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Response of Seed Germination and Seedling Emergence of Rice (*Oryza sativa* L.) Genotypes to Different Osmopriming Levels

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Abstract: The major deterrent factor for adoption of rice seedlings in paddy fields is poor synchronized germination and seedling establishment. Since seed priming could be an suitable approach for better crop stand, the main objective of this study was to determine the effectiveness of different seed priming strategies on germination and seedling parameters of rice. Seed priming consisted of osmopriming by PEG and NaCl in different levels. According to response of genotypes to mentioned treatments they classified into 3 classes by clustering analysis. After selection the genotypes from each cluster ANOVA were performed. The results of ANOVA indicated that priming and media levels significantly affected germination attributes in rice cultivars. In addition, selected genotypes differently responsed to priming media. The highest germination rate and percentage and radicle length in Tarom cultivar was recorded when 4 dS/m^{-1} NaCl priming was used. Also maximum total dry weight was recorded for 8702 line seedlings in -3 MPa of PEG priming. Good responses of Tarom to osmopriming could be derived from it's adaptation to environmental conditions. The results of this study suggest that osmopriming at lower concentration could improve seed and seedlings parameters of rice genotypes.

Key words: Germination · NaCl · Osmopriming · PEG · Rice

INTRODUCTION

Rice (*Oryza sativa* L.) being one of the most cultivated cereals in all over the world and it provides some 700 calories per person, mostly residing in developing countries [1]. Also world rice production must increase by approximately 1% annually to meet the growing demand for food that will result from population growth and economic development [2]. Thus, the present and future food security of Asia depends largely on the lowland rice production system and is a major user of fresh water [3]. However rice production is limited by many factors and is prone losing yield due to exposure to saline and drought conditions. These environmental limitations that impose water-deficit stress play a major role on plant productivity [4]. More than 30 percent of the irrigated land in the world is threatened by salinity [5]. Water salinity causes adverse effects on different physiological processes and thus markedly reduces plants growth [6, 7]. In view of current levels of the growing world population, it is estimated that there will be needed to increase food production up to 38% by 2025 and 57% by 2050 to sustain the food supply. Therefore, the attention should be focused on yield increase per unit of land rather than in the area cultivated [8]. Since well seed germination behavior is one of the important factors in successful agriculture, uneven or poor germination and subsequently inhomogeneous seedling growth can lead to great financial loses, reducing possibilities foe mechanization or lower prices of inhomogeneous plant batches [9]. So, one method for ensuring well and stabile seedlings in paddy field and satisfier grain yield, is treated the seeds of cultivars (seed priming) before or during the germination such as

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osmopriming and hydropriming [10]. Seed priming is a technique that germination processes begin but radicle emergence does not occur [11]. In fact, priming allows to some of the metabolic processes which necessary for germination to occur without actual germination. Primed seeds usually exhibit increased germination rate, higher germination uniformity, and sometimes greater total germination percentage [12]. Osmopriming is the most widely used type of seed priming in which seeds are soaked in aerated low water potential solutions. Examples of such osmotica used include polyethylene glycol (PEG), KNO_3, K_3PO_4, KH_2PO_4, $MgSO_4$, NaCl and manitol. PEG and NaCl, however, are the most commonly used osmoticum for rice priming [10, 13].

Many recently researches believed that seed priming of crops specially rice might be a influence way for better seedling establishment and growth [For example 14, 15]. Thus, the main objectives of this study were to analyze seed and seedling parameters under osmopriming by PEG 6000, NaCl and quantify the responses of contrast rice genotypes which commonly grown or recommended in north of Iran conditions.

MATERIALS AND METHODS

These experiments were conducted in two parts in seed technology laboratory of Sari Agricultural Sciences and Natural Resources University during 2008.

Germination Parameters: First step of experiment consists of osmotic priming by PEG 6000 and NaCl on seeds of 15 promising lines, improved and traditional cultivars which collected from Rice Research Institutes of Iran, Deputy of Mazandaran (Amol) in 2008. Some characteristics of these genotypes are shown in Table 1.

