

**THE POTENTIAL OF PRIMING AS A METHOD TO IMPROVE THE
QUALITY OF AEROBIC RICE SEED**

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ABSTRACT

Seed quality has a significant potential of increasing on-farm productivity which can be promote by priming. An experiment was conducted to study the effect of primed aerobic rice seeds on seed quality prior to storage and after storage of one month. This study was arranged in a Completely Randomized Design (CRD), with 4 replicates for each treatment. Aerobic rice seeds was treated with distilled water for 12 hour, distilled water for 24 hour, 1% potassium nitrate for 12 hour, 1% potassium nitrate for 24 hour, 2% potassium nitrate for 12 hour, 2% potassium nitrate for 24, 1% sodium chloride for 12 hour, 1% sodium chloride for 24 hour, 2% sodium chloride for 12 hour, 2% sodium chloride for 24 hour and unprimed seed act as control. After primed, the seed was dried back to its original moisture content and germination test was conducted. The remaining seed was stored in refrigerator at 2°C and 40% relative humidity for one month. After one month, the unprimed and primed aerobic rice seeds were once again subjected to germination test. The germination test was conducted for 14 days. The parameters evaluated in this study included final germination percentage, percentage of normal seeding, percentage of abnormal seedling, , plumule length, radicle length, seedling vigor index, seedling fresh weight as well as seedling dry weight. The results revealed that aerobic rice seeds treated with salt solution enhance the seed performance. Potassium nitrate gave highest final germination percentage, percentage of normal seedling, radicle length and seed vigor index meanwhile sodium chloride improve the rate of germination, plumule length, seedling fresh weight and reducing the percentage of abnormal seedling. After storage of one month, potassium nitrate gave highest result in all parameters except seedling fresh weight and seeding dry weight. This showed that potassium nitrate have excelled over all other priming agents including sodium chloride. Thus, priming the seed with potassium nitrate enhanced the seed quality and also after storage of one month.



POTENSI PEMPRIMAAN SEBAGAI KAEDAH UNTUK MENKKAJI PENYIMPANAN BIJI BENIH PADI AEROB

ABSTRAK

Kuliti biji benih memainkan peranan yang penting dalam meningkatkan produktiviti tanaman dengan menggunakan pemprimaan. Satu kajian telah dijalankan untuk mengkaji kesan pemprimaan terhadap biji benih aerob sebelum penyimpanan dan selepas penyimpanan selama satu bulan. Kajian ini disusun dalam Reka Bentuk Rawak Lengkap, dengan 4 replikasi bagi setiap rawatan. Biji benih aerob dirawat dengan air suling selama 12 jam, air suling selama 24 jam, 1% kalium nitrat selama 12 jam, 1% kalium nitrat selama 24 jam, 2% kalium nitrat selama 12 jam, 2% kalium nitrat selama 24 jam, 1% natrium klorida selama 12 jam, 1% natrium klorida selama 24 jam, 2% natrium klorida selama 12 jam, 2% natrium klorida selama 24 jam dan biji benih yang tidak dirawat sebagai kawalan. Selepas dirawat, biji benih dikeringkan sehingga ia mencapai kandungan kelembapan yang asal dan ujian percambahan dilakukan. Manakala lebih biji benih disimpan di dalam peti sejuk bersuhu 2 °C dan 40 % kandungan kelembapan. Sebulan kemudian, percambahan semua rawatan diuji semua. Ujian percambahan dilakukan selama 12 hari. Parameter yang dinilai dalam kajian ini termasuk peratusan percambahan, peratusan biji benih normal, peratusan biji benih tidak normal, purata masa percambahan, panjang plumul, panjang radikel, indeks kecergasan benih, berat basah anak benih berserta berat kering anak benih. Kajian menunjukkan pemprimaan biji benih aerob menggunakan solusi rendaman meningkatkan prestasi biji benih. Kalium nitrate memberikan keputusan tertinggi dalam peratusan percambahan, peratusan biji benih normal, panjang radikel dan indeks kecergasan benih manakala natrium klorida meningkatkan kadar percambahan, berat basah anak benih dan mengurangkan peratusan biji benih tidak normal. Selepas penyimpanan selama satu bulan, kalium nitrat memberikan keputusan tertinggi dalam semua parameter kecuali berat basah anak benih dan berat kering anak benih. Hal ini menunjukkan kalium nitrat cemerlang daripada semua pemprimaan termasuk natrium klorida. Oleh itu, pemprimaan biji benih dengan kalium nitrat meningkatkan kualiti benih dan selepas penyimpanan satu bulan.

