Predicting and improving seedling emergence of three vegetable crops

By

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LIST OF ABBREVIATIONS

AA	Accelerated ageing
ANOVA	Analysis of variance
AOSA	Association of official seed analysts
CV	Coefficient of variation
DIFF	Difference between the highest and lowest values
DF	Degree of freedom
FEP	Final emergence percentage
FGP	Final germination percentage
FSP	Final survival percentage
GC	Germination capacity
GE	Germination energy
GI	Germination index
ISTA	International seed testing association
LSD	Least significant difference
MGT	Mean germination time
MET	Mean emergence time
PEG	Polyethylene glycol
RH	Relative humidity
RH32	32% relative humidity
RH43	48% relative humidity
RH75	75% relative humidity
RH100	100% relative humidity
SE	Standard error
SGT	Standard germination test
SSAA	Saturated accelerated ageing
SVI	Seedling vigour index
TSE	Time spread of emergence
TSG	Time spread of germination

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ABSTRACT

The predictive values of standard and non-standard germination temperatures, standard accelerated ageing (AA) and saturated salt accelerated ageing (SSAA) tests were investigated. Germination tests were performed at standard and non-standard temperatures of 10°, 20°, 20°/30° and 30°C for cabbage and tomato, and 10°, 15°, 20° and 30°C for onion. The AA and SSAA tests were conducted using four relative humidities: standard AA (RH100), NaCl (RH75), $Ca(NO_3)_2$ (RH43) and $MgCl_2$ (RH32). Emergence trials were also conducted at a range of temperatures (winter, 15°/25° and 30°C) and media (Hygromix and soil) using seedling trays. Correlations were calculated to evaluate the relationship between laboratory and emergence test results. Three onion seed lots that have different vigour levels were primed with PEG or NaCl and were germinated and planted under varying temperatures and levels of salinity.

There were vigour differences among different seed lots of cabbage, onion and tomato seeds used in this study. Using the different laboratory tests, seed lots were distinguished as high, medium and low vigour seed lots. The low temperature germination test (10°C) for cabbage and onion; and 20°C for tomato were found to be effective for separation of seed lots according to their vigour levels. All SSAA vigour tests were also useful in differentiating seed lots based on their physiological stage of deterioration. Using the standard AA (RH100) the seed moisture content was high (29 to 45%), resulting in fungal growth. However, in the SSAA test the moisture content was below 14% and no fungal growth was observed. Ageing of seed lots using NaCl and $MgCl_2$ resulted in a low narrow range of moisture contents, but using $Ca(NO_3)_2$ the range of the moisture content between seed lots was higher.

For all crop seeds, there was no significant difference in the percentage emergence between the two growth media but larger seedlings were observed from the commercial growth medium (Hygromix). For cabbage, highly significant correlations were obtained between germination percentage at 10°C and RH32 and all emergence trials. In onion, the correlations were not consistent; highly significant correlations were observed from germination capacity (GC), 15°C, 20°C, 30°C, RH75 and RH32; and emergence parameters at specific conditions. However, the germination percentage from 30°C correlated significantly with the final emergence percentage and seedling vigour index. Germination of tomato seeds at 20°C and all SSAA had highly significant correlation with all emergence parameters. The germination rate parameters: MGT (mean germination time) and GI (germination

index) were valuable in categorising seed lots with moderate vigour levels, however, except GE in cabbage and onion, all other germination rate characters failed to correlate with any emergence parameters.

The radicle percentage germination (viability or GC) and final emergence percentage (FEP) were not enhanced by priming in all seed lots in the germination and emergence trials at all temperatures. The percentage of normal seedling was increased due to priming when seeds were subjected to low temperature and higher salinity levels. Priming was effective in improving the rate, uniformity of germination/emergence; and increased the seedling dry mass of onion seeds when grown under relatively cold environmental conditions. Priming was more beneficial for low vigour seed lots (seed lot A) than for high vigour seed lots (B, C). Priming also improved germination rate and uniformity at all salinity levels, but faster rate of emergence, more uniform and higher seedling dry mass were only observed when seeds were watered with low salinity levels (25 mM of NaCl). No significant difference was observed using NaCl or PEG as priming agent. Onion seeds are tolerant to salinity during germination and the effect of priming was more beneficial at emergence.

Keywords: Cabbage, onion, tomato, accelerated ageing test, saturated accelerated ageing test, seed vigour, seed priming, germination, emergence, rate, uniformity, percentage, viability, NaCl, PEG.

CHAPTER 1
GENERAL INTRODUCTION

Vegetable production is increasingly important to human beings. At present, vegetables are viewed as low caloric sources of essential vitamins necessary for health. They are particularly important in supplying certain constituents in which other food materials are deficient. Health conscious consumers need a greater variety of vegetable products in their diets (Salunkhe & Kadam, 1998).

Almost all vegetables produced commercially depend primarily on seeds for regeneration. As a result, for crop production seed quality is essential for providing a good stand. Another issue that makes vegetable seed quality so important is their high price. This is due to high quality control maintenance and high production cost. The specialized needs of controlled environments (such as greenhouses) and the use of transplants to assure rapid growth and uniform spacing in the field have also increased the cost of quality vegetable seeds (McDonald, 1998a).

Farmers need high quality seed since high quality seed is the basis of higher agricultural productivity. Furthermore, there is an interest among vegetable growers to predict emergence, due to an increasing need to schedule their crop in precision to meet market demands for continuity of high quality products. To ensure the success of any planting programme a regular and continuous supply of high quality seed is vital. Poorly adapted or low producing seed sources can result in plantation failures or considerable losses in production, because it is difficult to remedy these problems once a crop is established.

High yielding and good quality crops require rapid establishment of uniform plant stands under variable environmental conditions. Several factors besides seed quality can also influence stand establishment. These include soil physical characteristics, temperature, moisture, various cultural practices, weeds and diseases, which may all be limiting factors in establishing maximum stands and achieving the highest possible yields. Measures taken to increase stands include soil improvements, implementing cultural practices such as crop rotation, good seedbed preparation, appropriate seeding date, soil fertility, weed control and use of chemical and biological seed treatments, which can have a significant impact on the establishment of uniform crop stands. Combining seed treatments and

cultural/tillage practices to minimize environmental constraints can lead to maximum stands and yields in the production of high quality vegetable crops.

Germination, purity and health are the three criteria of seed quality and have been associated primarily with seedling emergence. Seed vigour appeared to be a fourth trait of seed quality, which is important in the context of field emergence (McDonald, 1998b). Successful stand establishment requires high quality and genetically pure seed that ensures rapid and uniform seedling emergence. Thus, seed industries are using tests ranging from genetic purity to vigour tests in order to insure the supply of quality seeds (Perry, 1980; McDonald, 1998b; Hampton, 2000).

The field performance of the plants can be measured by many parameters including: emergence, plant growth, vegetative and reproductive development and yield. All these parameters are directly or indirectly influenced by seed quality. Seed vigour can affect yield indirectly by reducing the plant density below the recommended population. Seed vigour can also affect crop yield directly by the rate and uniformity of emergence. Since high vigour seed lots emerge rapidly and more uniformly, they can have an advantage when performing certain cultural practices such as pesticides spraying (TeKrony & Egli, 1991).

Mostly standardized laboratory germination procedures are criticized as not predicting field performance very well (Ferguson, 1995). The standard germination test is conducted under optimum conditions for seed germination. Consequently, when field conditions at planting are near optimum, the results usually correlate well with field emergence. However, under sub-optimal field conditions, standard germination test results, usually overestimate field emergence. The problem with predicting field emergence is to accurately predict the weather at planting. As a result, seed scientists have emphasized the development of another seed quality parameter, namely seed vigour (McDonald, 1995). Non-standard temperatures were found useful for predicting emergence of vegetable seedlings under non-favourable conditions. Non-standard temperatures are also used to identify vigour differences among seed lots with similar standard germination percentages. Seed companies use thermogradient tables with a range of temperatures for determining vegetable seed vigour (McDonald, 1998a).

Vigour testing, if properly done could be one possible solution for predicting emergence. However, the vigour test does not predict performance for a particular set of fluctuations; rather, it predicts the general ability of a seed lot to germinate normally over a range of adverse conditions. Its purpose is to differentiate seed lots, with essentially equal germination, according to their ability to germinate. As seeds age and begin to deteriorate and die, vigour declines before germination test results decline (Hampton, 2000).

For predicting emergence of vegetable seedlings many vigour tests were introduced. The accelerated ageing test (AA) is one of the most acceptable vigour tests and was found to predict the emergence of large seeded agronomic crops. However, it has some limitations when used for small seeded vegetables due to rapid absorption of water during ageing. Jianhua and McDonald (1996) proposed a new method of ageing called the saturated salt accelerated ageing test (SSAA). This test is a modification of the standard AA test where saturated salts are used instead of water. The SSAA resulted in better results for small impatient seeds (Jianhua & McDonald, 1996; McDonald, 1997). The SSAA using different salts that produce low relative humidities may have a potential for better predicting emergence of small seeded vegetables, including cabbage (*Brassica oleracea* var. *capitata* L.), onion (*Allium cepa* L.) and tomato (*Lycopersicon esculentum* L.)

Enhancing field emergence of vegetable crops through the use of transplants, irrigation, mulching, and good soil preparation has been developed and is still developing. For a seed in order to germinate and emerge several problems are encountered. These include problems associated with the seed itself and with the soil environment (Dornbos, 1995b). Seed vigour is closely related to longevity as seeds of low vigour generally have shorter potential longevity than high vigour seeds (Ellis & Roberts, 1981). However, seed enhancement techniques involving hydration can improve vigour (Argerich & Bradford, 1989). Since the rate of germination and subsequent performance are positively correlated, improving germination and emergence improves both viability and vigour of a seed lot (Welbaum *et al.,* 1998a).

Seed enhancement is a post harvest improvement of germination, emergence and stand establishment by treating the seeds with different techniques before sowing. The most promising technique of improving the rate and uniformity of the seedling establishment is seed priming (Taylor *et al*., 1998). Seed priming can improve germination and seedling emergence in a number of

vegetable crops by reducing the time required for the seeds to emerge and produce seedlings. As a result, improved germination and emergence are important safeguard conditions against yield losses in direct-seeded crops and seedling transplants. Priming has practical implications in improving performance of vegetable crops under stressed environmental conditions such as extreme temperatures, salinity, and flooded or reduced water availability (McDonald, 2000). It can also be applied solely or in combination with other seed treatments including growth regulators and fungicides, and sowing techniques such as fluid drilling (Brocklehurst *et al*., 1987). The success of priming conditions depends on environmental conditions such as temperature, aeration and light, duration of treatment, osmotic potential of the solution, seed quality and drying.

Onion seeds are slow to germinate and emerge after sowing (Brewster, 1994). Heydecker and Coolbear (1977) reported that out of 31 common vegetable seeds, onion seeds ranked as the 29th slowest germinating crop. This results in smaller seedlings and plants, which are more vulnerable to soil borne diseases. The extended emergence periods predispose the planting bed to increased soil compaction and deterioration (Ellis, 1989). The slow and uneven germination of onion reduces the benefits of the present day highly mechanized agricultural farming system (Kim, 2000). Therefore, to improve vigour onion seeds were primed with PEG and NaCl, and germnation and emergence trials were conducted under varying temperatures and levels of salinity.

The aims of this experiment were:

- To evaluate the non-standard germination temperatures for their possible use as a vigour test.
- To predict emergence of cabbage, onion and tomato seeds by using the SSAA test.
- To improve vigour of onion seed lots under a varying temperature and salinity conditions.

CHAPTER 2
LITERATURE REVIEW

2.1. Seed vigour

Seed and seedling vigour are two different parameters. Seed vigour is the ability of a seed to germinate and grow rapidly to establish a normal seedling. Good seed vigour means rapid and uniform emergence and development of normal seedlings under a wide range of field conditions. While seedling vigour is the rapid growth of seedlings during cotyledon and early true leaf stages. Seed vigour is defined as an index of the extent of the physiological deterioration and/or mechanical integrity of a high germinating seed lot which governs its ability to perform in a wide range of environments (Hampton, 1995). ISTA (1999) also defined seed vigour as "the sum total of those properties of the seed which determine the level of activity and performance of seed or a seed lot during germination and seedling emergence". According to Ferguson (1995), seed vigour is not a single measurable property but a concept describing the interaction of several characteristics. This includes the rate and uniformity of germination and growth, tolerance of environmental stresses after sowing, and retention of performance capacity after storage.

2.2. Deterioration factors that influence seed vigour

The reduction in the ability of seeds to carry out the physiological functions that allow them to perform is called physiological deterioration (or ageing). This deterioration can begin before harvest and continues during harvest, processing and storage, at a rate greatly influenced by genetic, production and environmental factors (Hampton, 2000). Coolbear (1995) confirmed that the end point of this deterioration is ultimately death of the seed, but initially, the reduced ability of the seed to perform its physiological functions does not prevent germination. Seeds, therefore, lose vigour before they lose the ability to germinate, hence seed lots that have similar germination values can differ in their extent of deterioration, and so differ in seed vigour.

Smith and Berjak (1995) stated that symptoms of seed deterioration included a reduction in germination rate and uniformity, reduced tolerance to environmental stress and lower seedling emergence and growth. Although not all changes that occur during seed deterioration are understood, there exist some speculation on the probable sequence of events (Dornbos, 1995a). The

maximum vigour potential of a seed is attained at physiological maturity. Following physiological maturity, between the times of harvest and planting (storing, bagging and shipping), there is a considerable vigour loss, and deterioration can occur through physical damage, physiological decline and pathological infection (Dornbos 1995a; Copeland & McDonald, 2001).

Physical or mechanical damage to the seed can occur during harvesting, conditioning, transportation, and improper storage. Although the immediate effect of such damage on seed quality is generally not serious, the resulting effects of mechanical damage on seed longevity are problematic and of highly economically significance (Copeland & McDonald, 2001). At physiological maturity, seed generally have high moisture content, which is unsafe for storage. Therefore, seed is typically not harvested until it attains harvest maturity, where the moisture levels are low enough for safe storage, but high enough to minimize mechanical injury. Kulik (1995) reported that seeds that were exposed to severe environmental conditions at the period between physiological maturity and harvest maturity, resulted in poor seed quality.

Seed development includes a series of important sequential stages from fertilization, accumulation of assimilates, seed drying and dormancy. Each of these stages represents a change in morphological and physiological development that can change the seed's potential performance (Hampton, 2000). The initial phase of seed deterioration is seed degradation in which there is a reduction in ATP synthesis, respiration and biosynthesis rates, resulting in reduced emergence and development of abnormal seedlings (Dornbos, 1995a).

Seed-born pathogens can adversely affect germination and early seedling growth, reducing and delaying seedling emergence (Finch-Savage, 1995). Infection of plants and seeds by different plant pathogens (bacterial, fungal or viral) in the field or during storage can reduce vigour. This could directly be due to enzyme degradation and production of toxic substances, or indirectly by limiting the ability of the plant to produce normal seeds (Dornbos, 1995a). During storage, and transport, when humidity and temperature are not controlled, it may cause stress to the seed, speeding up deterioration through the infection of pathogens (Hampton, 2000). High and medium vigour seed lots are better able to withstand these stresses and therefore retain their ability to germinate for longer periods than low vigour seed lots. Loss of germination is dependent on species and storage or transport conditions and lead to low vigour seed lots. Poor storage conditions will always accelerate

vigour loss and controlled storage will reduce the rate of vigour loss, but not prevent it (Halmer & Bewley, 1984).

2.3. Seed vigour tests

Seed vigour tests are primarily used to evaluate the quality of seed lots. Based on these, decisions are made about the seed lots as to where and when to plant it (Ferguson, 1995). A vigour test should possess certain essential characteristics that can make it useful to the seed producer and grower. Copeland and McDonald (2001) have described these characteristics as: "a vigour test should be inexpensive, rapid, uncomplicated, reproducible and correlated with field performance". The last feature, which is "correlation of vigour test results with field performance" is the main reason for failure of vigour tests not to be accepted internationally. Nevertheless, most definitions of seed vigour emphasize the relationship between seed vigour and field performance. Matthews (1981) and Ferguson (1995) suggested that whenever possible, vigour tests should be related to field emergence results. Because, the aim of vigour tests is to act as a guide to the farmer.

Stress tests are well-known, accepted vigour tests for many crop seeds. They accelerate the seeds through the deterioration process. The relationship between germination after deterioration and field emergence, shows that the cause of low seed vigour is primarily due to ageing (Powell & Matthews, 1981). Deterioration due to ageing, can lead to the production of abnormal seedlings and dead seeds (McDonald, 1998b).

The identification of vigorous seed lots may allow growers to obtain acceptable seedling emergence in the field (McDonald, 1998b). Seed laboratory emergence is of practical implications, as the emergence of low vigour seed lots are considerably lower than higher vigour seed lots, particularly at lower temperatures. Slow emergence provides an opportunity for weed species to establish, hence affecting subsequent crop growth. Seed vigour tests are conducted to meet market demands for seed quality control and to assist growers in order to achieve an optimum plant population with precision sowing, achieving synchronous emergence (Dornbos, 1995a). McDonald (1995) emphasized that the ultimate value of any vigour test may be its ability to predict field performance. For that reason, a seed vigour test should provide a more accurate value than the germination test in terms of its potential field performance (Halmer, 2000).

2.3.1. Non-standard temperature as vigour test

The purpose of conducting vigour tests is to provide additional information on the ability of the seed to germinate and produce normal seedlings under variable soil and climactic conditions. Various seed vigour tests can be used to evaluate seed lots. The tests most commonly used to evaluate vigour, are measuring of germination at cooler temperatures (Smith & Varvil, 1984; Van de Venter & Lock, 1990). Many growers are seeding earlier to maximize yield and quality. With early planting, seeds are often planted in cool and wet soils, especially under minimum tillage or direct seeding. Because soil conditions may not be optimal, the seed must have good germination and vigour to establish uniform stands.

The standard germination test is conducted under favourable / ideal conditions and it can be used for predicting emergence under favourable ideal conditions (ISTA, 1999). However, field conditions are not always conducive. Most parts of the world experience either high or low temperatures at least for some months. Many seed companies use thermogradient tables as the preferred method of determining vegetable seed vigour (McDonald, 1998a). Jianhua and McDonald (1996) used a thermogradient table for screening varying quality of seed lots. The rates and total germination percentages, at non-standard temperatures (extreme temperatures), were used to identify differences in seed vigour among seed lots possessing the same standard germination percentages. Strydom and Van de Venter (1998) reported the potential use of non-standard germination (10°, 30° and 35°C) as a vigour test for cabbage seeds. Therefore, germination at non-standard germination conditions might be helpful in predicting emergence.

2.3.2. Accelerated ageing test (AA)

The standard accelerated ageing or accelerated ageing (AA) test was originally developed to estimate the longevity of seed in commercial storage, but later has been successfully related to field emergence and stand establishment in many vegetable species. The AA test is one of the most popular seed vigour tests due to its simplicity, ease of standardization and applicability to a wide range of vegetable crops (McDonald, 1998b). In this test, seeds are exposed for short periods of time to two environmental variables, namely high temperature and high relative humidity (RH) which cause rapid seed deterioration.

The AA conditions, which are favourable for testing a wide range of species and seed lots within a single species, may differ. Therefore, temperature, seed moisture content and duration of the treatment may be different for a particular seed lot (Osman, 1988). Nevertheless, for many species an ageing temperature of 41°C and ~100% RH for 72 hour is used. After this period of stress the germination of the seed lots are determined. This germination results are then compared with the germination results determined before performing the AA test (TeKrony, 1995). The principle of the AA test is that low quality seeds deteriorate more rapidly than high quality seeds.

2.3.3. Saturated accelerated ageing test (SSAA)

The primary use of the AA test has been limited to large seeded agronomic and vegetable crops. The test has been less studied for small seeded vegetables and when studies were conducted, correlations with seed quality were poor (Probert & Hay, 2000). This has been attributed to a large variation in initial seed moisture content as well as the ability of the small seeded vegetables to absorb moisture rapidly and achieve maximum moisture content after only one day.

Due to the problems experienced with the AA tests, Jianhua and McDonald (1996) proposed a new method of ageing, namely the saturated salt accelerated ageing test (SSAA). This test is the modification of the standard AA test by which saturated salts are used instead of water. Saturated NaCl and KCl solutions gave promising results when substituting water during the accelerated ageing of small seeds. The SSAA test help small seeds to absorb water more slowly by reducing the surrounding relative humidity, hence delays deterioration. Moreover, by lowering the relative humidity fungal growth was retard, which is a major cause of vigour loss during the standard AA test where distilled water is used (Jianhua & McDonald, 1996; McDonald, 1998b). McDonald (1997) reported that this test was found to reduce the rate of water absorption of small seeds and it was reproducible among laboratories applying vigour tests, which is one of the aims of a vigour test.

2.4. Predicting emergence of vegetable seed using vigour tests

The germination percentage of seeds is obtained from a standard germination test (SGT), which is indicative of the ability of seeds to produce a normal seedling under favourable conditions (ISTA, 1999). This test, commonly used to evaluate seed quality, is able to predict field emergence provided the conditions for emergence are favourable. Many authors found laboratory germination

tests to correlate well with field emergence of various vegetables under conducive environment. However, the SGT fails to give accurate information concerning a seed lot's field performance potential under unfavourable environmental conditions (Ferguson, 1995). This is due to: firstly, the lack of distinction between strong and weak seedlings. Those seeds that are considered as terminable might vary from weak to strong. This results in failure to categorize the quality since the SGT does not account for the progressive nature of seed deterioration, which has a major impact on stand establishment. Secondly, the SGT is conducted in artificially, standardized, environmentally controlled conditions, which rarely relate to field conditions (Copeland & McDonald, 2001).

In sowing to a stand the assumption is that all or at least a predictable proportion of the seeds sown will produce plants. The potential field emergence can accurately be predicted, thus simplifying the selection of good quality seed using various laboratory tests. A vigour test is one of the tests that can be used to predict field emergence of vegetable crops under wide range of environmental conditions (McDonald, 1998b). Seed vigour is different from seed viability; the later is a yes or no situation for germination, while seed vigour is an indication of growing ability of viable seed under sub-optimal conditions (Finch-Savage, 1995). The vigour test result indicates the seedling's probable performance in the field. Thus it is a more reliable and practical index for predicting stand establishment.

A number of laboratory tests are correlated with field emergence and various researchers used these as indicators of field emergence. Although all cultivars showed high germination percentages in the laboratory, there were large differences between them in terms of seedling emergence in the field, indicating that they differed in vigour. In general, poorer quality seeds will show symptoms typical of seed ageing such as low viability, reduced germination and emergence rates, poor resistance to sub optimum conditions and low seedling growth rate (Finch-Savage, 1995). However, in stressed environments, vigour tests can predict seedling emergence better as compared to SGT.

The effects of seed vigour on emergence and stand are especially critical in cabbage, where delayed emergence or missing plants may reduce yield and uniformity at harvest (Powell *et al.,* 1991; TeKrony & Egli, 1991). Unfavourable environments such as relatively low or high soil temperatures may also hinder seedling establishment of *Brassica* spp. This has resulted in emergence problems with cabbage, even when the seed lots exhibited adequate germination percentages in the standard

germination test (Wilson *et al*., 1992). Hampton (2000) conducted an experiment on *Brassica* spp. and the germination after the AA test of high vigour seed lots was similar to the pre-AA test germination. Whereas for low vigour seed lots, the post-AA test germination was considerably less than the pre-AA test germination. Considering these results, the SGT still remains the best predictor of field emergence of cabbage.

High vigour seed is crucial for onion, where the percentage and uniformity of emergence are the most important parameters for onion production. Predicting the field emergence of onion could increase quality of the bulb, since medium sized bulbs have high market value (Steenge, 2001). Kraak *et al*. (1984) subjected onion seeds to 60°C and 55% RH for 1.5 or 5.5 hours in the AA ageing test and compared the standard germination and field emergence over two years. He found that the AA test results were not correlated with field emergence of onion. In another experiment using the same ageing conditions, Bekendam *et al*. (1987) confirmed that there was no correlation between the results of germination after ageing and the mean emergence of individual seed lots before ageing. The final stage of ageing is a loss in viability and is preceded by a decline in vigour (Matthews & Powell, 1986). Even though variations in emergence occurred between sowings, no systematic behaviour exists between the seed lots to indicate any prospects for successful application of a vigour test. Consequently, emergence was highly correlated with laboratory germination percentage (Kraak *et al*., 1984).

2.5. Improving vigour of onion seeds though priming

Heydecker and Coolbear (1977) reported that out of 31 common vegetable seeds, onion seeds ranked as the 29th slowest germinating crop. This results in smaller seedlings and plants, which are more susceptible to biotic and a-botic factors (Ellis, 1989). Onion seeds are short-lived and most seed lots show reduced germination percentage in one year, since the vigour of seed lots deteriorates faster than germination percentage. High germinating seed lots may emerge slower and with reduced percentage particularly when they are planted under adverse conditions. Therefore, there is a need to improve vigour of onion seeds in order to repair reduced vigour.

Onions originated in central Asia and was domesticated 5000 years B.C. It ranks third after potato and tomato among total vegetable crop production area in the world. Onions are grown for fresh shoots green “salad” and bulbs and the aim of planting depends on the size of the bulb and stage of

consumption (Brewster, 1994). Onions are cool season crops that can withstand moderate freezing and may grow either by direct seeding in the field, by means of transplants or by using bulb sets. Onions can be grown under a wide range of day lengths ranging from short to long, in any country with day length greater than 12 hours photoperiod. Onion production is restricted in various countries by climate, labour and seed quality (Steenge, 2001).

The period between planting the seeds and the emergence of seedlings is critical for stand establishment of crops. Poor field stand establishment in commercial onion production has led to increased seeding rates and resulted in slower emergence of seedlings (Pill, 1995). For onion growers a simple method of seed priming for maintaining seed vigour was found to be highly beneficial (Parera & Cantliffe, 1994). After priming the rate, synchrony, and percentage of germination/emergence of seedlings were found to be higher for primed seeds as compared to the control. It can improve vigour especially under adverse conditions such as low/high temperatures, reduced water availability and salinity. The main obstacle for commercialising seed priming is that the response of species, cultivar, and even seed lots differs, therefore every seed lot needs specific priming conditions and one usually discovers this by trial and error, which makes priming tedious and unpractical (McDonald, 1998a; 2000).

2.5.1. Germination and priming

Germination can be defined as those events that begin with water uptake by the seed and end with the enlargement of the embryo and radicle protrusion from the seed coat. Generally, in the process of germination any seed that has no problem with dormancy and seed coat permeability shows a three-phase process of water uptake (Figure 1). When a dry seed is imbibed in water (0 PMa), the solutes inside the cell reduce the turgor potential and lead to rapid uptake of water from the surrounding solution (phase I), and as the water potential of the seed increases during imbibition the water content of the seed increases. The resistance of the cell wall to expansion, however, results in a turgor potential increase. When the seed water potential increases the water uptake decreases and the seed enters phase II (lag phase), in which only a small or negligible amount of water is absorbed by the seed over a relatively longer period of time. This allows the seed to complete all its physiological pre-germinative processes and make the seed ready for radicle emergence. Finally,

when the seed completed all pre-germinative embryonic processes in phase II, it transfers to phase III, which results in radicle protrusion from the seed coat (Bradford, 1986; Welbaum *et al*., 1998a).

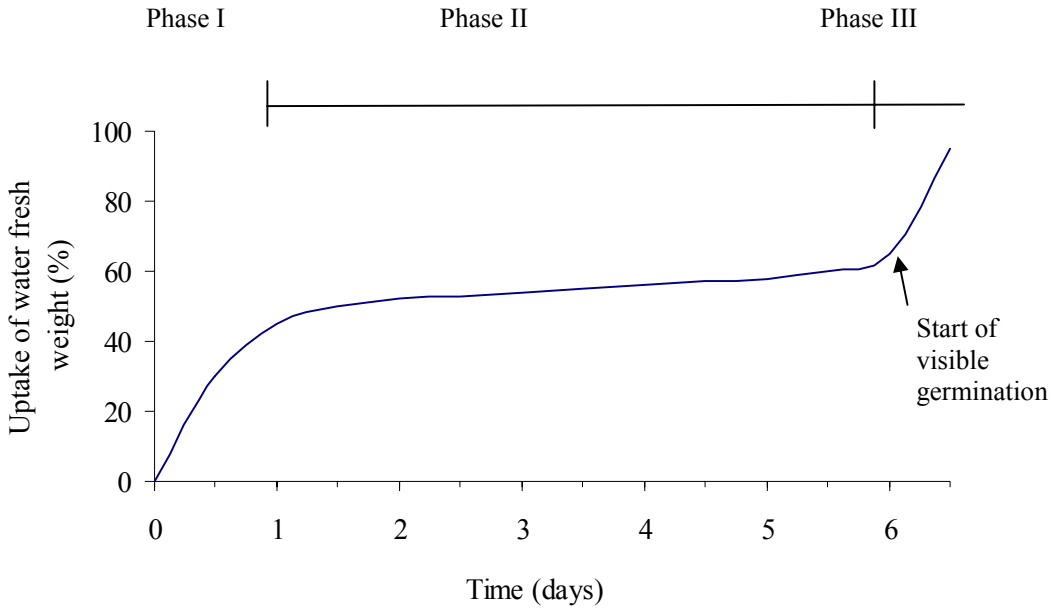

Figure 1: Triphasic water uptake pattern of germinating seeds (Copeland & McDonald, 2001)

Heydecker and Coolbear (1977) and Bradford (1986) defined priming as a technique of controlling hydration of seeds to a level that allows pre-germinative metabolic activities to carry on but that prevents radicle protrusion from the seed coat. Seed priming can improve seedling emergence in a number of vegetable crops by reducing the time required for the seeds to emerge and produce seedling transplants in a shorter period of time. Priming can be applied solely or in combination with other seed treatments including growth regulators and fungicides, and sowing techniques, such as fluid drilling (Brocklehurst *et al.*, 1987).

2.5.2. Factors affecting priming

Priming conditions are the same as the conditions that are favourable for germination, except during priming the last step of germination is prevented by using a solution with a low water potential (Akers & Holley, 1986). The main factors that affect the success of priming are temperature, aeration, osmotic potential of the solution, duration of treatment, seed quality and drying.

a) Temperature

The temperature required for priming is similar to that of germination and different seeds have different optimal temperatures within which they germinate. The optimal temperature is the point at which the maximum germination percentage is attained in the shortest period of time. However, a temperature lower or higher than the optimal delays germination due to a longer lag phase (Figure 1) (Bradford, 1995; Copeland & McDonald, 2001). The improvement benefits of priming onion seeds are greatly dependent on temperature.

