

ROLE OF GA₃ AND KNO₃ IN IMPROVING THE FREQUENCY OF SEED GERMINATION IN *PLANTAGO LANCEOLATA* L.

ERCUMENT OSMAN SARIHAN, ARIF IPEK, KHALID MAHMOOD
KHAWAR*, MEHMET ATAK AND BILAL GURBUZ

¹*Department of Field Crops,
Faculty of Agriculture, University of Ankara, 06110,
Diskapi, Ankara, Turkey*

Abstract

Seeds of *Plantago lanceolata* L., obtained from crops of 1992- 2001 were treated with 100, 200 and 400 ppm of GA₃ and 1000, 2000 and 4000 ppm of KNO₃ by incubating them at 20 °C under 16 h light photoperiod, along with control. Highest germination of 97% was obtained from the seed of the crop of 2001, which decreased to 78.8% in the seeds of the crop of 1992 in untreated control. It was found that age plays a significant role in the decline of frequency of seed germination, which was effectively improved with the use of GA₃ and KNO₃.

Introduction

Plantago lanceolata L., of the family *Plantaginaceae* is a perennial herb that grows upto 70 cm, has lanceolate leaves, deeply venated whitish flowers and has egg shaped fruit capsules. The plant dies to the ground each winter and sprouts anew from its fibrous tap root around mid-spring. The plant prefers saline and wet habitats upto 2200 meters. The leaves may grow up to about 15 cm long and 10 cm wide. (McCloud & Berenbaum, 2000). It is widely found in Turkey and most of the European countries (Davis, 1975). In Germany, France and Belgium it is cultivated on limited areas for forage (Karl, 1982). Recently, domesticated cultivars have been selected for pasture production in New Zealand (Sanderson & Elwinger, 2000a).

Medicinally, *P. lanceolata* is astringent, demulcent, emollient, cooling, vulnerary, expectorant, antimicrobial, antiviral, antitoxin and diuretic (Leung & Foster 1996) and is effectively used in treatment of tumors (Fracoise *et al.*, 1998). Traditionally, its leaf tea is used for diarrhoea, coughs and dysentery. Leaves are also applied to blisters, sores, swelling and insect stings. The mucilage from any plantain seeds may decrease cholesterol level in blood (Foster & Duke, 1990).

In practice, seed germination rate is very important for successful growing of any crop. Most crop seeds are probably dead after 25 years, even under favorable storage conditions (Martin & Leonard, 1967). However, there is a correlation between longevity of seed and storage conditions (Hughes & Metcalfe, 1972). The old seeds lose their germination ability, with the passage of time. However, it is possible to extend seed germination ability by using some seed germination promoters. Gibberellins and potassium nitrate (KNO₃) are used for breaking seed dormancy and promoting seed germination. Gibberellins are most directly implicated in the control and promotion of germination. Gibberellins promote growth by increasing plasticity of the cell wall followed by the hydrolysis of starch to sugar which reduces the potential in the cell,

*Institute of Biotechnology, Department of Field Crops, Faculty of Agriculture, University of Ankara, Dışkapı, Ankara, Turkey (Corr. author Tel: +90-312-5961286, +90 312 5961467-1467, Fax: +90-312-3179815, Email: kmkhawar@gmail.com, gurbuz@agri.ankara.edu.tr

resulting in the entry of water into the cell causing elongation (Arteca, 1996). There is evidence that natural gibberellin like substances appear during successive stages of after-ripening and germination. Gibberellins play an important role in the regulation of seed germination. KNO_3 is most widely used chemical for promoting germination. Solutions of 0.1 to 0.2 % KNO_3 are common in routine germination testing and are recommended by the Association of Official Seed Analysts and the International Seed Testing Association for germination tests of many species (Copeland & McDonald, 1995; Basra, 1994). The objective of the study was to establish a relationship between seed age and germination percentage in *Plantago lanceolata* with emphasis on simplicity and practicability. The protocol determines and evaluates reduction in germination due to metabolic changes affecting germination and offers effective, cheap and simple procedure using Gibberellic acid and KNO_3 to improve germination of long stored *P. lanceolata* seeds stored at room temperature.

Materials and Methods

Seed collection and storage

Seeds of *P. lanceolata* from the crops of 1991 to 1992, were collected from the nursery of perennial medicinal and aromatic plants of the department of Field Crops, Faculty of Agriculture, University of Ankara, Turkey and stored at room temperature.

GA_3 and KNO_3 treatment and germination

Batches of 100 seeds each contained in Petri dishes (100 x 10 mm) replicated four times, were treated with three doses each of 100, 200 and 400 ppm of GA_3 for 6 hours and 1000, 2000 and 4000 ppm of KNO_3 for 6 hours and were germinated in between moistened filter papers. The filter papers were humidified regularly with demineralized water. Seeds without any treatment were used as the control. They were incubated in a growth chamber (Sanyo MLR 350 H) at 20 °C under 16 h light photoperiod. The seed were scored for final germination after 21 days according to ISTA, (Anon., 1993).

