Korean J. Crop Sci. 44(3) : 263~266 (1999)

Optimum Conditions for Tobacco Seed Priming by PEG 6000

Tai-Gi Min*† and Byung-Moon Seo**

ABSTRACT

Tobacco (*Nicotiana tabacum* L. 'KF109') seeds were primed in polyethylene glycol 6000 (PEG 6000) solutions to determine a) what osmotic potential of the solution would be optimal for priming, i.e., critical potential level for preventing germination, and b) what temperature and duration would be the most effective in priming. The germination was completely prevented below -0.8 MPa of PEG 6000, that indicates a optimum water potential for seed priming. Seeds were primed for 0, 1, 2, 3, 5, 10 and 15 days at 15, 20 and 25℃, respectively, under the -0.8 MPa PEG 6000 solution to find out the most effective temperature and duration for priming. The effectiveness of priming, particularly in germination speed, was observed more distinctly when the primed seeds were germinated at 15℃ than 25℃. The greatest reduction of the time to 50% germination (T_{50}) was when the seeds were primed at 25℃. The reduction rate of the T_{50} was rapid when primed from 1 day to 8 days and then slowed down in the seeds primed for longer than 8 days. The time from 10 to 90% germination (T_{10-90}) increased in the primed seeds for longer than 8 days which showed the reversed effect of synchronous germination. However, T_{50} was reduced continuously in the seeds even primed over 8 days. Thus, the optimum condition for tobacco seeds priming with PEG 6000 solution was -0.8 MPa in osmotic potential of the solution at 25℃ for 8 days.

Keywords : tobacco, priming, PEG 6000, osmotic potential, time to germination, synchronous germination. T_{50}, T_{10-90}.

Tobacco seeds are usually sown in the seed bed in late February in greenhouses in Korea, that is under suboptimal temperature conditions for seed germination and seedling growth. It usually takes about 10-15 days to germinate and about 30 days from germination to first transplanting for tobacco seedlings. Careful management of the seed bed is necessary to get good seedling establishment during this period. Therefore, it is very important to enhance the germination speed of tobacco seeds and the seedling establishment during such a suboptimal temperature period.

Seed priming is a technique to treat seeds with low osmotic solutions to improve the rate, speed and uniformity of germination, especially under suboptimal temperatures (Heydecker and Coolbear 1977). Priming techniques rely on the controlled uptake of water to achieve a critical moisture content that activates metabolic activity in a controlled environment (Taylor, 1997). The controlled environmental factors for optimum seed priming are the water potential of priming medium, temperature and duration. Effective priming could be achieved by adjusting the osmotic concentration of the priming solution to a level just enough to prevent germination. Smith and Cobb (1991) reported that priming solutions with the highest osmotic potentials that prevented germination resulted in the greatest reduction of T_{50} in pepper seeds. Bodsworth and Bewley (1981) also reported that the osmotic potential of the priming solution (PEG) was an essential factor of effective priming for several different types of seeds. The water potential of the solution for priming by large molecular weight polymers such as PEG varied by species and ranged from -0.5 to -2.0 MPa (Khan et al., 1980, 1981). Priming temperatures ranged from 10 to 35℃, but 15 to 20℃ were most commonly used (Bradford, 1986). The period of time for the seeds in these conditions might range from less than 1 day to several weeks (Taylor and Harman, 1990).

PEG is commonly used for adjusting osmotic potential because it is inert and highly soluble in water, and gives consistent beneficial effects on seed germination in several species. PEG has widely been used to regulate water potential and it's formulas have been developed to adjust the water potential of a solution to a known concentration and temperature (Michel, 1983). Min (1993) confirmed the effectiveness of tobacco seed priming, but did not verify the optimum conditions for the priming. Thus, the object of this study was to determine the optimum water potential of PEG 6000 solution, temperature and duration for tobacco seed priming and to confirm the effects of the priming in suboptimal temperature conditions.

