wilkinson (1963) and Eberhart and Russell (1966). They used the analysis to detect and measure the magnitude of G x E in barley and maize respectively.

2.11 Concepts of stability

In the presence of G x E, the use of genotypic means across environments as criteria for selecting superior genotypes is not valid. This leads to the concept of stability of performance (Kang, 1998). Lin *et al.* (1986) classified stability into three types; Type 1: A genotype is regarded to be stable if its among environment variance is small. This is useful when the environments regarded are not very diverse and is equivalent to the static concept of stability (Becker and Léon, 1988). Type 2: A genotype is regarded to be stable if its response to environments is parallel to the mean response of all genotypes in the trial. Any genotype with the slope of the regression coefficient $b = 1$ will be assumed to be stable. Type 3: A genotype is regarded to be stable if the residual mean square (MS) from the regression model on an environmental index is small. The environmental index implicates the mean yield of all the genotypes in each location minus the grand mean of all genotypes in all locations. Type 3 is also part of the dynamic or agronomic stability concept according Becker and Léon (1988). Breeding for broad adaptability requires a different interpretation and approach to the stability analysis procedure than breeding for specific adaptability (Hildebrand and Poey, 1985). Methods of Eberhart and Russell (1966) and Tai (1971) can be used for estimating Type 3 stability.

Becker and Léon (1988) stated that all stability procedures based on quantifying G x E effects belong to the dynamic concept. This includes the procedures for partitioning the G x E of Wricke's (1962) ecovalence and Shukla's (1972) stability of variance procedures using the regression approach such as proposed by Finlay and Wilkinson (1963), Eberhart and Russell (1966) and Perkins and Jinks (1968) as well as non-parametric stability statistics.

Lin *et al.* (1986) defined four group of stability statistics. Group A is based on deviations from average genotype effect (DG), group B on G x E term (G x E), and group C and D on either DG or G x E. Group A and B represent sums of squares, and those of group C and D represent regression coefficient or

regression deviation. They integrated type 1, type 2 and type 3 stabilities with the four groups. Group A was regarded as type 1, groups B and C as type 2 and group D as type 3 stability models.

Lin and Binns (1988) proposed type 4 stability concepts on the basis of predictable and unpredictable non genetic variation; the predictable component related to locations and the unpredictable component related to years. They suggested the use of a regression approach for the predictable portion and the mean square for years x locations for each genotype as a measure of the unpredictable variation.

2.12 Statistical methods for measuring genotype x environment interactions

A combined analysis of variance procedure is the most common method used to identify the existence of genotype x environment interactions from replicated multi-location trials. If the genotype x environment interaction variance is found to be significant, one or more of the various methods for measuring the stability of genotypes can be used to identify the stable genotype(s). The statistics, which are used to identify stable genotypes, are classified into parametric and nonparametric. Parametric statistics are more useful when the data are continuous, while nonparametric statistics are used when the data are discontinuous. Nonparametric data analysis has the potential to reduce complex data to intuitive measures of stability (Nassar and Huhn, 1987).

2.12.1 Analysis of variance

Consider a trial in which the yield of G genotypes is measured in E environments each with R replicates. The classic model for analysing the total yield variation contained in GER observations is the analysis of variance (Fisher, 1918; Fisher, 1925). The within environment residual mean square measures the error in estimating the genotype means due to differences in soil fertility and other factors, such as shading and competition from one plot to another. After removing the replicate effect when combining the data, the GE observations are partitioned into two sources: (a) additive main effect for genotypes and environments and (b) non-additive effects due to genotype by environment interaction. The analysis of variance of the combined data expresses the observed (Y_{ij}) mean yield of the i^{th} genotype at the j^{th} environment as:

$$Y_{ij} = \mu + G_i + E_j + GE_{ij} + e_{ij} \dots \dots \dots (1)$$

Where μ is the general mean; G_i , E_j , and GE_{ij} represent the effect of the genotype, environment, and the genotype by environment interactions, respectively; and e_{ij} is the average of the random errors associated with the r^{th} plot that receives the i^{th} genotype in the j^{th} environment. The non-additivity interaction as defined in (1) implies that the expected value of the i^{th} genotype in the j^{th} environment (Y_{ij}) depends not only on the levels of G and E separately but also on the particular combination of levels of G and E (Crossa, 1990).

The major limitation in this analysis is that the error variances over environments should be homogeneous to test for genotypic differences. If error variances are heterogeneous, this analysis is open to criticism as the F-test of the genotype by environment interaction mean squares against the pooled error variances is biased towards significant results. A correct test for significance, by weighting each genotype mean by the inverse of its estimated variance, has been used by Yates and Cochran (1938) and Cochran and Cox (1957). This weighted analysis gives less weight to environments that have a high residual mean square. The disadvantage of weighted analysis is, however, that weights may be correlated to environment yield responses (high yielding environments showing higher error variance and low yielding sites presenting lower error variances) and this could mask the true performance of some genotypes in certain environments (Crossa, 1990).

One of the main deficiencies of the combined analysis of variance for multilocation trials is that it does not explore any underlying structure within the observed nonadditivity (genotype by environment interaction). The analysis of variance fails to determine the pattern of response of genotypes and environments. The valuable information contained in $(G-1)$ $(E-1)$ degrees of freedom is particularly wasted if no further analysis is done. Since the nonadditive structure of the data matrix has a non-random (pattern) and random (noise) component, the advantage of the additive model is lost if the pattern component of the nonadditive structure is not further partitioned into functions of one variable each (Crossa, 1990).

Analysis of variance of multilocation trials is useful for estimating variance components related to different sources of variation, including genotypes and genotype by environment interactions. In general, variance component methodology is important in multilocation trials, since errors in measuring the yield performance of a genotype arise largely from genotype by environment interactions. Therefore, knowledge of the size of this interaction is required to (a) obtain efficient estimates of the genotypic effects and (b) determine optimum resource allocations, that is the number of plots and locations to be included in future trials. In a

breeding program, variance component methodology is used to measure genetic variability and estimate the heritability and predicted gain of a trait under selection (Crossa, 1990).

2.12.2 Stability analysis: parametric approach

Stability analysis provides a general summary of response patterns of genotypes to environmental changes. A number of statistical procedures have been developed to enhance our understanding of genotype x environment interactions and its relationship to stability (Vijendra Das, 2005), but not all give easily interpretable results. Freeman (1973) termed the main type of stability analysis, joint regression analysis or joint linear regression (JLR). It involves the regression of the genotypic means on an environmental index. Joint regression analysis provides a means of testing whether the genotypes have characteristic linear responses to changes in environments. Joint regression analysis was first proposed by Yates and Cochran (1938) and then widely used and reviewed by various authors (Baker, 1988; Becker and Léon, 1988; Crossa, 1990; Eberhart and Russell, 1966; Finlay and Wilkinson, 1963; Freeman, 1973; Freeman and Perkins, 1971; Hardwick and Wood, 1972; Hill, 1975; Hohls, 1995; Lin *et al.*, 1986; Perkins and Jinks, 1968; Shukla, 1972; Westcott, 1986; Wright, 1971).

2.12.2.1 Regression analysis (b_i) and deviation mean square (s^2d_i)

Joint linear regression is a model used for analysing and interpreting the non-additive genotype x environment interaction of two way classification data. The genotype x environment interaction is partitioned into a component due to the linear regression (b_i) of the i^{th} genotype on environment mean, and a deviation (d_{ij}):

$$(GE)_{ij} = b_i E_j + d_{ij} \quad (2)$$

$$\text{And thus } Y_{ij} = \mu + G_i + E_j + (b_i E_j + d_{ij}) + e_{ij} \quad (3)$$

This model was first proposed by Yates and Cochran (1938) in their evaluation of barley yield trials. The methods divides the $(G - 1) (E - 1)$ df for interaction into $G - 1$ df for heterogeneity among genotype regression and the remainder $(G - 2) (E - 2)$ for deviation. Further details about interaction are obtained by regressing the performance of each genotype on the environmental means. Finlay and Wilkinson (1963)

determined the regression coefficient by regressing variety mean on environmental mean, and plotting the obtained genotype regression coefficients against the genotype mean yield.

In Finlay and Wilkinson's (1963) study, the adaptation of the whole population of varieties was facilitated by the use of a two dimensional plot (Scatter diagram), with mean yield and regression coefficient as coordinates of each variety. Finlay and Wilkinson (1963) defined a genotype with $b_i = 0$ as stable, while Eberhart and Russell (1966) defined a genotype with $b_i = 1$ to be stable. Perkins and Jinks (1968) regression coefficient is similar to Finlay and Wilkinson's (1963) except that the observed values are adjusted for environmental effects before the regression.

Eberhart and Russell (1966) proposed pooling the sum of squares for environments and genotype by environment interaction and subdividing it into a linear effect between environments (with 1 df), a linear effect for genotype x environment (with E, 2 df). In effect the residual mean squares from the regression model across environments is used as an index of stability, and a stable genotype is one in which the deviation from regression mean squares (S^2d_i) is small.

$$S^2d_i = 1/E - 2 [E_j (X_{ij} - X_i - X_j + X_{..})^2 - (b_i - 1)^2 E_j (X_j - X_{..})^2] \dots \dots \dots (4)$$

The regression approach has been shown to be the most useful for geneticists (Freeman and Perkins, 1971; Freeman, 1973; Hill, 1975; Westcott, 1986), but it should be noted that these authors have pointed out several statistical and biological limitations and criticisms.

The first statistical criticism is that the genotype mean (x-variable) is not independent from the marginal means of the environments (y-variable). Regressing one set of variables on another that is not independent violates one of the assumptions of regression analysis (Freeman, 1973; Freeman and Perkins, 1971).

The second statistical limitation is that errors associated with the slopes of the genotypes are not statistically independent, because sum of squares of deviation with (G-1) (E-2) df, cannot be subdivided orthogonally among the G genotypes (Cossa, 1990).

The third statistical problem is that it assumes a linear relationship between interaction and environmental means. When this assumption is violated, the effectiveness of the analysis is reduced, and results may be misleading (Mungomery *et al.*, 1974).

A major biological problem with regression genotype means on environmental means arises when only a few low or very high yielding sites are included in the analysis. The genotype fit may be determined largely by its performance in a few extreme environments, which in turn generates misleading results (Westcott, 1986). Regression analysis should be used with caution when the data set includes results from only a few high or low yielding locations (Crossa, 1990).

Becker and Léon (1988) noted when studying the most appropriate biometrical method, that the regression approach is of little use if the regression coefficient b_i is included in the definition of “stability”. For this reason b_i is generally viewed by authors not as a measure of stability but rather as additional information on the average response of a genotype to advantageous environmental conditions.