Significance was determined by analysis of variance (ANOVA) using SPSS for windows (v. 11. SPSS Inc USA) and differences between the means of the seed age and the treatments were compared by Duncan's multiple range test ($P < 0.05$). Data given in percentages were subjected to arcsine (\sqrt{X}) transformation before statistical analysis (Snedecor & Cochran 1967).

Results and Discussion

Frequency of seed germination of untreated seeds

The results indicated no seed dormancy but a sharp reduction in seed germination frequency as influenced by the seed age (Table 1). The seed germination reduced gradually with age with range of 78.8 to 97%. The data further indicated statistically similar frequency of seed germination from the seeds of 1999 to 2001; but different from those of 1995 to 1998, 1993-94 and 1992 ($p < 0.05$). The statistical data showed a sharp decline of germination within each group such that the highest seed germination of 97% from the seeds of 2001 reduced to 78.8%; the lowest from the seeds of 1992.

Table 1. Effects of GA₃ and KNO₃ treatments on seed germination of *Plantago lanceolata* belonging to the crops of 1991–2001.

Years	Control	Treatments					
		GA ₃ 100 ppm	GA ₃ 200 ppm	GA ₃ 400 ppm	KNO ₃ 1000 ppm	KNO ₃ 2000 ppm	KNO ₃ 4000 ppm
2001	97.0 a ^{1*} ± 0.408	92.8 ab2 ± 1.887	92.8 a2 ± 1.377	92.0ab2 ± 0.408	92.5 ab2 ± 0.866	93.3 abc2 ± 1.377	94.8 a12 ± 1.315
2000	95.5 a1 ± 0.289	94.8 a1 ± 0.479	93.3 a1 ± 0.250	93.0ab1 ± 0.707	93.5 ab1 ± 0.289	93.0 abc1 ± 0.408	92.8 ab1 ± 0.479
1999	94.3 a1 ± 0.750	94.3 ab1 ± 0.957	92.8 a1 ± 0.250	94.0a1 ± 0.577	91.3 bc12 ± 1.109	92.3 bc12 ± 1.315	90.5 bc 2 ± 0.866
1998	88.5 b2 ± 1.041	92.8 ab1 ± 1.377	92.5 a1 ± 0.957	90.3 ab12 ± 0.854	93.5 ab1 ± 1.041	94.3 abc1 ± 0.479	91.3 abc12 ± 0.854
1997	85.3 bc 2 ± 0.629	93.0 ab1 ± 1.155	93.0 a1 ± 1.080	91.3 ab1 ± 1.315	92.8 ab1 ± 1.797	92.0 bc1 ± 1.080	93.8 ab1 ± 1.315
1996	86.0 bc2 ± 1.291	94.3 ab1 ± 0.479	92.0 a1 ± 0.408	93.3 ab1 ± 0.479	95.3 a1 ± 1.377	96.0 a1 ± 0.408	94.0 ab1 ± 1.291
1995	85.0 bc2 ± 1.225	93.5 ab1 ± 1.893	92.0 a1 ± 1.225	91.0 ab1 ± 1.354	93.8 ab1 ± 1.493	94.8 ab1 ± 1.109	93.5 ab1 ± 1.190
1994	84.6 c2 ± 0.479	90.8 b1 ± 0.854	91.3 a1 ± 0.750	92.3 ab1 ± 0.854	88.8 c1 ± 2.097	91.0 c1 ± 1.472	91.8ab1 ± 1.250
1993	83.5 c2 ± 0.866	90.5 b1 ± 1.555	92.0 a1 ± 1.080	93.8 a1 ± 0.629	91.0 bc1 ± 1.080	90.8 c1 ± 1.315	93.0 ab1 ± 1.080
1992	78.8 d2 ± 0.629	86.3 c1 ± 1.931	87.5 b1 ± 2.723	89.8 b1 ± 1.652	84.3 d2 ± 1.315	85.0 d2 ± 1.225	88.3 c1 ± 1.185

*Letters show vertical groups, figures show horizontal groups in each treatment.

**Mean value followed by different letters and figures are significant at the 0.05 level.

Effects of GA₃ treatment on germination

A significant improvement in GA₃ treated seeds over untreated seeds was observed with a range of 86.3-94.8 % with 100 ppm, 87.5-93.3 % with 200 ppm and 89.8-94.0 % with 400 ppm treatment (Table 1). A visible improvement in frequency of seed germination of GA₃ treated seeds over control was observed on seeds belonging to the crops of 1992-1998. However, the GA₃ treatment in any concentration resulted in decline in the seed germination from the crops of 1999 to 2001. A 100 ppm GA₃ treatment improved seed germination of the seeds of the crops of 1995- 1998 and 400 ppm GA₃ treatment effectively improved seed germination of the crops of 1992 to 1994.

