IDENTIFICATION OF ACCELERATED AGING CONDITIONS FOR SEED VIGOR TEST IN RICE (*Oryza sativa* L.)

Suraj Chhetri

A Thesis Submitted in Partial Fulfillment of the Requirements for the

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การตรวจสอบความแข็งแรงเมล็ดพันธุ์ข้าวโดยวิธีเร่งอายุ

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วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรมหาบัณฑิต

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IDENTIFICATION OF ACCELERATED AGING CONDITIONS FOR SEED VIGOR TEST IN RICE (*Oryza sativa* L.)

Suranaree University of Technology has approved this thesis submitted in partial fulfillment of the requirements for a Master's Degree.

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SURAJ CHHETRI : การตรวจสอบความแข็งแรงเมล็ดพันธุ์ข้าวโดยวิธีเร่งอายุ [IDENTIFICATION OF ACCELERATED AGING CONDITIONS FOR SEED VIGOR TEST IN RICE (*Oryza sativa* L.)] อาจารย์ที่ปรึกษา : ผู้ช่วยศาสตราจารย์ ดร.ธวัชชัย ทีฆชุณหเถียร, 115 หน้า.

วิธีการตรวจสอบความแข็งแรงเมล็ดพันธุ์ข้าวโดยวิธีเร่งอายุที่ใช้ในประเทศไทยยังขาดความแม่นยำและไม่พบคำแนะนำของอุณหภูมิและระยะเวลาการเร่งอายุเมล็ดพันธุ์ข้าวในคู่มือการตรวจสอบความแข็งแรงเมล็ดพันธุ์ในระดับนานาชาติ วัตถุประสงค์หลักของการวิจัยครั้งนี้คือการตรวจสอบหาอุณหภูมิและระยะเวลาของการตรวจสอบความแข็งแรงเมล็ดพันธุ์ข้าวโดยวิธีเร่งอายุที่มีความแม่นยำ ส่วนวัตถุประสงค์รองคือการสำรวจหาวิธีอื่น ๆ ที่อาจมีประสิทธิภาพในการตรวจหาความแข็งแรงของเมล็ดพันธุ์ข้าว ทำการตรวจสอบคุณภาพเมล็ดพันธุ์ข้าวที่นิยมปลูกในประเทศไทย 3 พันธุ์ (ขาวดอกมะลิ 105 ชัยนาท 1 และ พิษณุโลก 1) จำนวน 24 ตัวอย่าง จากศูนย์เมล็ดพันธุ์ข้าวและศูนย์วิจัยข้าวทั่วประเทศไทยที่มีความแข็งแรงแตกต่างกัน ทำการตรวจสอบคุณภาพเมล็ดพันธุ์โดยวิธีตรวจสอบความงอกมาตรฐาน ความยาวรากต้นอ่อน ความยาวยอดต้นอ่อน ความยาวของต้นอ่อน ความงอกในแปลงปลูก ตรวจสอบน้ำหนักแห้งของต้นอ่อน ค่าการนำไฟฟ้าของสารละลาย และวิธีการเร่งอายุเมล็ดพันธุ์รวม 9 วิธีการ ที่อุณหภูมิ 42, 43 และ 44 องศาเซลเซียส ระยะเวลา 72, 96 และ 120 ชั่วโมง ผลการทดลองพบความแตกต่างของเปอร์เซ็นต์ความงอกและความแข็งแรงในกลุ่มของตัวอย่างเมล็ดพันธุ์ที่นำมาตรวจสอบ การวิเคราะห์ค่าสหสัมพันธ์ (r) พบว่าวิธีการตรวจสอบทุกวิธีการมีสหสัมพันธ์กันอย่างมีนัยสำคัญทางสถิติกับความงอกในแปลงปลูกโดยมีค่าสหสัมพันธ์ตั้งแต่ 0.55** ถึง 0.82** วิธีการตรวจสอบความแข็งแรงที่มีค่าสหสัมพันธ์กับความงอกในแปลงปลูกสูงที่สุด 3 วิธีการ ได้แก่ วิธีวัดค่าการนำไฟฟ้าของสารละลาย (r = -0.82**) วิธีเร่งอายุที่ 44 องศาเซลเซียส 72 ชั่วโมง (r = 0.78**) และวิธีวัดความยาวยอดของต้นอ่อน (r = 0.75**) ตามลำดับ เมื่อเปรียบเทียบระหว่างวิธีการวัดค่าการนำไฟฟ้าของสารละลายกับวิธีการเร่งอายุ 9 วิธีการ พบว่าค่าการนำไฟฟ้าของสารละลายมีค่าสหสัมพันธ์มากที่สุดกับวิธีการเร่งอายุที่ 44 องศาเซลเซียส 72 ชั่วโมง (r = -0.71**) ดังนั้น ในเบื้องต้นนี้สามารถแนะนำให้ใช้วิธีเร่งอายุที่ 44 องศาเซลเซียส 72 ชั่วโมง และวิธีวัดค่าการนำไฟฟ้าของสารละลายเป็นวิธีตรวจสอบความแข็งแรงเมล็ดพันธุ์ข้าวของประเทศไทย จากนั้นทำการทดลองสอบเทียบคำแนะนำดังกล่าวโดยใช้เมล็ดพันธุ์ข้าว 9 พันธุ์ จำนวน 60 ตัวอย่าง จากศูนย์เมล็ดพันธุ์ข้าวและศูนย์วิจัยข้าวทั่วประเทศ ผลการตรวจสอบคุณภาพเมล็ดพันธุ์ข้าวและวิเคราะห์ค่าสหสัมพันธ์เป็นไปในทางเดียวกันกับที่กล่าวมาแล้ว แต่พบว่ามีค่าสหสัมพันธ์สูงกว่าเดิม โดยค่าสหสัมพันธ์ความงอกในแปลงปลูกกับวิธีเร่งอายุที่ 44 องศาเซลเซียส 72 ชั่วโมง และค่าการนำ

ไฟฟ้าของสารละลาย เท่ากับ 0.89** และ -0.86** ตามลำดับ ส่วนค่าสหสัมพันธ์ระหว่างวิธีการเร่งอายุดังกล่าวกับค่าการนำไฟฟ้าของสารละลายเท่ากับ -0.77**

สาขาวิชาเทคโนโลยีการผลิตพืช ลายมือชื่อนักศึกษา_____

ปีการศึกษา 2552 ลายมือชื่ออาจารย์ที่ปรึกษา_____

SURAJ CHHETRI : IDENTIFICATION OF ACCELERATED AGING CONDITIONS FOR SEED VIGOR TEST IN RICE (*Oryza sativa* L.). THESIS ADVISOR : ASST. PROF. THAWATCHAI TEEKACHUNHATEAN, Ph.D., 115 PP.

RICE SEEDS/SEED VIGOR TEST/ACCELERATED AGING TEST/ CONDUCTIVITY TEST

The accelerated aging conditions commonly practiced in Thailand as seed vigor test in rice is not accurate and no recommended accelerated aging test conditions for rice seeds have been prescribed in international seed vigor testing handbooks. The main objective of this study was to investigate accurate combinations of temperature and time for the accelerated aging conditions for seed vigor test in rice. The minor objective was to explore other efficient vigor tests for rice seeds other than accelerated aging test. Twenty four rice seed lots of 3 common Thai varieties (Khao Dok Mali 105, Chai Nat 1, and Phitsanulok 1) of different vigor levels from different rice seed centers and rice research centers in Thailand were used in the experiments. The following tests were conducted: standard germination, seedling root length, seedling shoot length, total seedling length, field emergence, seedling growth rate, conductivity and accelerated aging at 42, 43 and 44^{o}C for 72, 96 and 120 hrs at each temperature. In the multiple correlation analyses, all tests showed highly significant correlations with the field emergence (r = 0.55** to 0.82**). The three single vigor tests that provided highest correlations with the field emergence were conductivity test (r = -0.82**), accelerated aging at 44^{o}C for 72 hrs (r = 0.78**) and seedling shoot length (r = 0.75**), respectively. Among the conductivity test and the 9 accelerated aging

conditions, the highest correlation (r = -0.71**) was observed between conductivity and accelerated aging at 44^{o}C for 72 hrs. Based on the result obtained, it can be concluded that the accelerated aging conditions at 44^{o}C for 72 hrs and conductivity test should be a preliminary recommendation for a vigor test in rice seeds of Thai varieties. Sixty seed lots of 10 rice varieties from different rice seed centers and rice research centers in Thailand were used to verify the above recommendation. The results were similar to the above findings, except that the correlations were higher among the tests. The results showed highly significant correlations between the accelerated aging test at 44^{o}C for 72 hours and the field emergence (r = 0.89**); and the conductivity test and the field emergence (r = -0.86**). The result also demonstrated highly significant correlation between the accelerated aging test at 44^{o}C for 72 hours and the conductivity test (r = -0.77**).

School of Crop Production Technology Student's Signature_____

Academic Year 2009 Advisor's Signature_____

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LIST OF ABBREVIATIONS

AA	=	Accelerated aging test
AOSA	=	Association of Official Seed Analysts
CNT	=	Chai Nat
CT	=	Conductivity test
DS	=	Dry season
ES	=	Extension seed
FE	=	Field emergence
FS	=	Foundation seed
ISTA	=	International Seed Testing Association
KDML	=	Khao Dok Mali
PSL	=	Phitsanulok
RL	=	Seedling root length
RS	=	Rainy season
SG	=	Standard germination test
SGR	=	Seedling growth rate test
SL	=	Seedling shoot length
SMP	=	Sangyod Muang Phatthalung
SPR	=	Suphan Buri
SPT	=	San Pa Tong
TL	=	Total seedling length

CHAPTER I

INTRODUCTION

1. 1 Rational

Rice is life for thousands of millions of people. In Asia alone, more than 2,000 million people obtain 60 to 70% of their calories from rice and its products (FAO, 2004). Nearly 90% of the world's rice is produced and consumed in Asia. Only about 9% of the total world harvest is produced outside Asia (FAO, 1998). Rice along with wheat and maize comprises of at least 75 percent of grain production. The world population is expected to increase by 3.68 billion between 1995 and 2050 and Asia will contribute some 2 billion**.** By the year 2025, about 760 million tons of paddy needs to be produced to meet the increasing demand due to increase in world population, this requirement is 35 percent more than actual rice production (Duwayri et al., 2000).

Rice is the most important food crop in Thailand. The total area under rice is estimated to be about 11 million ha representing approximately 40 percent of the cropped land area (Kupkanchanakul, 2000). Despite Thailand being one of the world’s largest rice producers and also the world's biggest rice exporter (FAO 2004), it still can be mentioned that the average yield as compare to other countries is much lower. The average yield is often less than 2,000 kg/ha (Kupkanchanakul, 2000) which is much less than other countries like Egypt (8,250 kg/ha), Australia (8,230), USA. (6,740 kg/ha) and China (6,170 kg/ha) (Duwayri, 2000). The attainment of high yields depends on many factors: variety, land preparation, weed control, fertilizer, disease

and insect control and seed quality. Furthermore, the need for high quality seeds has increased in recent years. Information about seed quality can benefit farmers in making decisions regarding the cost of seeds, quantity of seeds to plant, time of planting, and the need to replant.

One of the potential means of enhancing rice productivity is to ensure the quality of seeds for sowing. A good quality seeds are often free from diseases and posses high germinability and vigor. The benefits of various production inputs and improved farming technologies can be gained only when highly vigorous seeds are used for sowing. In the Philippines, use of quality seeds increased the rice yield to 12.6% (FAO, 2000). Since rice is self-pollinated therefore achievement of pure seed is relatively easy but obtaining high vigor seeds needs scientific managements and proper techniques for harvesting, processing, treatment, and storage (Seshu et al., 1987).

Rapid seed deterioration during storage is one of the important constraints encountered by rice growing farmers. Most of the seed companies and rice seed centers produce rice seeds and keep in stock for sell to the farmers. The storage seeds are at least kept for 6 months to 1 year in the warehouses. Although the germination test are being carried out, but test results are obtain from seeds which have been placed under favorable germination conditions. Seldom are these favorable conditions encountered in the fields and germination results often overestimate field emergence (AOSA, 1983). Therefore, it is of utmost important that seed vigor test is being carried out in time to time to check the seed quality and for proper disposal of storage seeds to prevent seed deterioration resulting into poor stand in actual field conditions.

Seed vigor is an index of seed quality. Seeds that have high vigor will have high quality. Seed vigor not only influences the productivity but also the storability of

seeds. Vigorous seeds can store well, produce uniform stands, develop into vigorous, and productive plants. To satisfy the need for a quick estimate of seed vigor in seed programs, many vigor tests have been developed. Of all vigor tests in use today, one of the most important is accelerated aging test.

Accelerated aging test is a stress test. The seeds are stressed prior to the germination test. Seeds are placed in temperature of 40-45°C and nearly 100% relative humidity for varying lengths of time, depending on the kinds of seeds, after which a germination test is made. The basis for this test is that higher vigor seeds tolerate the high temperature-high humidity treatment and thus retain their capability to produce normal seedlings in the germination test. It was first developed by Delouche (1965) quoted in AOSA (1983) for seed longevity. Since then several researchers have carried out and aging treatment has been recommended for wide range of crop species (AOSA, 1983). However, there are no suggested or recommended procedures for conducting rice seed vigor tests including accelerated aging test are available in the handbooks of vigor test from the International Seed Testing Association (ISTA, 1995) or Association of Official Seed Analysts (AOSA, 1983).

Although accelerated aging test is being carried out by various rice seed centers in Thailand as seed vigor test, there is no standard accelerated aging conditions among rice seed centers, for example Chang Mai Rice Seed Center uses 42^{o}C for 72 hrs, Nakhon Ratchasima Rice Seed Center uses 45^{o}C for 96 hrs, However, the aging variables they adopt in the laboratory test do not separate vigor levels of seed lots thus, affecting the accuracy of seed vigor estimation which seedmen use to make decision for seed storage duration and arrangement for sale release priority. Determination of seed vigor in rice seeds seems to be more important recently because some recent recommend varieties of rice tend to have low seed vigor. Germination percentages of

those varieties usually decrease immediately after ripening period [Singkanipa, 2008 (personal communication)].

Since, rice is one of the important cash crops in Thailand therefore it is of utmost importance to have proper vigor tests which could evaluate rice seed vigor and thus could improve seed quality, benefit farmers and increase rice production. Therefore, this study aims to investigate the proper accelerated aging conditions of rice seed of common varieties in Thailand.

1.2 Research objectives

1.2.1 To develop a proper testing condition of temperature and time for accelerated aging test as rice seed vigor test for Thai varieties.

1.2.2 To servey for possibility of alternative rice seed vigor tests other than accelerated aging test.

1.3 Scope and limitations of the study

This study included only recommended rice varieties in some of the best and popular rice varieties like KDML105 were included in the experiments. Attempts are made to include as many rice varieties and seed lots produced by rice seed centers and rice research centers in Thailand. This means the varieties included in the experiments were those varieties which have market requirement and need for seed multiplication. The seed lots obtained were produced in the normal multiplication fields of seed production of breeder seed, foundation seed, registered seed and extension seed. So, the outcome of the study can be certainly benefit rice seed centers and rice research centers in Thailand.

CHAPTER II

REVIEW OF LITERATURES

2.1 Introduction

Seed quality is one of the major factors that determines the success or failure of a crop. Seed vigor is a key element of seed quality. Seeds selected based on high vigor will produce more uniform, vigorous stand of plants resulting ultimately higher yields per area (Tomer and Maguire, 1990). Some prominent works related to seed vigor are summarized below.

2.2 Concept of seed vigor

The development of a satisfactory definition of seed vigor has been a central theme in the development of vigor tests. Without a definition, the ability to measure or test this undefined object becomes difficult. Fortunately, many definitions have been proposed and a study of their development presents the initially confusing and changing status in the expectations for seed vigor (Copeland and McDonald, 1995). As an example, Gelmond et al. (1978) defined seed vigor as potential ability of seeds to yield the maximum plant products at the earliest time under variable environmental field conditions. According to Bishnoi and Delouche (1980), seed vigor is an aspect of seed quality which control field stand establishment and vigor tests are required to obtain reliable assessments of field performance. Chin (1988) defined seed vigor as the sum total of those properties of seeds which determines the level of activities and performances of seed lots during germination and seedling emergences. Seed vigoras per Sundstrom et al. (1986) is the capacity of a seed lot to germinate quickly and

completely with subsequent uniform seedling development**.** Seshu et al. (1987), outlined vigor as a concept usually derived from the observation of differences between seed lots, and a wide definition based on the effect have to be considered more appropriate than one based on causes. Egli and TeKrony (1995) set vigor as indicator for measurement of future performance of seed lots which related to the ability of seeds to germinate and produce seedlings that will emerge from the soil into healthy vigorous plants. Seeds that perform well are termed “high vigor seeds,” while those that perform poorly are called “low vigor seeds”.

Association of Official Seed Analyst; AOSA (1983) defined seed vigor as those seed properties which determine the potential for rapid uniform emergence and development of normal seedlings under a wide range of field conditions. The definition of seed vigor adopted by AOSA (1983) is quite similar to the definition of International Seed Testing Association; ISTA (1995) which stated seed vigor as the sum total of those properties of seeds which determines the level of activities and performances of seed lots during germination and seedling emergences**.**

At the beginning, more importance was given to the physical characteristics of seeds which were thought to be associated with low vigor but more recent investigation have focused on the physiological causes of vigor differences, especially the role of seed aging and membrane integrity (Copeland and McDonald, 1995). Seed aging has to be recognized as the major cause of reduced seed vigor and viability, and involves the process of deterioration like the accumulation of degenerative changes until eventually the ability to germinate is lost (Powell, 2006)

Seed deterioration usually begins at physiological maturity and continues during harvest, processing and storage at the rate greatly influenced by genetic, production and environmental factors. The time ranges from days to many years. The

process of deterioration is generally progressive and sequential (AOSA, 1983). A hypothetical model has been developed by Delouche and Baskin (1973) which outlined the sequence of changes in seeds during deterioration. The first change that is believed to occur is the deterioration of cell membrane probably due to the oxidation of fatty acid chains within the phospholipids of the membrane (McDonald, 1999). Since membranes are essential for many metabolic events and enzyme synthesis, therefore deterioration of cell membranes leads to decrease in the amount of ATP formed as an energy source, as well as retard the synthesis of specific enzymes essential for growth, reduce respiration which provides seeds with the energy molecules required for growth. The accumulation of all these deleterious changes contributes to a gradual decline in germination rate culminating in a loss of seed lot uniformity.

Other associated events which occur during deterioration are the loss in storage capacity and the ability to resist diseases. Eventually this subtle expression of loss in seed quality is expressed by an increasing incidence of abnormal seedlings, reduction in the speed and uniformity of field emergence and ultimately reduced yield. All these changes contribute to gradual decrease in seed vigor and germination (AOSA, 1983).

The concept and importance of high vigor seeds has been well understood. Considerable researches have been done and still continuing to acquire knowledge into better understanding of seed vigor. Several approaches have been made to 1) identify high and low vigor seeds of wheat (Tomer and Maguire, 1990; Modarresi et al., 2002), pea (Hampton et al., 2004), pumpkin and zucchini (Dutra and Vieira, 2006), and kale (Komba et al., 2006); 2) adopt vigor test as a predictor of predict field emergence of corn (Singhabumrung and Juntakool, 2004), soybean (Torres et al., 2004; Egli and TeKrony 1995), french bean (Hampton et al., 1992), lentil and chick

pea (Fernandez and Johnston 1995), cotton (Bishnoi and Delouche 1980) and 3) identify new vigor test in small seeded crops such as flower seeds (impatiens) (Jianhua and McDonbald, 1996) and onion (Rodo and Filho, 2003).

2.3 Factors affecting seed vigor

Seed development covers several important maturation stages from fertilization to accumulation of nutrient, to seed drying and to dormancy. These stages represent a change in morphological and physiological development that can alter seeds performance potentials. The point at which the seeds achieve maximum dry weight is called physiological maturity (Copeland and McDonald, 1995). At this point seeds have great potential for germination and vigor (TeKrony et al. 1979). However, since seeds generally achieve physiological maturity at high moisture levels unsafe for storage, seeds are typically not harvested until it attains harvest maturity, when seed moisture content is low enough for safe storage. Between physiological maturity and harvest maturity, the seeds are essentially stored on the plants where they may be exposed to severe environmental conditions that adversely affect seed quality. Post harvest technologies like threshing, seed cleaning, seed treatments and storage conditions also effect seed deterioration or seed vigor. Seeds may die in a few days to years depending on the level of deterioration (Copeland and McDonald, 1995). Some important factors which effect seed vigor are explained briefly below.

2.3.1 Environment

Rice seeds obtained from plants given short day treatment germinate more rapidly than seeds from untreated plants, particularly at low temperature (Ota and Takeichi, 1966). According to Sato (1973), rice seeds of japonica variety, Norin 17, ripened at 20°C day temperature, high light intensity and low relative humidity

(R.H.) showed maximum germination. A 30°C day temperature harmed the seeds, but low night temperatures increased germinability. However, in the indica variety, IR8, the optimum conditions for increased in germination were a day temperature of 30 or 35°C combined with high light intensity and low R.H. Finding of Krishnan and Rao (2005) stated that warm weather conditions with high solar radiation and without excessive rains during grain filling stage gave the best seed yield with high quality.

In wheat, Macchia et al. (1986) observed maximum germination at 36°C but an optimum temperature for viability and germination energy was between 24°C to 28.5°C. Whereas finding of Kamana and Maguire (1992) revealed that, germination of six winter wheats was zero at 5 and gradually increased and reached maximum germination at 21°C and 25°C. But in western wheatgrass, Waldron et al. (2006), found three different environmental conditions had little effect on seed yield and seedling vigor. According to Woltz et al. (2006), freezing temperatures reduced germination and vigor of corn seeds and to obtain higher germination and vigor, the corn seeds should be harvested immature stage before the freezing event. In peas, maturation in cool temperatures led to the development of larger seeds which gave rise to taller plants and earlier flowering in comparison to those of the same varieties which had matured in warmer temperatures (Kant et al., 1973). Likewise Wang et al. (2006) also revealed that high temperature stress during pod development decreased pod fertility, seed set and seed yield in chick pea. In soybean, Spears et al. (1997) revealed that high temperature during seed development reduced seed vigor in the absence of associated mechanical injury and seed borne diseases. Similarly, Khalil et al. (2001) also found that seed germination was inversely proportional to temperature experience by mother plant during pod filling stage which resulted into decrease in germination by 10.7% with 1% increase in temperature in soybean seeds.

2.3.2 Nutrition and hormone

The crop response to N in terms of seed protein and germination potential increased with foliar spraying of gibberellic acid at 10 ppm in rice (Mukherjee and Prabhakar, 1980). Warraich et al. (2002) indicated that nitrogen application improved grain quality and vigor in wheat by improving grain protein contents which increased the final germination percentage and reduced the T_{50} and mean germination time. Where as in corn, Khan et al. (2005) stated that nitrogen application of 120 N kg/ha resulted into maximum plant height, number of plants per area and low mortality percent. Finding of Mugnisjah and Nakamura (1984) revealed that high level of P application (1,000 kg/ha) along with 80 NH_4Cl kg/ha to the mother plant influenced the vigor of resulting soybean seeds producing high vigor seeds, further they also studied combination effect of harvest days and P application and suggested that application of 0 or 250 kg/ha would result in high vigor seeds. Keiser and Mullen (1993) suggested that reduced Ca supply to the plants may reduce seed Ca concentration in addition to altering other seed nutrient which are associated to poorer seed germination in soybean. Similarly, according to Burton et al. (2000), decreased calcium levels in the nutrient medium reduced soybean leaf dry matter during seed fill, seed production, seed Ca concentration, seed germination, and increased the incidence of seedling disorders such as watery hypocotyle and epicotyl necrosis. Dordas (2006) stated that application of Bo improved seed germination and seed vigor by 27% in 2003 and up to 19% in 2004 as compared with control in Alfalfa.

2.3.3 Water stress

Sürek and Beser (1999) in their investigation found that rice varieties irrigated at 8 day interval after tillering initiation reduced grain yield due to decrease

in number of filled spikelets per panicle and 1,000 grain weight whereas highest value of grain yield was observed at continuous flooding irrigation. Boonjung and Fukai (1996), specified that occurrence of drought condition at vegetative stage had only small effect with reduction in yield of rice up to 30%, but when drought occurred during panicle development, the stress was severe resulting into 60% yield reduction and during grain filling stage the drought stress reduced yield to 40%. Work of Yang et al. (2001) suggested that water stress altered hormonal balance in rice grains during grain filling, especially a decrease in gas and an increase in ABA. In sorghum, water potential at field capacity in two varieties showed similar radical and coleoptile emergence but at low water potential there was reduction in radical and coleoptile emergence and radical length due to reduced water uptake (Gurmu and Naylor 1991). Where as in peanut, water deficit during the growing season of the parent plants produced seeds with fewer rapidly growing seedlings compared to well water plants (Ketring, 1991). According to Brevedan and Egli (2003), water stress induced acceleration of leaf senescence which led to reductions in seed size and yield of soybean.