2.12.2.2 Cultivar superiority measure (P_i)

Lin and Binns (1988) defined the superiority measure (P_i) of the i^{th} genotype as the mean square of distance between the i^{th} genotype and the genotype with maximum response as:

$$P_i = [n(Y_i - M_{..})^2 + (Y_{ij} - Y_i + M_j + M_{..})^2] / 2n$$

Where, Y_{ij} is the average response of the i^{th} genotype in the j^{th} environment, Y_i is the mean deviation of genotype i , M_j is the genotype with maximum response among all genotype in the j^{th} location, and n is the number of locations. The smaller the value of P_i , the less its distance to the genotype with maximum yield and the better the genotype. A pairwise genotype x environment interaction mean square between the maximum and each genotype is also calculated and it is similar to the method used by Plaisted and Peterson (1959), except that (a) the stability statistical are based on both the average genotypic effects and genotype x environment interaction effects and (b) each genotype is compared only with the one maximum response at each environment (Crossa, 1990).

2.12.3 Nonparametric approach

Nonparametric statistics for genotype x environment interactions based on ranks provided a useful alternative to parametric approaches currently used, which are based on absolute data. Some advantages of nonparametric statistics compared to parametric ones are: reduction or even avoidance of the bias caused by outliers, no assumption are needed about the distribution of the analysis values, homogeneity of variances, and additivity (linearity) of effects are not necessary requirements (Huhn, 1996). Further advantages are that nonparametric stability statistics are expected to be less sensitive to errors of measurement than parametric estimates and the addition or deletion of one or a few observations is not likely to cause great variation in the estimate as would be the case for parametric stability statistics (Nassar and Huhn, 1987).

2.13 Classification of genotype by environment interactions

Every factor that is a part of the plant has the potential to cause differential performance that is associated with G x E (Allard and Bradshaw, 1964). The type of genotype x environment interaction influences the nature of the cultivar the breeder eventually releases for the production region. The environment, can be complex, and so can the genotype of the plant. Consequently, the biological basis of G x E is complex by nature. The interaction between the genotype (cultivar) and the environment is ongoing. As the number of environments (n) and number of genotypes (m) increase, the possible G x E is given by $mn!/m!n!$ of this, there is theoretically only one genotype that is the best performer under all environments, odds that make a search for it futile (Acquaah, 2007). Statistically a G x E will arise when the difference in performance between the genotypes lacks consistency over the environments (Hill *et al.*, 1998):

$$A_1 - B_1 \neq A_2 - B_2 \text{ or } A_1 - B_1 - (A_2 - B_2) \neq 0$$

A significant interaction will arise when:

$$A_1 - B_1 - A_2 + B_2 \neq 0$$

Where A_1 , A_2 , B_1 and B_2 represent four genotypes.

The relative performance of a genotype across environment determines the importance of an interaction. There is no G x E when the relative performance among genotypes remain constant across environments (Hill *et al.*, 1998).

2.13.1 Basic types of genotype x environment interactions

2.13.1.1 No interactions

A no genotype x environment interaction occurs when one genotype e.g. A consistently performs better than the other genotype (B) by about the same magnitude across all the environments included in the test environment (Simmonds, 1979) (Figure 2.1).

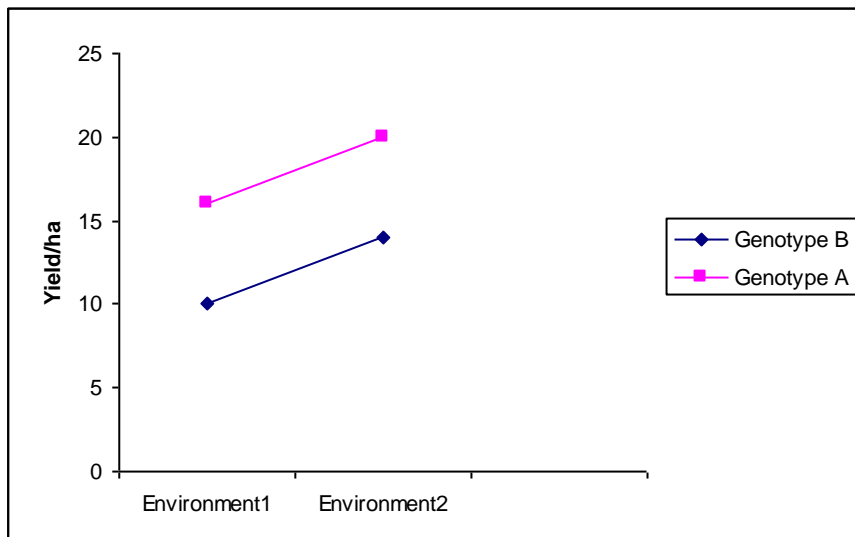


Figure 2.1. Non interaction effect of genotype by environment for yield response.

2.13.1.2 A non-crossover genotype by environment interaction

A non-crossover genotype by environment interaction is said to occur when genotype A consistently outperforms genotype B, across the entire test environment (Kang, 2005). However, the differential performance is not the same across the environment. That is, there is no change in rank, genotype A may exceed genotype B by 20 units in one environment and 60 units in another (Figure 2.2).

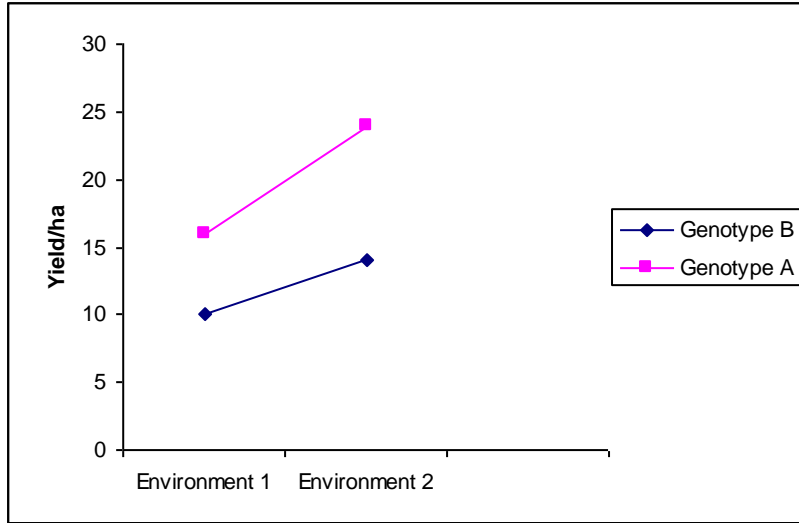


Figure 2.2. Non-crossover interaction effect of genotype by environment for yield response.

2.13.1.3 A crossover genotype x environment interaction

This is the most important genotype x environment interaction to plant breeders. A crossover genotype x environment interaction occurs when a genotype (A) is more productive in one environment, but a different genotype (B) is more productive in another environment. The basic test for crossover interaction (also called qualitative interaction) is to compare the performance of two genotypes in two environments and to determine if the difference in performance is significantly less than zero in one environment and significantly greater than zero in the other (Acquaah, 2007) (Figure 2.3).

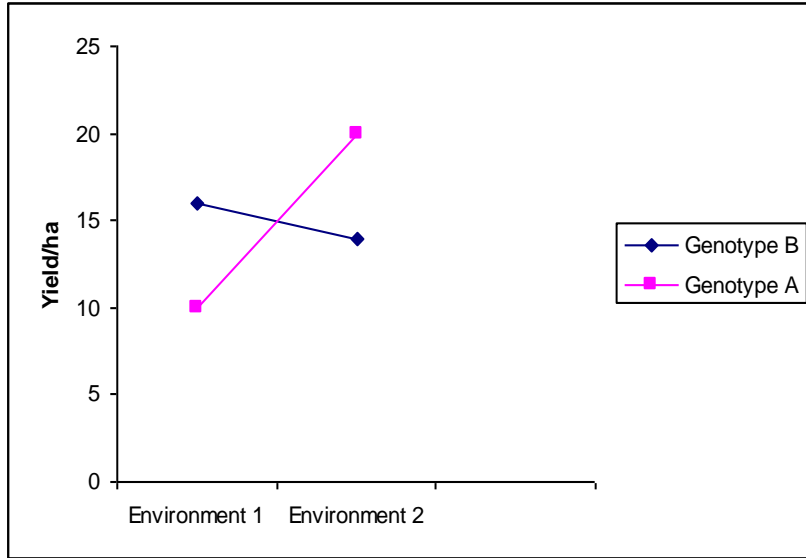


Figure 2.3 Crossover interaction effect of genotype by environment for yield response.

CHAPTER 3

GENOTYPE BY ENVIRONMENT INTERACTIONS AMONG SELECTED SOYBEAN GENOTYPES FOR YIELD AND YIELD COMPONENTS

3.1 INTRODUCTION

Soybean (*Glycine max* (L.) Merrill) is one of the world's most important legume crop. It serves as human diet, animal feed, soil fertility enhancement and for various other industrial applications. Currently, there is an increasing interest to use soybean for bio-fuel production due to the dwindling supply and increased price of fossil fuels.

Soybean cultivars with stabilizing yields and economic returns are required for large scale production. Currently South Africa produces approximately 190000 tons of commercial soy per annum. Between 60000-65000 tons are processed for human consumption and the remainder used mainly for animal feed (Smit and de Beer, 1991).

Yield stability of candidate cultivars across a range of production environments is important in plant breeding programs to make specific or wide area recommendation. Ideally candidate cultivars should have the genetic potential for superior performance under target growing conditions, and should also produce acceptable yields under less favorable environments. Therefore, a stable genotype can be referred to as the one that is capable of utilizing the resources available in high yielding environments and has a mean performance that is above average in all environments (Allard and Bradshaw, 1964; Eberhart and Russell, 1966). Plant breeders often apply genotype by environment interaction stability statistics to assess the performance of their crosses or advanced genotypes across environments. Selection of the best commercially suitable cultivars for a target group of environments is based on the information obtained from evaluation of cultivars grown in a sample of growing environments.

