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EFFECT OF PRE-SOWING HARDENING TREATMENTS ON GERMINATION, RELATIVE GROWTH RATE AND YIELD OF *PENNISETUM AMERICANUM* (L) AND *S. BICOLOR* (MOENCH)

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ABSTRACT
The effects of pre-sowing hardening treatment using 1 and 2 cycles of 1% calcium chloride, 50ppm ascorbic acid and 200ppm indole-3-butyric acid on embryo enlargement, relative growth rate and yields of Pennisetum americanum (cvs, ex Bornu, hairy compositae) and Sorghum bicolor (cvs, ksv11 and ksv12) were investigated. Seeds of the 2 cultivars of P. americanum and S. bicolor hardened with 2 cycles of water were employed as the controls. The 1 and 2 cycles of hardening treatments increased plumule and radicle lengths and relative growth rate of all the cultivars of P. americanum and S. bicolor with increases obtained for 2 cycles of hardening treatments being statistically significant. Yields of plants of 1 and 2 cycles ascorbic acid, calcium chloride and indole-3-butyric acid treated seeds were significantly higher than those of the control plants. For all the cultivars of P. americanum and S. bicolor, 2 cycles of hardening treatments were generally better than 1 cycle of hardening treatments because they induced greater plumule and radicle growth, relative growth rate and yield. Out of the 4 hardening agents used in the study, indole-3-butyric acid stimulated the greatest plumule and radical lengths, relative growth and yield and was as such the best pre-sowing hardening agent. It was followed by ascorbic acid while water was the poorest.

Keywords: Pre-sowing, hardening, germination, yield, pennistum americanum, Sorghum, bicolor

INTRODUCTION

The quantity and quality of plants' yields are affected by two factors. One is the availability of a suitable environment for crop establishment and growth while the other is the genetical, physiological and physical nature of seeds (Marthanova, 1962). One method of physiologically improving the quality of seeds prior to planting is by pre-sowing hardening treatments which employ repeated cycles of "soak and dry" treatments of seeds in water as well as in solutions of salts, phytohormones, vitamins and amino acids (Genkel and Tichomirov, 1981).

Pre-sowing hardening treatments have been found to stimulate extensive physiological changes in seeds which resulted in increases in germination (Marthanova, 1962; Bleak and Keller, 1972), plant growth and yield (Austin et al., 1969; Lush et al., 1981; Khan and Chatterjee, 1981). Pre-sowing hardening treatments were also found to cause resistance to drought and heat (May et al., 1962; Henckel, 1964) and control of physiological deterioration during storage (Basu and Nilanjana, 1979; Savino et al., 1979; Basu and Prativapal, 1980). Moreover, pre-sowing hardening treatments have been observed to induce greater amylase and proteinase activities in oats (Drennan and Berrie, 1962; Berrie and Drennan, 1971), increased sugar and protein contents of wheat and sorghum (Balasubramanian, 1976; Genkel and Tikhomirov, 1981) as well as increased heat tolerance of sugar-cane setts (Mohandas and Naidu, 1984).

Kadiri (1990) observed that presowing hardening treatments of sorghum bicolor with amino acids led to increases in photosynthetic and respiratory rates. Ajiboye, Ebofin, Atayese, Adedipe and Agboola (2007) using presowing treatments of soaking seeds in concentrated sulphuric acid for 5-15 minutes were able to terminate the dormancy in *Dialium guineensis* and *Prosopis africana*. Ibrahim and Nwobosi (1986) had earlier observed that presowing treatment accelerated germination in Teak and Gmelina. Akhtar, Ibrar and Aman (2008) observed that soaking of seed of *Spinacia oleracea* in gibberellic acid enhanced the seed germination and seedling growth.

The present study was undertaken in order to compare the effects of dilute solutions of ascorbic acid, calcium chloride and indole-3-butyric acid to that of water as pre-sowing hardening treatments of *Pennisetum americanum* L. cvs. Ex. Bornu and hairy compositae and *Sorghum bicolor* L. cvs. Ksv11 and ksv12.

MATERIALS AND METHODS

Six months old seeds of *P. americanum* (cvs. Ex. Bornu, hairy compositae) and Sorghum bicolor (cvs. ksv11, ksv12) were obtained from Institute of Agricultural Research, Ahmadu Bello University, Zaria. The seeds were kept in a cold room at 10^oC pending usage.

Pre-sowing hardening treatments of seeds: 500 seeds of each cultivar type were soaked in 200ml of each of the following solutions: distilled water, 50ppm ascorbic acid, 1% calcium chloride ($CaCl_2$) and 200ppm indole-3-butyric acid (IBA) for 4 hours at 28 ± 2^oC to complete 1 cycle of seed treatment. Seed treatments of 2 cycles were obtained by repeating the procedure for 1 cycle treatment twice. Seeds given 2 cycles of water treatments were employed as the controls.