Effect of KNO₃ treatments on germination

The treated seeds showed germination frequency range of 84.3-95.3, 85.0-96.0 and 88.3-94.8 % from 1000 ppm, 2000 ppm and 4000 ppm KNO₃ treatment, respectively. Like GA₃, any of the KNO₃ treatment also was ineffective to improve germination in the seeds of crops belonging to 1999 to 2001 over control (Table 1). However, all doses of KNO₃ improved germination frequency over control, on the seeds belonging to the crops of 1992-1998 periods. Although improvement in the germination of KNO₃ treated seeds was not consistent yet 2000 ppm KNO₃ treated seeds showed the highest germination percentage over control in general.

GA₃ and KNO₃ treatments showing decrease in germination in seeds of 1999-2001 may be due to endogenous or exogenous plant growth substances based primarily on corresponding shifts in threshold water potential for radicle emergence in line with the results of Ni & Bradford (1993). The progressing seed age resulted in corresponding decrease in the rate of germination especially from seeds belonging to the crops of 1991 to 1998, where less than 90 percent germination was very evident in line with the results of Tosserams *et al.*, (1997) and Davis & Robinson (2001), who observed reduction in seed germination with age. However, a decline in germination in aged seeds reported here was improved using either GA₃ or KNO₃ supporting the results of Saruhan *et al.*, (2002), who found that GA₃ application promoted the seed germination in *P. major*.

Sanderson & Elwinger (2000b), reported that the depth of planting was effective on seedling emergence and recorded 62 % of emergence value in this species. Beside the planting depth, the seed with high germination rate is necessary for good seedling emergence under field conditions. In conclusion, GA₃ treatment seems preferable over KNO₃ to obtain high germination in *P. lanceolata*.

Acknowledgements

We would like to thank Prof. Dr. Sebahattin Ozcan, Prof. Dr. Neset Arslan and Prof. Dr. Ibrahim Demir for their kind help in providing the material used in the study.

References

- Anonymous. 1993. ISTA–International Rules for Seed Testing, supplement 21.
- Arteca, R. N. 1996. Plant Growth Substances Principles and Applications. Chapter 3: Chemistry, Biological Effects and Mechanism of Action, p: 66. Chapman & Hall 115 Fifty Avenue New York, NY 10003.
- Basra, A. S. 1994. *Seed quality basic mechanism and agricultural implications*. Food Products Press. p: 386.
- Copeland, L.O. and M.B. McDonald. 1995. *Principles of seed science and technology*. Chapman and Hall, 3. edition, p: 408.
- Davis PH 1975 *Flora of Turkey and The East Aegean Islands*. Vol. 5. Edinburgh University Press.
- Davis, M.L. and D.L. Robinson. 2001. Biological effects of volatile emissions from cut grass. Biology Department, Bellarmine College, Louisville, KY, 40205. (<http://cas.bellarmine.edu/tietjen/PPT/Ecology/>)
- Foster, S. and J.A. Duke. 1990. *Medicinal Plants*. The Peterson Field Guide Series, Houghton Mifflin Company, 366 p, New York.
- Fraçoise F, A., Gargadennec, A. Gueiffier, J.L. Roussel and C. Andary. 1998. Effects of Cinnamic acid on polyphenol production in *Plantago lanceolata*. *Phytochem.* 49: 697-702.
- Hughes, H.D. and D.S. Metcalfe. 1972. *Crop production*. Macmillan publishing Co., 627 p, New York.
- Karl, E. 1982. *Arznei-und Gewürzpflanzen*. Wissenschaftliche Verlag-Gesellschaft, 221 p, Stuttgart.
- Leung, A.Y. and S. Foster. 1996. *Encyclopedia of Common Natural Ingredients Used in Food, Drugs, and Cosmetics*. 2nd ed. New York, John Wiley & Sons.
- Martin, J.H. and W.H. Leonard. 1967. *Principles of field crops production*. The Macmillan Company, 1044 p, New York..
- McCloud, E.S. and M.R. Berenbaum, 2000. Effects of spring and summer levels of UV-B radiation on the growth and reproduction of a temperate perennial forb. *Plant Ecol.*, 146: 61-6.

- Ni, B.R. and K.J. Bradford, 1993. Germination and dormancy of ascorbic acid and gibberellin deficient tomato (*Lycopersicon esculantum*) seeds. *Plant Physiol.*, 101: 607-17.
- Sanderson, M.A. and G.F. Elwinger. 2000a. Seedling development of chicory and plantain. *Agronomy J*, 92:69-74.
- Sanderson, M.A. and G.F. Elwinger. 2000b. Chicory and English plantain seedling emergence at different planting depths. *Agronomy J.*, 92:1206-10.
- Saruhan, N., A. Kadioğlu. and N. Durmuş. 2002. Alleviation of seed dormancy in *Plantago major*. *Israel J. Plant Sci.*, 50: 177-9.
- Snedecor G.W. and W.G. Cochran. 1967. *Statistical Methods*. The Iowa State University Press. Iowa, USA,
- Tosserams, M., E. Bolink and J. Rozema. 1997. The effect of enhanced ultraviolet-β radiation on germination and seedling development of plant species occurring in a dune grassland ecosystem. *Plant Ecology*, 128: 139-147.

(Received for publication 25 March 2003)