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LIST OF SYMBOLS, UNITS AND ABBREVIATIONS

°C	Degree of Celcius
%	Percentage
ANNOVA	Analysis of Variance
cm	Centimeter
FSA	Faculty of Sustainable Agriculture
IRRI	International Rice Research Institute
ISTA	International Seed Testing Association
KNO ₃	Potassium Nitrate
LSD	Least Significant Differences
MARDI	Malaysia Agricultural Research and Development Institute
NaCl	Sodium Chloride
No	Number
SAS	Statistical Analysis System
T	Treatment



CHAPTER 1

INTRODUCTION

1.1 Background

Rice is the staple food for most of the world's population, with Asia being the largest producer and consumer (Gumma *et al.*, 2011). Nowadays, the population of Asia is rapidly increasing than its ability to produce sufficient rice to meet the forthcoming demand (Papademetriou, 2000). Even in Malaysia, the self-sufficiency level was only 72% (Ahmad Zubir Ibrahim, 2012). In response to the 2008 crisis, the Malaysian government aimed at increasing food production and productivity to achieve self-sufficiency by providing adequate incentives and income to producers. This can be attained with some options such as identifying new areas of management, use of locally high yielding varieties and the use of good quality seeds for planting. (Mohd Dandan, 2016). Freitas *et al.* (2004) also emphasized that a successful farming is primarily depends on the use of high quality seeds.

Seeds are foundation of agriculture where it plays an important role in the production of agronomic and horticultural crops. The use of good quality seed helps greatly in higher production per unit area to attain food security of the country. Characteristics such as trueness to variety, germination percentage, purity, vigor, uniform establishment and appearance are important to farmers. However, seed are vary in quality which may due to genetic make-up, seed size, growing condition during plant establishment, age or maturity of seed and storage condition, etc. This variation may cause the crop grow erratically. When crop does not grow uniformly, it limits potential for mechanized harvesting as the maturity of rice was not evenly distributed. The timing and uniformity of field crop of field crop seeding emergence also alter their competitive advantage with weeds. This has an immediate impact upon the efficacy of herbicide applications, weeding strategies and other aspects of crop production that determined cost-effectiveness and impact on the environment. Hence, one of the



methods to increase the uniformity of crop establishment is seed priming. Seed priming is defined as seed enhancement in which seed immersed in water or low osmotic potential solution to trigger the germination metabolic activities but insufficient to allow radical emergence (Bradford, 1986; Bray, 1995; Farood *et al*, 2012). Various seed priming techniques have been developed, including hydro-priming (soaking in water), halo-priming (soaking in inorganic salt solutions), osmo-priming (soaking in solutions of different organic osmotica), thermo-priming (treatment of seeds with low or high temperatures), solid matrix priming (treatment of seed with solid matrices) and bio-priming (hydration using biological compounds).

1.2 Justification

Many researches have shown an overall advantages in increasing the quality of seed where it provide better germination percentage, uniform seedling emergence and high vigor (Parera and Cantliffe, 1994; Abdulrahmani *et al.*, 2007; Ghassemi-Golezani *et al.*, 2008). Earlier works showed that the success of seed priming is influenced by the complex interaction of factors including plant species, water potentiality of priming agent, duration of priming, temperature, seed vigour and storage conditions of the primed seeds (Parera and Cantliffe, 1994). A lot of work has been done on seed priming and results of these studies indicate well the importance of priming to get a good crop stand in many crops of tropical region such as rice, maize, sorghum and pigeon pea. Considering the above points, the effects of seed storing after seed priming on germination behaviours is still scanty. According to Mehdi and Mokthar (2012), storing of primed seed about 30-60 days improved germination characteristics compared to fresh primed seed. However, some of research shows poor performance of primed seed stored. A negative correlation between germination performances after storage was found by Liu *et al.* (1996). Furthermore, Nascimento (2002), reported decreases in germination and vigor of conditioned seeds after 12 months of storage.

1.3 Objective

The objective of this study has to determine the effect of priming method on the quality of aerobic rice seeds prior to storage and after storage of one month.