2.3.4 Herbicide management

Chemicals especially systemic ones used to control pest should be tested for any side effect on seed quality. Eastin (1980) studied the suitability of pre-harvest desiccants like diquat, glyphosate, paraquat and sodium chlorate, and found that none of the above desiccants reduced seed germination or seed weight in rice. Gangadhara and Kunhi (1979) in their studies revealed the use of chemical like 2, 4, 5 -trichlorophenoxy acetic acid did not reduced germination and seedling vigor of rice, corn, sorghum, finger millet and horse gram where as reduction in germination and seedling vigor was observed in tomato and brinjal. El-Daly (2006) indicated that high

dose of cyanox (10^{-3} M) lowered the percent germination in radish.

2.3.5 Seed maturity and stage of harvesting

Harvesting the seeds at the appropriate stage is important to achieve good quality seeds (Copeland and McDonald, 1995). The optimum time for harvesting has been considered to be 30-42 days after heading in the wet season and 28-34 days after heading in the dry season in rice (Seetanun and De Datta, 1973). Pre-harvest spraying of desiccants is a technique employed to enhance seed maturation (Seshu et al., 1987).

Eastin et al. (1973), revealed that black layer formation in the placental area of seeds as good indicators of physiological maturity or date of maximum dry weight accumulation in sorghum. Physiological maturity of seeds can be detected morphologically as well as formation of black layer in corn (Daynard and Duncan, 1969) where as milk line at stage 4 proved to be the best indicator of the time to harvest corn seeds for maximum physiological quality (Santo et al., 2005). Afuakwa and Crookston (1978), suggested black layer is more reliable indicator of physiological maturity where as milk line is useful in monitoring grain maturity prior to physiological maturity in corn.

TeKrony et al. (1979) found that in soybean, physiological maturity of entire plant, estimated from dry seed weight data occurred when 26% of the seeds were yellow and 35% of the pods were yellow or green. He further suggested that the occurrence on the main stem of one normally colored mature pod per plant represented a useful and acceptable indicator of physiological maturity. In common bean, maximum viability was achieved at moisture contents of 31-37% which was well beyond physiological maturity but maximum seed vigor was archived at physiological maturity (Muasya et al., 2002).

According to Chuntarachurd et al. (1984) yard long bean seeds harvested at 16 days after pollination produced highest seed weight and seed vigor. However, as per finding of Santipracha and Santipracha (1997) yard long bean seeds harvested at 18-20 days after flowering produced fresh pod yield of 6,337.5-6,437.5 kg/ha which were significantly higher than the seeds harvested at 14-16 days after flowering. In stakeless yard long bean, Teekachunhatean (2006) was found that physiological maturity was 20 days after anthesis. At physiological maturity, it showed 1) maximum seed dry weight of 163.37 mg/seed, 2) maximum seed germination and vigor, and 3) seed moisture content of 29.71%. However, maximum seed size which measure by seed length was 13.06 mm at 18 days after anthesis and reduce to 11.53 mm at physiological maturity. If harvesting was delayed to 30-34 days after anthesis, seeds lost germination rapidly. It was recommended to harvest the seeds at 22-26 days after anthesis.

2.3.6 Position of seeds

Food reserve accumulation and seed weight may vary with the position of the seeds on the panicle and on different tillers. Amatitka (1992) revealed that in indeterminate soybean, large seeds all from the base plant portion had the highest germinability and vigor followed by the medium seeds all from the middle portion. According to Thomas et al. (1987), immature seeds from secondary umbels took 6 days longer to germinate at 5°C and 2 days longer at 20°C than those from primary umbels in radish and seedling weight about 20 days after 50% emergence were greater from seeds from primary than secondary umbels. In celery also heavier seeds were produced from primary umbels and were less dormant than those from quarternary umbels. Similarly, Pereira et al. (2008) also revealed that carrot seeds from primary and secondary umbels had higher vigor and germination than tertiary umbels under

high temperature of 36°C**.** Alan and Eser (2007), concluded that fruits harvested from different layers (first, second and third) of pepper plants showed gradual decrease in fruit weight and seed yield throughout fructification, from the first to the third layer and seeds extracted from fruits of the first layer had higher germination and vigor, lower mean germination time than those from other layers. Seeds from the third layer caused decline in seed quality especially vigor with 69.3%.

2.3.7 Seed size and weight

Heavy rice seeds gave better germination, emergence and more resistance to deterioration (Akil and Araújo, 1977). Similar to this Roy et al. (2008) revealed, germination rate and seedling vigor index values increased with the increase of seed size in rice suggesting selection of larger seeds for good stand establishment. In contrast to above finding Krishnasamy and Seshu (1990) reported that seed weight negatively correlated with germination in rice. Lafond and Baker (1986) investigated the role of seed size, speed of emergence and rate of plant development in seedling vigor of nine spring wheat varieties and found that plants grown from small seeds emerged faster but accumulated less shoot dry weight than those from larger seeds and further suggested that seed size may be taken as one criteria for evaluating seedling vigor. Baalbaki and Copeland (1997) suggested that seed size significantly affected emergence and yield in wheat with small seeds producing lowest grain yields. From their work they recommended that selecting larger seeds would provide high quality seeds with the potential for increased yields in wheat under variable conditions. In corn also, large seeds resulted in maximum emergence per area, maximum plants height, maximum number of plants per area and low mortality percent **(**Khan et al., 2005).

In pea, large seeds gave rise to taller plants which flowered earlier

(Kant et al., 1973). As per finding of Amin (1999), mungbean seed size showed no significant effect on yield except large seeded plants flowered and matured earlier than small seeded plants producing heavier seeds. Similar trend was observed by Na Chiangmai et al. (2007) where large mungbean seeds had advantages only in sprout production than small seeds without any effect on final yield.

2.3.8 Mechanical injury

Mechanical injury during threshing, cleaning, handling and planting is considered as one of the most important factors influencing seed quality and thus seeds and seedling vigor. Machine threshed seeds are more injured than hand threshed ones because machine damages the embryo causing death.

According to Bourgeois (1993) a high vigor could be maintained in durum wheat seeds by threshing at low cylinder speed and low kernel water content. Similarly, Kantor and Webster (1967) observed, increased cylinder speeds of the thresher not only increased the percentage of cracked seeds but also reduced the germinability of ‘sound’ seeds and increased the percentage of abnormal seedlings in sorghum. Fessel (2006), evaluated sixteen samples of corn seeds in series of conditioning steps like reception, pre-cleaning, post gravity separator, post grader and after bagging and concluded that mechanical damage could occur at any stage of seed conditioning and was cumulative. According to Cicero et al. (1998), mechanical damage especially ruptures which caused restriction of nutrient translocation to the embryonic axis caused seed and seedling abnormalities in corn seeds.

Studies in bean and cabbage seeds showed that the external damages were much larger in bigger bean seeds than in smaller cabbage and the mechanical abuse was not only visible such as cracking seed coats and breaking seeds in both species but also invisible, internal injuries, which caused lower germination down to

60-70% and increased the number of abnormal seedlings (Duczmal, 1981). In cotton, handpicked and laboratory ginned lots were of better quality than mechanically harvested and commercial ginned seeds (Bishnoi and Delouche, 1980).

2.3.9 Aging

During aging declined in seed vigor, respiration rate, phosphatase activity and sugar content accompanied by a complete declined of alpha amylase activity are noticeable. The concentration and the number of amino acids and the RNA and DNA contents also show a similar reduction with higher RNAse activity (Seshu et al., 1987). Rise in respiration rate, phosphatase activity and sugar content accompanied by a complete decline of alpha amylase activity and RNA, DNA and protein content were noticeable in rice during seed deterioration. (Zhoe et al., 2002); Decrease of phospholipids, carbohydrates, sugars, proteins ascorbate and peroxidase activity with increased in fatty acids and malonaldehydes were observed during aging of corn seeds (Basavarajappa et al., 1991). Kalpana and Madhava Rao (1999) observed accumulation of reducing sugar in pigeon pea during aging which caused rapid seed deterioration. Likewise in french bean seeds, aging caused complete loss of nucleoli (Begnami and Cortelazzo, 1996); while aging of carrot (Al-Maskri et al., 2003) and cotton (Basra et al., 2003) seeds caused increase in lipid peroxidation leading to loss in germination and seed vigor.

2.3.10 Pathogens

Infection of plants and seeds by various plant pathogens, fungal, bacterial, or viral in nature, in the field or in storage, can reduce vigor directly through such mechanisms as enzymatic degradation, toxin production and growth regulation. Seed vigor loss because of infestation by pathogens could occur indirectly to the extent that pathogenic infection limits the ability of the seeds on the plants to develop normally

(Copeland and McDonald, 1995). Imolehin (1983) obtained a high negative correlation between rice seed infestation by different fungi and seed germination in the laboratory. According to Phat et al. (2005), seed germination of jasmine 85 rice affected by discoloration disease in both wet and dry seasons resulted into decrease number of filled grains/panicle and 1,000 grain weight**.** The presence of pathogens in the soil clearly had a deleterious effect on field emergence of soybean as field emergence of low vigor seed lots were reduced more by the non-sterile soil than the medium and high vigor seed lots (Hamman et al., 2002).

2.3.11 Seed treatment and priming

Seed treatment is one of the important quality aspects of seeds. Treated seeds are protected from pests that attack seeds and seedlings and can improve stand quality and increase yields also (Powell, 2006). Ventura and Garrity (1987)**,** recommended hot water treatment at 52°C for 15 min was physiologically safe for rice seeds regardless of variety as it increased field seedling vigor and tiller number over the untreated control. According to Bhattacharjee and Bhattacharyya (1989), pre-treatment of rice seeds with dikegulac-sodium (DK) a growth regulator at 1,000 and 2,000 µg/ml slowed down the rapid fall of germination and arrested the leakage of electrolytes, reduced the leaching of soluble carbohydrates and decrease the loss of RNA from seeds.

Biswas et al. (2000) indicated that certain strains of nonphotosynthetic diazotrophs, including rhizobia, could promote growth and vigor of rice seedlings**.** Nghiep and Gaur (2005), studied efficacy of various chemical seed treatment in rice seeds and found that seeds treated with Vitavax, Thiram and Mancozeb maintained germination above 80% after six months of storage. According to Farooq et al. (2004), thermal hardening in indica rice increased germination index, energy of germination,

radical and plumule growth rate, and root/shoot ratio. Seed treatment of coarse and fine rice with $CaCl_2$ gave higher values of final germination, emergence speed, energy of germination, higher root and shoot length and seedling fresh and dry weight than other chemicals like $KN0_3$, KCl and NaCl.

Hu et al. (2005) suggested that rice seeds primed with sands that contained 3.8% (v/w) water at 18°C for 72 hrs improved energy of germination, germination percentage, germination index and vigor index, resulting in higher seedling height, root length, number of roots and root dry weight as compare to control. As per Basra et al. (2006), rice seeds primed with 10 ppm ascorbic acid promote early germination and vigor enhancement. Rice seeds soaked in potassium and phosphorous solutions produced taller plants and had higher dehydrogenase activity suggesting higher viability and vigor of seeds (Bam et al., 2006)**.** In wheat, seeds primed with 25 ppm kinetin and 1% prostrate (2 hrs) treatments showed best results on seedling growth, fresh and dry weights under both saline and non-saline conditions (Afzal et al., 2005).

2.3.12 Genetic Variability

Differences in seed vigor usually arise as a result of various factors which effect seeds (Seshu et al., 1987) identified principal causes of vigor differences in rice and stated that seed dormancy may obscure the vigor potential of seed lots in laboratory test, but it should not be regarded as a component of vigor if seedling emergence is unaffected in field sowings. As per Siddique et al. (1988), different rice varieties have different tolerance level to aging. His studies indicated that out of 64 and 121 varieties tested during dry season and wet season in 1985, respectively, the most tolerance to 6 days aging at 43°C were found in each eco geographic races of *Oryza sativa* L. (indica, japonica and javanica) and also among photoperiod sensitive

and insensitive strains, upland and lowland types. According to Ali et al. (2003), germination of rice varieties in low temperature was influenced more by genotype than seed quality.

Krishnan and Surya Rao (2005) investigated 12 rice genotypes grown during 2000-2003 and found out that proportion of seed setting, seed leachate conductivity, potential of seed longevity, percentage of seed germination and proportion of seed discoloration were influenced more by environmental effects than by genotype. But in contrast, yield, panicle number, seed weight, and proportion of high-density grains were influenced more by genotypic than by environmental effects.

2.3.13 Other factors

Schweizer and Ries (1969) reported that seedling growth and grain yield were significantly correlated with seed protein in barley, likewise in wheat also they found seed protein positively correlated with subsequent seedling growth (r = 0.7335**). Pedersen and Toy (2001) investigated the effect of seeds and plant color on seed vigor and found out that in sorghum, purple plant color phenotypes had higher cold germination, higher germination after accelerated aging, and greater seedling elongation at 10 days than tan plant color phonotypes. Similar results were observed in peas by Woyke (1987) where green-yellow seeds had higher vigor as compare to yellow-green and yellow seeds. Finding of Atak et al. (2008) also revealed that dark green colored pea seeds germinated faster had highest shoot, root length and fresh weight resulting into higher seed vigor than light and medium colored seeds. Patil and Andrews (1986) studied response of cotton hard seeds to accelerated aging and found out that hard seeds were less susceptible to storage deterioration.

2.4 Classification of vigor test

The challenge of vigor testing has been to identify a measurable parameter which is common to seed deterioration. The most neglected area of seed study remains the physiological and biochemical characterization of seed deterioration. The principle reason for this lack of knowledge is that many events could create and contribute to loss in seed viability which leads to difficulty in identifying the cause and effect of a specific deteriorative response (Copeland and McDonald, 1995). Although it is impossible to state with accuracy which events occur during seed deterioration we can speculate on the probable sequence of changes as developed by Delouche and Baskin (1973) which suggested that seed deterioration is sequential and accumulative and outlines some of the major parameters which can be utilized in measuring seed vigor (AOSA, 1983)

Seed vigor test is more sensitive index of seed quality than germination test, any of the events which precede loss of germination could serve as vigor tests. Thus, according to the seed deterioration model developed by Delouche and Baskin (1973), the most sensitive vigor test might be the one which monitors membrane degradation as it is the first sign of seed deterioration. Besides this at the biological level, it involves biosynthesis of energy and metabolic compounds such as proteins, nucleic acids, carbohydrates, lipids, etc. which coordinates cellular activity, membrane integrity, and transportation and utilization of reserve foods. At the germination level, it involves speed and totality of germination, mechanical rupture force of seedlings, tolerance of seedlings to environmental stress and diseases resistance (AOSA, 1983). A vigor test can be a measurement of one or more of these events.

Hampton and Coolbear (1990) classified vigor tests into a) single test based on some aspects of germination behavior such as rate of germination, seedling growth

and seedling evaluation, cold tests, accelerated aging, and controlled deterioration, b) physiological or biochemical test as electrical conductivity and tetrazolium test and c) multiple testing, such as assessments based on more than one technique. AOSA (1983) classified vigor tests into a) biochemical test such as tetrazolium (TZ), conductivity, respiration, glutamic acid decarboxylase activity (GADA) and ATP content, b) stress test such as cold test, accelerated aging, cool germination, brick test and osmotic stress and c) seedling growth and evaluation tests such as seedling vigor classification, seedling growth rate, speed of germination

2.4.1 Biochemical test

2.4.1.1 Conductivity test

The conductivity test is a measurement of electrolytes leaking from seeds. Changes in the organization of cell membranes occur during the development of seeds prior to physiological maturity, seed desiccation before harvest, and during imbibition prior to germination (ISTA, 1995). As seed rehydrates during early imbibition, the ability of its cellular membranes to reorganize and repair any damage that may have occurred will influence the extend of electrolyte leakage from seeds. The greater the speed with which the seeds are able to re-establish their membrane integrity the lower the electrolyte leakage. Higher vigor seeds are able to reorganize their membranes more rapidly, and repair any damages to a greater extend, than lower vigor seeds. Consequently, electrolyte leakages measured from high vigor seeds are less than that measured from low vigor seeds. Low vigor seeds have been shown posses decreased membrane integrity as a result of storage deterioration and mechanical injury.

The conductivity test has been successfully used and recommended in corn and soybean (AOSA, 1983), pea (AOSA, 1983, ISTA, 1995), barley (Shephard et

al., 1989), rice (Krishnasamy and Seshu, 1990), cabbage (Taylor et al., 1995) and bean (Fernandez and Johnston, 1995).

The correlations between conductivity and field emergence had been studied among several crop seeds. Bedford (1974) observed conductivity test as good indicator for predicting field emergence in pea seeds, where as Bustamante et al. (1984) found no significant correlation between conductivity test and field emergence. Studies on other crop seeds such as lentil, bean and chickpea (Fernandez and Johnston, 1995), soybean (Makkawi et al., 1999) and french bean and mungbean (Hampton et al., 1992) showed that conductivity test could predict field emergence significantly. In rice, Chea (2006) observed that conductivity test was the only one seed vigor test among several other seed vigor tests which could predict field emergence of rice under different seed bed conditions.

2.4.1.2 Tetrazolium test

The tetrazolium test is commonly used to assess the viability of seeds and it can also be used to detect vigor differences (AOSA, 1983; ISTA, 1995). The test evaluates the presence and location of living tissue within the seeds through the reaction between a solution of 2, 3, 5-tripheny tetrazolium chloride (TZ) with active dehydrogenase enzymes. The TZ is reduced by enzymes to produce the red and stable substance triphenyl tetrazolium formazan. Hence, the presence of red staining indicates living tissue; dead tissue remains unstained. Vigor is evaluated through the appraisal of sound, weak and dead tissues as they relate to subsequent seedling development. This test has been used and recommended in seeds of corn, pea, soybean, cotton, and clover (AOSA, 1983). Amaritsut (2004) found that soybean seeds stained with 0.1% TZ was not reduced in germination percentage. The stained seeds could germinate into normal seedling properly. He developed the germination of TZ

stained seed technique with very accurate and concrete standard patterns of TZ for viability and vigor evaluation of soybean seeds.

2.4.1.3 Respiration activity

Seed germination and seedling growth require the use of metabolic energy acquired from respiration. This test is based on the assumption that seeds showing high respiration rate can produce vigorous seedlings (AOSA, 1983). Woodstock and Pollock (1965) quoted in AOSA (1983) reported that during the first few hours of imbibition, respiration was closely related with seedling growth rate in beans. Woodstock (1966) quoted in AOSA (1983) grouped corn seeds into high, medium and low vigor ones by using the differences observed in respiration rates.

2.4.1.4 Glutamic acid decarboxylase activity (GADA)

Glutamic acid decarboxylase (GADA) is an enzyme which was first observed to be activated in wheat seeds during imbibition and later it was recognized also as a better indicator of storability of seeds than the level of free fatty acids. Determination of this enzyme is known as GADA test. The procedure for measuring GADA is simple. The equipment consists of water bath or temperature control, manometers, a scale for measuring manometer fluid moments, ½ pint jars and a small grinder. Seeds are grinded and placed in the jar with buffered glutamic acid (Bedell, 2001). The glutamic acid decarboxylase (GADA) reacts with glumatic acid and glutamate is converted into aminobutyric acid which results in released of CO_2 (Hampton, 1995). The amount of CO_2 evolved reflects the level of enzymatic activity. Measurements are usually taken after 30 minutes at 26ºC (Bedell, 2001). High vigor seeds usually produced more amount of CO_2 and vise versa in low vigor seeds. Grabe (1964) quated in AOSA (1983) worked a lot in developing the GADA test to one of the potential values in vigor testing. He found that GADA was highly correlated with

seedling growth in corn.

2.4.1.5 Adenosine triphosphate (ATP) content

The energy for biochemical reactions in living cells are stored in high energy compounds such as ATP. ATP can be extracted from imbibed seeds by boiling water and measured by photometer. The ATP content is directly proportional to seed vigor levels. However, recent study has reported that ATP content was not correlated in corn, cucumber, onion and radish seeds (Styer et al., 1980 quoted in AOSA, 1983).

2.4.2 Stress test

2.4.2.1 Accelerated aging

Delouche and Baskin (1973) developed accelerated aging test procedure to measure seed storability and evaluate vigor. The technique involved the exposure of seeds to adverse levels of temperature (40-45ºC) and 100% R.H. for varying length of time followed by regular germination test. The seeds absorbed moisture from the humid atmosphere and aged rapidly due to high temperature. The basis for this test is that higher vigor seeds tolerate the high temperature-high humidity treatment and thus retain their capability to produce normal seedlings in the germination test. Accelerated aging test has been suggested and recommended in many crops such as wheat, sorghum, corn, beans, soybean, onion, radish and lettuce (AOSA, 1983; ISTA, 1995). Several studies in seeds of french bean (Pandey, 1989), wheat (Bhattacharyya et al., 1985), rice (Krisnasamy and Seshu, 1990, Ali et al., 2003), cotton (Basra et al., 2003), carrot (Al-Maskri et al., 2003), aubergine, cucumber and melon (Demir et al., 2004), beet root (Silva at al., 2006), kale (Komba et al., 2006), soybean (Torres et al., 2004), radish (Jain et al., 2006) have also proved that accelerated aging test could be successfully used as seed vigor test.

2.4.2.2 Cold test

The cold test expose seeds to cold temperatures (10ºC for 7 days) in non-sterile field soil at approximately 60-70% of water holding capacity prior to a 4-7 day grow out period in ideal conditions (25ºC). The moisture and temperature provided in the cold test simulate the adverse conditions that seeds might encounter in an early spring planting. Besides this two stress conditions, other factors like seed quality, genotype, pathogens and seed treatment may also affect seed vigor. The cold test attempts to measure the impact of the combined effects of all these factors and often represents the lowest emergence that would be expected from seed lots when planted under reasonably satisfactory field conditions, while the standard germination test represents the highest emergence potential that could be expected.

When germination obtained in the cold test is very close to that obtained in the standard germination test, the seed lots would be expected to emerge well over a wide range of soil moisture and temperature (ISTA, 1995) The cold test has been traditionally used for vigor testing of corn it is also used as ability to forecast seed performance in cotton (Bishnoi and Delouche, 1980). It has also been successfully used and recommended in crops such as corn (AOSA, 1983; ISTA, 1995).

2.4.2.3 Cool germination

The cold test was too severe for cotton; therefore cool germination test was developed especially for cotton to provide less demanding yet sufficiently severe test conditions to enable the separation of vigorous from less vigorous seeds. Cotton seeds are germinated in darkness at a constant temperature of 18ºC for seven days since the minimal temperature for cotton seeds germination is about 15ºC. Normal

cotton seedlings having a total length of 4 cm or longer after seven days of germination at 18ºC are considered high vigor (AOSA, 1983).

2.4.2.4 Brick test

Also called as hiltner test, seeds are planted on damp brick grit or sand or flower pot and covered with 3 cm damp brick grit and germinated in darkness at room temperature for a specific time. Seeds infected by pathogens fungi, injured or low in vigor are unable to penetrate the brick grit layers. The percentage of normal seedling emerged is considered an indication of the vigor (AOSA, 1983). The test has been suggested and recommended for corn, wheat, barley, oats and rye (ISTA, 1995).

2.4.2.5 Osmotic stress

When the seeds are sown in the field, they are often subjected to drought stress which results in poor field emergence. Such drought condition can be simulated in a laboratory test by using solution having specific osmotic potential like sodium chloride, glycerol, sucrose and polyethyleneglycol (PEG). The rate of germination under water stress conditions is markedly reduced; only vigorous seeds could tolerate a more sever osmotic stress (AOSA, 1983). The test has been successfully used in peas and sugar beet (McDonald, 1999).

2.4.3 Seedling growth and evaluation tests

2.4.3.1 Seedling vigor classification

This vigor test is similar to the standard germination test. The only difference between the two tests is that normal seedlings are further classified as “strong” and “weak”. The basis for the test is that the “strong” and “weak” classification provides means of separating seedlings free of deficiencies from those with deficiencies which are symptomatic of low vigor or reduced quality. The test has been successfully used and recommended in beans, soybean (AOSA, 1983; ISTA, 1995),

peanut and cotton (AOSA, 1983) and pea (ISTA, 1995).

