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Priming Effects on Germination of Aged Tobacco Seeds

Tai-Gi Min*†

**College of Natural Resources, Taegu University, Kyungsan, Kyungbuk 712-714, Korea*

ABSTRACT: Tobacco (*Nicotiana tabacum* L. cv KF109) seeds were artificially aged in a controlled environment of 45°C and 80% relative humidity condition for different duration up to 14 days before priming in polyethylene glycol 6000 solution of equivalent osmotic potential of -0.8 MPa for 8 days at 25°C. The seeds aged only and primed after aging were germinated at 15°C and 25°C to observe the priming effects on the germination of aged seeds at different temperature. The germination percentage of the aged seeds was rapidly dropped starting from 8 days of aging and mean germination time (T_{50}) was greatly increased, particularly in germination at 15°C. The germination capacity was greatly restored in the primed seeds after aging, particularly in the seeds of longer aging and germinating at 15°C.

Keywords **:** germination, polyethylene glycol. seed age, tobacco seed, priming, mean germination time

The promotive effects on germination of many species due to priming treatment have been well documented (Heydecker and Coolbear, 1977; Brocklehurst and Dearman, 1983). However, there was some controversy regarding the merits and effects of seed priming with respect to existing seed vigor. Matthews and Powell (1986) reported that presowing treatment of seeds such as priming could be of little benefit to low-vigor seed. They further suggested that the major benefit of seed treatments might lie in the direct influence they had in ensuring that only highly germinable and vigorous seedlots were given expensive treatments. Others have found, however, that low-quality seedlots derived greater benefit from presowing treatments. For example, preplant priming improved the performance of good and poor quality carrot seeds, but the improvements were greater in the poor seeds than in the more vigorous seeds (Khan, Abawi, and Maquire, 1992).

Priming has been known to accelerate germination in many types of aged seeds. An accelerated aging has been used as a means of circumventing the need for experiment analyses that would otherwise extend over many years of storage. Savino *et al.* (I979), for example, reported that aged pea seeds imbibed for I8 hours at 20°C, followed by drying, led to a significant increase in radicle length in subsequent germination assays. Goldsworthy *et al.* (1982) suggested that soaking for 30 minutes or less is sufficient to reinvigorate the aged wheat seeds. General experience with priming treatments has been that the vigor of old seeds can be enhanced. Sanches and Miguel (1983), however, argued that hydration-dependent repair processes could restore viability to aged embryos of *Datura ferox.* Thus, the effects and benefits of priming would be various depending on species and seed quality. The objective of this study was to observe how much priming effects could be expected from tobacco seeds artificially aged for different duration in different germination temperature conditions.

MATERIALS AND METHODS

Tobacco seeds, cv 'KF109' were used for this experiment. For artificially aging treatment, each batch of 2 g seed was placed on wire mesh trays in the plastic containers (11×11×4 cm) with controlled humidity of 80% and the containers put into 45°C incubator for 2, 4, 6, 8, 10, 12 and 14 days. The relative humidity (RH) inside the plastic containers was controlled by putting 40 ml of the mixtures of the glycerol and water in the bottom of the containers according to Forney and Brandl (1992). Each batch of the aged seeds were taken out from the incubator at two days interval and dried back to their original weight in forced air at 20°C. The seeds dried after ageing were primed in polyethylene glycol 6000 (PEG) solution of equivalent osmotic potential of -0.8 MPa (262 g/kg water) under fluorescent light at 25°C for 8 days and stored at 4°C until germination test (Min and Seo, 1999). Germination test were conducted at 15 and 25°C by the AOSA rules (1993). Count of the number of germinated seeds were made at 24-hour intervals for 20 days at 15°C, and 12 days at 25°C respectively, that is, until no further germination was observed ether at 15 or 25°C.

The mean time to germination(T50) was calculated from the following equation:

$$T_{50} = \Sigma TiXi/\Sigma Xi$$

where Xi is the number of newly germinated seeds at time Ti. The time used was the midpoint of the interval since the previous count.

†Corresponding author: (Phone) +82-53-850-6761 (E-mail) tgmin@biho.taegu.ac.kr <Received August 21, 2001>

RESULTS AND DISCUSSION

There were little loss of germination capacity until 8 days aging at 45°C, but the germination capacity was markedly dropped from 87% in 8 days ageing to 8% in 14 days aging (Fig. 1).

