INTERNATIONAL JOURNAL OF AGRICULTURE & BIOLOGY
1560–8530/2006/08–1–19–22
http://www.fspublishers.org

Germination and Early Seedling Growth as affected by Pre-Sowing Ethanol Seed Treatments in Fine Rice

M. FAROOQ[1], SHAHZAD M.A. BASRA, HEFEEZ-UE-REHMAN AND TARIQ MEHMOOD†
Department of Crop Physiology, University of Agriculture, Faisalabad-38040, Pakistan
†*Department of Agronomy, University of Agrid Agriculture, Rawalpindi, Pakistan*
[1]Corresponding author's e-mail:farooqcp@gmail.com

ABSTRACT

The present study was designed to investigate the possibility of rice seed invigoration by pre-sowing ethanol seed treatment. Fine rice (Super-Basmati) seeds were soaked in 1, 5, 10 and 15% (v/v) aerated solution of ethanol for 48 h. None of the seed could germinate/emerge following 10 and 15% (v/v) ethanol treatment. However, soaking rice seeds in 1 or 5% ethanol solution resulted in earlier and more uniform germination. Moreover higher leaf score was recorded in seedlings raised from 1% ethanol treated seeds. No improvement was recorded for final germination percentage, shoot length and seedling dry weight from any of the pre-sowing ethanol seed treatments.

Key Words: Fine rice; Ethanol; Germination; Seedling growth
Abbreviations: Time to 50 % germination= $T_{50,}$ Mean germination time= MGT, Energy of germination= GE, Final germination percentage= FGP

INTRODUCTION

Seed priming is a technique by which seeds are partially hydrated to a point where germination processes begin but radicle emergence does not occur (Heydecker & Coolbear, 1977; Bradford, 1986). Priming allows for some of the metabolic processes necessary for germination to occur without actual germination. In priming, seeds are soaked in different solutions with a high osmotic potential. This prevents the seeds from taking in enough water for radicle protrusion, thus suspending the seeds in lag phase (Taylor *et al.,* 1998). During the lag phase, the seeds are metabolically active and convert stored reserves for use during germination. This continues during the extended lag phase induced by priming. The seeds are then removed from the priming solution, rinsed with water and dried. Since they have already gone through the early germination processes, when they are planted they germinate faster than seeds that have not been primed.

Primed seeds usually exhibit increased germination rate, greater germination uniformity, and sometimes greater total germination percentage (Basra *et al*., 2002, 2003, 2004, 2005; Farooq *et al*., 2005, 2006). Increased germination rate and uniformity have been attributed to metabolic repair during imbibition (Burgass & Powell, 1984), a buildup of germination-enhancing metabolites (Basra *et al*., 2005), osmotic adjustment (Bradford, 1986), and, for seeds that are not redried after treatment, a simple reduction in the lag time of imbibition (Heydecker & Coolbear, 1977; Brocklehurst & Dearman, 1983).

Improved seed performance has been achieved by incorporating plant growth regulators during priming and other pre-sowing treatments of many crops (Miyoshi and Sato, 1997). GA_3 is well known to activate β-amylase for breakdown of starch stored in seeds to be utilized by growing embryos during germination. GA_3 and ethylene stimulate the elongation of mesocotyl, coleoptile and internodes of rice seedlings after germination (Lee *et al.,* 1999) while ABA promotes elongation of the mesocotyl of rice seedlings (Lee *et al*., 1999). Miyoshi and Sato (1997) applied kinetin and gibberellins on dehusked seeds of indica and japonica rice to study their effects on the germination under aerobic and anaerobic conditions. They found stimulatory effects of gibberellin on the germination of indica and japonica rice seeds under both conditions, while, under anaerobic conditions, the responses of dehusked indica and japonica rice seeds to kinetin and gibberellin differed, being negative with kinetin and positive with gibberellin. Under aerobic conditions, the stimulatory effects of kinetin on germination of dehusked seeds were greater than those of gibberellin.

Ethanol has been reported to have stimulatory effects on the germination of seeds of many plant species (Taylorson & Hendricks, 1979; Bewley & Black, 1982). In another study the dormancy problem in japonica rice was overcome by 0.5-5% ethanol treatment (Miyoshi & Sato, 1997a), which gave the idea of its possible role in vigor enhancement. The present study was therefore, planned to investigate the effects of pre-sowing ethanol seed treatments on the germination and early seedling vigor in fine rice.