Yield is a complex trait which is dependent on a number of interrelated characters and is highly influenced by many genetic factors as well as many environmental fluctuations (Joarder *et al.*, 1978). The presence of a significant genotype by environment interaction for quantitative traits such as seed yield can reduce the usefulness of subsequent analysis, restrict the significance of inferences that would otherwise be valid, and

seriously limit the feasibility of selecting superior genotypes (Flores *et al.*, 1998). Baker (1988) defined genotype by environment interaction as the failure of genotypes to achieve the same relative performance in different environments. However, in most cases, breeders look for a variety that has good mean performance over a wide array of environments and years and the concept of stability is overlooked. Such approach is reasonable if there is no genotype by environment interactions, but in most cases there are interactions. Some genotypes can have high yield in few environments and very low yield in other environments, showing better mean performance across environments. But few genotypes may have average yield that is stable over wider environments. Genotype by environment interaction constitutes an important limiting factor in the estimation of variance components and in the efficiency of selection programmes (Sprague, 1966). Genotype by environment interactions have been reported in soybean (Smit and de Beer, 1991) and other crops including potatoes (Steyn *et al.*, 1993), lucerne (Smit and de Beer, 1991; Smith and Smith, 1992), sunflower (De La Vega and Chapman, 2006) and wheat (Annicchiarico *et al.*, 2005).

Wider adaptability and stability are prime considerations in formulating an efficient breeding strategy. Irrespective of how it is defined or measured, one of the most critical questions for stability parameters is whether it is genetic (Lin and Binns, 1991). In the past, inheritance of some of these stability parameters was examined using quantitative genetic methods (Farshadfar *et al.*, 1999; Lin and Binns, 1991; Ortiz *et al.*, 2001; Sneller *et al.*, 1997; Zavalagarcia *et al.*, 1992). Knowledge of the pattern and magnitude of genotype by environment interaction and stability analysis is important for understanding the response of different genotypes to varying environments and for identification of stable and widely adapted and unstable but specifically adapted genotypes. Moreover, it is important for breeding new cultivars with improved adaptation to the environmental constraints prevailing in the target environments (Morales *et al.*, 1991).

Genotype by environment interaction is a major concern for a breeder, because such interaction confounds the selection of superior cultivars by altering their relative productiveness in different environments (Eagles and Frey, 1977). Varietal stability in yield with respect to wide range of environments is one of the most desired properties of genotypes to fit the crop under available cropping pattern. Stability analysis helps to measure the adaptability of different crop varieties to varying environments (Morales *et al.*, 1991). Stability statistics provide estimates of each genotype's contribution to the genotype by environment interaction complex and a basis for making selection decisions. Using this approach, many efforts have been made

towards developing statistical models for interpreting genotype by environment interactions in various crop species (Romagosa *et al.*, 1996; Crossa *et al.*, 1999; Campbell *et al.*, 2004; Malosetti *et al.*, 2004). These methods have enhanced the capacity of plant breeders to deal with genotype by environment interactions by providing a basic understanding of their physiological and environmental causes. Two genetic mechanisms were proposed by Via *et al.* (1995) to underpin stability i.e. the allelic sensitivity model and the gene regulation model. The allelic sensitivity model suggests that the constitutive genes themselves regulated in direct response to the environment, by activation of different alleles in different environments. In the gene regulation model, one or more regulatory loci are under the direct influence of the environment and the constitutive gene is switched on and off by the regulatory gene. As suggested by Kraakman *et al.* (2004), co-location of QTLs exhibiting QTL by environment interactions and QTLs for stability parameters would support the allelic sensitivity model. Where QTLs for stability parameters are detected at regions other than those for the trait itself, this would suggest a regulatory model.

In South Africa soybean yield and other agronomic traits are strongly affected by genotype by environment interactions (Smit and de Beer, 1991). Several other studies supported this including in maize (Laubscher *et al.*, 2000), wheat (Purchase, 1997; Purchase *et al.*, 2000), linseed (Adugna and Labuschagne, 2002), and Ethiopian mustard (Kassa, 2002). The objective of this study was to determine genotype by environment interactions of yield and important agronomic traits among ten selected soybean genotypes in two localities in Limpopo Province using stability parameters. Stable and promising genotypes may be identified for production and future breeding purposes across these or similar environments in South Africa.

3.2 MATERIAL AND METHODS

3.2.1 Study sites

Field experiments were conducted in Limpopo Province, South Africa under dryland conditions during the 2007/2008 and 2008/2009 growing seasons. Experiments were established at two localities namely at Syferkuil University of Limpopo's experimental farm situated in Capricorn District and at Gabaza farm in Mopani District near Tzaneen. Syferkuil is situated at 23° South and 29° East and an altitude of 1261.6 m above sea level. It has annual maximum temperature ranging from 28 °C to 30 °C and receives an average annual rainfall of 550 mm. This farm has sandy loam soil, of the Hutton form, Glenrosa family, with the pH ranging from 6.0-6.2. Gabaza farm (23° South and 30° East) lies at an altitude of 1100 m with an annual average rainfall of 700 mm. At Gabaza the annual average temperature ranges from 15 °C-37 °C and with clay-loam soil type. In general, soil, climatic, and biological conditions of the two locations varied considerably.

3.2.2 Plant materials and experimental design

Ten soybean genotypes were used for the study without artificial inoculation with rhizobium bacteria. The list and pedigree of the lines are indicated in Table 3.1. The first nine lines were supplied by the Agricultural Research Service of the United States Department of Agriculture. The experiments were laid out in a randomized complete block design with three replications. The plot size was 4.5 m X 1.5 m with an intra-row spacing of 30 cm and inter-row spacing of 75 cm.

Table 3.1. Name and pedigree of soybean lines used in the study.

Line	Pedigree/description
Harosoy 63	Harosoy(8) x Blackhawk
L82-1449	Harosoy(6) x D54-2437
L86-493	L2,Rps1 x (Harosoy(5) x D54-2437)Rps2 Rmd Rj2
Williams	Wayne x L57-0034 (Clark x Adams)
Clark	Lincoln(2) x Richland
Barc-4	Clark 63*8/Hardee
L81-4858	L6(2) x L63-1889
L72-2133	L12 x L63-1889,rj1
Barc-2	Clark 63(8) x (Hill x Clark)
Magoye	Local/promiscuous

3.2.3 Data collection and statistical analysis

During the study data were collected on seed yield, yield components and nodule formation. Germination percentage was determined by counting number of seed germinated per plot. The number of days to 50% flowering was determined when 50% of the plants per genotype had flowered. The number of nodules per plant and number of pods per plant were counted from five plants randomly selected from the inner rows. The collected nodules were weighed and expressed in grams. The numbers of active nodules was counted by observing cross sectioned internal color, nodules with reddish or pink internal colors were scored as active (effective) and those with green or white color were scored as non-active (ineffective) nodules. Days to physiological maturity and plant height were determined when 50% of individuals per genotype had matured. Above ground biomass was oven dried at 60 °C and 24 hours and weight using a standard balance scale. Seed yield was determined in grams during harvest from the two middle rows of each plot. Plot yield was later converted to kg/ha. The weight of 100 seed was determined in grams from randomly selected seeds after harvest. Temperature and rainfall were recorded from the nearest meteorological stations of each location. To estimate genotype by environment interactions a combined analysis of variance (ANOVA) of yield was conducted across environments using the ANOVA procedure (Agrobases, 2005). Mean comparisons among cultivars was performed using the LSD procedure at 5% probability level (Gomez and Gomez, 1984; Steele *et al.*, 1997). To assess yield stability, Eberhart and Russel's (1966) joint regression model was used and number of nodules, number of active nodules and seed yield of each genotype were regressed on the mean environmental yields. Accordingly, a cultivar was considered stable

when it showed a regression coefficient (b_i) close to unity and a deviation from regression (residual variance= $\sum s^2d_i$) close to zero. To compute the superiority index (P_i) (Lin and Binns, 1988) the maximum mean yield among all genotypes was noted at each location. Then for each genotype, the mean square difference between its yield and the maximum yield at that environment was determined.

3.3 RESULTS

3.3.1 Germination percentage

There were significant ($P<0.05$) differences due to genotype by location interaction with regards to germination percentage (Appendix 6.1). At Syferkuil during 2007 growing season genotypes Williams, Clark and L81-4858 had higher germination percentages both at 96.67%, while Barc-4 showed the lowest germination percentage of 83.33%. In 2008 growing season genotype Williams had 100% germination followed by L86-493 and Clark both with 96.67%. In this year Barc-4 was also the lowest in germination (86.67%) (Table 3.2).

At Gabaza during 2007 growing season genotype Harosay 63 had the highest germination percentage of 96.67% followed by Clark and L81-4858 both with 93.33% germination. While genotype Magoye had the lowest germination percentage of 63.33%. In 2008 growing season genotypes Harosay 63, Clark and L81-4858 had the highest germination percentages at 93.33%. While genotypes Magoye and Barc-2 displayed the lowest germination percentage of 63.33% and 73.33%, respectively (Table 3.2). Over all, Magoye had minimum germination at 63.33% followed by Barc-2 with 73.33%. The genotype with the highest germination percentage was Williams with 100% germination.

There were marked differences in the ranking of genotypes across locations and years. However, compared to other genotypes L81-4858 had relatively better and consistent germination percentage across both environments (Table 3.2). The overall mean of germination percentage of genotypes was 89.24 with a coefficient of variation of 12.63% (Appendix 6.1).

Table 3.2. Mean germination percentage and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	93.33	4	96.67	1	90.00	7	93.33	1
L82-1449	90.00	8	90.00	4	90.00	8	86.67	5
L86-493	93.33	5	83.33	7	96.67	2	83.33	7
Williams	96.67	1	90.00	5	100.00	1	86.67	6
Clark	96.67	2	93.33	2	96.67	3	93.33	3
Barc-4	83.33	10	90.00	6	86.67	10	90.00	4
L81-4858	96.67	3	93.33	3	96.67	4	93.33	2
L72-2133	93.33	6	83.33	8	93.33	5	83.33	8
Barc-2	93.33	7	80.00	9	90.00	9	73.33	9
Magoye	90.00	9	63.33	10	93.33	6	63.33	10
Mean	92.67		86.33		93.33		84.48	
LSD(0.05)	10.80		19.78		10.93		22.06	
CV%	8.23		16.18		8.27		18.07	

R = RANK.

3.3.2 Number of days to 50% flowering

Highly significant ($P < 0.01$) differences were detected due to genotype by location interactions for the number of days to 50% flowering (Appendix 6.2). Figure 3.1 shows flowering of soybean. At Syferkuil during 2007 growing season, genotype L72-2133 displayed shortest days to flowering (52 days) followed by Barc-4 (53 days) and Williams (54 days). Magoye took longer days to reach 50% flowering with 82 days, followed by genotypes Clark and Barc-2 both with 56 days. In 2008 growing season, Barc-4 and L72-2133 flowered early with 53 days followed by L81-4858 (54 days). During this year Magoye had delayed flowering (82 days) (Table 3.3).

At Gabaza during both years genotype Clark was the earliest to flower with 46 days followed by Harosoy 63 and Williams (48 days). Magoye was the last to attain 50% flowering with 69 days (Table 3.3).

