

Influence of High and Low Temperature Treatments on Seed Germination and Seedling Vigor of Rice (*Oryza sativa* L.)

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Abstract: In a laboratory study, The seeds of two rice cultivars included Fajer and Sherodi seeds were exposed thermal hardening (heating followed by chilling followed by heating; chilling followed by heating followed by chilling; heating followed by chilling or chilling followed by heating). Seed subjected to chilling followed by heating (C+H) resulted in improvement of germination percentage, increasing of germination index, seedling, radicle and stem length compared to the other treatments. In Fajer cultivar, the highest EC of seed leachates was observed from H+C. In Sherodi cultivar the highest EC of seed leachates was obtained in control seeds (untreated). In Sherodi cultivar, all seed treatments resulted in a lower EC of seed leachates compared to control.

Key words: Thermal hardening • Germination percentage • Seedling vigor and rice cultivar

INTRODUCTION

Seed invigoration treatments are developed to improve seed performance during germination and emergence. Seed hardening has been used for vigor enhancement in rice [1,2]. In hardening, seeds are exposed to alternate wetting and drying in distilled or tap water [3]. Aged seeds treated with osmoconditioning and hardening increased their total sugar content and α -amylase activity, but hardening was more effective than osmoconditioning. The α -amylase activity is positively correlated with total sugars and the germination rate [4]. Also dry-heat treatment of seeds is used to control external and internal seed borne pathogens such as fungi, bacteria, viruses and nematodes [5] and to break seed dormancy [6]. In general, the high temperature in this treatment reduced seed viability and seedling vigor, but the optimum temperature for breaking dormancy promote seed germination and seedling emergence in cereal crops [7] and cotton [8]. The degree of promotion of seed germination by dry - heat treatment showed wide intraspecific variation [9]. Presowing chilling treatments are being used effectively alone or with other invigoration techniques to shorten the period between planting and emergence and to protect the seeds from a biotic and biotic stresses during the critical phase of seedling establishment [10].

Farooq *et al.* [11] reported that japonica and indica rice seeds were exposed to dry heat treatment (namely 40°C for 72h and 60°C for 24) and chilling (-19°C) treatment for 72h. In indica rice, dry heat treatment at 40°C for 72 h resulted in increased vigor, whereas in japonica rice none of the treatments resulted in improved germination and seedling vigor. Also for coarse rice seed, the highest final germination percentage was noted in seeds subjected to chilling. Our study aimed to study the influence of high and low-temperature treatments on the germination and seedling vigor of both cultivars of rice.

MATERIAL AND METHODS

The Experiment was conducted in the laboratory of the Seed and Plant Certification and Registration Research Institute (SPCRI), Karaj, Iran in 2011 to determine The influence of high and low temperature treatments on seed germination and seedling vigor of two rice cultivars. Experimental units were arranged factorially in a completely randomized design (CRD) with three replications.

Seed Materials: Seeds of both cultivar, Fajer and Sherodi were used as the experimental material. Seeds were obtained from the Rice Research Institute (Golestan, Iran).

The initial seed moisture content of Fajer and Sherodi were 8.81 and 8.93 %, respectively (on dry weight basis).

Seed Treatments: The following seed treatment (thermal hardening) was integrated for vigor enhancement.

Heating: For dry heat treatment, 250g seeds of each cultivar were incubated at 40°C for 24h in an oven (Model memert-klibrier service INE 500, German). Seeds were incubated in glass jars tightly covered with lids.

Chilling: Seeds (250g) of both Fajer and Sherodi rice cultivars were sealed in polythene bags and placed in a refrigerator (Model Flocchetti Linea Ristorazione Luzzara (RE) Italy) at -20°C for 24 h.

The study consisted of the following treatment combinations: (i) H+C+H= heating followed by chilling followed by heating; (ii) C+H+C=chilling followed by heating followed by chilling; (iii) H+C= heating followed by chilling ; and (iv) C+H= chilling followed by heating .

