

**QUALITY ASSESSMENT OF PEA (*PISUM SATIVUM* L.) SEEDS
USING THE CONTROLLED DETERIORATION TECHNIQUE**

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A THESIS

BY

MD. LUTFOR RAHMAN

STUDENT NO.: 0905011

SESSION: 2009

SEMESTER: SUMMER, 2010



MASTER OF SCIENCE (M. S.)

**IN
HORTICULTURE**

DEPARTMENT OF HORTICULTURE

HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY UNIVERSITY

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Submitted to the Department of Horticulture

Hajee Mohammad Danesh Science and Technology University

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In partial fulfillment of the requirements for the degree of

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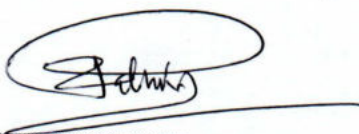
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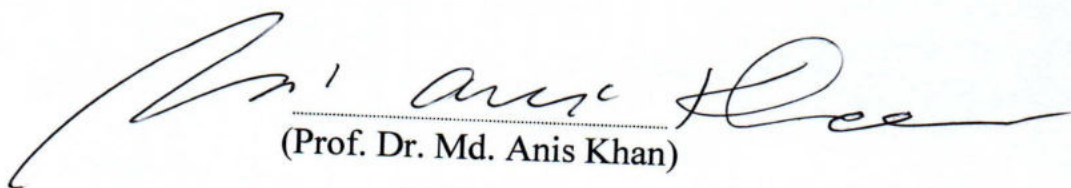
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DEDICATION

TO,

BELOVED PARENTS

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ABSTRACT

A three-factor experiment was set at the Horticulture Laboratory, Hajee Mohammad Danesh Science and Technology University, Dinajpur during April - June 2010 to study the effects of the controlled deterioration (CD) on the pea seeds at the constant temperature of 35°C. The 3 factors considered were: 3 pea seed sources; 3 ageing periods (0, 8 and 16 days); and 3 seed moisture contents (12, 16 and 20% MC). The 27 treatment combinations compared in the CRD with the 3 repetitions for the 8 parameters were: % germination, % abnormal seedlings, % dead seeds, % soil emergence and seedling evaluation test for the root and shoot lengths as well as their dry matter contents. Identical prototypes of notable (5-1% level) degradations were recorded everywhere. But the disparities were clear under the extreme stresses. Moreover, highly noteworthy (1% level) relations were traced amid all the traits ranging from -0.9847 (soil emergence x abnormal seedling) to 0.9623 (soil emergence x normal seedling). So, the CD technique was very effectual in judging the physiological statuses of the seed sources studied. Thus, the germination test might be add-on by a vigour test, the latter of which could be assessed by quantifying the seedlings' root and shoot lengths and/or their dry matter accumulations. Moreover, in the seed quality certification, the suitable limits of vigour for the chosen traits could also be got by this technique. But the seeds of several pea varieties should be exploited to fix-up the agreeable limits of the traits. Furthermore, to save time, the ageing period could be squeezed by raising the seed moisture content (MC).

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CHAPTER I

INTRODUCTION

Legumes play vital roles in the global agriculture, chiefly by providing the greatest amount of plant proteins to humans as well as animals (mostly various pulses), vegetables (beans and peas), vegetable oils (groundnut, soybean), ornamentals (lupines, garden pea, cassia, etc.), fodder (cow pea, clover, etc.), timber and firewood (diverse trees), fruits (tamarind, *Igna dulkis*, etc.), spices (fenugreek, *dalfiringi*, etc.), medicines (several spp.), natural dyes (indigo, etc.) and fibres (sun hemp). Even a few legumes are grown as cover and green manuring crops too. Moreover, all the legumes fix notable quantities of atmospheric nitrogen to soils utilizing the bacteria *Rhizobium* spp.- a unique credibility in the whole plant kingdom (Purseglove, 1977). And numerous leguminous species have major roles in the national economy of Bangladesh. Out of those, pea (*Pisum sativum* L.) is a cherished and nourishing winter vegetable as well as pulse crop in the country. In addition, its seeds are also utilized as snacks: fried seeds and *chotpoti*.

Seed quality affects the performance of any crop under the field conditions in several ways (Roberts and Osei-Bonsu, 1988; Roberts, 1986; Keefe and Draper, 1986). In addition, the initial seed quality also affects the storage life of seeds. So, an in-depth study of the seed quality is vital if crop productivity as well as the storage life of the seeds is to be enhanced or even maintained.

A single definition of the seed quality fit for all crops under varied agro-ecological conditions is neither possible nor usable. So, the elements of the seed quality should be taken into the accounts are as follows:

- 1) Percent seeds that germinate and, therefore, the initial seedling density.
- 2) Germination rates and the growth of seedlings, which would determine the sizes of the plants being produced.
- 3) Health condition of the seedlings obtained.
- 4) Uniformity of the seedlings grown and finally
- 5) Ultimate yield and the class of the produce.

Seed germination, vigour and size are the 3 crucial aspects of seed quality, which influence crop yield and grade through both the indirect and the direct effects (Ellis, 2004). A major component of the seed quality is the % germination determined as per the International Seed Testing Association (ISTA, 1999) and performed by any seed certification program all over the world on a routine basis. The germination rate under the laboratory conditions is usually a good estimator of the field emergence (Pourhadian and Khajehpour, 2010). Many seeds though germinate profusely under the ideal laboratory situations fail to emerge abundantly in the crop field (Wang *et al.*, 2004; Kolasinska *et al.*, 2000; Kulik and Yaklich, 1982). So, the aim of the laboratory germination test is to facilitate the global trade in seeds by proving a standard measure of the field planting value under the idyllic conditions.

Differences in the field emergences of the seed lots with the similar and acceptable levels of the laboratory germination percentage had been noticed in several legumes (Borba, 1987; Roberts, 1986; Perry, 1982; Hampton and Scott, 1982). As such variations are observed the standard laboratory germination test fails to be the effective indicator of the field establishment. Failure of the germination percentage to relate with the field emergence

leads to the very term '**Vigour**' at the 1950's ISTA Congress in Washington D. C. (Perry, 1978). The data evaluated by Brown and Mayer (1986) also showed that the seed germination continued at the slow rate after the fast period of the germination. Apparently, these slow germinating seeds had low vigour (Ranal and Santana, 2006; Brown and Mayer, 1986). Seed lots are considered to possess the low vigour when field emergence is low compared to other seed lots having the similar germination percentage.

Accelerated aging is a very responsive test in ranking the quality of seeds (Khan *et al.*, 2007; Hill *et al.*, 2007 and Kibinza *et al.*, 2006). And the controlled deterioration test is usually illustrated by the nonlinear courses of germination loss during the aging period (Kruse, 1999). Several factors are liable for the vigour of any seed. But little work has been carried out to study the effects of the ageing on the physiological statuses of pea seeds.

Keeping that view in mind, the present study was set with these two specific objectives:

- To investigate some of the significant consequences of the ageing using the controlled deterioration technique on the physiological statuses of the 3 pea seed sources (Thakurgaon Local/TL, Dinajpur Local/DL and Rangpur Local/RL) during their germination and the subsequent establishment of the seedlings and
- To relate the findings with the standard germination test for seed quality certification.

CHAPTER II

REVIEW OF RELATED LITERATURE

Different authors studied the quality and the ageing of various seed species. The valued conclusions of those important findings are reviewed critically and highlighted in this chapter selectively under different suitable headings for the ease of quick access.

1. Concept of the seed quality

The successful culture of any crop depends on the rapid establishment of an area of the foliage to intercept the incoming solar radiation. A measurement of the seed quality must, therefore, take accounts of (Perry, 1980):

- a. Percentage of the seeds producing the normal seedlings and
- b. Rate of the germination and the growth rate of the seedlings.

But the differences in the field emergence of the seedlings of various seed lots of any crop having the similar and high level of laboratory germination percentage might occur (Roberts, 1986; Perry, 1982). If such disparities were found, the laboratory germination test failed to be the effective mirror of the field emergences and the seeds were said to have dissimilar ranks of vigour.

Such a discrepancy is not a new finding, as more than 130 years ago Nobbe (1876) recognized that the properties of the individual seed, such as, the speed of germination and the seedling growth rate varied within a seed lot, and that the average of the seed lots frequently differed also. He gave the

German name 'TRIEBKRAFT' (literally meaning 'Driving force') to that happening, which correlates with the very English term 'SEED VIGOUR'.

Seed vigour has been described as the cover name given to the complex of the interrelated properties, which rendered the seeds versatile enough to germinate and flourish under the varied conditions (Heydecker, 1972; Perry, 1972). Vigour has also been used to describe a wide range of traits of the individual seed or seed lots including the activity of the enzymatic systems or oxygen uptake; the aspects of germination such as the rate and the uniformity, and the rate of seedling growth; the performance in the field with respect to percentage, the rate and the uniformity of seedling emergences, the plant growth and even the yield and grade of the produce (Perry, 1976).

Vigour has been defined in various ways including: the response to the stress conditions (Abdullah and Vanderlip, 1972); the relative stage of the seedling development (Adenikinju, 1972); the relative seedling growth rate measured by the dry weight accumulation (Lowe *et al.*, 1972); the seedling survival rate (Mukherjee *et al.*, 1971) and a length of time a seedling was able to exert an increasing upward force (Jensen *et al.*, 1972). Such considerations, however, has been excluded as merely mechanical, morphological or microbial factors and the high vigour has been given by others, a rather narrower definition, i.e. it was the physiological property of the individual seed (Perry, 1972).

From the above definitions, it is quite apparent that the seed vigour is a multi-component concept rather than a single clear-cut quantifiable property.

An agreed definition was not accepted for the seed vigour until 1977 when the ISTA's vigour committee anonymously adopted the following one:

'Seed vigour is the sum total of those properties of seeds, which determine the level of the activity and the performance of the seeds or a seed lot during the germination and the seedling emergence.' (Perry, 1978). It went on to specify those aspects of performance, which has been reported as showing the variations linked with the disparities in the seed vigour (Perry, 1978):

- a. biochemical processes and the reactions during the germination such as the enzymatic reactions and the respiratory activity;
- b. rate of the uniformity of the seedling emergence and the growth;
- c. rate and the uniformity of the seedling emergence and the growth in fields and, finally
- d. ability of the emergence of the seeds under the unfavourable locality.