Genotype L81-4858 showed better and consistent early flowering across both environments than other genotypes (Table 3.3). The overall mean for the ten soybean genotypes to reach 50% flowering were 55 days after planting with a coefficient of variation of 3.05% (Appendix 6.2).



Figure 3.1. Soybean during flowering.

Table 3.3. Mean number of days to 50% flowering and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	56.00	7	48.67	2	56.00	5	48.67	2
L82-1449	55.33	6	51.67	8	56.00	6	51.67	8
L86-493	54.67	5	51.67	9	56.00	7	51.67	9
Williams	54.33	3	48.67	3	55.00	4	48.67	3
Clark	56.67	9	46.67	1	56.00	8	46.67	1
Barc-4	53.33	2	51.33	6	53.00	1	51.33	6
L81-4858	54.33	4	48.67	4	54.00	3	48.67	4
L72-2133	52.67	1	50.00	5	53.00	2	50.00	5
Barc-2	56.00	8	51.33	7	56.00	9	51.33	7
Magoye	82.67	10	69.33	10	82.67	10	69.33	10
Mean	57.60		51.80		57.77		51.80	
LSD(0.05)	2.65		3.21		2.07		3.21	
CV %	3.25		4.38		2.53		4.38	

R = RANK.

3.3.3 Number of nodules

Highly significant ($P < 0.01$) differences were found due to genotype by location interactions for number of nodules among ten soybean genotypes (Appendix 6.3). At Syferkuil during 2007 growing season, genotype Harosoy 63 produced high number of nodules (13 nodules per plant), followed by Barc-2 (11 nodules per plant) and L82-1449 (10 nodules per plant). Magoye produced lowest number of nodules (7 nodules per plant) followed by L72-2133 (8 nodules per plant). At Syferkuil (2008) Harosoy 63 was the highest with 12 nodules per plant followed by Barc-2 and Williams both with 10 nodules per plant. Magoye (8 nodules per plant), Barc-4 (9 nodules per plant) and L72-2133 (9 nodules per plant) were the lowest with regards to number of nodules per plant (Table 3.4).

At Gabaza during 2007 growing season, Williams (9 nodules per plant), Harosoy 63 (8 nodules per plant) and L81-4858 (8 nodules per plant) produced high number of nodules per plant. Genotype Magoye (6 nodules per plant) and L86-493 (7 nodules per plant) produced lowest number of nodules per plant. At Gabaza (2008) Williams and Barc-4 produced high number of nodules per plant both with eight nodules per plant. Genotype Magoye and Barc-2 had six nodules per plant, which was the lowest in the trial (Table 3.4).

Compared to other genotypes Harosoy 63 and Williams had relatively better and consistent number of nodules across both environments (Table 3.4). The overall mean of eight number of nodules per plant was displayed by the genotypes with coefficient of variation of 3.48% (Appendix 6.3).

Table 3.4. Mean number of nodules per plant and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	12.67	1	8.33	2	11.67	1	7.33	6
L82-1449	10.33	3	7.67	7	9.33	6	7.33	3
L86-493	9.67	5	6.67	9	9.67	5	6.33	8
Williams	10.00	4	8.67	1	10.00	3	8.00	1
Clark	8.67	7	8.00	4	9.67	4	7.33	5
Barc-4	8.67	8	7.67	6	8.67	9	7.50	2
L81-4858	9.00	6	8.00	3	8.67	7	7.00	7
L72-2133	8.33	9	7.67	5	8.67	8	7.33	4
Barc-2	10.67	2	7.33	8	10.33	2	6.33	9
Magoye	7.33	10	6.33	10	8.00	10	6.33	10
Mean	9.53		7.63		9.47		7.07	
LSD(0.05)	1.65		1.47		1.22		1.34	
CV %	30.72		44.49		40.03		49.88	

R = RANK;

3.3.4 Nodule weight

There were significant ($P < 0.05$) interaction among genotypes by locations for nodule weight (Appendix 6.4). At Syferkuil (2007), Barc-4 and Magoye were ranked 1st and 2nd both with 3 grams per nodule per plant respectively. These were followed by L86-493 and L81-4858 both with 2.67 grams per nodule per plant. During 2008 growing season, Magoye and Barc-4 ranked high with regards to nodule weight of 3 grams per nodule per plant (Table 3.5).

At Gabaza (2007), genotypes L180-1449 and L86-493 were ranked high with regard to nodule weight at 1.33 and 1.33 grams per plant, respectively, followed by Magoye (3.0 grams) and Williams (2.67 grams).

Clark resulted in low nodule weight of 2.33 grams. During 2008 growing season, Magoye (3.0 grams) resulted in high nodule weight, followed by L86-493 (2.67 grams) and L180-1449 (2.67 grams). Genotype Clark was low with weight of 2.33 grams while Williams and Harosoy 63 had 2 grams nodule weight (Table 3.5).

There were marked differences in the ranking of genotypes across locations and years. However, compared to other genotypes Magoye had relatively better and consistent nodule weight across both environments (Table 3.5).

Table 3.5. Mean nodule weight (grams per plant) and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	2.33	7	2.00	10	2.33	7	2.00	9
L82-1449	2.00	10	3.33	1	2.00	10	2.67	3
L86-493	2.67	3	3.33	2	2.67	3	2.67	2
Williams	2.00	8	2.67	4	2.00	8	2.00	10
Clark	2.33	6	2.33	9	2.33	6	2.33	8
Barc-4	3.00	1	2.33	7	3.00	1	2.50	4
L81-4858	2.67	4	2.67	5	2.67	4	2.33	6
L72-2133	2.67	5	2.33	8	2.67	5	2.33	7
Barc-2	2.00	9	2.33	6	2.00	9	2.33	5
Magoye	3.00	2	3.00	3	3.00	2	3.00	1
Mean	2.47		2.63		2.47		2.41	
LSD(0.05)	2.57		3.06		2.57		2.73	
CV %	85.52		118.1		85.52		122.5	

R = RANK.

3.3.5 Number of active nodules

No significant interactions of genotype by location or genotype by year were detected. Significant ($P < 0.01$) differences were observed on year and location for number of active nodules (Appendix 6.5). Figure 3.2 depicts active nodules of soybean genotypes. At Syferkuil during 2007 growing season, genotype Harosoy 63 (7 active nodules) ranked first followed by Clark and Barc-2. Barc-4 (5 active nodules) and Magoye (4 active nodules) ranked 9th and 10th with regards to active nodules. At Syferkuil (2008) Harosoy 63 and Clark had six active nodules while Magoye (4 active nodules) and Barc-4 (5 active nodules) had a low number of active nodules (Table 3.6).

At Gabaza during 2007 growing season, genotype Barc-2 (6 active nodules) had a high number of active nodules followed by BARC-4 and L81-4858 both with five active nodules. L180-1449, L86-493 and Clark had a low number of active nodules. During 2008 growing season, L81-4858, L72-2133 and Williams had four active nodules per plant. Genotypes L86-493, Clark and Barc-2 had a low number of active nodules both with three active nodules (Table 3.6). Considering the rankings there were no consistency among genotypes across both environments (Table 3.6).



Figure 3.2. Photo showing active nodules in soybean plants during the experiment.

Table 3.6. Mean number of active nodules and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	6.67	1	4.33	5	5.67	1	3.33	5
L82-1449	6.00	4	3.33	8	5.33	5	3.33	6
L86-493	5.67	5	3.33	9	5.67	3	3.00	8
Williams	5.67	6	4.33	6	5.00	7	3.67	2
Clark	6.33	2	3.33	10	5.67	2	3.00	9
Barc-4	5.00	9	5.00	2	4.67	9	3.50	4
L81-4858	5.67	7	4.67	3	5.00	8	4.00	1
L72-2133	5.00	8	4.67	4	5.33	6	3.67	3
Barc-2	6.33	3	6.00	1	5.33	4	3.00	10
Magoye	4.33	10	4.00	7	4.00	10	3.33	7
Mean	5.67		4.30		5.16		3.38	
LSD(0.05)	3.66		3.78		2.70		3.27	
CV %	31.88		54.68		37.81		63.95	

R = RANK.

3.3.6 Days to 50% physiological maturity

There were highly significant differences ($P < 0.01$) with respect to genotype by location interactions for days to 50% physiological maturity (Appendix 6.6). The average time to 50% physiological maturity among genotypes varied from 105–159 days at all environments (Table 3.7). The highest number of days to attain 50% maturity were noted for genotypes Magoye, Clark and Barc-4. At Syferkuil during 2007 and 2008 growing season Magoye, Clark and Barc-4 were the last genotypes to attain 50% physiological maturity with 159, 113 and 112 days respectively. Genotypes Barc-2 and L72-2133 were first to attain 50% physiological maturity with 105 days each during 2007 and 2008 growing season (Table 3.7).

At Gabaza in all growing seasons Herosoy 63 and L82-1449 were the first genotypes to attain 50% physiological maturity with 105 days each. Genotypes L72-2133 and Magoye were the last to attain 50% physiological maturity with 109 and 124 days respectively.

Table 3.7. Mean number of days to 50% physiological maturity and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	107.33	5	105.67	9	107.33	5	105.67	9
L82-1449	106.00	6	105.33	10	106.00	6	105.33	10
L86-493	106.00	7	108.67	3	106.00	7	108.67	3
Williams	108.00	4	107.00	7	108.00	4	107.00	7
Clark	113.67	2	106.67	8	113.67	2	106.67	8
Barc-4	112.33	3	107.00	6	112.33	3	107.00	6
L81-4858	106.00	8	109.00	2	106.00	8	109.00	2
L72-2133	105.33	9	108.67	4	105.33	9	108.67	4
Barc-2	105.33	10	108.00	5	105.33	10	108.00	5
Magoye	159.33	1	124.67	1	159.33	1	124.67	1
Mean	112.93		109.07		112.93		109.07	
LSD(0.05)	6.25		3.88		6.25		3.88	
CV %	3.91		2.51		3.91		2.51	

R = RANK.

3.3.7 Plant height

This character showed highly significant ($P < 0.01$) interactions amongst genotype by year by location. Further significant ($P < 0.05$) interactions of genotype by year interactions were also observed with regard to plant height (Appendix 6.7). At Syferkuil during 2007 growing season genotype Magoye ranked first with plant height of 98.67 cm followed by Harosoy 63 and Clark (Table 3.8). Barc-2 had the shortest plant height at 38.67cm. During 2008 growing season Magoye and L82-1449 ranked 1st and 2nd with plant height at 92.67 cm and 53 cm, respectively. The shortest genotypes were Barc-2 with 38.67 cm followed by Barc-4 with 44.67 cm respectively. Plant height at Syferkuil ranged from 38.67 to 98.67 cm (Table 3.8).