Post-treatment Procedures: After treatment of seeds with a particular method and also for a particular duration, seed were sealed in polythene bags and stored in a refrigerator at 5°C until further use.

Germination Test and Seedling Emergence: 50 seeds from each treatment and Control were placed in 18 cm×6 cm germination box containing two layers of moistened blotters with distilled water. Seeds were placed in growth chamber (25°C) under normal light. The relative humidity was maintained at 70±5%. Germination was recorded daily according to the AOSA method [12].

Germination Percent (GP), MeanGermination Time (MGT) and germination index (GI) were calculated as described in Scott *et al.* [13] (equation 1, 2 and 3).

$$GP = \frac{\text{Total seeds germination after day 14}}{\text{Total number of planted seeds}} \quad (1)$$

$$MGT \text{ (day)} = \frac{\sum TiNi}{\sum Ni} \quad (2)$$

$$GI = \frac{\sum TiTi}{S} \quad (3)$$

Where Ti is the number of days after sowing, Ni is the number of seeds germination on i th day, $\sum Ni$ (equation 2) is the total number of germinated seeds and S (equation 3) is the total number of seeds used .

Final germination percentage, seedling length, radicle length, stems length and seedling dry weight were recorded 14 days after cessation of the experiment. Experimental data were analyzed using SAS [14] and treatment means were compared using Duncan's Multiple Range Test (DMRT) at 5% level.

Electrical Conductivity of Seed Leachates: After washing in distilled water, 5 g seeds was soaked in 50mL distilled water at 25°C .The electrical conductivity (EC) of the soak solution was measured at 0.5,1.0,1.5,2.0,6.0,11.0 and 24.0 h of soaking using a conductivity meter (model LF90 SER-NA31245385) and expressed as $ms.cm^{-1}$.

RESULTS

Germination: Seeds subjected to C+H germinated earlier than seeds subjected to other treatment combinations (Table 1). All treatments resulted in lower MGT than that of control (but satistically similar to that of the control). Seed treatments had significantly effects on final germination percentage. Also the highest GP was obtained in seed treated with C+H and C+H+C. Minimum GP was noted in seeds subjected to H+C and H+C+H. Maximum GI was recorded in seeds subjected to C+H that did not differ significantly from that of seeds subjected to the other treatments (Table 1).

Seedling Vigor: Radicle length affected by presowing treatments. Maximum radicle length was observed from seeds treated with C+H that did not differ significantly from that of seeds subjected to the other treatments and minimum radicle length was noticed in control seeds. However the highest seedling and stem length were recorded in seed treated with C+H, which was not significantly different from the control.

Treatments did not significantly affect the seedling dry weight. The interaction between seed treatments×cultivar had not meaningful effects on germination and seedling vigour.

Table 1: Effect of seed treatments on the germination vigour of rice cultivars

Seed treatments	Germination percentage	Seedling length (cm)	Stem length (cm)	Radicle length(cm)	GI	Mean Germination Time (MGT day)	Seedling dry weight(g)
C+H+C	94 a	17.02 a	9.75 a	7.26 ab	1.9a	2.03a	a 0.15
H+C+H	80.33 b	16.88 a	9.62 a	7.26 ab	1.84a	2.04a	a 0.15
C+H	94.66 a	17.95 a	9.93 a	8.01 a	1.97a	1.88a	0.15 a
H+C	89.33 b	15.89 a	8.77 a	7.11 ab	1.88a	2.04a	0.15 a
control	91.66 ab	16.27 a	9.56 a	6.7 b	1.91a	2.08a	0.15 a

Duncan's multiple range test at 0.05

* Means with same letter are not significantly different at 0.05, H+C+H=Heating followed by chilling followed by heating; H+C=Heating followed by chilling; C+H+C=Chilling followed by heating followed by chilling; C+H=Chilling followed by heating.GI, germination index; MGT, mean germination time.