* College of Natural Resources, Taegu Univ., Kyongsan 712-714, Korea, ** Nongwoo Seed Co., Kimhae 621-810, Korea.
*† Corresponding author : (E-mail) tgmin@biho.taegu.ac.kr (Phone) +82-53-850-6761. Received 30 July, 1999.

MATERIALS AND METHODS

Tobacco seeds of 'KF109' (*Nicotiana tabacum* L.) were used. Different water potentials of PEG 6000 solutions were prepared by Michel's formula, ranged from -0.1 to -1.0 MPa, -0.1 MPa increments (Michel and Kaufmann, 1973). Tobacco seeds, with three replicates of 100 seeds each, were germinated on the blotters in plastic containers (11 × 11 × 4 cm) with lids. The blotters were moistened with the different osmotic potentials of the PEG solutions at 25℃ to determine the critical moisture level just preventing the seed germination, that is known to be an optimum water potential for priming tobacco seeds. Inside the containers, 9 cm petri plates put top down to support the blotters and two edges of the blotters were submerged and continuously moistened with 40 ml of the PEG solution from the bottom of the containers.

Priming treatments were performed under the temperature controlled incubators at 15, 20, and 25℃ and for 1, 2, 3, 5, 10 and 15 days, respectively with -0.8 MPa PEG 6000 solution, that was found to be a optimum water potential (Fig. 1). PEG 6000 was used at concentrations of 241, 251 and 262 g/kg water, providing an osmotic potential of -0.8 MPa at 15, 20 and 25℃, respectively (Michel and Kaufmann, 1973). Batches of 2 g seeds were primed on the blotters in plastic containers (11 × 11 × 4 cm). The blotters were continuously moistened with the PEG solution in the same way as described as the above. The containers were tightly sealed with lids and placed in the temperature controlled incubators under light.

Germination tests were carried out in 15, 20 and 25℃ incubators after priming depending on the AOSA method (1993).

RESULTS AND DISCUSSION

Priming is a technique to hydrate seeds under controlled conditions and prevent the completion of germination. The first step for priming is to find a critical water level of the priming medium to prevent seed germination. The correlation was observed between the reduction of germination and the low osmotic potential of the priming solution (PEG) (Fig. 1). The germination rate of tobacco seeds planted on the blotters that were moistened with different concentrations of PEG solutions was continuously reduced depending on the reduced osmotic potential of the PEG solutions and finally germination was completely prevented at -0.8 MPa PEG solution. Thus, the critical level of the osmotic potential of PEG solution preventing germination was -0.8 MPa and the solution was used in all treatments of tobacco seed priming.

The temperature during the priming greatly affected the germination speed of the tobacco seeds after priming (Fig. 2). The fastest speed of germination was obtained when primed at 25℃ rather than those primed at 20 or 15℃. Though the physiological basis of this result is unknown, greater metabolic activity seems to occur in seeds during the priming at higher temperatures. The greater effectiveness of priming was observed in both germination rate and speed when germinated at 15℃, suboptimal temperature for tobacco seed germination (Fig. 2, B), compared to 25℃, that is generally considered to be an optimum temperature for germination (Fig. 2, A), i.e., when the primed seeds were germinated at 25℃, the germination was faster only about 1 day than control (non-primed). When the seeds were germinated at 15℃, the germination was faster about 7 days than the control.