At Gabaza during 2007 growing season Magoye was the highest with plant height at 64 cm followed by genotype Clark with 43 cm. The lowest genotypes with regards to plant height were Barc-4 and Barc-2 with heights of 34.33 cm and 28.33 cm respectively. Magoye (62.67 cm) and Clark (41.67 cm) again had highest plant heights. The shortest genotype was Barc-2 with height of 27 cm. The ranges for plant height at Gabaza were from 27 to 64 cm (Table 3.8).

Table 3.8. Mean plant height (cm) and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	58.67	2	41.33	5	53.00	4	39.67	7
L82-1449	55.33	4	35.67	8	55.00	2	34.00	8
L86-493	52.00	6	42.67	3	52.33	5	41.33	3
Williams	50.67	7	41.00	7	48.00	7	40.33	6
Clark	58.67	3	43.00	2	54.67	3	41.67	2
Barc-4	44.67	9	34.33	9	41.67	9	32.50	9
L81-4858	55.33	5	42.67	4	50.00	6	41.33	4
L72-2133	48.67	8	41.33	6	45.00	8	40.67	5
Barc-2	38.67	10	28.33	10	38.67	10	27.00	10
Magoye	98.67	1	64.00	1	92.67	1	62.67	1
Mean	56.13		41.43		47.10		40.38	
LSD(0.05)	8.26		10.29		26.72		10.77	
CV %	10.39		17.54		40.07		18.47	

R = RANK.

3.3.8 Number of pods per plant

The results showed highly significant ($P < 0.01$) interactions between genotype by location with respect to number of pods per plant (Appendix 6.8). During 2007 at Syferkuil Magoye (190 pods per plant) and Clark (134 pods per plant) had more number of pods per plant than other genotypes. Genotypes that had low number of pods per plant were L72-2133 (59 pods per plant) and Harosoy 63 (60 pods per plant). During 2008 Magoye (222 pods per plant) produced high number of pods per plant, followed by Clark (130 pods per plant) and Barc-2 (107 pods per plant), while Barc-4 (57 pods per plant) and Harosoy 63 (54 pods per plant) had the lowest number of pods per plant (Table 3.9).

At Gabaza during 2007 growing season, Magoye (143 pods per plant) and Barc-2 (71 pods per plant) produced high number of pods per plant. Genotypes Harosoy 63 (50 pods per plant) and Williams (43 pods per plant) had the lowest number of pods per plant. During 2008 Magoye (182 pods per plant) also obtained high number of pods per plant followed by Barc-2 (68 pods per plant) and L81-4858 (55 pods per plant). BARC-4 (44 pods per plant) and Williams (41 pods per plant) had lowest number of pods per plant (Table 3.9).

Table 3.9. Mean number of pods per plant and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	59.67	9	49.67	9	54.33	10	47.67	8
L82-1449	68.67	8	54.00	5	66.00	7	52.00	6
L86-493	110.33	4	52.67	8	106.00	4	54.33	4
Williams	109.33	5	43.33	10	104.67	5	41.00	10
Clark	134.00	2	54.00	6	130.00	2	53.00	5
Barc-4	69.33	7	54.00	4	56.67	9	43.50	9
L81-4858	96.67	6	56.67	3	95.33	6	55.33	3
L72-2133	58.67	10	53.33	7	58.00	8	51.33	7
Barc-2	111.33	3	70.67	2	107.00	3	68.00	2
Magoye	190.33	1	143.00	1	221.67	1	181.67	1
Mean	100.83		63.23		99.97		65.52	
LSD(0.05)	40.27		21.41		44.22		32.57	
CV %	28.21		23.92		31.24		34.41	

R = RANK.

3.3.9 Number of seeds per pods

A significant ($P < 0.05$) interaction was obtained amongst genotype by year by location. Also highly significant ($P < 0.01$) interaction was obtained amongst genotype by location for the number of seeds per pod (Appendix 6.9). At Syferkuil during 2007 growing season Harosoy 63 (3.33 seeds per pod) and Barc-2 (3.33 seeds per pods) produced highest number of seeds per pod. Genotypes Barc-4 (2 seeds per pods), Clark (2 seeds per pods), L81-4858 (2 seeds per pods) and L82-1449 (seeds per pods) were least with the number of seeds per pod. During 2008 growing season all genotype produced two number of seeds per pod, only L81-4858 produced 2.33 seeds per pod (Table 3.10).

At Gabaza during 2007 growing season, Barc-2 and Barc-4 produced 3.33 number of seeds per pod. L82-1449 and Magoye had the least number of seeds per pod (2.33 seeds per pods). During 2008 growing season almost all genotypes produced two seeds per pod, only L82-1449 produces 1.67 number of seeds per pod (Table 3.10).

Table 3.10. Mean number of seeds per pod and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	3.33	1	3.00	6	2.00	2	2.00	1
L82-1449	2.00	8	2.33	9	2.00	7	1.67	10
L86-493	3.00	4	3.00	4	2.00	3	2.00	3
Williams	2.67	6	3.00	7	2.00	4	2.00	4
Clark	2.00	10	3.00	3	2.00	5	2.00	5
Barc-4	2.00	9	3.33	1	2.00	6	2.00	6
L81-4858	2.00	7	3.00	5	2.33	1	2.00	2
L72-2133	3.00	3	2.67	8	2.00	8	2.00	8
Barc-2	3.33	2	3.33	2	2.00	9	2.00	9
Magoye	2.67	5	2.33	10	2.00	10	2.00	7
Mean	2.73		2.53		2.03		1.97	
LSD(0.05)	0.69		0.62		0.26		0.27	
CV %	10.29		10.17		8.98		9.57	

R = RANK.

3.3.10 Dry matter

Significant ($P < 0.01$) interactions were obtained amongst genotype by location and genotype by year for dry matter content per plant (Appendix 6.10). At Syferkuil during 2007 growing season, Barc-4 had 10 grams dry matter per plant and L72-2133 (14 grams). Magoye with 30.67 grams and Harosoy 63 with 28.33 grams were the best genotypes with the greatest dry matter yield. At Syferkuil (2008) Harosoy 63 (7.33 grams) and Barc-4 (7.33 grams) were the lowest genotypes in terms of dry matter. Genotype Magoye (29.67 grams) and Barc-2 (23.67 grams) had the best dry matter content (Table 3.11).

At Gabaza during 2007 growing season Barc-2 (10.67 grams) and Williams (11.33 gram) had the lowest dry matter content. Magoye with 21.33 grams and L82-1449 with 16.67 grams had the greatest dry matter. At Gabaza (2008) Barc-2 (6.67 grams) and L72-2133 (8.33 grams) yielded the lowest dry matter. Genotypes Magoye (19.33 grams) and L86-493 (14.67 grams) were again the best lines to attain high dry matter. Overall, the landrace Magoye was the best line for high dry matter content across all environments (Table 3.11).

Compared to other genotypes Magoye had relatively better and consistent dry matter across both environments (Table 3.11). The overall mean of dry matter of genotypes was 15.68 grams with the coefficient of variation at 29.85% (Appendix 6.10).

Table 3.11. Mean dry matter (gram/plant) and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	28.33	2	16.33	3	7.33	10	10.00	6
L82-1449	21.33	6	16.67	2	14.33	6	9.00	8
L86-493	14.67	8	14.67	5	14.67	5	14.67	2
Williams	16.33	7	11.33	9	14.00	7	10.67	5
Clark	23.33	4	15.33	4	20.67	3	10.00	7
Barc-4	10.00	10	14.00	6	7.33	9	11.00	4
L81-4858	22.00	5	12.67	8	19.33	4	11.33	3
L72-2133	14.00	9	14.00	7	11.33	8	8.33	9
Barc-2	24.67	3	10.67	10	23.67	2	6.67	10
Magoye	30.67	1	21.33	1	29.67	1	19.33	1
Mean	20.53		14.70		16.23		11.10	
LSD(0.05)	10.14		4.31		6.64		3.96	
CV %	34.88		20.70		28.88		24.69	

R = RANK.

3.3.11 Hundred seed weight

Significant ($P < 0.01$) genotype by location interactions were obtained for hundred seed weight (Appendix 6.11). Table 3.12. represents test seed weight from 100 randomly selected seed of each genotype. At Syferkuil during 2007 growing season Williams and L81-4858 had hundred seed weight of 23.33 g. Magoye had relatively low seed weight at 14.67 g, followed by L86-493 with 17.67 g. In 2008 growing season Williams showed the heaviest seed weight with 21.67 g and Magoye showed the lowest seed weight with 12.67 g followed by L82-1449.

At Gabaza during 2007 genotype Barc-4 showed the heaviest seed weight with 20.67 g followed by Clark with 18 g. Genotype Magoye showed the lowest seed weight with 13 g followed by Barc-2 with 16.33 g. In

2008 growing season genotype Barc-4 showed also heaviest seed weight with 20.5 g followed by Clark with 17.67 g. Magoye also showed the lowest seed weight at 12.33 g followed by L72-2133 with 15 g. The environment which showed the heaviest seed weight was Syferkuil during 2007 growing season (Table 3.12). Most genotypes showed relatively better and more consistent hundred seed weight at Syferkuil (Table 3.12).

Table 3.12. Mean hundred seeds weight (grams) and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	20.00	5	18.00	3	19.00	5	15.33	8
L82-1449	18.67	8	16.67	5	18.33	9	16.67	5
L86-493	17.67	9	15.33	7	18.67	8	16.00	6
Williams	23.33	1	14.33	9	21.67	1	15.67	7
Clark	19.33	7	18.00	2	18.67	7	17.67	2
Barc-4	20.00	6	20.67	1	19.00	6	20.50	1
L81-4858	23.33	2	16.67	4	21.67	2	17.33	3
L72-2133	20.67	3	15.33	8	19.33	3	15.00	9
Barc-2	20.67	4	16.33	9	19.33	4	16.67	4
Magoye	14.67	10	13.00	10	12.67	10	12.33	10
Mean	19.83		16.43		18.83		16.17	
LSD(0.05)	2.83		3.60		2.80		3.05	
CV %	10.09		15.46		10.51		13.05	

R = RANK.

