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Relationship between Seed Vigour and Electrolyte Leakage in Rice Seeds with Different Grain-filling Period

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ABSTRACT

The conductivity test is a measure of electrolytes leakage from plant tissue. The shorter the maturation period after heading was the greater electrical conductivity (EC) of rice seed. The polymer-coated seed was not different in EC compared with non-coated seed. As soaking time of rice seed increased, EC increased gradually. The EC varied from 9.9 to 20.7μS $cm^{-1}g^{-1}$ for control plots and from 21.3 to 41.7μS $cm^{-1}g^{-1}$ for heat-killed seeds which were produced by autoclaving seeds at 121℃ for 20 minutes. The germination speed (the rate of 5th day) of rice seed was 94% at control plot, 83% at low temperature and 20% at high temperature. Besides, germination percentage was 95% for the control, 92% for the low temperature treatment and 39% for the high temperature treatment. The EC was negatively correlated ($r=-0.771^{**}$) with germination percentage at low temperature. Water uptake in seeds of 30, 40, 50 days after heading (DAH) was greater than that of 20 DAH.

Plant height of seedlings was 9.84 cm for the control but 4.32 cm for the high temperature treatment, and the tallest for polymer-coated seed. Dry weight of seedlings was 0.841 g for the control and 0.287 g at high temperature. Besides, the polymer-coated seed was heavier than non-coated seed. The number of roots was largest from 40 to 50 DAH and polymer-coated seed, but was decreased from 20 to 30 DAH. The length of roots was 20.52 cm at control plot and 19.89 cm polymer-coated seed but 8.68 cm for the low temperature treatment and 7.28 cm for the high temperature treatment.

Key words : electrical conductivity, seed vigour, grain-filling period, heat-killed seeds, germination, polymer-coated seed, water uptake, temperature.

Although seeds with high germination percentage are sown on farms, the emergence of seedling can be low, and sometimes even require re-seeded. If bad weather conditions occur before or after seeding, poor seedling stand, and delayed maturation can cause lower yields. In these cases, seed with high seed vigor has an advantage compared with the seed with low seed vigor.

In predicting seed vigor and emergence rate under various field conditions, conductivity test can be used before seeding. The conductivity test for electrolytes is a method which measures the degree of leakage of electrolytes such as amino acids (Harman & Granett, 1972), proteins (McKersie & Stinson, 1980), sugar (Takayanagi & Murakami, 1969), phenolics (Samad & Pearce, 1978) and inorganic compounds; potassium, phosphate, magnesium (McKersie & Stinson, 1980; Samad & Pearce, 1978). It was first adopted to test the viability of cotton seed by Presely (1958). It was later developed into the vigor test for the prediction of field emergence rate of wrinkled-seeded garden peas, soybeans (Matthews & Bradnock, 1968), peas, and French bean (Loeffler et al., 1988; Tao, 1978). Changes in the organization of cells occur when dry seed takes up water during the development of seed to physiological maturity (Abdul-Baki, 1980).

The main components of this cell organization are the cell membranes, and cell membrane integrity can be considered to be a fundamental cause of difference in seed vigor. A seed lot with low germination percentage yields a large quantity of electrolytes after soaking and vigor is low and field emergence is likely to be poor.

Factors influencing the conductivity test in pea and soybean include ion content of the soak water, soaking temperature, the soak period, seed moisture content, and seed size (Duke et al., 1986; Ellis et al., 1990; Eua-umpon, 1991; Hampton et al., 1992; Polock et al., 1969). Tao (1978) also suggested that only uninjured soybean seeds should be tested, but Loeffler et al. (1988) concluded that seeds should be randomly selected from the pure seed fraction of the seed lots. Similarly, AOSA (1983) suggested that seed treatment was unnecessary before testing, but Loeffler et al. (1988) reported that seed treatment with fungicides had little effect on conductivity results. The 'Seed Vigour Testing Handbook' (AOSA, 1983) and 'Handbook of Vigour Test Methods' (ISTA, 1987) contain recommended methods for conductivity test of soybean and peas, respectively.

Until now, few seed vigour researches by EC in rice seed have been done. Accordingly, this study was conducted to investigate the relationship between electrolytes leakage and seed vigour in seeds of different maturation in rice.

MATERIALS AND METHODS

'Donjinbyeo', a japonica type was used for the experiments. Rice seedlings were transplanted on May 15, and headed on August 25. The rice seeds were harvested

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at 10 day intervals from 20 to 50 DAH (Sep. 14, Sep. 24, Oct. 2 and Oct. 14). The seeds of 40 DAH were coated with polymer (Sacrust).