Perry (1970) categorized the disparities in the seed vigour into two groups:

- a. the intrinsic factors due to the genetic make-up and
- b. the disparities induced by the external factors interacting on the genotypes.

The author also concluded that the maximum possible vigour was determined by the genotype but could be modified.

The causes of variations in the vigour are several and diverse, and the commonly known factors influencing the altitude of the vigour are listed here to clarify the concept further (Perry, 1978):

- a. genetic constitutions i.e. the species related and also varietals;
- b. growing environments and the nutrition of the mother plants;
- c. stage of maturity at harvest;
- d. deterioration and the ageing and
- e. pathogens.

The seed deterioration affects many sub-cellular systems. So, the loss of seed vigour has many symptoms. It is, consequently, not surprising that many different vigour tests measure quite diverse properties, which might be more or less vital depending on the circumstances in the field after sowing. Hence, a number of esteemed workers argued that as an effective indicator of field performance, the vigour could be more reliably assessed on the basis of the "multiple criteria". In other words, several different types of tests should be carried out on each seed lot, and the results should be combined in some arbitrary way to provide a single vigour index for seed lots (Roberts, 1986).

Thus, to summarize, the seed vigour is a concept, rather than a specific property of a seed or seed lot. Several factors might alone or jointly influence the level of the vigour. Essentially, the vigour is affected by the genetic make-up of a seed. But the efficiency by which a seedling will establish is modified by the physiological and biochemical constituents of the seed or seed lot and is influenced by the environment during production, processing, handling, storage and even after sowing in the fields.

2. Importance of the seed vigour

Eminent scientists are working globally to discover the package of technology to produce the high vigour seeds and maintain its utmost level

during the storage. The reason for those valued works is that the vigour has immense consequences on the crop production. In support of the declaration, the following points could be pinpointed.

2.1 High and uniform field emergence as well as seedling establishment

The optimum plant population in a unit area is essential to achieve the high yield of any crop; it is particularly vital when there is under population. The low vigour seed could fail to sustain in the field and it happens primarily if there are stresses or sub-optimal conditions in the field (Roberts, 1986). Perry (1982) described the reduced establishment as well as the seedling growth associated with the low vigour seeds of peas and Brassicas grown in the protected environment and in the field sown vegetable crops. Humpton and Scott (1982) also observed poor emergence resulting in the reduced plant stand with the low vigour pea seeds. Roberts (1986) mentioned that the effects of the low vigour on the emergence would likely be a greater problem in those species where there is evidence of deteriorative changes occurring in the seeds well in advance of death, e.g. soybean. Because its seeds became defective about 1.7 normal deviates before death (Ellis and Roberts, 1981). Other legumes also showed the relatively higher potential of abnormal seedlings and thereby reduced % germination for the same reason. In cowpeas and chickpeas, for instance, the seeds appeared to become defective about 0.9 normal deviates before death (Osei-Bonsu, 1981). In contrast, other species, such as, in cereals, the seeds become defective in terms of producing seedling abnormalities and the reduced germination only a

relatively short time before death, e.g. 0.3 normal deviates both in wheat and maize (Ellis and Roberts, 1980). Hence, the abnormal seedlings in the leguminous seeds and pulses are very common than those of cereals.

So, the high uniform field emergences and the good seedling establishment could be got from the high vigorous seeds, chiefly under the sub-optimal field conditions.

2.2. Good health of the seedling/crop

If poor quality seeds are sown, the seedlings are also of the low vigour (Delouche and Baskin, 1973). In addition, the low vigour seeds are more prone to pests and diseases and produced non-uniform stands. Nonetheless, the dissimilarities in vigour amid individual plants are obvious even at the seedling and the early growth stages (Roberts, 1986). And from the unhealthy and the non-uniform plants, a good harvest is quite unexpected.

2.3 Effect of the seed vigour on the crop yield

There is controversy about yield reduction due to the use of the low vigour seeds. Reduced yields were obtained using both the low vigour pea (Humpton and Scott, 1982) and cotton seeds (Bishnoi, 1981). These authors mentioned that the drop in the yields was basically a function of the reduced plant stands. Roberts (1986) experienced that the potential loss of the yield from the poor emergence could be exacerbated because the emergence might be patchy; even if the average number of plants/unit area is ideal, an irregular distribution of the plants could lead to the reduced yield. In contrast, nevertheless, no disparities were found in the yields amid the low and the high vigour peas (Humpton and Scott, 1982) and soybean (Egli and

Tekrony, 1979) at equal plant densities. Oppositely, at the identical plant stands, the high vigour cotton seeds gave 21% more yield than the low vigour seeds (Bishnoi, 1981). Differences in the seed vigour were found more likely to affect the final yield at the identical plant densities following the late sowing when the growth period was shortened (Khan and Roberts, 1986). Roberts (1986) showed that the decline in the yield from the low vigour seeds was less when the plants were grown at the higher densities than at the lower stand; he testified that phenomenon as the greater rivalry between the plants from the more vigorous lots in the high population densities compared to the less vigorous ones.

2.4 Seed vigour and the storage

It is tricky to imagine how humans would sustain if it were impractical to store seeds from one season to the next one. Most of the global agriculture depends on the simple code that once seeds are reaped and dried, those could retain their viability for certain period depending on the storage conditions. From a purely monetary view, the seed industry would have a difficult time balancing the supply and the demand if it could not '**Carry over**' some seeds for the next season. Similarly, plant breeders need long-term storage so that the breeding materials could be sustained for up to 10 or more years. That is feasible to the extreme level by the gene banks where the indefinite preservation of the seeds (= genes) is the ultimate goal (Ross, 1986).

The utmost seed quality might occur at the physiological maturity stage, i.e. when the seed attains the topmost dry weight (Siddique *et al.*, 1987). But the seeds start to deteriorate as soon as those have reached the peak maturity on

the mother plant at the rate depending mainly on the seed MC and the prevailing temperature (Roberts and Osei-Bonsu, 1988). Even after reaping at the physiological maturity stage and storing the high vigour seeds in the most suitable setting, the vigour will fall due to the natural ageing (Powell *et al.*, 1984; Ross, 1980).

Nonetheless, the rate of the reduction of vigour due to the ageing depends on the prevailing temperature and the seed MC. In this link, Harrington (1972) developed two rules of thumb relating the effects of the seed MC (as a function of the storage atmospheric relative humidity) and the storage temperature to the rate of the ageing of the seed:

- 1) For each 1% increase in the seed moisture content, the life-span of the seed is halved. The rule is valid between 5-14% moisture contents (fresh weight basis) and

- 2) For each 5°C rise in temperature, the lifespan of the seed is halved.

This rule is acceptable amid 0-50°C.

It is vital to note that the total loss of the germination capacity is the ultimate event produced by the ageing process (Roberts and Osei-Bonsu, 1988). This process is partly a function of the reduction in the seed vigour, which might be hastened by the adverse environments.

In summary; for the successful crop production as well as the storage, the seeds possessing the high vigour have the greater opportunity of:

- a. achieving the establishment in the sub-optimal field conditions;
- b. attaining a good crop harvest;
- c. producing a homogeneous crop and
- d. storing well under certain specific conditions.

3. Review on the controlled deteriorations of different seeds

The controlled deterioration technique is globally used to appraise the physiological conditions of various seeds. Different authors worked globally with the seeds of the assorted crops using this technique. So, the review of those lucrative findings has been done critically to gain the knowledge of the broad spectrum with the view to support the discussion critically and citing that in this episode selectively under diverse but suitable captions.

3.1. General review

Kruse (1999) had an experiment on the vigour of different seeds using the controlled deterioration test to estimate the power of that test by applying the assumptions about the nonlinear course made to establish the Ellis and Roberts' seed viability equation. The calculations showed that during the aging period, the divergences in the germination between the seed lots was increased according to the assumed normal distribution while the sampling error of the germination test was augmented as per the binomial distribution.

3.2. Controlled deteriorations of different seeds

3.2.1 Pea seeds

Maristela *et al.* (2007) verified the influence of the low storage temperatures on the electrical conductivity in the seeds of two pea cultivars. The studied temperatures were: 10, 20, 25, 20/10 and 25/10°C. The physiological potentials of the seeds were evaluated at the interval of 3 months during the 18-month storage period using the germination, accelerated aging and electrical conductivity tests. The electrical conductivity test was not a good

indicator for the degree of the seed deterioration stored at the low temperatures. The seed deterioration at 10°C did not seem to be directly related to the loss of the membrane integrity, possibly due to the membrane repair or the reform during the storage at that temperature.

Three pea seed lots of the variety Greenshaft were aged for 0, 2, 4, 8 and 16 days at 12, 16 and 20% MC through the CD at 35°C (Iqbal, 1989). The fates were assessed through the electrical conductivity; standard germination, soil emergence, and seedling evaluation test for the root and shoot lengths and their dry matter contents. Similar patterns of notable declines were recorded in all the cases. But the differences of the seed physiological conditions among the lots were clearly detected under the extreme conditions (16% MC, 16days; or 20% MC, at least 8days). Moreover, highly notable relations were found among all the traits studied. The results vividly showed that the standard germination test might be added-on by a vigour test, the latter of which could be assessed from the root and shoot lengths and/or dry matter contents. Moreover, the suitable limits of the seed vigour for the various traits for the seed quality assurance might also be obtained from the CD. Finally, to save time, the ageing period might be reduced raising the seed MC.