2.4.3.2 Seedling growth rate

Vigorous seeds are able to efficiently synthesize new materials and rapidly transfer new products to the emerging embryonic axis, resulting in increased dry weight accumulation. The seedling growth rate test is based on this concept and vigor results are expressed as mg dry weight/seedling. Seeds are germinated according to the standard germination test but kept in the dark germinator. After 7 days normal seedlings with cotyledons or endosperms removed are oven dried at 80ºC for 24 hrs and weight to determined seedling dry weight. The test has been suggested and recommended in corn and soybean seeds (AOSA, 1983; ISTA, 1995).

2.4.3.3 Speed of germination

Speed of germination is an important vigor aspect because it provides a reasonably good index of vigor. A faster germination rate will facilitate early seedling establishment, a clear advantage under direct sown rice culture (Seshu et al., 1987) Seed lots with similar total germination often vary in their rate of germination and growth. Unlike in standard germination test, the normal seedlings are evaluated on daily basis starting from first count till the final count and calculated as follows

$$Speed\ of\ germination = \frac{number\ of\ \ normal\ seedlings}{days\ of\ \ first\ count} + \ldots\ldots + \frac{number\ of\ \ normal\ seedlings}{days\ of\ \ last\ count}$$

It is found that the higher the number, the higher the seed vigor. The advantage of the speed of germination test is that very little work is required when compared to the standard germination test.

2.5 Correlation between field emergence and laboratory tests

Most definitions of seed vigor emphasize the relationship between seed vigor test and field emergence and many studies have demonstrated that this association exists. Consequently, the ultimate value of any seed vigor test would be its ability to predict field emergence. Field emergence depends on interaction between the seed vigor and seed bed environment, under favorable field conditions only the germination in the laboratory correlates to the field emergence but the correlations declined under stress field conditions (Tomer and Maguire, 1990).

Venter van de et al. (1993) observed in wheat seeds that percent germination at low temperature (9ºC) had higher correlation (r = 0.71**) with percent field emergence than other seed vigor tests like accelerated aging test (r = 0.13) and complex stressing vigor test (r = 0.50*). Baskin et al. (1993) compared standard germination and cold test for predicting field emergence in sorghum. They observed that both tests showed high correlations with field emergence under favorable soil condition (r = 0.848** for cold test and r = 0.825** for standard germination test) but under unfavorable soil conditions the correlation between standard germination and field emergence decreased (r = 0.501**), whereas cold test still showed almost the same correlation coefficient with field emergence (r = 0.845**). According to Lovato et al. (2005), the cold test at 10ºC for 7 days and accelerated aging test at 45ºC for 72 hrs provided higher correlation (r = 0.75** and r = 0.79**, respectively) where as standard germination test poorly correlated (r = 0.51**) with field emergence in corn seeds. Similar results were obtained in two experiments by Noli et al. (2008) where they observed highest correlation (r = 0.66** and 0.73**) between cold test at 10ºC for 7 days and field emergence in corn seeds. As per Ilbi et al. (2009), cool germination test at 18ºC could be used as alternative test for seed vigor test in corn as it

provided high correlation (r = 0.890**) with field emergence and with cold test (r = 0.828**).

Bustamante et al. (1984) studied the correlation of field emergence and seed quality in pea seeds and reported that water stress (r = 0.78**), controlled deterioration (r = 0.91**) and laboratory soil emergence (0.94**) were the good seed vigor indices for predicting field emergence where as conductivity (r = -0.51) failed to predict field emergence. In contrast to this, Bedford (1974) studied 12 pea varieties and concluded that conductivity test could be a good indicator for predicting field emergence (r = -0.809**). Fernandez and Johnston (1995) performed bulk conductivity test in lentil, bean and chickpea and found that bulk conductivity test correlated well with field emergence. However, Makkawi et al. (1999) did not observe any correlation between conductivity test and field emergence in one of his two experiments instead standard germination provided high correlation (r = 0.811** and r = 0.555*) in both the test conditions with field emergence in lentil seeds. Vieira et al. (2004), studied the relationship between electrical conductivity and soybean seedling emergence and recommended that under optimum field conditions, seed lots with up to 110 $\mu S\ cm^{-1}g^{-1}$ of EC would establish adequate field stands.

Amaritsut (2004) developed standard patterns of TZ for vigor test in soybean. He proposed that high vigor and medium vigor seeds according to TZ test would represent field emergence percentage. It was proven that high correlations were found between the TZ vigor test results (r = 0.985**) and field emergence (r = 0.994**).

2.6 Accelerated aging as seed vigor test

2.6.1 History

Accelerated aging as a test for seed quality was first developed by Delouche (1965) quoted in AOSA (1983) at the seed technology laboratory, Mississippi State University, USA. It was initially developed as a test to estimate the longevity of seeds in warehouse storage. Subsequent studies have verified the accuracy of this test in predicting the life span of a number of different seed species under a range of storage conditions (Delouche and Baskin, 1973). In 1970, Baskin proposed using the accelerated aging test to predict stand establishment of peanut and suggested that accelerated aging test might have additional utility as a test for predicting seed performance other than storability. Since then other studies have shown that accelerated aging test functions equally well in forecasting stand establishment of seeds of various crops.

2.6.2 Principle of accelerate aging test

The accelerated aging test functions by exposing seeds to the most important environmental conditions which influence seed deterioration; high temperatures (40 to 45ºC) and high relative humidity (greater than 90%) for short periods of time (48 hrs or longer depending on the species). Under these extreme conditions, the rate of deterioration is greatly enhanced (AOSA, 1983). It has been proposed by Delouche and Baskin (1973) that the declined following accelerated aging is proportional to the initial physiological potential of the seeds. High vigor seeds would show only small decreases in germination after accelerated aging treatment while low vigor seeds would demonstrate marked decreases. Similarly, Delouche and Baskin (1973) suggested that the germination response after accelerated aging was related to the performances of seed lots in the field under a wide range of environmental conditions.

2.6.3 Seed deterioration processes after accelerated aging

The two most important factors that influence the life span of seeds are relative humidity and temperature. The effect of relative humidity (and its subsequent effect on seed moisture) and temperature are highly interdependent. Most crop seeds lose their viability at relative humidity approaching 80% and temperatures of 25 to 30ºC (Copeland and McDonald, 1995). Accelerated aging is a physiological stress test that permits controlled deterioration of seeds (Begnami and Cortelazzo, 1996) due to exposer to high temperature and high relative humidity (greater than 90%) (ISTA, 1995). Seed moisture content and high temperature influence seed metabolism. High relative humidity increases seed moisture, which results in biochemical events such as increase hydrolytic enzyme activity, free fatty acids where as high temperature serves to enhance the rate at which many enzymatic and metabolic reaction occurs, increases the metabolic activity of hydrolyzed substrates and enzymes causing more rapid rate of deterioration (Copeland and McDonald, 1995).

Some researchers studied the deterioration process after seeds were subjected to accelerated aging test. The deleterious changes that occur during the process of aging have been described in detail on many occasions The first sign of seed deterioration was membrane degradation (Delouche and Baskin, 1973). The first change that is believed to occur is the deterioration of cell membranes, probably due to oxidation of fatty acids chains within the phospholipids of the membrane (McDonald, 1999). This leads to a reduction in the integrity of cell membranes and the first clear expression of deterioration, an increase in solute leakage from the seeds (AOSA, 1983). Electron microscope studies have revealed the disruption of sub-cellular organization, reduced enzyme activity, decreased in the rate and efficiency of respiration and over all reduction in synthesis of macromolecules. Finally, before the germination of a population begins to decline, an increase in the incidence of

chromosome abnormalities, observed in the root tips of germination of aged seeds, suggested damaged DNA (Powell, 2006).

Pauratsamee (2008) also revealed that in soybean seeds, membrane degradation was the first sign of seed deterioration after accelerated aging due to lipid peroxidation. As per Perez and Arguello (1995), accelerated aging caused changes in membrane integrity associated with seed deterioration in embryonic axis of peanut resulting in leakage of vital electrolytes out of the seeds. Jain et al. (2006) found that the protein content declined under accelerated aging condition in radish may be due to either decreased rate of synthesis of proteins or increased degradation activity of proteinases, or the combination of both. The declined in the total protein content due to impaired protein biosynthetic activity with the gradual loss of seed viability have also been reported in pigeon pea (Madhava Roo and Kalpana, 1994); declined in viability, vigor, lipoxygenase and acid phosphatase activity and lipid content in cotton (Freitas et al., 2006).

2.6.4 Benefits

Accelerate aging test is very popular and most frequently used in seed testing laboratories because it is rapid, simple, inexpensive, no sophisticate equipments are needed and it could be done by any one without training (AOSA, 1983). Besides, seed vigor test for predicting field emergence it can also be used in evaluating storage potential crop seed. Surveys of seed testing laboraties in North America have shown that the accelerated aging test is one of the most frequently used vigor tests (TeKrony, 1983; Ferguson, 1990 quoted in AOSA (1995). The accelerated aging method has been proved as indicator of seed vigor in wide range of crop species and has been successfully related to field emergence and stand establishment. AOSA

(1983) and ISTA (1995) have recommended common conditons for accelerated aging test as seed vigor test in various crops as presented in table 2.1

Table 2.1 AOSA (1983) and ISTA (1995) common recommendations for conditions of accelerated aging test for vigor test in variables among crop seeds.

Crop	AOSA (1983)		ISTA (1995)	
	Temp (°C)	Hour	Temp (°C)	Hour
Tall Fescue	40	72	41	72
Lettuce	40	72	41	72
Garden bean	42	72	41	72
Bean and Onion	42	72	41	72
Corn	42	96	45	72
Sweet corn	-	-	41	72
Sorghum	45	72	43	72
Wheat	45	48	41	72
Radish	45	48	45	48
Soybean	41	64	41	72
Onion	42	72	41	72
Tomato	-	-	41	72
Mungbean	-	-	45	96
French bean	-	-	45	48
Pepper	-	-	41	72
Cotton	-	-	45	72
Water melon	45	72	45	144

2.6.5 Specific accelerated aging conditions

Although, AOSA (1983) and ISTA (1995) have recommended common conditions for accelerated aging test as seed vigor test in various crop (table 2.2), the recommendations are only common or general conditions and many seed crops are not included. Currently, there have been the evidences that specific crop varieties or types need specific accelerated aging conditions for the most accurate vigor test results.

Aging conditions either 43°C for 72 hrs or 45°C for 72 hrs provided better separation of wheat seed lots into vigor levels than 41°C for 72 hrs as suggested by ISTA (Modarresi et al. 2002). According to Santipracha et al. (1997), accelerated aging at 42°C for 96 hrs for corn seeds as recommended by AOSA (1983) showed only some reduction in germination and suggested that the aging conditions should be at 44°C for 96 hrs to evaluate vigor of hybrid corn seeds in humid tropics. The accelerated aging conditions of 41°C for 72 hrs internationally accepted for seed vigor test of soybean could also separate pea seeds in different vigor levels (Hampton et al., 2004). Hampton et al. (1992) suggested that accelerated aging at 45°C for 96 hrs and 45°C for 48 hrs could separate mungbean and french bean seeds into different vigor levels. Komba et al. (2006) evaluated accelerated aging test in brassica species and suggested that aging conditions of 41°C for 72 hrs which is presently used for *Brassica naphus* L. was too sever for kale (*B. oleracea* L. var. *acephala* DC) and recommended aging conditions of 41°C for 48 hrs should be used due to its short and suitable time. In order to find out the best accelerated aging conditions in aubergine, cucumber and melon seeds, Demir et al. (2004) evaluated series of temperature (40°C and 45°C) and time (24, 48, 72, 96, 120 and 144 hrs) combinations and found out that

aging conditions of 45°C for 72 hrs for cucumber seeds, 45°C for 120 hrs for melon seeds and 45°C for 48 hrs for aubergine seeds gave best separation between the lots.

In contrast to the above finding Torres and Filho (2003) concluded that the aging conditions of 38 or 41°C for 72 and 96 hrs provided differences in physiological quality of melon seeds. In pumpkin and zucchini seeds, aging conditions of 41°C for 96 hrs gave the best result in evaluating the potential of physiological seed quality (Dutra and Vieira, 2006). Aging of the flax seeds over a period of 42°C for 72 hrs resulted in complete loss of germination and failed to separate seeds into vigor levels and greater difference in seed vigor levels was observed at the aging conditions of 42°C for 48 hrs (Diederichsen and Jones-Flor, 2005). Series of accelerated aging conditions were evaluated in chilli, eggplant and tomato and the best aging conditions as seed vigor test were 45°C for 72 hrs, 40°C for 96 hrs and 45°C for 48 hrs, respectively (Thakan, 2004). According to Rodo and Filho (2003), the aging conditions of 41°C for 48 and 72 hrs could be used in evaluating the potential of physiological quality of onion seeds.

2.6.6 Prediction of field emergence

Various investigation relating the results of the accelerated aging test to field emergence among crop seeds revealed that accelerated aging test could predict field emergence such as in seeds of Wheat (Tomer and Maguire, 1990), sweet corn (singhabumrung and Juntakol, 2004), soybean (Egli and TeKrony 1995; Torres et al., 2004), cotton (Bishnoi and Delouche 1980), watermelon (Mavi and Demir 2007), pepper (Sundstrom et al. 1986) and rice (Chea, 2006).

Among the three test, high correlations were observed between field emergence and seedling vigor test (r = 0.983**), accelerated aging at 45°C for 48 hrs

(r = 0.714**) and standard germination test (r = 0.698**) in wheat seeds (Tomer and Maguire, 1990). In contrast to the above finding, Venter van de et al. (1993) did not find any correlation between accelerated aging conditions of 45°C for 48 hrs (r = 0.13 and r = 0.18) and field emergence in wheat seeds. In sweet corn, accelerated aging at 41°C for 96 hrs and 43°C for 72 hrs were the best filed emergence predictor (r = 0.96**) followed by conductivity test (r = 0.95**) and complex stress vigor test (r = 0.91**), respectively (Singhabumrung and Juntakool, 2004). Egli and TeKrony (1995) studied standard germination, accelerated aging test at 41°C for 72 hrs and cold test in soybean seeds for 10 years under different field conditions and demonstrated that soybean seed lots with germination after aging higher than 80% or standard germination above 95% had a high probability of producing adequate seedling emergence under a relatively high variation of environmental conditions. However, different result was obtained by Torres et al. (2004), where soybean seeds with germination after aging (42°C for 48 hrs) was above 90% showed field emergence more than 80% (r = 0.94**). According to Bishnoi and Delouche (1980), the cold test and accelerated aging test (42°C for 144 hrs) were adequate for predicting the levels of deterioration and field performances of cotton seeds. In watermelon seeds also accelerated aging at 45°C for 120-144 hrs provided highest correlation with field emergence (Mavi and Demir, 2007).

2.6.7 Prediction of seed storage

The storability of seeds in a specific environment is largely determined by its inheritance and pre-storage history. Inherent differences in longevity among species and varieties are biological facts over which seed specialist have no control. These differences, however, must be recognized and taken into account in planning for storage. The pre-storage history of seeds however is controllable. Timely harvesting

and threshing, prompt and adequate drying and careful handling minimize quality losses from field exposure, high moisture content, and mechanical damage and contribute to a seed history favorable for storage (Delouche et al. 1973).

Accelerated aging was first used as test to estimate longevity of seeds in warehouse storage (Delouche, 1965 quoted in AOSA (1983)). The seeds that had a high survival after accelerated aging stored well, while the seeds that were severely reduced in germination by accelerated aging declined rapidly in storage (Delouche and Baskin, 1973). Results from 6-years study involving many lots of 16 different seed kinds showed that germinative responses after accelerated aging were highly correlated with responses in storage under a variety of conditions for periods up to 3 years (Delouche and Baskin, 1973). According to Santipracha et al. (1993), accelerated aging at 44ºC for 96 hrs had better correlation to the quality of corn seeds packed in paper bags and stored for one year. Similarly in mungbean Santipracha et al. 1993 (a) and 1993 (b) reported that accelerated aging of 43ºC for 96 hrs gave the best longevity evaluation in the humid tropics. A temperature of 37ºC caused accelerated aging of soybean seeds and provides a satisfactory model for seed deterioration under normal storage conditions (Likhatchev et al. 1984).

2.6.8 Restriction

Certain precaution needs to be taken to reduce variability in test results, like initial moisture had been reported to be a factor causing variation in test results. In using testing apparatus like sealed jar or box method, variations in the distance between seeds and water surface can cause variations in test results (Tao, 1979 quoted in AOSA (1983). Seeds should be either treated or untreated (ISTA, 1995; AOSA, 1983). Variation in temperature and other environmental conditions are also critical for accelerated aging test. A 1ºC difference in temperature during the period of the

standard germination test probably has little effect on the percent germination obtained, but a 1ºC difference in temperature for an extended period during accelerated aging test might have a considerable effect on the deterioration of seeds (Copeland and McDonald, 1995). Therefore, strict procedure needs to be followed to avoid any variation in the result. Sufficient evidence has now accumulated to show that repeatable results can be achieved using accelerated aging as seed vigor test. Only slight modifications in temperature control, sample size or aging time will cause variation in final seed moisture or germination which will limit the acceptance of vigor test (AOSA, 1983)

From the previous researches and suggestions on the use of accelerated aging as seed vigor test in many crop seeds and with the popularity of this test in rice seed centers in Thailand, the scope of using the accelerated aging as vigor test in rice should be greatly enhanced.

2.6.9 Accelerated aging conditions for rice seed vigor test

Few researchers have been conducted in accelerated aging test as rice seed vigor test and there are no suggested or recommended accelerated aging test variables available in the handbook on vigor testing from the ISTA (1995) or the AOSA (1983). Patin and Gutormson. (2009) studied 10 rice seed lots to examine possible vigor tests for rice seeds. They evaluated several seed vigor tests such as sand germination, cold germination, cool germination, accelerated aging and conductivity test in rice seeds. Based on their finding, it was concluded that cold test and accelerated aging test might be suitable in evaluating vigor in rice seeds. Similarly, Chea (2006) evaluated several vigor tests such as seedling growth rate, seedling growth test, osmotic stress test, accelerated aging at 41ºC for 84 hrs and conductivity test and their use in predicting field emergence in 2 rice varieties (KDML105 and

RD6) of Thailand. He observed that accelerated aging, conductivity test, seedling growth test and standard germination test could be used in predicting field emergence of rice seeds but the accuracy was strongly dependent on field environment and cultural practices.

Some seed analysts and organizations have been using accelerated aging as seed vigor test in rice as presented in (table 2.2).

Table 2.2 Accelerated aging test conditions used in different rice seed laboratories as seed vigor test.

No.	Aging Condition	Source	Reference
1	42ºC, 72 hrs	Chaingmai, Rice Seed Center, Thailand	Singkanipa (2008)
2	45ºC, 96 hrs	Nakhon Ratchasima Rice Seed Center, Thailand	Singkanipa (2008)
3	41ºC, 72 hrs	CABI Abstract, USA.	Patin and Gutormson (2009)
4	41ºC, 84 hrs	Khon Kaen University, Thailand	Chea (2006)

Although the rice seed centers in Thailand have been doing the accelerated aging test as routine rice seed vigor test but there is no standard aging conditions which could predict filed emergence (Singkanipa, 2008). The accelerated aging conditions of 41ºC for 72 hrs of Mid-West Seed Service, South Dakota, America may not be applicable in Thailand because of difference in genetic constituents, paddy field environment and growing techniques.

CHAPTER III

MATERIALS AND METHODS

3.1 Experiments

Experiment I. Identification of accelerated aging conditions for seed vigor test in rice (*Oryza sativa* L).

Experiment II. Verification of recommended accelerated aging test condition and conductivity test as rice seed vigor tests.

3.2 Period of study

The research works of experiment I and II were carried out from July 2008 to March 2009 and April 2009, respectively.

3.3 Place of study

1) All the laboratory tests were conducted at Seed Technology Laboratory, Center for Science Equipment and Technology building 3 at Suranaree University of Technology, Nakhon Ratchasima Province, Thailand.

2) The field emergence tests were conducted at the University Farm at Suranaree University of Technology, Nakhon Ratchasima Province, Thailand.

3.4 Experiment I. Identification of accelerated aging conditions for seed vigor test in rice

The accelerated aging conditions currently practiced in Thailand are not

accurate, therefore the objective of this experiment was to develop a proper testing condition of temperature and aging period for accelerated aging test as rice seed vigor test of various recommended Thai varieties. Standard germination, field emergence, several seed vigor tests and 9 accelerated aging conditions were tested and statistic correlations with field emergence were observed.

3.4.1 Statistical design

Completely Randomized Design of 24 seed lots with 3 replications were used in experiment I. The rice varieties used in experiment I were 1) Khao Dawk Mali 105 (KDML105), 2) Chai Nat 1 (CNT1), and 3) Phitsanulok1 (PSL1). Each variety consisted of 8 seed lots with different vigor levels ranging from 40-80% as determined by field emergence test. Nine accelerated aging conditions of seeds are presented in table 3.1. The quality of selected seed lots were determined by standard germination test, seedling root length, seedling shoot length, total seedling length, seedling growth rate test, conductivity test and field emergence test.

3.4.2 Seed procurement

One hundred sixty four seed lots of different rice varieties were procured from various rice seed centers and rice research centers in Thailand. The seed lots were produced in the multiplication fields either at research stations and contract growers' fields. The seeds were cleaned by oblong screen (1.785 X 12.7 mm) and South Dakota seed blower. The seeds were air dried in the laboratory before packing for storage. The seed moisture measured by electrical moisture meter (lab model Steinlite 900, Seedburo Equipment Company, USA.) ranged from 10-12%. To prevent seed deterioration, seed lots were packed in sealed plastic bags and stored in cold storage at 5°C and 60% R.H. until used in the experiments. A preliminary field emergence test was conducted and accordingly 24 seed lots of three rice varieties of

different vigor levels ranging from 40-80% of field emergence were selected for the experiment I (table 3.1).

3.4.3 Seed testing procedures

Working sample were drawn from each seed lots and tested for

1) Standard germination test (SG)

2) Field emergence test (FE)

3) Five vigor tests

   3.1) Seedling root length (RL)

   3.2) Seedling shoot length (SL)

   3.3) Total seedling length (TL)

   3.4) Seedling growth rate test (SGR)

   3.5) Conductivity test (CT)

4) Accelerated aging test (AA) at 9 conditions as mentioned in table 3.2

The following procedures were followed for each quality test

3.4.3.1 Standard germination test (SGT)

Three replications of 50 seeds were drawn from each seed lot. The seeds were germinated by between papers (BP) method at 25^{o}C and 8 hours light in the germinator. Seedling evaluation were done at 5 and 10 days after incubation following the procedure for standard germination test as described in the ISTA rules for seed testing (ISTA, 1999).

3.4.3.2 Field emergence test (FE)

Three replications of 50 seeds were planted in concrete plots (19.0 m x 0.80 m) in plastic roofed house. The soil was sandy clay loam. The seeds were hand planted (50 seeds per row), in September 2008. The plots were covered with plastic nets and rotenticide (flocoumafen; common name) was applied in and around the plots

to prevent bird and rat damages, respectively. The plots were watered immediately after planting the seeds through pipe and every day thereafter. Healthy seedlings were counted at 15 days after sowing.

3.4.3.3 Seedling root length (RL)

Between paper (BP) germination test was used. Three replications of 25 seeds were germinated between germination paper towels at 25^{o}C in the dark germinator. Each replication consisted of three paper towels, two below the seeds and one covering the seeds. The seeds were placed at the center of the paper towels in a straight line with the radicle end towards the bottom of the towels. Once the seeds were covered with the third towel, the paper towels were folded from both sides towards the middle in a rectangle shape of 13 cm in width. To prevent the bending of paper towels only 5 sets of paper towels were placed in a plastic box (W x L x H = 18.5 x 27.5 x 10.0 cm), then put in a black plastic bag to create dark germination. The plastic boxes were placed in a 45º upright position in the germinator. The seedling root length of only normal seedlings were measured in cm at 7 days after planting. Means of seedling root length then were calculated in cm per seedling (ISTA, 1995)

3.4.3.4 Seedling shoot length (SL)

The same samples used in the determination of seedling root length as described in 3.4.3.3 were used to measure seedling shoot length. Only normal seedlings were measured for seedling shoot length in cm. Means of seedling shoot length then were calculated in cm per seedling**.**

3.4.3.5 Total seedling length (TL)

Mean of total seedling length of each seed lot was determined by the accumulation of seedling root and shoot length means of each seedling as demonstrated in 3.4.3.3 and 3.4.3.4, respectively.