Priming does not extend the predetermined temperature range, but allows a greater portion of the seed to germinate at temperatures which were previously inhibitory. Priming onion seeds at 15°C was found to improve rate and uniformity of emergence without significantly reducing the percentage of emergence (Ali *et al.*, 1990; Brewster *et al*., 1991; Gray *et al.*, 1991; Drew *et al*, 1997). For primed onion seeds, the optimal temperature extended from 10° to 18°C. Within this range all seeds had a higher germination percentage and geminated in a shorter period of time (Ali *et al.*, 1990). Seeds, which were primed at 10°C, germinated quicker than the seeds primed at 24°C. Similar trend was reported by Haigh *et al*. (1986) after onion seeds were primed with KNO_3 + K_3PO_4 at 15°, 20° and 25°C in -1.6 MPa, where seeds emerged faster at 15°C in comparison to higher temperatures. This implies that priming is more effective at relatively lower temperatures than at higher temperatures.

b) Oxygen availability

Priming, similar to germination is a process related to living cells and requires spending of energy. The energy requirement of the living cells to initiate, process or mobilize the resources in the seed to make the seed ready for radicle emergence is dependent on oxygen. Most seeds germinate in an atmosphere containing about 20% of oxygen, but in priming, seeds can even germinate at lower oxygen levels (Copeland & McDonald, 2001). Priming reduces the seed's sensitivity to lower oxygen levels.

Onion seeds primed with PEG-6000 reduced germination and this reduction may be due to insufficient oxygen in the solution. The low oxygen level may encourage anaerobic respiration in some seeds, leading to toxic levels of ethanol (Furutani *et al.*, 1986). Aerated solutions of PEG or

salts give more benefits in comparison to using moist surface media and this may help seed companies in commercially priming large quantities of seeds (Akers & Holley, 1986; Bujalski *et al.*, 1989).

c) Osmotic potential

The duration of the three consecutive phases during the germination process are dependent on the amount of water as well as the species, which are the major event in the success of priming (Bray, 1995). Priming prevents the seeds from entering phase III by extending the time of the lag phase at which most of the physiological and pre-germinative metabolic processes occur and make the seed ready to allow radicle protrusion from the seed coat (Bradford, 1995; Bray, 1995).

The osmotic potential of dry seeds is very low usually between -350 to –50 MPa, and when dry seeds are soaked in pure water with an osmotic potential of zero (0 MPa) or any priming solutions (>-2.0 MPa), a large amount of water will be absorbed by the seeds until equilibrium is reached. The rate of imbibition is dependent on the seed coat permeability and seed contact area (Bradford, 1995). No difference in the pattern of water uptake was observed whether seeds were treated with water or priming solutions using osmotic potential of 0 (water), -0.5, -1.0, -1.5, -2.0, -3.0 and -4.0 MPa. Finch-Savage and Phelps (1993) reported that the period between phase II and III is the time most sensitive to moisture content. The failure of entering phase III due to a lack of moisture is the limiting step, which determines the osmotic potential for radicle emergence.

Gray *et al.* (1990) confirmed that onion seeds that were treated within a range of -1.0 to -2.0 MPa showed lower mean germination time than the untreated seeds. According to Haigh and Balow (1987), onion seeds that were primed in PEG at an osmotic potential -1.75 MPa were found to prevent germination of seeds during the priming treatment. Priming the seeds in higher negative osmotic potential (-3.0 and -4.0 MPa) did not affect the mean germination time in PEG. As the time of imbibition extended the mean germination time of primed seeds increased as compared to non-primed seeds (Gray *et al.*, 1990).

d) Duration of treatment

The duration of priming process depends on the priming agent, osmotic potential and temperature for a particular seed lot. If seeds germinated during the priming process then the seeds will lose their viability when dried. This could be due to irreversible embryo damage (Parera & Cantliffe, 1994; McDonald, 2000). The lag phase of the germination process takes 7 to 14 days, which allow the initial or complete pre-germinative metabolic events but is not sufficient for radicle emergence (Copeland & McDonald, 2001).

Priming seeds for 14 days resulted in reduced mean and spread of germination time for both low and high vigour seed lots (Gray *et al.*, 1991; Khan, 1992). Up to four weeks duration of treatment the beneficial advantage of priming increases with increase in time, although the rate is small between 14 and 28 days (Gray *et al.*, 1990). Ali *et al.* (1990) reported that when seeds were primed at -1.49 MPa no germination occurred in the first week, but as the time of soaking extended from 1 to 4 weeks the number of seeds, which germinated during priming increased. Furutani *et al.* (1986) used ten days of priming in order to avoid over priming of seeds and to reduce or inhibit the number of seeds that germinated during priming.

e) Seed quality

Germination of all seeds declines over time. So the germination rate listed on a seed bag is only valid for the year in which it was purchased. Just how quickly germination decreases after the first year depends on the type of seed and how it was stored. Field performance of primed seeds could be related to seed quality difference among seed lots. There was a clear effect of seed quality on the percentage and rate of emergence (Wheeler & Ellis, 1994). The response of seed priming differs from seed lot to seed lot and from cultivar to cultivar. The rate of pre or post seedling emergence and growth of onion is mainly affected by the genetic character or vigour of the seed lot but not by the environmental growth factors (Wheeler & Ellis, 1991). According to Gabriel *et al.* (1997) high seed quality is expressed as percentage of seeds germinated, rate of germination and seed vigour of a seed lot. These parameters are directly or indirectly related to the success or failure of an individual seed lot's stand performance and its further production.

f) Drying

In most experiments with priming, seeds are transferred immediately from the priming osmotica or chemical to the germination media and germination was improved as compared to non-treated seeds. When seeds are subjected to germination without drying, the seeds have already ended the phases II (lag) and I (hydration), hence the seeds only need a rapid water uptake to enter phase III for radicle emergence. Primed and dried-back seeds subjected to germination, need partially to repeat phases I and II of the germination process before radicle appearance can occur (Bradford, 1986; Pill, 1995; Welbaum *et al.*, 1998b). Through priming initial physiological and biochemical events are stimulated, even primed and dried-back seeds are in a more advanced stage of the germination process than the untreated seeds (McDonald, 2000).

Seeds that were dried after priming were found to have fewer normal seedlings and higher mean germination time than non-dried seeds (Gray *et al.*, 1991). Primed and dried-back seeds germinate and emerge slower compared to non-dried and primed seeds. However, all primed seeds showed faster germination and emergence than untreated seeds. In the same experiment primed and stored seeds showed negative effect on seedling growth, even if no influence on percentage of emergence was observed. Normal seedlings emerged in the beginning but with time and successive growth poor stand establishment with short and stunted roots were observed (Gray *et al.*, 1990). Generally, better improvement in germination was showed in unstored primed seeds as compared to stored seeds (Drew *et al.*, 1997). Primed onion seeds can be dried-back to their original moisture content, even though the germination and emergence are delayed as compared to priming and surface drying (Bujalski & Nienow, 1991).

CHAPTER 3
GERMINATION TRIAL

3.1. Introduction

Seed testing is the cornerstone of all other seed technologies. It is the means, by which seed viability and all other physical factors that regulate the use and maintenance of seeds, are measured. Everything that is done to the seeds should have a track record to ensure high quality seed. Seed tests, which forms part of the track record, can tell if the seeds of a crop are worth collecting, if handling procedures were correct and give an indication of the potential number of seedlings available for regeneration (Copeland & McDonald, 2001).

Germination tests are seed tests, which indicate the potential of a seed to produce a harvestable plant in the field. It is performed under controlled laboratory conditions by using specialized germinators and growth substrates, resulting in the best possible seed performance. Germination testing is designed to estimate the maximum number of seeds that will produce a normal seedling and give results that are as repeatable as possible. These results can be used to compare the quality of different seed lots and also estimate the field planting value thereof. When field conditions are near optimum, the standard germination test may accurately predict seed emergence and seedling establishment in the field. However, when environmental conditions are not favourable, additional tests, like a seed vigour test, are needed to provide accurate information on potential crop establishment.

Standard germination tests are conducted at a standardised temperature and will differ between crops. Seeds can, however, germinate at a wide range of temperatures, but the germination percentage is drastically reduced at extreme temperatures. The range of temperatures at which the maximum germination percentage occurs, differs among crops as well as with seed quality. In general, the temperature range becomes narrower as a seed lot deteriorates (Ellis & Roberts, 1981). Germination of seeds under non-standard temperatures can thus be used as a vigour test for differentiating low and high vigour seed lots, where low vigour seeds have a narrower temperature range than high vigour seed lots.

The aim of this trial was to evaluate the use of non-standard temperatures during germination to test its capability to predict the emergence of cabbage, onion and tomato planted under wide range of environmental conditions.

3.2. Materials and Methods

3.2.1. Seed source

For this experiment three crops, namely cabbage, onion and tomato were used. A total of 21 seed lots, six seed lots of cabbage, seven seed lots of onion, and eight seed lots of tomato were obtained from Hygrotech Seed Company. Perry (1984) recommended the use of seed lots with similar germination capacities (viability). Since the correlation coefficient is dependent on the degree of freedom (n-2), he suggested the use of at least six seed lots for better correlation of the results. The seed lots represent different cultivars and all seed lots have commercially acceptable germination percentages. The seed lots are represented by alphabetical letters (A to H) and details of the seed lots are presented in Table 1. All the seed lots were stored in sealed containers in a cold room at 6-8°C and the required amount of seeds was taken from the store just prior to the start of the experiment to be conducted.

Table 1: Cultivars, allocated seed lot names and standard germination percentages of cabbage, onion and tomato seeds used in the germination trial

Cabbage			Onion			Tomato		
Cultivar	Seed	Germination[1] %	Cultivar	Seed	Germination %	Cultivar	Seed	Germination %
Copenhagen Marke	A	88	Red Creole	A	91	Rodade	A	91
Copenhagen Marke	B	91	Red Creole	B	95	Rodade	B	89
Kaapse spits	C	84	Red Creole	C	94	Rodade	C	97
Kaapse spits	D	87	Gold Rush	D	85	Rodade	D	85
Savoy Express	E	89	Gold Rush	E	86	Hytec	E	88
Savoy Express	F	96	Primavera	F	89	NUN 7488	F	90
			Primavera	G	90	BHN 4	G	99
						Top Roma	H	76

[1]The germination percentages are the percentages as determined by Hygrotech with SGT.

3.2.2. Germination at standard and non-standard temperatures

The standard temperature for germination is different for each crop. The three crops were incubated at various standard temperatures according to the rules of the International Seed Testing Association (ISTA, 1999). A standard 20°C was included for tomato, because some South African Seed Companies used this temperature as a standard temperature for some local cultivars of tomato. This resulted in standard temperatures of 20° and 20°/30°C for cabbage and tomato, and 15° and 20°C for onion. Together with the standard germination temperatures, two temperatures (10° and 30°C) were used for all the crops. For the constant temperature regimes (10°, 15°, 20°and 30°C), seeds were incubated in the presence of constant light. For the alternating temperature regime of 20°/30°C, the higher temperature coincides with an 8-hour light period and the lower temperature coincides with 16-hour dark period. This was done in accordance with the ISTA rules.

Germination tests should be conducted in cabinets or rooms that meet the exact requirements of temperature and light, in order to make accurate and repeatable estimates (Copeland & McDonald, 2001). To comply with this, germination tests were performed at the Department of Botany, where Labcon germination cabinets were used. Germination was determined by placing fifty seeds, per treatment combination, on a double layer of filter paper (Whateman #1) in a plastic Petri dish (90mm in diameter), moistened with 5ml of distilled water while extra water was added when needed.

Germination for the non-standard temperatures was defined as emerged, normal seedlings. Final counts for normal seedlings were recorded according to ISTA rules (1999) at 10, 12 and 14 days after incubation for cabbage, onion and tomato respectively. The final germination percentage was calculated.

For germination at standard temperatures, germinated seedlings were counted daily and germination was defined as visible radicle protrusion through the seed coat. Seeds that did not germinate within the range of days specified by ISTA for each crop were considered as non-viable. From the daily counts (visible radicle) the following variables were calculated.

- Germination capacity (GC) or Viability = the ratio between the number of seeds with visible radicle protrusion and the total number of seeds used, times 100. (1)

- Germination energy (GE) = the ratio between total number of seedlings germinated at the 3rd day and the total number of seeds used, times 100. (2)

Mean germination time (MGT) and germination index (GI) were calculated according to Ellis and Roberts (1980) where the expressions were:

- Mean germination time $(MGT) = \frac{\sum (n\,d)}{\sum n}$ (3)

- Germination index (GI) = $\sum \left[\frac{n_1}{d_1} + \frac{n_2}{d_2} + \ldots + \frac{n_n}{d_n} \right]$ (4)

Where n = number of seeds germinated on day (d);

d = serial number of the day

∑n = total number of germinated seeds

3.2.3. Statistical analysis

The Petri dishes were placed in a completely randomised design with four replications. Data were subjected to analysis of variance and means were compared using LSD_{Tukey} (SAS, 1999). Simple correlation coefficients were performed on treatment means to determine the relationship between the emergence trial and germination test results (Chapter 5). ANOVA tables are included in the appendix and are indicated in the text by the letter A before the table number.

3.3. Results and Discussion

3.3.1. Cabbage

The cabbage seed lots (A-F) were ranked based on final germination percentage at four incubating temperatures (10°, 20°, 20°/30° and 30°C); while germination capacity (GC), germination energy (GE), mean germination time (MGT) and germination index (GI) were calculated from the radicle protrusion records at the standard temperature of 20°C (Table 2). In addition, the mean (average of all values), CV (coefficients of variation), LSD_{Tukey} at α=0.05 and DIFF (range between the highest and lowest values) are also available. Similarly, these features are presented in Tables 3 and 4 for onion and tomato respectively.

Table 2: Ranking of cabbage seed lots, based on the results of laboratory germination tests

Seed lot	Germination (%)				GC	GE	MGT	GI
	10°C	20°C[3]	20°/30°C[3]	30°C	%	%	(Days)	
A	79.5a[1](5)[2]	90.5ab(3)	89.5ab(4)	90.0ab(4)	93.0a (3)	88.5ab(4)	2.10b (4)	25.05c (4)
B	91.0a (2)	92.5ab(2)	91.0ab(2)	92.0a (2)	95.0a (2)	93.5a (2)	1.73a (2)	31.17ab(2)
C	56.0b (6)	82.0b (6)	78.0c (6)	81.0b (6)	86.0a (6)	65.0c (6)	3.11c (6)	15.59d (6)
D	82.0a (4)	87.5ab(5)	84.0bc(5)	87.0ab(5)	91.0a (5)	79.0b (5)	2.84c (5)	17.59d (5)
E	85.0a (3)	89.0ab(4)	90.5ab(3)	91.5a (3)	92.5a (4)	91.5ab(3)	1.88ab (3)	27.49bc(3)
F	96.0a (1)	96.0a (1)	94.5a (1)	96.0a (1)	97.0a (1)	97.0a (1)	1.72a (1)	32.61a (1)
Mean	81.66	89.50	87.92	89.58	92.33	85.75	2.23	24.91
CV	12.35	6.14	4.59	4.96	5.74	7.15	5.60	8.52
F value	7.73**	2.82*	8.64*	5.32**	1.90^{NS}	14.96**	92.74**	43.78**
LSD_{Tukey}	22.68	12.35	9.07	9.98	11.92	13.78	0.28	4.77
DIFF	40.0	14.0	16.5	15.0	11.0	32.0	1.41	17.07

[1]Values in each column followed by the same letters were not significantly different. [2]Values in parenthesis are the ranking order of the seed lots. [3] Standard germination temperatures.[NS] Not significant, *significant at α=0.05, and **significant at α=0.01. GC (germination capacity), GE (germination energy), MGT (mean germination time) and GI (germination index).

There were significant differences between the cabbage seed lot means for all the parameters except for germination capacity (Table A1). The results of the standard germination test (20°C) indicated that all cabbage seed lots used in this study had commercially acceptable seed quality with germination percentages above 80%. The germination percentage of the seed lots was significantly different with seed lot C (lowest germination %) being significantly lower than that of seed lot F (highest germination %). There was, however, no significant difference among intermediate seed lots.

There was relatively small differences between the mean germination percentages at 20°, 20°/30° and 30°C, with a difference of less than 2%. However, when the mean germination percentage of these temperatures was compared to that of 10°C, the differences ranged from 6 to 8%. When the mean GC was compared with the mean germination percentages of 20°, 20°/30° and 30°C the difference was low (3 to 5%), but when compared to germination percentage at 10°C, the difference was higher (11%). The difference between germination capacity and germination percentage at different temperatures was mostly due to abnormal seedling counts. The abnormal seedling counts were higher at non-standard stress temperatures, and the number increased substantially as the

temperature reduced to 10°C. Seed deterioration starts with reduction in germination followed by increasing number of abnormal seedlings and finally death of the seed (Priestley, 1986).

Seed lots A, D and E were the only seed lots not consistent in their ranking order. Seed lots A and E were interchanging between one another, ranking as third and fourth for all the parameters except germination at 10°C where seed lot D ranked as number four. Seed lots B and F had a consistent ranking order as being first and second respectively and can be grouped as high vigour seed lots, while seed lot C ranked as number six being a low vigour seed lot. The remaining seed lots (A, D, E) had moderate parameter values and can be grouped as medium vigour seed lots. High vigour seed lots (B, F) did not differ much in terms of germination capacity and germination percentage at various temperatures, while the same was not true for the low vigour seed lot (C). For example, there was no difference between germination at 10°C and 20°C for seed lot F, while there was a 26% difference for seed lot C. Also, when comparing the results from the 10°C and that of the GC there was only a 1% difference for seed lot F, but a 30% difference for seed lot C.

The inconsistent ranking order of seed lots with intermediate vigour is an indication of vigour differences among seed lots. For intermediate (A, D and E) and low (C) vigour seed lots the mean average germination percentage was lowest at 10°C as compared to other temperatures. However, there was no large difference among the other germinating temperatures (Table 2). This implies that 10°C could be a good vigour test for determining the quality of a seed lot.

Ellis and Roberts (1980) suggested the use of germination rate as a second aspect of vigour in seed quality tests. The concept is that low vigour (deteriorated) seeds take a longer time to germinate as compared to high vigour seed lots. Coolbear (1995) confirmed that the slower germination from low vigour (partially deteriorated) seeds could be due to the additional time needed to undertake self repair. In agreement with this, the low vigour seed lot (C) had the longest MGT, while the high vigour seed lots (B and F) had the shortest MGT (Table 2). The germination energy and germination index followed the same pattern, namely higher GE and GI for the high vigour seed lots (B and F) in comparison to the low vigour seed lot (C) (Table 2). More importantly, however, the MGT as well as GE and GI results can be helpful in further separating the intermediate vigour seed lots. With the aid of these parameters, seed lots A, D and E can be ranked as numbers three, four and five respectively (Table 2). The germination percentage (20°, 20°/30° and 30°C), GC and GE of seed lot

D were consistently lower than that of seed lots A, B, E and F while the MGT and GI of seed lot D was very close to that of the lowest ranking seed lot (C). Therefore, the seed lots can be grouped as B and F having high vigour, A and E having medium vigour and C and D having low vigour.

3.3.2. Onion

There were highly significant difference among the seed lot mean values in all of the parameters used (Table A2). The lowest mean percentage germination, and the highest variation in germination percentage were observed at 10°C (Table 3). This can be due to a slower metabolism rate and lower temperatures. Copeland and McDonald (2001) also stated that low temperatures could cause a reduction in the denaturation rate of proteins, which are essential for germination. The highest mean germination percentage was recorded at 20°C, followed by 15°C and 30°C (Table 3). At 20°C only a few abnormal seedlings were found, causing the GC to be only 3% higher than the germination percentage at 20°C (Table 3).

Table 3: Ranking of onion seed lots, based on the results of laboratory germination tests

Seed lot	Germination (%)				GC	GE	MGT	GI
	10°C	15°C[3]	20°C[3]	30°C	%	%	(Days)	
A	80.0ab[1](4)[2]	86.0bc(4)	90.5abc(4)	87.0ab(4)	93.0abc(4)	71b (4)	4.29b(5)	10.06c(4)
B	83.0a (1)	92.5ab(2)	94.5a (1)	91.0a (1)	96.5ab (2)	91a (1)	2.56c (1)	12.77a(1)
C	81.0a (3)	94.5a (1)	93.5a b (2)	89.0a (2)	97.5a (1)	82ab(3)	3.04c (3)	11.06b(2)
D	71.5bc(6)	82.0c (6)	84.5d (7)	86.0ab(5)	88.5c (7)	52c (6)	4.49b(6)	9.38c (6)
E	78.0ab(5)	84.0c (5)	86.0cd (6)	82.5bc(6)	89.5c (6)	54c (5)	4.24b(4)	9.46c (5)
F	63.0c (7)	81.0c (7)	88.5bcd(5)	77.5c (7)	91.5bc (5)	22d (7)	6.63a (7)	7.16d (7)
G	82.5a (2)	87.0bc(3)	91.5abc(3)	89.0a (3)	95.0ab (3)	84ab(2)	2.95c (2)	11.03b(3)
Mean	77.0	86.71	89.85	86.0	93.14	65.14	4.03	10.13
CV	5.21	3.47	2.68	2.37	2.55	9.98	11.16	3.69
F value	13.19**	11.56**	9.57**	15.25**	8.47**	55.9**	37.49**	88.00**
LSD_{Tukey}	9.22	6.91	5.54	5.45	5.45	14.95	1.03	0.86
DIFF	20.0	13.5	10.0	13.5	9.0	69.0	4.06	5.61

[1]Values in each column followed by the same letters were not significantly different. [2]Values in parenthesis are the ranking order of the seed lots. [3]Standard germination temperatures. **Significant at α=0.01. GC (germination capacity), GE (germination energy), MGT (mean germination time) and GI (germination index).

As for the cabbage seed lots, the onion seed lots also differed in terms of vigour, despite commercially acceptable germination percentages at standard germination temperature (20°C). According to the germination percentages at the different temperatures and GC, seed lots B, C, and G were ranked as either first/second/third. However, the rest of the seed lots had no consistent ranking. As overall ranking of seed lots based on all parameters in Table 3, seed lots can be grouped as high (B, C and G), medium (A, D and E) and low (F) vigour. This again confirms that most of the parameters could be used as a vigour test to distinguish vigour differences between seed lots, thus providing a more sensitive index of seed quality than germination test alone. This has been stated by Hampton (1995) as being one of the objectives of vigour testing.

3.3.3. Tomato

None of the tomato seeds incubated at 10°C had normal seedlings. The results are in agreement with those obtained by Özbingöl *et al*. (1998) where the base temperature for tomato germination used was 9°C, and no normal seedlings were recorded within 30 days. Therefore, only the data from three (20°, 20°/30° and 30°C) germinating temperatures will be presented. There were significant differences between the seed lots for all the parameters as shown in Table A3.

Some South African seed companies also use 20°C for the SGT of tomatoes. According to the present results (Table 4), better germination was, however, obtained when 20°/30°C were used as suggested by the ISTA. The SGT (20°/30°C) percentage of seeds germinated and with normal seedlings according to ISTA rules varied from 76% (seed lot H) to 98.5% (seed lot G). The difference between the final normal germination percentage (20°/30°C) and emerged radicle percentage (GC) was highest for low vigour lots than medium and high vigour lots. For seed lot G there was only 1% difference between germination percentage and viability (GC). However, for lower vigour seed lots (example seed lot H) the GC was 86.5% and germination was 76% and there was 10.5% difference (Table 4). The differences get bigger when seeds were germinated at non-favourable temperatures i.e. for seed lot H the difference increased to 18.5% when viability (GC) and germination at 20°C were compared.

Based on the ranking orders in Table 4, the seed lots can be grouped into three categories namely: high (C, G), moderate (A, B, D, F) and low (E, H) vigour seed lots. For all the parameters, seed lots C and G ranked either as first or second and seed lot H was always last. However, there was some

inconsistency in the intermediate (A, B, D, F) vigour seed lots. According to AOSA (1983), most vigour tests have no difficulty in differentiating low and high vigour seed lots. The problem lies on distinguishing of seed lots with moderate vigour levels.

Table 4: Ranking of tomato seed lots, based on the results of laboratory germination tests

Seed lot	Germination (%)			GC	GE	MGT	GI
	20°C	20°/30°C[3]	30°C	%	%	(Days)	
A	88.0ab[1](4)[2]	90.5bc (3)	86.5b(5)	93.0abcd(4)	77.5c (5)	2.37a(3)	18.92b (4)
B	82.0b (6)	86.0c (7)	86.0b(6)	94.0abc (3)	83.5bc(3)	2.73b (5)	18.45b (5)
C	93.0ab (2)	96.5ab (2)	97.0a(2)	97.0ab (2)	95.5a (1)	2.21a (1)	22.60a (2)
D	83.0b (5)	90.0c (4)	88.0b(3)	88.0cd (7)	82.5c (4)	2.39a (4)	19.58b (3)
E	80.0bc (7)	88.5c (6)	84.5b(7)	89.0cd (6)	39.5e (7)	4.75c (7)	10.16cd (7)
F	88.0ab (3)	88.5c (5)	87.5b(4)	91.0bcd (5)	57.0d (6)	4.61c (6)	11.30c (6)
G	99.5a (1)	98.5a (1)	98.5a(1)	99.0a (1)	94.5ab(2)	2.22a (2)	23.15a (1)
H	68.0c (8)	76.0d (8)	73.0c(8)	86.5d (8)	11.0f (8)	5.66d (8)	8.83d (8)
Mean	85.19	89.25	87.63	92.25	67.56	3.37	16.57
CV	6.86	2.93	2.83	3.36	7.12	3.48	6.23
F value	10.36**	27.47**	40.20**	7.87**	152.48**	517.57**	125.41**
LSD_{Tukey}	13.68	6.12	5.68	7.25	11.26	0.27	2.42
DIFF	31.5	22.5	25.5	12.5	84.5	3.45	14.77

[1]Values in each column followed by the same letters were not significantly different. [2]Values in parenthesis are the ranking order of the seed lots. [3]Standard germination temperature. **Significant at α=0.01. GC (germination capacity), GE (germination energy), MGT (mean germination time) and GI (germination index).

When the parameters of germination rate (GE, MGT and GI) were used for ranking order of seed lots, the ranking order had some inconsistency only for medium vigour seed lots. Seed lots with high germination percentages (C, G), emerged faster (MGT) and they had a higher germination energy (GE) and index (GI). Seed lots with low germination percentages (E, H) emerged slower and had a lower germination energy and index than the medium and high vigour seed lots. All the intermediate seed lots (A, B, D and F) performed moderately. However, there were high irregularities in ranking order among each other. Seed lot F was grouped as a fifth according to the SGT (20°/30°C), but it ranked, as sixth, third and fourth according to GE, MGT and GI (Table 4) respectively. Therefore, there is no assurance that a high vigour seed lot will geminate faster or that a low vigour seed lot

will germinate slower. However, as shown from other seed lots, there is a tendency for seed lots with a high germination percentage to geminate faster.

3.4. Conclusions

There were vigour difference among the seed lots of all three crops used in this study. High vigour seed lots perform well at both standard and non-standard temperatures, while low vigour seed lots perform poorly when being incubated at adverse temperatures. When the germination percentages at different incubating temperatures (10°, 15°, 20°, 20°/30° and 30°C) were compared, the low incubation temperature (10°C) was best to separate cabbage and onion seed lots with different levels of vigour. At this low temperature (10°C) no normal tomato seedlings were recorded, while an incubation temperature of 20°C was found to be best suited for distinguishing between tomato seed lots at different stages of physiological deterioration.

Some South African companies are using 20°C as the germination temperature in standard germination tests for tomatoes. According to the present results, 20°/30°C as suggested by the ISTA for the SGT, resulted in much better germination percentages than using 20°C. Germination at 20°C was better than the suggested 20°/30°C and 15°C for cabbage and tomato respectively, and could be used as an alterative if the two suggested temperatures are unavailable when performing a SGT.

With the use of non-standard temperatures for germination, one can easily distinguish between high and low vigour seed lots, but not between the moderate vigour seed lots. To differentiate seed lots with moderate vigour, the rate of germination parameters (MGT, GI and GE) can be used. However, since rate of germination is highly temperature (Ellis & Roberts, 1980) and moisture dependent, it needs careful regulation to ensure accurate results.

It is also important to note that in calculating the GE, MGT and GI, all seeds with emerged radicle were counted. Therefore a seed lot can have a short MGT or high GI even if there were few normal seedlings. Therefore, application of these tests as vigour tests without determining the standard germination percentage, may lead to incorrect conclusions.

CHAPTER 4
ACCELERATED AGEING TEST

4.1. Introduction

Some seed lots may consistently emerge poorly despite having high and acceptable laboratory germination. These lots have come to be described as low vigour seed and the major cause of low vigour is ageing (Matthews, 1980). Vigour test of vegetable seeds relate better to emergence in the field under environmental stressed conditions than the results of the standard germination test. This has lead many vegetable seed companies to use one or more vigour tests in their quality control performances (Dornbos, 1995a).

Vigour tests were introduced to predict stand establishment under a wide range of environmental conditions. The use of seed vigour, as a parameter of seed quality, has received much attention in the past four decades. Even though many vigour tests were proposed, only a few have attained acceptance by seed analysts and seed testing organizations. One test that has been accepted is the accelerating ageing (AA) test (Copeland & McDonald, 2001). Seeds of most species can be stored for short periods of time without affecting their viability. However, seeds gradually lose viability during storage. Ageing which occurs over time during storage is called natural ageing (Bewely & Black, 1994). However, seeds can also be aged artificially by subjecting them to high temperatures and relative humidity.

The AA test is considered standardized and correlates well with field emergence under a variety of seedbed conditions (TeKrony, 1995). One of the advantages of this test is that temperature and duration of ageing are easy to manipulate in obtaining the best accelerated ageing conditions for a specific crop. As a result, research continues to further define the ideal test conditions for various crops (Ferguson, 1995).