3.2.2 Chick pea seeds

Kapoor *et al.* (2010) watched the accelerated ageing in five varieties of chick pea with the objective of studying the consequences and the relationships of the process of deterioration over the short periods. All the varieties were investigated to consider whether there were genetical differences for

tolerance to the ageing conditions. The effect of the ageing on the seed viability and the seedling vigour were explored. The overall results showed that the seed quality in chick pea deteriorated following the ageing. All the physiological parameters: % germination, seedling root length, shoot length and, therefore, the vigour index in all the varieties were decreased notably. An increase in the MC during the ageing decreased the seed viability, % germination as well as the rate and the vigour were also linked with the biochemical changes associated with the seed ageing. The study also exhibited differential responses amid varieties in terms of the ageing.

Kabir *et al.* (2005) set a study with two chick pea varieties having 3 seed MC (1, 2, 16 and 20%) subjected to 4 ageing periods (0, 4, 8 and 16days) using the controlled deterioration skill at 35°C. Identical patterns of significant degradations of the seed qualities were recorded for all the parameters considered viz. % germination, % seedling emergence, and seedling evaluation for root length, shoot length, number of secondary root lets, and root and shoot dry matter contents. The variety Nabin was better over Hyprocola. The findings proved that the controlled deterioration was an effective technique to study the seed physiology of chick pea. Moreover, the degrees of changes of those attributes could be effectively used for devising the germination and vigour test for seed quality insurance and the clue for developing the variety having the genetically higher seed quality.

Bahadur *et al.* (2005) set an experimentation to ascertain the effects of 5 ageing periods (0, 4, 8, 12 and 16days) having 3 seed MC (12, 16 and 20%)

on the physiological changes of groundnut seed quality. The seeds were deteriorated by taking those in a sealed test tube and kept in a water-bath at 35°C. Different traits pertaining to the seed quality were measured through the standard germination, soil emergence and seedling evaluation test for 7 parameters: root and shoot lengths, root and shoot weights (fresh and dry) and number of root-lets (≥ 1 cm). Correlation matrix was also set. It was found that all the traits were notably affected with the increasing MC and the ageing periods. Nevertheless, as the deterioration conditions were intensified, the seed quality also became poorer identically. Moreover, remarkable links were obtained among most of the studied features.

3.2.3 Cauliflower seeds

The profiles of protein synthesized *in vivo* in the radicle tips were compared among the un-aged, the artificially aged (controlled deterioration: 10% MC at 42°C for 3 weeks) and the artificially aged and subsequently osmoprimed cauliflower seeds (Fujikura *et al.*, 2008). The germination and the initiation of the incorporation of 35 S-methionine into the radicle tips were delayed due to the controlled ageing. The osmopriming accelerated the processes and also alleviated the delay. Proteins were found whose expressions were linked with the rate of the germination and reduced by the ageing process.

3.2.4 Watermelon seeds

Mavi and Demir (2007) compared the emergence of 19 watermelon seed lots of the laboratory germination > 90% in the stressful settings of the low temp. (LTE, early spring), mechanical stress (MSE, 6cm deep sowing) and salt

stress (SSE, 200 mM NaCl). The % emergence ranged amid 12-54 for LTE, 19-75 for MSE and 43-91% in 2005; and 35-80 at LTE, 13-82 at MSE and 45-92% at SSE in 2006. The radicle emergences of the lots were determined after the CD at 40 and 45°C in 2005, or at 45 and 47°C in 2006, with 20 or 24% MC for 48, 72, 96, 120, 144 hours in 2005 and 48, 72, 96, 120, 144, 168 hours in 2006. The accelerated ageing test was also conducted at the same temperatures and the ageing durations in both the years. The germination range of the seed lots was increased in both the CD and the AA as the ageing period was extended.

3.2.5 Cucumber seeds

Demir and Mavi (2008) evaluated the seedling emergence of 16 seed lots (7 in 2005 and 9 in 2006) of cucumber at the low temperature (LTE), mechanical stress (MSE, 8 cm sowing) and salt stress (SSE, 200 mM NaCl) plantings. The emergence percentage ranged from 29-76 for LTE, 46-86, for MSE and 59-92 in 2005 and 35-86 at LTE, 28-83 at MSE and 54-93% at SSE in 2006. The radicle emergence of the lots was determined after the CD at 45°C with 20 or 24% MC for 48, 72, 96, 120, 144h. The accelerated ageing test was also conducted at the same temperature and duration in both the years. The germination after the large number of combinations of the CD and the AA was correlated with LTE, MSE and SSE was noted. The initial laboratory germination was considerably related to LTE, MSE and SSE in 2005 but not in 2006. The optimum CD conditions of 48h and 20% MC at

45°C and the accelerated ageing at 96h and 45°C were suggested as a useful vigour test conditions for the cucumber seeds.

3.2.6 Squash (*C. maxima*) seeds

Kazim and Ibrahim (2005) from a study on the CD test (45°C and 24% MC) assessed the seed vigour of 3 winter squash seed lots. The germination after the CD was correlated with the seedling growth parameters under the salt stress conditions (50, 100, and 150 mM NaCl). The % germination after CD for 24, 48, and 72h were all ≥ 94 , and those treatments were unsuccessful in ranking the lots. Nonetheless, the germination of the seeds after the CD for 96h ranged from 25 - 75% for the 3 lots. The seedling emergence was $> 90\%$ at both 50 and 100 mM NaCl and did not differ among the lots. But at the 150 mM NaCl, the emergence differed among the lots, and the CD for 96h germination were positively and drastically related to the mean seedling emergence time, the seedling fresh and dry weights, the hypocotyl length and the cotyledon width. The CD test could be used to predict the seedling performance of the seed under the salt stress conditions.

3.2.7 Bitter gourd seeds

Hsu *et al.* (2003) had a study on the CD with both the priming (mixing seeds with moist vermiculite for 36h at 20°C) and the hot water soaking (soaking seeds for 4h in water at 40°C) of bitter gourd seeds. The CD improved the percent and the mean emergence time (MET) at the sub-optimal temperature of 20°C. The percent emergence was decreased but the MET was increased by the aging. The percent and the speed of the emergence were squeezed

greatly after the accelerated aging in primed and particularly in the hot water-soaked seeds than in the controlled seeds and finally, retarded the emergence performance of the aged seeds.

3.2.8. Lettuce seeds

Hill *et al.* (2007) conducted an experiment on the seeds of two varieties of lettuce to determine how the priming affected those in relation to the viability equation. The seeds primed in the polyethylene glycol 8000 (-1.45 MPa, 24h at 15°C) and the non-primed seeds were first adjusted to 6 and 9% MC and then stored at 48 and 38°C for up to 30 days, in that sequence. The storage conditions (6% MC and 48°C; 9% MC and 38°C) were predicted by the viability equation to result in the equal longevities. Later, the viability assayed at 20°C revealed that the non-primed seeds in both the storage environments exhibited the similar losses over the time, thus validating the Ellis-Roberts' equation and the use of these conditions to apply different but equal aging stress. Nevertheless, the primed seeds of both the cultivars declined faster than the non-primed ones

Patricia *et al.* (2005) observed that the high seed quality was essential for the optimum stand establishment in lettuce. As a result, it was necessary to have seed vigour tests, which permitted rapid, objective and accurate evaluation of the seed quality. The study evaluated the physical and the physiological seed quality components of 4 seed lots of 6 lettuce varieties. The seeds were evaluated for seedling emergence under the greenhouse conditions, standard germination, seed physiological aspects, the Saturated Salt Accelerated

Aging (SSAA) test and the Seed Vigour Imaging System (SVIS). The results showed that the large-seeded varieties had higher percentage of germination; superior SSAA values; more SVIS indices; and better, rapid and uniform greenhouse emergence. It was finally opined that the SSAA and SVIS tests were the practical and the accurate criterion of knowing lettuce seed quality and to separate the high and the low quality seeds.

3.2.9. Onion seeds

Rodo and Filho (2003) carried out a research work on the vegetable seed vigour to identify the reliable procedures for the accelerated aging and the controlled deterioration tests to rank the onion seed lots according to their physiological potentials. The six seed lots were evaluated in the laboratory for the germination, first count, seedling vigour classification, traditional CD (24% MC, 45°C and 24h) and seedling emergence tests. The CD test was best to assess the physiological potential of the onion seeds, and was advised for the seed quality control.

Khan *et al.* (2004) set a study on the accelerated ageing with the indigenous onion seeds. The seeds exhibited very short life and lost their viability within 1-2 years. The study was also conducted to investigate the vigour and the viability in relation to the germinability and the seed membrane damage. The seeds lost their germinability, vigour and viability progressively with the ageing treatment. The % germination was reduced up to 43%, radicle length 1.7cm and germination speed 6.68-index (GSI). The ageing time and the treatment were taken to get 50% germination, while the leaching from the

viable seeds (control) was negligible. The loss of the viability in seeds after the ageing appeared relating to the increased membrane destruction (loss of membrane integrity). That membrane integrity loss might be responsible for the decreased germinability, vigour and ultimately viability.

3.2.10 Chilli seeds

Basak *et al.* (2006) studied the emergences of 13 commercial seed lots of 6 chilli cultivars. The March sowing seeds gave a low temperature emergence (LTE) from 63 - 93%. The physiological germination (radicle of 2 mm long) of the lots was determined after the CD at 45°C and at 18, 20 and 22% MC for 24, 48, 72 and 96h. The % germinations ranged from 74 - 92% for 24h at 18% MC and 3 and 3 - 93% for 96h at 22% MC and were appreciably and optimistically correlated with LTE (6 out of 12 correlations) and HTE (9 out of 12 correlations). The germination after the CD at the MC 22% for 24h was strongly interrelated with HTE ($r = 0.86$, $P < 0.001$) and LTE ($r = 0.73$, $P < 0.001$). The laboratory germination after 4 and 8-month storage at 5°C and 8% MC were closely correlated (mostly $P < 0.001$) with the CD germinations for all but the 3 CD conditions, more so than was the initial laboratory germination. The mean emergence time (MET) in LTE was inversely related to the CD germination (24h, 22% MC) for 3 lots of the cultivar and 4 lots of other cultivars. The slower emerging lots had the lower germinations.