MATERIALS AND METHODS

Seed materials. Seeds of fine rice cultivar (Super-Basmati)

were obtained from Rice Research Institute, Kala Shah Kakoo, District Sheikhupura, Pakistan. The initial seed moisture contents were 8.34% (on dry weight basis).

Seed treatments. The seeds were soaked in 1, 5, 10 and 15% (v/v) aerated solution of ethanol for 48 h. The ratio of seed weight to solution volume was 1:5 (g mL^{-1}) (Farooq *et al*., 2006). After treating, seeds were dried near to their original weight, sealed in polythene bags and then stored in refrigerator at 7°C±1 for further use.

Germination test. Seeds (15 in each) were placed in Petri dishes between layers of moist Whatman 45 at 27°C in an incubator. The completely randomized design with four replications was used. Germination was observed daily according to the AOSA method (AOSA, 1990). The time to get 50% germination (T_{50}) was calculated according to the following formulae of Coolbear *et al*. (1984) modified by Farooq *et al*. (2005) as under:

$$T_{50} = t_i + \frac{\left(\frac{N}{2} - n_i\right)(t_j - t_i)}{n_j - n_i}$$

Where N is the final number of germination and n_i, n_j cumulative number of seeds germinated by adjacent counts at times t_i and t_j when $n_i < N/2 < n_j$.

Mean germination time (MGT) was calculated according to the equation of Ellis and Roberts (1981) as under:

$$MGT = \frac{\sum Dn}{\sum n}$$

Where n is the number of seeds, which were germinated on day D, and D is the number of days counted from the beginning of germination.

Energy of germination was recorded at 4th day after planting. It is the percentage of germinating seeds 4 days after planting relative to the total number of seeds tested (Farooq *et al.,* 2005).

Seedling emergence. Control and treated seeds were sown in 20 x 20 cm plastic trays (25 in each) having moist sand, replicated four times and were placed in growth chamber (Vindon, England) in completely randomized design. Day and night lengths were kept 15 and 9 h with 30°Cand 24°C temperatures, respectively. Relative humidity was maintained at 70%. Root and shoot length, and seedling fresh and dry weights were recorded 16 days after sowing. Number of leaves at harvest was designated as leaf score.

RESULTS

Pre-sowing ethanol seed treatments significantly ($p < 0.05$) affected the germination and early seedling growth in fine rice (Fig. 1-4). However, none of the seed could germinate/emerge following 10 and 15% (v/v) ethanol treatment.

Fig. 1. Influence of Pre-sowing Ethanol seed treatments on the (a) time to 50% germination and (b) Mean germination time

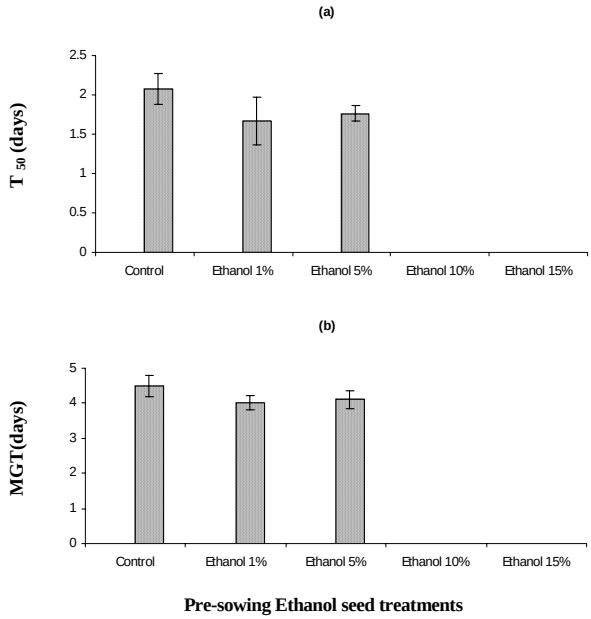
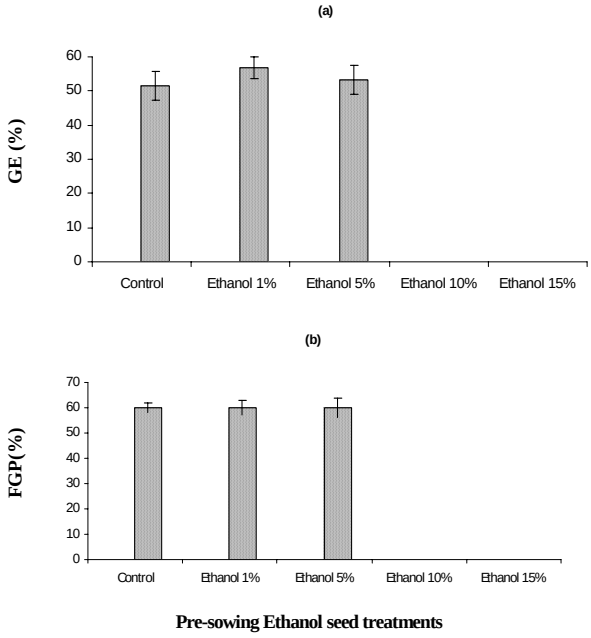