3.4.3.6 Seedling growth rate test (SGR)

Three replications of 25 seeds were germinated between papers at 25^{o}C in a 45^{o} upright position in the dark germinator as described in 3.4.4.3. After 7 days the seedlings were evaluated. Abnormal seedlings and dead seeds were discarded. Normal seedlings were cut free from seeds and oven dried at 80^{o}C for 24 hrs. After the drying period the seedlings were weighed to 3 decimal places. The total weight of dry seedlings per roll of germination paper towels was than divided by total number of normal seedlings. The seedling growth rate was expressed in gram per plant as described by ISTA (1955) seed vigor testing handbook.

3.4.3.7 Conductivity test (CT)

Three replications of 25 uninjured seeds of each seed lot were weighed to 2 decimal places. The seeds of each replication were placed in a 200 ml beaker and 75 ml of deionized water was added. The seeds were gently stirred by stirring rod to ensure that all seeds were completely immersed and evenly distributed. All beakers were covered by aluminum foil to reduce contaminations. The beakers were placed at the constant temperature of 20°C for 24 hours. The electrical conductivity of leachates of each replication was measured by using conductivity meter (model Consort C 831, Cole-Parmer Instrument Company, Belgium) and conductivity per gram of seed weight was calculated ($\mu S\ cm^{-1}g^{-1}$) and recorded as per the AOSA (1983) seed vigor testing handbook.

3.4.3.8 Accelerated aging test (AA)

Three replications of 50 seeds of each seed lot were placed one layer deep in wire mesh baskets (8 cm in diameter and 9 cm in length including legs which the legs are 3.5 cm in height), One hundred twenty ml of water was then added to the plastic box (10 cm in diameter and 10.5 cm in height). To prevent the dropping of

condensed vapour from the lid into the seeds, one piece of blotter paper of circular shape was placed on eash basket. The lid of the plastic box was then tightly closed and placed into incubator at temperature and hours as presented in table 3.2. Subsequent to aging treatments the seeds were germinated as the procedure described in 3.4.3.1, the standard germination test.

3.4.4 Statistical analysis

Analysis of variances were performed on the data with the Statistical Package for Social Sciences (SPSS) version 15 (Little and Hills, 1972). Duncan's multiple range test (DMRT) was used in the mean comparisons. Correlation co-efficients between all test results were calculated to observe the relationships of all tests.

3.5 Experiment II. Verification of recommended accelerated aging test condition and conductivity test as rice seed vigor test

According to the results of experiment I, accelerated aging condition at 44ºC for 72 hrs was recommended for accelerated aging test as seed vigor test in rice. Furthermore, conductivity test was also recommended to use as supplement or substitute test for rice seed vigor test. However, it was necessary to verify these recommendations before making finale suggestions. In order to verify the above recommendations as seed vigor tests in rice, high variations of rice seed lots and varieties of different vigor levels must be included in both recommended vigor tests and observations of correlation coefficients with field emergence were observed.

3.5.1 Statistical design

A completely randomized design of 60 seed lots of rice varieties and 3 vigor tests; field emergence, conductivity test and accelerated aging condition at 44ºC

for 72 hrs, were included in experiment II.

3.5.2 Seeds procurement

From 164 seed lots procured from various rice seed centers and rice research centers as mentioned in 3.4.2, 60 seed lots of 9 rice varieties which were not yet used in experiment I were selected for experiment II (table 3.3). Before selection, seed lots were tested for field emergence to identify vigor levels. The selected lots had field emergence percentages in the range of 40-85.

3.5.3 Seed testing procedures

Working samples were drawn from each seed lots and tested for

(1) Field emergence test

(2) Conductivity test

(3) Accelerated aging at 44ºC for 72 hours

The testing procedures of these tests were the same as described in 3.4.3.2, 3.4.3.7 and 3.4.3.8, respectively.

3.5.4 Statistical analysis

Analysis of variances were performed on the data with the Statistical Package for Social Sciences (SPSS) version 15 (Little and Hills, 1972). Duncan's multiple range test (DMRT) was used in the mean comparisons. Correlation co-efficients between all test results were calculated to observe the relationships of all tests.

Table 3.1 Twenty four seed lots of 3 rice varieties included in experiment I.

No.	Lot No.	Variety	Seed Source	Seed Class	Production Season and Year
1	8	CNT1	Pattani Rice Seed Center	ES	RS 07
2	24	CNT1	Surat Thani Rice Seed Center	FS	RS 08
3	34	CNT1	Khon Kaen Rice Seed Center	ES	DS 08
4	36	CNT1	Khon Kaen Rice Seed Center	ES	DS 08
5	54	CNT1	Phatthalung Rice Seed Center	RS	DS 08
6	71	CNT1	Phatthalung Rice Seed Center	FS	RS 08
7	72	CNT1	Phatthalung Rice Seed Center	FS	RS 08
8	85	CNT1	Nakhon Sawan Rice Seed Center	RS	RS 08
9	5	KDML105	Nakhon Ratchasima Rice Seed Center	ES	RS 07
10	46	KDML105	Chiang Mai Rice Seed Center	FS	RS 07
11	48	KDML105	Chiang Mai Rice Seed Center	FS	RS 07
12	9	KDML105	Nakhon Ratchasima Rice Seed Center	FS	RS 08
13	82	KDML105	Phitsanulok Rice Seed Center	FS	RS 08
14	88	KDML105	Nakhon Ratchasima Rice Seed Center	ES	RS 08
15	90	KDML105	Nakhon Ratchasima Rice Seed Center	ES	RS 08
16	106	KDML105	Nakhon Ratchasima Rice Seed Center	ES	RS 08
17	128	PSL1	Phitsanulok Rice Seed Center	FS	RS 08
18	129	PSL1	Phitsanulok Rice Seed Center	FS	RS 08
19	130	PSL1	Phitsanulok Rice Seed Center	FS	RS 08
20	131	PSL1	Phitsanulok Rice Seed Center	FS	RS 08
21	132	PSL1	Phitsanulok Rice Seed Center	ES	RS 08
22	133	PSL1	Phitsanulok Rice Seed Center	FS	RS 08
23	134	PSL1	Phitsanulok Rice Seed Center	ES	RS 08
24	135	PSL1	Phitsanulok Rice Seed Center	ES	RS 08

Table 3.2 Nine accelerated aging conditions used in experiment I.

Treatment	Temperature (ºC)	Hour
1	42	72
2	42	96
3	42	120
4	43	72
5	43	96
6	43	120
7	44	72
8	44	96
9	44	120

Table 3.3 Sixty seed lots of 10 rice varieties included in experiment II.

No.	Lot No.	Variety	Seed Source	Seed Class	Production Season and Year
1	14	CNT1	Surat Thani Rice Seed Center	FS	DS 08
2	15	CNT1	Surat Thani Rice Seed Center	FS	DS 08
3	20	CNT1	Surat Thani Rice Seed Center	FS	DS 08
4	29	CNT1	Surat Thani Rice Seed Center	FS	RS 08
5	33	CNT1	Surat Thani Rice Seed Center	FS	RS 08
6	35	CNT1	Khon Kaen Rice Seed Center	ES	DS 08
7	38	CNT1	Khon Kaen Rice Seed Center	ES	DS 08
8	40	CNT1	Khon Kaen Rice Seed Center	ES	DS 08
9	53	CNT1	Phatthalung Rice Seed Center	RS	DS 08
10	57	CNT1	Phatthalung Rice Seed Center	RS	DS 08
11	65	CNT1	Phatthalung Rice Seed Center	RS	DS 08
12	69	CNT1	Phatthalung Rice Seed Center	RS	DS 08
13	70	CNT1	Phatthalung Rice Seed Center	FS	RS 08
14	73	CNT1	Phatthalung Rice Seed Center	FS	RS 08
15	75	CNT1	Phatthalung Rice Seed Center	FS	RS 08
16	79	CNT1	Phatthalung Rice Seed Center	FS	RS 08
17	83	CNT1	Nakhon Sawan Rice Seed Center	RS	-
18	84	CNT1	Nakhon Sawan Rice Seed Center	RS	-
19	86	CNT1	Nakhon Sawan Rice Seed Center	RS	-
20	87	CNT1	Nakhon Sawan Rice Seed Center	RS	-

Table 3.3 Sixty seed lots of 10 rice varieties included in experiment II. (continued)

No.	Lot No.	Variety	Seed Source	Seed Class	Production Season and Year
21	46	KDML105	Chiang Mai Rice Seed Center	FS	RS 07
22	47	KDML105	Chiang Mai Rice Seed Center	FS	RS 07
23	82	KDML105	Phitsanulok Rice Seed Center	-	-
24	93	KDML105	Nakhon Ratchasima Rice Seed Center	-	-
25	98	KDML105	Nakhon Ratchasima Rice Seed Center	-	-
26	110	KDML105	Nakhon Ratchasima Rice Seed Center	-	-
27	114	KDML105	Nakhon Ratchasima Rice Seed Center	-	-
28	118	KDML105	Nakhon Ratchasima Rice Seed Center	-	-
29	121	KDML105	Nakhon Ratchasima Rice Seed Center	-	-
30	128	PSL1	Phitsanulok Rice Seed Center	-	-
31	129	PSL1	Phitsanulok Rice Seed Center	-	-
32	130	PSL1	Phitsanulok Rice Seed Center	-	-
33	132	PSL1	Phitsanulok Rice Seed Center	-	-
34	134	PSL1	Phitsanulok Rice Seed Center	-	-
35	44	SPT	Phrae Rice Seed Center	FS	RS 08
36	45	SPT	Phrae Rice Seed Center	FS	RS 08
37	80	SMP	Phatthalung Rice Seed Center	FS	RS 07
38	81	SMP	Phatthalung Rice Seed Center	FS	RS 07
39	49	IR6	Chiang Mai Rice Seed Center	FS	RS 07
40	50	IR6	Chiang Mai Rice Seed Center	FS	RS 07

Table 3.3 Sixty seed lots of 10 rice varieties included in experiment II. (continued)

No.	Lot No.	Variety	Seed Source	Seed Class	Production Season and Year
41	51	IR6	Chiang Mai Rice Seed Center	FS	RS 08
42	52	IR15	Chiang Mai Rice Seed Center	FS	-
43	136	IR15	Phitsanulok Rice Seed Center	FS	-
44	137	IR15	Phitsanulok Rice Seed Center	-	-
45	138	IR15	Phitsanulok Rice Seed Center	-	
46	139	IR15	Phitsanulok Rice Seed Center	FS	-
47	144	SPR1	Suphan Buri Rice Research Center	BS	RS 07
48	147	SPR1	Suphan Buri Rice Research Center	BS	DS 08
49	148	SPR1	Suphan Buri Rice Research Center	FS	DS 08
50	149	SPR1	Suphan Buri Rice Research Center	FS	DS 08
51	152	SPR2	Suphan Buri Rice Research Center	BS	RS 07
52	153	SPR2	Suphan Buri Rice Research Center	BS	RS 07
53	154	SPR2	Suphan Buri Rice Research Center	BS	RS 07
54	155	SPR2	Suphan Buri Rice Research Center	BS	RS 07
55	157	SPR2	Suphan Buri Rice Research Center	FS	RS 07
56	158	SPR2	Suphan Buri Rice Research Center	FS	DS 08
57	159	SPR2	Suphan Buri Rice Research Center	FS	DS 08
58	163	SPR3	Suphan Buri Rice Research Center	FS	DS 08
59	164	SPR3	Suphan Buri Rice Research Center	FS	DS 08
60	160	SPR3	Suphan Buri Rice Research Center	FS	DS 08

CHAPTER IV

RESULTS

4.1 Experiment I. Identification of accelerated aging conditions for seed vigor test in rice.

In order to identify the accelerated aging conditions for seed vigor test in rice, 24 seed lots of 3 rice varieties of different vigor levels were tested for standard germination test, field emergence, 9 accelerated aging conditions and several vigor tests. The statistical differences of means were examined using Duncan's multiple range test and correlation coefficients among all the tests were observed.

4.1.1 Seed quality of 24 rice seed lots of 3 varieties tested by standard germination, field emergence, 5 seed vigor tests and 9 accelerated aging conditions.

Seed quality of 24 seed lots determined by standard germination, field emergence, 5 seed vigor tests and 9 accelerated aging conditions are shown in table 4.1-4.4 and appendix table 1 and 2. The germination percentages of 24 seed lots ranged from 44.00 to 99.33% with average of 88.69% and very highly significant differences ($p < 0.01$) were observed among means of 24 seed lots. When seeds were planted in the field, field emergence percentages showed significant difference ($p < 0.05$) between means of seed lots. From the results of 5 seed vigor tests; seedling root length, seedling shoot length, total seedling length, seedling growth rate and conductivity test, each test showed highly significant differences among its seed lot means except seedling growth rate test which was non-significant. Each accelerated

aging condition also showed highly significant difference among 24 seed lots (table 4.2 and 4.4).

According to high coefficient of variation (C.V.) of 4 accelerated aging conditions (44ºC, 120 hrs, 43ºC, 120 hrs, 44ºC, 96 hrs and 44ºC, 120 hrs) (table 4.2 and 4.4), therefore the data were transformed by square root method.

4.1.2 Correlation coefficients amongst standard germination test, field emergence, 5 vigor tests and 9 accelerated aging conditions of 24 seed lots of rice.

Correlation coefficients of standard germination test, 5 seed vigor tests and 9 accelerated aging conditions of 24 seed lots of rice are shown in table 4.5. The relationships among standard germination test, field emergence, conductivity test and accelerated aging conditions are presented in figure 4.1-4.17.

Highly significant correlation (r = 0.64**) was observed between standard germination and field emergence tests. All seed vigor tests; seedling root length (r = 0.56**), seedling shoot length (r = 0.75**), total seedling length (r = 0.63**), seedling growth rate test (r = 0.55**) and conductivity test (r = -0.82**) provided highly significant correlations with field emergence. Correlations of 9 accelerated conditions with field emergence were also highly significant (r = 0.55** to 0.78**).

Among standard germination test, 5 seed vigor tests and 9 accelerated aging conditions, the three highest correlations with field emergence were conductivity test (r = -0.82**) followed by accelerated aging condition at 44ºC for 72 hrs (r = 0.78**) and seedling shoot length (r = 0.75**), respectively. According to conductivity test, seedling shoot length showed highest correlation (r = -0.82**) followed by accelerated aging condition at 44ºC for 72 hrs (r = -0.71**).

Negative correlations were always observed between conductivity test and standard germination test and other seed vigor tests. This is because low germination and vigor seeds give high amount of leakage of electrolytes (measured in $\mu S\ cm^{-1}g^{-1}$), in contrast high vigor seeds give low amount of leakage of electrolytes.

Standard germination test showed highly significant correlations with all tests except accelerated aging test conditions at 44ºC for 96 and 120 hrs. Highly significant correlation coefficients were observed between standard germination test and seedling shoot length (r = 0.93**), total seedling length (r = 0.92**), seedling root length (r = 0.88**), and accelerated aging at 43ºC for 72 hrs (r = 0.80**). Correlation coefficient of standard germination test and field emergence test was only 0.64** which indicated the deficiencies of standard germination test on vigor determination for prediction of field emergence.

From the results of maximum correlation coefficients (r) of accelerated aging condition at 44ºC for 72 hrs (r = 0.78**) and conductivity test (r = -0.82**) with field emergence obtained in experiment I, accelerated aging condition at 44ºC for 72 hrs is recommended as preliminary recommendation for rice seed vigor test and conductivity test is also recommended as alternative or substitute test for rice seed vigor test. Seedling shoot length seems to be good rice vigor test, but was not included in the recommendation, as it is a time consuming and tedious test, therefore it may not be practical as rice seed vigor test in rice seed laboratories.

4.2 Experiment II. Verification of recommended accelerated aging test condition and conductivity test as rice seed vigor tests.

According to the highest correlation coefficients (r) showed among field emergence, conductivity test and accelerated aging condition at 44ºC for 72 hrs,

therefore accelerated aging test at 44ºC for 72 hrs and conductivity were recommended for rice seed vigor tests in rice. Conductivity test is rapid, simple and do not need personal skill for result analysis, so besides accelerated aging test, conductivity test may be used as alternative or substitute test for rice seed vigor test. Sixty seed lots of six rice varieties which were not used in experiment I, representing various rice seed centers and rice research centers in Thailand were used in experiment II to verify the recommendations made from the results of experiment I.

4.2.1 Seed quality of sixty rice seed lots (10 varieties) tested by field emergence, conductivity test and accelerated aging condition at 44ºC for 72 hrs.

Seed sources and quality of sixty seed lots determined by field emergence, conductivity test and accelerated aging condition are shown in table 4.6 and 4.7. Field emergence percentages ranged from 13.33 to 85.00% with mean of 55.15 %. The field emergence showed very highly significant differences among seed lots. Highly significant differences among sixty seed lots were also observed in conductivity test results. The germination percentages after accelerated aging test ranged from 0.00 to 90.66% and germination percent mean was 51.04%. The accelerated aging test also showed highly significant differences in seed quality among sixty seed lots.

4.2.2 Correlation coefficients among field emergence, conductivity tests and accelerated aging condition at 44ºC for 72 hrs of 60 seed lots of rice (10 varieties).

Similar correlations among all tests as found in experiment I were also observed in experiment II, but the correlations in experiment II were higher than in experiment I. This was due to less variation among test results as the inclusion of

many seed lots (60) in experiment II. Correlation coefficients among conductivity test, accelerated aging at 44ºC for 72 hrs and field emergence of 60 seed lots of 10 rice varieties are shown in table 4.8.

The accelerated aging at 44ºC for 72 hrs showed highest correlation with field emergence (r = 0.89**) followed by conductivity test (r = -0.86**). Highly significant correlation (r = -0.77**) was observed between accelerated aging at 44ºC for 72 hrs and conductivity test.

From the above findings, we can recommend that accelerated aging at 44ºC for 72 hrs and conductivity test are vigor tests of common rice varieties in Thailand. Conductivity test can be used as alternative or substitute test in rice seed vigor.

Table 4.1 Standard germination and vigor tests of 24 seed lots of 3 rice varieties, data sorted according to varieties and minimum to maximum percentages of standard germination test.

Variety	Lots	Vigor Test						
		SG (%)	**FE** (%)	**RL** (cm)	**SL** (cm)	**TL** (cm)	**SGR** (mg/plant)	**CT** (µS cm-¹g-¹)
CNT1	36	44.00 h [1]	31.33 e	2.84 h	0.79 g	3.63 i	0.160 b	81.98 a
CNT1	72	80.67 g	48.67 c-e	8.49 g	3.67 e-f	12.16 g-h	4.467 a	31.36 d-g
CNT1	34	82.00 f-g	66.00 a-e	9.29 f-g	3.95 a-f	13.23 f-h	4.125 a	28.25 e-g
CNT1	71	83.33 e-g	61.33 a-e	8.53 g	3.36 f	11.89 h	3.096 a-b	33.34 d-e
CNT1	85	90.67 b-e	79.33 a-c	9.62 f-g	4.31 a-f	13.93 e-h	5.436 a	27.76 e-g
CNT1	54	93.33 a-d	76.00 a-d	11.19 d-f	4.44 a-e	15.64 c-f	5.257 a	26.76 e-g
CNT1	8	94.00 a-d	85.33 a-b	10.65 d-g	4.44 a-e	15.09 c-g	5.004 a	23.57 g
CNT1	24	94.67 a-d	86.67 a	11.02 d-f	4.78 a-d	15.81 b-f	4.430 a	23.12 g
KDML105	48	88.67 c-g	42.00 d-e	10.94 d-f	3.60 e-f	14.54 d-h	3.246 a-b	34.15 d-e
KDML105	46	88.67 c-g	65.33 a-e	10.79 d-f	4.09 c-f	14.88 c-h	5.166 a	32.97 d-f
KDML105	90	89.33 c-f	49.33 b-e	10.24 e-g	3.92 d-f	14.16 e-h	4.908 a	35.15 d-e
KDML105	5	90.67 b-e	72.00 a-d	11.99 b-e	4.43 a-e	16.42 b-e	5.759 a	24.56 f-g
KDML105	82	94.67 a-d	76.67 a-d	12.80 a-d	4.95 a-c	17.75 a-c	6.582 a	27.54 e-g
KDML105	106	96.00 a-c	54.67 a-e	12.38 a-e	4.94 a-c	17.32 a-d	4.366 a	44.18 b-c
KDML105	9	98.67 a-b	84.00 a-c	14.28 a	5.23 a	19.51 a	4.960 a	24.87 f-g
KDML105	88	99.33 a	80.00 a-c	13.55 a-c	5.11 a-b	18.66 a-b	5.933 a	24.83 f-g
PSL 1	135	83.33 e-g	64.67 a-e	11.40 c-f	3.65 e-f	15.05 c-g	6.645 a	44.10 b-c
PSL 1	132	87.33 d-g	60.67 a-e	10.34 e-g	3.51 e-f	13.86 e-h	4.920 a	37.28 c-d
PSL 1	134	88.00 c-g	56.00 a-e	13.65 a-b	3.75 e-f	17.41 a-d	6.555 a	48.04 b
PSL 1	131	89.33 c-f	60.00 a-e	10.44 e-g	3.61 e-f	14.06 e-h	6.080 a	38.09 c-d
PSL 1	133	91.33 a-e	60.67 a-e	11.11 d-f	3.72 e-f	14.83 c-h	4.924 a	36.80 c-d
PSL 1	129	92.67 a-d	57.33 a-e	10.33 e-g	3.85 d-f	14.18 e-h	4.539 a	47.52 b
PSL 1	130	94.00 a-d	52.00 a-e	11.37 c-f	4.13 b-f	15.51 c-f	5.348 a	35.16 d-e
PSL 1	128	94.00 a-d	70.00 a-d	12.01 b-e	4.24 a-f	16.25 b-f	5.593 a	33.54 d-e
Mean		**88.69**	**64.17**	**10.8**	**4.02**	**14.82**	**4.895**	**35.21**
F test		**	*	**	**	**	ns	**
C.V. (%)		**4.87**	**28.25**	**10.74**	**12.9**	**10.66**	**38.7**	**12.48**

[1] = Means in the same column that followed by the same letters are not significantly different according to DMRT.

*, **, ns = Significant difference at $p < 0.05$, $p < 0.01$ and non-significant, respectively.

Table 4.2 Accelerate aging test of 9 conditions of 24 seed lots of 3 rice varieties, data sorted according to varieties and minimum to maximum percentages of standard germination test of Table 4.1.