1.4 Hypothesis

H_0 : There was no significant difference in the effect of priming method on seed quality of aerobic rice seeds prior to storage and after storage of one month

H_A : There was significant difference in the effect of priming method on seed quality of aerobic rice seeds prior to storage and after storage of one month

CHAPTER 2

LITERATURE REVIEW

2.1 Rice

2.1.1 Taxonomy

Cultivated rice comprises *Oryza sativa* and grass *Oryza glaberrima*. These plants are indigenous to Tropical and Subtropical Southern Asia and South Eastern Africa respectively (Linares, 2002). Out of the two cultivated species, *Oryza sativa* is most widely grown (Office of the Gene Technology Regulator, 2005) where it feeds more than half of the world's population. The taxonomy of *Oryza sativa* is shown in Table 2.1.

Table 2.1 Taxonomy of *Oryza sativa*

Kingdom	Plantae
Division	Magnoliophyta
Class	Liliopsida
Order	Cyperales
Family	Poaceae
Genus	<i>Oryza</i>
Species	<i>sativa</i>

Sources: Adapted from Office of the Gene Technology Regulator, 2005



2.1.2 Rice production in Malaysia

In the year 2020 with the increase of 2% per year in the population growth, Malaysia will have population of 32.37 million. It is reported that Malaysia rice consumption is 85.5kg per person per year (Sariam and Badrulhadza, 2016), at an average of two and half plates of rice per day (Norimah, 2008). With increasing population and consumption, the gap between domestic supply and demand need to be narrowed with imported rice (Yogambigai *et al.*, 2015). Malaysia had not reached self-sufficiency level in which a total of 876,100 t of rice (34% of our country's rice necessity) was imported from countries such as Vietnam and Thailand (Sariam and Badrulhadza, 2016). The target for the country's sufficiency level of rice often is currently at a minimum level of 65%. National Agro-Food Policy (2011-2015) targeted the national rice sufficiency level will be more than 70% and that stage will be monitored from time to time.

Other than the wetland rice, the rice production can be executed in other area such as wasteland and other suitable land area where paddy can be planted including area without standing water throughout the whole growing stage like the paddy field. Paddy that does not need standing water or ponding is known as Aerobic Rice. MARDI plans to introduce aerobic rice cultivation over 1,000 hectares nationwide (Borneo Post, 2014).

2.1.3 Aerobic Rice

Aerobic rice is a newly introduced cultivation system in Malaysia that requires less water production system as compared to wetland rice. Moreover, it aimed to maximize crop productivity by growing plants in aerobic system without or less flooding (Sariam and Badrulhadza, 2016). So, Based on Bouman (2005), aerobic rice holds promise for farmers that have access to limited water sources to grow flooded lowland rice as it uses less water in land preparation, less evaporation and less transpiration than flooded fields. Besides, the efficiency of water usage can be increased up to 20%.