The AA test incorporates many of the important traits desired in a vigour test. Initially it was proposed as a method to evaluate seed storability. This test exposes seeds for short periods of time (3 to 4 days) to the two environmental variables that cause rapid seed deterioration, namely high temperature (41°C) and humidity (around 100%). The seeds are then removed from the stress conditions and placed under optimum germination conditions. The selection, maintenance, and use

of control samples for each species tested are essential in vigour testing. High vigour seed lots will withstand these extreme stress conditions and deteriorate at a slower rate than low vigour seed lots.

The AA test is one of the most acceptable vigour tests and was found to predict the emergence of large seeded agronomic crops. However, it had some limitations when it was used for small seeded vegetables due to rapid absorption of water by the small seeds during ageing. As a result of the above limitations, the standard AA test was modified by using saturated salt solutions instead of water (Jianhua & McDonald, 1996). Since the relative humidity of saturated salt solutions is lower than that of water ($\leq 100\%$) smaller seeds may absorb water more slowly. As the relative humidity of the accelerated ageing chamber declines, seed moisture content decreases. Due to the use of saturated salts the authors call it the saturated salt accelerated ageing (SSAA) test. This test was used in distinguishing vigour differences between seed lots of impatiens (*Impatiens wallerana* Hook) seeds (Jianhua & McDonald, 1996). Therefore, the SSAA could also be helpful in predicting emergence of small seeded vegetables under field conditions. In this experiment different saturated salts, with a wide range of relative humidity, were used to predict the field emergence of cabbage, onion and tomato.

The objectives of this study were:

- To evaluate different saturated salts and relative humidities for use in the SSAA.
- To evaluate the predictive value of the SSAA vigour test for the emergence of cabbage, onion and tomato seed lots.

4.2. Materials and Methods

4.2.1. Test conditions

The same seed lots used in Chapter 3 (Table 1) were again used for this experiment. The accelerated ageing vigour test requires the monitoring and controlling of time, temperature, and moisture for the duration of the test. For all species, a Labcon incubator with a temperature of 41^{o}C was used as an outer chamber, and all the seed lots were aged for 72 hours. The standard AA and SSAA tests were conducted according to TeKrony (1995) and McDonald (1997). All other equipment and procedures were according to the recommendations for a standard accelerated ageing test. In the use of the SSAA, saturated salt solutions were used instead of distilled water. The inner chamber boxes and

screen trays were washed thoroughly in a 20% Sodium Hypo-Chlorite solution and then dried. This was done after each use, to prevent fungal contamination.

4.2.2. Moisture content

The initial seed moisture of each seed lot was determined using the method appropriate for the specific crop (ISTA, 1999). Cabbage and onion seeds were placed in an oven at a temperature of 103°C for 18 hours, and tomato at 130°C for one hour. Percentage fresh mass was determined by using equation 5 and all seeds of all crops had a moisture content ranging from 7 to 10% (fresh mass basis).

$$\%\ Fresh\ mass = \frac{mass\ before\ drying - mass\ after\ drying}{mass\ before\ drying}\ x\,100 \qquad (5)$$

4.2.3. Procedure

Four different saturated salt solutions, producing different relative humidities were used. Detailed information on the salts and their RH is presented in Table 5. The saturated solutions were prepared by dissolving salts in distilled water and salt was added until saturation. Then, 40ml of distilled water (for standard AA) or saturated salt solution (for SSAA) was added to each ageing box.

Table 5: Relative humidity of distilled water and the different saturated salt solutions that were used in the accelerated ageing test

	RH (%)	Test
Distilled water	RH100	Standard AA
NaCl	RH75	SSAA
$Ca(NO_3)_2$	RH43	SSAA
$MgCl_2$	RH32	SSAA

From each seed lot, 300 seeds (mass ± 1 gram) were placed on a screen tray and spread out in a single layer (Figure 2). The screen trays were inserted into the inner chambers (plastic boxes) containing 40ml of distilled water or saturated salt solution. Each inner chamber was covered by a lid (cover). The inner chambers (ageing boxes) were placed in an incubator (outer chamber) and the seeds were aged at 41°C for 72 hours. To maintain a uniform and constant temperature, the door of the incubator remained closed for the entire ageing period.

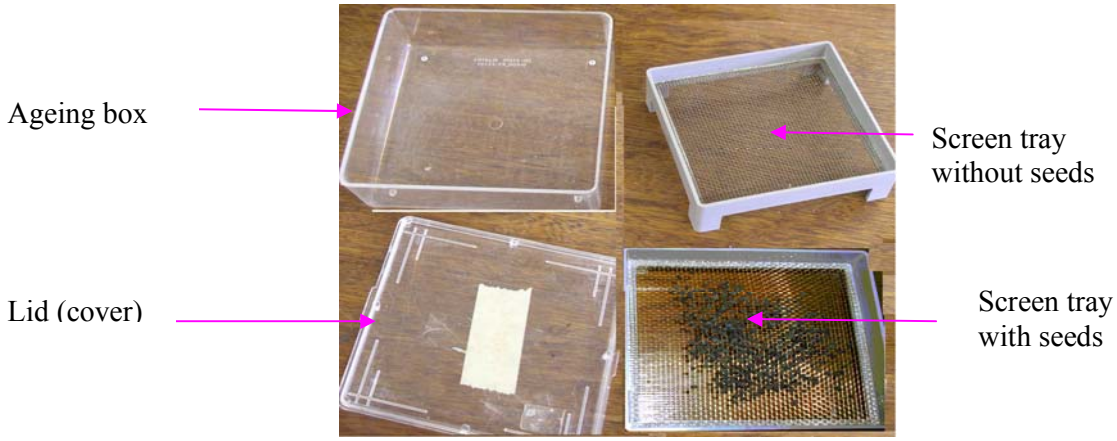

Figure 2: Illustration of the ageing box, lid (cover) and screen tray used in the AA and SSAA tests

After aged seeds were removed from the inner chamber, the moisture content of the seed was determined and a standard germination test (ISTA, 1999) was conducted. For determining the moisture content, two replicates of fifty seeds from each seed lot were weighed immediately after removing them from the incubator. Four replicates of fifty seeds from each seed lot and untreated control were placed in 9cm Petri dishes, using two layers of filter paper (Whatman #1), for the germination test. Cabbage and onion seeds were germinated at 20^{o}C and tomato at an alternating temperature of 20/30^{o}C. Normal seedlings were recorded, according to ISTA (1999) rules, at day 10, 12 and 14 for cabbage, onion and tomato seeds respectively.

4.2.4. Statistical analysis

The Petri dishes were arranged in a completely randomised design with four replications. The data was analysed using SAS, a statistical analysis software package (SAS, 1999). The LSD_{Tukey} was calculated for comparison of the seed lot means at α=0.05. ANOVA tables are included in the appendix and are indicated in the text by the letter A before the table number.

4.3. Results and Discussions

4.3.1. Moisture content

The differences in the initial seed moisture content can affect the degree of deterioration. In addition to vigour, Hampton (1995) reported that additional factors such as seed permeability, testa integrity and position of seed within the outer chamber can also affect ageing. The initial seed moisture content for all three crops ranged from 6.5 to 10.4% (fresh mass basis).

Table 6: Moisture content (fresh mass basis) of seeds after being aged at different RH's

Crop	Control	RH100	RH75	RH43	RH32
Cabbage	6.5-8.3 (±1.8)	29.8-38.7(±8.9)	8.1-10.7(±2.6)	7.4-8.8(±1.4)	5.3-6.7(±1.4)
Onion	8.2-9.8 (±1.6)	34.4-45.2 (±10.8)	9.7-13.1(±3.4)	8.5-12.2(±3.7)	6.6-7.2(±0.8)
Tomato	7.1-10.4(±3.3)	34.7-39.2 (±4.5)	11.6-13.9(±2.3)	9.2-11.9(±2.7)	6.0-7.1(±1.1)

For all seed lots, the seed moisture content increased as the RH increased (Table 6). The moisture content of RH32 was lower than the control, this may be due to the storage environment of the cold room. In all conditions, the moisture content of cabbage seeds was lower as compared to that of the other crop seeds. This could be due to relatively higher oil content of cabbage seeds, which might not release the moisture during oven drying. Copeland and McDonald (2001) also reported similar problems for *Brassicas* in which cabbage belongs and suggested the use of a higher oven temperature.

The deviation in seed moisture content in the standard accelerated ageing test (RH100) was above 4% for all crop seeds (Table 6). According to Rodo and Marcos-Filho (2003), a variation of seed moisture content above 4% between seed lots was out of tolerance. Powell (1995) reported variation in seed moisture content (ranging from 11.8 to 24.0%) among onion seeds lots, after 24 hours of accelerated ageing (100% relative humidity at 45ºC). TeKrony (1995) suggested seed moisture contents ranging from 39 to 44% for cabbage, 40 to 45% for onion and 44 to 46% for tomato. These results verified that the standard AA test results in a high variation of seed moisture content. Hence, it is not advisable to age small vegetable seeds such as cabbage and onion using the standard AA test (RH100 and 72 hours) (McDonald, 1997). However, for tomato seeds, the standard AA can be used as an alternative vigour test since the range was smaller (Table 6).

In all the crops the moisture content after the SSAA (RH75, RH43 and RH32) was lower than 14%. Thus, the salts were effective in lowering the moisture content of seeds. After the SSAA test, the range of moisture content between the seed lots for each crop was less than 4% (Table 6). According to Rodo and Marcos-Filho (2003), the maximum range in seed moisture content after the saturated salt accelerated ageing test using NaCl was 1.1%, and they recommend variations in seed moisture content up to 3 to 4% after ageing, as tolerable. Therefore, the present results reconfirmed the reports of Jianhua and McDonald (1996) that salt solutions induce seeds to absorb water at a lower speed, resulting in lower seed moisture contents.

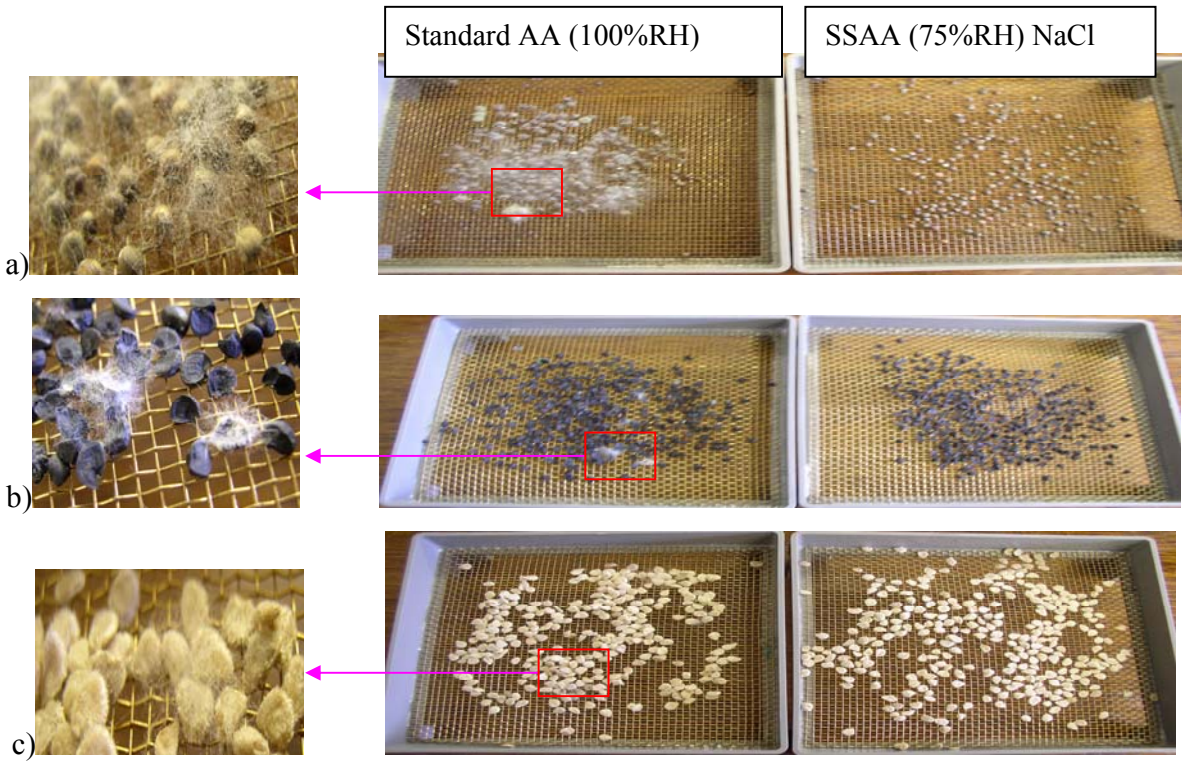

Figure 3: Fungal growth on a) cabbage, b) onion and c) tomato seeds subjected to the standard AA (RH100) and SSAA (RH75) tests

Fungal growth was observed on the seeds in the standard AA test (RH100) (Figure 3), this was encouraged by the high moisture content of the seeds resulting from the high relative humidity. According to Kulik (1995) fungi can attack and destroy seeds over a wide range of RH (65 to 100%) and temperature (4 to 45°C). High (>30%) moisture content seeds may germinate, and at moisture contents of 18 to 30% rapid deterioration due to micro-organism activity may occur. The fungal

growth was more severe in cabbage, followed by onion and tomato seeds. This could be due to the relative smaller size of cabbage seeds as compared to that of onion and tomato.

On the other hand, no fungal growth was observed using saturated salts (Figure 3). Bewely and Black (1994) reported that fungal growth was absent below a RH of 68%, hence they suggested fungal growth is not responsible for deterioration that occur below a moisture content of 13% for starchy seeds and below 8% for oily seeds. Rodo and Marcos-Filho (2003) also found no fungal growth in onion seeds when seeds were aged using saturated salt solutions of NaCl. Similarly, no fungal growth was reported in impatiens (*Impatiens wallerana* Hook) seeds by Jianhua and McDonald (1996) after seeds were aged with NaCl, KCl and NaBr.

4.3.2. Ageing conditions

The results of different ageing conditions on the standard germination of the seeds of all three crops used are presented in Table 7. For all the crops, the germination percentage varied significantly among the different ageing conditions (relative humidities) (Table A5). Irrespective of crop and ageing condition the mean germination percentage of aged seeds was lower as compared to the control seeds. The mean germination percentage difference was highest for cabbage (50.5%) followed by onion (27.86%) and tomato (13.94%). Cabbage seed lots were more affected by the different ageing conditions, which implies that cabbage seed lots used in this study differ in vigour more widely as compared to onion and tomato. The reason for the huge variation could also be due to the genetic makeup of the cabbage seeds, and it is unlikely that all types of crop seeds will respond the same to the SSAA tests.

The analysis of variance for ageing condition, seed lot and their interaction for all crops is shown in Table A6. In all crops, there were highly significant difference in standard germination parentages for the different ageing conditions. For cabbage and onion seeds, the germination percentages for all ageing conditions were significantly lower than that of the control (Table 7), with the exception of the germination percentage after ageing at RH32, which was not significant. Ageing tomato seeds at RH32, RH75, and RH100 did not significantly affect the germination percentages as compared to that of control. For cabbage and onion, the highest germination percentage was recorded at the control followed by RH32, RH75, RH43 and lowest at RH100 (standard AA). However, for tomato seeds the lowest parentage was obtained from RH43. Powell (1995) found seed lots that absorb

moisture rapidly reaching high moisture contents after only one day, to have a lower germination percentage as compared to high vigour seed lots that absorb water relatively slow.

Table 7: Comparison of different relative humidity ageing tests on the germination percentage of cabbage, onion and tomato

Ageing RH	Germination %		
	Cabbage	Onion	Tomato
Control (SGT)	90.17a[1] (1)[2]	90.50a (1)	89.19a (1)
Water (RH100)	39.67c (5)	62.64c (5)	82.56a (4)
NaCl (RH75)	69.50b (3)	73.36b (3)	82.63a(3)
$Ca(NO_3)_2$ (RH43)	69.42b (4)	63.43c (4)	75.25b (5)
$MgCl_2$ (RH32)	79.92ab (2)	87.29a (2)	85.13a (2)
Means	69.73	75.44	82.95
CV	23.19	16.04	12.56
F value	32.72**	32.47**	7.60**
LSD_{Tukey}	12.94	8.94	7.19
DIFF	50.5	27.86	13.94

[1]Values in each column followed by the same letters were not significantly different.
[2]Values in parenthesis are the ranking order of the ageing tests. **Significant at α=0.01.

As shown in Table 8 to 10, high vigour seed lots performed well before and after ageing. However, some seed lots (low vigour) in spite of their high standard germination percentage show low germination percentages after ageing. The significant "ageing x seed lot" interaction indicates that there was vigour difference between seed lots of all crops used in this study (Table A6). These results are in agreement with Powell (1995) who found interaction between ageing and seed lot of onion seeds. Seed ageing is the loss of seed quality with time (Coolbear, 1995). Ageing of a seed is designated generally by reduced germination percentage and rate, slower growth, increased susceptibility to environmental stresses and reduce resistance to storage condition under adverse conditions (Benjamin, 1990).

4.3.3. Seed lots

a) Cabbage

The vigour percentage varied significantly between different seed lots within each ageing condition (Table A7). The ranking order of seed lots of cabbage according to percentage germination, after different ageing conditions is shown in Table 8. Germination percentage (control), means, CV, F value, LSD_{Tukey} (α=0.05) and DIFF are also presented. Similarly, these features are presented in Tables 9 and 10 for onion and tomato respectively.

The overall cabbage mean germination percentage after RH100 (standard AA) was the lowest (39.7%) followed by RH43 (69.4%), RH75 (69.5%) and highest at RH32 (79.9%). The low percentage germination at the standard AA test may also reflect the infection of seed lots with pathogens rather than only physiological state of the seed.

Table 8: Ranking of cabbage seed lots based on germination percentages after different ageing conditions

Seed lot	Germination %					
	Control	RH100	RH75	RH43	RH32	Mean
A	88.5ab[1](4)[2]	29.0d (4)	69.0c (4)	74.0a (5)	79.0a (5)	67.9c (4)
B	93.0ab (2)	61.0b(2)	80.5ab(2)	80.5a (2)	83.0a (3)	79.6b (2)
C	83.5b (6)	8.5f (6)	42.0d (6)	25.0b (6)	65.5b (6)	44.9d (6)
D	91.5ab (3)	49.0c (3)	73.5bc(3)	79.0a (3)	84.5a (2)	75.5b (3)
E	88.0ab (5)	19.0e (5)	64.5c (5)	76.0a (4)	82.5a (4)	66.0c (5)
F	96.5a (1)	71.5a (1)	87.5a (1)	82.0a (1)	85.0a (1)	84.5a (1)
Mean	90.17	39.67	69.5	69.42	79.92	
CV	5.07	11.17	6.29	6.03	5.12	2.82
F value	3.90*	126.22**	52.01**	110.18**	13.0**	203.31**
LSD_{Tukey}	10.27	9.91	9.82	9.40	9.19	4.42
DIFF	13.0	63.0	45.5	57.0	19.5	39.5

[1]Values in each column followed by the same letters were not significantly different. [2]Values in parenthesis are the ranking order of the seed lots. *Significant at α=0.05, and **significant at α=0.01.

Germination assessed with the SGT ranged from 83.5 to 96.5%. The germination after ageing was lower and ranged from 8.5 to 87.5%. For high vigour (F, B) seed lots, the mean germination between SGT and germination after ageing were equivalent. However, for seed lots C (low vigour) the difference was 75.0%, 41.5%, 58.5% and 18.0% at RH100, RH75, RH43 and RH32 respectively. Therefore, the ranking order of seed lot C was not only consistently low, but also substantially affected by ageing. The present results, therefore, verify that germination percentage declines only after a significant loss of vigour. Hence, it is recommended that the SGT should be supplemented with a seed vigour test which is an important criterium for predicting emergence of a seed lot (McDonald, 1995).

When the germination percentage of all ageing conditions was compared, seed lots B and F ranked as first and second, and seed lot C last. Consequently, seed lots (B, F) can be grouped as high vigour seed lots and seed lot C as low vigour seed lot. Seed lots A, D and E had moderate values and can be grouped as intermediate vigour seed lots.

b) Onion

The ranking orders of onion seed lots according to standard germination percentage, and germination percentage after different ageing conditions are shown in Table 9. The germination percentage of the SGT was highest as compared to the germination percentage of all aged seeds. The SGT results ranged from 86 to 96% with a difference of 10%, however, germination after AA ranged from 33 to 94% with a difference of 61%. Germination after AA at RH43 (49%) exhibited the widest range followed by RH100 (39%), RH75 (26%), and RH32 (14.5%) (Table 9).

There were significant differences between seed lot means at all ageing conditions (Table A7). The largest difference between the standard germination (control) and ageing vigour test results was observed in low vigour seed lots when seeds were aged at RH43 (Table 9). Seed lot F had a germination percentage of 89.5% but it dropped to 33% when it was aged with RH43. In high vigour seed lots, such as seed lot B the difference was only 14%.

The overall average germination percentage at RH32 was lower than the standard germination percentage (control) by only 3% (Table 9). With the exception of the low vigour seed lot (seed lot F) all other seed lots, irrespective of their vigour level had only slightly lower germination

percentage as compared to the SGT results. The germination percentage of seed lot F was 89.5% at SGT and 79% at RH32 with a difference of 10.5%. The difference between the SGT and other ageing treatments were the highest for low vigour seed lots.

Table 9: Ranking of onion seed lots based on germination percentages after different ageing conditions

Seed lot	Germination %					
	Control	RH100	RH75	RH43	RH32	Mean
A	90.5bc[1](4)[2]	65.0b (4)	77.0abc(4)	69.0a (5)	89.0ab (4)	78.1b (4)
B	96.0a (1)	81.0a (1)	85.0a (1)	82.0a (1)	92.0a (2)	87.2a (1)
C	94.0ab (2)	66.0b (3)	82.5ab (3)	71.5a (3)	93.5a (1)	81.5b (3)
D	86.0c (7)	42.0c (7)	59.0d (7)	45.0b (6)	82.5cd (6)	62.9d (6)
E	87.0c (6)	59.5b (5)	65.5bcd(5)	69.5a (4)	85.5bc (5)	72.1c (5)
F	89.5bc (5)	45.0c (6)	61.5cd (6)	33.0b (7)	79.0d (7)	62.9d (7)
G	90.5bc (3)	80.0a (2)	83.0ab (2)	74.0a (2)	89.5ab (3)	83.4ab(2)
Mean	90.50	62.64	73.36	63.43	87.29	
CV	2.34	9.29	10.41	8.99	2.96	3.07
F value	11.44**	27.61**	8.39**	38.08**	16.34**	70.91**
LSD_{Tukey}	4.84	13.38	17.56	13.10	5.94	5.33
DIFF	10.0	39.0	26.0	49.0	14.5	24.3

[1]Values in each column followed by the same letters were not significantly different.
[2]Values in parenthesis are the ranking order of the seed lots. **Significant at α=0.01.

In all the ageing vigour tests, seed lots B, C and G, had the highest values and ranked as first, second or third even with no consistent ranking order among one another. Other seed lots had no consistent ranking order. Nevertheless, in all the three ageing vigour tests, seed lot F had the lowest germination percentage except at RH43 and can be considered as low vigour. Seed lots A, D and E had intermediate values and can be grouped as medium vigour seed lots.

The use of different relative humidities as an alternative for ageing was helpful in separating seed lots of onion based on their physiological stages of deterioration. As a result, a convenient ageing test can be performed based on the environmental conditions under which the seeds will be planted. According to Rodo and Marcos Filho (2003), the use of saturated salt accelerated ageing using NaCl

(RH75) was found successful for differentiating seed lots of onion that had similar germination percentage but different levels of vigour.

c) Tomato

The results of different ageing tests conducted under laboratory conditions on eight seed lots of tomato are presented in Table 10. The vigour percentages differed between seed lots for all ageing tests. The SGT (control) results were higher as compared to that of aged seeds and ranged from 76.5 to 98.5% with a difference of 22% between the lowest and highest values. The overall mean germination percentage of all eight tomato seed lots aged at RH32 revealed the highest germination percentage, followed by RH100, RH75 with the lowest at RH43. The largest difference (40.5%) was observed after seeds were aged at RH43, followed by the standard AA or RH100 (36%), RH75 (29%) and smallest at RH32 (27%).

Based on the results presented in Table 10, seed lots G and C had consistently the highest germination percentage for the SGT as well as germination percentage after ageing and can be grouped as high vigour seed lots. Seed lot E and H, had considerably lower germination percentages at both control and for all ageing conditions. Seed lot F ranked consistently as third in the control and all ageing conditions except when it was aged with RH43 where it ranked fifth. The rest of the seed lots (A, B, D) had inconsistent ranking order and they can be grouped as moderate vigour seed lots.

In tomato, the germination percentages after ageing at the standard AA (RH100), RH32 and RH75 were not significantly different (Table 6). All three ageing conditions can be used for separation vigour difference among tomato seed lots. Similarly, Panobianco and Marcos-Filho (2001) reported efficient use of the standard accelerated ageing test (41°C and 72h) and saturated salt test using NaCl (RH75) for detection vigour difference among seed lots of tomato. In addition, the efficiency of NaCl to detect different levels of seed physiological potential among seed lots was also verified for other cops such as green pepper (Panobianco & Marcos-Filho, 2001).

Table 10: Ranking of tomato seed lots based on germination percentages after different ageing conditions

Seed lot	Germination %					
	Control	RH100	RH75	RH43	RH32	Mean
A	89.5bc[1](5)[2]	79.5c (6)	84.0c (4)	78.0b (3)	86.0bcd (4)	83.4cd
B	87.0c (6)	83.0bc (5)	83.5c (5)	73.5b (6)	85.0bcd (5)	82.4d
C	94.5ab(2)	92.0ab (2)	93.0ab (2)	90.0a (2)	93.5ab (2)	92.6b
D	86.5c (7)	83.0bc (4)	74.0d (6)	74.5b (4)	83.0cd (6)	80.2d
E	90.0bc (4)	77.5c (7)	73.5d (7)	59.5c (7)	79.0d (7)	75.9e
F	91.0bc (3)	89.5ab(3)	87.0bc (3)	74.0b (5)	89.0abc (3)	86.1c
G	98.5a (1)	97.0a (1)	97.5a (1)	96.5a (1)	96.5a (1)	97.2a
H	76.5d (8)	59.0d (8)	68.5d (8)	56.0c (8)	69.0e (8)	65.8f
Mean	89.19	82.56	81.19	75.25	85.13	
CV	3.23	5.10	4.61	6.11	4.41	1.85
F value	20.12**	30.21**	27.90**	35.11**	20.99**	159.55**
LSD_{Tukey}	6.74	9.86	8.92	10.77	8.79	3.60
DIFF	22.0	38.0	29.0	40.5	27.5	31.4

[1]Values in each column followed by the same letters were not significantly different. [2]Values in parenthesis are the ranking order of the seed lots. **Significant at α=0.01.

4.4. Conclusions

In conclusion, the basic requirement of vigour test is the ability to provide consistent ranking of seed lots in terms of potential performance (Perry, 1984). There was no clear relation between SGT results and germination percentages after ageing test. Seed lots perform differently after different ageing tests as compared to SGT, some were affected severely (poor vigour), some slightly (medium vigour) while others remained unaffected (high vigour).

After physiological maturity, seeds will start to deteriorate at a rate, which is dependent on temperature and moisture (Probert & Hay, 2000). According to vigour test committee reported on by TeKrony (1995) the temperature of the outer chamber must be kept constant and he recommended a water-jacketed incubator capable of maintaining a constant temperature. He, however, said that alternative outer ageing chambers could be used, as long as the temperature is well controlled and

constant. For the present study, as an alternative ageing chamber, Labcon incubator with a constant temperature (41°C) was used. Therefore, the low germination percentages that were obtained after ageing seeds at RH43 could be due to the fluctuation in relative humidity with minor fluctuations in temperature. Using $Ca(NO3)_2$, Winiston and Bates (1960) reported a loss of some or all of the water due to hydration as the temperature rose to ±42°C. Therefore, it is important to know the transition points, so that salts should not be used **at** or **close** to the critical temperature.

After ageing $MgCl_2$ (RH32), the high vigour seed lots ones did not show any difference in their germination values, compared to the SGT results. In all crops, the overall mean germination percentage using $MgCl_2$ was not significantly different from that of the control seeds. During the accelerated ageing test seeds must be subjected to high temperature and relative humidity but in this case a low relative humidity was used ($MgCl_2$ ±32%RH). Consequently, seeds might not age since one of the factors i.e. relative humidity was low and seeds were aged only with high temperature.

NaCl (RH75) had relatively consistent results as compared to other ageing conditions in terms of seed moisture content. In this test, unlike that of standard AA no fungal growth was observed. In general, since the relative humidity of NaCl is constant at a range of temperatures, this salt can be used even with incubators (outer chambers), which are less precise in temperature regulations. Winiston and Bates (1960) suggested the use of this salt for reducing error introduced by fluctuations in temperature.

CHAPTER 5
EMERGENCE TRIAL

5.1. Introduction

Inadequate seedling emergence will reduce crop yield and in most situations, no amount of effort and expense later on crop development can compensate for this effect. Seedling emergence is the result of a complex interaction between seed quality and seedbed environment (Perry, 1984). Consequently, field emergence can be lower than the germination percentage that was obtained through the standard germination test. This resulted in the development of vigour testing that can be used to predict emergence of seedlings at a wider range of environmental conditions.

Although germination is a complex process, it has only three requirements: water, favourable temperature, and oxygen. Seeds are regularly sown into soil at varying temperature and moisture content (Benjamin, 1990). Consequently, growers started to use transplants by raising seedlings in containers where they can control the temperature and amount of water. The amount of water can easily be managed at low cost, however, to control the temperature a larger amount of initial and running capital is required. Due to these reasons, temperature has been taken as the limiting factor in seedlings production for the trials in this Chapter.

Vigour tests do not predict stand or percentage emergence easily, because the difference in the stand establishment depends on the year, season and location. Consequently, vigour test provides a measure of relative performance against other seed lots tested under similar conditions (McDonald, 1994). High vigour lots can be stored for longer periods of time without losing their germination ability. Therefore, if two seed lots have equal germinations, the one with the lower vigour might be considered for use first, because the germination of this lot will likely decrease faster than a lot of higher vigour. Vegetable crops such as cabbage, onion and tomato are planted either directly or using transplants. In both cases, high vigour seeds that can withstand a wider range of environmental variations are important in obtaining uniform, synchrony seedlings at the recommended plant population.