3.2.11 Canola/rape (*Brassica napus*) seeds

Mohsen *et al.* (2008) conducted a study on the canola seeds to investigate the effects of the accelerated ageing periods (0, 24, 48, 72 and 96h) on the

germination and the seedling growth of 4 cultivars at 40°C and 100% RH. The seeds were placed in the petri dish on the Whatman paper and then set in a germinator at 25°C in dark. The results showed that with the increase in time of the ageing, all the germination parameters became lessened. The aging decreased the quality of the seeds, and resulted in the agricultural and economic losses. Nevertheless, the sensitivity of the cultivars to the ageing periods was different.

3.2.12 Cotton seeds

Cotton seeds were exposed to the high temperature (45-48°C) and relative humidity (80-90%) for 2, 3, 10 and 20 days. The treated and the untreated seeds were judged for various vigour tests. The germination percentage was decreased with the accelerated ageing. There was an increase in the root and the shoot lengths up to 3 days of aging but with the further increase in the accelerated ageing, the seeds failed to germinate (Maqsood *et al.*, 2000).

3.2.13 Barley seeds

Pavlina *et al.* (2006) determined the potential differences in the seed quality of spring and winter barley in relation to varieties (4-winter and 2-spring) and growing conditions (6 locations, 2 years). Three tests were performed on each of the seed sample: (1) the standard germination test; (2) the low temperature and drought stress germination test and (3) the germination test in a salt solution. The germination of the winter varieties was influenced by the provenance, both the measures of vigour by year, and significant discrepancies between varieties were found in all the 3 traits. The spring

varieties differed only in the germination and the vigour in one year when the influence of the provenance prevailed. The results showed different effects of the variety and the provenance on the seed vigour. This could be promising for the breeding of high vigour barley. Some preliminary results were also obtained using the CD as a vigour testing method for barley at 45°C and 24% MC for 24h.

3.2.14 Sunflower seeds

Kibinza *et al.* (2006) investigated whether the loss of the viability of the sunflower seed was affected by the embryo MC during the seed pretreatment at 35°C. The non-dormant seeds were equilibrated at the MC of the embryonic axis ranging from 0.037 - 0.605g H₂O g⁻¹ dry matter for 1 day at 15°C, and kept at 35°C for various durations up to 14 days before the germination were assayed at 15°C. As expected, the higher the MC, the faster was the seed deterioration. There existed a negative linear relationship between the time taken for the germination to drop to 50% (P₅₀) and the embryonic axis MC ranging from 0.108 - 0.438g H₂O g⁻¹ DM. The higher the energy metabolism during the ageing, the lower was the seed viability. The loss of the seed viability was associated with an accumulation of H₂O₂ and malondialdehyde (MDA). When there was a sub-linear relation amid the H₂O₂ content in the embryonic axis and the seed viability, malondialdehyde accumulation only occurred when 50% of the population died within 7 days, i.e. when MC was > 0.248g H₂O g⁻¹ DM.

The transgenic seeds of sunflower showed the high basal thermo tolerance persistence during the early hours of the imbibitions. The seeds also showed the significantly improved resistance to the CD in a stable and transgenic-dependent manner. Furthermore, the over-expression of the HaHSFA9 had no detrimental effects on the plant growth or the development, including the seed morphology and the total seed yield. The results agreed with the previous work tentatively associating the HSP gene expression with the phenotypes important for the seed longevity. The findings might help to improve seed longevity in vital crops (Raúl *et al.*, 2006).

3.2.15 Bixa (*Bixa orellana*) seeds

Nelson *et al.* (2008) predicted the storability of the scarified and the non scarified seeds of bixa through the accelerated aging test. The accelerated aging, carried out at $40 \pm 1^{\circ}\text{C}$ and $98 \pm 2\%$ RH for 20 days revealed that the scarified seeds lost its 50% germination in the 6th day of ageing whereas in the non-scarified seeds the germination % increased up to 4 days of ageing. The study also predicted that the storability of the scarified seeds was lesser than the non-scarified seeds. In the scarified seeds, the % germination became reduced progressively with the advances in the ageing period while in the non scarified seeds; the germination was enhanced initially and then diminished with the advances in the aging period.

3.2.16 Bamboo (*Dendrocalamus strictus*) seeds

Ravikumar *et al.* (2004) investigated the accelerated ageing of bamboo seeds at $42 \pm 1^{\circ}\text{C}$ and 100% RH for 1 -8 days. The seeds lost viability and changed their biochemical constituents. The reductions in the contents of sugars, starch, proteins and lipids were found. The decrease in the activity of the peroxidase as well as the acid and alkaline phosphatase was also noted. The increase in the total free amino acid contents and the activity of the amylase confirmed the degradation of the seed reserves as well as the viability.

3.2.17 Forage (legumes and grasses) species

Wang *et al.* (2004) studied the suitability of the various laboratory vigour tests to rank the quality of commercial seed lots, and to predict the seedling field emergence (FE) of 4 forage species. The CD test generally better indicated seed lot's vigour for the test species than did the standard germination (SG) test. From that study and the previous works on forage species, the author opined that the EC test for forage legumes and the CD tests for both the forage legumes and the grasses had the potentials to be developed as the improved vigour tests for ranking seed-lots' quality and predicting the seeding performances.

CHAPTER III

MATERIALS AND METHODS

The materials utilized and the methods exploited in conducting this work are presented here under different headings including their brief descriptions.

1. Location

The experiment was conducted at the Horticulture Laboratory, Hajee Mohammad Danish Science and Technology University (HSTU), Dinajpur.

2. Duration

The experimentation was carried out during April - June 2010.

3. The seed sources

Three different sources of pea seeds of the local varieties were obtained from the three diverse sources of the northern Bangladesh: Rangpur, Dinajpur and Thakurgoan. The original and the re-designation of those three seed sources are given below:

Original seed sources	Re-designated seed sources
Thakurgoan Local	TL
Dinajpur Local	DL
Rangpur Local	RL

Having the seeds, cleaning was done to remove the cracked, broken and abnormal seeds; other seeds; foreign matters; etc. by hand picking. Then the pure seed fraction was transferred to three separate polyethylene bags, labeled, sealed and kept at the room temperature until exploited.

At the beginning of the experimentation, the following 12 selective traits (Table 1) of the three seed sources were measured to have their comparative bench mark information.

Table 1. Quantitative aspects of the 3 pea seed sources used in the study

Serial nos.	Quantitative aspects considered	Three seed sources		
		TL	DL	RL
1	Initial moisture content (% fresh wt. basis)	12.22	13.30	14.70
2	Thousand seed weights (g)	254.66	241.86	236.55
3	Percent germination (paper towel)	91.00	91.00	89.00
4	Percent soil emergences (course sand)	80.00	79.00	79.00
5	Seedling characteristics (paper towel)			
5. a	Normal seedlings (%)	91.00	91.00	89.00
5. b	Abnormal seedlings (%)	4.00	5.00	4.00
5. c	Death seeds (%)	5.00	4.00	6.00
5. d	Root length (cm)	13.60	13.47	13.35
5. e	Shoot length (cm)	7.13	7.06	6.98
5. f	Root dry matter (g/100 seedlings)	2.62	2.57	2.52
5. g	Shoot dry matter (g/100 seedlings)	2.46	2.38	2.33

4. Experimental design

The completely randomized design (CRD) with three replications was used throughout the experiment.

5. Factors exploited

The three factors and their levels judged in the study were as follows:

Serial nos.	Factors	Levels	Short descriptions
1	Seed sources	3	TL, DL and RL
2	Seed moisture contents (%)	3	12, 16 and 20
3	Ageing periods (days)	3	0, 8 and 16

6. Treatment combinations

The 27 treatment amalgamations compared were as follows (Table 2).

Table 2. The 27 treatment combinations compared for the study

Treatment Combinations	Descriptions of the treatment groupings			
	Seed sources	X	Seed MC (%)	X Ageing periods (days)
TC ₁	RL		12	0
TC ₂	"		"	8
TC ₃	"		"	16
TC ₄	"		16	0
TC ₅	"		"	8
TC ₆	"		"	16
TC ₇	"		20	0
TC ₈	"		"	8
TC ₉	"		"	16
TC ₁₀	TL		12	0
TC ₁₁	"		"	8
TC ₁₂	"		"	16
TC ₁₃	"		16	0
TC ₁₄	"		"	8
TC ₁₅	"		"	16
TC ₁₆	"		20	0
TC ₁₇	"		"	8
TC ₁₈	"		"	16
TC ₁₉	DL		12	0
TC ₂₀	"		"	8
TC ₂₁	"		"	16
TC ₂₂	"		16	0
TC ₂₃	"		"	8
TC ₂₄	"		"	16
TC ₂₅	"		20	0
TC ₂₆	"		"	8
TC ₂₇	"		"	16

7. Number of seeds used per replication

For both the standard germination and the soil emergence tests, randomly selected 100 seeds per replication were used.

8. Determination of the initial moisture content (MC) of the seeds

Three primary seed samples from 3 diverse places of each of the polyethylene bags were taken and the composite samples were coarsely grounded with a hand grinder (Model: G-WON). From this sample, three sub samples (forming the three replications); each consisting of nearly 5g (weighed up to two decimal places) were taken using an electric balance and kept in a 50ml beaker. That was then set in the constant high temperature oven running at $130 \pm 1^{\circ}\text{C}$ for one hour (ISTA, 1999). After that, the beaker was put in a desiccator to cool down. Thirty minutes later, the sample was weighed using the electric balance up to two decimal places. The % MC on the fresh weight basis of the sample was calculated out using the formula-

$$\text{MC (\%)} = \frac{\text{Loss of weight from the seed sample}}{\text{Initial weight of the seed sample}} \times 100$$

Finally, the mean values obtained from the three replicates were used as the moisture contents for each seed source.

9. Adjustment of the seed moisture contents at the desired levels

Firstly, the required number of seeds for each sub-lot was counted and then was weighed to two decimal places. Finally, the expected weight of the seeds at the given MC was calculated out by the formula-

$$EW = \frac{100 \times DW}{100 \times EMC}$$

Here,

EW = Expected weight of the seed at the given moisture content

DW = Dry weight of the seed sample used and

EMC = Expected moisture content to be reached at

Then the adjustment was done in two phases. Of these, the 1st one was applicable to all the sub-lots (sources) and the 2nd one was used as needed.