There was little reduction in percent germination of the seeds aged only and primed after aging with increasing the duration of aging until 8 days when germinated at 25, but in the seeds aged for 14 days, percent germination was significantly reduced not only in priming after aging, but also in ageing only compared with control (Fig. 2A). When germinated at 15°C, there was a progressive declining in percent germination on the seeds primed after aging with increasing duration of aging (Fig. 2B). Particularly, the seeds aged for 14 days were not germinated at all until 14 days after sowing, whereas the seeds primed after aging began to germinate from 3 days after sowing and reached about 50% of final germination. Prolonged aging resulted in progressive increase in mean germination time (T_{50}) for both seeds aged and primed after aging compared with control (0 day aging) when germinated at 15°C and 25°C. T_{50} of the seed aged only was much longer than that of the seeds primed after aging in all aging durations and germination temperatures. However, when the seeds were germinated at 15°C, priming of the aged seeds tremendously decreased in T_{50}s compared with germination at 25°C (Fig. 3). That means that the priming effect was greater in the suboptimal temperature of 15°C than in the optimal temperature of 25°C for germination.

Final germination was generally much higher in the seeds primed after aging than the seeds aged only at both germination temperature of 25°C and 15°C but the difference of the germination percentage between the seeds aged only and primed after aging was greater when germinated at 15°C than 25°C. The priming effect on the final germination of the

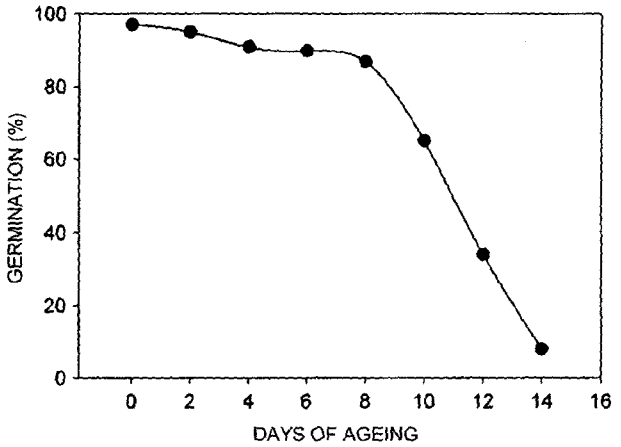

Fig. 1. Changes of germination rate of tobacco seeds by aging treatment (6 days after sowing at 25°C).

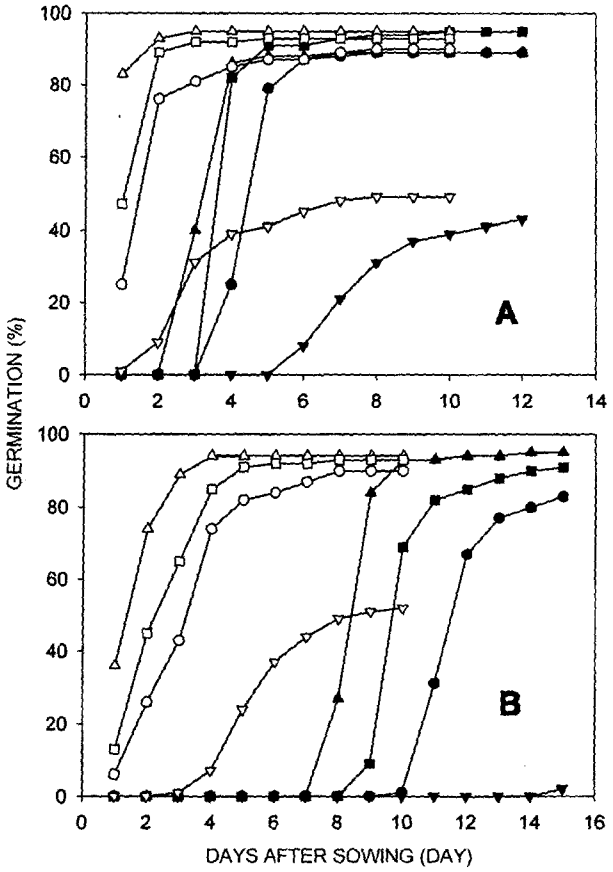

Fig. 2. Effects of priming after aging, and aging only on germination at 25°C(A) and 15°C(B). Open symbols represent aging and priming, and closed symbols represent aging only. (△▲ , control (0 day aging+no priming); □■ , seed primed after aging and, aged only for 4 days; ○● , seed primed after aging, and aged only for 8 days; ▽▼ , seed primed after aging, and aged only for 14 days).