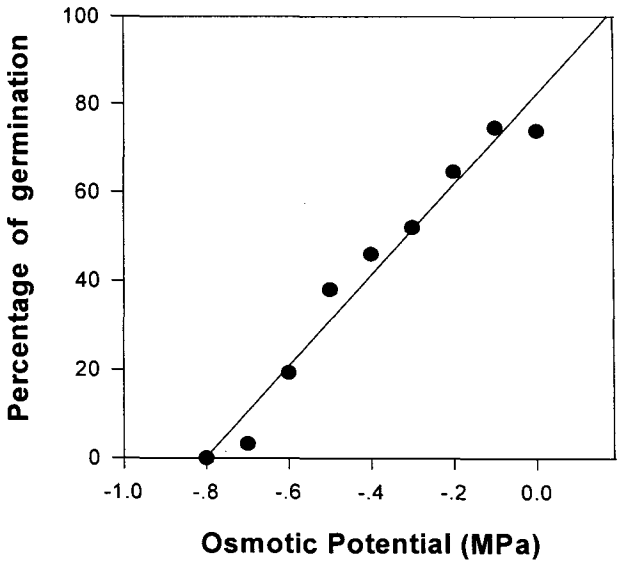

Fig. 1. Correlation between the germination of tobacco seeds and the osmotic potentials of PEG 6000 solution (12 days after planting at 25℃).

The germination tests showed that the duration of priming was also important; i.e., the longer period primed, the faster germination occurred (Fig. 3). Priming for 15 days was the most effective in germination speed than priming for less than 15 days. The greater effectiveness of priming was observed also when the seeds were germinated at 15℃ than 25℃, i.e., the germination of the non-primed seeds (0 day) were retarded about 1 day compared to those primed for 15 days and germinated at 25℃(Fig. 3, A), while germination was retarded about 7 days when the seeds were germinated at 15℃(Fig. 3, B). Smith and Cobb (1991) reported also that the longer period of priming had greater priming effects.

Priming of seeds in low osmotic solutions of PEG

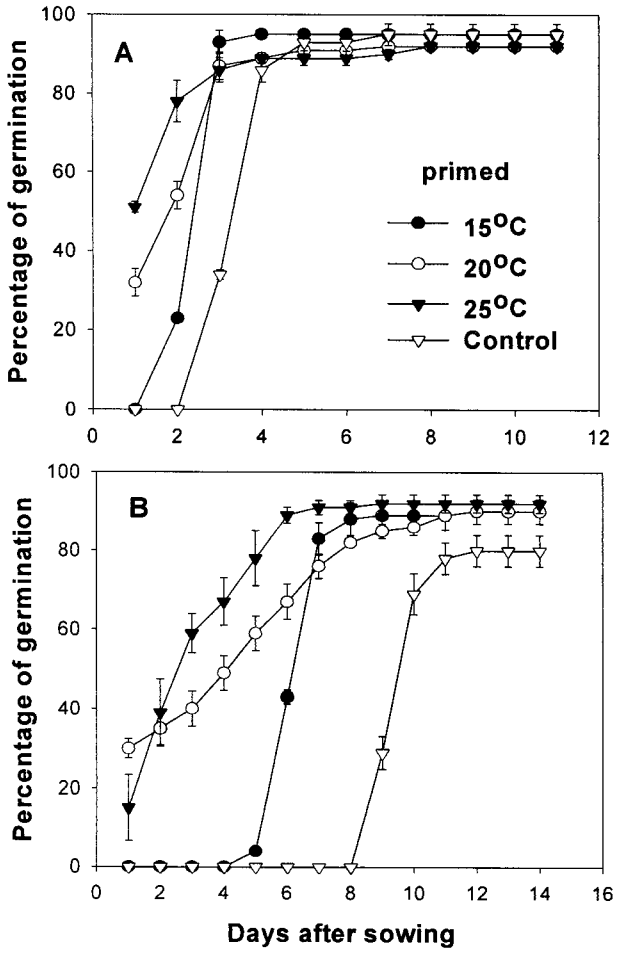

Fig. 2. Effect of priming temperatures on the germination of tobacco seeds primed for 15 days and germinated at 25℃ (A) and 15℃ (B). Bars represent standard errors.