Hence, the aim of this experiment was to correlate the germination and vigour test results (Chapter 3 and 4) with the different emergence parameters that were obtained from various growing temperatures and media. The emergence trial of all crops was determined in three different

environmental conditions and two media. The number of emerged seedlings was counted daily until no further seedlings appeared.

5.2. Materials and Methods

5.2.1. Seed source

For this experiment the same seed lots and crops, which were listed in Chapter 3 (Table 1) in the germination test, were used.

5.2.2. Temperature

All the experiments were conducted in 2003 on the University of Pretoria Experimental Farm. Three different environmental conditions, in reference to temperature (high, favourable and low) were used. One of the experiments was conducted in open area in winter at the coldest months (June-July) of the year. In the open field the temperature data was recorded at the weather station on the Experimental Farm. The minimum, mean and maximum temperatures were 5.01°, 11.93° and 19.06°C respectively. The remaining two trials were conducted in controlled environmental conditions of 15°/25°C (favourable) and 30°C (hot).

5.2.3. Planting media

Seedling emergence can be restricted due to the formation of crusts on the soil surface (Benjamin, 1990), therefore, for a single seed lot different media can result in varied emergence. Cabbage, onion and tomato grow well in a wide variety of soils, but a well-drained sandy loam with high organic matter content is preferred. All these crops can be established by direct seeding to the field or by using transplants. To investigate if the growing media has an effect on vigour (percentage and rate of emergence) of seed lots, two media were used.

A commercial medium “Hygromix” was obtained from Hygrotech Seed Company and a red clay soil from the Experimental Farm. The commercial medium was used for comparison in the production of transplants. Generally, a commercially prepared soil-less medium is used for containerised-transplant production. Although, field soils are generally not used, due to poor drainage, and chances of contamination with diseases and weed seeds, soil was used in this experiment. The reasons being: firstly all direct-sown vegetable crops use soil as a medium of

growth. Secondly, since soil is easily accessible it can be used by resource poor farmers and lastly it can also be used as an alternative growth medium for countries where commercial media are not available. Nutrient analysis and textural classification of the soil were done by the laboratory of the Department of Plant Production and Soil Science at the University of Pretoria. Based on the results of the percentages of the textural classes, the soil was classified as sand clay loam (Brady & Weil, 1999). The soil nutrient and soil textural composition analysis are given in Table A8, in the appendix.

5.2.4. Procedure

All the experiments were conducted during 2003 at the Hatfield Experimental Farm of the University of Pretoria. In this experiment 128 cavity seedling trays were used and before use the trays were sterilized with 10% Sodium Hypo-Chlorite. The trays' cavities were filled either with commercial growth medium "Hygromix" or soil. Before use, the soil was sieved by using 2mm of sieve. Fifty seeds per seed lot were used and one seed per cavity was sown at a depth of 1 to 1.5 cm, which is the recommended seeding depth for the three crops that were used in this trial. Two seed lots were sown per tray as illustrated in Figure 4. The trays were watered daily with approximately equal amount of water. In order to keep the trays moist they were irrigated with a hand spray once a day during cold (winter) and twice a day during favourable (15°/25°C) and hot (30°C) conditions.

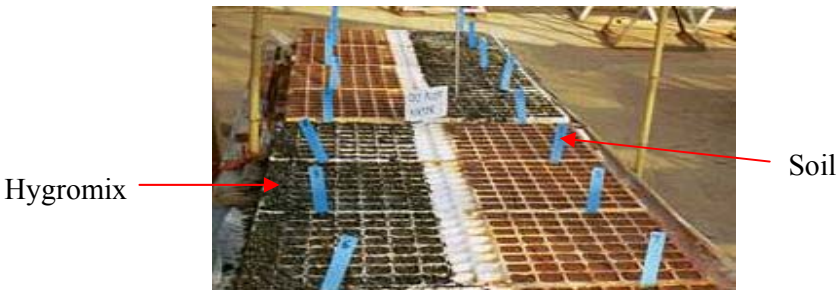

Figure 4: Sample trays used in the emergence trial

Emerged seedlings were counted daily until emergence ceased. From the daily counts final emergence percentage, mean emergence time and seedling vigour index were calculated according to the following formulas.

- Final emergence percentage (FEP) = the ratio between the number of seeds emerged and the total number of seeds used times 100. (6)
- Mean emergence time (MET) according to the expression (Ellis & Roberts, 1980):

$$MET = \frac{\sum (n\,d)}{\sum n} \quad (7)$$

Where n = number of seedlings emerged on day (d);

d = serial number of the day

∑n = total number of emerged seedlings

- Seedling vigour index was calculated after recording the number of seedlings emerged daily by using the following equation (Ruan *et al.*, 2002).

$$Seedling\,vigour\,index = \frac{number\,of\ seedlings\ emerged}{number\,of\ days\,of\ first\,count} + \frac{number\,of\ seedlings\,emerged}{number\,of\ days\,of\,\text{second}\,count} + \ldots + \frac{number\,of\ seedlings\ emerged}{number\,of\ days\,of\ last\,count} \quad (8)$$

Dry mass was determined by sampling 10 plant shoots per replicate and drying the shoots in an oven at 65°C for 72 hours. The average of ten plants was reported in milligram (mg) per plant.

5.2.5. Statistical analysis

A factorial experiment with three crops, 21 seed lots and two growing media was used. There were four replicates of each treatment allocated at random with separate blocks of each species. Data were analysed with a computer based SAS program software package (SAS, 1999) and LSD_{Tukey} was calculated at α=0.05. Simple linear correlation coefficients were determined between laboratory and different emergence test results. The correlation analysis was based on average values for each seed lot. Vigour tests that predict emergence best were determined using stepwise comparison and the models were developed at α=0.01 (Gomez & Gomez, 1984). ANOVA tables are included in the appendix and are indicated in the text by the letter A before the table number.

5.3. Results and Discussions

5.3.1. Cabbage

In cabbage, for all the variables, there were significant differences between planting temperatures (Table A9). In all variables the winter emergence trial had significantly performed lower as compared to the other emergence trials (Table 11). Further, with the exception of mean emergence time (MET) there was no significant differences in any of the other variables for the 15°/25°C and 30°C emergence trials. Seedlings at 30°C had significantly a lower MET than 15°/25°C, and emerged faster by approximately one day.

Table 11: Final emergence percentage, mean emergence time, seedling vigour index and seedling dry mass of cabbage seed lots planted under different temperatures and growth media

Source	Final emergence percentage (%)	Mean emergence time (days)	Seedling vigour index	Seedling dry mass (mg/plant)
Temperature				
Winter	70.88b[1]	12.11c	3.10b	242.29b
15°/25°C	88.17a	7.88b	5.92a	316.71a
30°C	85.12a	6.45a	6.86a	319.98a
Growth media				
Hygromix	82.22a	9.0b	5.20b	365.82a
Soil	80.56a	8.62a	5.39a	220.15b

[1]**Values in each column followed by the same letters were not significantly different.**

There was no significant difference in FEP (final emergence percentage) of cabbage seeds between the two growth media (Table A9), though the FEP in Hygromix was slightly higher than that of soil (Table 11). In contrast, the overall mean SVI (seedling vigour index) of seed lots in soil was significantly higher as compared to Hygromix. In spite of the higher SVI and faster emergence (lower MET), seeds that were planted in soil had significantly lower seedling dry mass than Hygromix. This could be primarily due to the low water holding capacity of the soil as compared to Hygromix, and hence most nutrients were leached down. Rodo and Marcos-Filho (2003) used expanded vermiculite to protect excessive evaporation from trays. Therefore, in the present experiment when seeds were planted at high temperature, vermiculite might be helpful for reducing evaporation, because Hygromix has vermiculite, but in low proportions in its composition. Lastly

since the seedlings were watered with pressurized sprayer the soil was also slightly compacted as compared to Hygromix. In line with this, Dornbos (1995a) suggested uneven emergence could be encouraged by limited soil moisture and soil compaction.

The results of the emergence trials and ranking orders of six seed lots of cabbage according to FEP and seedling vigour index are shown in Tables 12 and 13, respectively. In addition to the ranking order, means, CV, F value, LSD_{Tukey} (α=0.05) and DIFF are also presented. Similarly, these features are presented in Tables 16 and 17 for onion; and Tables 20 and 21 for tomato respectively.

Table 12: Ranking of cabbage seed lots based on FEP recorded from various emergence trials

Seed lot	Germination (%)	Emergence (%)			
		Winter	15°/25°C	30°C	Mean
A	90.5ab[1] (3)[2]	58.8cd (5)	84.3bc (5)	80.0b (5)	74.4c (5)
B	92.5ab (2)	81.3ab (2)	91.3ab (3)	89.3a (2)	87.3ab (2)
C	82.0b (6)	48.8d (6)	75.0c (6)	70.5c (6)	64.8d (6)
D	87.5ab (5)	70.3bc (4)	91.5ab (2)	88.3ab (4)	83.3b (4)
E	89.0ab (4)	77.8ab (3)	90.8ab (4)	88.8ab (3)	85.8ab (3)
F	96.0a (1)	88.5a (1)	96.3a (1)	94.0a (1)	92.9a (1)
Mean	89.50	70.88	88.17	85.13	
CV (%)	6.14	9.54	5.93	4.61	4.15
F value	2.82*	19.30**	8.23**	18.64**	36.28**
LSD_{Tukey}	12.35	15.29	11.75	8.82	7.59
DIFF	13.5	39.75	21.25	18.5	28.17

[1]Values in each column followed by the same letters were not significantly different. [2]Values in parenthesis are the ranking order of the seed lots. *Significant at α=0.05, and **significant at α=0.01.

There was a significant difference in the emergence percentage (Table A12) and seedling vigour index (Table A13) between seed lots as well as a significant interaction between temperature and seed lot (Table A9). This implies that there was a vigour difference among cabbage seed lots and some seed lots performed better at some temperatures. However, at ideal conditions seed lots with the same vigour should perform consistently in all temperatures. Perry (1984) reported similar emergence of seed lots at favourable environmental conditions, but differently at unfavourable

conditions. He concluded that the interaction between seed lot and environment was due to the inconsistent response of seed lots to a specific environment.

There was a significant difference between emergence percentages of seed lots of cabbage for all trials (Table A12). The emergence percentage range was highest at the winter trial (cold) and lowest at the 15/25°C trial (favourable) (Table 12). When the mean FEPs of all trials were compared, seed lots C and A were poorest and seed lot F was the best. Based on the emergence percentage, seed lots can be distinguished as high (F), medium (B, D, E) and poor (A, C) vigour seed lots. In the SGT seed lot A had relatively high percentage (91%) and ranked as third, however, in the emergence trial seed lot A ranked as fifth. This reconfirmed the principle of vigour testing, which states that high germinating seed lots may perform differently in the field (Coolbear, 1995). The causes of low emergence percentage could be mostly due to deterioration resulted from hostile environment.

Seedling vigour index (SVI) varied considerably among different emergence trials (between temperatures, media and seed lots) as seen in Table A13. As overall, the highest mean SVI were observed at 30°C followed by 15°/25°C and lowest in the winter emergence trial (Table 13). The low SVI at winter was due to the low temperature and the seedlings emerged slowly as compared to the others. Concerning the growth media, the highest SVI is observed in soil since seedlings emerged earlier in soil than in Hygromix. However, seedling development was higher in Hygromix than soil (Table 11). McNertnery (1989) reported that early development in the germination process is more dependent on seed vigour than later development. Later development is influenced more by environmental factors.

Generally, the SVI had a similar trend as the FEP and the highest SVI (Table 13) was obtained in seed lot F and lowest in seed lot C. However, the SVI of the intermediate seed lots was not consistent throughout all temperatures and media. This can be explained by differences in seed vigour between seed lots. The effect of cultivar might also play a great role in determining these parameters. Since cultivars differ in their resistance to adverse conditions they can emerge well under a range of environmental conditions while others may not. All cultivars did not respond systematically to either temperature or growth media in both parameters. For example seed lot A (90.5%) and B (92.5%) had similar germination percentages, however, in the emergence trial the performance of seed lot A was poorer.

Table 13: Ranking of cabbage seed lots based on SVI calculated from daily counts of various emergence trials

Seed lot	Winter		15°/25°C		30°C		Mean
	Hygro	Soil	Hygro	Soil	Hygro	Soil	
A	2.47cd[1](5)[2]	2.50cd (5)	4.81b (5)	5.14bc(5)	6.28b (5)	5.86c (5)	4.51c (5)
B	3.75ab (2)	3.70ab (2)	6.39a (3)	6.89a (1)	7.12b (2)	7.27b (4)	5.86b (2)
C	1.71d (6)	1.86d (6)	4.23b (6)	4.19c (6)	4.62c (6)	5.03c (6)	3.61d (6)
D	3.04bc (4)	3.10bc (4)	6.40a (2)	6.58ab(4)	6.89b (4)	7.56ab(3)	5.59b (4)
E	3.53ab (3)	3.24abc(3)	6.18a (4)	6.73ab(3)	7.09b (3)	7.86ab(2)	5.77b (3)
F	4.14a (1)	4.18a (1)	6.64a (1)	6.86a(2)	8.31a (1)	8.48a (1)	6.43a (1)
Mean	3.10	3.11	5.78	6.06	6.72	7.01	
CV (%)	11.23	14.04	5.52	11.95	7.09	6.44	3.60
F value	26.35**	14.56**	39.45**	9.76**	26.26**	33.40**	118.99**
LSD_{Tukey}	0.79	0.98	0.72	1.63	1.07	1.02	0.43
DIFF	2.43	2.32	2.41	2.70	3.69	3.45	2.83

[1]Values in each column followed by the same letters were not significantly different. [2]Values in parenthesis are the ranking order of the seed lots. **Significant at α=0.01.

The correlation coefficients between laboratory (germination and vigour) and emergence tests (FEP and SVI) results of six seed lots of cabbage are shown in Table 14. Since there was no significant difference between the two media in terms of FEP, correlations were calculated only for the planting temperatures. The SVI of the two growth media was significantly different and the correlations were determined for separate media and temperature as seen in Table 14. There were highly significant (α=0.01) correlations between the germination percentages at 10°C and all emergence trials (FEP and SVI). On the contrary, Strydom and Van De Venter (1998) reported correlation between germination percentage at 10°C and winter emergence trial but not with other trials. The contrasting result could be since they used some seed lots with low (below 80%) germination percentages, but in the current study seed lots that have standard germination below 80% were excluded from calculating correlation coefficients. All the germination percentage at different temperatures (20°, 20°/30° and 30°C) and GC (germination capacity) had also significant correlation (α=0.05) with most emergence trials.

Table 14: Correlation coefficients between laboratory test and emergence test results of cabbage seeds

Laboratory tests	Emergence percentage				Seedling vigour index					
					Temperature			Growth media		
	Winter	15°/25°C	30°C	Mean	Winter	15°/25°C	30°C	Hygro	Soil	Mean
Germination %										
10°C	0.94**	0.96**	0.96**	0.96**	0.96**	0.90*	0.94**	0.96**	0.93**	0.95**
20°C[1]	0.84*	0.84*	0.83*	0.85*	0.86*	0.71	0.82*	0.83*	0.78	0.81*
20°/30°C[2]	0.84*	0.81*	0.82*	0.83*	0.88*	0.72	0.82*	0.85*	0.78	0.82*
30°C	0.89*	0.87*	0.87*	0.89*	0.91*	0.77	0.88*	0.89*	0.84*	0.86*
Germination rate										
GE	0.85*	0.83*	0.84*	0.85*	0.87*	0.75	0.83*	0.85*	0.80	0.82*
MGT	-0.78	-0.70	-0.72	-0.75	-0.80	-0.63	-0.71	-0.74	-0.69	-0.72
GI	0.82*	0.71	0.73	0.78	0.84*	0.64	0.73	0.77	0.72	0.74
Viability test										
GC	0.87*	0.87*	0.86*	0.87*	0.90*	0.77	0.85*	0.88*	0.82*	0.85*
Ageing tests										
RH100	0.80	0.78	0.78	0.80	0.83*	0.76	0.77	0.81*	0.77	0.79
RH75	0.86*	0.91*	0.90*	0.90*	0.90*	0.84*	0.88*	0.91*	0.86*	0.88*
RH43	0.81*	0.91*	0.91*	0.87*	0.83*	0.85*	0.87*	0.88*	0.85*	0.86*
RH32	0.91*	0.98**	0.97**	0.95**	0.92**	0.90*	0.97**	0.96**	0.93**	0.95**

[1, 2]Standard germination test for cabbage according to ISTA. *Significant at α=0.05, **significant at α=0.01 and all the rest are not significant.

Germination energy had a significant correlation with both MGT (mean germination time) and GI (germination index) at α=0.05, however, failed to correlate with all emergence trials. Matthews (1980) also reported non-significant correlation between MGT and percentage emergence of cabbage seeds. Thus there is no guarantee for all fast germinating seed lots to have high emergence percentage. In some situations seed lots with high rate of germination may have low emergence percentages. This could be due to the genetic character of the seed lots and due to the environmental growth conditions of the mother plants.

All ageing results, except standard AA (100RH) have correlation with all emergence trials (FEP and SVI) of cabbage. Similar to the current study, Powell (1995) reported non-significant correlations between standard AA and field emergence results. The germination percentage at RH32 was highly correlated (α=0.01) with most emergence trials. The high correlation could be due to the relatively higher germination percentage after SSAA (RH32).

When the correlations between the laboratory and SVI results of two growth media were compared, more significant correlations were shown with Hygromix than soil. As observed in Figure 5, the best predictive equations for the final emergence percentage was found to be the germination percentage at 10°C, and for the seedling vigour index the germination percentage after ageing at RH32.

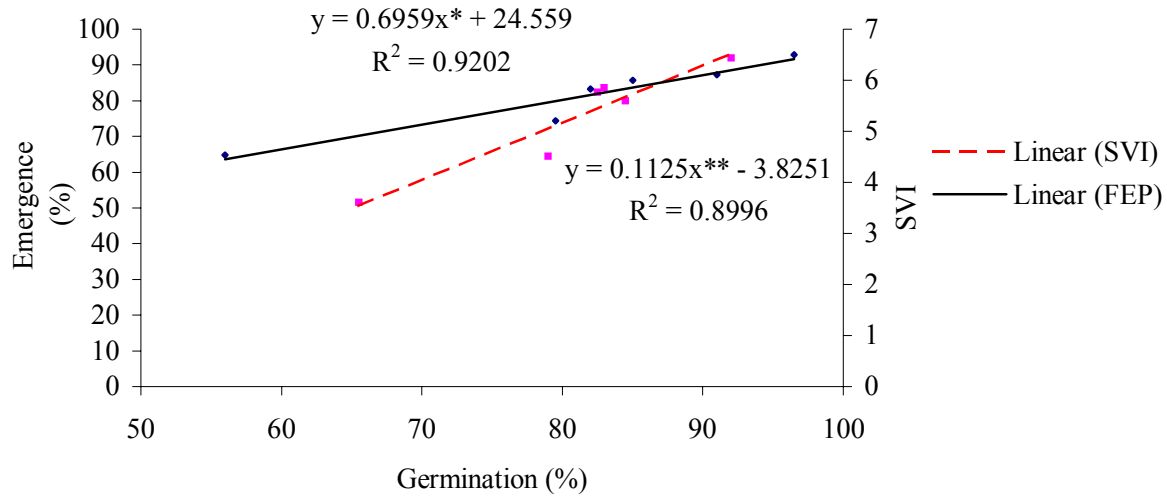

Figure 5: Best equations for predicting FEP: final emergence percentage (*germination % at 10°C); and SVI: seedling vigour index (**germination % after ageing of RH32) of cabbage seeds planted under a range of temperatures

5.3.2. Onion

The ANOVA of temperature and growth media of the onion emergence trials is shown in Table A10. For all parameters, there were significant differences among the temperatures (Table 15). Similar to cabbage, there was statistically significant difference between growth media for all parameters of onion seeds, except for the emergence percentage (Table A10). Seed lots emerged from the soil significantly faster than from Hygromix and they had higher SVI. However, higher seedling dry mass was obtained from Hygromix as compared to that of soil.

Table 15: Final emergence percentage, mean emergence time, seedling vigour index and seedling dry mass of onion seed lots planted under different temperatures and growth media

Source	Final emergence percentage (%)	Mean emergence time (days)	Seedling vigour index	Seedling dry mass (mg/plant)
Temperature				
Winter	79.64c[1]	22.25c	1.86a	86.51b
15/25 °C	86.85a	14.38b	3.11b	95.32a
30 °C	83.82b	11.63a	3.66c	102.83a
Growth media				
Hygromix	83.67a	16.43b	2.82b	102.31a
Soil	82.98a	15.75a	2.93a	87.45b

[1] **Values in each column followed by the same letters were not significantly different.**

The performance of seed lots was not consistent (Table 16) in all temperatures, possibly as a result of vigour difference among seed lots used in this study. This was also illustrated more clearly by other parameters used (MET and SVI). The interaction between temperature and seed lot was significant for MET and SVI. On the contrary, there was no interaction between temperature and seed lot in seedling dry mass (Table A10). This could be due to the growth conditions or vigour of seed lots. This demonstrated that some late emerged seed lots, can still perform well in the later stages of seedling development.

Table 16 shows the ranking of seven seed lots of onion based on the FEP. The standard germination test is good for predicting emergence of seed lots when onion seeds are planted at favourable environmental conditions. The emergence trial at 15°/25°C demonstrated the narrowest range (12%) and in the SGT the difference was only 10%, which is similar to the difference obtained from the

emergence trial at a favourable temperature (15°/25°C). However, for other emergence trials, irrespective of growth media, the germination percentage range was about 20%. Although all seed lots showed higher germination in the laboratory, there was a wide range in emergence percentage at non-favourable temperatures (winter and 30°C), indicating that they differed in vigour. Seed lots that have similar germination percentages may not perform the same under adverse conditions and thus a difference in vigour can be expected. Difference in emergence and growth of seed lots that have similar germination percentage can be explained by differences in seed vigour (Hampton, 1981).

Table 16: Ranking of onion seed lots based on FEP recorded from various emergence trials

Seed lot	Germination (%)	Emergence (%)			
		Winter	15°/25°C	30°C	Mean
A	90.5abc[1](4)[2]	81.8ab (4)	89.5ab (3)	89.8ab (3)	87.0a (3)
B	94.5a (1)	86.5a (2)	91.3a (2)	90.0ab (2)	89.3a (2)
C	93.5ab (2)	86.8a (1)	93.5a (1)	91.0a (1)	90.3a (1)
D	84.5d (7)	78.5abc(5)	82.3bc (5)	82.0bc (5)	80.9bc (5)
E	86.0cd (6)	72.5bc (6)	82.0bc (6)	73.8c (7)	76.1cd (6)
F	88.5bcd(5)	67.8c (7)	81.3c (7)	73.8c (6)	74.3d (7)
G	91.5abc(3)	83.8ab (3)	86.3abc(4)	86.5ab (4)	85.5ab (4)
Mean	89.85	79.64	86.50	83.82	
CV (%)	2.68	6.30	4.08	4.53	2.81
F value	9.57**	8.29**	7.47**	15.61**	29.51**
LSD_{Tukey}	5.54	11.53	8.12	8.74	5.37
DIFF	10.0	19.0	12.25	17.25	16.0

[1]Values in each column followed by the same letters were not significantly different. [2]Values in parenthesis are the ranking order of the seed lots. **Significant at α=0.01.

The ranking order of seven seed lots of onion based on SVI is presented in Table 17. These seed lots had a similar ranking order as those presented in Table 16, when seed lots were ranked according to FEP. The separation of seed lots was the same as that of FEP. Seed lot B and C showed the highest FEP and SVI in all emergence trials at all temperatures and growth media and they can be grouped as high vigour seed lots. Seed lot A and G had moderate performance (medium vigour) while D, E and F had consistently poor performances (low vigour). These results are in accordance to that

found in the previous chapters. High vigour seed lots had the same ranking order as the SGT and emergence trials. However, irregularities were observed in ranking order among the intermediate and low vigour seed lots between SGT and emergence trials.

Table 17: Ranking of onion seed lots based on SVI calculated from daily counts of various emergence trials

Seed lot	Winter		15°/25°C		30°C		Mean
	Hygro	Soil	Hygro	Soil	Hygro	Soil	
A	1.90ab[1](4)[2]	1.98ab (4)	3.15bc(3)	3.34ab (3)	3.93ab(2)	4.13a (1)	3.08bc(3)
B	2.15a (1)	2.14a (1)	3.63a (1)	3.82a (1)	4.12a (1)	4.12a (2)	3.33a (1)
C	2.10a (2)	2.09a (2)	3.50ab (2)	3.73a (2)	3.50bc(4)	3.94a (3)	3.21ab(2)
D	1.80ab (5)	1.79abc(5)	2.66d (5)	3.00bc (5)	3.49bc(5)	3.76ab(4)	2.75d(5)
E	1.58bc (6)	1.63bc (6)	2.58d (6)	2.80c (7)	3.05c (7)	2.95c (7)	2.43e(6)
F	1.37c (7)	1.39c (7)	2.56d (7)	2.80c (6)	3.15c (6)	3.28bc(6)	2.42e(7)
G	2.02a (3)	2.08a (3)	2.82cd(4)	3.12bc (4)	3.73ab(3)	3.72ab(5)	2.92cd(4)
Mean	1.85	1.89	2.99	3.23	3.62	3.70	
CV (%)	8.58	9.71	6.68	7.36	6.23	7.59	3.02
F value	13.02**	9.53**	19.95**	12.77**	12.88**	9.84**	68.64**
LSD_{Tukey}	0.36	0.42	0.46	0.55	0.52	0.65	0.20
DIFF	0.78	0.75	1.07	1.03	1.07	1.19	0.91

[1]Values in each column followed by the same letters were not significantly different. [2]Values in parenthesis are the ranking order of the seed lots. **Significant at α=0.01.

The correlations between different laboratory (germination and vigour) and emergence test (FEP and SVI) results of onion seeds are listed in Table 18. Germination percentage at 15° and 30°C were correlated significantly with most emergence trials (FEP and SVI). However, germination at 10°C was only correlated with winter emergence trial of both parameters, while germination at 20°C correlated with only emergence at 15°/25°C (favourable). Similarly, Smith and Varvil (1984) reported the use of a cool test in predicting field performance in adverse condition. Germination at standard temperatures (15° and 20°C) and the GC had highly significant correlations with FEP at favourable temperature (15°/25°C). This proved that standard germination percentage is a good tool for predicting emergence of onion seed lots when planted under conducive environmental

conditions. It has been reported that the standard germination test is an excellent predictor of field emergence under ideal conditions (TeKrony, 2003). The use of GC (viability) may be helpful for predicting emergence of onion seed lots when planted under suitable environmental conditions. In case of very favourable environmental conditions the abnormal seedlings might develop to good transplant seedlings. The use of GC (radicle protrusion percentage) may avoid disagreement arising because of subjective assessment of normal seedlings (Matthews, 1980).

When correlations were determined between emergence test results and laboratory germination rate parameters, GE had good correlation with all emergence trials. The MGT correlated negatively only with the winter emergence trial, while the GI correlated positively with both adverse planting conditions (winter and 30°C). Most emergence test results had no significant correlation with the standard AA (RH100). Similarly, Kraak *et al.* (1984) reported non-significant correlation between AA test (at 55%RH and 20°C for 14 days) results and field emergence percentage of onion. The germination percentage after ageing using the RH43 was only correlated (α=0.05) with the winter emergence trial. However, germination results after ageing of RH75 (NaCl) and RH32 ($MgCl_2$) were significantly correlated with stress emergence trials (winter and 30°C) and were highly correlated with the favourable (15°/25°C) emergence trial (α=0.01). Rodo and Marcos-Filho (2003) reported close relation between SSAA test using NaCl (75RH) and seedling emergence at 25°C.

As overall correlation, germination at all four temperatures (Table 18) had significant correlation with both growth media, though the values were higher for Hygromix than for soil. All the germination and vigour test parameters except 10°C, MGT, standard AA, and RH43 results had significant correlations with both growth media. Generally there was no pronounced difference in correlation between the two growth media used. However in most cases the correlation values were slightly higher for Hygromix than soil (Table 18).

Table 18: Correlation coefficients between laboratory test and emergence test results of onion seeds

Laboratory tests	Emergence percentage				Seedling vigour index					
					Temperature			Growth media		
	Winter	15°/25°C	30°C	Mean	Winter	15°/25°C	30°C	Hygro	Soil	Mean
Germination %										
10°C	0.84*	0.73	0.72	0.78*	0.87*	0.66	0.60	0.75	0.70	0.73
15°C[1]	0.85*	0.93**	0.82*	0.88**	0.83*	0.93**	0.69	0.88**	0.83*	0.86*
20°C[2]	0.72	0.88**	0.75	0.79*	0.72	0.86*	0.72	0.84*	0.78*	0.81*
30°C	0.98**	0.79*	0.89**	0.92**	0.99**	0.79*	0.83*	0.90**	0.89**	0.90**
Germination rate										
GE	0.90**	0.77*	0.82*	0.86*	0.93**	0.73	0.79*	0.85*	0.83*	0.84*
MGT	-0.80*	-0.62	-0.69	-0.73	-0.85*	-0.58	-0.68	-0.73	-0.71	-0.72
GI	0.84*	0.72	0.74	0.79*	0.88**	0.74	0.77*	0.84*	0.81*	0.82*
Viability test										
GC	0.76*	0.90**	0.80*	0.83*	0.75	0.85*	0.71	0.84*	0.79*	0.82*
Ageing tests										
RH100	0.73	0.69	0.63	0.70	0.77*	0.65	0.57	0.72	0.65	0.68
RH75	0.85*	0.88**	0.82*	0.87**	0.86*	0.82*	0.74	0.91*	0.87*	0.84*
RH43	0.76*	0.72	0.64	0.72	0.80*	0.67	0.53	0.88*	0.65	0.68
RH32	0.77*	0.89**	0.86*	0.86*	0.75	0.86*	0.84*	0.96**	0.86*	0.87*

[1]Standard germination test for onion according to ISTA. *Significant at α=0.05, **significant at α=0.01 and all the rest are not significant.

When the best predictive models were determined using stepwise comparison the germination percentage at 30°C was found to be highly significant (α=0.01) and it was best for the overall mean final emergence percentage and seedling vigour index (Figure 6).