Effect of hardening treatments of embryo growth: Twenty five (25) seeds of each treatment and control were sown in petri dishes in six replicates per treatment at 28 ± 2^oC on sterile filter paper and cotton wool moistened with 5ml of sterile distilled water. At 96 hours after germination, 6 seedlings were picked up at random from each petri dish and the lengths of the resulting radicles and plumules were measured with a metre rule.

Determination of relative growth rate and yield of treated and control plants: The index of growth used was the relative growth rate as described by Kadiri (1990). Treated seeds and controls were planted in 30 x 12cm polyethylene bags with each bag containing 2.6kg of sterilized garden soil (pH 6.8). The sowing was replicated six times and the polyethylene bags arranged randomly in an open field with an ambient temperature of 29 ± 3^oC, relative humidity of 58 ± 10%, wind speed of 5 ± 2kph and photoperiod of 11 ± 1h daily. The emergent plants were harvested twice at 3 weeks and 6 weeks after sowing, the harvested plants dried at 80^oC for 2 days and their dry weights measured and used for the relative growth rate calculation. Relative growth rate was calculated using the formula:

$$\text{Relative growth rate} = \frac{\text{Log}_e \text{ Total dry wt}_2 - \text{Log}_e \text{ Total dry wt}_1}{t_2 - t_1}$$

wt_2 = weight at second harvest t_2

wt_1 = weight at first harvest t_1

At 12 weeks after sowing, the spikes of treated and control *P. americanum* plants and the panicums of treated and control *S. bicolor* plants were harvested and their fresh weights taken as the yields of the various plants.

RESULTS AND DISCUSSION

The plumule lengths, radical lengths, relative growth rates and yields of the 2 cultivars of each of *S. bicolor* and *P. americanum* treated with one and two cycles of ascorbic acid, calcium chloride and indole-3-butyric acid were greater than those of the control seeds treated with 2 cycles of water Tables 1 and 2). The only exceptions were relative growth rate for 1 cycle ascorbic acid and 1 cycle calcium chloride treatments and plumule and radical length for 1 cycle IBA, 1 cycle calcium chloride and 1 cycle ascorbic treatments for *S. bicolor* (Tables 1 and 2). All the seeds given 2 cycles of ascorbic acid, calcium chloride and indole-3-butyric acid hardening treatments had significantly higher plumule and radicle lengths than those of the control seeds (Tables 1 and 2). A similar result of significantly greater relative growth rate and yields for plants of 2 cycles ascorbic acid, calcium chloride and indole-3-butyric acid treated seeds when compared with control plants of water-treated seeds was observed (Tables 1 and 2). The present finding of increased embryo extension due to hardening treatments with calcium chloride, ascorbic acid and indole-3-butyric acid is in line with the findings of Zubenko 1959, Heydecker 1977, Basra, et al., 1990 and Akhtar et al., 2008. Moreover, similar increases in plant relative growth rates due to hardening treatments were obtained by Mumunoy (1975) for Melon, Basu and Prativapal (1980) for wheat, Mohandas and Naidu (1984) for sugar-cane and Ajiboye et al. (2007) for *Dialium guineensis* and *Prosopis africana*.

Two cycles of hardening treatments of the 2 cultivars of *P. americanum* and S. bicolor stimulated greater plumule and radicle elongation, relative growth rate and yields than 1 cycle of hardening treatments (Tables 1 and 2). Genkel et al. (1964) using barley, maize, sunflower, carrot, sugarcane and tomato observed similar increases in plant growth rates as the number of hardening treatment cycles was increased Akhtar et al. (2008) obtained similar results for *Spinacia oleracea*.

The enhancement of plumule and radicle elongation by the pre-sowing hardening treatments of ascorbic acid, calcium chloride and indole-3-butyric acid could be attributed to extensive physiological reorganization in hardened seeds which results in softening of the seed coat. Moreover, the increased relative growth rates and yields by pre-sowing hardening treatments using the 3 chemicals could be due to greater stimulation of biochemical activities in the hardened seeds.

Out of the hardening agents used in the study, indole-3-butyric acid was consistently the best, followed by ascorbic acid and calcium chloride respectively while water which was employed for the control seeds was the least (Tables 1 and 2). This could be due to the fact that IBA is a plant hormone while ascorbic acid is a vitamin. Hormones are believed to promote biochemical activities faster than vitamins (Ibrahim and Nwobosi, 1986).

On the basis of the findings in the present study, pre-sowing hardenings of the seeds of *P. americanum* and *S. bicolor* using indole-3-butyric acid and ascorbic acid as hardening agents hold great potential for improving germination and growth as they produced greater physiological effects than calcium chloride or water.