Variety	Lots	Accelerated Aging Condition								
		42 °C 72 hrs (%)	**42 °C 96 hrs (%)**	**42 °C 120 hrs (%)**	**43 °C 72 hrs (%)**	**43 °C 96 hrs (%)**	**43 °C 120 hrs (%)**	**44 °C 72 hrs (%)**	**44 °C 96 hrs (%)**	**44 °C 120 hrs (%)**
CNT1	36	13.33 g[1]	1.33 k	0.00 f	29.33 e	11.33 g	4.00 d	0.00 i	0.00 f	0.00 h
CNT1	72	73.33 a-e	65.33 b-e	42.00 a-d	79.33 a-c	77.33 a-c	33.33 b-c	27.33 g-h	4.67 e-f	2.00 g-h
CNT1	34	44.67 f	41.33 g-j	20.67 c-e	50.00 d	58.00 c-f	58.67 a-c	56.67 b-g	38.00 a-d	19.33 b-f
CNT1	71	73.33 a-e	57.33 c-g	28.67 b-e	86.67 a-b	72.66 a-d	54.66 a-c	35.00 f-h	12.67 de	1.33 g-h
CNT1	85	90.00 a-b	57.33 c-g	45.33 a-d	86.00 a-b	80.67 a-c	81.33 a	78.00 a-b	32.67 a-d	20.67 b-f
CNT1	54	78.00 a-d	73.33 a-c	70.00 a	85.33 a-b	86.67 a-b	48.00 a-c	79.33 a-b	70.00 a	45.33 a-b
CNT1	8	81.33 a-c	51.33 c-g	48.00 a-c	90.67 a-b	66.00 a-f	64.00 a-c	59.00 b-f	32.67 b-d	6.00 d-h
CNT1	24	84.00 a-b	82.00 a-b	53.33 a-b	90.67 a-b	90.00 a	84.00 a	95.33 a	65.33 a-b	62.00 a
KDML105	48	62.67 d-e	31.33 j	22.00 c-e	71.33 b-c	47.33 d-f	28.00 c	21.33 h-i	13.33 d-f	4.00 d-h
KDML105	46	85.33 a-b	48.00 e-j	28.00 b-e	79.33 a-c	43.33 e-f	36.67 a-c	52.00 b-g	34.67 a-d	22.67 b-d
KDML105	90	79.33 a-d	50.67 d-i	21.33 c-e	80.00 a-c	74.67 a-d	63.33 a-c	42.00 d-h	22.00 c-e	0.00 h
KDML105	5	65.33 c-e	35.33 h-j	28.00 b-e	64.67 c-d	58.00 c-f	60.00 a-c	67.33 a-e	38.67 a-d	26.00 b-c
KDML105	82	86.67 a-b	57.33 c-g	32.00 b-e	92.00 a	78.00 a-c	79.33 a	76.67 a-c	59.33 a-c	40.67 a-b
KDML105	106	72.00 b-e	64.00 b-f	54.67 a-b	78.67 a-c	68.00 a-f	56.00 a-c	57.33 b-f	16.67 d-e	14.00 b-g
KDML105	9	82.67 a-c	68.00 a-d	29.33 b-e	91.33 a	65.33 a-f	64.00 a-c	60.67 b-f	28.00 a-d	17.33 c-g
KDML105	88	90.67 a	84.00 a	58.00 a-b	83.33 a-c	88.00 a-b	48.00 a-c	56.67 b-g	6.00 e-f	4.00 f-h

Table 4.2 Accelerate aging test of 9 conditions of 24 seed lots of 3 rice varieties, data sorted according to varieties and minimum to maximum percentages of standard germination test of Table 4.1. (continued)

Variety	Lots	Accelerated Aging Condition								
		42 °C 72 hrs (%)	**42 °C 96 hrs (%)**	**42 °C 120 hrs (%)**	**43 °C 72 hrs (%)**	**43 °C 96 hrs (%)**	**43 °C 120 hrs (%)**	**44 °C 72 hrs (%)**	**44 °C 96 hrs (%)**	**44 °C 120 hrs (%)**
PSL 1	135	79.33 a-d	62.00 c-f	12.00 e	77.33 a-c	69.33 a-e	56.00 a-c	39.67 e-h	29.33 b-d	10.67 c-g
PSL 1	132	80.00 a-d	32.00 i -j	11.33 e	80.00 a-c	42.00 f	62.00 a-c	62.33 b-f	15.33 d-e	3.33 e-h
PSL 1	134	74.00 a-e	41.33 g-j	38.00 a-e	74.67 a-c	63.33 a-f	64.67 a-c	70.67 a-d	40.00 a-d	3.33 d-h
PSL 1	131	74.67 a-e	45.33 f-j	18.00 d-e	81.33 a-c	74.67 a-d	50.00 a-c	47.33 c-h	33.33 a-d	13.33 b-g
PSL 1	133	60.00 e	41.33 g-j	30.67 b-e	64.67 c-d	58.00 c-f	42.00 a-c	58.00 b-f	9.33 d-f	10.00 c-h
PSL 1	129	84.00 a-b	55.33 c-g	26.00 b-e	80.00 a-c	72.00 a-d	55.33 a-c	50.00 b-h	24.00 c-e	6.00 c-h
PSL 1	130	81.33 a-c	46.67 e-j	30.00 b-e	82.67 a-c	77.33 a-c	72.67 a-b	62.67 b-f	15.33 d-e	8.67 c-h
PSL 1	128	82.00 a-c	56.67 c-g	39.33 a-d	80.00 a-c	61.33 b-f	45.33 a-c	68.00 a-e	31.33 a-d	20.00 b-e
Mean		**74.08**	**52.03**	**32.78**	**77.47**	**65.97**	**54.64**	**55.14**	**28.03**	**15.03**
F test		**	**	**	**	**	**	**	**	**
C.V. (%)		**12.2**	**19.2**	**24.57**	**12.6**	**21.16**	**22.22**	**27.98**	**35.55**	**50.77**

[1] = Means in the same column that followed by the same letters are not significantly different according to DMRT.
** = Significant difference at $p < 0.01$.

Table 4.3 Standard germination and vigor tests of 24 seed lots of 3 rice varieties, data sorted according to varieties and minimum to maximum percentages of field emergence test.

Variety	Lots	Vigor Test						
		SG (%)	FE (%)	RL (cm)	SL (cm)	TL (cm)	SGR (mg/plant)	CT ($\mu S\ cm^{-1}g^{-1}$)
CNT1	36	44.00 h [1]	31.33 e	2.84 h	0.79 g	3.63 i	0.160 b	81.98 a
CNT1	72	80.67 g	48.67 c-e	8.49 g	3.67 e-f	12.16 g-h	4.467 a	31.36 d-g
CNT1	71	83.33 e-g	61.33 a-e	8.53 g	3.36 f	11.89 h	3.096 a-b	33.34 d-e
CNT1	34	82.00 f-g	66.00 a-e	9.29 f-g	3.95 a-f	13.23 f-h	4.125 a	28.25 e-g
CNT1	54	93.33 a-d	76.00 a-d	11.19 d-f	4.44 a-e	15.64 c-f	5.257 a	26.76 e-g
CNT1	85	90.67 b-e	79.33 a-c	9.62 f-g	4.31 a-f	13.93 e-h	5.436 a	27.76 e-g
CNT1	8	94.00 a-d	85.33 a-b	10.65 d-g	4.44 a-e	15.09 c-g	5.004 a	23.57 g
CNT1	24	94.67 a-d	86.67 a	11.02 d-f	4.78 a-d	15.81 b-f	4.430 a	23.12 g
KDML105	48	88.67 c-g	42.00 d-e	10.94 d-f	3.60 e-f	14.54 d-h	3.246 a-b	34.15 d-e
KDML105	90	89.33 c-f	49.33 b-e	10.24 e-g	3.92 d-f	14.16 e-h	4.908 a	35.15 d-e
KDML105	106	96.00 a-c	54.67 a-e	12.38 a-e	4.94 a-c	17.32 a-d	4.366 a	44.18 b-c
KDML105	46	88.67 c-g	65.33 a-e	10.79 d-f	4.09 c-f	14.88 c-h	5.166 a	32.97 d-f
KDML105	5	90.67 b-e	72.00 a-d	11.99 b-e	4.43 a-e	16.42 b-e	5.759 a	24.56 f-g
KDML105	82	94.67 a-d	76.67 a-d	12.80 a-d	4.95 a-c	17.75 a-c	6.582 a	27.54 e-g
KDML105	88	99.33 a	80.00 a-c	13.55 a-c	5.11 a-b	18.66 a-b	5.933 a	24.83 f-g
KDML105	9	98.67 a-b	84.00 a-c	14.28 a	5.23 a	19.51 a	4.960 a	24.87 f-g
PSL 1	130	94.00 a-d	52.00 a-e	11.37 c-f	4.13 b-f	15.51 c-f	5.348 a	35.16 d-e
PSL 1	134	88.00 c-g	56.00 a-e	13.65 a-b	3.75 e-f	17.41 a-d	6.555 a	48.04 b
PSL 1	129	92.67 a-d	57.33 a-e	10.33 e-g	3.85 d-f	14.18 e-h	4.539 a	47.52 b
PSL 1	131	89.33 c-f	60.00 a-e	10.44 e-g	3.61 e-f	14.06 e-h	6.080 a	38.09 c-d
PSL 1	132	87.33 d-g	60.67 a-e	10.34 e-g	3.51 e-f	13.86 e-h	4.920 a	37.28 c-d
PSL 1	133	91.33 a-e	60.67 a-e	11.11 d-f	3.72 e-f	14.83 c-h	4.924 a	36.80 c-d
PSL 1	135	83.33 e-g	64.67 a-e	11.40 c-f	3.65 e-f	15.05 c-g	6.645 a	44.10 b-c
PSL 1	128	94.00 a-d	70.00 a-d	12.01 b-e	4.24 a-f	16.25 b-f	5.593 a	33.54 d-e
Mean		**88.69**	**64.17**	**10.8**	**4.02**	**14.82**	**4.895**	**35.21**
F test		**	*	**	**	**	ns	**
C.V. (%)		**4.87**	**28.25**	**10.74**	**12.9**	**10.66**	**38.7**	**12.48**

[1] = Means in the same column that followed by the same letters are not significantly different according to DMRT.

*, **, ns = Significant difference at $p < 0.05$, $p < 0.01$ and non-significant, respectively.

Table 4.4 Accelerate aging test of 9 conditions of 24 seed lots of 3 rice varieties, data sorted according to varieties and minimum to maximum percentages of field emergence test of Table 4.3.

Variety	Lots	Accelerated Aging Condition								
		42 °C 72 hrs (%)	**42 °C 96 hrs (%)**	**42 °C 120 hrs (%)**	**43 °C 72 hrs (%)**	**43 °C 96 hrs (%)**	**43 °C 120 hrs (%)**	**44 °C 72 hrs (%)**	**44 °C 96 hrs (%)**	**44 °C 120 hrs (%)**
CNT1	36	13.33 g[1]	1.33 k	0.00 f	29.33 e	11.33 g	4.00 d	0.00 i	0.00 f	0.00 h
CNT1	72	73.33 a-e	65.33 b-e	42.00 a-d	79.33 a-c	77.33 a-c	33.33 b-c	27.33 g-h	4.67 e-f	2.00 g-h
CNT1	71	73.33 a-e	57.33 c-g	28.67 b-e	86.67 a-b	72.66 a-d	54.66 a-c	35.00 f-h	12.67 de	1.33 g-h
CNT1	34	44.67 f	41.33 g-j	20.67 c-e	50.00 d	58.00 c-f	58.67 a-c	56.67 b-g	38.00 a-d	19.33 b-f
CNT1	54	78.00 a-d	73.33 a-c	70.00 a	85.33 a-b	86.67 a-b	48.00 a-c	79.33 a-b	70.00 a	45.33 a-b
CNT1	85	90.00 a-b	57.33 c-g	45.33 a-d	86.00 a-b	80.67 a-c	81.33 a	78.00 a-b	32.67 a-d	20.67 b-f
CNT1	8	81.33 a-c	51.33 c-g	48.00 a-c	90.67 a-b	66.00 a-f	64.00 a-c	59.00 b-f	32.67 b-d	6.00 d-h
CNT1	24	84.00 a-b	82.00 a-b	53.33 a-b	90.67 a-b	90.00 a	84.00 a	95.33 a	65.33 a-b	62.00 a
KDML105	90	79.33 a-d	50.67 d-i	21.33 c-e	80.00 a-c	74.67 a-d	63.33 a-c	42.00 d-h	22.00 c-e	0.00 h
KDML105	106	72.00 b-e	64.00 b-f	54.67 a-b	78.67 a-c	68.00 a-f	56.00 a-c	57.33 b-f	16.67 d-e	14.00 b-g
KDML105	5	65.33 c-e	35.33 h-j	28.00 b-e	64.67 c-d	58.00 c-f	60.00 a-c	67.33 a-e	38.67 a-d	26.00 b-c
KDML105	82	86.67 a-b	57.33 c-g	32.00 b-e	92.00 a	78.00 a-c	79.33 a	76.67 a-c	59.33 a-c	40.67 a-b
KDML105	88	90.67 a	84.00 a	58.00 a-b	83.33 a-c	88.00 a-b	48.00 a-c	56.67 b-g	6.00 e-f	4.00 f-h
KDML105	9	82.67 a-c	68.00 a-d	29.33 b-e	91.33 a	65.33 a-f	64.00 a-c	60.67 b-f	28.00 a-d	17.33 c-g
KDML105	48	62.67 d-e	31.33 j	22.00 c-e	71.33 b-c	47.33 d-f	28.00 c	21.33 h-i	13.33 d-f	4.00 d-h
KDML105	46	85.33 a-b	48.00 e-j	28.00 b-e	79.33 a-c	43.33 e-f	36.67 a-c	52.00 b-g	34.67 a-d	22.67 b-d

Table 4.4 Accelerate aging test of 9 conditions of 24 seed lots of 3 rice varieties, data sorted according to varieties and minimum to maximum percentages of field emergence test of Table 4.3. (continued)

Variety	Lots	Accelerated Aging Condition								
		42 °C 72 hrs (%)	**42 °C 96 hrs (%)**	**42 °C 120 hrs (%)**	**43 °C 72 hrs (%)**	**43 °C 96 hrs (%)**	**43 °C 120 hrs (%)**	**44 °C 72 hrs (%)**	**44 °C 96 hrs (%)**	**44 °C 120 hrs (%)**
PSL 1	130	81.33 a-c	46.67 e-j	30.00 b-e	82.67 a-c	77.33 a-c	72.67 a-b	62.67 b-f	15.33 d-e	8.67 c-h
PSL 1	134	74.00 a-e	41.33 g-j	38.00 a-e	74.67 a-c	63.33 a-f	64.67 a-c	70.67 a-d	40.00 a-d	3.33 d-h
PSL 1	129	84.00 a-b	55.33 c-g	26.00 b-e	80.00 a-c	72.00 a-d	55.33 a-c	50.00 b-h	24.00 c-e	6.00 c-h
PSL 1	131	74.67 a-e	45.33 f-j	18.00 d-e	81.33 a-c	74.67 a-d	50.00 a-c	47.33 c-h	33.33 a-d	13.33 b-g
PSL 1	132	80.00 a-d	32.00 i -j	11.33 e	80.00 a-c	42.00 f	62.00 a-c	62.33 b-f	15.33 d-e	3.33 e-h
PSL 1	133	60.00 e	41.33 g-j	30.67 b-e	64.67 c-d	58.00 c-f	42.00 a-c	58.00 b-f	9.33 d-f	10.00 c-h
PSL 1	135	79.33 a-d	62.00 c-f	12.00 e	77.33 a-c	69.33 a-e	56.00 a-c	39.67 e-h	29.33 b-d	10.67 c-g
PSL 1	128	82.00 a-c	56.67 c-g	39.33 a-d	80.00 a-c	61.33 b-f	45.33 a-c	68.00 a-e	31.33 a-d	20.00 b-e
Mean		**74.08**	**52.03**	**32.78**	**77.47**	**65.97**	**54.64**	**55.14**	**28.03**	**15.03**
F test		**	**	**	**	**	**	**	**	**
C.V. (%)		**12.2**	**19.2**	**24.57**	**12.6**	**21.16**	**22.22**	**27.98**	**35.55**	**50.77**

[1] = Means in the same column that followed by the same letters are not significantly different according to DMRT.

** = Significant difference at $p < 0.01$.

Table 4.5 Correlation coefficients (r) of standard germination, field emergence and accelerated aging test of 9 conditions and other seed vigor tests of 24 seed lots of 3 rice varieties.

Test	SG	RL	SL	TL	SGR	CT	Accelerated Aging Condition									FE
							42°C 72 hrs	42°C 96 hrs	42°C 120 hrs	43°C 72 hrs	43°C 96 hrs	43°C 120 hrs	44°C 72 hrs	44°C 96 hrs	44°C 120 hrs	
SG	1.00	0.88 **	0.93 **	0.92 **	0.73 **	-0.76 **	0.84 **	0.69 **	0.59 **	0.80 **	0.70 **	0.65 **	0.71 **	0.38	0.33	0.64 **
RL		1.00	0.86 **	0.99 **	0.80 **	-0.58 **	0.71 **	0.57 **	0.45 *	0.64 **	0.52 **	0.54 **	0.64 **	0.36	0.26	0.56 **
SL			1.00	0.92 **	0.69 **	-0.82 **	0.77 **	0.77 **	0.66 **	0.74 **	0.71 **	0.67 **	0.74 **	0.46 *	0.47 *	**0.75** **
TL				1.00	0.79 **	-0.66 **	0.75 **	0.64 **	0.53 **	0.69 **	0.59 **	0.60 **	0.69 **	0.40	0.33	0.63 **
SGR					1.00	-0.62 **	0.75 **	0.50 *	0.32	0.61 **	0.59 **	0.61 **	0.65 **	0.45 *	0.26	0.55 **
CT						1.00	-0.68 **	-0.69 **	-0.56 **	-0.68 **	-0.69 **	-0.65 **	**-0.71 ****	-0.48 *	-0.48 *	**-0.82** **
42°C /72 hrs							1.00	0.74 **	0.50 *	0.94 **	0.72 **	0.65 **	0.61 **	0.34	0.26	0.61 **
42°C /96 hrs								1.00	0.74**	0.75 **	0.86 **	0.48 *	0.53 **	0.37	0.44 *	0.66 **
42°C /120 hrs									1.00	0.54 **	0.68 **	0.32	0.60 **	0.40	0.46 *	0.56 **
43°C /72 hrs										1.00	0.74 **	0.64 **	0.56 **	0.37	0.30	0.61 **
43°C /96 hrs											1.00	0.65 **	0.58 **	0.42 *	0.39	0.55 **
43°C /120 hrs												1.00	0.79 **	0.54 **	0.44 *	0.64 **
44°C /72 hrs													1.00	0.74 **	0.71 **	**0.78** **
44°C /96 hrs														1.00	0.86 **	0.62 **
44°C /120 hrs															1.00	0.63 **
FE																1.00

*, ** = significant difference at $p < 0.05$ and $p < 0.01$, respectively.

Table 4.6 Field emergence, accelerate aging test at 44°C for 72 hrs and conductivity of 60 rice seed lots of 10 varieties, data sorted by varieties and minimum to maximum percentages of field emergence test.

No.	Variety	Lot No.	FE (%)	CT (µS $m^{-1}g^{-1}$)	AA (%)
1	CNT1	40	22.66 w-y [1]	46.86 b-d	12.66 z
2	CNT1	38	26.00 v-y	39.29 c-g	12.66 z
3	CNT1	75	35.00 r-x	33.95 e-m	15.33 y-z
4	CNT1	79	41.66 o-v	32.85 e-n	34.00 u-x
5	CNT1	70	52.00 j-r	24.10 j-r	36.66 t-x
6	CNT1	35	52.33 i -r	33.00 e-n	36.00 t-x
7	CNT1	15	53.33 h-q	23.30 k-r	44.00 q-w
8	CNT1	57	58.66 e-o	16.52 r	78.00 a-d
9	CNT1	14	63.67 d-m	19.61 o-r	46.33 p-v
10	CNT1	73	64.33 c-m	16.74 r	70.00 b-h
11	CNT1	53	65.00 c-m	17.00 r	74.66 b-e
12	CNT1	83	66.33 b-l	23.99 j-r	53.33 k-q
13	CNT1	20	67.66 a-k	21.47 m-r	51.00 m-r
14	CNT1	29	72.33 a-g	17.33 q-r	88.00 a
15	CNT1	87	74.33 a-f	28.11 g-r	80.66 a-c
16	CNT1	69	75.66 a-e	16.45 r	83.00 a-b
17	CNT1	84	76.66 a-e	18.42 p-r	65.33 d-l
18	CNT1	86	81.66 a-d	18.48 p-r	82.66 a-b
19	CNT1	33	82.33 a	18.50 p-r	90.66 a
20	CNT1	65	85.00 a	16.83 r	88.66 a
21	KDML105	47	39.33 p-w	36.50 c-j	33.33 u-x
22	KDML105	118	47.33 m-t	41.06 c-f	48.66 n-t
23	KDML105	114	51.33 j-r	39.43 c-g	51.00 l-r
24	KDML105	121	58.33 e-o	28.00 g-r	55.00 i-q
25	KDML105	93	65.66 c-m	28.13 g-r	54.33 i-q
26	KDML105	82	69.66 a-j	21.41 m-r	73.66 b-f
27	KDML105	46	71.33 a-h	21.20 m-r	68.66 c-i
28	KDML105	110	71.66 a-h	23.04 k-r	65.66 d-k
29	KDML105	98	84.33 a-b	24.11 j-r	73.66 b-f
30	PSL1	130	50.00 k-s	25.49 i-r	54.00 j-q
31	PSL1	134	54.00 g-p	22.80 k-r	50.66 m-s
32	PSL1	132	58.33 e-o	25.38 i-r	58.66 g-p
33	PSL1	128	62.00 e-n	22.57 l-r	61.33 e-o
34	PSL1	129	74.33 a-f	25.87 h-r	61.66 e-o
35	SPT	45	40.00 o-w	38.68 c-h	62.00 e-n
36	SPT	44	53.66 g-p	26.83 g-r	56.00 h-q
37	SMP	80	54.00 g-p	27.60 g-r	63.33 e-m
38	SMP	81	54.66 g-p	41.08 c-f	47.33 o-u
39	IR6	50	13.33 y	62.10 a	0.00 z

Table 4.6 Field emergence, accelerate aging test at 44^{o}C for 72 hrs and conductivity of 60 rice seed lots of 10 varieties, data sorted by varieties and minimum to maximum percentages of field emergence test. (continued)

No.	Variety	Lot No.	FE (%)	CT (µS $m^{-1}g^{-1}$)	AA (%)
40	IR6	49	40.66 o-v	42.79 b-f	37.00 s-x
41	IR6	51	49.33 k-s	35.75 c-k	58.00 g-q
42	IR15	136	34.66 r-x	38.99 c-g	0.00 z
43	IR15	139	43.66 n-v	37.59 c-i	37.66 n-v
44	IR15	52	54.33 g-p	20.59 n-r	31.66 w-x
45	IR15	138	54.66 g-p	26.83 g-r	35.00 t-x
46	IR15	137	64.33 c-m	30.23 f-q	52.66 k-q
47	SPR1	144	21.66 x-y	48.23 b-c	0.00 z
48	SPR1	147	28.00 u-y	48.13 b-c	10.00 z
49	SPR1	149	66.66 b-l	27.01 g-r	68.00 c-j
50	SPR1	148	73.66 a-f	21.89 l-r	66.66 d-k
51	SPR2	155	30.00 t-y	54.37 a-b	25.66 x-y
52	SPR2	154	33.00 s-x	44.57 b-e	32.66 v-x
53	SPR2	153	48.00 l-s	34.94 d-l	38.33 r-x
54	SPR2	152	54.00 g-p	25.50 i-r	46.66 p-v
55	SPR2	159	56.00 f-p	27.16 g-r	64.00 d-m
56	SPR2	157	61.66 e-n	30.56 f-p	63.33 e-m
57	SPR2	158	71.00 a-i	25.10 i-r	72.00 b-g
58	SPR3	163	35.33 q-x	42.25 c-f	26.00 x-y
59	SPR3	164	45.00 n-u	37.64 c-i	54.66 i-q
60	SPR3	160	53.33 h-q	32.45 e-o	59.66 f-p
Mean			**55.15**	**29.94**	**51.04**
F test			**	**	**
C.V. (%)			**16.77**	**21.49**	**14.20**

[1] = Means in the same column that followed by the same letters are not significantly difference according to DMRT.

** = Significant difference at p <0.01.

Table 4.7 Field emergence, accelerate aging test at 44°C for 72 hrs and conductivity of 60 rice seed lots of 10 varieties, data sorted by minimum to maximum percentages of field emergence test.