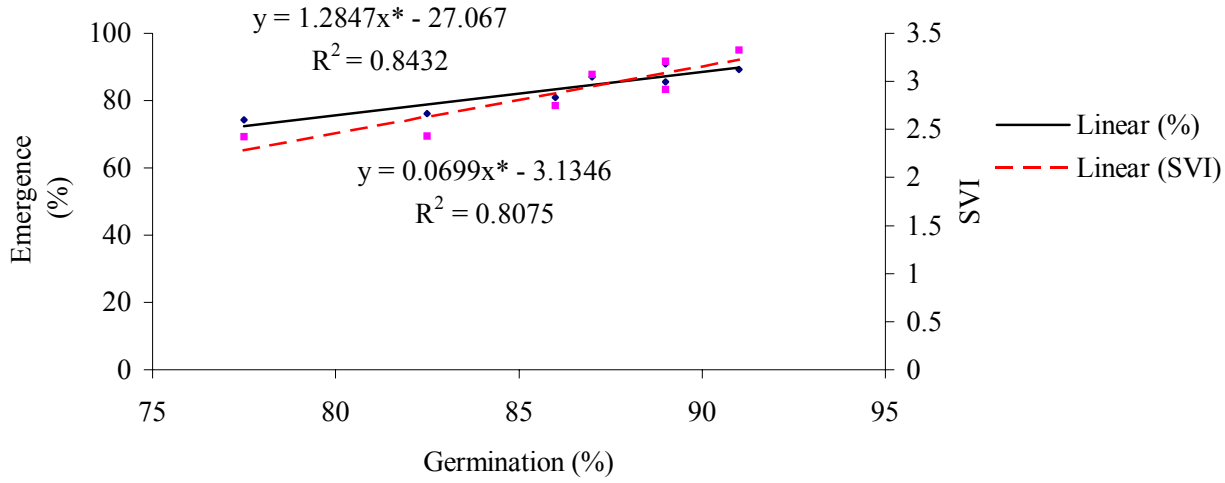

Figure 6: Best equations for predicting over all FEP (final emergence percentage) and (SVI seedling vigour index), *germination % at 30°C of onion seeds planted under a range of temperatures

5.3.3. Tomato

None of the tomato seeds emerged in winter. The analysis of variance for the two planting temperatures, two growth media and seed lots and their interaction is presented Table in A11, in the appendix. None of temperature trials revealed significant difference in percentage emergence (Table 19). On the contrary, seedlings at 15°/25°C emerged four days later and had lower seedling vigour index and seedling dry mass as compared to seedlings from 30°C.

The FEP (final emergence percentage) and SVI (seedling vigour index) were not significantly different whether seeds were planted in Hygromix or soil. However, there was significant MET and seedling dry mass differences among the two growth media. Seedlings emerged from the soil significantly faster than Hygromix and had a higher SVI. In contrast, the seedling dry mass of Hygromix was higher when compared to that of soil. With the exception of MET (mean emergence time), which was significant at α=0.05, all other parameters had non-significant "temperature x seed lot" interaction. The results of the two temperature emergence trials did not show any pronounced

vigour difference amongst seed lots, since these temperatures were optimum or near optimal. Even though the emergence percentage was lower compared to the SGT results, almost all seed lots had consistent performance throughout both temperature trials and growth media. This demonstrates that even poor vigour seed lots can also perform well under favourable environmental conditions (Coolbear, 1995).

Table 19: Final emergence percentage, mean emergence time, seedling vigour index and seedling dry mass of tomato seed lots planted under different temperatures and growth media

Source	Final emergence percentage (%)	Mean emergence time (days)	Seedling vigour index	Seedling dry mass (mg/plant)
Temperature				
15/25°C	85.66a[1]	16.94b	2.66b	192.49a
30°C	84.78a	10.52a	4.13a	177.57b
Growth media				
Hygromix	85.84a	14.03b	3.34a	190.78a
Soil	84.59a	13.45a	3.45a	179.30b

[1]Values in each column followed by the same letters were not significantly different.

The FEP and SVI of the eight seed lots averaged for two temperatures and two growth media are presented in Table 20. No emergence was recorded in winter, since tomato is a warm season crop. The range between the highest and the lowest values in the emergence trial was similar to the standard germination percentage. Thus, under favourable environmental conditions all seed lots, irrespective of their vigour level, performed similar to the standard germination percentage. It is also more likely if seed lots differ in germination, it will also in emergence. However, even seed lots of the same germination differ in the seedling emergence. This can be explained by seed lot difference in vigour, since seedling emergence is the effect of both seed quality and environment (Perry, 1984).

Based on the information in Table 20, the performance of different seed lots concerning SVI is similar to FEP. Seed lot C and G were ranked as first and second and seed lot H was ranked lowest at both parameters in all emergence trials. However, since the environment was near optimum for tomato the emergence at these trials did not reveal better separation of seed lots than the SGT. For comparison, the SGT results are presented in Table 20 and only seed lot H had germination percentage of below 85%. Therefore, most seed lots were considered as high germinating seed lots.

Table 20: Ranking of tomato seed lots based on the FEP and SVI recorded from various emergence trials

Seed lot	Germination (%)	Emergence (%)			Seedling vigour index (SVI)		
		15°/25°C	30°C	Mean	15°/25°C	30°C	Mean
A	91bc (3)	84.8a[1](6)[2]	84.3ab(5)	84.5b (6)	2.53cd (6)	4.17b (5)	3.35bc(6)
B	86c (7)	87.3a (4)	86.5ab(3)	86.9ab(4)	2.58bcd(5)	4.17b (4)	3.38bc(5)
C	97ab (2)	90.5a (2)	87.0ab(2)	88.8ab(2)	2.89bc (3)	4.44ab(2)	3.67b(2)
D	90c (4)	86.0a (5)	84.3ab(6)	85.1b (5)	2.73bcd(4)	4.24ab(3)	3.48bc(4)
E	88c (6)	82.8ab(7)	82.0bc(7)	82.4b (7)	2.41d (7)	4.01b (7)	3.21c(7)
F	89c (5)	89.5a (3)	86.5ab(4)	88.0ab(3)	2.94ab (2)	4.14b (6)	3.54bc(3)
G	99a (1)	92.5a (1)	94.8a (1)	93.6a (1)	3.29a (1)	4.83a (1)	4.06a(1)
H	76d (8)	72.0b (8)	73.5c (8)	72.5c (8)	1.94e (8)	3.05c (8)	2.50d(8)
Mean	89.25	85.66	84.78		2.66	4.13	
CV	2.93	5.75	5.46	4.06	6.41	6.35	4.83
F value	27.47**	6.69**	6.86**	12.61**	22.28**	14.76**	29.40**
LSD_{Tukey}	6.12	11.53	10.82	8.10	0.40	0.61	0.38
Diff	22.5	20.5	21.25	21.13	1.35	1.78	1.56

[1]Values in each column followed by the same letters were not significantly different. [2]Values in parenthesis are the ranking order of the seed lots. **Significant at α=0.01.

The standard germination percentages for seed lot B and F were 89 and 86% and the emergence percentage at 15°/25°C was 89 and 92% respectively (Table 20). It is clear from seed lots B and F that the percentage emergence of these seed lots, when planted under favourable conditions was higher than the standard germination test. Therefore, the use of GC based on radicle emerged should not be completely avoided as a test in some crops such as tomato. Wurr and Fellows (1984) reported lower standard germination than emergence of lettuce and they suggested that the reason was mostly due to fungal growth at the laboratory germination test. Coolbear (1995) reported the main problem for not showing vigour differences in the favourable planting conditions could be due to recovering of seeds from some of the damages caused during deterioration. Thus, some abnormal seedling may emerge if planted under favourable environments.

The correlation coefficients between the laboratory (germination and vigour) tests and emergence (FEP and SVI) test results of tomato seed lots are shown in Table 21. There was no significant difference between the two growth media and for calculating the correlation the average results of the media was taken. Seed lot H was excluded from calculation correlation (r) because its relatively low germination percentage (less than 80%) might affect the value of r. Matthews (1980) suggested application of vigour only for seeds that have germination percentage above the minimum. Germination from standard and non-standard temperatures correlated with both emergence parameters. Especially, the germination percentage at 20°C had highly significant correlation with the all FEP and SVI results. According to Demir and Samit (2001) germination at 18°C was found as the best vigour test in separating seed lots of cotton and had a good correlation with emergence of tomato.

Table 21: Correlation coefficients between laboratory test and emergence test results of tomato seeds

Laboratory tests	Emergence (%)			Seedling vigour index (SVI)		
	15°/25°C	30°C	Mean	15°/25°C	30°C	Mean
Germination %						
20°C	0.96**	0.95**	0.98**	0.87**	0.91**	0.92**
20°/30°C[1]	0.83*	0.82*	0.84*	0.76*	0.90**	0.86**
30°C	0.86*	0.82*	0.86*	0.84**	0.93**	0.91**
Germination rate						
GE	0.61	0.59	0.62	0.54	0.74	0.66
MGT	-0.37	-0.43	-0.42	-0.31	-0.61	-0.47
GI	0.57	0.61	0.61	0.49	0.73	0.64
Viability test						
GC	0.73	0.79*	0.79*	0.63	0.81*	0.74*
Ageing tests						
RH100	0.93**	0.84*	0.91**	0.97**	0.86**	0.95**
RH75	0.91**	0.88**	0.92**	0.80*	0.80*	0.83*
RH53	0.91**	0.88**	0.92**	0.83*	0.93**	0.90**
RH12	0.97**	0.89**	0.95**	0.89**	0.87**	0.91**

[1]Standard germination test for tomato according to ISTA. *Significant at α=0.05, **significant at α=0.01 and all the rest are not significant.

Percentage viability had no correlation with 15°/25°C emergence trial. Moreover, none of the three germination speed characters (GE, MGT and GI) had significant correlation with any of the emergence trials. The correlations between the results of all germination percentages after ageing using different relative humidity and the emergence trial results (FEP and SVI) are significant. The overall FEP and SVI had significant correlations with all AA test results. Panobianco and Marcos-Filho (2001) suggested that both the standard and SSAA test using NaCl could be used effectively in detecting vigour differences among seed lots of tomato. There was highly significant correlation between germination percentage after ageing with RH32 and all emergence trials (FEP and SVI). The best predictive equation for both FEP and SVI was the germination percentage at the standard (RH100) accelerated-ageing test (Figure 7).

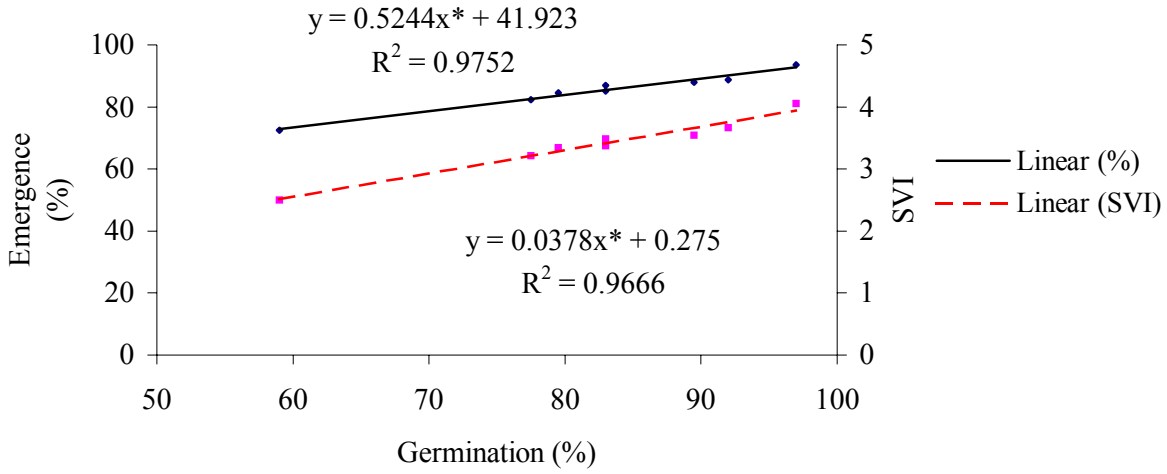

Figure 7: Best equations for predicting over all FEP (final emergence percentage) and SVI (seedling vigour index); *germination percentage after the standard AA test of tomato seeds planted under a range of temperatures

5.4. Conclusions

Testing a seed lot over a range of environmental conditions may provide a more reliable estimate of how the seed lot will perform over a range of germination environments as was done in this chapter than taking data resulted from a single extreme environment. The data obtained from the current experiment indicates that temperature was an important factor-influencing emergence. A wide range of emergence values were revealed in each trial, the poorest and slowest emergence occurred in the winter trial. Emergence also varied between seed lots and the highest was observed in favourable

temperature and the values decreased under stressed environmental conditions. These differences in the emergence were indicative of seed vigour difference among seed lots.

There was no significant difference in terms of percentage and rate of emergence between the two growth media (Hygromix and soil) used in this study. Therefore, soil can be used as an alternative medium for raising seedling transplants in places where commercial growth media are not accessible. However, since larger seedlings were harvested from Hygromix than soil, it needs careful assessment in terms of all cultural practices such as fertilization and irrigation. These results could also be applicable for direct seeding of these vegetables in soil.

Differences in vigour of seed lots were reflected in their emergence in the seedling trays. Seeds with lower vigour emerged slower. The emergence of low vigour seeds also spread over a longer time as evidenced by low seedling vigour index. Irrespective of temperature (winter or controlled) and growth media (Hygromix or soil), this study revealed that it is important to determine quality of a seed lot before planting. If not, using a low vigour seed lot may endanger the success of producing more uniform and vigorous seedlings. Therefore, choosing a high vigour seed lot may yield uniform and satisfactory number of transplants.

As it is observed from the values of the correlation coefficients, there were many significant values between different germination/vigour test and emergence test results. The best vigour tests that can predict emergence over wider range conditions (temperature and growth media) are different for different crops. For cabbage the best predictors were germination at 10°C and SSAA test using $MgCl_2$ (RH32), for onion the SGT at 15°C, SSAA test using NaCl (75%RH) and $MgCl_2$ (32%RH) and for tomato germination at 20°C, and all ageing tests (standard and SSAA).

Most laboratory tests were found to be good vigour tests for tomato as compared to cabbage and onion seeds. This was due to failure of tomato seeds to emerge in the winter trial. Therefore, it cannot be said that a wide ranges of environment were used in tomato. Finally, the standard germination test and the germination capacity (viability) results should not be ignored at all, because, though they had lower values they showed good correlations, particularly with the favourable emergence trial.

CHAPTER 6
IMPROVING VIGOUR OF ONION SEEDS THROUGH PRIMING

6.1. Introduction

Farmers demand high quality commercial seed lots when planting directly into the field. Usually the germination capacity of onion seed lots should be high to be competitive on the seed market. In addition to seed lots having a high germination capacity, it is also very important that they have uniform field emergence, since this improves plant establishment and uniformity of the crop at harvest. Seeds should, therefore, germinate and emerge quickly and uniformly throughout the field so that light, water and soil nutrients may be utilized at optimum level with maximum efficiency. If crops emerge and grow slowly after germination, they often become stunted (Benjamin, 1990). Such plants are usually more susceptible to damage by pests and diseases, and they produce less yield. Thus, starting with a vigorous seedling is of crucial importance.

For crop production, the seedbed environment may not always be conducive for rapid germination and seedling growth. This could be due to physical stress such as temperature extremes, moisture surplus or scarcity, salinity and soil crusting, and biological stresses including insects and pathogens. In addition to seed quality, these stresses can severely affect the plant at early stand establishment (Ali *et al*., 1990). Uniform emergence is the product of high vigour seeds; while erratic and slow emergence reflects low vigour seed. To avoid unpredictable effects of field conditions on emergence of seeds differing in vigour, growers preferably produce vegetable seedling transplants in greenhouses. These seedlings are produced in trays and growing conditions are well controlled to ensure rapid, uniform and high percentage emergence. However, most of the time seedling transplants are not uniform and this reduce the success of producing vegetable seedling transplants in greenhouses (McDonald, 2000). Variation in time of seedling emergence has a large effect on the size of individual plants and this in turn greatly influences the outcome of the subsequent competition between individuals for growth resources (Benjamin, 1990).

Onion seeds are short-lived and most seed lots show reduced germination within one year. This implies that a farmer will have to sow 2.5 kg ha^{-1} low quality seeds to produce 250,000 plants instead of only one kg ha^{-1} of high quality seed (Stumpf *et al*., 1996). To minimize this effect, growers started to raise seedlings using a flat bare root system but this has its own problems such as

difficulty in separation and resulted in deterioration of seedlings during transplanting. To avoid the problem of bare root transplants, it was replaced by using seedling transplant plugs. In order to attain the recommended plant population and to grow more seedlings in a single greenhouse, all the seeds in each tray cell must germinate rapidly and uniformly (McDonald, 2000).

Onion seeds are slow to germinate and emerge after sowing and the rates depend greatly on temperature. Heydecker and Coolbear (1977) reported that out of 31 common vegetable seeds, onion seeds ranked as the 29th slowest germinating crop. This results in smaller seedlings and plants, which are more vulnerable to soil borne diseases. The extended emergence periods predispose the planting bed to increased soil compaction (Ellis, 1989). This slow and uneven germination reduces the benefits of the present day highly mechanized agricultural farming system. This requires rapid, uniform and high number of emergent seedlings, because the time taken by seeds to germinate and seedlings to establish affect the total amount and quality of marketable yield and maturity (Kim, 2000).

Low temperature can inhibit or delay emergence and increase exposure to biological and physical stresses. Priming can reduce or overcome these stresses by improving stand establishment and seedling vigour as well as reducing the vulnerability to soil crusting and damping off (Murray *et al*., 1992).

Priming is defined as a technique of controlling hydration of seeds to a level that allows pre-germinative metabolic activities to carry on but that prevents radicle protrusion from the seed coat (Bradford, 1986). Seed priming has been successful in improving seed vigour of many vegetable and agronomic crops, leading to rapid and uniform germination and seedling emergence. It can improve vigour especially under adverse conditions such as low/high temperatures, reduced water availability and salinity (Khan, 1992; Parera & Cantliffe, 1994; McDonald, 2000). For onion growers, a simple method of seed priming for maintaining or enhancing seed vigour was found to be highly beneficial (Parera & Cantliffe 1994).

The aim of this experiment was to investigate whether priming can improve vigour of onion seed lots and the effect of priming on the vigour of onion seed lots with different germination percentages.

6.2. Materials and Methods

6.2.1. Seed source

Three seed lots of onion Cultivar "Red Creole" (A, B and C) which are the same as mentioned in Chapter 3 were used. To prevent fungal growth during priming the seed lots were treated with 5% Sodium Hypo-Chloride for five minutes.

6.2.2. Priming treatment

The priming and germination experiments were conducted at the Department of Botany. For the priming treatment, sixty seeds from each seed lot were placed in a 90 mm diameter Petri dish containing a double layer of filter paper (Whatman # 1). Seed lots were moistened with 5 ml of polyethylene glycol (PEG-6000). The solutions were chosen from results of experiments conducted to determine the optimal priming concentration by Ali *et al.* (1990). The PEG solution was prepared by dissolving 342 g of PEG in 1000 ml of distilled water corresponding to –1.5 MPa water potential at 15°C as described by Michel (1983). The formula is given below in equation 9.

$$\Psi = 0.130\,[PEG]^2\,T - 13.7\,[PEG]^2 \qquad (9)$$

Where; PEG is the concentration of PEG expressed as grams per litre of water, and T is the temperature in °C. The calculated water potential is in Mega Pascal (MPa).

To maintain the osmotic potential constant at –1.5 MPa, the solutions were changed every 48hrs and evaporation was reduced by sealing the Petri dishes properly with Parafilm. The four replicates of sixty seeds per Petri dish were arranged in a completely randomised design in a Labcon growth chamber at 15°C for seven days, kept in darkness. The duration of priming was seven days because previous reports found that to be the optimal duration for onion priming (Ali *et al*., 1990; Gray *et al.*, 1990; Gray *et al.*, 1991). The priming treatments were designed in such a way that all the germination tests could start at the same time.

6.2. 3. Seed drying

Drying is a necessary pre-requisite for safe storage and handling of the primed seed. High moisture content during storage encourages fungal growth. Therefore, to increase the life span, primed seeds must be dried to a low moisture content (mostly to their original moisture content prior to storage)

(Khan, 1992). Dried seeds have also an advantage over fresh seeds in the ease of handling during sowing either using machinery or manually.

As a result, seeds were washed under running tap water for five minutes and then left to dry at room temperature. After priming was completed seeds were either surface-dried for two hours or dried-back to their original moisture content. To check if primed seeds reached their original moisture content, the mass of four replicates of 100 control seeds was taken. Primed seeds were weighed frequently until it reached the same mass as the control seeds.

6.2.4. Germination test

For the germination test, the primed seeds that were dried for two hours (surface-dried) were used. Fifty primed and non-primed (control) seeds were placed in 9 mm diameter Petri dishes containing two layers of filter paper (Whatman #1). When necessary the filter papers were re-moistened with distilled water. The tests were carried out under the following constant temperatures: 10°, 20° and 30°C in light.

Germination was recorded daily for twelve days and counts were made when the radicle became visible. The final count for the number of normal seedlings was recorded at day 12 and the final germination percentage of normal seedlings (FGP) was calculated according to ISTA rules (1999). The GC, MGT and GE were calculated according to the equations (1-3) used in Chapter 3. The time spread of germination (TSG) was also calculated as the log variance of the mean germination times (Orchard, 1977).

6.2.5. Emergence test

The seedling emergence of onion seeds is slow and the rate of seedlings emergence depends greatly on temperature (Brewster, 1994). To evaluate the effect of priming on optimal and sub-optimal temperatures, two separate seedling emergence trials were performed in the open field from July - October 2003 at the Hatfield Experimental Farm. One experiment was conducted during the winter (cold environment) and the other during spring (favourable environment). In the first experiment, seeds were planted at the 4th of July, and in the second experiment at the 30th of August. The air temperature was recorded by the weather station on the farm and the minimum, mean and maximum

temperatures are shown in Table 22. The second experiment was selected as being favourable, since the optimal range of temperature for onion emergence is from 13° to 28°C (Brewster, 1994).

Table 22: Minimum, mean and maximum temperatures of the two emergence trials

Experiment	Planting conditions	Minimum	Mean	Maximum	Planting date
First (winter)	Cold	6.1°C	12.9°C	19.8°C	July 4, 2003
Second (spring)	Favourable	13.7°C	20.6°C	27.9°C	August 30,2003

For the emergence test, fifty seeds each of primed and surface-dried (dried for 2 hours), primed and dried-back (dried for 48 hours) and control (non-primed) seeds were planted in seedling trays with 128 pyramidal-shaped cells. Prior to planting the trays were filled with a growing medium consisting of sand and bark at a 2:1 ratio, by volume. One seed was sown to each cell cavity at a depth of 1 to 1.5cm. A factorial design of three seed lots (A, B and C) and three treatments (primed 2hr drying, primed 48hr drying and control) were placed in a randomised design with four replicates. The trays were watered daily with equal amount of water using a pressurized hand sprayer. Trays were inspected daily and emerged seedlings were recorded at 24 hours interval until no further emergence occurred.

The FEP, MET and SVI were calculated using the same equations (6, 7, and 8) as in Chapter 5. Time spread of emergence (TSE) was also calculated as the variance of emergence times (Orchard, 1977). The seedlings were harvested four weeks (30 days) after sowing. Ten plants per treatment combination and replication were sampled at random, and the shoots were cut at the surface. Following harvesting, the shoots were dried in an oven at 65°C for 72 hours. For weighing the dry mass, an analytical scale balance (with three digits after the decimal point) was used and the ten plant samples were weighed collectively. The mean dry mass per plant was then calculated and the results are given in milligrams.

6.2.6. Statistical analysis

To improve homogeneity before analysis of variance, the energy of germination and time spread of germination data were transformed using arcsine $\sqrt{\%}$ and log respectively. The transformed data was used only to determine significance, but in all other comparisons the original data was used. Finally, all the calculated germination and emergence variables and dry mass were analysed by using the

computer based SAS program (SAS, 1999). ANOVA tables are included in the appendix and are indicated in the text by the letter A before the table number.

6.3. Results and Discussions

Most works which were reported on the past showed improved vigour traits due to priming. The enhanced vigour parameters were: 1) germination and emergence percentage, 2) rate of germination and emergence, 3) uniformity of germination and emergence and 4) seedling growth. Therefore, the effect of priming on each vigour parameter will be discussed separately.

6.3.1. Germination test

a) Germination percentage

The GC (Table 23) was not significantly different for the three onion seed lots (Table A14), nor was significantly affected by temperature and priming. All the onion seed lots used, had a high germination percentage (Table 1) resulting in high FGPs, without being significantly different. Priming did, however, improve the FGP on the seeds, while the two extreme temperatures (10°C and 30°C) caused a reduction in FGP (Table 25).

Table 23: Effect of temperature, seed lot and priming on the GC and FGP of onion seeds

Temperature	GC (%)	FGP (%)	Seed lot	GC (%)	FGP (%)	Priming	GC (%)	FGP (%)
10°C	95.5a[1]	83.1c	A	95.3a	86.9a	Primed	95.7a	89.3a
20°C	96.4a	92.5a	B	95.6a	89.4a	Control	95.4a	87.3b
30°C	95.2a	87.0b	C	96.1a	88.7a	-	-	-
Mean	95.58	88.33	Mean	95.58	88.33	Mean	95.58	88.33
CV (%)	2.48	4.21	CV (%)	2.48	4.21	CV (%)	2.48	4.21
LSD_{Tukey}	1.65	2.59	LSD_{Tukey}	1.65	2.59	LSD_{Tukey}	1.12	1.76

[1]Values followed by different letters were significantly different at α=0.05. GC (germination capacity) and FGP (final germination percentage).

For both parameters the highest percentages were recorded at the favourable temperature (20°C), even though the GC was not significantly different from the other germination temperatures (Figure 8(a)). In contrast, the interaction of "temperature x priming treatment" of the FGP was significant

(Table A14). The lowest and significant FGP was observed for non-primed seeds that were germinated at 10°C and the highest for both primed and control seeds that were germinated at 20°C (Figure 8(b)). The response to priming is temperature dependent. Priming did not improve germination at optimal (20°C) and supra-optimal (30°C) temperatures. At these temperatures (20° and 30°C), even though the seed lots attained highest vigour level, the more marked and economically beneficial is shown at sub-optimal temperature (10°C).

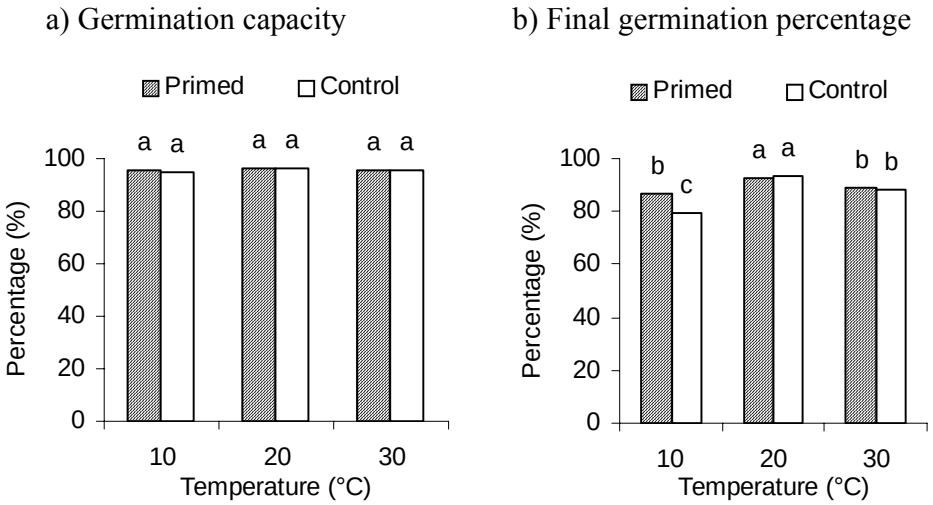

Figure 8: Effect of priming on a) germination capacity (GC) and b) the final germination percentage (FGP) of onion seeds

These results are in agreement with most studies that were conducted previously by using PEG as solutions for priming of onion (Brocklehurst & Dearman 1983a; Haigh & Barlow, 1987; Ali *et al*., 1990; Kim, 2000) even though different priming and germination temperatures were used. These results are also supported by Cantliffe (1989) who stated priming cannot bring dead seeds back to life, therefore, the germination percentage could not increase. Priming also had no effect on the percentage germination of other vegetable crops including tomato, leek, pepper and celery (Khan, 1992; Parera & Cantliffe, 1994; McDonald, 2000).

However, there are conflicting reports about priming of onion in the improvement of final radicle or normal percentage germination. Furutani *et al.* (1986) reported a reduction in germination percentage when onion seeds were primed with PEG 6000 for two days. Gray *et al.* (1991) also stated a reduction in the final radicle germination percentage after onion seeds were primed with

PEG for 7, 10 and 14 days. Similar results were also reported for various vegetable seeds (Welbaum *et al.,* 1998b). These contrasting results of increase, no effect or decrease, in percentage could be due to:

Aeration

Furutani *et al.* (1986) suggested that a reduction in germination might be due to the high viscosity of the PEG 6000 solution, which makes aeration difficult and leads to insufficient oxygen supply. Low oxygen levels may encourage anaerobic respiration in some seeds leading to the production of toxic levels of ethanol. Gray *et al.* (1991), in another experiment showed that the germination of onion seeds, which were primed in PEG 6000 was lower as compared to untreated seeds. This could be due to PEG molecules that have penetrated the seed coat and interfered with the seed internal metabolic processes. Akers and Holley (1986) found a higher germination percentage by using aerated solutions of PEG. In line with this, more benefits of aerated solution of PEG were confirmed over non-aerated solution (Bujalski *et al.*, 1989). Bujalski and Nienow (1991) introduced large scale priming by injecting bubbles of oxygen when priming larger amounts of seeds. This may have more practical advantage for seed companies and commercial farmers.

Seed quality

Murray *et al*. (1992) recommended that the difference in the percentage of germination could be due to the original differences in the quality of the seed lots. The response of a particular seed lot to priming is controlled largely by its initial quality, and this proves that the effect of priming may not improve any genetic character (Brocklehurst & Dearman, 1983a). According to Drew *et al.* (1997), 12 onion seed lots, which were primed with PEG 6000, showed clear differences in terms of the final germination percentage. A seed lot's specific response to priming might be different from that of other seed lots in view of the fact that the seed lots might have different vigour levels. For this reason, in the current experiment lower germination percentage at sub-optimal temperatures may imply that seed lot A is lower in vigour as compared to the other onion seed lots used.