3.3.12 Seed yield

This character showed highly significant ($P < 0.01$) genotype by location interactions (Appendix 6.12). The average for seed yield ranged from 2895 kg/ha to 5428.47 kg/ha at all environments (Table 3.13). At Syferkuil during 2007 growing season genotypes which obtain highest seed yield were L81-4858 with 7411 kg/ha followed by Clark with 6777.67 kg/ha. Harosoy 63 with 4732.33 kg/ha and Magoye with 2499 kg/ha obtained lowest with regard to seed yield. In 2008 growing season L81-4858 (7425 kg/ha) and Clark (6791.67 kg/ha) had highest seed yield. Genotypes Harosoy 63 (4758.33 kg/ha) and Magoye (2208.33 kg/ha) obtained least seed yield.

At Gabaza during 2007 growing season Clark and Harosoy 63 produced highest seed yield at 3687.33 kg/ha and 3604 kg/ha respectively. Genotypes L81-4858 and Magoye were the least with regard to seed yield. In 2008 growing season genotypes Clark and Harosoy 63 were the highest with regard to seed yield with 3683.33 kg/ha and 3600 kg/ha and L81-4858 and Magoye were the least genotypes with 2258.33 kg/ha and 2083.33 kg/ha. The highest seed yield/ha during 2007 and 2008 was obtained at Syferkuil (Table 3.13). Compared to other genotypes Clark had relatively better and more consistent seed yield across both environments (Table 3.13). The overall mean of seed yield of genotypes was 4177.85 with the coefficient of variation at 27.45% (Appendix 6.12).

Table 3.13. Mean seed yield (kg/ha) and ranks of ten soybean genotypes when evaluated at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Location and year							
	Syferkuil 2007	R	Gabaza 2007	R	Syferkuil 2008	R	Gabaza 2008	R
Harosoy 63	4732.33	9	3604.00	2	4758.33	9	3600.00	2
L82-1449	4979.00	7	2987.33	5	4866.67	7	2983.33	4
L86-493	4844.33	8	2629.00	7	4858.33	8	2625.00	7
Williams	5265.67	6	3445.67	3	5333.33	6	3441.67	3
Clark	6777.67	2	3687.33	1	6791.67	2	3683.33	1
Barc-4	5865.67	4	2989.00	4	5875.00	4	2762.50	6
L81-4858	7411.00	1	2262.33	9	7425.00	1	2258.33	9
L72-2133	5852.67	5	2489.00	8	5866.67	5	2485.00	8
Barc-2	6057.33	3	2987.33	6	6300.00	3	2983.33	5
Magoye	2499.00	10	2087.33	10	2208.33	10	2083.33	10
Mean	5428.47		2916.83		5428.33		2895.00	
LSD(0.05)	1934.22		1363.54		1978.99		1441.72	
CV %	25.17		33.02		25.75		34.47	

R = RANK.

3.4 Stability measures

Two stability measures were conducted on number of nodules, number of active nodules and seed yield. The regression (Eberhart and Russell, 1966) and cultivar superiority (Lin and Binns, 1988) statistics were applied to estimate stability among genotypes.

3.4.1 Eberhart and Russell's joint regression analysis

Eberhart and Russell's (1966) procedure involves the use of joint linear regression where the yield of each genotype is regressed on the environmental mean yield. This stability is the most widely used and published statistical procedure in plant breeding for providing the genotypic stability. The analysis of variance for the

regression model is presented in Table 3.14. The sums of squares due to environments and genotype by environment are portioned into environments (linear), genotype by environment (linear) and deviations from the regression model. The genotype's performance is generally expressed in terms of three parameters, mean yield (\bar{x}), regression coefficient (b) and the deviation (S^2d) from the regression. According to this model a stable genotype should have a high mean yield, $b = 1$ and $S^2d = 0$. It is however specifically the deviation from the regression (S^2d) which is used as a measure of a genotype's stability across environments.

The results showed highly significant ($P \leq 0.01$) differences due to genotype by environment interactions for number of nodules and seed yields. This indicated that genotype performance for number of nodules and seed yield when tested across different environments was not consistent (Table 3.14) (Appendices 6.13, 6.14 and 6.15). There were no significant differences for number of active nodule.

The estimates of the measures of adaptability and stability using Eberhart and Russell (1966) model are shown in Table 3.15. According to this model, with regards to seed yield genotypes Barc-4 and Barc-2 were considered to be relatively stable with better performance of the seed yield, since they had regression coefficients (b) of close to one across environments. Harosoy 63 and L86-493 were unstable genotypes with regression coefficients (b) of lower than one across environments.

With regard to number of nodules genotype Clark and Harosoy 63 were stable, but had regression coefficients (b) of lower or higher than one across environments. Barc-2, Williams and L82-1449 were considered to be relatively stable genotypes with better number of nodules.

The results also showed that with regards to number of active nodules L86-493 and Clark were relatively stable with better number of active nodules, since they had regression coefficient of close to one across environments (Table 3.15). In general, Barc-2 was the most stable, with high mean seed yield (Table 3.14) and number of active nodules at Syferkuil during 2007 and 2008 (Table 3.15).

Table 3.14. Mean and ranks (R) of seed yield (kg/ha) and number of active nodules of genotypes at Syferkuil and Gabaza during 2007 and 2008.

Genotype	Seed yield								Number of active nodules							
	2007				2008				2007				2008			
	Syferkuil		Gabaza		Syferkuil		Gabaza		Syferkuil		Gabaza		Syferkuil		Gabaza	
	Yield	R	Yield	R	Yield	R	Yield	R	Number	R	Number	R	Number	R	Number	R
Harosoy 63	4732.33	9	3604.00	2	4758.33	9	3600.00	2	4.67	1	2.33	4	3.67	1	1.33	4
L82-1449	4979.00	7	2987.33	5	4866.67	7	2983.33	5	4.00	3	1.33	6	3.33	2	1.33	4
L86-493	4844.33	8	2629.00	7	4858.33	8	2625.00	7	3.67	4	1.33	6	3.67	1	1.00	5
Williams	5265.67	6	3445.67	3	5333.33	6	3441.67	3	3.67	4	2.33	4	3.00	4	1.67	2
Clark	6777.67	2	3687.33	1	6791.67	2	3683.33	1	4.33	2	1.33	6	3.67	1	1.00	5
Barc-4	5865.67	4	2989.99	4	5875.00	4	2762.52	6	3.00	5	3.00	2	2.67	5	1.50	3
L81-4858	7411.00	1	2262.33	9	7425.00	1	2258.33	9	3.67	3	2.67	3	3.00	4	2.00	1
L72-2133	5852.67	5	2489.00	8	5866.67	5	2485.00	8	3.00	5	2.67	3	3.33	2	1.67	2
Barc-2	6057.33	3	2987.33	6	6300.00	3	2983.33	4	4.33	2	4.00	1	3.33	2	1.67	2
Magoye	2499.00	10	2087.33	10	2208.33	10	2083.33	10	2.33	6	2.00	5	2.00	6	1.33	4
LSD _(0.05)	1934.22		1363.54		1978.99		1441.72		1.66		1.78		1.70		1.27	

Table 3.15. Stability measure of ten soybean genotypes for three characters.

Genotype	Seed Yield		Number of Nodules		Number of Active Nodules	
	b	S ² d	b	S ² d	b	S ² d
Harosoy 63	0.4545	-475666.96	2.0238	-0.2755	1.7680	-0.3503
L82-1449	0.7704	-471640.80	1.0783	-0.2344	1.3812	-0.0464
L86-493	0.8843	-475659.90	1.4504	-0.4092	1.5627	0.2322
Williams	0.7379	-475009.89	0.7904	-0.4520	0.8603	-0.1972
Clark	1.2326	-475527.61	0.7113	-0.1733	1.5627	0.2322
Barc-4	1.1517	-457499.07	0.4996	-0.4635	0.3867	-0.2619
L81-4858	2.0509	-474959.42	0.6630	-0.3422	0.7261	-0.2350
L72-2133	1.3412	-475473.01	0.4687	-0.4306	0.3867	-0.2619
Barc-2	1.2702	-463675.56	1.7168	-0.4531	0.9787	1.0830
Magoye	0.1063	-454276.19	0.5978	-0.3121	0.3867	-0.2619

3.4.2 Cultivar superiority measure

Table 3.16 indicates cultivar superiority measure and the ranks of ten soybean genotypes for three characters. The superiority measure (P_i) of cultivars is estimated by the squares of differences between an entry mean and maximum entry mean, summed and divided by twice the number of locations (Lin and Binns, 1988). According to Lin and Binns (1988) for cultivar superiority measure (P_i) analysis, the genotype with low or small P_i value are considered to be the more stable. The stability measure indicates that Clark, L81-4858 and Barc-2 were the most stable genotypes with regard to seed yield and L86-493 and Magoye had high P_i values which means they were not stable (Table 3.16). The cultivar superiority measure for number of nodules allocates Harosoy 63 followed by Williams and Barc-2 as the most stable genotypes. Genotypes L72-2133 and Magoye were not stable, since they had a high P_i value with regard to number of nodules (Table 3.16). Further, L81-4858 and Barc-2 were considered to be stable in terms of number of active nodules while L82-1449 and Magoye had high P_i values (Table 3.16).

Compared to other genotypes Barc-2 had relatively better and consistent stability in terms of superiority measure for number of nodules, number of active nodules and seed yield.

Table 3.16. Cultivar superiority measure and the ranks of ten soybean genotypes for three characters.

Genotype	Seed Yield (Pi)	Rank	Number of Nodules (Pi)	Rank	Number of Active Nodules (Pi)	Rank
Harosoy 63	1787963.50	8	0.0706	1	0.6250	3
L82-1449	1679748.50	7	1.5500	4	1.5000	9
L86-493	1927213.25	9	2.4736	5	1.3750	7
Williams	1136706.38	6	1.2397	2	0.7500	4
Clark	100172.25	1	2.6122	6	1.3750	8
Barc-4	847641.13	4	3.2813	8	0.7500	5
L81-4858	507656.25	2	2.9897	7	0.3750	1
L72-2133	965642.00	5	3.6606	9	0.7500	6
Barc-2	509867.63	3	1.2975	3	0.3750	2
Magoye	7058104.13	10	6.2811	10	2.2500	10

3.4 DISCUSSION

The study found 50% flowering to show highly significant ($P < 0.01$) genotype by location interactions among ten selected soybean genotypes (Appendix 6.2). There were short number of days to 50% flowering at Gabaza during 2007 and 2008 than at Syferkuil. This could be attributed to high temperature during flowering at Gabaza (Table 3.17). In this study, in all environments genotype L72-2133 took minimum days to flowering (50 days) and Magoye took maximum days (82 days) (Table 3.3). Although specific factors affecting flowering and pod set were not studied, temperature was reportedly the dominant factor suggested to contribute to early flowering in plants (Lawn and Hume, 1985). Mann and Jaworski (1970) reported that during flowering and pod set, temperatures as high as 30°C favoured greater pod set but temperatures above 40°C severely limited pod formation.