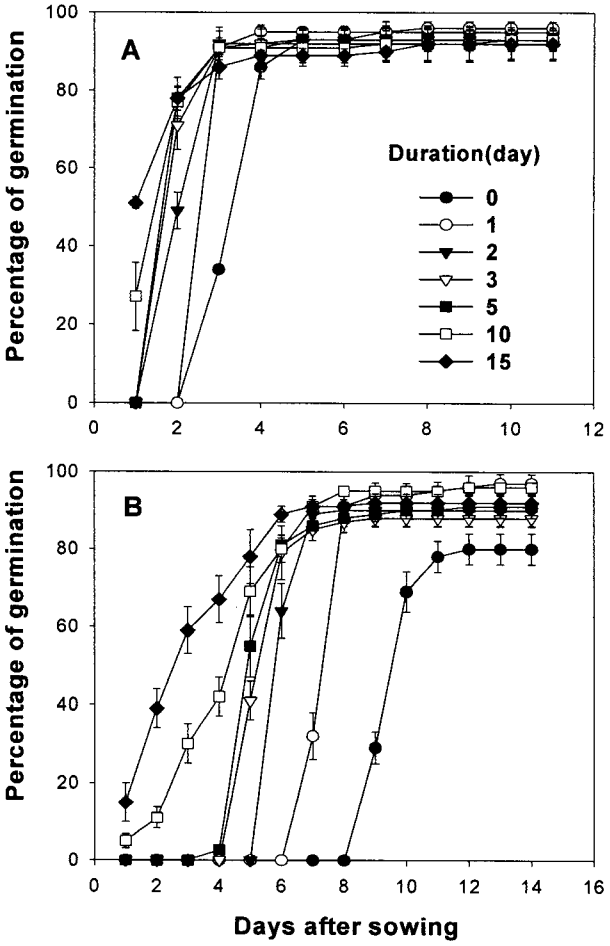

Fig. 3. Effect of priming durations(days) on the germination of tobacco seeds primed at 25℃ and germinated at 25℃ (A) and 15℃ (B). Bars represent standard errors.

has been shown to improve the germination speed of several seeds, such as pepper, tomato, sweet corn, snap bean, table beet, sugar beet and watermelon at suboptimal temperatures in laboratory and/or field planting(Khan, et. al., 1995; Khan, 1992). Primed tobacco seeds also showed more effective germination speed at suboptimal low temperature (15℃) than normal conditions (25℃) in this study.

Relationships between T_{50} (time to 50% germination) and $T_{10\ 90}$ (time from 10 to 90% germination) in different priming temperatures and durations when the seeds were germinated at 15℃ are shown in Fig. 4. T_{50} was reduced as the priming duration increased, regardless of any priming temperature. Priming at 25℃ was more effective than at 15℃ or 20℃ to decrease the germination time. T_{50s} were decreased rapidly until 8 days priming but showed less effectiveness in priming for longer than 8 days. In contrast to T_{50}, $T_{10\text{-}90}$ were increased according to increase of the priming durations, that were represented as less synchronous germination if the longer periods of priming were performed over any limitations. $T_{10\text{-}90}$ increased rapidly after about 6-8 days priming. This indicates that if tobacco seeds were primed longer than 6-8 days, the beneficial effect of synchronous germination was rather reversed. The phenomenon of the reversed beneficial effects of priming is referred to as "over priming" (Ely and Heydecker, 1981). Thus, priming for 8 days satisfied both, time to germination(T_{50}) and the synchronous germination($T_{10\text{-}90}$), when the primed seeds were germinated at 15℃, a suboptimal temperature for germination.