EC test of seed (AOSA, 1983) : the test was conducted by soaking 25 seeds in 500 ml conical flasks with 75 ml deionized water and with 4 replicates. EC of control seeds was measured after 2, 4, 8, 12 and 24 hours at room temperature (25℃). The other seeds were autoclaved at 121℃ for 20 minutes for heat-kill (Hill et al., 1988).

The water uptake of seeds was measured at 2, 4, 8, 12 and 24 hours after sowing at room temperature (25℃). Seed germination percentage were measured at 20, 30, 40 and 50 DAH with three replicates of 50 seeds in pH 7.0 paper towel (30×60 cm, Anchor Co.) in a 25℃ germinator. The test of germination speed (Burris and Mcgee, 1991) was measured on the 5th day after seeding and seedling growth was measured on the 14th day after seeding. For low temperature germination, seeds were kept for 7 days at 10℃ germinator, the speed of seed germination was measured on the 5th day after seeding at 25℃ and seedling growth was measured on the 14th day after seeding with three replicates of 50 seeds in pH 7.0 paper towel. For high temperature germination, seed germination speed was measured on the 5th day after seeding at 25℃, accelerating aging treatment for 2 days at 45℃, 100% moisture with three replicates of 50 seeds in pH 7.0 paper towel.

RESULTS AND DISCUSSION

Germination and EC of rice seed with different grain-filling periods

When soaked for 24 hours, seeds harvested 20~30 DAH showed ECs of 25.5~29.1 μS cm^{-1}g^{-1}, and germination percentage of 90~94% (Table 1). Seeds harvested 40~50 DAH showed EC of 16.7~17.1 μS cm^{-1}g^{-1} and germination percentage of 98%. The EC of 40~50 DAH seeds was lower than that of 20~30 DAH, and germination percentage increased by 6%. Also, polymer-coated seeds showed an EC of 15 μS cm^{-1}g^{-1}, and germination percentage of 98%, suggesting these two characters were not different between polymer-coated and non-coated seeds.

The premature seeds had greater electrolyte leakage than the fully matured seeds, showing the similar results that early-harvested seeds had greater leakage because of early disintegration of cellular membranes (Harman & Granett, 1972).

The electrolyte leakage with different grain-filling periods

Time course of the electrolyte leakage measured by EC under 25℃ is shown in Fig, 1. As soaking time increased, the EC increased gradually. The EC increased from 9.9 (2 hours) to 20.7 μS cm^{-1}g^{-1} in 24 hours for the control. The EC of 20~30 DAH seeds was 12.8~12.9 μS cm^{-1} g^{-1}, and was 7.0~17.1 μS cm^{-1}g^{-1} for 40~50 DAH seeds.

Table 1. Germination percentage and electrical conductivity of rice seed with different grain-filling period.

Days after heading	Germination percentage (%)	Conductivity (μS cm^{-1} g^{-1})
20	90 b†	29.1 a
30	94 ab	25.5 ab
40	98 a	16.7 c
50	98 a	17.1 c
PC‡	98 a	15.1 c
Mean	96	20.7

† ; Duncan's multiple range test at 5 % level.
‡ PC ; polymer-coated seed (Sacrust).

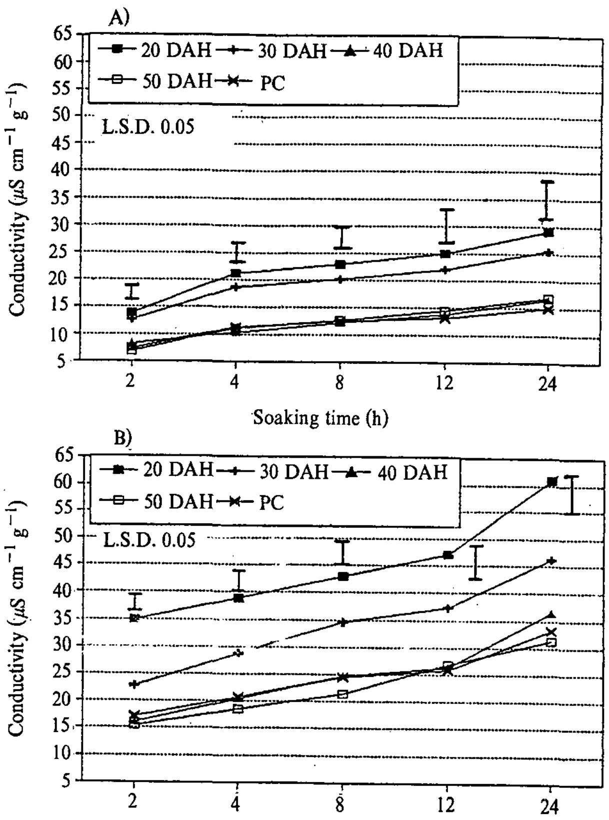

Fig. 1. Time course of electrical conductivity of rice seed with different grain-filling period.
A) control plot. B) heat-killed seeds.
*Heat-killed seeds were produced by autoclaving seeds at 121℃ for 20 min.