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Table 1:Effect of various cycles of ascorbic acid, calcium chloride ($CaCl_2$) and indole-3-butyric acid (IBA) on embryo enlargement and relative growth rate of *Pennisetum americanum* cvs. ex Bornu and hairy composite

Seed Treatment and No. of cycles	*P. americanum* cv. Ex. Bornu Plumule length (cm)	Radical length (cm)	Relative growth rate (g/g/wk)	Yield spike fresh wt/plant (g)	*P. americanum* cv hairy compositae Plumule length (cm)	Radical length (cm)	Relative growth rate (g/g/wk)	Yield spike fresh wt/plant (g)
Water control (2cycles)	2.8 ± 0.2	5.8 ± 0.3	0.38 ± 0.02	44.6 ± 2.1	3.4 ± 0.2	6.3 ± 0.3	0.41 ± 0.02	46.2 ± 2.4
Ascorbic acid (1cycle)	*3.6 ± 0.3	*9.5 ± 0.4	*0.45 ± 0.02	*60.2 ± 2.1	*4.3 ± 0.3	*10.2 ± 0.3	0.45 ± 0.03	*64.5 ± 2.6
Ascorbic acid (2cycles)	*4.7 ± 0.3	*11.3 ± 0.5	*0.51 ± 0.03	*65.2 ± 1.8	*6.0 ± 0.4	*11.6 ± 0.4	*0.52 ± 0.04	*69.1 ± 1.9
$CaCl_2$ (1 cycle)	*3.5 ± 0.3	*7.8 ± 0.3	0.42 ± 0.03	*56.3 ± 2.5	*4.0 ± 0.2	*9.4 ± 0.3	0.43 ± 0.03	*60.3 ± 2.4
CaCl2 (2 cycles)	*3.8 ± 0.3	*10.6 ± 0.4	*0.49 ± 0.03	*61.5 ± 2.2	*5.2 ± 0.3	*11.0 ± 0.4	*0.50 ± 0.03	*65.6 ± 2.0
IBA (1 cycle)	*4.9 ± 0.4	*9.9 ± 0.4	*0.48 ± 0.02	*64.5 ± 2.6	*4.7 ± 0.2	*10.4 ± 0.3	*0.49 ± 0.03	*67.3 ± 2.5
IBA (2 cycles)	*5.2 ± 0.4	*12.4 ± 0.5	*0.54 ± 0.03	*69.4 ± 2.2	*6.2 ± 0.4	*12.9 ± 0.4	*0.58 ± 0.04	*71.4 ± 3.5

- Significantly different from the control at 5% probability using Duncan's multiple range test
Data are means of six replicates ± S. E.

Table 2: Effect of various cycles of ascorbic acid, calcium chloride ($CaCl_2$) and indole-3-butyric acid (IBA) on embryo enlargement and relative growth rate of *Sorghum bicolor* cvs. KSV11 and KSV12

Seed Treatment and No. of cycles	*S. bicolor*cv. cv. KSV11 Plumule length (cm)	Radicle length (cm)	Relative growth rate (g/g/wk)	Yield panicle fresh wt/plant (g)	*S. bicolor* cv. KSV12 Plumule length (cm)	Radicle length (cm)	Relative growth rate (g/g/wk)	Yield panicle fresh wt/plant (g)
Water control (2cycles)	3.0 ± 0.0	4.0 ± 0.2	0.21 ± 0.02	54.3 ± 2.1	3.0 ± 0.2	4.0 ± 0.3	0.20 ± 0.01	50.6 ± 2.4
Ascorbic acid (1cycle)	*3.5 ± 0.2	4.5 ± 0.3	*0.32 ± 0.02	*71.1 ± 3.0	3.4 ± 0.2	4.3 ± 0.3	*0.31 ± 0.02	*66.7 ± 3.2
Ascorbic acid (2cycles)	*3.9 ± 0.2	*5.1 ± 0.3	*0.34 ± 0.03	*77.5 ± 2.9	*3.8 ± 0.3	*5.0 ± 0.3	*0.33 ± 0.03	*74.4 ± 2.6
$CaCl_2$ (1 cycle)	3.2 ± 0.1	4.2 ± 0.3	*0.29 ± 0.03	*66.2 ± 2.7	3.3 ± 0.3	4.2 ± 0.3	*0.29 ± 0.02	*61.4 ± 2.6
CaCl2 (2 cycles)	*3.6 ± 0.2	*4.8 ± 0.3	*0.32 ± 0.03	*72.6 ± 3.1	*3.7 ± 0.3	*4.8 ± 0.3	*0.31 ± 0.03	*69.7 ± 2.8
IBA (1 cycle)	*3.8 ± 0.2	*4.8 ± 0.4	*0.34 ± 0.02	*75.7 ± 2.8	*3.6 ± 0.2	4.5 ± 0.2	*0.32 ± 0.01	*73.2 ± 2.9
IBA (2 cycles)	*4.4 ± 0.3	*5.4 ± 0.4	*0.37 ± 0.03	*84.5 ± 3.3	*4.0 ± 0.3	*5.2 ± 0.3	*0.35 ± 0.03	*80.6 ± 3.8

- Significantly different from the control at 5% probability using Duncan's multiple range test
Data are means of six replicates ± S. E.