Conclusively, the optimum conditions for tobacco seed priming by PEG 6000 solution were: 1) the critical osmotic potential of the solution was -0.8 MPa. 2) the most effective temperature for priming to

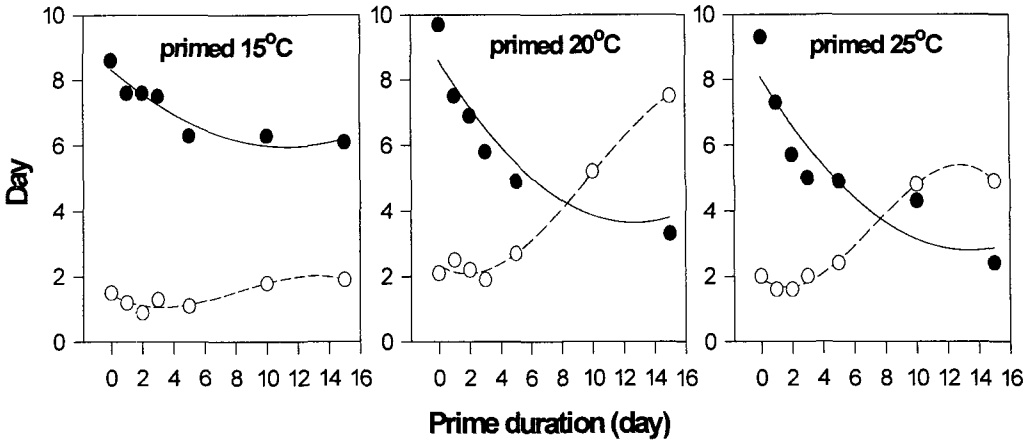

Fig. 4. Relationship between T_{50} (●) and T_{10-90} (○) depending on the priming temperatures and durations for tobacco seeds when germinated at 15℃. (T_{50}=time to 50% germination, T_{10-90}= time from 10 to 90% germination).

enhance germination time was 25℃. 3) the optimum duration of priming was 8 days, considering the beneficial effects of the germination speed and uniformity. After all, this study for tobacco seed priming indicated that effective priming was strongly dependent on both the priming temperature and the duration of the treatments, and the priming effect was more distinct at suboptimal temperatures.

ACKNOWLEDGEMENTS

This paper was partly supported by "The Research and Development Promotion Center for Agriculture and Forestry".

REFERENCES

Association of Official Seed Analysts(AOSA). 1993. Rules for testing seeds. Journal of Seed Technology. 16(3).

Bodsworth, S. and J. D. Bewley. 1981. Osmotic priming of crop species with polyethylene glycol as a means of enhancing early and synchronous germination at cool temperatures. Can. J. Bot. 59: 672-676.

Bradford, K. J. 1986. Manipulation of seed water relations via osmotic priming to improve germination under stress conditions. HortScience 21(5): 1105-1112.

Ely, P. R. and W. Heydecker. 1981. Fast germination of parsley seeds. Scientia Hort. 15: 127-136.

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

Khan, A. A., N. H. Peck and Samimy C. 1980/1981. Seed osmo-conditioning: physiological and biochemical changes. Israel Journal of Botany 29: 133-144.

Khan, A. A. 1992. Preplant physiological seed conditioning. Horti-cultural Reviews 13: 131-181.

Khan, A. A., S. Ilyas and W. Ptasznik. 1995. Integrating low water potential seed hydration with other treatments to improve cold tolerance. Annals of Botany 75: 13-19.

Michel, B. E. and M. R. Kaufmann. 1973. The osmotic potential of polyethylene glycol 6000. Plant Physiology 51: 914-916.

Michel, B. E. 1983. Evaluation of the water potentials of solutions of polyethylene glycol 8000 both in the absence and presence of other solutes. Plant Physiol. 72: 66-70.

Min, T. G. 1993. Effects of pre-sowing treatments of tobacco seed on germination and seedling uniformity. Korean J. Crop Science. 38(6): 507-512.

Smith, P. T. and B. G. Cobb. 1991. *Accelerated germination of* pepper seed by priming with salt solutions and water. HortScience 26(4): 417-419.

Taylor, A. G. 1997. Seed Storage, Germination and Quality. (In: The Physiology of Vegetable Crops, ed by Wien H. C.) pp. 1-36.

Taylor, A. G. and G. E. Harman. 1990. Concepts and technologies *of selected seed treatments*. Annual Review of Phytopathology 28: 32-339.