Conditions during priming

For the same seed lot, the benefit obtained by priming depends on the treatment conditions such as water potential, temperature and duration. The relationship between temperature, concentration and

osmotic potential of PEG is not linear and this makes PEG priming more difficult than when salts are used (Michel, 1983). Since there is no guideline for determining the best condition for a particular species, the optimal priming treatment is determined by trial and error. However, in the present experiment the same conditions were applied to all seed lots.

Definition of germination percentage

Even though in this study, the final radicle percentage (GC) and final germination percentages (FGP) were used, only highly significant improvement was recorded for the final germination percentage at low germination temperatures. In most previous reports no improvements in germination percentage were reported (McDonald, 2000). This could be due to differences in the interpretation of "germination" since germination was defined as emerged radicle instead of normal seedlings as in the ISTA guidelines.

b) Rate of germination

There were significant differences in the MGT and GE among temperature, seed lot and priming agent (Table A15). As overall comparison, fast germination was observed for seed lot B followed by C and A (Table 24). The MGT of primed seeds was reduced by 2.4 days as compared to the control seeds, and the GE of primed seeds was increased by 21%. When the overall temperatures were compared the slowest and significantly so, germination was obtained at 10°C, as compared to 20° and 30°C.

Table 24: Effect of temperature, seed lot and priming treatment on the GE and MGT of onion seeds

Temperature	GE (%)	MGT (Days)	Seed lot	GE (%)	MGT (Days)	Priming	GE (%)	MGT (Days)
10°C	57.3[49.9][1]b[2]	5.25a	A	74.9[61.8]c	4.25a	Primed	91.2[73.9]a	2.87b
20°C	92.8[75.2]a	3.11b	B	87.4[71.0]a	3.08c	Control	70.4[59.0]b	4.51a
30°C	92.3[74.32]a	2.66c	C	80.1[66.6]b	3.74b	-	-	-
Mean	[66.46]	3.69	Mean	[66.46]	3.69	Mean	[66.46]	3.69
CV (%)	[5.60]	6.28	CV (%)	[5.60]	6.28	CV (%)	[5.60]	6.28
LSD_{Tukey}	[2.59]	0.16	LSD_{Tukey}	[2.59]	0.16	LSD_{Tukey}	[1.76]	0.11

[1]Value in the parenthesis is transformed data. [2]Values followed by different letters were significantly different at α=0.05. GE (germination energy) and MGT (mean germination time).

The "temperature x seed lot x priming" interaction of GE was highly significant (Table A15). Primed seeds showed higher germination energy than non-primed seeds at all temperatures. The highest germination energy was observed for primed seed lots that were germinated at 20°C followed by 30° and 10°C (Figure 9(a)). However, the highest difference between primed and control seeds was observed at 10°C. Even with this constraint, seed lot B attained the highest germination energy. When the difference in GEs between primed and control seeds were compared, priming of seed lot A was the most advantageous followed by seed lot C and B.

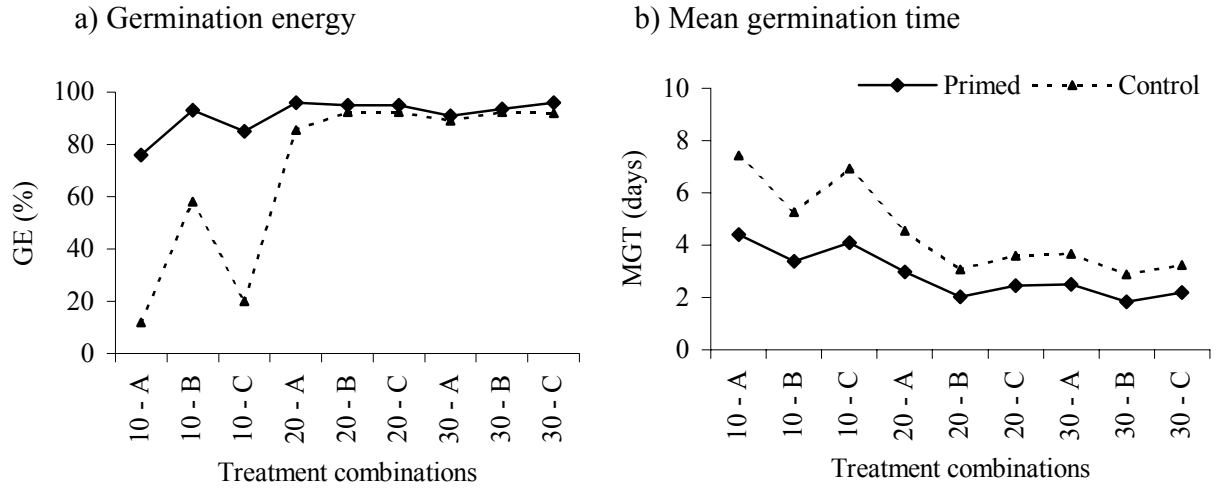

Figure 9: Effect of priming on a) germination energy (GE) and b) mean germination time (MGT), of three onion seed lots germinated at 10°, 20° and 30°C

When the germination energy was used as a parameter for comparing primed and control seeds, Goobkin (1989) reported greater germination energy for primed onion seeds after seeds were treated with potassium salts. Ruan *et al.* (2002) also found higher germination energy for primed rice seeds than untreated control seeds. For this trait, significant benefits were gained when primed seed lots were germinated at cooler temperatures. This is confirmed by the present results where the GE of primed seeds that were germinated at 10°C were significantly increased by 65, 35 and 64% for seed lots A, B and C respectively, as compared to the control seeds.

Priming accelerated germination of all three seed lots of onion (Figure 9(b)). The longest MGT was observed for non-primed seeds germinated at 10°C, while the shortest was found for primed seeds at 30°C (Figure 9(b), Figure 10). At 10°C, priming accelerated germination by 3.0, 2.0 and 2.8 days

for seed lots A, B and C respectively (Figure 10). Irrespective of type of seed lot, primed seeds germinated earlier by 2.6 days, 1.2 days and 1.0 day than control seeds when incubated at 10°, 20° and 30°C respectively. Priming significantly reduced the MGT of all seed lots at all temperatures (Figure 9(b), Figure 10). Priming reduced the germination time of the low vigour seed lot (A) more than that of the high vigour seed lot (B), since priming can repair some of the deteriorated tissue of low vigour seed lots (Welbaum *et al*., 1998a).

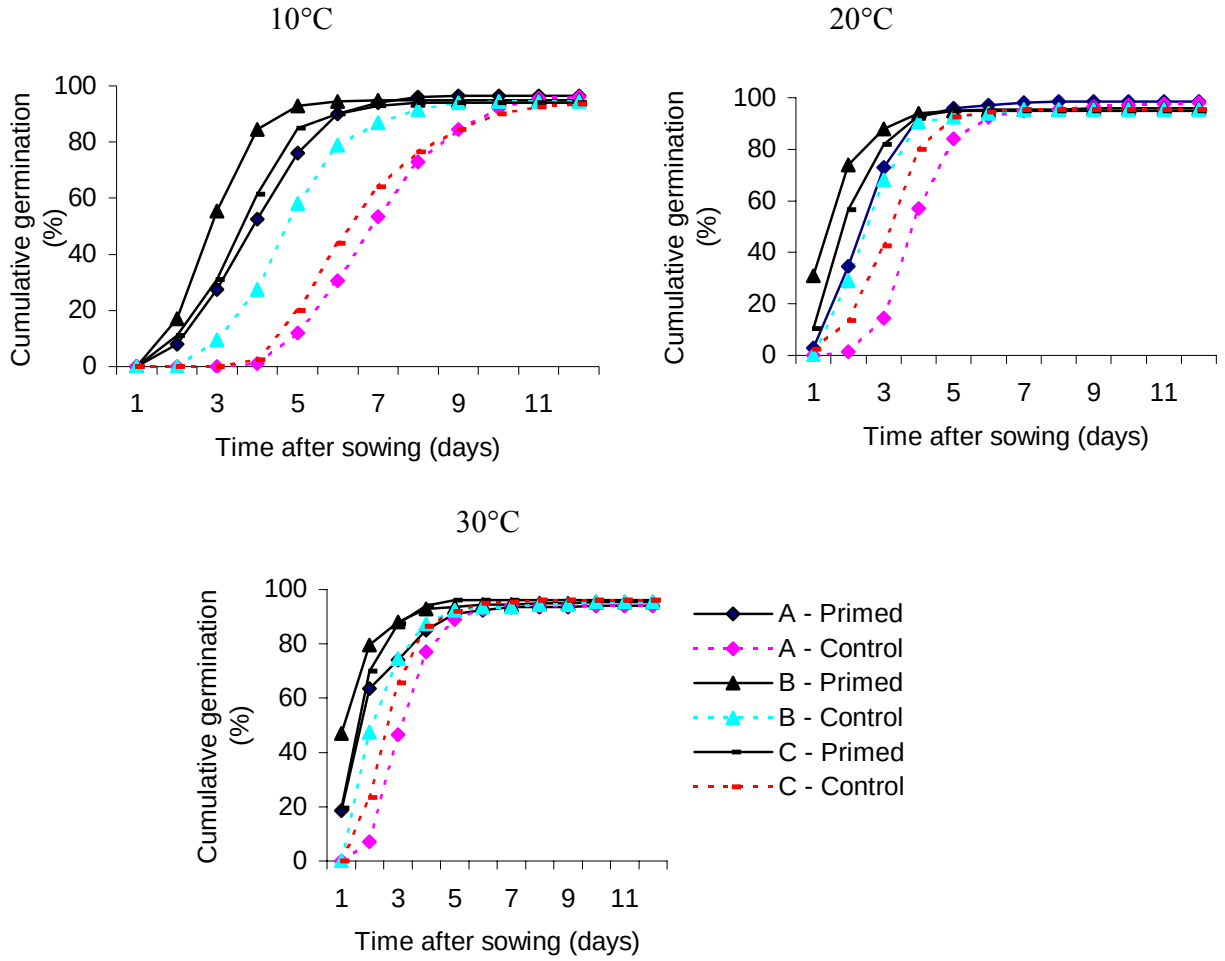

Figure 10: Effect of priming on the cumulative radicle germination percentage of three onion seed lots germinated at 10°, 20° and 30°C

c) Uniformity of germination

Time spread of germination (TSG) is expressed as $\log_e$ (variance), where the variance is the square of the standard deviation of the mean germination times. A low value of $\log_e$ (variance) indicates a narrow time spread of germination (more uniform). There were significant differences among temperature, seed lot and priming treatment in terms of TSG (Table A15). All seed lots were significantly different among one another. More uniform germination was observed from seed lot B, primed seeds, that were germinated at 30°C. Irrespective of priming treatment and temperature, seed lot B had the narrowest spread of germination time followed by seed lot C and A (Table 25).

Table 25: Effect of temperature, seed lot and priming on the TSG [Log_e (variance)] of onion seeds

Temperature	TSG	Seed lot	TSG	Priming	TSG
10°C	0.26[1.56][1] a[2]	A	-0.22[1.11] a	Primed	-1.066[0.43] b
20°C	-0.73[0.59] b	B	-0.90[0.56] c	Control	0.011[1.28] a
30°C	-1.12[0.40] c	C	-0.47[0.89] b	-	- -
Mean	[0.85]	Mean	[0.85]	Mean	[0.85]
CV (%)	[13.57]	CV (%)	[13.57]	CV (%)	[13.57]
$\text{LSD}_{\text{Tukey}}$	[0.081]	$\text{LSD}_{\text{Tukey}}$	[0.081]	$\text{LSD}_{\text{Tukey}}$	[0.055]

[1]Value in the parenthesis is the transformed data. [2]Values followed by different letters were significantly different at α=0.05. TSG (time spread of germination).

There were significant interactions among temperature, seed lot and priming treatment for TSG (Table A15). However, seeds that were primed had significantly lower TSG's than that of control seeds at all germination temperatures. Irrespective of temperature and seed lot, priming significantly reduced the time spread of germination (Table A15). Thus, all primed seed lots showed more uniform germination than the control seeds at all temperatures (Figure 11). The statistical analysis for this parameter at different temperatures showed that whether primed or not, seeds had a significantly narrower spread of germination time at higher germination temperatures. The narrowest TSG was observed from primed seeds that were germinated at 30°C followed by 20°C and the widest at 10°C. Brocklehurst and Dearman (1983b) reported a reduction of time spread when onion seeds were primed with PEG-6000. Bujalski and Nienow (1991) also found a lower TSG from primed seeds than that of untreated seeds when seeds were primed using filter paper. Therefore, these results conform that priming can lead to more uniform germination.

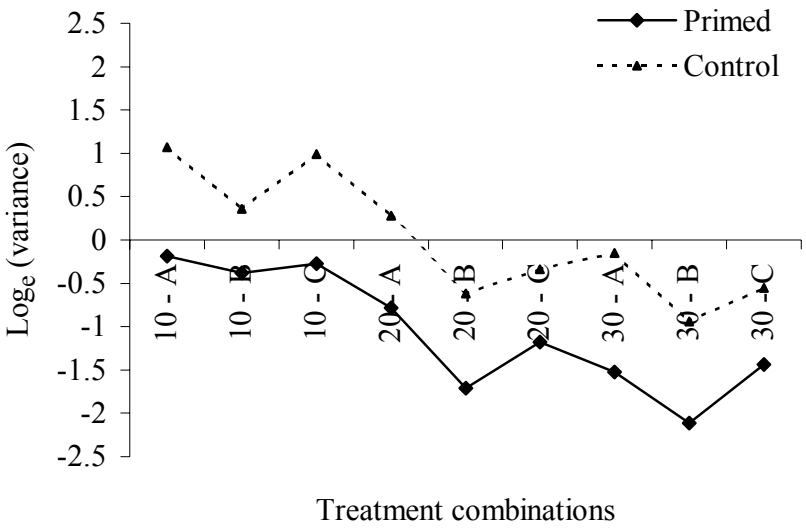

Figure 11: Spread of germination time for primed and control seeds of three onion seed lots germinated at 10°, 20° and 30°C

6.3.2. Emergence test

a) Emergence percentage

The analysis of variance for the final emergence percentage (FEP) is shown in Table A16. Seeds that were planted at favourable temperatures had a greater emergence percentage than under cold conditions (Table 26). Neither seed lots nor priming treatment were significantly different among one another. However, though not significant in both conditions, seed lot C had the highest FEP followed by seed lots B and A. Drying primed seeds had no significant effect on the final emergence percentage, whether seeds were planted at low or suitable temperatures.

The present results are in agreement with that of Haigh *et al.* (1986) who reported no effect on the emergence percentage after primed tomato and carrot seeds were dried for 28 days. Priming also had no effect on the FEP of seeds planted at a minimum temperature of 10°C (Brocklehurst & Dearman, 1983b). Drying back primed seeds had no effect on emergence percentage of onion seeds when seeds were dried for 48 hours at 15°C to moisture content of 10.4 %, but reduced the emergence percentage of carrot and celery seeds (Brocklehurst & Dearman, 1983b). On the contrary, Haigh *et al.* (1986) reported a reduction in the final field emergence of onion seeds when the primed seeds were air-dried for one day as compared to unprimed ones. Similarly, under harsh low temperature

field conditions, primed maize seeds dried at 15°C and 20°C had significantly lower final emergence percentage than those dried at higher temperature and non-primed seeds (Parera & Cantliffe, 1992).

Table 26: Effect of temperature, priming and drying on the FEP (%) of onion seeds

Seed lot	Cold	Favourable	Priming	Cold	Favourable
A	80.0a[1]	91.7a	Control	81.0a	92.8a
B	81.8a	92.0a	Surface-dried	82.5a	92.18a
C	83.0a	93.3a	Dried-back	81.3a	92.0a
Mean	81.6	92.3	Mean	81.6	92.3
CV (%)	4.26	4.42	CV (%)	4.26	4.42
LSD_{Tukey}	3.52	4.13	LSD_{Tukey}	3.52	4.13

[1]Values followed by different letters were significantly different at α=0.05.

b) Rate of emergence

The ANOVA of MET (mean emergence time) and SVI (seedling vigour index) of the two planting conditions are shown in Tables A17 and A18 respectively. At both conditions the shortest MET was observed in seed lot B followed by seed lots C and A. Nevertheless, even though there was highly significant difference between seed lots under favourable conditions, the difference was only 0.6 days, which has no practical implications. It can, therefore, be suggested to rather select a seed lot with a high germination than the one with the shortest MET, since one day difference in emergence may not have that much of an impact on later development.

Priming had decreased the MET of all seed lots as compared to the control at both planting conditions. Even though the highest reduction was observed when seeds were dried for two hours. Primed and surface-dried seeds reduced the MET by 3.9 and 1.4 days for cooler and favourable conditions respectively. Whereas, primed and dried-back seeds reduced the MET by only 1.7 days at cooler and by 0.8 days at favourable conditions (Table 27). Bujalski *et al.* (1989) reported 1.5 days earlier emergence from seeds that were surface-dried as compared with primed seeds dried to their original moisture content. Primed and dried seeds emerged faster by one day than untreated seeds (Khan, 1992).

Table 27: Effect of temperature, priming and drying on the MET and SVI of onion seeds

Cold conditions					
Seed lot	MET (Days)	SVI	Priming treatment	MET (Days)	SVI
A	16.17a[1]	2.67b	Control	17.25a	2.39c
B	14.44c	2.94a	Surface-dried	13.36c	3.18a
C	15.51b	2.67b	Dried-back	15.51b	2.71b
Mean	15.37	2.76	Mean	15.37	2.76
CV (%)	3.39	5.39	CV (%)	3.39	5.39
LSD_{Tukey}	0.53	0.15	LSD_{Tukey}	0.53	0.15
Favourable conditions					
A	11.63a	4.07a	Control	12.08c	3.89c
B	11.01b	4.23a	Surface-dried	10.69a	4.35a
C	11.39a	4.08a	Dried-back	11.26b	4.14b
Mean	11.34	4.13	Mean	11.34	4.13
CV (%)	2.60	4.59	CV (%)	2.60	4.59
LSD_{Tukey}	0.30	0.19	LSD_{Tukey}	0.30	0.19

[1]Values followed by different letters were significantly different at α=0.05. MET (mean emergence time) and SVI (seedling vigour index).

Under sub-optimal (winter) conditions, the effect of priming on the MET was by far greater than under favourable conditions (Table 27). Brocklehurst and Dearman (1983b) confirm that when onion seeds were planted at low temperatures, the mean seedling emergence time was significantly reduced through priming, by up to 8.8 days. Similarly, Brewster *et al.* (1991) reported a six days earlier emergence from primed onion seeds than non-primed seeds. According Murray *et al.* (1992) and Khan (1992) primed seeds emerged faster than non-primed seeds by two days when seeds were planted at relatively favourable temperature (15°C).

At both planting conditions, seed lot B had the highest SVI but it is only significantly different from the other seed lots under cold conditions (Table 27). No significant SVI's were observed among the seed lots at favourable planting conditions even though the highest SVI was observed from seed lot B. Perry (1984) also reported no significant vigour differences among high germinating seed lots when seeds were planted at more conducive conditions. Seeds that were planted at favourable conditions revealed higher SVI's as compared to lower temperature conditions, but when the

difference was compared the biggest difference was observed between seed lots planted under cold conditions (Table 27). In both planting conditions, priming increased the SVI of all seed lots, especially when seeds were primed and surface dried. Irrespective of drying, primed seeds had significantly higher SVI's than non-primed seeds at both planting conditions. When seeds that were primed and dried for two hours and primed and dried for two days were compared, seeds that were dried for the shorter period had significantly higher SVI's than primed seeds dried for relatively longer period of time. Similar results were reported by Ruan *et al.* (2002) after rice seeds were primed using $CaCl_2$.

The delayed time in emergence of seeds that were dried to their original moisture content would be mostly due to the time needed for re-imbibitions. Much of the improvement was lost after drying when seeds were primed using filter paper. The delay in germination following drying is typically the time required for re-imbibitions by the seeds as reported by previous workers (Brocklehurst & Dearman, 1983b). Brocklehurst *et al.* (1987) suggest that the improvement due to priming was as a result of imbibitions of water alone and no physiological modification of the tissue was observed.

c) Uniformity of emergence

Uniform emergence is important for ensuring bulb development to the right size, since a widely spread emergence creates difficulties in deciding the optimal application time of cultural practices, spraying of pesticides and harvesting (Brewster, 1994). Therefore, for successful onion production, uniform emergence is very important. Uniform emergence is also essential for raising seedlings and allows accurate time of transplantation and increased market value of the seedlings as they have good and uniform look (McDonald, 2000). In both planting conditions, the time spread of emergence (TSE) of seeds was significantly different among seeds lots and priming treatments (Table A19).

Table 28: Effect of temperature, priming and drying on the TSE [Log_e (variance)] of onion seeds

Seed lot	Cold	Favourable	Priming	Cold	Favourable
A	2.54a[1]	2.19a	Control	2.85a	2.22a
B	2.42b	2.06b	Surface-dried	2.26c	2.05b
C	2.53a	2.12ab	Dried-back	2.39b	2.10ab
Mean	2.50	2.12	Mean	2.50	2.12
CV (%)	4.05	5.46	CV (%)	4.05	5.46
LSD_{Tukey}	0.10	0.12	LSD_{Tukey}	0.10	0.12

[1]Values followed by different letters were significantly different at α=0.05.

The lowest and significantly so, TSE was observed from seed lot B followed by seed lots C and A (Table 28). At both conditions seed lot B emerged more uniformly than seed lots A and C. The TSE was used as a vigour test parameter and the seed lots with a relatively lower value are considered as more vigorous as compared to seed lots with higher values (Copeland & McDonald, 2001). As a result, the shortest time spread was for seed lot B, this implies that seed lot B is more vigorous than seed lots C and A.

There was highly significant difference among priming treatments at both planting conditions. Under cooler conditions, primed seeds that were planted after two hours of drying, had a significantly shorter time spread of emergence than primed seeds and dried-back for 48 hours, and the control seeds (Table 28). At both plantings, the lowest TSE was observed from primed seeds that were dried for two hours, followed by primed and dried-back, and then the control seeds. Consequently, all primed seeds emerged more uniformly than the control seeds. Similar results were reported by Brocklehurst and Dearman (1983b) under the same conditions using PEG-6000. Gray *et al.* (1991) also reported reduced TSE when onion seeds were primed with different PEG concentration (PEG 600 to 8000) and surface-dried at room temperature. Similarly, Haigh *et al.* (1986) observed about two-third reduction in time spread of emergence of onion seeds due to priming as compared to that of untreated seeds and the maximal enhancement was at lower temperatures. Therefore, the present results reconfirmed that priming has more advantages in yielding more uniform emergence of onion seedling at low temperature growing conditions.

d) Seedling growth

The analysis of variance for seedling dry mass is available in A20, in the appendix. At both planting conditions, the seedling dry mass was not significantly different among seeds lots (Table 29). These results are consistent with that of Ellis (1989) who found no relative seedling dry mass differences among three seed lots of onion cultivar "White Lisbon" and the relative seedling growth was unaffected by seed quality. Primed and surface dried seeds had significantly higher seedling dry mass than that of control and primed and dried-back seeds only at low temperature planting condition. Priming could, however, not significantly improve the seedling dry mass under favourable conditions (Table 29). Similarly, no significant difference was revealed between control and dried-back seeds at cold temperatures. The present results are consistent with Khan (1992) where seeds were planted at 10/20°C resulting in a higher top fresh mass of primed seeds than the untreated control when seedlings were harvested 15 days after sowing. Brocklehurst and Dearman (1983b) also found significantly higher fresh seedling mass for primed seeds than for the control seeds at 10°C when the seedlings were harvested after 12 days after sowing.

Table 29: Effect of temperature, priming and drying on seedling dry mass (mg) of onion

Seed lot	Cold	Favourable	Priming	Cold	Favourable
A	81.17a[1]	200.83a	Control	73.50b	199.50a
B	82.92a	206.58a	Surface-dried	95.08a	207.42a
C	81.67a	201.17a	Dried-back	77.17b	201.67a
Mean	81.92	202.86	Mean	81.92	202.86
CV (%)	11.37	5.21	CV (%)	11.37	5.21
LSD_{Tukey}	9.4	10.70	LSD_{Tukey}	9.4	10.70

[1]Values followed by different letters were significantly different at α=0.05.

The relative growth of onion seedlings is not influenced by priming (Brocklehurst & Dearman, 1983b). This experiment confirmed that even though priming had increased the seedling dry mass, the higher dry mass that was observed from primed seeds is only due to the effect of priming on MET. Since primed seeds emerged faster, they are also expected to have a bigger plants, especially at the early vegetative growth stage. Therefore, the effect of priming on seedling mass is the result of the initial faster emergence time.

Benefits to plant stand include earlier, more even and healthier stand establishment leading to improved crop uniformity. This would allow for seeding rates to be reduced by 10% and improve weed management. Transplant production with primed seeds results in higher percentage of transplants with a reduced cycle time (Kim, 2000). Early emergence of onion seedlings resulted in larger seedlings, earlier maturity increased marketable and total bulb mass as compared to seedlings that emerged one week later (Murray *et al*., 1992). Therefore, seed priming may enhance bulb yield of onion if priming resulted in earlier seedling emergence.

6.4. Conclusions

Seed priming did improve some seed vigour properties leading to ***rapid*** and ***uniform*** germination and emergence. when the differences of the primed and control seeds were compared, it was clear that priming is more advantageous for low vigour seed lot (seed lot A) than high vigour seed lots (seed lot B). The present results demonstrated that the performance of onion seed lots was improved through priming especially if the seed lot has a relatively low vigour. However, the germination percentage must not be commercially unacceptable since priming can only repair physiologically damaged seeds but cannot transform dead seeds in to viable seeds.

Priming improved most characteristics of vigour of all seed lots at all temperatures even though the most prominent improvement of priming on germination and emergence was recorded at lower temperautre (10°C). Nevertheless, the enhancement was revealed better for germination than for emergence. Drying primed seeds to their original moisture content has slowed emergence as compared to the surface-dried seeds. As a result, it is advisable to plant primed onion seeds after surface drying as this improves handling and mechanical sowing without decreasing the added benefits of priming.

CHAPTER 7
IMPROVING VIGOUR OF ONION SEEDS UNDER SALINE CONDITIONS

7.1. Introduction

Salinity is one of the most severe problems in plant production, since high levels of salts in solution inhibit the growth and development of plants. The effects differ depending on climate, soil water, and type of plant and the plant's stage of development. Saline soils are soils that contain large amounts of soluble salts of Na, Ca and Mg with chloride, sulphate and bicarbonate. Out of these salts, NaCl salinity is mostly common in arid and semi-arid areas of Africa (Szabolcs, 1994). Soils are considered saline if their electrical conductivity (EC) exceeds 4 mS cm^{-1}.

Onion is an important vegetable crop, which does not grow very well under saline conditions (Mangal *et al.,* 1989). Very little genetic variation for salinity has been found, despite with numerous cultivars tested. Generally salt tolerance is high at germination, but very low during seedling growth while it increases again when the plants reach the three to five leaf stage. Salinity reduces bulb diameter, bulb mass, root growth, plant height, and number of leaves per plant (Wannamaker & Pike, 1987). Onion may mature a week earlier but with reduced yield when grown under saline conditions.

The emergence force of seedlings can be reduced by an increase in soil water salinity, hence increasing the time required to exert the maximum force (Benjamin, 1990). Seed priming was found to reduce the mean time of germination and emergence and increase stand uniformity of onion in non-saline areas (Haigh *et al*., 1986; Khan, 1992). Priming also enhanced germination and stand establishment of tomato and asparagus (Pill *et al.,* 1991), soybean (Umezawa *et al*., 2000), sugar beet (Ghoulam & Fares, 2001) and canola (Zheng *et al.,* 1998) seeds under saline conditions. Priming may have potential to alleviate the problems experienced with salinity in onion as well. Thus, the objective of this investigation was to determine the effect of PEG and NaCl as pre-sowing treatments on the improvements of seed germination and seedling emergence of onion at stressed temperature and at different salinity levels.

7.2. Materials and Methods

7.2.1. Seed source

To reduce the effect of genotype and difference among seed lots, only one seed lot of cultivar "Red Creole" was used. This seed lot was selected based on its performance in the germination and emergence trials in Chapter 6.

7.2.2. Priming treatment

PEG priming was conducted in the same way as in Chapter 6, but for NaCl priming seeds were treated using 331 g of NaCl per litre of water for seven days. The NaCl solution was prepared according to the methods used by Cochrane (1994). Both priming agents were calculated to give an osmotic potential of -1.5 MPa. After priming the seeds were dried for two hours at room temperature before they were subjected to the germination or emergence tests.

7.2.3. Germination test

Germination tests were conducted on four replicates of 50 seeds each. Primed and control seeds were transferred in Petri dishes with double layers of Whatman #1 filter paper moistened with 5 ml of different salinity levels 0 (distilled water), 100, 200 and 300 mM NaCl resulting in electrical conductivities (EC) of 0, 9.1, 17.5 and 25.7 mS cm^{-1} respectively. EC was calculated using equation 10 (Szabolcs, 1994).

$$\text{Log } T_{salt} = 0.990 + 1.055 \log EC \qquad (10)$$

Where T_{salt} is the concentration of NaCl in mM, and EC is the electric conductivity.

The Petri dishes with seeds were incubated at 10°C in the light. This temperature was selected based on previous results (Chapter 6), since priming noticeably improved when seeds were germinated under cold stressed conditions. A seed was regarded as germinated when the radicle had pierced the seed coat and radicle emergence was scored daily for a total of 20 days. The final count for the number of normal seedlings was recorded at day 20 and the final germination percentage of normal seedlings (FGP) was calculated. The GC and MGT were also calculated according to the equations (1 and 3) used in Chapter 3 and the time spread of germination (TSG) as the log variance of the mean germination times (Orchard, 1977).

7.2.4. Emergence test

The emergence trial was conducted in a greenhouse under controlled environmental conditions and natural daylight. The details of the temperature and relative humidity can be seen in Figure 12. Seedling trays with 128 pyramidal-shaped cells were used. The trays were filled with a commercial medium consisting of sand and bark at a ratio of 2:1 by volume. One seed per cell was sown to a depth of 1 to 1.5 cm.