Also, the EC increased to 21.3~41.7 μS cm^{-1}g^{-1} for the heat-killed treatment. Those of 20~30 DAH seeds were 22.8~60.9 μS cm^{-1}g^{-1} and 15.4~36.6 μS cm^{-1}g^{-1} for

Table 2. Germination speed and germination percentage of rice seed with different grain-filling period.

Days after heading	Germination speed (%)			Germination percentage (%)		
	C†	HT‡	LT§	C	HT	LT
20	86 b¶	14 c	70 c	90 ab	29 c	78 c
30	90 ab	16 c	81 b	94 ab	41 b	90 ab
40	98 a	28 ab	83 b	98 a	46 b	98 a
50	98 a	8 cd	84 b	98 a	22 c	96 ab
PC#	98 a	35 a	97 a	98 a	56 a	97 ab
Mean	94	20	83	95	39	92

† C ; Control plot.
‡ HT ; 5th day after 2 days at 45℃.
§ LT ; 5th day after 7 days at 10℃.
¶ Duncan's multiple range test at 5% level.
# PC ; polymer-coated seed (Sacrust).

40~50 DAH seeds. Accordingly, the EC decreased by 7.4~24.3 μS cm^{-1}g^{-1} in 40~50 DAH seeds. Compared with the control treatment, heat-killed seeds showed increased electrolyte leakage was increased.

It has been shown that high EC due to greater electrolyte leakage was derived from imperfect formation of seed coat and breaking of the seed coat by high temperature in seeds with shorter grain-filling (Hill et al., 1988; Kuo, 1989; McDonald et al., 1988; McKerise & Senaratna, 1983; Powell & Mattews, 1981).

In addition, the EC of polymer-coated seed was shown 7.5~15.1 μS cm^{-1}g^{-1} for 24 hours at control plot, and 17.2~33.3 μS cm^{-1}g^{-1} for the heat-killed treatment.

Germination speed and germination percentage with temperature treatments

The germination speed and germination percentage of rice seeds with different maturation period is shown in Table 2. The germination speed measured by germination on the 5th day after seeding was 94% in the control plot, and 20% in high temperature treatment and 83% in low temperature treatment.

As compared with control plot, average germination speed decreased by 11% in the low and 74% in the high temperature treatments. The germination speeds were 28%, and 35% in 40 DAH without and with polymer-coated seed in high temperature treatment, respectively. The other treatments were 8% to 16%. Germination percentage was 95% for the control, 92% for the low temperature, and 39% for the high temperature treatment. In the heat-killed treatment, germination percentage was decreased, for the high temperature destroyed the function of seed vigor according to hydration of plasma membrane with phospholipid (Hill et al., 1988; Kuo, 1989).

Correlation between EC and germination percentage

The EC was negatively correlated (r=−0.586**) with germination percentage in the control treatment, was not

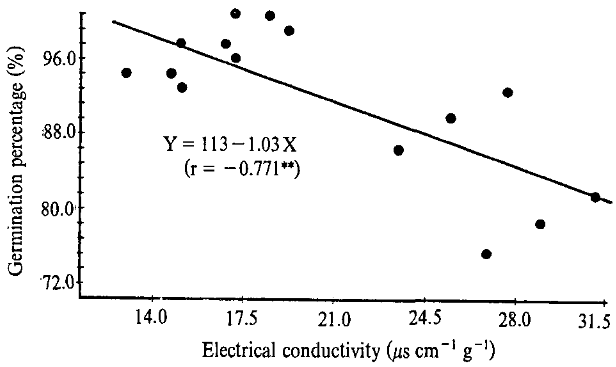

Fig. 2. Relationship between electrical conductivity and germination percentage (Low temperature) of rice seed with different grain-filling period.

Table 3. Simple correlation coefficients between electrical conductivity and germination percentages of rice seed with different grain-filling period.