9.1. Increasing the seed moisture contents

Adequate water was taken in a desiccator until it reached the level of 4cm below its bottom neck. A metallic wire net was set at the neck. On the net, a piece of cotton cloth was placed. The seeds were spread onto the cloth and the lid was set properly. Finally, the desiccator was kept at the room temperature for vapourization of the water. Weighing of the seeds was done time to time to check whether the desired weight was reached by absorbing the water vapour (ISTA, 1999). Under the experimental conditions, it took one to seven days to reach the desired moisture levels of the seeds.

9.2. Decreasing the seed moisture contents

In some cases, the seeds gained slightly higher MC than expected. Hence, the seeds were re-dried. Its method was as follows.

Sufficient quantity of dried blue silica gel (a desiccant) was taken in a desiccator until it reached the level of 4cm below its bottom neck. A metallic wire net was placed at the neck. On the net, a piece of cotton cloth was placed. The seeds were spread onto the cloth and the lid was set on the desiccator properly. Finally, the desiccator was kept at the room temperature.

Time to time weighing of the seeds was done to check whether the desired weight was reached by loosing the moisture from the seeds and absorbing that by the dry silica gel (ISTA, 1999). Under the experimental conditions, it took one to three days to reach the desired moisture levels of the seeds.

10. Process of the controlled deterioration (CD)

The degradation was performed by taking the seeds in the test tubes, sealed, set in beakers containing sufficient water to cover the seed-height, put into an electric oven for the pre-set incubation periods as per the experimental design. The details were as follows:

Treatment wise 100 seeds for each replication were divided into three parts by numbers (33 + 33 + 34). Each part was taken in a test tube (15cm long, 2.5cm internal diameter). A small piece of tissue paper was formed in the shape of a ball and wrapped around with a small piece of polyethylene sheet. Then the mass was put in the mouth of the test tube and pressed sufficiently to make the tube airtight. Such three test tubes were marked with a glass marker as the one replication for each treatment. The test tubes were placed in the beaker (1000ml) and sufficient water was added to cover the seed-height. The beaker was set in an electric oven. The temperature of the oven was adjusted to $35^{\circ}\text{C} \pm 1^{\circ}\text{C}$, which was run for the 3 required incubation periods (0, 8 and 16days) as per the experimental design. During the deterioration, a small quantity of water was added now and then to the beaker to keep the water level high enough to cover the seed-height. So, the whole thing was set to act as a hot water bath.

11. Tests utilized to evaluate the seed quality following the deterioration, parameters noted and the methods of data collection

The following tests were exploited for this purpose:

- A) Standard germination (4 ply rolled paper towel method) for-
 - a. Normal seedlings (vis-à-vis percent germination)
 - b. Abnormal seedlings and
 - c. Dead seeds
- B) Soil emergence (seed-flat containing sterilized coarse sand) and
- C) Seedling evaluation test (7days old normal seedlings obtained from the standard germination test) for:
 - a. Length of root
 - b. Length of shoot
 - c. Dry matter of roots (100 seedlings basis)
 - d. Dry matter of shoots (100 seedlings basis) and
- D) The above 8 parameters were also correlated with one another.

12. Standard germination test

The rolled paper towel method (4 ply) was used for it. In each towel 25 seeds were set centrally lengthwise 2cm apart from one another and then rolled as needed. The towels were then tied loosely with a piece of thread. Those were set on a plastic tray in the upright manner. The tray was kept on the table at the room temperature. Light watering was done as needed with a plastic jet bottle. After 7days, the seedlings were evaluated according to ISTA (1999). For this, data were collected for normal seedlings, abnormal seedlings and dead seeds. The results were then expressed as the percentage and the transformed values were exploited for the statistical analyses.

13. Soil emergence test

The soil emergence test was conducted using the heat-sterilized coarse sand as the substratum. The particulars of that were as follows:

Sufficient coarse sand was sieved to discard all the organic substances, and soil particles > 0.8 and $< 0.05\text{mm}$ in diameter. The sieved sand was sterilized in an electric oven running at 100°C for 24 hours. Later on, the sand was cooled down. A rectangular wooden seed flat ($1.0 \times 0.5 \times 0.15\text{M}$) was filled in with the sterilized sand. Then the seeds were inserted in the sand $5 \times 5\text{cm}$ apart in all the directions and also at the depth of 5cm . In this way 100 seeds were placed in the seed flat for each replication. Light watering was done as needed with a plastic jet bottle. After 14 days of setting the emergence was recorded. Finally, the result was expressed as the percentage and the transformed values were used for the statistical analyses.

14. Seedling (7days old normal seedlings) evaluation test

For this, the normal seedlings from the standard germination test were used. Then the root and shoot lengths, and their dry matters were measured.

Firstly, the normal seedlings were selected. The seed residues were separated from the seedlings with a sharp knife. Roots and shoots were also separated, spread on a table and allowed to wither for two hours to facilitate the measurement of the roots and shoots with a 30cm plastic scale. The roots and the shoots were then taken separately in 50ml beakers and lastly dried in an electric oven at the constant temperature of 85°C for 48 hours (ISTA, 1999). Finally, those were weighed with an electric balance up to the two decimal places and expressed on the 100-seedling basis for the statistical analyses.

15. Statistical analyses

The results were evaluated using the analysis of variance (ANOVA) while the paired means was compared by the 'T' test. In addition, the simple correlation was studied. The MSTAT-C package was exploited for this.

CHAPTER IV

RESULTS AND DISCUSSIONS

The results of the present study being reported are accessible in this chapter under the 9 diverse headings and in the 2 tables. Moreover, the findings are also discussed and supported by the experiences of a range of scientists investigated the quality of different seeds all over the world, specially the legumes using the controlled deterioration technique.

1. Normal seedlings

Due to the controlled deterioration, the % normal seedlings vis-à-vis the % germination was decreased notably at the 5% level of probability (Appendix 1) from as high as 90.67 (TC₁₉) to just 39.00% in (TC₉), i.e., up to 51.67% due to the treatment combinations (Table 3). As the conditions of the ageing, i.e. seed MC from 12-20% and the ageing periods as of 0-16 days were gradually intensified, the fall in the % normal seedlings was also in the same track.

The integrity of the cellular membranes, enzymatic roles, metabolic functions and also translocation activities were increasingly hampered due to the degradation. So, the seeds lost their credibility to produce adequate number of normal seedlings as found in the germination test (Roberts, 1972; Khan *et al.*, 2004). Iqbal and Smith (1996) noted such reduced germination in pea seeds following the controlled declining. Bahadur *et al.* (2005) obtained parallel results in groundnut seeds weakened through the controlled degradation. Kabir *et al.* (2005) and Kapoor *et al.* (2010) recorded the similar happenings too with such an ageing study of chick pea seeds.

Table 3. Various traits obtained from the standard germination and the soil emergence tests of the 3 pea seed sources as influenced by the controlled deterioration technique

Treatment combinations	Natures of the combinations	Normal seedlings (%)	Abnormal seedlings (%)	Dead seeds (%)	Soil emergence (%)
TC ₁	RL M ₁₂ A ₀	88.00 a-d	7.00 m	5.00 i-k	76.67 c-e
TC ₂	RL M ₁₂ A ₈	84.00 ef	10.00 kl	6.00 h-k	73.67 f
TC ₃	RL M ₁₂ A ₁₆	77.00 hi	13.67 h-j	9.33 e-g	66.67 h
TC ₄	RL M ₁₆ A ₀	86.33 c-e	8.00 lm	5.67 h-k	76.00 d-f
TC ₅	RL M ₁₆ A ₈	78.67 h	14.00 h-j	7.33 f-i	67.33 gh
TC ₆	RL M ₁₆ A ₁₆	62.67 l	22.00 c	15.33 c	57.33 jk
TC ₇	RL M ₂₀ A ₀	86.00 de	8.00 lm	6.00 hijk	74.00 f
TC ₈	RL M ₂₀ A ₈	67.33 k	19.00 d-f	13.67 cd	54.67 l
TC ₉	RL M ₂₀ A ₁₆	39.00 o	36.00 a	25.00 a	36.67 m
TC ₁₀	DL M ₁₂ A ₀	89.00 a-c	6.33 m	4.67 i-k	79.33 ab
TC ₁₁	DL M ₁₂ A ₈	86.67 b-e	8.00 lm	5.33 i-k	76.00 d-f
TC ₁₂	DL M ₁₂ A ₁₆	82.00 fg	11.33 jk	6.67 g-j	69.67 g
TC ₁₃	DL M ₁₆ A ₀	89.33 ab	6.67 m	4.00 jk	78.00 a-d
TC ₁₄	DL M ₁₆ A ₈	75.33 i	15.00 g-i	9.67 ef	61.67 i
TC ₁₅	DL M ₁₆ A ₁₆	67.33 k	19.67 c-e	13.00 cd	57.00 jkl
TC ₁₆	DL M ₂₀ A ₀	88.67 a-d	7.67 lm	3.67 k	75.33 ef
TC ₁₇	DL M ₂₀ A ₈	64.33 l	21.33 cd	14.33 cd	55.00 kl
TC ₁₈	DL M ₂₀ A ₁₆	49.67 n	34.33 a	20.33 b	40.67 m
TC ₁₉	TL M ₁₂ A ₀	90.67 a	6.00 m	3.33 k	80.00 a
TC ₂₀	TL M ₁₂ A ₈	88.33 a-d	7.33 lm	4.33 gk	77.00 b-e
TC ₂₁	TL M ₁₂ A ₁₆	86.00 de	8.33 lm	5.67 h-k	76.00 d-f
TC ₂₂	TL M ₁₆ A ₀	88.00 a-d	7.33 m	3.67 k	77.67 a-e
TC ₂₃	TL M ₁₆ A ₈	79.33 gh	12.33 i-k	8.33 f-h	67.33 gh
TC ₂₄	TL M ₁₆ A ₁₆	71.67 j	16.33 f-h	12.00 de	58.00 j
TC ₂₅	TL M ₂₀ A ₀	89.00 a-c	7.00 m	4.67 i-k	79.00 a-c
TC ₂₆	TL M ₂₀ A ₈	71.00 j	17.33 e-g	11.67 de	63.00 i
TC ₂₇	TL M ₂₀ A ₁₆	53.00 m	27.00 b	21.00 b	46.33 m
CV (%)		2.26	11.72	13.70	2.44

DL=Dinajpur Local, TL=Thakurgaon Local, RL=Rangpur Local M=Moisture and A=Ageing.