Fig. 2. Influence of Pre-sowing Ethanol seed treatments on the (a) energy of germination and (b) final germination percentage



Ethanol seed treatments in 1 and 5% solutions resulted in lower T_{50} and MGT compared with control (Fig. 1a, 1b). Although none of the treatments could enhance the germination percentage (2b) however, 1% ethanol treatment resulted in improved energy of germination (1a). Similarly,

Fig. 3. Influence of Pre-sowing Ethanol seed treatments on the (a) shoot and (b) root length

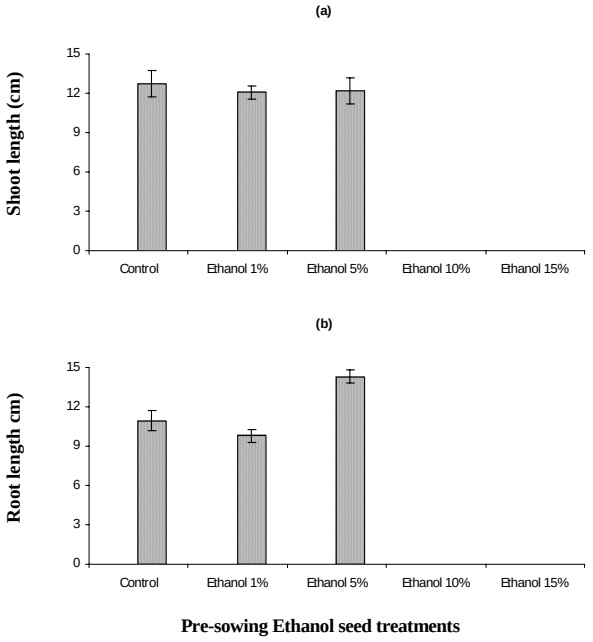
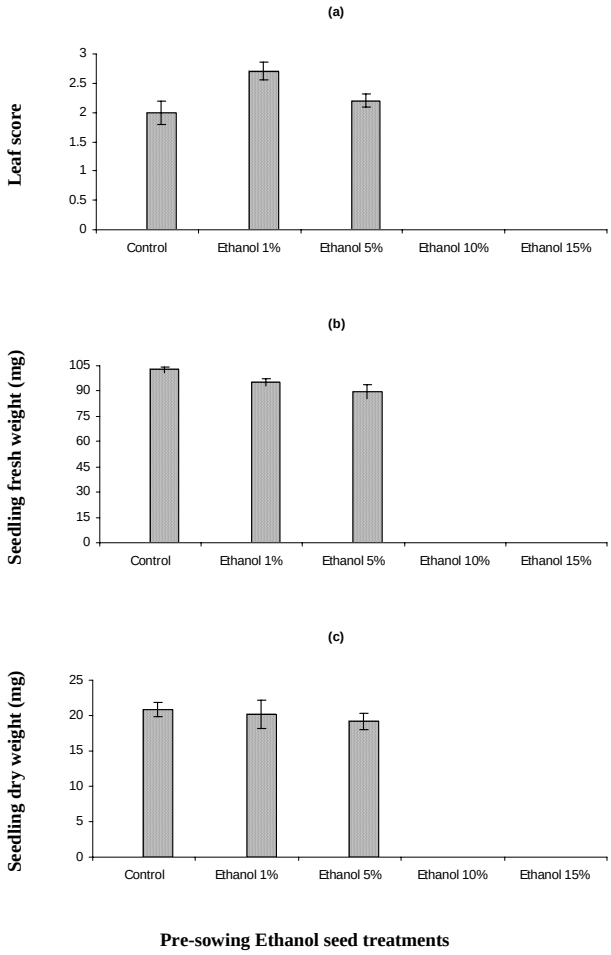