Characters	Electrical conductivity	Germination percentage (%)	
		Control plot	High temperature
Control plot	−0.586*		
High temperature†	−0.349	0.370	
Low temperature‡	−0.771**	0.942**	0.447

† 5th day after 2 days at 45℃.
‡ 5th day after 7 days at 10℃.

significantly correlated in the high temperature and was negatively correlated (r=−0.771**) in the low temperature treatment (Table 3, Fig. 2). In mungbean and soybean conductivity was significantly correlated with both germination percentage and thousand seed weight (Eua-umpon, 1991). But there were no significant correlations for French bean seed lots. Seed moisture content was not related to other quality components in any species (Hill et al., 1988).

Water uptake of seeds

The water absorption of seeds was rapid up about until 8 hours, followed by relatively slow absorption (Table 4). Water uptake in seeds of 30, 40, 50 DAH was greater than that of 20 DAH. The polymer-coated seed was faster in water uptake compared with non-coated seed in the same maturation period.

Seedling growth of seeds with different grain-filling periods and temperature treatments

Plant height of seedlings was 9.84 cm for the control treatment, 4.83 cm for the low temperature treatment and 4.32 cm for the high temperature. The plant height of 40 DAH was the tallest, and polymer-coated seed yield taller seedlings compared with non-coated seed. Total dry weight of seedling was 0.841 g for the control treatment, 0.400 g for the low temperature and 0.287 g for the high temperature treatment. Seedling dry weights of seed more than 30 DAH were not significantly different, but it was significantly lower for the 20 DAH treatment. The polymer-coated seed was heavier than non-coated seed (Table 5).

The number of roots of more mature seed than 30

Table 4. Time course of water uptake (%) of rice seed with different grain-filling period (25℃).

Days after heading	Soaking time (hrs)					
	2	4	8	12	24	48
20	15.70	17.53	20.38	22.36	24.95	28.93
30	16.10	19.45	23.43	24.52	27.81	30.83
40	16.70	18.31	22.54	24.35	26.20	32.13
50	16.07	18.61	23.30	24.86	26.15	33.06
PC†	19.48	25.31	30.34	32.13	34.35	39.47
Mean	16.81	19.84	24.00	25.64	27.89	32.88

† polymer-coated seed (Sacrust).

Table 5. Changes of plant height and dry weight of seedling growth of rice seed with different grain-filling period.

Days after heading	Plant height (cm)			Seedling dry weight (g /100 plants)		
	Control	HT†	LT‡	Control	HT	LT
20	8.67 bc§	2.61 bc	2.92 b	0.666 c	0.087 c	0.130 cc
30	9.37 b	3.64 b	3.85 b	0.856 ab	0.343 ab	0.382 b
40	10.61 ab	3.79 b	4.37 b	0.889 ab	0.348 ab	0.454 ab
50	9.37 b	3.43 b	3.65 b	0.859 ab	0.282 ab	0.447 ab
PC¶	11.18 a	8.13 a	9.36 a	0.935 a	0.375 a	0.587 a
Mean	9.84	4.32	4.83	0.841	0.287	0.400

† 14th day after 2 days at 45℃.
‡ 14th day after 7 days at 10℃.
§ Duncan's multiple range test at 5 % level.
¶ PC ; polymer-coated (Sacrust).

Table 6. Changes of number of roots and root length of seedling growth of rice seed with different grain-filling period.

Days after heading	Number of roots (cm)			Root length (cm)		
	Control	HT†	LT‡	Control	HT	LT
20	3.7 c§	1.6 cd	1.7 c	14.29 c	2.80 c	2.53 d
30	6.1 ab	2.9 c	3.9 b	24.40 a	6.25 b	5.75 c
40	6.2 ab	4.3 ab	5.2 ab	21.87 ab	5.18 b	8.40 b
50	6.2 ab	4.1 ab	4.5 ab	19.79 ab	4.03 b	7.42 b
PC¶	6.7 a	4.5 a	5.8 a	22.25 ab	18.12 a	19.30 a
Mean	5.78	3.48	4.22	20.52	7.28	8.68

† HT ; 14th day after 2 days at 45℃.
‡ LT ; 14th day after 7 days at 10℃.
§ Duncan's multiple range test at 5 % level.
¶ PC ; polymer-coated seed (Sacrust).

DAH treatment were not different. Seed more than 40 DAH was significantly greater compared with 20~30 DAH, and the number of roots with polymer-coated seed was not different in the same maturation of non-coated seed. The length of roots was, in order, an average of 20. 5 cm for the control, 19.9 cm for the polymer-coated treatment, 8.7 cm for the low temperature treatment and 7.3 cm for the high temperature treatment. The length of roots in 30 DAH compared with 20 DAH were longer, but those of more than 30 DAH were the same regardless of grain-filling periods. The formation of roots in polymer-coated seed at the same maturation was remarkably increased (Table 6).

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