In a column means bearing the same letter(s) do not differ significantly as per DMRT.

The decline was also prominent in the seed sources though their magnitudes were overall less in TL (TC₁₉ - TC₂₇) gone after by DL (TC₁₀ - TC₁₈) while it was the least in RL (TC₁ - TC₉). These results showed that TL was the best seed among the sources explored. That might be due to the fact that TL had the heaviest thousand seeds, i.e. 254.66 while those were 241.86 and 236.55g in DL and RL, in that order (Table 1). The results are also in full agreement with that of Patricia *et al.* (2005) who showed that the large seeded varieties had higher percentage of germination. Note that the bold seeds in any species are always considered as the high vigorous ones by global seed technologists due to the more food reserve.

The differences in producing the % normal seedlings might also be linked with the heredity of the seed sources (Appendix 1). While working with the ageing of five chick pea varieties, Kapoor *et al.* (2010) found that the % germination declined notably having differential responses amid varieties.

Low seed moisture content is another vital issue for the seed quality. And TL had the least initial MC (12.22) too than the rest 2 sources, i.e. 13.30 and 14.70% for DL and RL, respectively (Table 1). So, it might play another imperative role in varying the % normal seedlings among the 3 sources. Note that, the utmost seed quality may occur at the physiological maturity stage, i.e. when the seed attains the topmost dry weight (Siddique *et al.*, 1987). But the seeds start to deteriorate naturally as soon as those have reached the peak

maturity on the mother plants at the rate depending basically on the seed MC and the prevailing temperature (Roberts and Osei-Bonsu, 1988).

Lastly, the treatment combinations TC₁, TC₄ and TC₇ were statistically parallel among themselves. Similarly, the treatments TC₁₀, TC₁₃ and TC₁₆ were statistically alike with one another. In addition, the amalgamations TC₁₉, TC₂₂ and TC₂₅ were also same too with each other. So, it proved that just rise in the seed MC at the zero incubation had no outcome on this trait.

Finally, all the three seed sources were quite alike for the % normal seedlings under the least stress conditions (12% MC and 0 day incubation in TC₁, TC₁₀ and TC₁₉). But as the adverse situations were most severe (20% MC and 16 days ageing in TC₉, TC₁₈ and TC₂₇), their physiological divergences were clear from the germination percentages (Table 3). So, the results paved the way to opine further that to watch such disparities in the seed sources of very close physiological statuses, the ageing should be done under the extreme conditions. Iqbal and Smith (1996), Bahadur *et al.* (2005) and Kabir *et al.* (2005) also had similar findings regarding this phenomenon.

2. Abnormal seedlings

The percent abnormal seedlings ranged outstandingly at the 1% level of opportunity (Appendix 1) from as high as 36.00% in TC₉ to as low as only 6.00 in TC₁₉, i.e. up to 30.00% on account of the treatment amalgamations (Table 3). The integrity of the cellular membranes, enzymatic reactions, metabolic roles and also translocation activities were increasingly hindered

due to the ageing. So, the seeds lost their credibility to produce adequate normal seedlings yielding more and more abnormal seedlings as found in the germination test (Roberts, 1972; Khan *et al.*, 2004). As the conditions of the deterioration, viz. the seed MC from 12-20% and the ageing periods as of 0-16 days were strengthened, the rise in abnormalities in the 3 sources were also in the same trail. Nonetheless, the magnitude was less in TL i.e. TC₁₉-TC₂₇ (6.00-27.00) gone after by DL i.e. TC₁₀-TC₁₈ (6.33-34.33) while in RL i.e. TC₁-TC₉ it was at the top (7.00-36.00%). The results again revealed that TL was the best one among the 3 seed sources and might happen once more due to the bold seeds and the low initial seed MC as noted before.

Again, Roberts (1986) mentioned that the effects of low vigour would likely be a great problem in those species where the manifestation of deteriorative changes occurred in the seeds well in advance of the death, e.g. soybean: a legume. Because its seeds became defective about 1.7 normal deviates before the death (Ellis and Roberts, 1981). Other legumes also showed relatively high risk of having abnormalities and thereby amplified the oddities for the same trait, e.g. in cowpeas and chickpeas, seeds became defective about 0.9 normal deviates before the death (Osei-Bonsu, 1981). Contrarily, in the cereals, the seeds became faulty and produced abnormalities only a relatively short time before the death: 0.3 normal deviates both in wheat and maize (Ellis and Roberts, 1980). So, the abnormalities in the legumes are quite common than the cereals and other seeds. Bahadur *et al.* (2005) had similar results with the

ageing of the groundnut seeds. Moreover, it is widely accepted that the loss of germination is almost the last stage of the ageing: the final catastrophe i.e. the death as proceed by the more subtle stages (Heydecker, 1972). So, the 3 seed sources had the % more abnormalities than the % dead seeds.

The differences in producing the abnormal seedlings might also be related to the genotypes (Appendix 1). Kapoor *et al.* (2010) dealing with the ageing of 5 chick pea varieties found that the deformities in the varieties were decreased notably having varied responses. Kabir *et al.* (2005) also had such genetic disparities with the controlled weakening of 2 chick pea varieties.

Lastly, the treatments TC₁, TC₄ and TC₇ were statistically identical among themselves. Again, the combinations TC₁₀, TC₁₃ and TC₁₆ were alike with one another. Even, the combinations TC₁₉, TC₂₂ and TC₂₅ were statistically same too with each other. All these 9 treatment combinations had 12-20% seed MC but no pessimistic effects. So, it again proved that just increase in the seed MC at the zero incubation periods had no effect on this trait.

Finally, all the 3 seed sources were alike for the % abnormal seedlings under the least stress (12% MC and 0 day in TC₁, TC₁₀ and TC₁₉). But as the stress conditions were extreme (20% MC and 16days for TC₉, TC₁₈ and TC₂₇), their physiological divergences became clear from the rise in the % abnormalities (Table 3). So, to study such disparities in the seed sources with very close physiological statuses, the ageing must be done under the extreme settings. Iqbal and Smith (1996), Bahadur *et al.* (2005) and Kabir *et al.* (2005) had also similar findings about these consequences.

3. Dead seeds

Due to the CD, the % dead seeds rose up notably at the 1% level of probability ((Appendix 1) from as low as 3.33 (TC₁₉) to as high as 25.00% (TC₉) i.e. up to 21.67% in consequence of the treatments (Table 3). Nonetheless, TC₉ (25.00) was followed by TC₂₇ (21.00) and TC₁₈ (20.33%) and the latter 2 were statistically similar with one another. The amplification in the % dead seeds was also significantly countable in the 3 seed sources although the extents were less in TL i.e. TC₁₉ - TC₂₇ (3.33 - 21.00%) gone after by DL i.e. TC₁₀ - TC₁₈ (4.67 - 20.33%) while it was at the top in the entire sub groups of RL i.e. TC₁ - TC₉ (5.00- 25.00%). These results revealed that in this trait TL stood 2nd while DL was 1st and RL was 3rd among the sources explored. But in terms of all other traits studied, TL was in the 1st position. Again TC₂₇ and TC₁₈ were statistically in the same group.

The reliability of the cellular membranes, enzymatic roles, synthesis activities and also translocation processes were increasingly impaired due to the ageing (Roberts, 1972; Khan *et al.*, 2004). Thus, the seeds lost their reliability to produce any seedlings leading to the rise in the dead toll. Again, Roberts (1986) mentioned that the effects of the low vigour would likely be a great problem in the species where the manifestation of the deteriorative changes occurred in the seeds well in advance of the death, e.g. soybean. Because its seed became defective about 1.7 normal deviates before its death (Ellis and Roberts, 1981). Other legumes also showed relatively high risk of

having abnormalities and thereby amplified oddities for the same trait, e.g. in cowpeas and chickpeas, the seeds became defective about 0.9 normal deviates before the death (Osei-Bonsu, 1981). Contrarily, in the cereals, the seeds became faulty and produced abnormalities only a relatively short time before the death: 0.3 normal deviates both in wheat and maize (Ellis and Roberts, 1980). Moreover, it is widely accepted that the loss of germination is almost the last stage of the deterioration: the final catastrophe i.e. the death as proceed by the more subtle stages (Heydecker, 1972). So, the abnormalities in the legumes are quite common and it might be the main cause of the less % dead seeds than the % abnormal seedlings in all the cases of the 3 sources. Finally, the differences in the % dead seeds amid the 3 sources might also be linked with the genotypes used as stated earlier for germination.

4. Percent soil emergence

The percent emergence vis-à-vis field establishment was declined notably at the 1% level of possibility (Appendix 1) from as high as 80.00 (TC₁₉) to as low as 36.67% (TC₉) i.e. up to 43.33% as a result of the treatments (Table 3). Moreover, TC₉ was statistically at par with TC₁₈ (40.67) and TC₂₇ (46.33%).

As the weakening conditions of the seeds in terms of the seed MC and the ageing days were exaggerated, the drop in the soil emergence was also in the same track. The decline was also remarkable in the 3 sources. But the highest enormity in deprivation was less in TL i.e. 46.33 in TC₂₇ followed by DL i.e. 40.67 in TC₁₈ while in RL it was the least i.e. 36.67% in TC₉. These

results revealed that TL was statistically paramount than the rest 2 sources exploited. That might also happen due to the bold seeds in TL over DL and RL as well (Table 1) as bold seeds are always treated as the high vigorous ones by the global seed scientists as pointed before.