No.	Variety	Lot No.	FE (%)	CT (µS $m^{-1}g^{-1}$)	AA (%)
1	IR6	50	13.33 y	62.10 a	0.00 z
2	IR15	136	34.66 r-x	38.99 c-g	0.00 z
3	SPR1	144	21.66 x-y	48.23 b-c	0.00 z
4	SPR1	147	28.00 u-y	48.13 b-c	10.00 z
5	CNT1	40	22.66 w-y [1]	46.86 b-d	12.66 z
6	CNT1	38	26.00 v-y	39.29 c-g	12.66 z
7	CNT1	75	35.00 r-x	33.95 e-m	15.33 y-z
8	SPR2	155	30.00 t-y	54.37 a-b	25.66 x-y
9	SPR3	163	35.33 q-x	42.25 c-f	26.00 x-y
10	IR15	52	54.33 g-p	20.59 n-r	31.66 w-x
11	SPR2	154	33.00 s-x	44.57 b-e	32.66 v-x
12	KDML105	47	39.33 p-w	36.50 c-j	33.33 u-x
13	CNT1	79	41.66 o-v	32.85 e-n	34.00 u-x
14	IR15	138	54.66 g-p	26.83 g-r	35.00 t-x
15	CNT1	35	52.33 i -r	33.00 e-n	36.00 t-x
16	CNT1	70	52.00 j-r	24.10 j-r	36.66 t-x
17	IR6	49	40.66 o-v	42.79 b-f	37.00 s-x
18	IR15	139	43.66 n-v	37.59 c-i	37.66 n-v
19	SPR2	153	48.00 l-s	34.94 d-l	38.33 r-x
20	CNT1	15	53.33 h-q	23.30 k-r	44.00 q-w
21	CNT1	14	63.67 d-m	19.61 o-r	46.33 p-v
22	SPR2	152	54.00 g-p	25.50 i-r	46.66 p-v
23	SMP	81	54.66 g-p	41.08 c-f	47.33 o-u
24	KDML105	118	47.33 m-t	41.06 c-f	48.66 n-t
25	PSL1	134	54.00 g-p	22.80 k-r	50.66 m-s
26	KDML105	114	51.33 j-r	39.43 c-g	51.00 l-r
27	CNT1	20	67.66 a-k	21.47 m-r	51.00 m-r
28	IR15	137	64.33 c-m	30.23 f-q	52.66 k-q
29	CNT1	83	66.33 b-l	23.99 j-r	53.33 k-q
30	PSL1	130	50.00 k-s	25.49 i-r	54.00 j-q
31	KDML105	93	65.66 c-m	28.13 g-r	54.33 i-q
32	SPR3	164	45.00 n-u	37.64 c-i	54.66 i-q
33	KDML105	121	58.33 e-o	28.00 g-r	55.00 i-q
34	SPT	44	53.66 g-p	26.83 g-r	56.00 h-q
35	IR6	51	49.33 k-s	35.75 c-k	58.00 g-q
36	PSL1	132	58.33 e-o	25.38 i-r	58.66 g-p
37	SPR3	160	53.33 h-q	32.45 e-o	59.66 f-p
38	PSL1	128	62.00 e-n	22.57 l-r	61.33 e-o
39	PSL1	129	74.33 a-f	25.87 h-r	61.66 e-o

Table 4.7 Field emergence, accelerate aging test at 44°C for 72 hrs and conductivity of 60 rice seed lots of 10 varieties, data sorted by minimum to maximum percentages of field emergence test. (continued)

No.	Variety	Lot No.	FE (%)	CT (µS $m^{-1}g^{-1}$)	AA (%)
40	SPT	45	40.00 o-w	38.68 c-h	62.00 e-n
41	SMP	80	54.00 g-p	27.60 g-r	63.33 e-m
42	SPR2	157	61.66 e-n	30.56 f-p	63.33 e-m
43	SPR2	159	56.00 f-p	27.16 g-r	64.00 d-m
44	CNT1	84	76.66 a-e	18.42 p-r	65.33 d-l
45	KDML105	110	71.66 a-h	23.04 k-r	65.66 d-k
46	SPR1	148	73.66 a-f	21.89 l-r	66.66 d-k
47	SPR1	149	66.66 b-l	27.01 g-r	68.00 c-j
48	KDML105	46	71.33 a-h	21.20 m-r	68.66 c-i
49	CNT1	73	64.33 c-m	16.74 r	70.00 b-h
50	SPR2	158	71.00 a-i	25.10 i-r	72.00 b-g
51	KDML105	82	69.66 a-j	21.41 m-r	73.66 b-f
52	KDML105	98	84.33 a-b	24.11 j-r	73.66 b-f
53	CNT1	53	65.00 c-m	17.00 r	74.66 b-e
54	CNT1	57	58.66 e-o	16.52 r	78.00 a-d
55	CNT1	87	74.33 a-f	28.11 g-r	80.66 a-c
56	CNT1	86	81.66 a-d	18.48 p-r	82.66 a-b
57	CNT1	69	75.66 a-e	16.45 r	83.00 a-b
58	CNT1	29	72.33 a-g	17.33 q-r	88.00 a
59	CNT1	65	85.00 a	16.83 r	88.66 a
60	CNT1	33	82.33 a	18.50 p-r	90.66 a
Mean			**55.15**	**29.94**	**51.04**
F test			**	**	**
C.V. (%)			**16.77**	**21.49**	**14.2**

[1] = Means in the same column that followed by the same letters are not significantly difference according to DMRT.

** = Significant difference at $p < 0.01$.

Table 4.8 Correlation coefficients of field emergence, conductivity test and accelerated aging tests at 44ºC for 72 hrs of 60 rice seed lots of 10 varieties.

Test	FE	AA	CT
FE	1.00	0.89**	-0.86**
AA		1.00	-0.77**
CT			1.00

** = Significant different at $p < 0.01$.

CHAPTER V

DISCUSSIONS

5.1 Deficiencies of standard germination test

It has been known for a long time that germination test has a lot of deficiencies on vigor determination. This deficiency was also observed in this experiment. In tables 4.1 and 4.3, mean of standard germination was 88.69% but when the seeds were planted in the field, the mean of field emergence decreased to 64.17%. Some individual seed lots showed very high standard germination percentage but in actual field conditions the seedling emergence reduced drastically showing low seed vigor for example seed lot no. 46 and 48 of KDML105 variety had same standard germination of 88.67% but in actual field condition these seed lots gave different field emergence percentages of 65.33 and 42.00%, respectively. Similarly in CNT1 variety lot no. 72 and 34 had standard germination of 80.67 and 82.00%, respectively but field emergence percentage reduced to 48.67 and 66.00, respectively. The result justify that standard germination can not be used to predict field emergence and standard germination can not be used as seed vigor determination in rice.

Baskin et al. (1993) studied relationship between standard germination test and field emergence of sorghum under favorable and unfavorable field conditions. Standard germination percent of seed lots ranged from 63% to 99% with a mean of 89.5%. Under favorable condition the field emergence percent ranged from 69% to 97% with a mean of 86.5% and highly significant correlation (r = 0.825**) was observed with standard germination test. Under unfavorable field condition (cold wet

soil condition) the mean field emergence percent decreased to 65.9% and low correlation coefficient (r = 0.501**) was observed between standard germination test and field emergence. Similarly in cotton, Bishnoi and Delouche (1980) observed that only vigor tests like accelerated aging test at 42ºC at 144 hrs and cold test which simulated adverse field conditions were effective in predicting field emergence and no significant correlation between standard germination and field emergence was observed.

However, the objectives of standard germination test and vigor tests are in different purposes. Standard germination test is universally accepted and used as seed quality test. The test methodology has been standardized so that test results are reproducible within and among laboratories. The information provided by the test, i.e., the germination percentage of a seed lot, is equally useful as an index of quality in seed trade negotiations, certification, seed control activities and in in-house quality assurance and control programs. The test results establish the maximum plant producing potential of seed lots and correlate quite well with emergence under favorable field conditions. But seldom are these conditions encountered in the field and germination results therefore are always higher than field emergence percentages.

5.2 Accelerated aging test

5.2.1 Designs for the best combinations of accelerated aging conditions

We can conclude from the table 2.2 that accelerated aging condition is basically time against temperature. When the temperature is increased time can be reduced and in the opposite way when the temperature is decreased time must be increased to create the same stress condition. By adjusting these two conditions, seed testing agencies could find a perfect match of time and temperature for accurate

accelerated aging test. However, it is suggested that aging duration should not be too long for faster test results. Very high temperatures will also decrease testing time but create too extreme stress which result in high variation of test results. Another very important aspect of suitable duration of aging the test could be done during convenient time and which would fit in routine working hours. As at the end of aging hours, seeds must be tested for germination immediately. The laboratory officers should have enough time to finish the germination test before the end of working hours.

From the information in table 2.2 it seems that cereal seeds are more tolerance to high stress conditions. Similar result was obtained in this experiment also where aging condition of 44ºC for 72 hrs is suitable and perfect combination which could predict field emergence in rice seed as seed vigor test which could fit in seed laboratory daily work schedule.

5.2.2 Confirmations and verifications of recommendation of accelerated aging condition

Our results showed that accelerated aging test condition at 44ºC for 72 hrs could predict field emergence and could be used as seed vigor test in generally recommended common Thai rice varieties. But before the aging condition is being standardized by seed agencies or rice seed centers, verifications of accelerated aging condition needs to be done especially for new release and specific varieties and different rice types.

Our finding of accelerated aging at 44ºC for 72 hrs as seed vigor test in rice is for generally recommended common Thai varieties but not for specific varieties. In experiment 1, the observations of correlation coefficients between field emergence test and 9 accelerated aging conditions of each variety showed that maximum correlation coefficients between field emergence test and accelerated aging

test were found at 44ºC and 72 hrs (r = 0.93**), 44ºC and 96 hrs (r = 0.88**) and 45ºC and 120 hrs (r = 0.88**) in CNT1, KDML 105 and PSL1, respectively. When the verification of aging at 44ºC and 72 hrs was done in experiment 2 with 10 rice varieties, the correlation coefficient among field emergence test and accelerated aging of some varieties were highly significant but some of them were non-significant with high correlation coefficients while some of them show very low correlation coefficients. The correlation coefficients of field emergence and accelerated aging at 44ºC and 72 hrs for each variety were shown as follow; CNT 1 = 0.91**, KDML105 = 0.93**, PSL1 = -0.15, SPT = -0.51, IR6 = 0.99, IR15 = 0.87, SPR1 = 0.99**, SPL2 = 0.95** and SPL3 = 0.95.

Usually one rice seed center will produce only 2-3 varieties of rice. To be more accurate of accelerated aging results for specific rice varieties, it is recommend that seed centers should verify this accelerated aging condition or conduct experiments for new conditions. In the same manner, the suggested aging condition needs to be verified in new release varieties in the future. For those seed agencies or rice seed centers which deal many rice varieties, verifications of the accelerated aging condition is also recommended.

In order to reduce works and times used in verification processes, rice seed centers can add accelerated aging, conductivity test and field emergence in some routine seed testings and pool data from different rice seed centers for statistical analysis to determine correlation coefficients.

Currently there have been the evidences that specific plant varieties or types need specific accelerated aging variables for the most accurate vigor test results. Komba et al., (2006) observed that aging variables of 41°C for 72 hrs currently suggested as seed vigor test in brassica species was too sever for kale (*B. oleracea* L.

var *acephala* DC) which differentiated seeds into low vigor only and recommended aging conditions of 41°C for 48 hrs as seed vigor test in kale due to its short and suitable time. According to Santipracha et al. (1997), accelerated aging at 44°C for 96 hrs was the best combination of time and temperature for seed vigor test in widely used hybrid corn in humid tropics than the aging conditions of 42°C for 96 hrs and 45°C for 72 hrs for corn seeds as recommended by AOSA (1983) and ISTA (1995), respectively.

5.3 Benefits and limitations of conductivity test

In this research we observed that conductivity test could predict field emergence of rice seed and could be seed as seed vigor in rice.

The integrity of cell membranes, determined by deteriorative biochemical changes and/or physical disruption, can be considered the fundamental cause of differences in seed vigor which are indirectly determined as electrolyte leakage during the conductivity test (AOSA, 1983). The loss in membrane integrity and the leakage of electrolytes are the first symptoms of seed deterioration (McDonald 1999). Thus, the measurement of electrolytes or conductivity test should be most important and effective seed vigor test as it can detect between high and low vigor seed at very early seed deterioration stage.

The conductivity test offers a quick (24 hrs), objective vigor test that can be conducted easily on most seed testing laboratories with minimum expenditure for equipment and training of personnel. Physically injured and mechanical damaged seeds can influence the results. Initial seed moisture is another source of variation in conductivity test results. Treated seeds should be avoided for conductive test.

According to advantages and disadvantages of conductivity test as described

above rice seed analyst should consider using this test in certain cases or even use multiple seed vigor tests to obtain maximum informations on rice seed vigor.

5.4 Urgent need for vigor tests in rice seed

Few researches in rice seed vigor tests have been conducted so far and at present there is no specific recommended accelerated aging test conditions or even other vigor test in rice seeds. The followings are the reports which show the awareness of some researchers who think that having vigor tests in rice seeds is necessary.

Bradford (1988) form Department of Vegetable Crops, University of California evaluated different rice seed lots with high germination percentages through accelerated aging test and compared the result with field emergence. He observed that accelerated aging test could provide additional useful information about seedling vigor and seed germination of 40% after accelerated aging test correlated well with field emergence but not below that.

Patin and Gutormson (2009) at Mid-West Seed Services. Inc. South Dagota, USA, reported that cold and accelerated aging test methods could discriminate between 10 rice samples with low seed vigor and those with high standard germination percentages. From their finding, they suggested that cold and accelerated aging test could be suitable seed vigor test in rice and should be verified with actual field emergence.

Chea (2006) did his studies at Khon Kaen University, Thailand, which performed tests on 2 rice varieties (KDML105 and RD6) to find possible seed vigor tests which could predict field emergence. He evaluated several seed vigor tests of dry and pre-germinating seeds under dry and wet seed bed conditions. He suggested that standard germination test, accelerated aging test (41^{o}C, 84 hrs) seedling growth rate

and conductivity test could be used in predicting field emergence in rice seeds.

The above suggestions support our finding that accelerated aging test could be used as an accurate seed vigor test in rice. However Bradford (1988), and Patin and Gutormson (2009) might not have used specific accelerated aging condition because the condition was not mentioned in their reports. In the near future, it is anticipated that specific recommendations of accelerated aging variables of rice seeds should be widely explored by several researchers.

Currently, no suggested or recommended procedures for conducting rice seed vigor tests are available in the handbooks on vigor testing from International Seed Testing Association (ISTA, 1999) or the Association of Official Seed Analysts (AOSA, 1983). There could be various reasons for not giving awareness in conducting seed vigor tests in rice. Probably rice seeds may have good longevity and store well for one growing season. However, currently there could be an urgent need to have suitable seed vigor tests in rice because of following reasons.

Some recent recommend varieties of rice in Thailand seem to have low seed vigor due to physiological or genetic deformities. Germination percentages of those varieties usually decrease immediately after ripening period (Singkanipa, 2008; personal communication). Seed vigor tests for such rice varieties could provide information to seed agencies for seed quality management.

For old varieties, seed vigor tests are important as high grain quality for export is very important and vigorous seeds can improve rice production to meet high quality standards. Besides the quality of grains the amount of production also increases to meet high demand for export which more amount of seed requirements are needed. Supply of high vigor seeds for export varieties to the farmers will give proper plant populations with uniform seedling emergence, tolerance to adverse environmental

conditions, uniform maturity and finally increasing the grain yields. Therefore, by using high vigor rice seeds, farmers can gain benefits by receiving higher income from higher yield which will minimize input costs.

Today most of the farmers practiced informal seed system (seed villages or self-saved seeds). They use their own seeds every year. The seeds are weak and of low vigor due to no replacement of commercial seeds for many years. This could decrease over all rice production of the country. A proper seed vigor test could prevent this problem and replacement of low vigor seeds with high vigor seeds to them from rice seed centers in every 3-4 years could improve rice production. High amount of rice seed requirement leads to great increase of seed production of rice seed centers and thus may risk in having low seed quality in the system. Therefore, rice seed centers need to have efficient quality control system to ensure seed quality including seed vigor determination.

Climate changes (global warming) also have effect on seed vigor of rice, it creates adverse climatical conditions which could affect rice seed vigor thereby reducing seed longetivity in ambient storage. According to global warming, farmers can not any more expect optimum conditions in all stages of rice culture such as seeding, planting, flowering, harvesting etc. Only high seed vigor can help farmers to overcome the negative effects from climatic changes. Planting rice seed varieties which are tolerance to aging would withstand adverse field conditions providing healthy seedlings and optimum plant populations.

Changes in post-harvest technologies of rice seed production create negative effects in rice seed vigor. Rice seeds are harvested at high moisture content while lack of farmer drying facilities and raw seed drying at seed centers must be well organized. Some seed growers use combines to harvest rice seeds. Harvesting by combines is

done at high seed moisture content. Some time on the combines, seeds are packed in small bags and subsequently the bags are thrown from the harvesters into the fields waiting many hours in the sun to be transported to temporary storage houses. Moreover, farmers do not have drying yards for quick drying so seeds must be directly transported to seed centers for drying. Currently, seed centers have received higher amount of high moisture raw seeds into the processing plants which are more complicate in drying management and increase risk for having low vigor seeds.

5.5 Impact of test results on seed development program in Thailand

Currently, no standardized rice seed vigor tests is available in Thailand. Therefore, the findings of both accurate vigor tests; accelerated aging test at 44ºC for 72 hrs and conductivity test, recommended for rice varieties in Thailand could have following useful impacts on seed development program in Thailand.

5.5.1 Decisions making for seed management and marketing

Seed companies can use vigor informations in their quality assurance program as a marketing strategy. Seed companies can win the confidence of the client and gain good image in seed market with the supply of good quality and high vigor seeds.

Seed company management must make many technical decisions in the course of business operations. High vigor rice seeds can be stored well for a long time. One of the important decisions that must be made each season involves the determination of which seed lots among those held in inventory should be marketed first and which can be safely held for carryover in the event of a weak market or to insure an adequate seed supply the following year (Delouche and Baskin, 1973). Seed companies can make decision on timely sale of stock seeds as per seed vigor levels.

Low vigor seeds can be sold first in the market. Seed blending of different vigor levels is considered as another alternative for seed companies to avoid inventory losses. Low vigor seeds can be blended with high vigor seeds (within the minimum germination percentages required for individual crop) and supply in the market for sale. Seed manager can also decide to withhold low vigor seeds from processing for further quality determinations and consideration for blending or even consider upgrading among low vigor seeds on the basis of seed weight by using gravity separator. Low vigor seed could also be supplied to farmers along with seed treatments like fungicides as preventative measures from fungal attack and diseases, so that seeds are still produce optimum seedlings.

Seed lot vigor information is also important for sowing seeds in differential environmental conditions (ISTA, 1995), for example in harsh environmental conditions (rainfed areas, early growing season which may experience drought, broadcasting technique is used in seed sowing) etc., a good seed quality with high vigor seed lots should be supplied as they can withstand those stress conditions resulting into rapid, uniform and high percentage of seedling emergence which would serve well in giving good yield. In case of good environmental conditions (transplanted rice areas, irrigated areas, good soil and good climate) low vigor seed lots can still be used and produce satisfactory population of seedlings.

Seed storage managers can use seed vigor information in their decision making such as rice seed lots can be arranged as per vigor levels in the store for better inventory management. Management can also decide on disposal of low vigor rice seeds by various programs like sales promotion, sale in subsidized rate or use in farmer’s demonstration plots.

5.5.2 Seed purchasing

When vigor of rice seeds is accurately distinguished, vigor levels could be used as a pricing criteria in seed purchasing policy. That could be a win-win situation for seed companies. In one hand farmers are happy to get good price, they get more encouraged to produce high vigor seeds and more farmers may come up to take seed production scheme. In the other hand, seed companies can increase selling price of high vigor seeds in the market.

5.5.3 Seed storage management

Adequate provisions for storage of seeds are a common feature of successful seed production marketing programmes regardless of their geographical location. Seeds in storage represent not only a programme or company's potential return on substantial investments in research and development, production, facilities, operations and promotion, but also an input vital for continued agricultural production. Accelerated aging test has been successfully used in predicting storage potential of different crop species under wide range of storage conditions (Delouche and Baskin, 1973). Abba and Lovota (1999) observed accelerated aging test at 42ºC for 96 hrs could predict corn longetivity for one year under storage condition at 20ºC 45% R.H.. Similar results were obtained by Basu et al. (2004) which stated that accelerated aging test condition at 40ºC for 168 hrs could predict storage potential of corn for 8 months under ambient storage condition. Santipracha et al. (1993) observed accelerated aging at 44ºC for 96 hrs could be used for predicting storage potential of corn seeds packed in paper bags for at least one year. Research in accelerated aging test for rice seed storability should be explored.

5.6 Rice varieties screening for plant breeding program

Humid tropics climates (with high temperatures and high relative humidity) are conducive to rapid seed storability deterioration. The degree of tolerance for accelerated aging conditions has been related to the survivability of the seeds in storage (Delouche and Baskin, 1973). Accelerated aging conditions could help breeders determine differences in a variety's potential to resist seed deterioration during storage. Siddique et al. (1988) used accelerated aging test to screen different rice varieties tolerance to aging and developed a rapid screening test that can evaluate rice seeds tolerance for adverse environmental conditions. In their studies they evaluated 185 rice varieties (116 indicas, 39 japonicas, and 30 javanicas) which were subjected to accelerated aging test at 43ºC for 2, 4, and 6 days. They observed that varieties with superior tolerance for the aging conditions were found among each eco-geographic races of *Oryza sativa* L. (indica, japonica, and javanica), photoperiod-sensitive and insensitive strains, upland and lowland types, salt tolerant, and tungro virus-resistant varieties. Their results indicated that rice varieties differ significantly in their ability to maintain seed viability through artificial aging.

From our finding, plant breeders in Thailand should experiment the use of accelerated aging test condition at 44ºC for 72 hrs to screen out Thai rice varieties for aging tolerance varieties or lines which could help in developing improved varieties with the potential for maintaining longer and stronger seed viability in storage.

CHAPTER VI

CONCLUSION AND RECOMMENDATION

6.1 Conclusions

1) The accelerated aging test condition at 44ºC for 72 hrs was accurate and suitable rice seed vigor test which showed highly significant correlation with field emergence.

2) Conductivity test can be used as substitute or alternative seed vigor test in rice as it also showed highly significant correlation with field emergence and accelerated aging test at 44ºC for 72 hrs.

3) Seedling shoot length could also be a good rice seed vigor test as it also significantly correlated with field emergence but it is not recommended as it is very time consuming method.

6.2 Recommendations

1) Accelerated aging conditions at 44ºC and 72 hrs is recommended as vigor test in rice seeds for common Thai cultivars. However, following points need to be taken into consideration before the aging condition is standardized.

1.1) Seed agency needs to verify this recommended accelerated aging test condition for general Thai rice varieties.

1.2) For the most accurate aging test conditions for specific varieties, new release cultivars or eco-geographic races of rice, the accelerated aging test at 44ºC for 72 hrs needs to be verified and re-tested.

2) The conductivity test can be used as substitute to accelerated aging test or alternative test for rice seed vigor test.

3) Future researches need to be done on suitability of accelerated aging test and conductivity test to predict storage potential of rice seeds.

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APPENDIX

Appendix Table 1 Standard germination, field emergence and accelerated aging test of 9 conditions and other seed vigor tests of 24 seed lots of 3 rice varieties, data sorted according to varieties and minimum to maximum percentages of standard germination test.