The results showed that there were relatively good number of nodules across all environments (Table 3.4). Adequate nodulation requires about 7-14 nodules per plant (Bohner, 2007). The relative efficiency of nodule initiation can be defined as the number of bacteria required to induce a number of nodules in the initially susceptible region of the root (Bhuvanewari *et al.*, 1983). The fewer the cells required, the more efficient that particular strain-host combination is in the initiation of the first nodules. Many studies suggested that there were good possibilities for increasing nodulation, nitrogen fixation and crop yield of legumes in the

field by inoculation with *Rhizobium* and *Azospirillum* or *Azotobacter* (Iruthayathas *et al.*, 1983; Milic *et al.*, 1993; Plazinski *et al.*, 1984; Singh and Subba, 1979).

Table 3.17. Minimum and maximum temperatures (°C) for each location of the experiments averaged over 2007 and 2008

Month	Syferkuil				Gabaza			
	2007/8		2008/9		2007/8		2008/9	
	Min	Max	Min	Max	Min	Max	Min	Max
November	15.00	27.02	15.89	26.98	16.17	29.54	16.00	30.00
December	16.32	26.21	17.45	29.00	16.80	32.23	16.80	33.87
January	16.60	27.04	18.42	29.17	16.00	32.73	16.60	32.00
February	16.00	29.30	16.59	28.28	16.47	30.95	16.00	30.86
March	14.95	26.74	13.31	27.35	15.00	30.74	16.03	30.67
April	12.00	26.00	13.06	27.24	15.00	29.08	15.45	30.23
Average	15.15	27.05	15.79	28.00	15.90	30.88	18.91	31.27

Table 3.18. Total rainfall (mm) for each location of the experiments averaged over 2007/8 and 2008/9

	Syferkuil		Gabaza	
	2007/8	2008/9	2007/8	2008/9
November	97.0	80.0	107.0	94.5
December	155.4	112.5	138.0	117.0
January	70.0	80.9	90.0	90.0
February	121.4	109.7	133.8	119.9
March	250.1	201.2	340.6	310.2
April	132.3	115.5	167.4	150.1
Average	137.7	116.7	162.8	147.0

This study showed that genotypes varied with regards to physiological maturity. Magoye took 159 days at Syferkuil and 125 days at Gabaza during 2007 and 2008 growing seasons. Magoye was also the last to reach 50% flowering and had delayed physiological maturity (Table 3.3 and Table 3.7).

In this study soybean genotypes showed considerable variation in plant height. Differences existed among genotypes for the trait that ranged from 27 to 98.67 cm across all environments (Table 3.8). The shortest genotype recorded was Barc-2 across almost all environments, while the tallest was Magoye (Table 3.8). Paul *et al.* (2003) found similar results. Many genotypes were observed to be taller at Syferkuil during 2007 and 2008 growing seasons.

The number of pods per plant is related to number of flowers produced, the proportion of flowers that initiated pods, and the proportion of pods that survived to produce grain bearing pods. The number of pods per plant varied between the tested genotypes. Magoye produced the highest number of pods per plant (Table 3.9). This results was due to the fact that genotype Magoye was taller than other genotypes and more smaller leaves were produced during the branching stage as well as during flowering and pod formation. The mean of genotypes ranged from 41 to 221.67 pods per plant. In average across all environment, genotypes with the least pods per plant were Williams, L72-2133 and Harosoy 63. The highest mean number of pods per plant differ significantly from the genotypes that had the lowest number of pods per plant.

The number of seeds per pod is the most important component in determining yield in several legume crops (Pandey and Gritton, 1975). Generally, variation in the number of pods per plant depends on type of legume species. However the number of pods per plant produced or maintained to final harvest, depends on environmental conditions and management practices (Knott and Tolukdar, 1971). The genotypes differently reacted to different environments but majority of them produced higher number of seeds per pod during 2007 growing season at Syferkuil and Gabaza. The results revealed that there were decreases in number of seeds per pod during 2008 growing season at Syferkuil and Gabaza.

The results shows that the genotypes with the highest 100 seed weight were Williams and L81-4858 at Syferkuil and Barc-4 and Clark at Gabaza during 2007 and 2008 growing seasons. This attributed to good quality seeds and higher yields. Hundred seed weight is a measure of seed size. Magoye had the lowest

100 seed weight due to small seed type, and not because of poor yield. Hot dry weather during seed maturation can also result in poor quality (FAO, 1994). Seed quality is however, sensitive to temperature during the seed filling period, because high temperature can differentially affect the various processes involved in seed filling (Table 3.17).

Ayisi (2000), reported that increased seed yield resulted from large seeds. The yield of a given genotype changed with the changes of environment. Therefore evaluation over a wide range of environments needs to be emphasized in relation to individual traits or a combination of traits to yield. The study revealed that yield potential of each genotype changed with the varying environments. In this study seed yield ranged from 2895 to 5428,47 kg/ha. Soybean seed yield ranged from 1560 to 3268 kg/ha as reported by Subhan and Edwards (2001). Akande *et al.* (2007) also reported soybean seed yield of between 1017.24 and 2133.01 kg/ha. The highest yielding genotype across all growing seasons was L81-4858. Genotype Magoye had the lowest seed yield at all environments.

Among the two environments during the two growing seasons, seed yield in all two environments indicate high variation between the genotypes, as rankings were different from one environment to the other. Syferkuil was the best with regards to seed yield. This is contributed to the main environmental factors interacting with genotypes like topography, soil structure and nutrient availability. There were increase in seed yield at Syferkuil during 2008 growing season. At Gabaza during 2008 growing season there was a decrease in seed yield. These could be the indication that temperature and rainfall were favourable to yield that were relatively better at Syferkuil during all growing seasons. Soybean genotypes at Gabaza during all growing season were the earliest to reach 50% days to flowering, physiological maturity and dry matter. These revealed that temperature were favorable for the three traits not for seed yield and other soybean traits (Table 3.17 and Table 3.18).

The tested soybean genotypes had variable seed yield across environments. This was the indication of sensitivity to change to the environment, mainly temperature and rainfall. The study suggests that most genotypes had the capacity to yield high. There were variations in ranks among genotypes for various soybean traits at the same or different environments showing that environment had a strong influence on traits which were quantitatively inherited. Thus, it is important to identify suitable genotypes to synchronize majority of soybean traits with the climatic conditions for maximum yield achievement.

In the present study, the genotype by environment interactions when tested against pooled error were found significant for number of nodule and seed yield characters, indicating that number of nodule and seed yield were highly influenced by the change in environments leading to extension of analysis for estimating stability parameters. The linear portion of genotype by environment interactions were highly significant ($P < 0.01$) for number of nodule and seed yield. Thus the prediction of the genotypes in the environments appeared to be feasible for the two traits under study. Hossain *et al.* (2003) and Singh *et al.* (1995) also found significant linear and non-linear components interactions in soybean.

Analysis of the stability parameters of individual genotypes indicated that there were certain genotypes in the study whose performance was not predictable or they were unstable. Genotypes Clark, Barc-4 and Barc-2 showed a regression coefficient close to one with regards to seed yield, which means that these genotypes are less responsive to favorable environments but should perform well in a more predictable and stable manner. They also showed low P_i with regards to seed yield. Genotype L81-4858 had b-value greater than one and had small P_i , which implies that this genotype should perform better in increasingly favourable environments. The genotypes Harosoy 63, L82-1449, L86-493, Williams and Magoye had b-values less than one and higher P_i values, which suggests that these genotypes will perform better in less favourable environments.

The partitioning of variance components showed that the genotype by environment interactions were mainly due to predictable environmental factors (locations) as indicated by their significance as opposed to the unpredictable factors (years). When genotype by environment interactions are caused by predictable environmental factors, the ultimate goal should be to develop specific cultivars for specific environments (Adungna and Labuschagne, 2002), rather than breeding for stable cultivars that will perform well across all target environments. These location dependent differences may be attributed to environmental variables, such as temperature, rainfall, altitude, and soil characteristics of the test site. However, different genotypes emerged as “winners” in different locations.

CHAPTER 4

SUMMARY AND CONCLUSIONS

The yield and yield components of soybean traits were affected by interactions between genotype and environment. The environmental conditions such as temperature, soil characteristics, rainfall etc. need to be understood in order to achieve stable and increased yield.

Soybean genotype caused large differences in plant height, number of pod per plant, dry matter, 100 seed weight and seed yield. Soybean seed yield was highly variable in this study because of the range of soil types used and environmental conditions. L81-4858 and Clark were the highest yielding genotypes. Magoye was the best genotype with regards to number of pods per plant and plant height across environments. However, Magoye had lower seed per pod and small seed size.

With regards to production, Syferkuil was found to be the best location for soybean cultivation. Low seed yield at Gabaza was probably due to high temperature and poor soil fertility.

The study showed that there were significant genotype by location interactions on 50% days to flowering, number of nodules, number of pods per plant, plant height, days to 50% physiological maturity, dry matter, 100 seed weight and seed yield. Presence of significant interactions makes it difficult for plant breeders to decide the variety for recommendation on inadequate number of locations and years can increase the chance of a wrong decision. If the number of genotypes, locations and years increased, data handling would be a very difficult task, particularly in case of significant interactions. Therefore, further analysis is needed to simplify these interactions.

Two stability parameters were used to determine stable yield, and this aided in enhancing the prediction of genotype performance. Eberhart and Russell's (1966) procedure assisted in selecting the most stable genotypes i.e. Clark, Barc-4 and Barc-2. Multilocational trials help to estimate yields accurately and understanding of genotype with year effect and do not really give the stability of the yield. An assessment of the stability of a crop to a new set of target environments needs to be conducted over years and location to

provide conclusive results. In the current study, results reported from two locations and two years of investigation were sufficiently encouraging. This may serve as an indication that soybean researchers in Limpopo Province should make a more substantial investigation to make specific recommendations. Consequently, the findings of the study must be regarded as a preliminary step that is in need of further confirmation. Therefore it is recommended that the trials be strengthened to determine if genotypes will be stable across other representative locations and years. It will then be easier to select stable genotypes that can be used for targeted breeding.

The study found that most of the tested genotypes performed better at Syferkuil than at Gabaza. Clark, Barc-4 and Barc-2 were the best genotypes which were stable with regards to all characters and had high seed yield and number of active nodules. Barc-2 was selected as the most stable genotype for seed yield, nodule formation and other agronomical characters that can be grown in Limpopo Province or other similar environments.