The veracity of the cellular membranes, enzymatic activities, synthesis processes and also translocation roles were increasingly disrupted due to the degradation (Roberts, 1972; Khan *et al.*, 2004). Hence, the ageing also reduced the shoot length (Table 4). Moreover, the weak seedlings might not have sufficient strength to come out of the sand from the 5cm depth giving the poor % soil emergence. Iqbal and Smith (1996), Bahadur *et al.* (2005), Kabir *et al.* (2005) and Kapoor *et al.* (2010) working with the ageing of different legume seeds had also identical observations. Furthermore, there could be differential responses amid genotypes. Low seed MC is another factor for quality seeds. And TL had also the least initial seed MC than the rest 2 sources. That issue might also have a vital role for disparities in the % soil emergence capacities as cited previously for the 4 traits.

5. Root length

The root was stunted markedly at the 1% level of possibility (Appendix 1) from as high as 12.98 (100.00) in TC₂₂ to as down as 9.14cm (70.41%) in TC₉ i.e. up to 29.58 % on account of the treatments (Table 4). Nonetheless, TC₂₂ was in the same statistical group of TC₁₉ (12.97), TC₂₅ (12.97), TC₁₀ (12.91) as well as TC₁₃ (12.90cm). As the weakening conditions of the seeds, i.e. the seed MC and the ageing days were embellished, the depletion in the root length also happened in the same manner.

Table 4: Evaluation of the seedling parameters of the 3 pea seed sources as influenced by the controlled deterioration skill

Treatment combinations	Natures of the combinations	Root length (cm)	Shoot length (cm)	Root dry matter (g)	Shoot dry matter (g)
TC ₁	RL M ₁₂ A ₀	12.82b-d	6.37b	2.40c	2.19c
TC ₂	RL M ₁₂ A ₈	12.20g	6.30b-d	2.24f	2.06f
TC ₃	RL M ₁₂ A ₁₆	11.12n	6.04hi	2.05h	1.88i
TC ₄	RL M ₁₆ A ₀	12.80d	6.34bc	2.39cd	2.18cd
TC ₅	RL M ₁₆ A ₈	11.36kl	6.11fg	2.16g	1.88i
TC ₆	RL M ₁₆ A ₁₆	10.22r	5.54m	1.92j	1.77k
TC ₇	RL M ₂₀ A ₀	12.81cd	6.31b-d	2.39c-e	2.17cd
TC ₈	RL M ₂₀ A ₈	11.24m	5.70l	2.09h	1.79jk
TC ₉	RL M ₂₀ A ₁₆	9.41r	4.68p	1.80k	1.54m
TC ₁₀	DL M ₁₂ A ₀	12.91ab	6.36b	2.52b	2.27b
TC ₁₁	DL M ₁₂ A ₈	12.40f	6.22e	2.34de	2.11ef
TC ₁₂	DL M ₁₂ A ₁₆	11.31lm	5.79jk	2.09h	1.90i
TC ₁₃	DL M ₁₆ A ₀	12.90abc	6.36b	2.55b	2.28b
TC ₁₄	DL M ₁₆ A ₈	11.29i	6.12f	2.26f	2.07f
TC ₁₅	DL M ₁₆ A ₁₆	9.55p	5.73kl	1.98l	1.82j
TC ₁₆	DL M ₂₀ A ₀	12.84b-d	6.28c-e	2.52b	1.28b
TC ₁₇	DL M ₂₀ A ₈	11.45jk	5.98i	2.17g	1.98g
TC ₁₈	DL M ₂₀ A ₁₆	9.37q	4.88o	1.93ij	1.55m
TC ₁₉	TL M ₁₂ A ₀	12.97a	6.47a	2.63a	2.33a
TC ₂₀	TL M ₁₂ A ₈	12.56e	6.32bc	2.40c	2.13de
TC ₂₁	TL M ₁₂ A ₁₆	11.38j-l	6.05g-i	2.17g	1.96gh
TC ₂₂	TL M ₁₆ A ₀	12.98a	6.47a	2.62a	2.32ab
TC ₂₃	TL M ₁₆ A ₈	11.70h	6.25de	2.34e	2.10ef
TC ₂₄	TL M ₁₆ A ₁₆	10.67o	5.86j	2.15g	1.92hi
TC ₂₅	TL M ₂₀ A ₀	12.97a	6.47a	2.63a	2.31ab
TC ₂₆	TL M ₂₀ A ₈	11.46j	6.08f-h	2.43c	2.00ab
TC ₂₇	TL M ₂₀ A ₁₆	9.51p	5.11n	2.14g	1.59l
CV (%)		0.66	0.66	0.39	0.39

DL= Dinajpur Local, TL= Thakurgaon Local, RL= Rangpur Local M= Moisture and A= Ageing. In a column means bearing the same letter(s) do not differ significantly as per DMRT.

Moreover, the reliability of the cellular membranes, metabolic processes, enzymatic roles and the translocation actions were even more damaged due to the weakening (Roberts, 1972; Khan *et al.*, 2004). So, the seedlings from such seeds could lose their capability to produce long roots. Iqbal and Smith (1996) working with the ageing of different legume seeds also found notable decrease in the root length.

Furthermore, the differences in the root length might also be linked with the heritable natures of the seed sources. Bahadur *et al.* (2005), Kabir *et al.* (2005) and Kapoor *et al.* (2010) working with the ageing of different legume seeds also found notable decrease in the root length in different varieties.

The declining trend was also remarkable in the 3 sources. But it was least in TL i.e. TC₁₉ - TC₂₇ from 12.97 - 9.51 cm than the others. The results further established that TL was statistically superior to the rest 2 sources.

Finally, all the seed sources were fairly similar for their root lengths under the least stress conditions (12% MC and 0 day incubation in TC₁, TC₁₀ and TC₁₉). But as the adverse conditions were most severe (20% MC and 16 days ageing for TC₉, TC₁₈ and TC₂₇), their physiological divergences were clear from their root lengths (Table 4). But the roots became stunted notably even at 8 days of ageing irrespective of the MC. So, the results set the base to opine further that to watch such disparities in the roots of the seed sources of very alike physiological habits, the ageing should be done under the medium to the extreme situations. Iqbal and Smith (1996), Bahadur *et al.* (2005) and Kabir *et al.* (2005) also had similar arguments about this phenomenon.

6. Shoot length

The shoot became dwarf distinctly at the 5% level of possibility (Appendix II) from as high as 6.47 in TC₂₅ (and TC₂₂) to as low as 4.68cm in TC₉ i.e. up to 27.66% because of the treatments (Table 4). As the weakening conditions of the seeds, i.e. the seed MC and the ageing days were inflated, the depletion in the shoot length also occurred in the same manner. Again, the reliability of the cellular membranes, enzymatic activities, metabolic paths and also translocation routes were ever more damaged due to the worsening (Roberts, 1972; Khan *et al.*, 2004). So, the seedlings from such weak seeds could lose their competence to have the long shoots. Iqbal and Smith (1996), Bahadur *et al.* (2005), Kabir *et al.* (2005) and Kapoor *et al.* (2010) working with the ageing of different legume seeds also found notable decreases in the shoot lengths. Again, the differences might also be associated with the seed sizes and the inherent qualities of the seed sources utilized as narrated previously in connection with the other parameters.

The declining trend was also remarkable in the 3 pea seed sources. But the reduction was least in TL i.e. TC₁₉ - TC₂₇ from 6.47 - 5.11 compared to those of DL i.e. TC₁₀-TC₁₈ from 6.36-4.88 and RL i.e. TC₁-TC₉ from 6.37-4.68cm; the results further confirmed that the seed source TL was quite superior to the rest 2 supplies.

Finally, all the 3 seed sources were rather identical for the shoot length under the least stress conditions (12% MC and 0 day incubation in TC₁, TC₁₀ and TC₁₉). But as the adverse conditions were most severe (20% MC and 16 days ageing for TC₉, TC₁₈ and TC₂₇), their physiological divergences were clear from their shoot lengths (Table 4). So, the results founded the pedestal to conclude again that to study such disparities in the seed sources of very alike physiological natures, the ageing should be done under the severe stresses. Iqbal and Smith (1996), Bahadur *et al.* (2005) and Kabir *et al.* (2005) also had similar arguments about this occurrence.

7. Root dry matter

The root dry matter also went down distinctly at the 1% level of possibility (Appendix 11) from as high as 2.63 in TC₂₅ to as downhill as 1.80mg in TC₉ i.e. up to 31.56% owing to the treatment amalgamations (Table 4). As the weakening conditions of the seeds, i.e. the seed MC and the ageing days were exaggerated, the depletion in the root dry matter synthesis also occurred in the same style. Again, the reliability of the cellular membranes, enzymatic functions, translocation roles and also metabolic processes were ever more damaged due to the ageing (Roberts, 1972; Khan *et al.*, 2004). So, the seedlings from such seeds could loss their competence to the long roots as well as the higher root dry matter accumulation. Again, the divergences in the root dry matter might also be deeply associated with the genotypes of the 3 seed sources explored. Iqbal (1989), Bahadur *et al.* (2005), Kabir *et al.*

(2005) and Kapoor *et al.* (2010) working with the ageing of various legume seeds also found notable decrease in it.

The declining trend was also remarkable in the 3 sources. But the fall was least in TL i.e. TC₁₉ - TC₂₇ from 2.63-2.14mg compared to others. These results notably confirmed that TL was statistically finer than the rest 2 cases.

Finally, all the 3 seed sources were somewhat akin for the root dry matters under the least stresses (12% MC and 0 day ageing in TC₁, TC₁₀ and TC₁₉). But as the adverse conditions were most severe (20% MC and 16 days ageing for TC₉, TC₁₈ and TC₂₇), their physiological divergences were clear from their root dry matter build-up (Table 4). So, the results gave the foot to opine additionally that to judge such disparities in the seed sources of very close physiological natures, the degradation should be done under the severe settings. Iqbal and Smith (1996), Bahadur *et al.* (2005) and Kabir *et al.* (2005) also had similar attitudes regarding this happening.