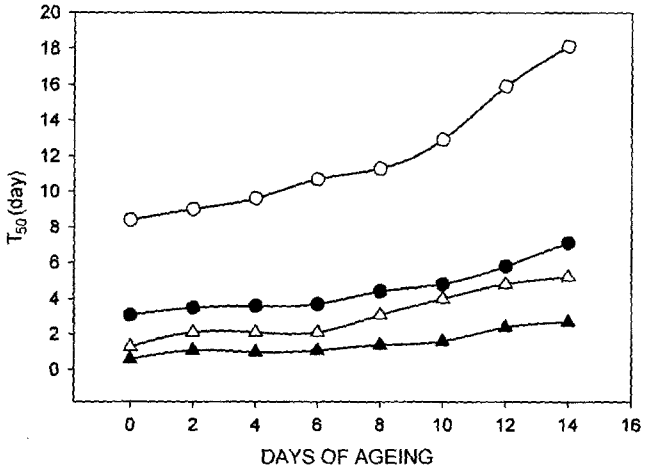

Fig. 3. Effects of priming after aging on mean germination time (T_{50}). (▲, seed primed after aging, and germinated at 25°C; ● , seed aged only, and germinated at 25°C; △ , seed primed after aging, and germinated at 15°C; ○ , seed aged only, and germinated at 15°C).

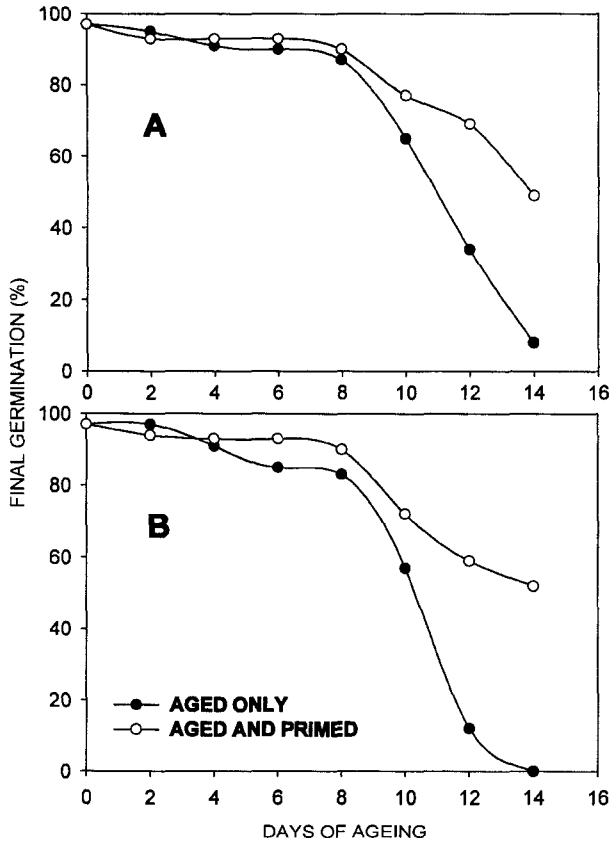

Fig. 4. The effects of aging only, and priming after aging treatment on final germination of tobacco seeds germinated at 25°C(A) and 15°C(B).

aged seeds was greater with increasing length of aging, particularly, when germinated at 15°C (Fig. 4). The priming effect was not great until 8 days of aging but increased from the seeds aged longer than 8 days.

Presowing treatment of seed in low water potential solutions of PEG and salts has been shown to improve the performance of a number of seeds at supoptimal temperatures in laboratory or field planting (Khan, 1992). Khan (1995) stated that the cold tolerance effect of priming could be derived from the activation or synthesis of large number of enzymes and greater mobilization of storage reserves before planting. Berjak and Villers (1972) suggested the experimentally-accelerated process of aging caused membrane aberrations which appeared when the cells first imbibed water, and also caused a delay in the onset of the developmental and metabolic events leading to germination, but there was much evidence of repair of this damage at later stages of germination; a temporarily increased rate of production of mitochondria, RNA, protein, and DNA replication. In view of these results, aged tobacco seeds needed longer time for germination due to the temporary impairment of metabolic processes caused by membrane aberrations and by the necessity for repair mechanisms to operate in order to compensate for the damage. However, the recovery of germinability in primed tobacco seeds after aging indicated that the repair process might be completed during the priming. In addition, the distinctive priming effect on the germination of aged tobacco seeds at suboptimal temperature might be attributed to the fact that the seeds just aged took longer time for repair of damages at suboptimal temperature than at optimal temperature.

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