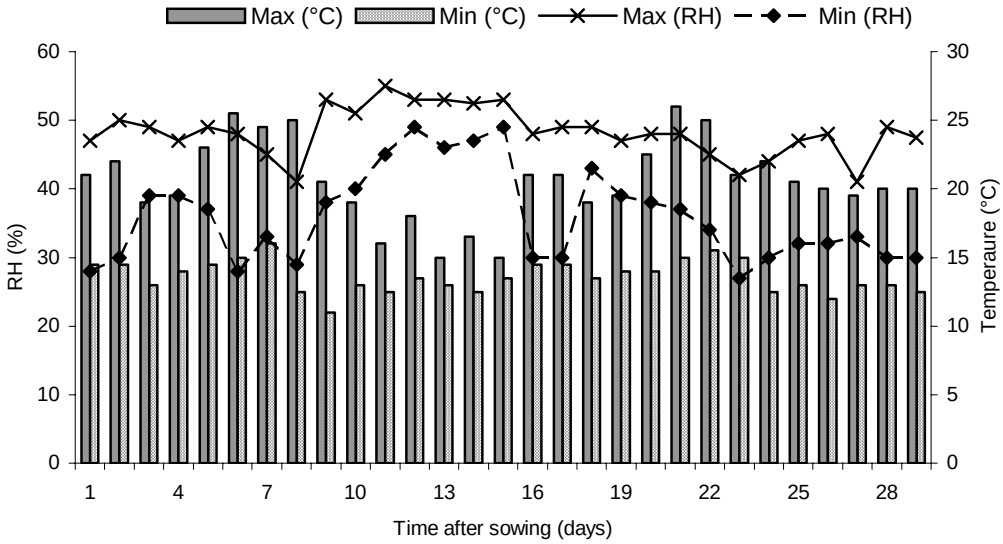

Figure 12: Temperature and relative humidity in the greenhouse during the onion emergence trial under saline conditions

The trays were watered once a day with one of four-salinity levels namely, tap water, 25, 50 and 75 mM of NaCl. After two weeks the tap water was replaced with a nutrient solution and the salinity levels were prepared by taking in to account the salinity levels of the nutrient solution. The EC of the treatments was measured using EC-meter and the values are presented in Table 30. Daily emergence counts of the number of seedlings visible above the medium surface were recorded. Plant dry mass was determined 30 days after sowing the same as Chapter 6.

Table 30: Electrical conductivity of the different salinity levels used in the emergence trial

	Salinity levels (mM of NaCl)			
	0	25	50	75
Water EC (mS cm^{-1})	0.31	3.02	4.79	7.14
Nutrient solution EC (mS cm^{-1})	2.10	4.66	6.86	8.83

7.2.5. Statistical analysis

The experimental design used for germination test was a completely randomised three (two priming treatments plus a control) x four (salinity levels) factorial design replicated four times. The emergence trial was arranged in a randomised split plot design with four blocks. The different irrigation levels represented the main plots and the priming treatments represented the sub pots. GC and FGP data were transformed using arcsine $\sqrt{\%}$ before statistical analysis. The transformed data was used only to determine significance, but in all other comparisons the original data was used. The data were analysed using the SAS software package (SAS, 1999) and LSD_{Tukey} (at α=0.05) was calculated for determining significance among treatment means. ANOVA tables are included in the appendix and are indicated in the text by the letter A before the table number.

7.3. Results and Discussions

The improvement of vigour due to priming on different parameters of germination and emergence: percentage, rate, uniformity and seedling growth under range of saline conditions are presented separately.

7.3.1. Germination test

a) Germination percentage

Highly significant differences for germination capacity (GC) and final germination percentages (FGP) were observed among the different salinity levels (Table A21). As overall analysis, the highest GC and FGP were obtained from seeds germinated using distilled water, and both variables showed a gradual decline with increasing salinity (Table 31). However, only the GC of seeds germinated at 300 mM NaCl was significantly affected, while the FGP was significantly decreased by all levels of salinity (Figure 16). According to Wannamaker and Pike (1987), the reduction in GC at high salinity levels was due to inhibition of the radicle from protruding through the seed coat. Various authors also found a linear decrease in onion seed germination as the salinity increased (Wannamaker & Pike, 1987; Miyamoto 1989; Yildirim *et al.,* 2002).

Miyamoto (1989) also reported no significant reduction in percentage radicle germination up to 27 mS cm^{-1} when seeds were geminated at 15°/25°C. But for current results, significant difference was observed when seeds were germinated starting at 17.5 mS cm^{-1} (200 mM) (Table 31). This could be

due to the low (10°C) temperature used for the germination. The interaction between salinity and priming was highly significant (Table A21) and the interactive effects can be seen in Figure 13(a). There was significant increase in GC of the primed onion seeds when they germinated at high salinity conditions (300 mM).

Table 31: Effect of salinity and priming on the GC and the FGP of onion seeds

Salinity levels			Priming treatments		
Salinity level (mM NaCl)	GC (%)	FGP (%)	Treatment	GC (%)	FGP (%)
0	94.5[77.4] [1]a[2]	85.7[68.0] a[2]	Control	73.1[61.6] b	40.4[32.3] b
100	93.8[76.0] ab	78.8[63.0] b	PEG	81.6[67.3] a	48.9[42.9] a
200	89.2[71.3] b	28.5[27.3] c	NaCl	80.9[67.0] a	56.4[46.2] a
300	36.7[36.6] c	1.2[4.5] d	-	-	-
Mean	[65.31]	[40.45]	Mean	[65.31]	[40.45]
CV (%)	[6.76]	[9.76]	CV (%)	[6.76]	[9.76]
LSD_{Tukey}	[4.85]	[4.34]	LSD_{Tukey}	[3.81]	[3.41]

[1]Value in the parenthesis is the transformed data. [2]Values followed by different letters were significantly different at α=0.05, GC (germination capacity) and FGP (final germination percentage).

Using a priming agent under control and relatively low salinity conditions (100 mM), did not significantly improve the FGP (Figure 13(b)). At medium (200 mM) to high (300 mM) salinity conditions, no normal seedlings were recorded for non-primed seeds. However, using a priming agent under these conditions could prevent total seedling loss. In agreement to these results, higher percentage of germination was reported from primed seeds of tomato (Wiebe & Muhyaddin, 1987; Pill *et al.,* 1991), sugar beet (Ghoulam & Fares, 2001) and canola (Zheng *et al.,* 1998), when seeds were germinated under cold temperature conditions using saline water.

The GC was not affected as the salinity concentration increased. However, the final normal germination percentage was lower as the level of salinity increased, and in many seeds even the radicle protruded but it failed to fulfil the prerequisite to be graded as normal seedling. The number of abnormal seedlings was higher, when seeds were germinated at higher levels of salinity.

Which priming agent to use depends on the level of salinity. Although there was no significant difference in the FGP of the two priming agents, the use of NaCl under low to medium salinity conditions could be recommended, while PEG could be recommended for use under high saline conditions. Under this saline conditions, however, only 3% normal seedlings were observed.

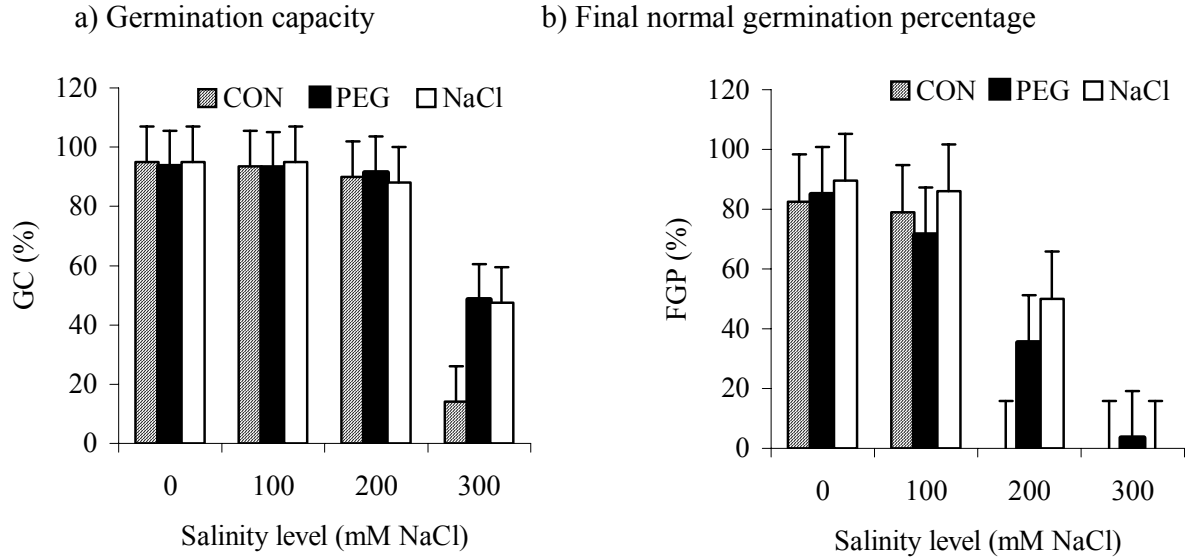

Figure 13: Interactive effects of salinity and priming on a) the germination capacity and b) the final germination percentages of onion seeds germinated under a range of salinity levels (the bar "I" is standard error (SE))

b) Rate of germination

The rate or speed of germination is given as the MGT (the reciprocal of rate of germination). In general, the MGT was significantly affected by the level of salinity (Table A22). There was a direct relation between salinity and MGT and as the level of salinity increased the MGT also increased. The fastest germination was observed when seeds were germinated with distilled water, while slowest germination occurred at highest salinity level (Table 32). Similarly, Wannamaker and Pike (1987) reported one to three day delay in germination of onion seeds as the salt concentration increased from zero to 25 mS cm^{-1}. Miyamoto (1989), also observed slower onion seed germination with increased levels of salinity.

Table 32: Effect of salinity level and priming on MGT (mean germnation time) of onion seeds

Salinity level (mM NaCl)	MGT (days)	Priming treatment	MGT (days)
0	4.91d[1]	Control	11.34a
100	7.62c	PEG	7.44c
200	10.14b	NaCl	8.12b
300	13.19a	-	-
Mean	8.97	Mean	8.97
CV (%)	4.40	CV (%)	4.40
LSD_{Tukey}	0.43	LSD_{Tukey}	0.34

[1]Values followed by different letters were significantly different at α=0.05.

There was highly significant differences in MGT among priming treatments and the interaction "salinity x priming" was highly significant (Figure 14; Table A22). The MGT of the control seeds was significantly longer than that of primed seeds (Table 32). When the two priming solutions were compared, PEG-primed seeds germinated faster than NaCl-primed seeds at all the levels of salinity (Figure 14). Pill *et al.* (1991) also reported that PEG primed tomato seeds germinated five days sooner than the untreated control seeds when seeds were germinated at 10°C. In agreement with these results, faster germination of primed seeds than non-primed seeds were reported for cucumber (Passam & Kakouriotis, 1995).

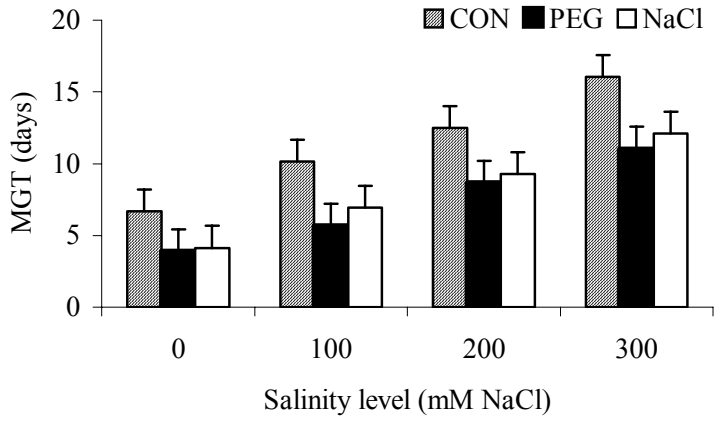

Figure 14: Interactive effects of salinity and priming on mean germination time (MGT) of onion seeds germinated under a range of salinity levels (the bar "I" is SE)

c) Uniformity of germination

The uniformity (TSG) of onion seeds was significantly affected by the level of salinity and priming agent used (Table 33) and the interaction of salinity and priming treatment was highly significant (Figure 15; Table A22). This implies that the response of onion seeds to germination under a range of salinity levels was dependent on whether seeds were primed or not. In general, significant and more uniform germination was obtained from primed than non-primed seeds, with the narrowest TSG being observed with distilled water when seeds were primed with PEG (Table 33).

Table 33: Effect of salinity level and priming on TSG (time spread of germination) of onion seeds

Salinity level (mM NaCl)	TSG $\log_e$ (variance)	Priming treatment	TSG $\log_e$ (variance)
0	0.27a[1]	Control	1.95a
100	1.14b	PEG	0.96b
200	1.78c	NaCl	1.25c
300	2.34d	-	-
Mean	1.39	Mean	1.39
CV (%)	8.84	CV (%)	8.84
LSD_{Tukey}	0.14	LSD_{Tukey}	0.11

[1]Values followed by different letters are significantly different at α=0.05.

The benefits of priming markedly reduced as the salt concentration increased to 300 mM (Figure 15). At higher salt concentrations, the germination of seeds was less uniform because seeds subjected to these salt concentrations need a longer period of time to emerge. Pill *et al.* (1991) also reported less uniform germination under stressed than favourable conditions.

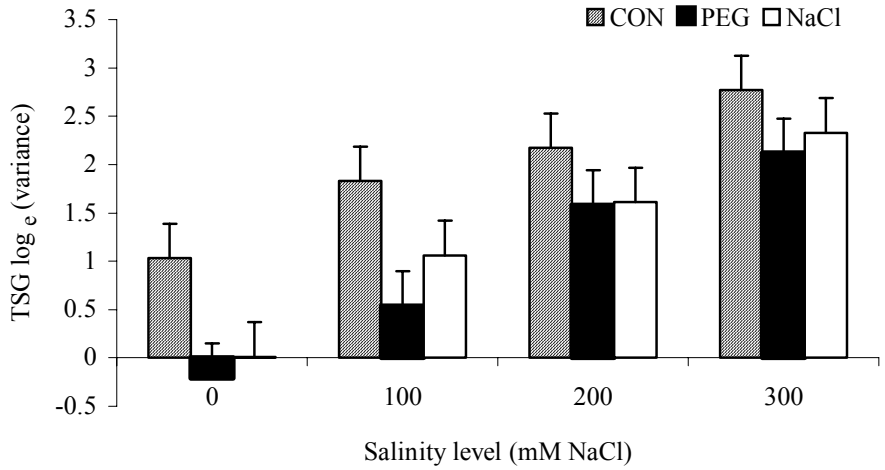

Figure 15: Interactive effects of salinity and priming on the time spread of germination (TSG) of onion seeds germinated under a range of salinity levels (the bar "I" is SE)

7.3.2. Emergence test

a) Emergence percentage

The final emergence percentage (FEP) and final survival percentage (FSP) have a similar trend. For both these parameters, the highest percentage was observed when seeds were irrigated with freshwater and the percentages decreased at higher salinity levels (Table 34; Table A23). Similar results were reported by Miyamoto (1989).

Table 34: Effect of salinity level and priming on FEP (final emergence percentage) and FSP (final survival percentage) of onion seeds

Salinity levels			Priming treatments		
Salinity (mM NaCl)	FEP (%)	FSP (%)	Treatment	FEP (%)	FSP (%)
0	85.8a[1]	83.7a	Control	76.2a	66.4a
25	82.3a	78.3b	PEG	74.9a	67.0a
50	73.7b	63.0c	NaCl	76.2a	68.8a
75	61c	44.5d	-	-	-
Mean	75.7	67.4	Mean	75.7	67.4
CV (%)	6.38	4.32	CV (%)	7.45	7.54
LSD_{Tukey}	6.17	3.71	LSD_{Tukey}	4.63	3.95

[1]Values followed by different letters were significantly different at α=0.05.

The FEP was not significantly different whether seedlings were watered with fresh water or 25 mM NaCl solution. Miyamoto (1989) reported a significant decrease in the final emergence percentage when the salt level of the irrigation water exceeds 4.9 mS cm^{-1} (50 mM of NaCl). In agreement with this, a significant reduction in FEP was observed (with the 50 mm NaCl solution) at an EC above 4.7 mS cm^{-1} (Table 34). The FSP was, however, significantly different for all salinity levels. When using irrigation water with a salt concentration of 75 mM NaCl, the FEP and FSP were reduced by 29% and 47% respectively, as compared to fresh water (Figure 16). Therefore, irrigating of onion seedlings with saline water have not only reduced the emergence percentage, but also killed off some of the seedlings. The percentage of dead seedlings progressively increased as the EC of the irrigation water increased (Figure 16) with 17% dead seedlings at the highest salinity treatment. Miyamoto (1989) found a 35% mortality rate when onion seedlings were irrigated with water at an EC of 8.9 mS cm^{-1}.

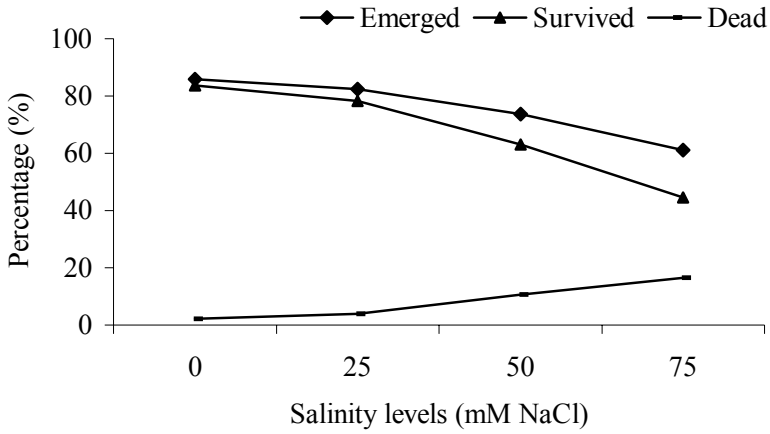

Figure 16: Percentage emergence and survival of onion seeds, irrigated with a range of salinity levels

Priming with either solution had no significant profit in improving the percentage emergence or survival of the seedlings. Similarly, Cayuela *et al.* (1996) reported no significant influence on emergence percentage of NaCl-primed tomato seeds when seeds were planted under salt stress conditions. In contrast, Sivritepe *et al*. (2003) irrigated melons with water ranging in EC from 9 to 18 mS cm^{-1} (NaCl) and reported an increased emergence percentage due to priming. Crops can thus be expected to react differently to priming.

b) Rate of emergence

The MET (mean emergence time) of seeds that were watered with different salinity levels was significantly different (Table A24). The rate of emergence was lower as the salinity concentration of the irrigation water increases; hence the highest MET was obtained from seeds irrigated with a 50 and 75 mM of NaCl solution (Figure 17). Similar results were observed in onion (Miyamoto, 1989), cucumber (Passam & Kakouriotis, 1995), tomato (Cayuela *et al.,* 1996), melon (Sivritepe *et al*., 2003) and canola (Zheng *et al.,* 1998).

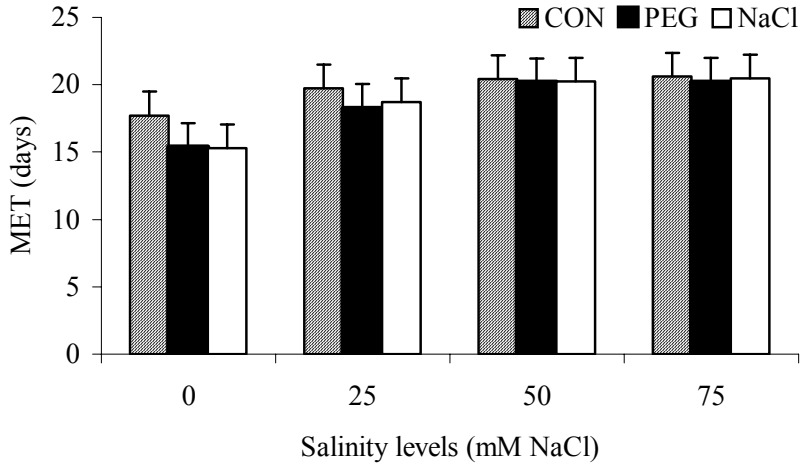

Figure 17: Effect of PEG and NaCl priming on MET (mean emergence time) of onion seeds irrigated with water containing a range of salinity levels (the bar "I" is SE)

There was a significant interaction between levels of salinity and priming (Table A24), and the highest MET was observed at higher salt concentrations. Irrespective of the priming solutions used, priming reduced the MET for all fresh and saline irrigated conditions. However, no significant difference was revealed between PEG and NaCl primed seeds. In addition, no significant difference was found between primed and control seeds, when seeds were watered with saline water.

Significant reduction in MET was obtained through priming when seeds were watered with fresh water and low salinity levels (25 mM). When the difference in MET was compared, priming accelerated emergence of onion seeds by 2.4 and 1.3 days when seeds were watered with fresh water and 25 mM respectively. However, at higher concentration the difference was only 0.2 days for 50 mM and 0.3 days for 75 mM (Figure 17). Therefore, marked and significant improvement in the rate of emergence through priming was obtained when seeds were irrigated with fresh water or water

with a low salt concentration (25 mM of NaCl). There was no influence of priming at higher salinity levels, since above a certain threshold all seeds emerged slowly and the advantage of priming, namely to reduce the lag phase of germination (Bradford, 1986) may be lost over time. Similar results were reported by Wiebe and Muhyaddin (1987) where PEG-primed tomato seeds responded less strongly at high EC.

c) Uniformity of emergence

No significant difference in time spread of emergence was found among different salinity levels (Table A24). Priming significantly decreased the time spread of emergence at all levels of salinity, though there was no significant difference between the two priming solutions (Table 35).

Table 35: Effect of priming on time spread of seedling emergence (TSE) of onion seeds

Priming treatment	TSE $\log_e$ (variance)
Control	3.04a[1]
PEG	2.86b
NaCl	2.93ab
Mean	2.94
CV (%)	4.46
LSD_{Tukey}	0.14

[1]Values followed by different letters were significantly different at α=0.05.

All primed seeds, whether primed with PEG or NaCl emerged at a narrower time span as compared to the untreated control seeds. The highest difference in TSE between primed and control seeds was observed at low salinity level; but the benefits decreased as the concentration of the irrigation water increased (Figure 18).

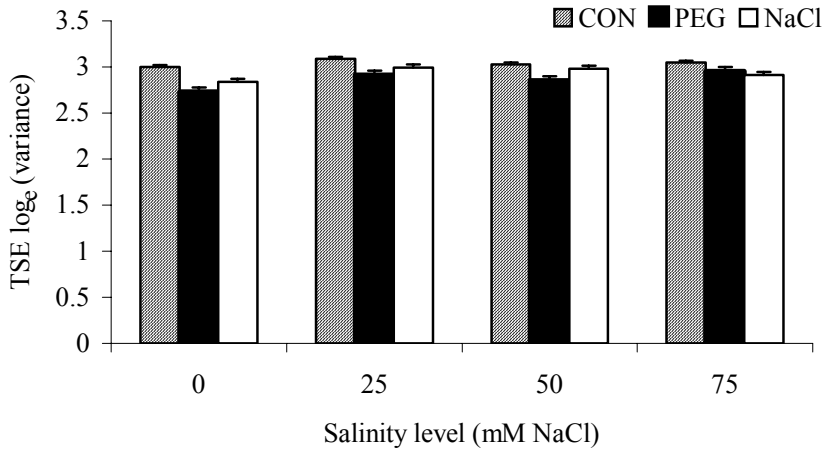

Figure 18: Effect of priming on TSE (uniformity) of onion seeds irrigated with water containing a range of salinity levels (the bar "I" is SE)

d) Seedling growth

The seedling dry mass was significantly different for the different salinity levels (Table A25). The highest dry mass was obtained from the seedlings irrigated with fresh water and seedling dry mass significantly reduced as the level of salinity level of the irrigation water increased (Table 36). Similarly, Mangal *et al.* (1989) reported a gradual decrease in the growth of onion seedlings as the concentration of EC went up from 0 to 10 mS cm^{-1} (with 2 mS cm^{-1} intervals). Sivritepe *et al*. (2003) also reported decreased seedling dry mass due to an increase in NaCl concentration.

Table 36: Effect of salinity level and priming on seedling dry mass of onion

Salinity level (mM NaCl)	Seedling dry mass (mg)	Priming treatment	Seedling dry mass (mg)
0	129.5a[1]	Control	72.4b
25	97.7b	PEG	94.0a
50	73.5c	NaCl	92.5a
75	44.5d	-	-
Mean	86.3	Mean	86.3
CV (%)	4.8	CV (%)	15.4
LSD_{Tukey}	5.3	LSD_{Tukey}	11.2

[1]Values followed by different letters were significantly different at α=0.05.

Severe injury, which resulted in stunted growth and dead tissue (Figure 19(a)), was caused by exposing the seedlings to high salinity conditions. Reduced growth in a plant occurs when the salinity exceeds the threshold level for a particular crop. Plants differ in their response to salinity and onion can be classified as a salt sensitive crop during the seedling stage. Salt sensitive crops have limited capacity to adjust and can be injured relatively easy at low salt concentrations. According to Blaylock (1994), the yield of salt sensitive plants can decrease by 25, 50 and 100% in soils with a salinity of 1.4-2.7 mS cm^{-1}, 2.6-4.2 mS cm^{-1} and >8 mS cm^{-1} respectively. In the present study, however, the percentage of reduction in seedling dry mass using 75 mM NaCl (EC = 6.9) was above 150%. This could be due to the use of nutrient solution, starting 12 days after sowing, by which the EC of the irrigation water was increased from 6.9 to 8.8 mS cm^{-1} (Table 30). Furthermore, it can also be a result of salt accumulation over time. This cannot be confirmed because no measurements were taken. However, in view of the fact that the media used was sand, no problem with drainage and thus salt accumulation was expected. However, toxicity and nutritional imbalances (higher Na^+ and Cl^-) may be the causes for lower growth of seedlings at saline conditions (Blaylock, 1994).

Priming significantly increased the dry mass of onion seedlings as compared to non-primed control seeds (Figure 19(b)). An increase in seedling dry mass of melon was also found by Sivritepe *et al*. (2003) after seeds were primed with NaCl solution. The highest increment in dry mass, due to priming was observed at lower levels of salinity, and the values deceased as the salt concentration in the water increased (Figure 20). No improvement in seedling dry mass was, however, observed at higher concentrations (75 mM = 6.9 mS cm^{-1}). This could be expected since priming could not reduce the MET (Figure 20) under very saline conditions. The higher seedling dry mass from primed than non-primed seeds was as result of faster emergence of primed seedlings (Brocklehurst & Dearman, 1983b). Sivritepe *et al*. (2003) reported no improvement in dry mass of melon due to priming when seeds were planted at 18 mS cm^{-1}. Furthermore, the authors also showed no significant differences in seedling dry mass as the salt concentration of the water increased.

Figure 19: Effect of a) salinity level and b) priming agent on onion seedling growth

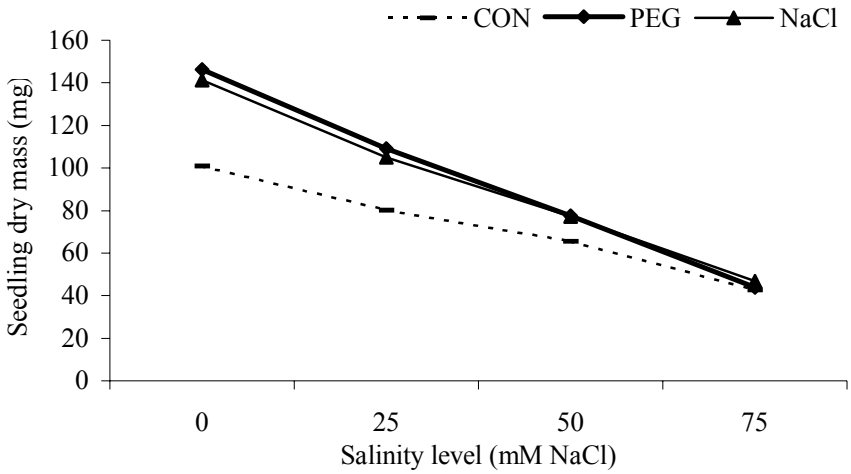

Figure 20: Effect of priming on seedling dry mass of onion, irrigated with a range of salinity levels

7.4. Conclusions

The results obtained from the current trial suggest that the emergence of onion seeds can be improved by priming when the seeds are subjected to different salinity levels and in particular where onion seeds are planted in relatively cool environment. The benefits of priming with PEG or NaCl at low salinity levels were not different. Both can be used effectively to alleviate salinity levels for attaining faster germination and more uniform seedlings, which result in larger seedlings with good market value. The stage of sensitivity of onion seeds to salinity shows to be at emergence rather than at germination. The present results further confirm that seedling growth of onion is even more sensitive to high salinity than emergence. Therefore, it is also advisable to treat onion at its seedling stage so as to produce more resistant seedlings that may survive more easily in saline soils or when irrigated with saline water.

CHAPTER 8
GENERAL DISCUSSION

The predictive values of standard and non-standard germination temperatures, standard accelerated ageing (AA) and saturated salt accelerated ageing (SSAA) tests were investigated. Germination tests were performed at standard and non-standard temperatures of 10°, 20°, 20°/30° and 30°C for cabbage and tomato, and 10°, 15°, 20° and 30°C for onion. The AA and SSAA tests were conducted using four relative humidities: standard AA (RH100), NaCl (RH75), $Ca(NO_3)_2$ (RH43) and $MgCl_2$ (RH32). Emergence trials were also conducted at a range of temperatures (winter, 15°/25° and 30°C) and media (Hygromix and soil) using seedling trays. Correlations were calculated to evaluate the relationship between laboratory and emergence test results.