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6. APPENDICES

Appendix 6.1 Analysis of variance for germination percentage among ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	3213.75	357.08	2.66**	0.0067
Year (Y)	1	7.95	7.95	0.06 ^{ns}	0.8032
Location (Loc)	1	1707.53	1707.53	12.73**	0.0005
G x Loc	9	2560.66	284.52	2.12*	0.0281
G x Y	9	130.23	14.47	0.11 ^{ns}	0.9993
G x Y x Loc	9	78.49	8.72	0.07 ^{ns}	0.9999
Replication in Loc and Year	8	276.25	34.53	0.26 ^{ns}	0.7043
Residual	72	9657.08	134.13		
Total	118	17631.93			
Grand mean 89.24	R- squared 45.23%		C.V. 12.97%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.2 Analysis of variance for days to 50% flowering among ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	6013.083	668.12	241.20**	0.0000
Year (Y)	1	1.748	1.75	0.63 ^{ns}	0.4170
Location (Loc)	1	1165.601	1165.60	420.79**	0.0000
G x Loc	9	296.674	32.96	11.90**	0.0000
G x Y	9	7.028	0.78	0.28 ^{ns}	0.9733
G x Y x Loc	9	24.858	2.76	1.00 ^{ns}	0.4073
Replication in Loc and Year	8	12.585	1.57	0.57 ^{ns}	0.3182
Residual	72	199.42	2.77		
Total	118	7720.99			
Grand mean 54.58	R- squared 97.42%		C.V. 3.05%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.3 Analysis of variance for number of nodules among ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	67.141	7.46	5.22**	0.0000
Year (Y)	1	2.592	2.59	1.81 ^{ns}	0.1713
Location (Loc)	1	136.752	136.75	95.63**	0.0000
G x Loc	9	38.184	4.24	2.97**	0.0030
G x Y	9	5.244	0.58	0.41 ^{ns}	0.9157
G x Y x Loc	9	4.197	0.47	0.33 ^{ns}	0.9574
Replication in Loc and Year	8	7.878	0.98	0.69 ^{ns}	0.2261
Residual	72	103.29	1.43		
Total	118	365.28			
Grand mean 3.44	R- squared 71.72%		C.V. 3.48%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.4 Analysis of variance for nodule weight among ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	9.316	1.04	3.70**	0.0004
Year (Y)	1	0.356	0.36	1.27 ^{ns}	0.2521
Location (Loc)	1	0.103	0.10	0.37 ^{ns}	0.5370
G x Loc	9	5.629	0.63	2.23*	0.0217
G x Y	9	0.776	0.09	0.31 ^{ns}	0.9650
G x Y x Loc	9	1.068	0.12	0.42 ^{ns}	0.9063
Replication in Loc and Year	8	2.211	0.28	0.99 ^{ns}	0.0929
Residual	72	20.29	0.28		
Total	118	39.75			
Grand mean 0.50	R- squared 48.96%		C.V. 10.58%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.5 Analysis of variance for number of active nodules among ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	12.5	1.39	1.08 ^{ns}	0.3498
Year (Y)	1	14.377	14.38	11.14 ^{**}	0.0010
Location (Loc)	1	73.252	73.25	56.78 ^{**}	0.0000
G x Loc	9	20.177	2.24	1.74 ^{ns}	0.0762
G x Y	9	7.285	0.81	0.63 ^{ns}	0.7413
G x Y x Loc	9	6.038	0.67	0.52 ^{ns}	0.8349
Replication in Loc and Year	8	6.753	0.84	0.65 ^{ns}	0.2494
Residual	72	93.08	1.29		
Total	118	233.46			
Grand mean 2.64	R- squared 60.13		C.V. 4.30%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.6 Analysis of variance for number of days to 50% physiological maturity among ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	12139.135	1348.79	195.76**	0.0000
Year (Y)	1	1.034	1.03	0.15 ^{ns}	0.6917
Location (Loc)	1	513.101	513.10	74.47**	0.0000
G x Loc	9	3329.248	369.92	53.69**	0.0000
G x Y	9	59.948	6.66	0.97 ^{ns}	0.4312
G x Y x Loc	9	91.518	10.17	1.48 ^{ns}	0.1432
Replication in Loc and Year	8	8.201	1.03	0.15 ^{ns}	0.8676
Residual	72	495.8	6.89		
Total	118	16637.98			
Grand mean 110.91	R- squared 97.02%		C.V. 2.37%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.7 Analysis of variance for plant height among ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	6954.026	772.67	6.12**	0.0000
Year (Y)	1	739.753	739.75	5.86*	0.0151
Location (Loc)	1	3406.736	3406.74	26.99**	0.0000
G x Loc	9	763.034	84.78	0.67 ^{ns}	0.6990
G x Y	9	2789.217	309.91	2.45*	0.0116
G x Y x Loc	9	3191.564	354.62	2.81**	0.0045
Replication in Loc and Year	8	597.575	74.70	0.59 ^{ns}	0.2975
Residual	72	9089.59	126.24		
Total	118	27531.5			
Grand mean 46.31	R-squared 66.98%	C.V. 24.26%			

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.8 Analysis of variance for number of pods per plant amongst ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	159536.715	17726.30	28.29**	0.0000
Year (Y)	1	29.782	29.78	0.05 ^{ns}	0.8234
Location (Loc)	1	38647.722	38647.72	61.68**	0.0000
G x Loc	9	18060.51	2006.72	3.20**	0.0016
G x Y	9	4126.284	458.48	0.73 ^{ns}	0.6421
G x Y x Loc	9	173.468	19.27	0.03 ^{ns}	1.0000
Replication in Loc and Year	8	3634.47	454.31	0.73 ^{ns}	0.2018
Residual	72	45112.7	626.57		
Total	118	269321.65			
Grand mean 82.53	R- squared 83.25%		C.V. 3.03%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.9 Analysis of variance for number of seeds per pod among ten soybean genotypes tested over two years and locations with three replications^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	6.132	0.68	0.57 ^{ns}	0.7907
Year (Y)	1	253.064	253.06	212.66 ^{**}	0.0000
Location (Loc)	1	20.616	20.62	17.32 ^{**}	0.0001
G x Loc	9	9.732	1.08	0.91 ^{ns}	0.4821
G x Y	9	4.485	0.50	0.42 ^{ns}	0.9199
G x Y x Loc	9	26.485	2.94	2.47 [*]	0.0112
Replication in Loc and Year	8	3.454	0.43	0.36 ^{ns}	0.5518
Residual	72	85.88	1.19		
Total	118	409.85			
Grand mean 3.47	R- squared 79.04%		C.V. 31.44%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.10 Analysis of variance for dry matter among ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	1698.472	188.719	8.62**	0.0000
Year (Y)	1	453.581	453.581	20.72**	0.0000
Location (Loc)	1	883.953	883.953	40.37**	0.0000
G x Loc	9	859.307	95.479	4.36**	0.0001
G x Y	9	440.347	48.927	2.23*	0.0284
G x Y x Loc	9	178.206	19.801	0.90 ^{ns}	0.5260
Replication in Loc and Year	8	407.957	101.989	4.66**	0.0020
Residual	72	1664.04	21.9		
Total	118	6585.87			
Grand mean 15.68	R- squared 74.73%		C.V. 29.85%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.11 Analysis of variance for hundred seed weight among ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	385.639	42.85	9.10**	0.0000
Year (Y)	1	10.993	10.99	2.33 ^{ns}	0.1206
Location (Loc)	1	272.797	272.80	57.92**	0.0000
G x Loc	9	156.836	17.43	3.70**	0.0004
G x Y	9	12.473	1.39	0.29 ^{ns}	0.9692
G x Y x Loc	9	17.4	1.93	0.41 ^{ns}	0.9130
Replication in Loc and Year	8	31.392	3.92	0.83 ^{ns}	0.1459
Residual	72	339.11	4.71		
Total	118	1226.64			
Grand mean 17.83	R- squared 72.35%	C.V. 12.17%			

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.12 Analysis of variance for seed yield among ten soybean genotypes tested over two years and locations with three replications ^a.

Source	Df	SS	MS	F- value ^b	Pr> F
Genotype (G)	9	71285185.1	790576.1	5.90**	0.0000
Year (Y)	1	3270.7	3270.7	0.00 ^{ns}	0.9608
Location (Loc)	1	189255799.5	189255799.5	141.04**	0.0000
G x Loc	9	48746171.9	5416241.3	4.04**	0.0003
G x Y	9	230444.5	25604.9	0.02 ^{ns}	1.0000
G x Y x Loc	9	71241.8	7915.8	0.01 ^{ns}	1.0000
Replication in Loc and Year	8	11200947.5	2800236.9	2.09 ^{ns}	0.0908
Residual	72	101984282.33	1341898.45		
Total	118	422777343.3			
Grand mean 4177.85	R- squared 75.88%	C.V. 27.45%			

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.13. Analysis of variance for linear regressions of seed yield among ten soybean genotypes^a.

Source	Df	SS	MS	F-value ^b	Pr> F
Total	119	103230948.0			
Genotypes	9	23410279.0	2601142.1	447.24**	0.0000
E + in G x E	30	79820669.0	2660689.0		
E (linear)	1	63247582.8			
G x E (linear)	9	16456766.0	1828529.6	314.40**	0.0000
Pooled deviation	20	116320.2	5816.0		
Residual	80	38060388.0	475754.85		
Grand mean 4161.0	R-squared 99.85%		C.V 28.71%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.14. Analysis of variance for linear regressions of number of active nodule among ten soybean genotypes^a.

Source	Df	SS	MS	F-value ^b	Pr> F
Total	119	53.375			
Genotypes	9	5.625	0.625	1.59 ^{ns}	0.1856
E + in G x E	30	47.750	1.592		
E (linear)	1	31.675			
G x E (linear)	9	8.209	0.912	2.32*	0.0562
Pooled deviation	20	7.866	0.393		
Residual	80	32	0.40		
Grand mean 2.63	R-squared 83.53%		C.V 41.73%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference

Appendix 6.15. Analysis of variance for linear regressions of number of nodule among ten soybean genotypes^a.

Source	Df	SS	MS	F-value ^b	Pr> F
Total	119	85.136			
Genotypes	9	22.543	2.505	22.35**	0.0000
E + in G x E	30	62.592	2.086		
E (linear)	1	47.497			
G x E (linear)	9	12.854	1.428	12.74**	0.0000
Pooled deviation	20	2.242	0.112		
Residual	80	37.34	0.47		
Grand mean	R-squared		C.V		
3.43	96.42%		34.50%		

^a Df=degrees of freedom; SS=sum of squares; MS=mean squares

^b * and ** denote significant differences at P<0.05 and P<0.01 probability levels, respectively; ns=non-significant difference