2.2 Rice Seed Structure

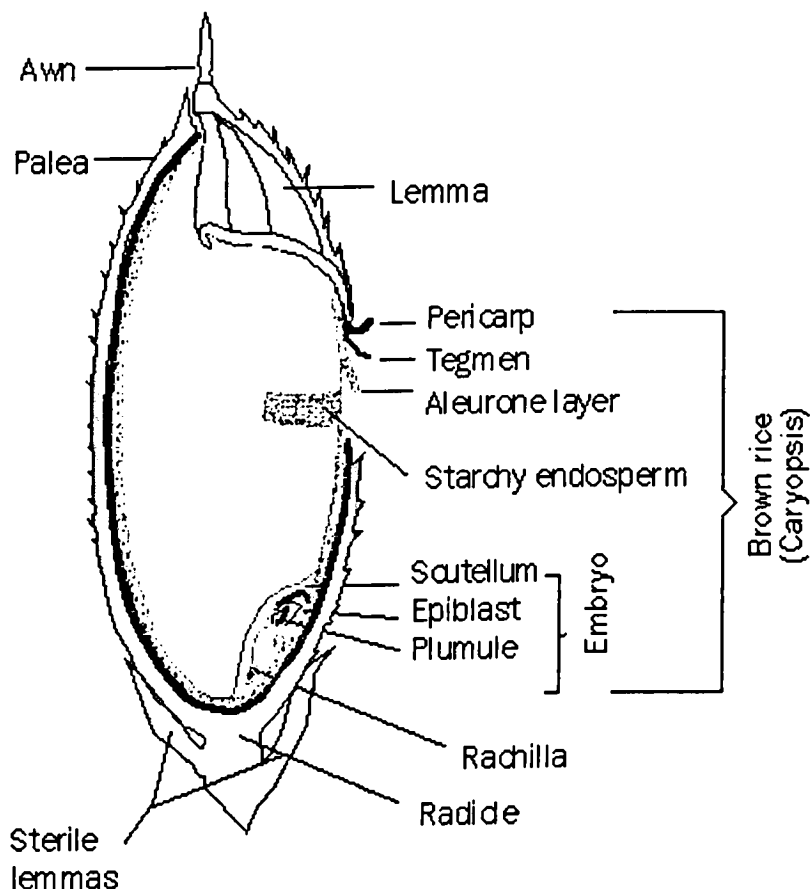


Figure 2.1 The structure of the rice seed

Source: Bienvenido, 1993

Seed is a complex structure that develops from an ovule and. Maturity a seed has an embryo, variable amounts of endosperm and protective layers of tissue on the surface of the seed, all enclosed by the seed coat or testa derived from the integuments. Rice seed or paddy seed comprises of a husk as the outer protective part and the edible grain on the inside part. Rice caryopsis also called brown rice, consists of pericarp, germ or embryo, endosperm, tegmen and aleurone layer (Bienvenido, 1993). Li (2003) stated that the embryo contains plumule, radicle and mesocotyl. Aleurone layer encloses the endosperm which lies beneath the tegmen (Chang, 1965). Size of rice seed is classified as with extra-long (>7.50 mm), long (6.61 mm – 7.50 mm), medium (5.51 mm – 6.60 mm) and short (4.50 mm) according to Bienvenido (1993).

2.3 Seed Quality

Good quality seed has a significant potential of increasing on-farm productivity and enhancing food security. Good rice seed quality comprises lower seed rate, better emergence (>70%), more uniformity, less replanting, vigorous and free from insects and diseases (IRRI, 2016). Recently, many techniques have been used to optimize seedling establishment under the changing environments. These techniques performed on seeds after harvest but prior to sowing that are generally described in the industry and scientific literature as 'seed enhancements' or 'seed treatments' (Irfan *et al.*, 2016). Seed enhancement is defined as treatments that improve germination, seedling growth or facilitate the delivery of seeds and other material prior to sowing. In other words, it is a value-added technique on a given lot of seed. The purpose of these treatments is to shorten the time between planting and emergence. There are three general methods which are priming, seed coating technologies and integration of these methods to enhance seed quality (Iqbal *et al.*, 2014).

2.4 Germination

Germination is one of the most important and the first stages of plant growth which can determine the quality of a seed lot (Richman *et al.*, 2006). Several definitions have been offered to explain seed germination. ISTA defined germination as the active growth of the embryo which results in the rupture of the seed coat (ISTA, 2006). Some people defined it as the emergence of radicle through the seed coat. Besides, germination also can be defined as the measure on how well a seed produces normal and healthy seedlings under favourable conditions for a specific period (Richman *et al.*, 2006). Measurement of germination can provide valuable information about the start, rate, uniformity and final percentage of germination. It is reported that results from germination test can be used to compare the quality of the seed (ISTA, 2007). The germination percentage and rate vary among and within individuals and populations of a species and for seeds in different stage of dormancy break (Baskin and Baskin, 2014). Germination percentage expresses the proportion of the total number of seeds that are alive which is determined through controlled tests and actual counts of the number of seeds that germinate. The minimum acceptable standard for germination percentage of rice seed is 70% (IRRI, 2016).

According to Richman *et al.* (2006), normal and abnormal seedlings are counted on the fourteenth to determine the germination percentage. Normal seedlings as defined by the ISTA rules show the potential for continued development into satisfactory plants when grown in good quality soil and under favourable conditions of moisture, temperature and light. To classify a normal seedling, it must comply with of the categories which are intact seedlings, seedlings with slight defects or deficiencies and seedlings with secondary infection. Meanwhile, abnormal seedlings possess of retarded growth of the seedling as a whole, short and stubby roots, primary leaf missing, etc. It is reported by Bewey and Black (1994) that germination occurs if three conditions exist: the embryo must be alive, seed dormancy must be overcome, and the proper environmental conditions must exist for germination.

2.5 Seed Vigor

Seed vigor is an important quality attribute of the potential field performance of a seed lot in the field or in storage. The ISTA congress in 1977 adopted the definition of seed vigour as the sum total of those properties of the seed which determine the level of activity and performance of the seed or seed lot during germination and seedling emergence. Rapid establishment and high vigor resulted in faster development, earlier flowering and maturity and higher yields.

2.6 Seed Priming

Seed priming is defined as seed enhancement in which seed immersed in water or low osmotic potential solution to trigger the germination metabolic activities but insufficient to allow radical emergence (Bradford, 1986; Bray, 1995; Farood *et al.*, 2006). Further drying of primed seed to its original weight or moisture content prior storage or any test is necessary as to reduce contamination occur.