The seeds were surface sterilized in a 1:10 (v/v) dilution of commercial hypochlorite bleach for 10 min and rinsed several times with distilled water. Then 100 uniformly seeds of each varieties were allowed to four levels of polyethylene glycol (PEG 6000) solution (0, -3, -6 and -9 Mpa) and NaCl solution (0, 4, 8 and 12 dS/m^{-1}) for 36 h at 25°C. After priming, seeds from each treatments rinsed several times with distilled water and placed on 9 cm-diameter Petri dishes on Whatman filter paper that was moistened with 10 ml distilled water, then Petri dishes were kept at 25°C with 12 h photoperiod in a germinator (Iran Khodsaz, IKHRH, IRAN) for germination up to seven days. Radicle protrusion of 2 mm was scored as germination.

Table 1: Some characteristics of studied genotypes

No	Name of genotypes	Type	Origin
1	Tarom	Traditional cultivar	Iran
2	Dashti Sard		
3	Sadri		
4	Fajr	Improved cultivar	IRRI
5	Nemat		Iran
6	843		
7	8405		
8	8702		
9	8803		
10	8805	Promising line	IRRI
11	8808		
12	8810		
13	8811		
14	8815		
15	8816		

Germination index (GI) and germination rate (GR) of seeds and vigor index (VI) of seedlings were calculated according to the following equations:

GI = By dividing the number of germinating seeds each day by the number of days and summing the values;

GR = [Number of germinated seeds / number of germination days];

VI = [Seedling length (cm) × germination percentage]

Seedling Parameters: Second step consist of seedling parameters study. Thus, after germination the primed seeds, ten uniformly seedlings were selected and placed to each Petri dish and kept at 25°C with 12 h photoperiod in germinator until 7 days after germination. At the end of seventh day all parameters such as radicle and shoot lengths and dry weight of seedlings were recorded. Wards mean was used for clustering the rice varieties based on their morphological traits. The analysis of variance was done by Statistical Analysis System [16] and MSTATC used for means comparison.

RESULTS AND DISCUSSION

Based on average values of morphological traits between 0 to 5 distances, the 15 varieties were classified into three clusters as, low, medium and high in responses to seed priming (Figure 1).

Based on Table 2, the first cluster (low response) include of four varieties (8816 and 843 lines, Fajr and Dashti Sard cultivars), the second cluster (medium response) include seven genotypes (8805, 8808, 8810, 8803 and 8702 lines, Nemat and Sadri cultivars) and The third cluster (highest response) contained of four varieties (8811, 8815 and 8405 lines and Tarom cultivar).

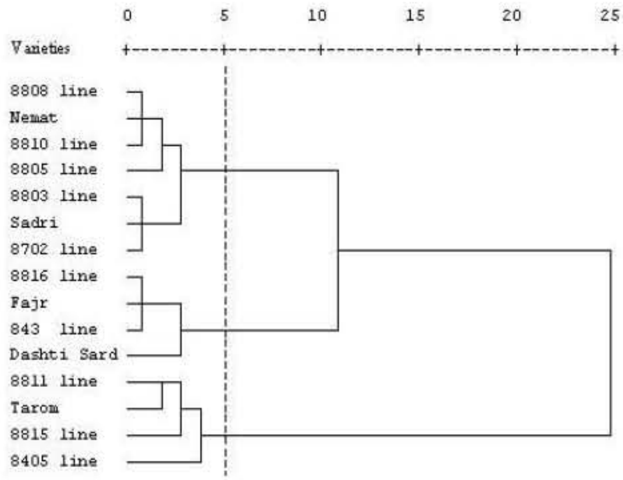

Fig. 1: Dendrogram for 15 genotypes of rice using average of morphological traits