There were vigour differences among the seed lots of all three crops used in this study. The seed lots of cabbage, onion and tomato were grouped as high, moderate and low vigour seed lots. High and low vigour seed lots generally perform consistently at both standard and non-standard temperatures and after ageing with different relative humidities, while there was some inconsistence in the intermediate vigour seed lots. High vigour seed lots perform well in all temperatures and ageing conditions with consistent values. However, low and medium vigour seed lots perform poorly when being incubated at adverse temperatures (10°C) and aged at higher RH's (RH100, RH75, RH43). Therefore, with the use of germination percentages of non-standard temperatures, and different ageing conditions (RH's) one can easily separate between high and low vigour seed lots, but not between the moderate vigour seed lots. To distinguish seed lots with intermediate vigour, the rate of germination parameters (MGT, GI and GE) can be used successfully. However, since rate of germination is highly temperature and moisture dependent, it needs careful regulation to ensure accurate results. It is also important to note that in calculating the GE, MGT and GI, all seeds with an emerged radicle were counted. Therefore a seed lot can have a short MGT or high GI even if there were few normal seedlings. Therefore, application of these tests as vigour tests without determining the standard germination percentage, may lead to incorrect conclusions.

For most seed lots, the ranking order according to germination rate parameters had similar trend with that of germination percentages at standard and non-standard temperatures, and germination percentages after ageing with different relative humidities. Low vigour seeds take longer time to

germinate as compared to high vigour seed lots. The slower germination from low vigour seeds could be due to the additional time needed to undertake self repair. However, there is no assurance for a high vigour seed lot to germinate faster though high germinating seed lots will most likely geminate faster.

For all seed lots the seed moisture content increased as the RH of the ageing increases. The RH in the standard AA (100RH) was above 29%, but in all crops the moisture content after SSAA was lower than 14%, which is not favourable for growth of fungus. Therefore, the all salts were effective in lowering the moisture content of seeds and no fungal growth was observed using SSAA. The deviation in seed moisture content in the standard accelerated ageing (RH100) test was above the tolerance level of 4% for all crop seeds, while in the SSAA it was below 4%. The fungal growth was more severe in cabbage, followed by onion and in tomato seeds. This could be due to the small size of cabbage seeds, enhancing absorption of water and stimulating faster deterioration.

The germination percentage was very close to the standard germination percentage when seeds were aged using RH32. At this relative low humidity the seeds might not deteriorated as fast as with higher humidities. The low germination percentage that was obtained after ageing seeds at RH43 could be due to the fluctuation in relative humidity with minor fluctuation in temperature.

NaCl (RH75) had relatively consistent results as compared to other ageing conditions in terms of seed moisture content. In general, since the relative humidity of NaCl is constant at a range of temperatures, this salt can be used even with incubators (outer chambers), which are less precise in temperature regulations. The use of this salt can also be used for reducing error introduced by fluctuations in temperature. Therefore, it is advisable to use NaCl (RH75) as a vigour test in the SSAA.

The results of the two temperature emergence trials (15°/25° and 30°C) did not show any pronounced vigour difference in the seed lots, since these temperatures were optimum or near optimal. This demonstrates that even poor vigour seed lots can also perform well under favourable environmental conditions and the standard germination test is good for predicting emergence of seed lots when onion seeds are planted at favourable environmental conditions. Although all seed lots

showed higher germination in the laboratory, there was a wide range in emergence percentage at non-favourable temperatures (winter and 30°C), indicating that they differed in vigour.

All parameters of cabbage, except MGT, GI and RH100 have significant correlations with various emergence trials (FEP and SVI). The germination percentages at 10°C and SSAA (RH32) have highly significant correlations with the mean emergence trial. In onion, highly significant correlations were observed between standard temperatures (15° and 20°C), GC and RH75 and RH32 with favourable emergence trial. This proved that standard germination percentage is a good tool for predicting emergence of onion seed lots when planted under conducive environments. None of the emergence trials of onion had significant correlation with the standard AA (RH100) and RH43 results. In tomato, the germination percentage at 20°C, standard AA and all SSAA test results had highly significant correlation with most emergence trials (PEP and SVI). The germination percentage from the standard temperature (20/30°C), 30°C and GC had also significant correlations with some emergence trials.

The use of GC may avoid disagreement arising because of subjective assessment of normal seedlings (Matthews, 1980). When tomato seed lots (B and F) were planted under favourable conditions the percentage emergence was higher than the standard normal germination test. Therefore, the use of GC based on emerged radicle rather than normal seedlings should not be completely avoided. Since low vigour may not show differences under favourable planting conditions, seeds may recover from some of the damages caused due to deterioration. Thus, some abnormal seedling may emerge if planted under encouraging environments.

When the germination rate parameters were correlated with emergence trials, for cabbage and tomato, GE had good correlation with all emergence trials. Nevertheless, MGT (negatively) and GI (positively) were correlated only with the winter emergence trial. In tomato, however, none of the germination rate characters (GE, MGT and GI) had significant correlations.

There was no significant difference in terms of percentage and rate of emergence between the two growth media (Hygromix and soil) used in this study. Therefore, soil can be used as an alternative medium for raising seedling transplants in places where commercial growth media are not accessible. These results could also be applicable for direct seeding of these vegetables in soil.

Onion seeds are short-lived and they lose their viability and vigour within one year and this resulted in slow, non-uniform emergence when seeds are planted under non-variable conditions, such as extreme temperatures. To investigate whether priming can improve vigour of onion, three seed lots onion that have different vigour levels were evaluated by primed with PEG-6000 or NaCl and were dried for 2 or 48-hrs at room temperature. Primed and non-primed seeds were subjected to a varying temperatures for germination (10°, 20° and 30°C); and winter (cold) and spring (favourable) for emergence trial.

Priming has no effect on germination capacity (final radicle percentage germination or viability) and in all onion seed lots this parameter was not improved through priming. However, priming did improve all parameters of germination percentage (FGP), rate (MGT, GE) and uniformity (TSG). All parameters of emergence except percentage were also improved though priming at all temperatures. However, marked, economic benefits are shown at the sub-optimal temperature (10°C). The enhancement due to priming was more pronounced in low vigour seed lots (C) than high vigour seed lots (B).

The highest GC, FGP, FEP, FSP (final survival percentage) and seedling dry mass were obtained from seeds germinated using distilled water. The fastest germination/emergence (low MGT/MET and high GE) and more uniform germination/emergence (low TSG and TSE) were observed when seeds were germinated with non-saline water. In all the variables, the benefits reduced gradually with increasing in levels of salinity. The number of abnormal and dead seedlings was higher when onion seeds were germinated at higher salt concentrations. Priming with either solution (PEG or NaCl) had no significant benefit in improving the over all percentage emergence or survival of the seeds.

Priming improved all germination rate and uniformity parameters used in the present study. Marked and significant improvement of all parameters through priming was obtained when seeds were irrigated with fresh and low salinity levels (25mM). The benefits of priming with PEG or NaCl at low salinity levels were not different. Thus, both can be used effectively to alleviate salinity levels for attaining faster germination and more uniform seedlings, which result in larger seedlings with good market value.

SUMMARY

To provide growers with high quality, seeds have been tested for their germination, purity and health. The standard germination percentage of seeds was obtained at favourable conditions and as a result it fails to give accurate information concerning a seed lot's field performance potential under unfavourable environmental conditions. Vigour test was introduced as a fourth character for better prediction of seedling emergence under a wide range of field conditions. AA vigour test is one of the tests that can be used to predict field emergence of large seeded vegetable crops. However, it is not successful in predicting emergence of small seeded vegetables and thus SSAA was introduced. In this study, the predictive values of the standard and non-standard temperature germination tests, the standard AA and SSAA tests were evaluated for cabbage, onion and tomato seeds. Seeds were also subjected to an emergence trial under a range of temperatures.

In all seed lots of the three crops, there were differences in vigour. Low temperature was best for separation of vigour difference according to their stages of physiological deterioration, 10°C for cabbage and onion seed lots and 20°C for tomato seed lots. The rate of germination can be used in distinguishing between seed lots according to their vigour levels. MGT (mean germination time), and GI (germination index) can also be used in differentiating seed lots with moderate vigour. A wide range of emergence was revealed at each trial, the poorest and slowest emergence occurred for the winter trial. Emergence percentage also varied between seed lots and the highest was observed at favourable temperature and the value decreased at stressed environmental conditions. These differences in the emergence were indicative range of seed vigour levels exits among seed lots.

Generally seed lots perform differently after different ageing tests as compared to the SGT, poor vigour seeds were affected severely, medium slightly while high vigour remained unaffected. In all crops, the overall mean germination percentage with $MgCl_2$ (RH32) was not significantly different from that of the control (SGT) results especially for high vigour seed lots. Low germination percentage was recorded after seeds were aged with RH100 (standard AA) and RH43 ($Ca(NO_3)_2$) in cabbage and onion. In tomato, however, the standard AA test was not significant from RH75 and RH32, but RH43 resulted in significantly lower germination percentage as compared to others. At the standard AA, there was a high seed moisture content, leading to high fungal growth. Using the saturated salt solutions, the seed moisture content was less than 14% and no fungal growth was

observed. NaCl (RH75) and $MgCl_2$ (RH32) had relatively consistent results as compared to the other ageing conditions in terms of seed moisture content.

The FEP of seeds growing at Hygromix or soil was not significantly different for any crop. However, larger seedlings were harvested from Hygromix than soil. For all crops, low vigour seed lots emerged more slowly and the final emergence percentage was lower as compared to high vigour seed lots. Differences in vigour of seed lots were reflected in their emergence in the seedling trays.

The best vigour tests that can predict emergence over a wider range of conditions (temperature and growth media) are different for different crops. In cabbage the best predictors are germination at 10°C and SSAA test using $MgCl_2$ (32%RH), for onion the SGT at 15°C, SSAA test using NaCl (75%RH) and $MgCl_2$ (32%RH) and for tomato germination at 20°C, and all ageing tests (standard and SSAA). Most laboratory tests were found to be best vigour tests for tomato as compared to cabbage and onion seeds.

Onion seeds are short-lived and they lose their viability and vigour within one year. Seeds lose vigour before they lose their ability to germinate, therefore, seed lots that have similarly high germination values can differ in the extent of deterioration, so as in vigour. Priming was found to improve the vigour of onion seed lots by maintaining some of these physiological deteriorations. As a result, the effect of priming on seed vigour of three onion seed lots that has different vigour levels (Chapter 5) were evaluated. This was done by subjecting primed and non-primed seeds to a varying temperature and salinity levels for germination and emergence trials.

In general, in seeds of higher quality (such as seed lot B) the possibility of improving vigour is naturally limited compared to seeds of lower quality (seed lot A). However, the increase of all parameters except germination and emergence percentages observed in all lots, at relatively cooler conditions. Seed priming is an effective means of improving vigour and is more advantageous for relatively low vigour seed lots (seed lot A) than high vigour seed lots (B and C).

Priming improved most characteristics of vigour of all seed lots at all temperatures even though it is more marked at lower temperature (10°C). The difference between high and low vigour seed lots is that high vigour seed lot can yield high percentage of germination and emergence; rapid germination/emergence and more uniform germination and emergence under adverse conditions.

For all seed lots the enhancement was revealed better in laboratory germination tests than emergence trials at a range of temperatures. Drying primed seeds to their original moisture content has slowed emergence as compared to surface dried seeds.

Germination and emergence percentages of onion seeds decreased gradually with an increase in levels of salinity. As the concentration of salts increased seedlings emerged from the seed coat but they were abnormal, with the number of dead seedlings the emergence trial also increase. Priming has improved the percentage of germination only at higher salt concentrations, but the same was not true for emergence percentage.

Onion seeds whether primed or not germinated/emerged at a slower rate, less uniform and seedling dry mass was lower as the concentration of salinity increased. Priming has reduced the mean germination time and time spread of germination significantly at all levels of salinity. However, significant reduction in mean emergence time and the time spread of emergence was revealed only at lower salinity level. Primed seeds, as compared to the control, showed higher seedling dry mass when irrigated with lower salinity concentrations. Thus, higher seedling dry mass of primed seeds was the result of earlier emergence as compared to the control.

The results obtained from this experiment can be used in improving emergence of onion under different temperature and salinity levels in particular when onion seeds are planted at relatively colder environments. The benefits of priming with PEG or NaCl have no difference and both can be effectively used to alleviate low salinity levels for attaining faster and more uniform seedlings, which result in larger seedlings with good market value. The stage of sensitivity of onion seeds to salinity shows to be at emergence rather than at germination. The present results further confirm that the seedling growth of onion is even more sensitive to salinity than emergence. Therefore, it is also worthwhile to treat onion at their seedling stages so as to produce more resistant seedlings that can survive easily in saline soils or when irrigated with saline water.

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APPENDIX

Table A1: Analysis of variance for **cabbage** seed lots germinated at different temperatures

Source	F value	Pr > F	CV (%)	Grand mean
10°C	7.73	0.0005	12.35	81.66
20°C	8.64	0.0003	4.59	89.50
20°/30°C	2.82	0.0474	6.14	87.92
30°C	5.32	0.0036	4.96	89.58
GC	1.90	0.1440	5.74	92.33
GE	14.96	< 0.0001	7.15	85.75
MGT	92.12	< 0.0001	5.62	2.23
GI	43.78	< 0.0001	8.52	24.91

Df = 5

Table A2: Analysis of variance for **onion** seed lots germinated at different temperatures

Source	F value	Pr > F	CV (%)	Grand mean
10°C	13.19	< 0.0001	5.21	77.0
15°C	11.56	< 0.0001	3.45	86.71
20°C	9.57	< 0.0001	2.68	89.85
30°C	15.25	< 0.0001	2.76	86.0
GC	8.47	< 0.0001	2.54	93.14
GE	55.19	< 0.0001	9.98	65.14
MGT	34.49	< 0.0001	11.16	4.03
GI	88.0	< 0.0001	3.69	10.13

Df = 6

Table A3: Analysis of variance for **tomato** seed lots germinated at different temperatures

Source	F value	Pr > F	CV (%)	Grand mean
20°C	10.36	< 0.0001	6.86	85.19
20°/30°C	27.47	< 0.0001	2.93	89.25
30°C	40.20	< 0.0001	2.83	87.63
GC	7.87	< 0.0001	3.36	92.25
GE	152.48	< 0.0001	7.12	67.56
MGT	571.57	< 0.0001	3.48	3.37
GI	125.41	< 0.0001	6.23	16.57

Df = 7

Table A4: ANOVA of interaction "incubation temperature x seed lot" for cabbage, onion and tomato seed lots

Source	Df	F value	Pr > F	CV (%)	Grand mean
Cabbage					
Temperature	3	8.02	0.0001	-	-
Seed lot	5	20.27	<0.0001	-	-
Temp x Lot	15	2.11	0.0188	7.43	87.17
Onion					
Temperature	3	93.39	<0.0001	-	-
Seed lot	6	38.69	<0.0001	-	-
Temp x Lot	18	3.81	<0.0001	3.56	84.89
Tomato					
Temperature	2	8.52	0.0005	-	-
Seed lot	7	48.31	<0.0001	-	-
Temp x Lot	14	0.97	0.4899	4.54	87.36

Table A5: ANOVA of cabbage, onion and tomato seeds for the final germination percentage after ageing with different RH

Crop	F value	Pr > F	CV (%)	Mean
Cabbage	32.72	<0.0001	23.19	69.73
Onion	32.47	<0.0001	16.04	75.44
Tomato	7.60	<0.0001	12.56	82.95

Df = 4

Table A6: ANOVA of different ageing conditions (relative humidities), seed lot and their interaction for the final germination percentage of cabbage, onion and tomato

Source	Df	F value	Pr > F	CV (%)	Grand mean
Cabbage					
Ageing conditions	4	456.53	<0.0001	-	-
Seed lot	5	209.82	<0.0001	-	-
Ageing x Lot	20	23.28	<0.0001	6.21	69.73
Onion					
Ageing conditions	4	175.18	<0.0001	-	-
Seed lot	6	70.11	<0.0001	-	-
Ageing x Lot	24	8.44	<0.0001	6.91	75.44
Tomato					
Ageing conditions	4	54.49	<0.0001	-	-
Seed lot	7	124.09	<0.0001	-	-
Ageing x Lot	28	4.37	<0.0001	4.37	82.95

Table A7: ANOVA for germination percentage after ageing at different RH for seed lots of cabbage onion and tomato

Source	F value	Pr > F	CV (%)	Grand mean
Cabbage (Df = 5)				
Control	3.90	0.0143	5.07	90.17
RH100	126.22	<0.0001	11.17	39.67
RH75	52.01	<0.0001	6.29	69.50
RH53	110.18	<0.0001	6.03	69.42
RH32	13.00	<0.0001	5.12	79.92
Mean	203.31	<0.0001	2.82	69.33
Onion (Df = 5)				
Control	11.44	<0.0001	2.34	90.50
RH100	27.61	<0.0001	9.29	62.64
RH75	8.39	0.0001	10.41	73.36
RH53	38.08	<0.0001	8.99	63.43
RH32	16.34	<0.0001	2.96	87.29
Mean	70.91	<0.0001	3.07	75.44
Tomato (Df = 5)				
Control	20.12	<0.0001	3.23	89.19
RH100	30.21	<0.0001	5.10	82.56
RH75	27.90	<0.0001	4.61	82.64
RH53	35.11	<0.0001	6.11	75.25
RH32	20.99	<0.0001	4.41	85.13
Mean	159.55	<0.0001	1.85	82.95

Table A8: Soil particle and nutrient analysis

Soil nutrient analysis						Particle size analysis			
PH water	P (mg/kg)	Ca (mg/kg)	K (mg/kg)	Mg (mg/kg)	Na (mg/kg)	Medium sand %	Silt %	Clay %	Total %
6.9	3.0	441	24	192	139	72.7	3.3	24.5	100.5

Table A9: ANOVA of temperature, growth media, seed lot and their interactions for the different parameters of emergence of **cabbage** seeds

Source	Df	Emergence (%)		MGT (days)		Seedling vigour index		Seedling dry mass (mg)	
		F value	Pr > F	F value	Pr > F	F value	Pr > F	F value	Pr > F
Temperature	2	93.20	<0.0001	895.33	<0.0001	806.94	<0.0001	138.13	<0.0001
Seed lot	5	56.51	<0.0001	43.07	<0.0001	113.61	<0.0001	42.84	<0.0001
Temp x Lot	10	3.51	0.0005	2.91	0.0029	3.16	0.0014	2.93	0.0028
Growth media	1	2.28	0.1341	11.41	0.0001	5.70	0.0187	1138.9	<0.0001
Temp x Media	2	0.04	0.9646	0.25	0.7814	1.53	0.2203	35.43	<0.0001
Lot x Media	5	0.02	0.9997	0.87	0.5061	0.44	0.8174	4.66	0.0007
Temp x Lot x Media	10	0.09	0.9999	0.96	0.4810	0.91	0.5267	1.87	0.0574
Grand mean		81.39		8.81		5.29		292.99	
CV (%)		8.14		7.72		9.02		13.42	

Table A10: ANOVA of temperature, growth media, seed lot and their interactions for the different parameters of emergence of **onion** seeds

Source	Df	Emergence (%)		MGT (days)		Seedling vigour index		Seedling dry mass (mg)	
		F value	Pr > F	F value	Pr > F	F value	Pr > F	F value	Pr > F
Temperature	2	21.65	<0.0001	724.26	<0.0001	998.01	<0.0001	12.03	<0.0001
Seed lot	6	31.32	<0.0001	49.07	<0.0001	65.64	<0.0001	27.18	<0.0001
Temp x Lot	12	1.40	0.1742	11.39	<0.0001	4.63	<0.0001	1.05	0.4088
Growth media	1	0.65	0.4223	42.91	<0.0001	11.90	0.0008	29.85	<0.0001
Temp x Media	2	0.24	0.7860	3.80	0.0251	3.96	0.0215	1.27	0.2839
Lot x Media	6	0.08	0.9977	0.60	0.7326	0.32	0.9361	0.69	0.6561
Temp x Lot x Media	12	0.15	0.9995	0.98	0.4679	0.27	0.9923	0.31	0.9870
Grand mean		83.32		16.09		2.88		90.49	
CV (%)		6.67		4.20		7.57		18.57	

Table A11: ANOVA of temperature, growth media, seed lot and their interactions for the different parameters of emergence of **tomato** seeds

Source	Df	Emergence (%)		MGT (days)		Seedling vigour index		Seedling dry mass (mg)	
		F value	Pr > F	F value	Pr > F	F value	Pr > F	F value	Pr > F
Temperature	1	0.53	0.4693	2161.3	<0.0001	602.28	<0.0001	40.05	<0.0001
Seed lot	7	13.0	<0.0001	29.49	<0.0001	27.61	<0.0001	22.63	<0.0001
Temp x Lot	7	0.32	0.9454	6.90	<0.0001	1.40	0.2156	1.41	0.2092
Growth media	1	1.08	0.3019	17.40	<0.0001	3.73	0.0564	23.63	<0.0001
Temp x Media	1	4.98	0.0280	12.54	0.0006	0.04	0.8386	0.63	0.4297
Lot x Media	7	0.28	0.9615	2.28	0.0340	0.46	0.8614	0.17	0.9898
Temp x Lot x Media	7	0.27	0.9640	2.40	0.0261	0.14	0.9945	0.63	0.7273
Grand mean		85.22		13.74		4.0		185.03	
CV (%)		8.0		5.70		9.97		15.68	

Table A12: ANOVA of **FEP** (final emergence percentage) of cabbage, onion and tomato seeds planted under range of temperatures

Source	F value	Pr > F	CV (%)	Grand mean
Cabbage (Df = 5)				
Winter	19.30	<0.0001	9.54	70.88
15/25°C	8.23	0.0003	5.93	88.17
30°C	18.64	<0.0001	4.61	85.13
Mean	36.28	<0.0001	4.15	81.39
Onion (Df = 6)				
Winter	8.29	0.0001	6.30	79.64
15/25°C	7.47	0.0002	4.08	86.50
30°C	15.61	<0.0001	4.53	83.82
Mean	29.51	<0.0001	2.81	83.82
Tomato (Df = 7)				
15/25°C	6.69	0.0002	5.75	85.66
30°C	6.86	0.0002	5.46	84.78
Mean	12.61	<0.0001	4.06	85.22

Table A13: ANOVA of **SVI** (seedling vigour index) of cabbage, onion and tomato seeds planted under range of temperatures

Source	F value	Pr > F	CV (%)	Grand mean
Cabbage (Df = 5)				
10°C - Hygromix	26.35	<0.0001	11.28	3.11
10°C - Soil	14.56	<0.0001	14.04	3.10
15/25°C - Hygromix	29.47	<0.0001	6.21	5.78
15/25°C - Soil	9.29	0.0002	12.05	6.06
30°C - Hygromix	6.72	<0.0001	7.09	6.72
30°C - Soil	33.40	<0.0001	6.44	7.01
Mean	118.99	<0.0001	3.60	5.29
Onion (Df = 6)				
10°C - Hygromix	13.02	<0.0001	8.58	1.85
10°C - Soil	9.53	<0.0001	9.71	1.87
15°/25°C - Hygromix	19.95	<0.0001	6.68	2.99
15°/25°C - Soil	12.77	<0.0001	7.36	3.23
30°C - Hygromix	12.88	<0.0001	6.23	3.62
30°C - Soil	9.84	<0.0001	7.59	3.70
Mean	68.64	<0.0001	3.02	2.88
Tomato (Df = 7)				
15°/25°C	22.28	<0.0001	6.41	2.66
30°C	14.76	<0.0001	6.35	4.13
Mean	29.40	<0.0001	4.83	3.40

Table A14: ANOVA of **GC** (germination capacity) and **FGP** (final germination percentage) for temperature, seed lot, priming treatment and their interactions of onion seeds germinated under range of temperatures

Source	Df	GC		FGP	
		F value	Pr > F	F value	Pr > F
Temperature (Temp)	2	2.23	0.1176	44.27	<0.0001
Seed lot (Lot)	2	0.80	0.4537	2.85	0.0665
Temp x Lot	4	2.58	0.0472	0.88	0.4831
Priming treatment	1	0.25	0.6208	5.20	0.0266
Temp x Priming	2	0.13	0.8795	7.74	0.0011
Lot x Priming	2	0.07	0.9331	0.47	0.6280
Temp x Lot x Priming	4	0.04	0.9969	0.41	0.8013
Grand mean		95.58		88.33	
CV (%)		2.48		4.21	

Table A15: ANOVA of **GE** (germination energy), **MGT** (mean germination time) and **TSG** (time spread of germination) for temperature, seed lot, priming treatment and their interactions of onion seeds germinated under range of temperatures

Source	Df	GE		MGT		TSG	
		F value	Pr > F	F value	Pr > F	F value	Pr > F
Temperature (Temp)	2	355.89	<0.0001	828.94	<0.0001	674.18	<0.0001
Seed lot (Lot)	2	36.44	<0.0001	153.29	<0.0001	134.13	<0.0001
Temp x Lot	4	20.44	<0.0001	10.89	<0.0001	18.07	<0.0001
Priming treatment	1	288.18	<0.0001	902.46	<0.0001	984.03	<0.0001
Temp x Priming	2	140.42	<0.0001	76.32	<0.0001	187.76	<0.0001
Lot x Priming	2	8.44	0.0006	9.29	0.0003	63.12	<0.0001
Temp x Lot x Priming	4	3.24	0.0186	3.54	0.0123	17.87	<0.0001
Grand mean		66.46		3.69		0.85	
CV (%)		2.59		6.28		13.57	

Table A16: ANOVA of **FEP** (final emergence percentage) of onion seed lot, priming treatment and their interaction planted at cold and favourable conditions

Source		Cold		Favourable	
	Df	F value	Pr > F	F value	Pr > F
Seed lot (Lot)	2	2.27	0.1231	0.56	0.5777
Priming treatment	4	0.61	0.5482	0.14	0.8700
Lot x Priming	4	0.20	0.9352	0.52	0.7217
Grand mean		81.61		92.33	
CV (%)		4.26		4.42	

Table A17: ANOVA of **MET** (mean seedling emergence time) of onion seed lot, priming treatment and their interaction planted at cold and favourable conditions

Source		Cold		Favourable	
	Df	F value	Pr > F	F value	Pr > F
Seed lot (Lot)	2	33.44	<0.0001	13.57	<0.0001
Priming treatment	4	168.03	<0.0001	67.57	<0.0001
Lot x Priming	4	0.51	0.7307	0.51	0.7301
Grand mean		15.37		11.34	
CV (%)		3.39		2.60	

Table A18: ANOVA of **SVI** (seedling vigour index) of onion seed lot, priming treatment and their interaction planted at cold and favourable conditions

Source		Cold		Favourable	
	Df	F value	Pr > F	F value	Pr > F
Seed lot (Lot)	2	13.13	0.0001	2.65	0.0892
Priming treatment	4	85.04	<0.0001	18.14	<0.0001
Lot x Priming	4	0.86	0.4991	0.13	0.9722
Grand mean		2.76		4.13	
CV (%)		5.39		4.59	

Table A19: ANOVA of **TSE** (time spread of emergence) of onion seed lot, priming treatment and their interaction planted at cold and favourable conditions

Source		Cold		Favourable	
	Df	F value	Pr > F	F value	Pr > F
Seed lot (Lot)	2	5.24	0.0120	4.19	0.0261
Priming treatment	4	113.18	<0.0001	6.02	0.0069
Lot x Priming	4	1.18	0.3406	0.27	0.8941
Grand mean		2.50		2.12	
CV (%)		4.05		5.46	

Table A20: ANOVA of **seedling dry mass** of onion seed lot, priming treatment and their interaction planted at cold and favourable conditions

Source		Cold		Favourable	
	Df	F value	Pr > F	F value	Pr > F
Seed lot (Lot)	2	0.11	0.8941	1.12	0.3416
Priming treatment	4	18.46	<0.0001	1.80	0.1852
Lot x Priming	4	0.07	0.9901	0.35	0.8406
Grand mean		81.92		202.86	
CV (%)		11.37		5.21	

Table A21: ANOVA of **GC** (germination capacity) and **FGP** (final germination percentage) of onion seeds primed with different priming agents and germinated at range of salinity levels

Source		GC		FGP	
	Df	F value	Pr > F	F value	Pr > F
Salinity level	3	230.59	<0.0001	719.91	<0.0001
Priming treatment	2	8.29	0.0011	54.12	<0.0001
Salinity x Priming	6	9.01	<0.0001	34.84	<0.0001
Grand mean		65.31		40.45	
CV (%)		6.76		9.76	

Table A22: ANOVA of **MGT** (mean germination time) and **TSG** (time spread of germination) of onion seeds primed with different priming agents and germinated at range of salinity levels

Source	Df	MGT		TSG	
		F value	Pr > F	F value	Pr > F
Salinity level	3	964.65	<0.0001	630.08	<0.0001
Priming treatment	2	446.36	<0.0001	276.61	<0.0001
Salinity x Priming	6	4.87	0.001	9.86	<0.0001
Grand mean		8.97		1.39	
CV (%)		4.40		8.84	

Table A23: ANOVA of **FEP** (final emergence percentage) and **FSP** (final survival percentage) of onion seeds primed with different priming agents and planted at range of salinity levels

Source	Df	FEP			FSP		
		F value	Pr > F	CV (%)	F value	Pr > F	CV (%)
Block	3	0.8413			2.2442	0.1523	
Salinity level	3	62.8739	<0.0001	6.38	439.0548	<0.0001	4.32
Salinity error	9						
Priming treatment	2	0.2618			0.9387		
Salinity x Priming	6	0.4782		6.38	0.7280		7.54
Salinity x Priming error	24						
Grand mean		75.7			67.4		

Table A24: ANOVA of **MET** (mean emergence time) and **TSE** (time spread of emergence) of onion seeds primed with different priming agents and planted at range of salinity levels

Source	Df	MET			TSE		
		F value	Pr > F	CV (%)	F value	Pr > F	CV (%)
Block	3	0.2499		3.09	1.6919	0.2376	7.04
Salinity level	3	138.6492	<0.0001		3.2065	0.0762	
Salinity error	9						
Priming treatment	2	11.7881	0.0003		7.5494	0.0029	
Salinity x Priming	6	3.0085	0.0245	3.66	0.4979		4.46
Salinity x Priming error	24						
Grand mean			18.94			2.94	

Table A25: ANOVA of **seedling dry mass** of onion seeds primed with different priming agents and planted at range of salinity levels

Source	Df	Seedling dry mass		
		F value	Pr > F	CV (%)
Block	3	4.5650	0.0331	4.32
Salinity level	3	902.0796	<0.0001	
Salinity error	9			
Priming treatment	2	13.3323	0.0001	
Salinity x Priming	6	2.4344	0.0557	15.35
Salinity x Priming error	24			
Grand mean			86.3	