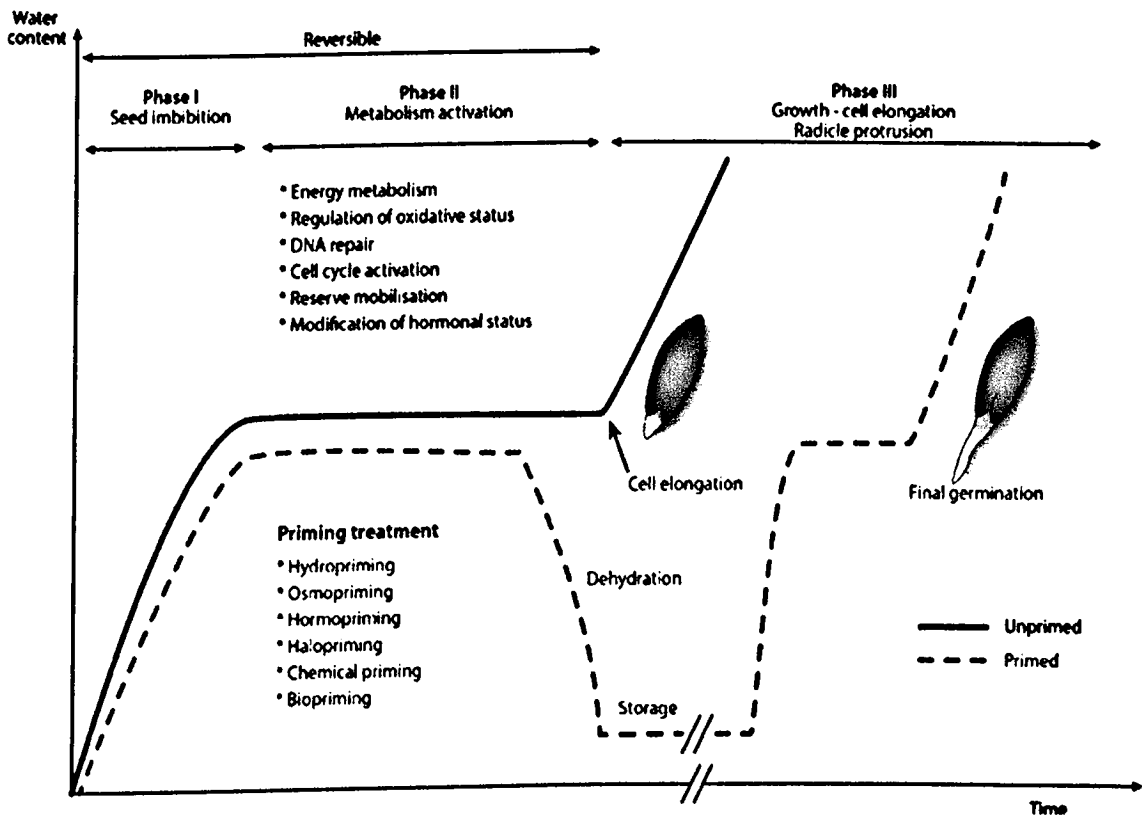


Figure 2.2 Seed hydration curves and germinating phases in unprimed and primed seeds

Sources: Stanley *et al.*, 2016

In the time of priming, three phases of water uptake occur in seed as shown in Figure 2.2 (Bewley and Black, 1994; Bray, 1995; Iqbal *et al.*, 2014). Rapid imbibition of water in seeds occurs in phase 1 because of high water potential difference between dry seeds and water. Some metabolic activities take place at this phase. Second phase is the lag phase, during which, little water uptake causes little change in fresh weight of seeds. This is the time period of food reserve in seed degraded, cell membrane reorganized and biosynthesis of starch occur needed for germination. Only germinating seeds are capable of entering phase three which is germination phase where radicle emergence and then root growth and seedling growth continue. Priming prevents seeds from entering Phase-III of hydration by extending and holding seeds within the lag phase (Taylor *et al.*, 1998).