Table 2: Average value of traits in rice varieties by clustering

Cluster	Genotype	Averages of traits value
First cluster		
(Low response)	8816 Line	26.68
	Fajr	26.07
	843 Line	27.45
	Dashti Sard	24.41
Second cluster	8808 Line	30.20
(Medium response)	Nemat	30.10
	8810 Line	29.73
	8805 Line	31.14
	8803 Line	29.06
	Sadri	28.91
	8702 Line	28.74
Third cluster		
(High response)	8811 Line	34.95
	8815 Line	38.47
	8405 Line	33.09
	Tarom	36.70

Therefore, for analysis of variance one genotype was selected from each cluster (according to their resemblance in traits value which considered in Table 1). The representative genotypes after clustering were 8702 as promising line, Fajr as an improved and Tarom as traditional cultivars.

Analysis of variance indicated that priming media and levels significantly affected germination attributes of rice cultivars (Table 3 and 4). In addition, representative genotypes differently affected by priming media. Generally, based on interaction effects (Table 3), osmopriming by PEG could improved root length in Fajr (Low response genotype) and root and shoot length and vigor index in 8702 line (medium response group). However, among genotypes and concentrations both priming treatments could not significantly improve the germination index, percentage and rate compared to unprimed treatment, although the responses of all varieties to different priming levels and types was not much similar. The highest germination rate and percentage in Tarom cultivar was not significantly affected by different levels of NaCl priming (Table 4). In terms of root length, priming in NaCl up to 4 ds/m^{-1} significantly improved root length and vigor index in Tarom cultivar compared to 8702 line and Fajr cultivar (Table 4).

Among varieties, priming media and concentrations the highest vigor index was related to Fajr cultivar which exposed to -3 MPa of PEG solution (Table 3 and 4). As evident from the Table 3 and 4, the highest increasing percentage of total dry weight of seedling was belonged to 8702 line in 4 ds/m^{-1} of NaCl solution (with 28%) in compared to their controls.

Direct seeding could be an attractive alternative to transplanting of rice, but poor germination, uneven crop stand and high weed infestation are among the main constraint to its adoption [17]. Seed priming is an affective technique for rapid and uniform seed germination of several cereal crops specially rice [12, 18]. It was the main derived from this study that, different genotypes can have different response to priming and different priming techniques can have various effects on seed germination and seedlings parameters of rice varieties.

Also results indicated that, well responding of Tarom (as traditional cultivar in north of Iran) to priming can be related to adaptability potential of this cultivar to costal regions in north of Iran where widely cultivated. It seems that, for most evaluated germination parameters, NaCl priming was more effective than osmopriming by PEG.

Table 3: Effect of osmopriming by PEG on seed and seedling parameters of rice varieties

Treatments	Traits	GI*	GP	GR	RL (mm)	SL (mm)	VI	TDW (mg)
Varieties	PEG							
8702 Line	Control (0)	10.45a	94.50a	13.50a	25.31ef	29.30b	516.00b	39.50bc
	-3 MPa	8.25bc	89.00ab	12.71ab	41.93b	34.31a	679.00a	42.25bc
	-6 MPa	7.35cd	89.00ab	12.60ab	36.81c	22.34c	523.00b	39.00bd
	-9 MPa	6.94c-e	88.00ab	12.57ab	29.40de	13.30d	376.00cd	35.00d
Fajr	Control (0)	8.97b	96.5a	13.78a	38.81bc	31.74ab	680.00a	43.00b
	-3 MPa	8.09b-d	95.00a	13.57a	52.17a	25.34c	736.00a	43.50b
	-6 MPa	5.89e	72.00bc	10.28bc	30.24d	13.15d	318.00d	36.25cd
	-9 MPa	6.79de	82a-c	11.71a-c	20.56g	4.74e	206.00e	34.25d
Tarom	Control (0)	10.80a	83.25a-c	11.89a-c	22.00fg	29.25b	426.00c	54.50a
	-3 MPa	4.28f	78.50-c	11.21a-c	7.31h	3.49ef	87.00f	35.00d
	-6 MPa	2.58g	66.50c	9.50c	4.79hi	2.71ef	65.00fg	33.25d
	-9 MPa	0.52h	1.50d	0.21d	1.10i	0.50f	0.37g	21.25e
S.O.V†								
Variety (A)		**	**	**	**	**	**	ns
Osmopriming (B)		**	**	**	**	**	**	**
A×B		**	**	**	**	**	**	**
CV (%)		13.9	18.99	19	12.36	12.73	13.8	12.6