8. Shoot dry matter

The shoot dry matter became markedly less at the 5% level of possibility (Appendix 11) from as much as 2.33 in TC₁₉ (100%) to as poor as 1.54mg (66.09%) in TC₉ i.e. up to 33.90% due to the treatments studied (Table 4).

As the ageing conditions of the seeds, i.e. the seed MC and the incubation days were inflated, the lessening in the gathering of shoot dry matter also occurred in the same line. As the integrity of the cellular membranes, enzymatic reactions, metabolic functions and also translocation activities

were gradually slowed down owing to the deprivation (Roberts, 1972; Khan *et al.*, 2004), the seeds could lose their competence to have long shoots and consequently bound to produce less amount of dry matter in it. Iqbal and Smith (1996), Bahadur *et al.* (2005), Kabir *et al.* (2005) and Kapoor *et al.* (2010) working with the ageing of diverse legume seeds also found notable decrease in the shoot dry matters. Again, the differences in its dry matter might also be governed by the genotypes of the seed sources explored.

The declining trend was also amazing in the 3 sources. But the rates were slightest in TL i.e. TC₁₉ - TC₂₇ from 2.33 - 1.59mg compared to the others. The results finally established that the seed source TL was clearly superior among the 3 seed sources exploited in the present experimentation.

Again, all the 3 seed sources were quite parallel for the shoot dry matter accumulations under the least adverse situations (12% MC and 0 day ageing in TC₁, TC₁₀ and TC₁₉). But as the stress conditions were most intensified (20% MC and 16 days ageing for TC₉, TC₁₈ and TC₂₇), their physiological divergences were vivid from their shoot dry matter production (Table 4). So, the results opened the avenue to remark again that to assess such disparities in the seed sources of very identical physiological ranks, the weakening should be done under the extreme settings. Similar experiences pertaining to this phenomenon had also been focused by Iqbal and Smith (1996), Bahadur *et al.* (2005) and Kabir *et al.* (2005) working with the controlled weakening of diverse legume seeds.

9. Correlation studies

Highly significant ($P > 0.001$) associations were found between all the parameters compared and the figures ranged, overall, from -0.9847 to 0.7060 (Table 5). Of those relationships, 12 were negative while 16 were positive. Out of those, the topmost positive value was 0.9828 amid the dead seeds x abnormal seedlings while the least positive figure was 0.7060 for the shoot dry matter x normal seedlings. On the other hand, the utmost negative value was -0.9847 for the soil emergence x abnormal seedlings but the poorest negative integer was -0.7908 in case of the shoot dry matter x dead seeds. Another vital finding was that among all the values pertaining to the normal seedlings (i.e. germination percentage), its affiliation with the soil emergence was at the climax, i.e. 0.9623. So, it vividly pinpointed that the standard germination test is really an effective indication of the field emergence. While dealing with the CD of pea seeds, Iqbal and Smith (1996) also found such positive and negative affinities among the different studied parameters. And the common findings for Iqbal (1989) and the present study were of similar natures. Bahadur *et al.* (2005) dealing with the ageing of groundnut seeds had also found identical associations among the common traits. Roberts and Osei-Bonsu (1988) argued that when care was taken to assess the liaison in various biologically meaningful terms, it was clear that most of the vital attributes of seed vigour were closely associated to one another.

Table 5. Correlation matrix for the 8 quantitative aspects of the 3 pea seed sources studied following the controlled deterioration skill

	Normal seedling	Abnormal seedling	Dead seed	Soil emergence	Shoot length	Root length	Shoot dry matter
Abnormal seedling	-0.9575						
Dead seed	-0.9632	0.9828					
Soil emergence	0.9623	-0.9847	-0.9821				
Shoot length	0.9033	-0.9488	-0.9444	0.9329			
Root length	0.7745	-0.8599	-0.8629	0.8821	0.8619		
Shoot dry matter	0.7060	-0.7954	-0.7908	0.8135	0.8321	0.9212	
Root dry matter	0.8330	-0.9061	-0.9057	0.9108	0.9373	0.9418	0.9302

Note: all the values are highly significant ($P < 0.001$)

CHAPTER V

SUMMARY AND CONCLUSIONS

A three-factor laboratory study was set during April - June 2010 to assess the consequences of the controlled deterioration on the pea seeds of 3 sources: Rangpur Local (RL), Dinajpur Local (DL) and Thakurgaon Local (TL). The seeds were deteriorated under the controlled situation for the 3 ageing periods (0, 8 and 16 days) having the 3 seed moisture contents (12, 16 and 20% MC) but aged only at 35°C. The outcomes judged using the 8 parameters were: standard germination (rolled paper towel method), soil emergence (sterilized coarse sand in a seed flat) and seedling evaluation test for their root and shoot lengths as well as the dry matter contents. Moreover, simple correlation coefficients were also found out amid different pairs of traits.

The aims of the study were to interpret the phenomena of the physiological ageing of pea seed in respect of the quality and to relate those to the current legal quality requirements assessed using the standard germination test conducted under the controlled laboratory conditions, which is globally used on the schedule basis for any seed quality certification program.

From the standard germination test viewpoint, all the 3 seed sources studied were the quite pleasing ones; initially those were nearly identical in the % germination (89.00-91.00) and almost same type of quantitative information was also noted within the control sub-lots (0 day of incubation irrespective of the seed MC) for not only the standard germination, but also with the soil emergence test. So, on the bases of the results obtained from the standard

germination test, the 3 seed sources were reasonably okay. In contrast, when their shoot and root lengths as well as the root dry matter contents were considered, differences were clearly notable even with no incubation.

As the 3 seed sources having almost parallel germination percentages were subjected to CD, those started to decline at different rates measured by the 8 diverse parameters. These differences were least in all the cases where both the seed MC and the ageing periods were low. So, the mild adverse conditions failed to predict their differences in their physiological statuses as those were very close. Nonetheless, when the seeds were subjected to the CD under the more adverse conditions in terms of the higher MC and the longer ageing period, the variations became quite clear. Hence, for the seed sources sharing more or less the comparable variability in terms of % germination, the severe adverse conditions were needed to distinguish those on their physiological bases: in the majority of the cases, deterioration for 16 days and at 20% MC, were required.

From the results it was fairly clear that the seed source TL was the most vigorous one. On the contrary, the source DL occupied the second place while the source RL was the last one in the queue in terms of the quality. Such significant positions could probably be due to disparities in the thousand seed-weights: 254.66, 241.86 and 236.55g in those sources, respectively. Note that the bold sized seeds in any crop variety are always considered the high vigorous ones by the seed technologist all over the globe and so, sieving of seeds with screens of slots of variable dimensions is done during their processing to have only those ones.

Highly significant ($P > 0.001$) correlations were found between all the 8 parameters studied and the figures ranged, overall, from -0.9847 to 0.7060 (Table 5). Of those relationships, 16 were positive while 12 were negative. Again, among the 7 values for the normal seedlings, the topmost figure was 0.9623 with the soil emergence indicating that the standard germination test is an effective mirror of the field emergence.

The results further pointed out that all the tests performed during the course of the study gave comparable as well as consistent consequences. As such, the uses of all other tests might be limited to specific situations where those either substitute the standard germination test, or complement it. For example, in the developing countries where the labour is cheap as well as plentiful but funding, equipment and technical hands are limited, the same seeds could be tested using the standard germination test and the normal seedlings obtained from that test could then be evaluated for their root and shoot characteristics. So, the controlled deterioration technique is a unique skill to study the physiological statuses of the seeds even having initially very close natures.

Nonetheless, further research works should be done for the consistency of the acceptable limits of each of the parameter for the seeds of all the cultivated varieties of pea as those traits might differ among themselves. Finally, to save time, the ageing period could also be squeezed by increasing the seed moisture contents.

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APPENDICES

Appendix I: Analysis of variance of the standard germination of 3 seed source as affected by controlled deterioration

Sources of variations	DF	Mean Square Values			
		Normal seeding	Abnormal seedling	Dead seed	Soil emergence
S	2	192.05 ^{**}	68.48 ^{**}	29.64 ^{**}	125.15 ^{**}
M	2	2240.91 ^{**}	831.15 ^{**}	412.79 ^{**}	1985.59 ^{**}
S × M	4	5.79 ^{NS}	5.52 ^{NS}	1.61 ^{NS}	22.13 ^{**}
A	2	3560.49 ^{**}	1275.15 ^{**}	642.08 ^{**}	3063.59 ^{**}
S × A	4	63.38 ^{**}	26.35 ^{**}	10.01 ^{**}	11.02 ^{**}
M × A	4	626.24 ^{**}	238.13 ^{**}	118.38 ^{**}	461.02 ^{**}
S × M × A	8	8.43 [*]	2.39 ^{**}	1.92 ^{**}	10.86 ^{**}
Error	54	3.04	2.69	2.67	2.63 ^{**}

* = Significant at 5% level

**= Significant at 1% level

^{NS} = Not significant

S = Seed Source

M = Seed moisture content

A = Ageing period

Appendix II: Analysis of variance of the seedling traits of 3 seed source as affected by controlled deterioration technique

Sources of variations	DF	Mean Square Values			
		Shoot length	Root length	Shoot dry matter	Root dry matter
S	2	0.247 ^{**}	0.487 ^{**}	0.355 ^{**}	0.128 ^{**}
M	2	1.742 ^{**}	3.433 ^{**}	0.048 ^{**}	0.235 ^{**}
S × M	4	0.057 ^{**}	0.007 [*]	0.013 ^{**}	0.004 ^{NS}
A	2	5.235 ^{**}	32.385 ^{**}	1.635 ^{**}	1.613 ^{**}
S × A	4	0.012 ^{**}	0.027 ^{**}	0.003 [*]	0.007 ^{**}
M × A	4	0.684 ^{**}	0.815 ^{**}	0.012 ^{**}	0.077 ^{**}
S × M × A	8	0.020 ^{**}	0.035 ^{**}	0.024 ^{**}	0.022 ^{**}
Error	54	0.002	0.003	0.001	0.001

* = Significant at 5% level

**= Significant at 1% level

^{NS} = Not significant

S = Seed Source

M = Seed moisture content

A = Ageing period