Many priming method have been introduced which include water (hydropriming), PEG (osmopriming), salt (halopriming) and others. In general, seed priming have shown an overall advantage such as improve seedling vigor, germination rate and seedling uniformity in field crop (Heydecker and Coolbaer, 1977; Bradford, 1986; Kaya *et al.*,

2006). It was reported that it improved germination rate and seedling uniformity in rice (Lee, 1998; Farooq *et al.*, 2006). Several literatures revealed that seed priming significantly improved seed size, vigor and primed crops produced large seeds as compared to non-primed crops (Basra *et al.*, 2003; Hussain *et al.*, 2013). Seed size significantly affected the germination, seedling emergence and large seeds germinate earlier and faster emerge on soil surface as compared to small size seeds (Larsen and Andreasen, 2004; Willenborg *et al.*, 2005). Priming had shown many advantages as compared to non-primed seeds. The priming effects depended on the nature of priming agent, priming duration, concentration of priming agent and the variety of seed (Jeong *et al.*, 2000). In laboratory experiment, characters of final germination percentage, mean germination time, root length, shoot length and seedling dry weight were affected significantly by cultivar, priming and salinity (Goudarz *et al.*, 2012). Pre-soaking the seeds in different treatments followed by incubation at 25/10 °C (light: dark; 16:8 h) revealed variation in germination percentage, mean germination time (MGT), germination rate and seed vigour index.

2.6.1 Hydropriming

Water priming or called as hydropriming is a simple and economical treatment as it used water to soak the seeds for a period of time at favourable temperature (Fujikura *et al.*, 1993). It is a very important seed treatment technique for rapid germination and uniform seedling establishment in various grain crops (Abebe and Modi, 2009). "On-farm" seed priming (soaking seeds in water prior to sowing) has been shown to be effective in producing early germination, better establishment and increased yields in a wide range of crops in diverse environments (Rashid *et al.*, 2006). Hydropriming was found to be the effective method for improving seed germination of pinto bean, especially when the seeds were hydrated for 14 hours as compared to 21 hours. The extended priming duration negatively affected laboratory and field performance of pinto bean cultivar (Kazem *et al.*, 2010). In basil (*Ocimum basilicum* L.) under saline conditions, the seedling vigor, germination percentage and seedling dry weight was found to increase due to hydropriming (Farahani and Maroufi, 2011). Dexfuli *et al.* (2008) presented hydropriming as suitable, cheap and easy seed invigoration treatment for inbred lines of maize, especially when germination is affected by salinity and drought stress. Primed crops produced 80 % more above-ground

biomass (3.3 vs. 1.9 t ha⁻¹), 264 % more pod yield (1.0 vs. 0.28 t ha⁻¹) and 415 % more grain (0.36 vs. 0.07 t ha⁻¹) than did non-primed crops (Rashid *et al.*, 2004).

2.6.2 Potassium Nitrate

It is reported that seed priming with nitrate solution stimulates germination that resulted better germination and seedling emergence in maize (Hanegave *et al.*, 2011), and tomato (Tulio *et al.*, 2013). Besides, Farooq *et al.* (2005), reported that nitrate solutions helped in shortening time required for spread of germination. Moreover, nitrate broke the seed dormancy in *Arabidopsis thaliana* seeds by reducing abscisic acid levels (Matakiadis *et al.*, 2009). Potassium nitrate priming had been shown benefit in tomato seeds which acted promoting a faster germination in tomato (Tulio *et al.*, 2013). They further explained that the salt solution removing the dormancy of tomato seed. In Canola (*Brassica napus* L.), this priming increasing maximum final germination percentage, germination index, seedling fresh and dry weight over the unprimed seed (Farzin and Leila, 2012).

Moreover, Kattimani *et al.* (2009), indicated that primed seeds with nitrate solutions produced vigorous seedlings, more dry matter accumulation and root length in compared with non-primed seeds. Mohammadi (2009), found that among the priming treatments, primed seeds with potassium nitrate showed highest values for all traits. Comparison between potassium nitrate and non-primed seed also further explained where potassium nitrate increased germination percentage, germination rate and seedling dry weight by 28.3, 129.4 and 58.1 percentage respectively. Kaya *et al.* (2006), indicated that priming sunflower seeds with KNO₃ led to increasing of germination percentage in drought and salinity stresses. Another report presented the KNO₃ primed seed show the highest radical length than water and urea priming in *Zea mays* (Hadi *et al.* 2012).

2.6.3 Sodium Chloride

Seed priming with different salts, especially NaCl, have shown to improve germination and growth of many crops under stressed conditions (Mohammadi 2009). Mohammadi (2009) reported that NaCl priming of canola seeds increase germination percentage, germination rate and seedling dry weight as compare with unprimed seeds. However, some studies reported that KNO₃ primed seeds excelled over all other priming agents

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