Fig. 4. Influence of Pre-sowing Ethanol seed treatments on the (a) leaf score, (b) seedling fresh weight and (c) seedling dry weight



no treatment could improve the shoot length (Fig. 2a) but elongated roots were noted in rice seedlings raised from 5% ethanol treatment (Fig. 3b).

Although, 1% ethanol treatment resulted in higher leaf score (Fig. 4a), maximum seedling fresh weight was recorded in untreated seeds (Fig. 4b), however none of the ethanol seed treatments could improve the seedling dry weight (Fig. 4c).

DISCUSSION

This study revealed that employing ethanol treatments at lower concentrations can invigorate fine rice seeds. Ethanol seed treatments significantly affected the germination and early seedling growth in fine rice. Higher ethanol concentrations completely inhibited the germination and emergence, although earliness and uniformity in germination was recorded following 1 and 5% ethanol treatments (Fig. 1, 2) that might be the result of dormancy breakdown, as fresh seeds were used during the investigation. The earlier and better-synchronized germination is associated with increased metabolic activities in the soaked seeds (Basra *et al*., 2005). Earlier, Miyoshi and Sato (1997a) had also reported effectiveness of ethanol treatments in dormancy breakdown in rice. Stimulation of the germination of caryopses by ethanol was reported in *Panicum dichotomiflorum* (Taylorson & Hendricks, 1979) and *Avena* spp. (Corbineau *et al*., 1991). Two different mechanisms by which ethanol might break dormancy have been proposed; Taylorson and Hendricks (1979) suggested that the stimulatory effect of ethanol might involve modification of the properties of a membrane(s). Ethanol might also be involved metabolically in the stimulation of germination, as a respiratory substrate. It might accelerate germination by promoting the uptake of oxygen (Fidler, 1968; Adkins *et al*., 1984) and increasing levels of fructose 2, 6-bisphosphate which has been suggested to stimulate glycolysis in dormant seeds of *Avena sativa* (Larondelle *et al*., 1987). Complete germination inhibition in seeds subjected to 10 and 15% ethanol treatments was probably due to its toxicity. In earlier studies, higher concentrations of KNO_3 have been proved toxic in wheat (Basra *et al.,* 2002) and rice (Basra *et al.,* 2003). This toxicity results in injury to the cellular organelles and membranes of wheat (Singh & Gill, 1988). Improved leaf score and root length might due to the possible role of ethanol (at very low concentration) in rapid cell division (Miyoshi & Sato, 1997a).

The present study suggests that ethanol have certain stimulatory effects on the germination and early seedling growth of fine rice at very low concentration and higher concentration is toxic and may even completely inhibit the germination. However, further research is required in this

regard to explore the best concentration of ethanol solution and the soaking duration.

Acknowledgments. Authors acknowledge the Higher Education Commission, Govt. of Pakistan, for financial support of the project.

REFERENCES

Adkins, S.W., G.M. Simpson and J.M. Naylor, 1984. The physiological basis of seed dormancy in *Avena fatua* VI. Respiration and the stimulation of germination by ethanol. *Physiol. Plant.*, 62: 148–52

Association of Official Seed Analysis (AOSA). 1990. Rules for testing seeds. *J. Seed Technol.,* 12: 1–112

Basra, S.M.A., M. Farooq and A. Khaliq, 2003. Comparative study of pre–sowing seed enhancement treatments in indica rice (*Oryza sativa* L.*). Pakistan J. Life and Soc. Sci.,* 1: 5–9

Basra, S.M.A., M. Farooq, K. Hafeez and N. Ahmad, 2004. Osmohardening: A new technique for rice seed invigoration. *Int. Rice Res. Notes,* 29: 80–1

Basra, S.M.A., M. Farooq, R. Tabassum and N. Ahmad, 2005. Physiological and biochemical aspects of seed vigor enhancement treatments in fine rice (*Oryza sativa* L.). *Seed Sci. Technol.,* 33: 623–8

Basra, S.M.A., M.N. Zia, T. Mahmood, I. Afzal and A. Khaliq, 2002. Comparison of different invigoration techniques in wheat (*Triticum aestivum* L.) Seeds. *Pakistan J. Arid. Agric*., 5: 11–6

Bewley, J.D. and M. Black, 1982. *Physiology and biochemistry of seeds in relation to germination,* Vol–II. Viability, dormancy, and environmental control. Springer–Verlag, Berlin.