Variety	Lots	Vigor Test							Accelerated Aging Condition								
		SG (%)	FE (%)	RL (cm)	SL (cm)	TL (cm)	SGR (mg/plant)	CT ($\mu S\ cm^{-1}g^{-1}$)	42 °C 72 hrs (%)	42 °C 96 hrs (%)	42 °C 120 hrs (%)	43 °C 72 hrs (%)	43 °C 96 hrs (%)	43 °C 120 hrs (%)	44 °C 72 hrs (%)	44 °C 96 hrs (%)	44 °C 120 hrs (%)
CNT1	36	44.00 h[1]	31.33 e	2.84 h	0.79 g	3.63 i	0.16	81.98 a	13.33 g	1.33 k	0.00 f	29.33 e	11.33 g	4.00 d	0.00 i	0.00 f	0.00 h
CNT1	72	80.67 g	48.67 c-e	8.49 g	3.67 e-f	12.16 g-h	4.467	31.36 d-g	73.33 a-e	65.33 b-e	42.00 a-d	79.33 a-c	77.33 a-c	33.33 b-c	27.33 g-h	4.67 e-f	2.00 g-h
CNT1	34	82.00 f-g	66.00 a-e	9.29 f-g	3.95 a-f	13.23 f-h	4.125	28.25 e-g	44.67 f	41.33 g-j	20.67 c-e	50.00 d	58.00 c-f	58.67 a-c	56.67 b-g	38.00 a-d	19.33 b-f
CNT1	71	83.33 e-g	61.33 a-e	8.53 g	3.36 f	11.89 h	3.096	33.34 d-e	73.33 a-e	57.33 c-g	28.67 b-e	86.67 a-b	72.66 a-d	54.66 a-c	35.00 f-h	12.67 d-e	1.33 g-h
CNT1	85	90.67 b-e	79.33 a-c	9.62 f-g	4.31 a-f	13.93 e-h	5.436	27.76 e-g	90.00 a-b	57.33 c-g	45.33 a-d	86.00 a-b	80.67 a-c	81.33 a	78.00 a-b	32.67 a-d	20.67 b-f
CNT1	54	93.33 a-d	76.00 a-d	11.19 d-f	4.44 a-e	15.64 c-f	5.257	26.76 e-g	78.00 a-d	73.33 a-c	70.00 a	85.33 a-b	86.67 a-b	48.00 a-c	79.33 a-b	70.00 a	45.33 a-b
CNT1	8	94.00 a-d	85.33 a-b	10.65 d-g	4.44 a-e	15.09 c-g	5.004	23.57 g	81.33 a-c	51.33 c-g	48.00 a-c	90.67 a-b	66.00 a-f	64.00 a-c	59.00 b-f	32.67 b-d	6.00 d-h
CNT1	24	94.67 a-d	86.67 a	11.02 d-f	4.78 a-d	15.81 b-f	4.43	23.12 g	84.00 a-b	82.00 a-b	53.33 a-b	90.67 a-b	90.00 a	84.00 a	95.33 a	65.33 a-b	62.00 a
KDML	48	88.67 c-g	42.00 d-e	10.94 d-f	3.60 e-f	14.54 d-h	3.246	34.15 d-e	62.67 d-e	31.33 j	22.00 c-e	71.33 b-c	47.33 d-f	28.00 c	21.33 h-i	13.33 3d-f	4.00 d-h
KDML	46	88.67 c-g	65.33 a-e	10.79 d-f	4.09 c-f	14.88 c-h	5.166	32.97 d-f	85.33 a-b	48.00 e-j	28.00 b-e	79.33 a-c	43.33 e-f	36.67 a-c	52.00 b-g	34.67 a-d	22.67 b-d
KDML	90	89.33 c-f	49.33 b-e	10.24 e-g	3.92 d-f	14.16 e-h	4.908	35.15 d-e	79.33 a-d	50.67 d-i	21.33 c-e	80.00 a-c	74.67 a-d	63.33 a-c	42.00 d-h	22.00 c-e	0.00 h
KDML	5	90.67 b-e	72.00 a-d	11.99 b-e	4.43 a-e	16.42 b-e	5.759	24.56 f-g	65.33 c-e	35.33 h-j	28.00 b-e	64.67 c-d	58.00 c-f	60.00 a-c	67.33 a-e	38.67 a-d	26.00 b-c
KDML	82	94.67 a-d	76.67 a-d	12.80 a-d	4.95 a-c	17.75 a-c	6.582	27.54 e-g	86.67 a-b	57.33 c-g	32.00 b-e	92.00 a	78.00 a-c	79.33 a	76.67 a-c	59.33 a-c	40.67 a-b
KDML	106	96.00 a-c	54.67 a-e	12.38 a-e	4.94 a-c	17.32 a-d	4.366	44.18 b-c	72.00 b-e	64.00 b-f	54.67 a-b	78.67 a-c	68.00 a-f	56.00 a-c	57.33 b-f	16.67 d-e	14.00 b-g
KDML	9	98.67 a-b	84.00 a-c	14.28 a	5.23 a	19.51 a	4.96	24.87 f-g	82.67 a-c	68.00 a-d	29.33 b-e	91.33 a	65.33 a-f	64.00 a-c	60.67 b-f	28.00 a-d	17.33 c-g
KDML	88	99.33 a	80.00 a-c	13.55 a-c	5.11 a-b	18.66 a-b	5.933	24.83 f-g	90.67 a	84.00 a	58.00 a-b	83.33 a-c	88.00 a-b	48.00 a-c	56.67 b-g	6.00 e-f	4.00 f-h
PSL1	135	83.33 e-g	64.67 a-e	11.40 c-f	3.65 e-f	15.05 c-g	6.645	44.10 b-c	79.33 a-d	62.00 c-f	12.00 e	77.33 a-c	69.33 a-e	56.00 a-c	39.67 e-h	29.33 b-d	10.67 c-g
PSL1	132	87.33 d-g	60.67 a-e	10.34 e-g	3.51 e-f	13.86 e-h	4.92	37.28 c-d	80.00 a-d	32.00 i -j	11.33 e	80.00 a-c	42.00 f	62.00 a-c	62.33 b-f	15.33 d-e	3.33 e-h
PSL1	134	88.00 c-g	56.00 a-e	13.65 a-b	3.75 e-f	17.41 a-d	6.555	48.04 b	74.00 a-e	41.33 g-j	38.00 a-e	74.67 a-c	63.33 a-f	64.67 a-c	70.67 a-d	40.00 a-d	3.33 d-h
PSL1	131	89.33 c-f	60.00 a-e	10.44 e-g	3.61 e-f	14.06 e-h	6.08	38.09 c-d	74.67 a-e	45.33 f-j	18.00 d-e	81.33 a-c	74.67 a-d	50.00 a-c	47.33 c-h	33.33 a-d	13.33 b-g
PSL1	133	91.33 a-e	60.67 a-e	11.11 d-f	3.72 e-f	14.83 c-h	4.924	36.80 c-d	60.00 e	41.33 g-j	30.67 b-e	64.67 c-d	58.00 c-f	42.00 a-c	58.00 b-f	9.33 d-f	10.00 c-h
PSL1	129	92.67 a-d	57.33 a-e	10.33 e-g	3.85 d-f	14.18 e-h	4.539	47.52 b	84.00 a-b	55.33 c-g	26.00 b-e	80.00 a-c	72.00 a-d	55.33 a-c	50.00 b-h	24.00 c-e	6.00 c-h
PSL1	130	94.00 a-d	52.00 a-e	11.37 c-f	4.13 b-f	15.51 c-f	5.348	35.16 d-e	81.33 a-c	46.67 e-j	30.00 b-e	82.67 a-c	77.33 a-c	72.67 a-b	62.67 b-f	15.33 d-e	8.67 c-h
PSL1	128	94.00 a-d	70.00 a-d	12.01 b-e	4.24 a-f	16.25 b-f	5.593	33.54 d-e	82.00 a-c	56.67 c-g	39.33 a-d	80.00 a-c	61.33 b-f	45.33 a-c	68.00 a-e	31.33 a-d	20.00 b-e
Mean		**88.69**	**64.17**	**10.8**	**4.02**	**14.82**	**4.895**	**35.21**	**74.08**	**52.03**	**32.78**	**77.47**	**65.97**	**54.64**	**55.14**	**28.03**	**15.03**
F test		**	*	**	**	**	ns	**	**	**	**	**	**	**	**	**	**
C.V. (%)		**4.87**	**28.25**	**10.74**	**12.9**	**10.66**	**38.7**	**12.48**	**12.2**	**19.2**	**24.57** [1]	**12.6**	**21.16**	**22.22** [2]	**27.98**	**35.55** [3]	**50.77** [4]

[1] = Means in the same column that followed by the same letters are not significantly different according to DMRT.
*, **, ns = Significant difference at $p < 0.05$, $p < 0.01$ and non-significant, respectively.

Appendix Table 2 Standard germination, field emergence and accelerated aging test of 9 conditions and other seed vigor tests of 24 seed lots of 3 rice varieties, data sorted according to varieties and minimum to maximum percentages of field emergence test.

Variety	Lots	Vigor Test							Accelerated Aging Condition								
		SG (%)	FE (%)	RL (cm)	SL (cm)	TL (cm)	SGR (mg/plant)	CT ($\mu S\ cm^{-1}g^{-1}$)	42 °C 72 hrs (%)	42 °C 96 hrs (%)	42 °C 120 hrs (%)	43 °C 72 hrs (%)	43 °C 96 hrs (%)	43 °C 120 hrs (%)	44 °C 72 hrs (%)	44 °C 96 hrs (%)	44 °C 120 hrs (%)
CNT1	36	44.00 h[1]	31.33 e	2.84 h	0.79 g	3.63 i	0.16	81.98 a	13.33 g	1.33 k	0.00 f	29.33 e	11.33 g	4.00 d	0.00 i	0.00 f	0.00 h
CNT1	72	80.67 g	48.67 c-e	8.49 g	3.67 e-f	12.16 g-h	4.467	31.36 d-g	73.33 a-e	65.33 b-e	42.00 a-d	79.33 a-c	77.33 a-c	33.33 b-c	27.33 g-h	4.67 e-f	2.00 g-h
CNT1	71	83.33 e-g	61.33 a-e	8.53 g	3.36 f	11.89 h	3.096	33.34 d-e	73.33 a-e	57.33 c-g	28.67 b-e	86.67 a-b	72.66 a-d	54.66 a-c	35.00 f-h	12.67 d-e	1.33 g-h
CNT1	34	82.00 f-g	66.00 a-e	9.29 f-g	3.95 a-f	13.23 f-h	4.125	28.25 e-g	44.67 f	41.33 g-j	20.67 c-e	50.00 d	58.00 c-f	58.67 a-c	56.67 b-g	38.00 a-d	19.33 b-f
CNT1	54	93.33 a-d	76.00 a-d	11.19 d-f	4.44 a-e	15.64 c-f	5.257	26.76 e-g	78.00 a-d	73.33 a-c	70.00 a	85.33 a-b	86.67 a-b	48.00 a-c	79.33 a-b	70.00 a	45.33 a-b
CNT1	85	90.67 b-e	79.33 a-c	9.62 f-g	4.31 a-f	13.93 e-h	5.436	27.76 e-g	90.00 a-b	57.33 c-g	45.33 a-d	86.00 a-b	80.67 a-c	81.33 a	78.00 a-b	32.67 a-d	20.67 b-f
CNT1	8	94.00 a-d	85.33 a-b	10.65 d-g	4.44 a-e	15.09 c-g	5.004	23.57 g	81.33 a-c	51.33 c-g	48.00 a-c	90.67 a-b	66.00 a-f	64.00 a-c	59.00 b-f	32.67 b-d	6.00 d-h
CNT1	24	94.67 a-d	86.67 a	11.02 d-f	4.78 a-d	15.81 b-f	4.43	23.12 g	84.00 a-b	82.00 a-b	53.33 a-b	90.67 a-b	90.00 a	84.00 a	95.33 a	65.33 a-b	62.00 a
KDML	48	88.67 c-g	42.00 d-e	10.94 d-f	3.60 e-f	14.54 d-h	3.246	34.15 d-e	62.67 d-e	31.33 j	22.00 c-e	71.33 b-c	47.33 d-f	28.00 c	21.33 h-i	13.33 3d-f	4.00 d-h
KDML	90	89.33 c-f	49.33 b-e	10.24 e-g	3.92 d-f	14.16 e-h	4.908	35.15 d-e	79.33 a-d	50.67 d-i	21.33 c-e	80.00 a-c	74.67 a-d	63.33 a-c	42.00 d-h	22.00 c-e	0.00 h
KDML	106	96.00 a-c	54.67 a-e	12.38 a-e	4.94 a-c	17.32 a-d	4.366	44.18 b-c	72.00 b-e	64.00 b-f	54.67 a-b	78.67 a-c	68.00 a-f	56.00 a-c	57.33 b-f	16.67 d-e	14.00 b-g
KDML	46	88.67 c-g	65.33 a-e	10.79 d-f	4.09 c-f	14.88 c-h	5.166	32.97 d-f	85.33 a-b	48.00 e-j	28.00 b-e	79.33 a-c	43.33 e-f	36.67 a-c	52.00 b-g	34.67 a-d	22.67 b-d
KDML	5	90.67 b-e	72.00 a-d	11.99 b-e	4.43 a-e	16.42 b-e	5.759	24.56 f-g	65.33 c-e	35.33 h-j	28.00 b-e	64.67 c-d	58.00 c-f	60.00 a-c	67.33 a-e	38.67 a-d	26.00 b-c
KDML	82	94.67 a-d	76.67 a-d	12.80 a-d	4.95 a-c	17.75 a-c	6.582	27.54 e-g	86.67 a-b	57.33 c-g	32.00 b-e	92.00 a	78.00 a-c	79.33 a	76.67 a-c	59.33 a-c	40.67 a-b
KDML	88	99.33 a	80.00 a-c	13.55 a-c	5.11 a-b	18.66 a-b	5.933	24.83 f-g	90.67 a	84.00 a	58.00 a-b	83.33 a-c	88.00 a-b	48.00 a-c	56.67 b-g	6.00 e-f	4.00 f-h
KDML	9	98.67 a-b	84.00 a-c	14.28 a	5.23 a	19.51 a	4.96	24.87 f-g	82.67 a-c	68.00 a-d	29.33 b-e	91.33 a	65.33 a-f	64.00 a-c	60.67 b-f	28.00 a-d	17.33 c-g
PSL1	130	94.00 a-d	52.00 a-e	11.37 c-f	4.13 b-f	15.51 c-f	5.348	35.16 d-e	81.33 a-c	46.67 e-j	30.00 b-e	82.67 a-c	77.33 a-c	72.67 a-b	62.67 b-f	15.33 d-e	8.67 c-h
PSL1	134	88.00 c-g	56.00 a-e	13.65 a-b	3.75 e-f	17.41 a-d	6.555	48.04 b	74.00 a-e	41.33 g-j	38.00 a-e	74.67 a-c	63.33 a-f	64.67 a-c	70.67 a-d	40.00 a-d	3.33 d-h
PSL1	129	92.67 a-d	57.33 a-e	10.33 e-g	3.85 d-f	14.18 e-h	4.539	47.52 b	84.00 a-b	55.33 c-g	26.00 b-e	80.00 a-c	72.00 a-d	55.33 a-c	50.00 b-h	24.00 c-e	6.00 c-h
PSL1	132	87.33 d-g	60.67 a-e	10.34 e-g	3.51 e-f	13.86 e-h	4.92	37.28 c-d	80.00 a-d	32.00 i-j	11.33 e	80.00 a-c	42.00 f	62.00 a-c	62.33 b-f	15.33 d-e	3.33 e-h
PSL1	133	91.33 a-e	60.67 a-e	11.11 d-f	3.72 e-f	14.83 c-h	4.924	36.80 c-d	60.00 e	41.33 g-j	30.67 b-e	64.67 c-d	58.00 c-f	42.00 a-c	58.00 b-f	9.33 d-f	10.00 c-h
PSL1	135	83.33 e-g	64.67 a-e	11.40 c-f	3.65 e-f	15.05 c-g	6.645	44.10 b-c	79.33 a-d	62.00 c-f	12.00 e	77.33 a-c	69.33 a-e	56.00 a-c	39.67 e-h	29.33 b-d	10.67 c-g
PSL1	128	94.00 a-d	70.00 a-d	12.01 b-e	4.24 a-f	16.25 b-f	5.593	33.54 d-e	82.00 a-c	56.67 c-g	39.33 a-d	80.00 a-c	61.33 b-f	45.33 a-c	68.00 a-e	31.33 a-d	20.00 b-e
PSL1	131	89.33 c-f	60.00 a-e	10.44 e-g	3.61 e-f	14.06 e-h	6.08	38.09 c-d	74.67 a-e	45.33 f-j	18.00 d-e	81.33 a-c	74.67 a-d	50.00 a-c	47.33 c-h	33.33 a-d	13.33 b-g
Mean		**88.69**	**64.17**	**10.8**	**4.02**	**14.82**	**4.895**	**35.21**	**74.08**	**52.03**	**32.78**	**77.47**	**65.97**	**54.64**	**55.14**	**28.03**	**15.03**
F test		**	*	**	**	**	ns	**	**	**	**	**	**	**	**	**	**
C.V. (%)		**4.87**	**28.25**	**10.74**	**12.9**	**10.66**	**38.7**	**12.48**	**12.2**	**19.2**	**24.57**[1]	**12.6**	**21.16**	**22.22**[2]	**27.98**	**35.55**[3]	**50.77**[4]

[1] = Means in the same column that followed by the same letters are not significantly different according to DMRT.
*, **, ns = Significant difference at $p < 0.05$, $p < 0.01$ and non-significant, respectively.

Appendix Table 3 Mean squares of standard germination, field emergence and other seed vigor tests of 24 seed lots of 3 rice varieties.

Source of variation	df	SG	FE	RL	SL	TL	SGR	CT
Treatment	23	344.52**	607.33*	15.21**	2.29**	27.63**	5.59 ns	462.98**
Error	48	18.61	328.61	1.35	0.27	2.50	3.59	19.33
Total	71	36.13	935.94	15.56	2.56	30.13	9.18	482.31
C.V. (%)		4.87	28.25	10.74	12.90	10.66	38.57	12.48

*, **, ns = Significant difference at $p < 0.05$, $p < 0.01$ and non-significant, respectively.

Appendix Table 4 Mean squares of accelerate aging test of 9 conditions of 24 seed lots of 3 rice varieties.

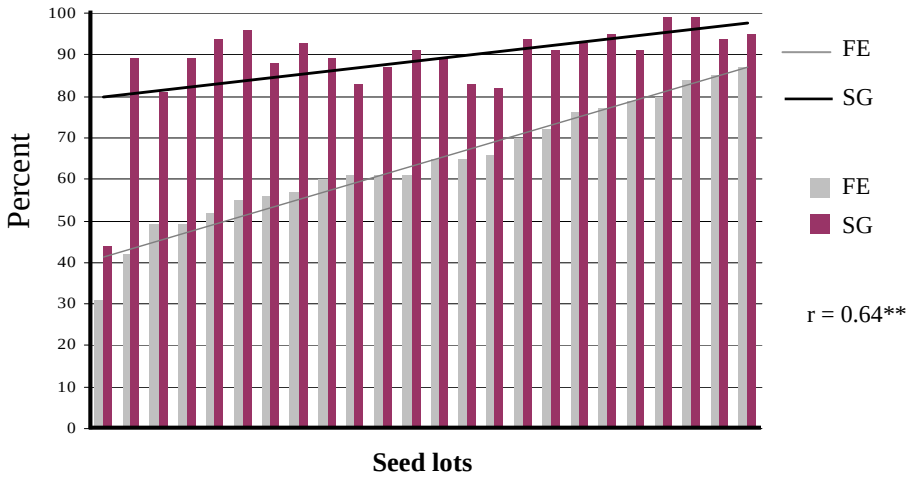
Source of Variation	df	Accelerate Aging Condition								
		42 °C 72 hrs	42 °C 96 hrs	42 °C 120 hrs	43 °C 72 hrs	43 °C 96 hrs	43 °C 120 hrs	44 °C 72 hrs	44 °C 96 hrs	44 °C 120 hrs
Treatment	23	828.85**	949.50**	805.99**	590.37**	912.64**	961.45**	1,276.29	1,009.45**	732.87**
Error	48	81.67	99.44	227.89	94.78	194.78	397.94	238.00	258.22	136.33
Total	71	906.85	1,048.94	1,033.88	685.15	1,107.42	1,359.39	1,514.29	1,267.67	869.20
C.V. (%)		12.20	19.20	24.57	12.60	21.16	22.22	27.98	35.55	50.77

** = Significant difference at $p < 0.01$.

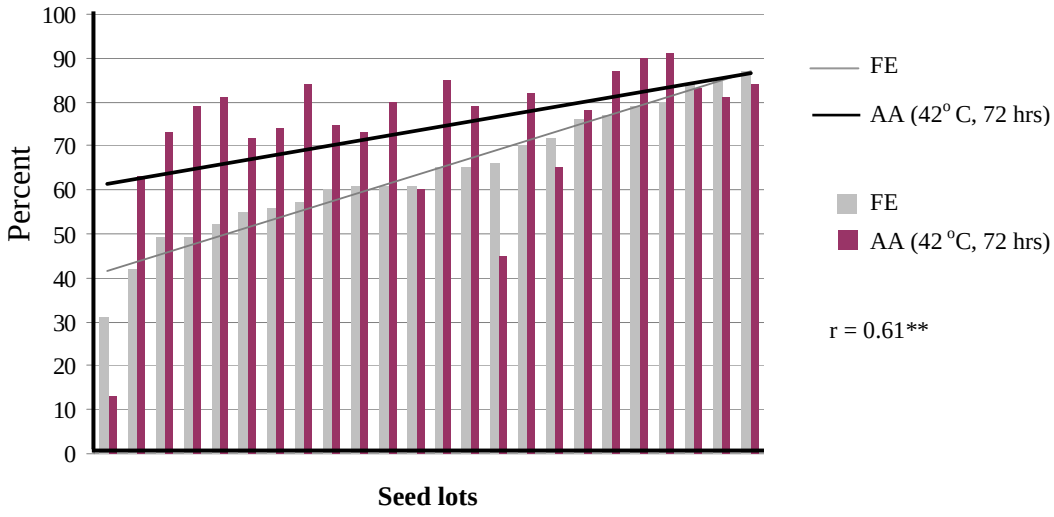
Appendix Table 5 Mean squares of accelerate aging, standard germination, field emergence and accelerate aging at 44 ^{o}C for 72 hrs of 60 rice seed lots of 10 rice varieties.

Source of variation	**df**	**FE**	**CT**	**AA**
Treatment	23	837.03**	321.16**	1,542.52**
Error	48	85.58	41.42	52.68
Total	71	922.61	362.58	1,595.20
C.V. (%)		16.77	21.49	14.20

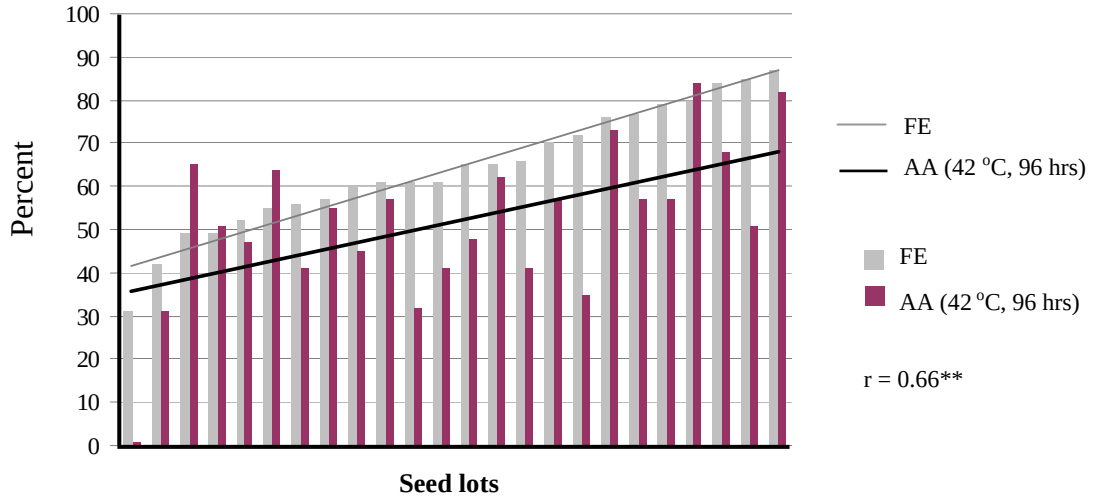
** = Significant difference at $p < 0.01$.