Table 2: Effect of seed treatments on seed leachates (ms.cm⁻¹) of rice cultivars

Cultivar	Seed temperature treatment	Soaking period(h)						
		0.5	1	1.5	2	6	11	24
Fajer	C+H+C	0.16d	0.22c	0.256c	0.256c	0.29d	0.31f	0.33f
Fajer	H+C+H	0.106n	0.12n	0.216m	0.216m	0.24l	0.37e	0.43n
Fajer	C+H	0.18c	0.21d	0.246e	0.246e	0.35c	0.51b	0.62b
Fajer	H+C	0.206b	0.25b	0.346b	0.346b	0.45a	0.58a	0.66a
Fajer	control	0.276a	0.3a	0.356a	0.356a	0.36b	0.42c	0.44m
Sherodi	C+H+C	0.166e	0.16f	0.166f	0.166f	0.33e	0.41c	0.43n
Sherodi	H+C+H	0.146m	0.176e	0.186n	0.186n	0.28f	0.37e	0.47e
Sherodi	C+H	0.146m	0.146m	0.156h	0.156h	0.24n	0.26m	0.31l
Sherodi	H+C	0.106n	0.116l	0.136l	0.136l	0.25m	0.25m	0.28n
Sherodi	control	0.156f	0.176e	0.216m	0.216m	0.25m	0.35e	0.48c

Duncan's multiple range test at 0.05

* Means with same letter are not significantly different at 0.05, H+C+H=Heating followed by chilling followed by heating; H+C=Heating followed by chilling; C+H+C=Chilling followed by heating followed by chilling; C+H=Chilling followed by heating.

Electrical Conductivity of Seed Leachates: In Fajer cultivar, the highest EC of seed leachates was observed with H+C and minimum EC of seed leachates was noted in seed subjected to C+H+C. In Sherodi cultivar, the highest EC of seed leachates was obtained in control seeds (untreated) and minimum EC of seed leachates was found in seeds subjected to H+C. In Sherodi cultivar, all seed treatments resulted in a lower EC of seed leachates compared to control.

DISCUSSION

Thermal hardening had a significant effect on the germination percentage and radicle length (Table 1). Earlier, synchronized germination and higher germination percentage was observed in treated seeds compared with control seeds, as evidenced by a lower time to start germination and higher GI, as well as radicle length (Table 1) the effect of dry-heat treatment on seed germination depended on dry-heat intensity and duration of exposure. seedling length, stem length and seedling dry weight remained unaffected, but higher seedling and stem length was recorded in seeds treated by C+H. These findings are in line with Ruan *et al.* [15] That reported seed

treatments were not able to invigorate rice seed, but resulted in a higher energy of germination and germination index compared with untreated seeds. Maximum GP and GI were noticed from seeds treated with C+H. The beneficial aspects of seed hardening are primarily due to pre-enlargement of the embryo [16], improvement of the germination rate [17] which are attributed to alternating wetting and drying processed [8]. Also Farooq *et al.* [18] reported that heating followed by chilling followed by heating resulted in decreased mean germination time, time to start germination as well as increased germination index, radicle and plumule length in rice seeds. Higher enhancement was found from the seeds subjected to chilling followed by heating (C+H) treatment. The highest EC of seed leachates was recorded in Fajer cultivar seeds subjected to H+C, which may be the result of membrane rupture during the heating followed by chilling. Also in Fajer cultivar minimum EC of seed leachates was observed the seeds subjected to C+H+C, which may be the result of membrane repair and a lower rate of seed leachates (Table 2). In contrast, Farooq *et al.* [18] reported that the highest EC of seed leachates was related the seeds treated with C+H+C. In Sherodi cultivar the highest EC of seed

leachates was obtained from the control seeds (untreated) and minimum EC of seed leachates was found from seeds subjected to H+C. The rice cultivars responded differently to seed treatments, that indicates genetic differences between the two rice cultivars.

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