Bradford, K.J., J.J. Steiner and S.E. Trawatha, 1990. Seed priming influence on germination and emergence of paper seed lots. *Crop Sci.,* 30: 718–21

Brocklehurst, P.A. and J. Dearman, 1983. Interactions between seed priming treatments and nine seed lots of carrot, celery and onion. I. Laboratory germination. *Ann. App. Biol*., 102: 577–84

Burgass, R.W. and A.A. Powell, 1984. Evidence for repair processes in the invigoration of seeds by hydration. *Ann. Bot*., 53: 753–7

Coolbear, P., A. Francis and D. Grierson, 1984. The effect of low temperature pre–sowing treatment on the germination performance and membrane integrity of artificially aged tomato seeds. *J. Exp. Bot.,* 35: 1609–17

Corbineau, F., B. Gouble, S. Lecat and D. Come, 1991. Stimulation of germination of dormant oat (*Avena sativa*.) seeds by ethanol and other alcohols. *Seed Sci. Res.,* 1: 21–8

Ellis, R.A. and E.H. Roberts, 1981. The quantification of ageing and survival in orthodox seeds. *Seed Sci. Technol.,* 9: 373–409

Farooq, M., S.M.A. Basra, K. Hafeez and N. Ahmad, 2005. Thermal hardening: a new seed vigor enhancement tool in rice. *J. Integ. Pl. Biol.,* 47: 187–93

Farooq, M., S.M.A. Basra, K. Hafeez and E.A. Warriach, 2004. The influence of high and low temperature treatments on the seed germination and seedling vigor of coarse and fine rice. *Int. Rice Res. Notes,* 27: 69–71

Farooq, M., S.M.A. Basra and K. Hafeez, 2006. Rice seed invigoration by osmohardening. *Seed Sci. Technol*., 34: 181–7

Fidler, J.C., 1968. The metabolism of acetaldehyde in plant tissues. *J. Exp. Bot.,* 58: 41–51

Heydecker, W. and P. Coolbear, 1977. Seed treatments for improved performance – survey and attempted prognosis. *Seed Sci. Technol*., 5: 353–425

Larondelle, Y., F. Corbineau, M. Dethier, D. Come and H.G. Hers, 1987. Fructose 2, 6–bisphosphate in germinating oat seeds. A biochemical study of seed dormancy. *European J. Bioch.,* 166: 605–10

Lee, S.S., J.H. Kim and S.B. Hong, 1999. Effect of priming and growth regulator treatments of seed on emergence and seedling growth of rice. *Korean J. Crop Sci.,* 44: 134–7

Miyoshi, K. and T. Sato, 1997. The effects of Kinetin and Gibberellin on the germination of dehusked seeds of indica and japonica rice (*Oryza sativa* L.) under anaerobic and aerobic conditions. *Ann. Bot.,* 80: 479–83

Miyoshi, K. and T. Sato, 1997a. The effects of ethanol on the germination of seeds of Japonica and Indica rice (*Oryza sativa* L.) under anaerobic and aerobic conditions. *Ann. Bot*., 79: 391–5

Singh, H. and H.S. Gill, 1988. Effect of Seed treatment with slats on germination and yield of wheat. *Agric. Sci. Digest*, 8: 173–5

Taylor, A.G., P.S. Allen, M.A. Bennett, K.J. Bradford, J.S. Burris and M.K. Misra, 1998. Seed enhancements. *Seed Sci. Res*., 8: 245–56

Taylorson, R.B. and S.B. Hendricks, 1979. Overcoming dormancy in seeds with ethanol and other anesthetics. *Planta,* 145: 507–10

(Received 10 July 2005; Accepted 23 November 2005)