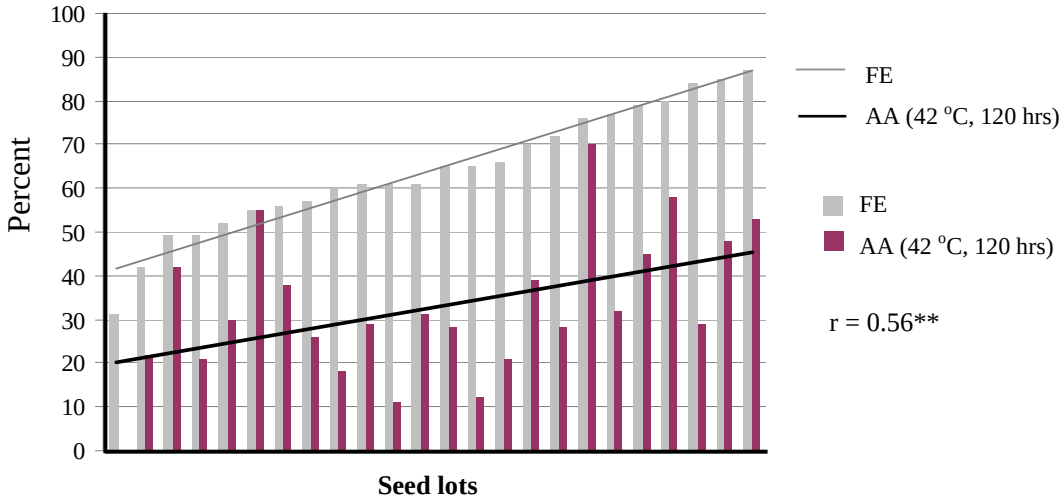
Appendix Figure 1 Relationship between standard germination (SG) and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



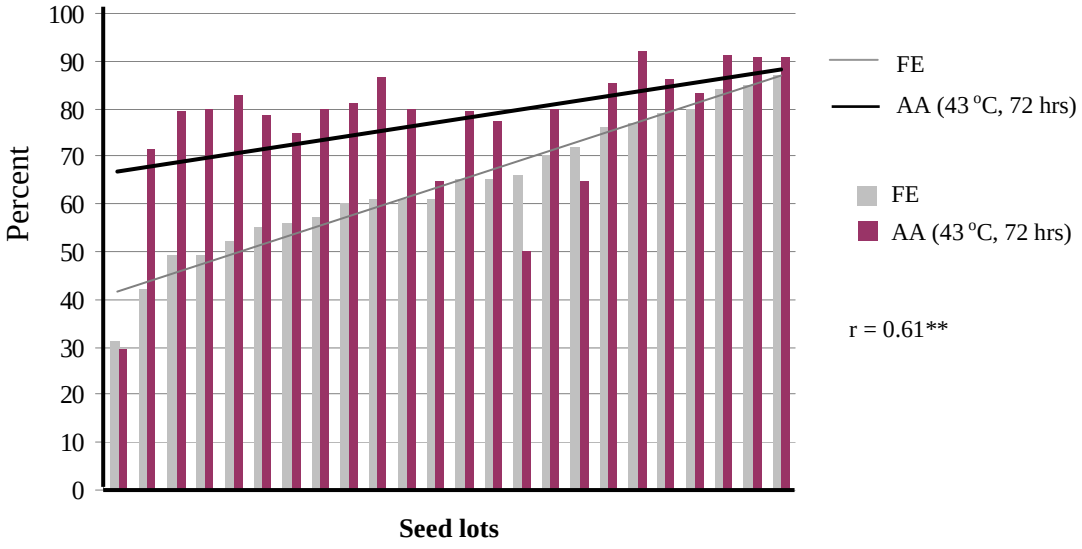
Appendix Figure 2 Relationship between accelerate aging (AA) at 42° C for 72 hrs and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



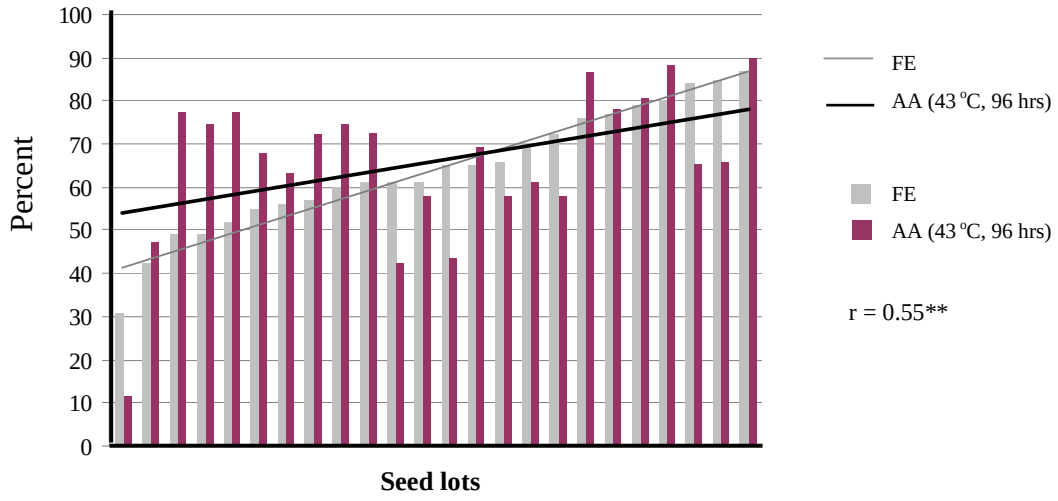
Appendix Figure 3 Relationship between accelerate aging (AA) at 42° C for 96 hrs and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



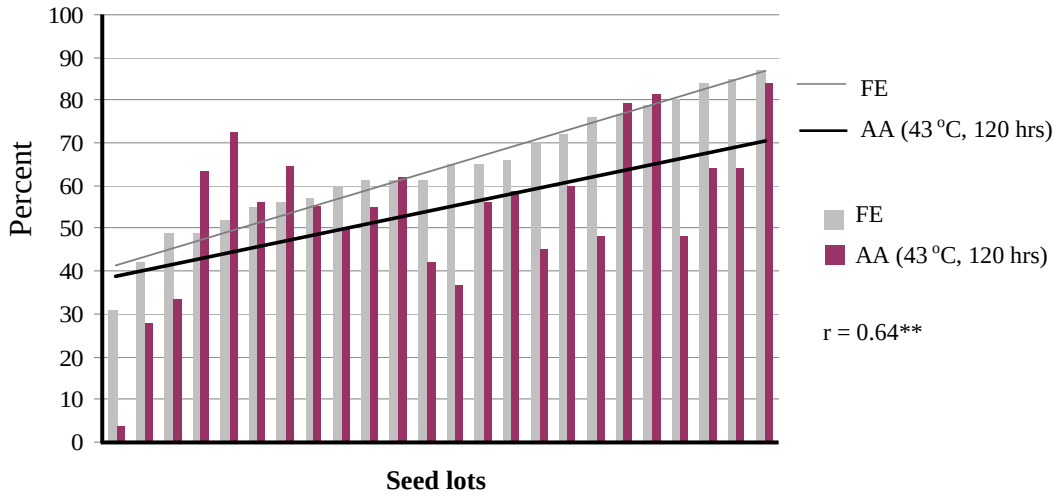
Appendix Figure 4 Relationship between accelerate aging (AA) at 42° C for 120 hrs and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



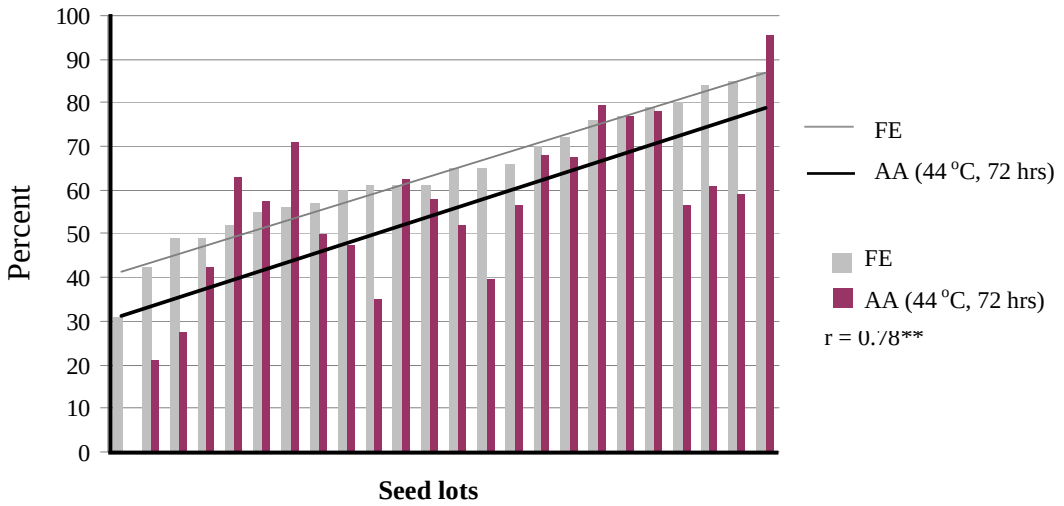
Appendix Figure 5 Relationship between accelerate aging (AA) at 43° C for 72 hrs and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



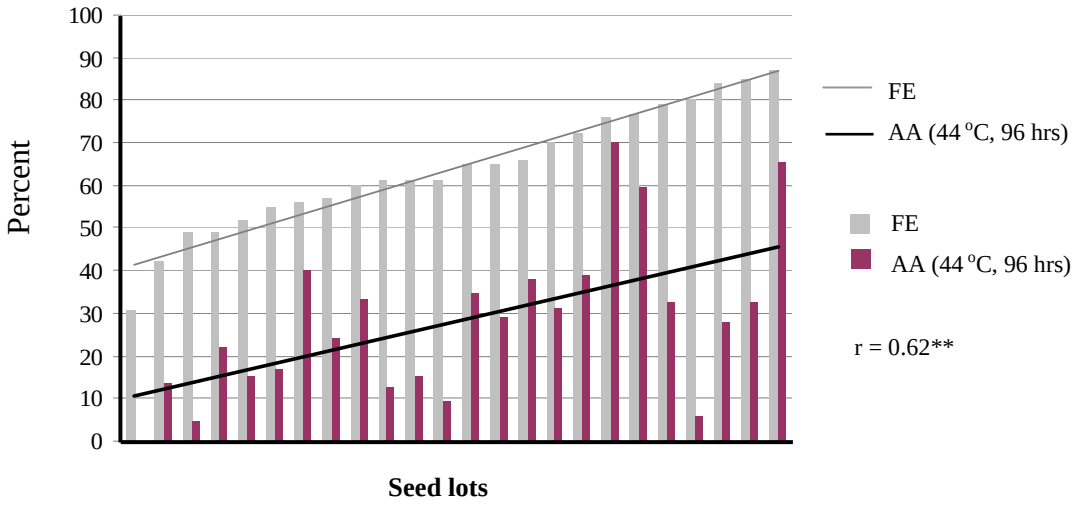
Appendix Figure 6 Relationship between accelerate aging (AA) at 43° C for 96 hrs and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



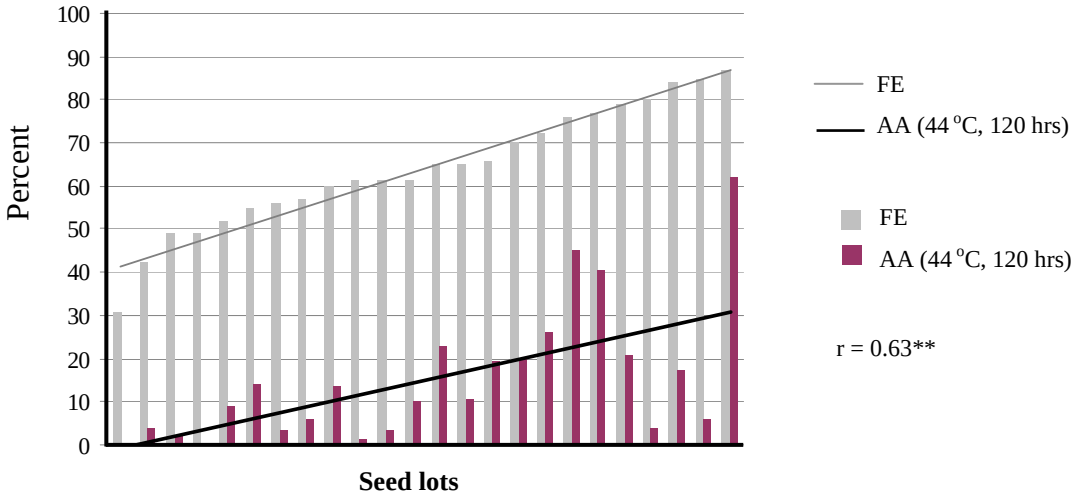
Appendix Figure 7 Relationship between accelerate aging (AA) at 43° C for 120 hrs and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



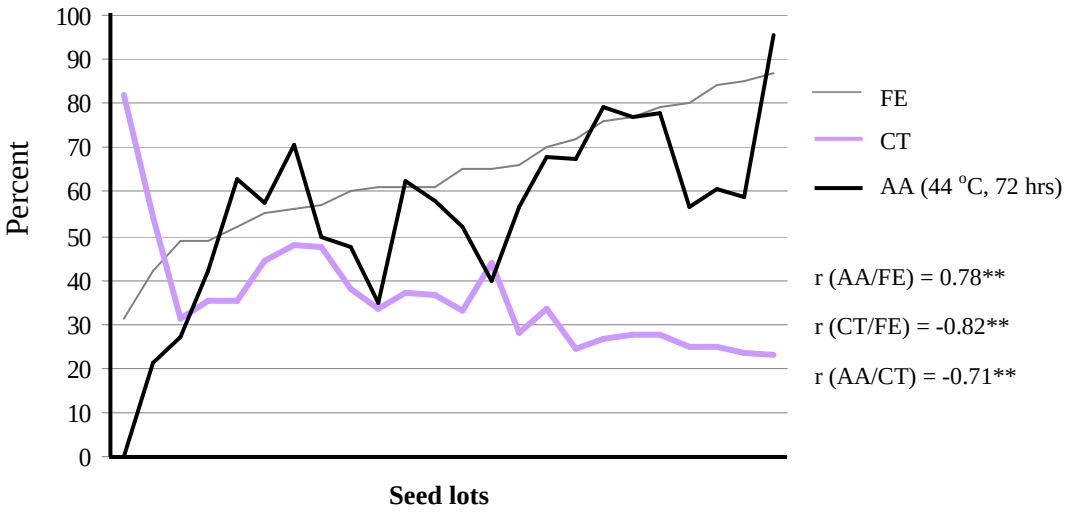
Appendix Figure 8 Relationship between accelerate aging (AA) at 44° C for 72 hrs and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



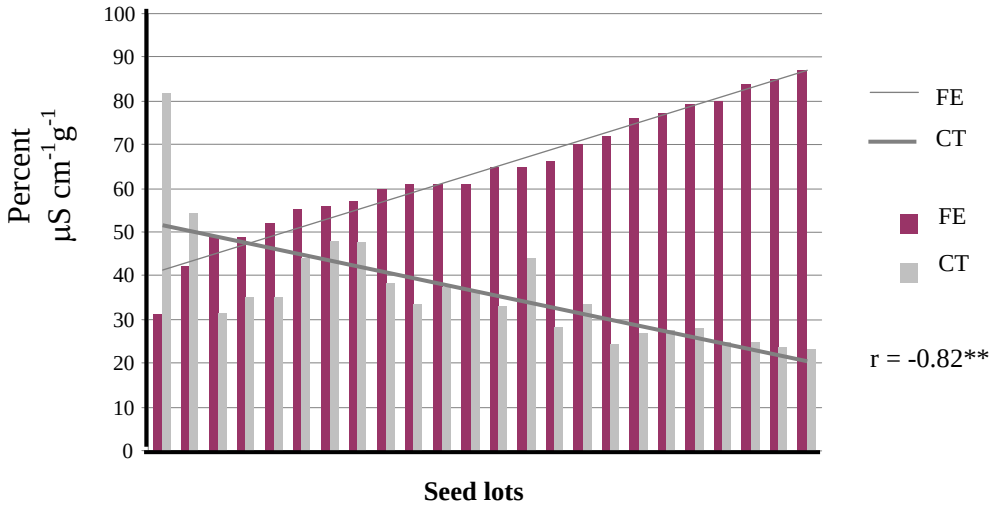
Appendix Figure 9 Relationship between accelerate aging (AA) at 44° C for 96 hrs and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



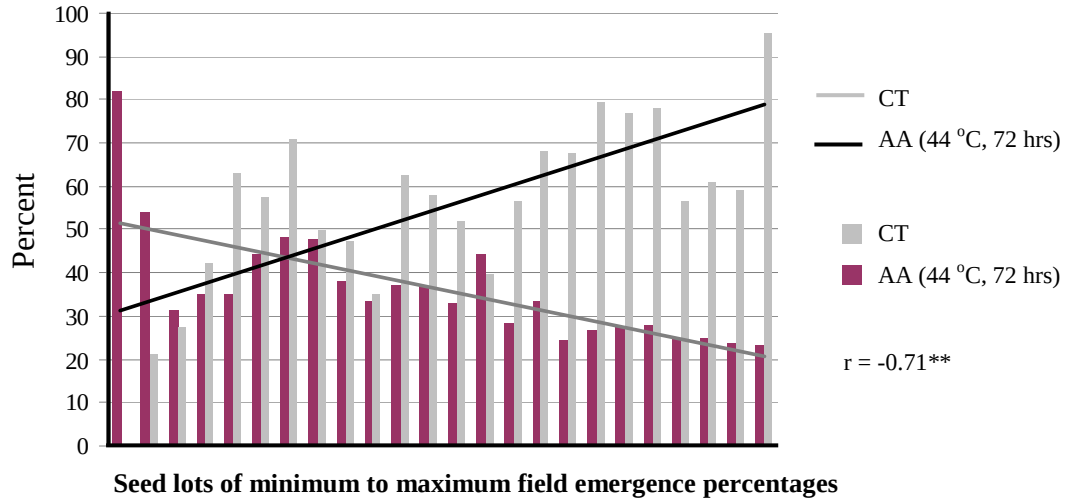
Appendix Figure 10 Relationship between accelerate aging (AA) at 44° C for 120 hrs and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages.



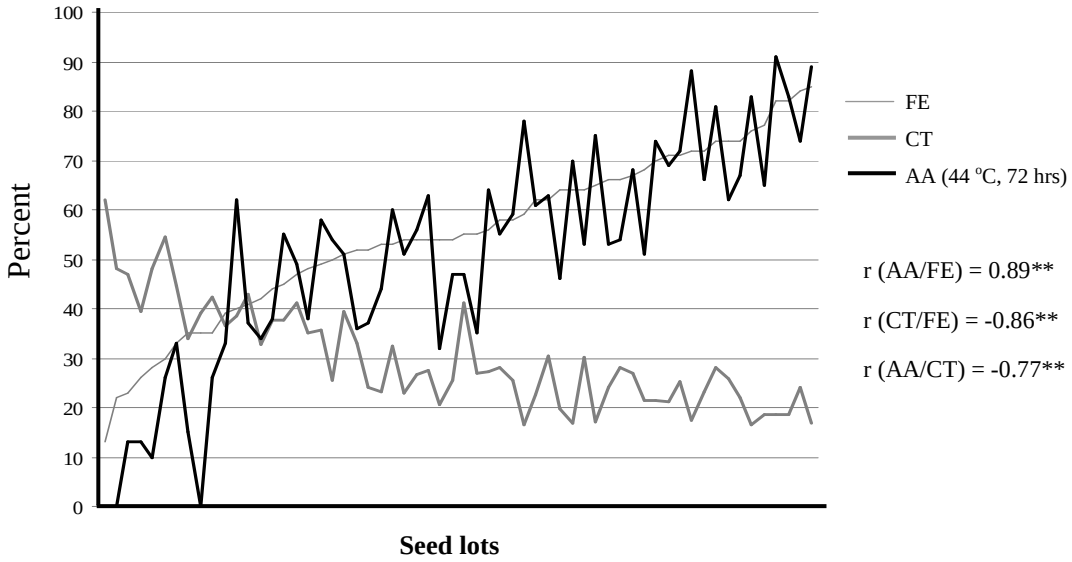
Appendix Figure 11 Relationship among accelerate aging (AA) at 44^{o}C for 72 hrs, conductivity (CT) and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages.



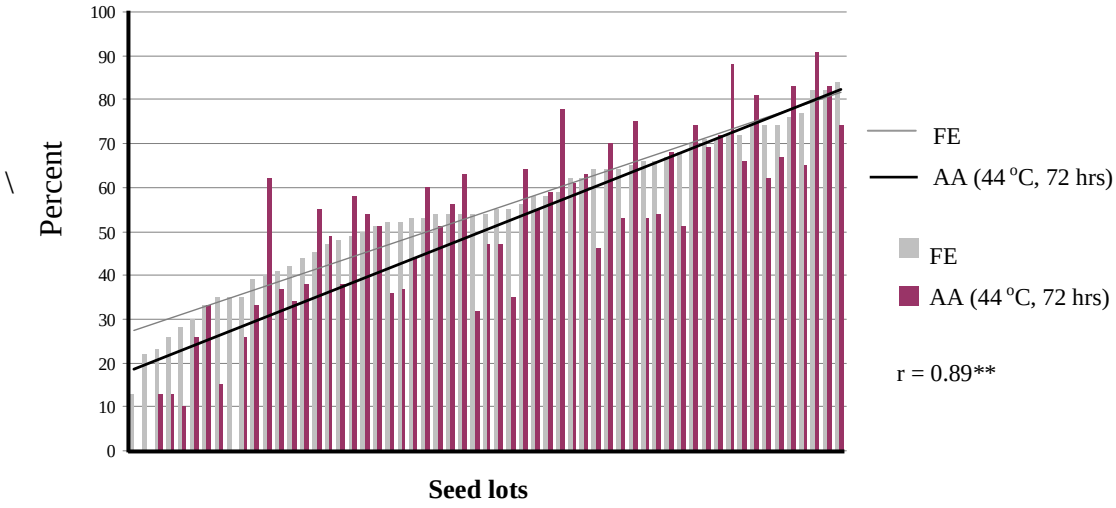
Appendix Figure 12 Relationship between conductivity test (CT) and field emergence (FE) of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



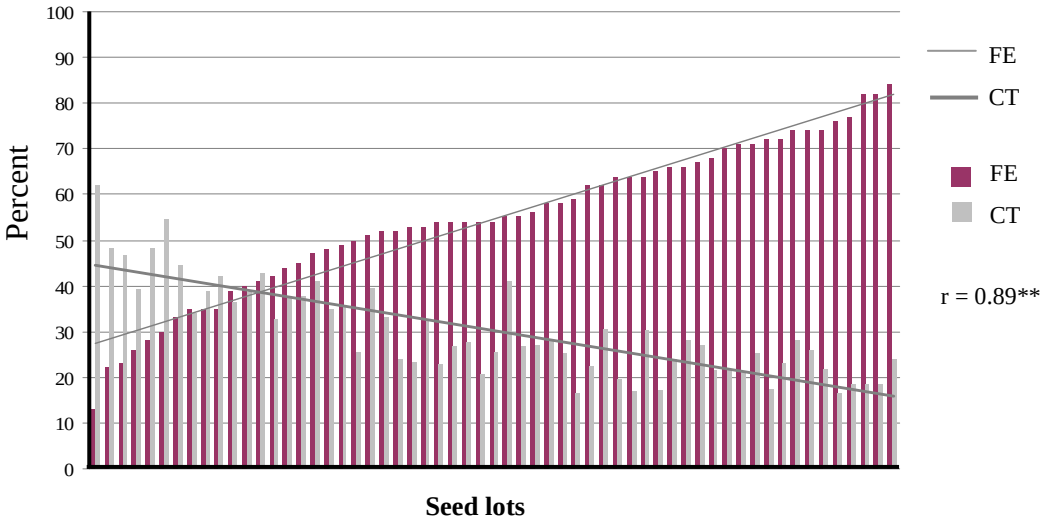
Appendix Figure 13 Relationship between conductivity test (CT) and accelerate aging (AA) at 44°C 72 hrs of 24 seed lots of 3 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



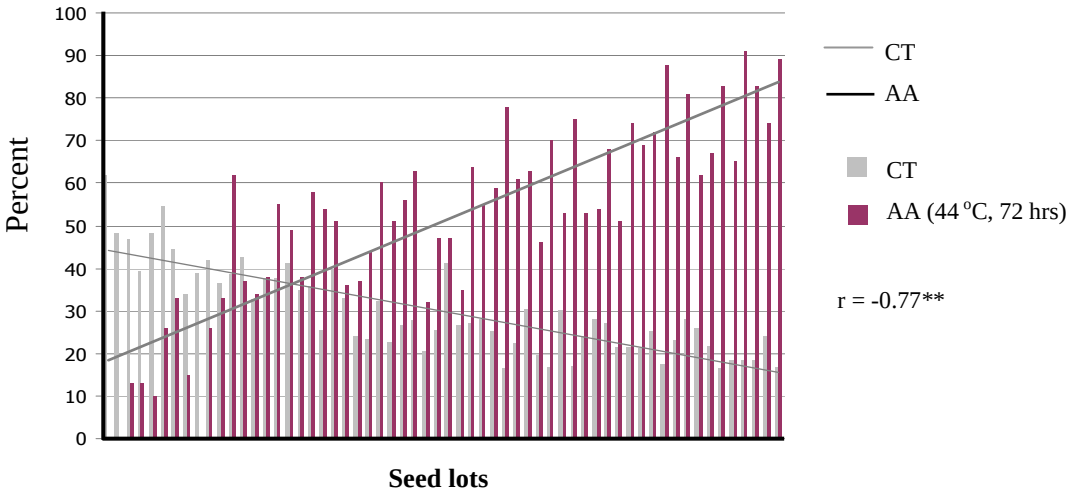
Appendix Figure 14 Relationship among accelerate aging (AA) at 44°C for 72 hrs, conductivity (CT) and field emergence (FE) of 60 seed lots of 10 rice varieties arranged from minimum to maximum field emergence.



Appendix Figure 15 Relationship between accelerate aging (AA) at 44°C for 72 hrs and field emergence (FE) of 60 seed lots of 10 rice varieties arranged from minimum to maximum field emergence percentages.



Appendix Figure 16 Relationship between conductivity (CT) at 44°C for 72 hrs and field emergence (FE) of 60 seed lots of 10 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.



Appendix Figure 17 Relationship between conductivity (CT) at 44^{o}C 72 hrs and accelerate aging (AA) of 60 seed lots of 10 rice varieties arranged from minimum to maximum field emergence percentages as shown by histogram and linear graphs.

BIOGRAPHY

Mr. Suraj Chhetri was born on January 17th 1974 in Samtse province, Bhutan. He received Bachelor degree in Agriculture in 1999 from College of Agriculture, Maharashtra, India. After graduation he was employed in Druk Seed Corporation (Government Undertaking), under Ministry of Agriculture.

Since 1999, he has successfully served Druk Seed Corporation in various capacities. Till 2003, he served as regional manager responsible for temperate vegetable seed production and marketing. In 2004, he worked as general manager RSG (Registered Seed Grower) scheme, coordinating seed production all over the country through registered grower's seed village scheme. He also worked as general sale manager and prior to his study; he was working as field crop seed coordinator responsible for seed production and marketing of field crops.