GI= Germination index, GP= Germination percentage, GR= Germination rate, RL= Radicle length,
SL= Shoot length VI= Vigor index, TDW= Total dry weight
*In each column data with the same letter are not significantly different
† ns, not significant, **, significant difference at 0.01 probability level

Table 4: Effects of osmopriming by NaCl on seed and seedling parameters of rice varieties

Treatments	Traits	GI	GP	GR	RL (mm)	SL (mm)	VI	TDW (mg)
Varieties	NaCl							
8702 Line	Control (0)	10.75a	91.00a	13.00a	17.38c	30.13a-c	434.00c	40.75cd
	4 ds m-1	8.83b	87.00a	12.42a	10.54d	30.12a-c	346.00d	52.25b
	8 ds m-1	5.19de	63.00d	9.00d	1.44e	5.97d	48.00e	39.75d
	12 ds m-1	5.62de	69.00cd	9.85cd	2.10e	5.51d	52.00e	33.25e
Fajr	Control (0)	9.25b	89.00a	12.71a	24.88b	32.57ab	513.00b	44.25cd
	4 ds m-1	7.27c	84.00ab	12.00ab	7.69d	25.73c	283.00d	4150cd
	8 ds m-1	5.49de	70.00b-d	10.00b-d	2.66e	5.60d	59.00e	33.75e
	12 ds m-1	3.34fg	38.00e	5.42e	1.10e	3.43d	17.00e	31.25e
Tarom	Control (0)	11.22a	92.25a	13.18a	28.32b	32.72ab	562.00b	58.75a
	4 ds m-1	5.94cd	92.75a	13.25a	41.32a	34.87a	705.00a	57.25a
	8 ds m-1	4.21ef	85.00a	12.14a	10.32d	27.82bc	323.00d	50.75b
	12 ds m-1	1.97g	79.50a-c	11.36a-c	3.57e	5.07d	68.00e	45.25c
S.O.V								
Variety (A)		**	**	**	**	**	**	**
Osmopriming (B)		**	**	**	**	**	**	**
A×B		**	**	**	**	**	**	**
CV (%)		16	12.96	12.59	20.42	18.07	19.06	7.44

GI= Germination index, GP= Germination percentage, GR= Germination rate, RL= Root length, SL= Shoot length, VI= Vigor index, TDW= Total dry weight. In each column data with the same letter are not significantly different **, Significant difference at 0.01 probability level

The present results are in accordance with observation of Basra [12] who suggested that seed germination and vigor in rice increased with osmoconditioning. However, osmopriming has been shown to activate processes related to germination, for instance, by affecting the oxidative metabolic such as increasing superoxide dismotase (SOD) and peroxidase (POD) [19] or by the activation of ATPase as well as acid phosphatase and RNA syntheses [20]. Therefore, increasing seedling dry weight might be due to synchronized germination and improved DNA, RNA synthesis during seed treatments and also the increasing shoot and root lengths by osmopriming may be due to nuclear replication in root and shoot.

CONCLUSION

The results of this experiment indicate that osmopriming by NaCl and PEG especially at lower concentration could improve some parameters of rice seed germination and seedlings growth. However, previous study [16] have shown that soaking seed for more than 12 h (36 in current experiment) using any priming media tended to reduce rate and extent of germination, suggesting that optimum soaking time for rice may be less than 36 hours and need to further investigation. Finally, for the next studies Tarom cultivar was recommended as a tolerant genotype.

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