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Standardizing the Duration of Seed Priming with GA_3 for Invigoration of Artificially Aged Seed of Sorghum

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ABSTRACT

Initially seed of sorghum variety, NTJ-5, was subjected to accelerated aging by exposing them to 45ºC and 95% relative humidity for 24, 48, 72 and 96 h. Seed aging resulted in reduction in germination, seedling length and seedling vigour index. Accelerated aged seed along with unaged seed was treated with GA_3 50 ppm for different durations (0, 3, 6, 9 and 12 h) to identify the best duration of priming for seed invigoration in sorghum. Results disclosed that hormonal priming had the capacity to mitigate the deteriorating effect of aging. Hormonal priming of sorghum seed with GA_3 50 ppm for 9 h caused maximum improvement in germination and seedling growth potential of aged seed with low initial seed quality.

Keywords: *Accelerated aging, GA_3 50 ppm, Germination, Seedling growth, Seed priming and Sorghum.*

Sorghum is the fifth most important crop among the coarse cereals. It is cultivated in semi-arid regions with great ability for yield improvement and abiotic stress tolerance. In plants heat is a key type of abiotic constraint responsible for the accumulation of Reactive Oxygen Species (ROS), which are harmful to plant cells leading to DNA, proteins, lipids and membrane damage (Gosavi *et al*., 2014). Seed deterioration is a natural phenomenon during storage.

Accelerated aging is used to determine storage quality and germination characteristics by mimicking natural aging conditions for crops like wheat (Galleschi *et al*., 2002), rice (Kapoor *et al*., 2011), cotton (Goel *et al*., 2003) and chickpea (Kapoor *et al*., 2010). Aging causes a progressive decline in biological processes leading to degeneration and death. Seed deterioration results in reduced germination capacity, viability and stunted seedling growth. Germination of aged and non-aged seeds can be enhanced by using seed priming technique.

Priming is a pre-sowing invigouration technique that increases germination and seedling growth predominantly under stress conditions like drought, salinity and heat (Sedghi *et al*., 2010). Priming agents like water, polyethylene glycol (PEG), potassium nitrate (KNO_3), calcium chloride ($CaCl_2$), gibberellic acid (GA_3) and salicylic acid (SA) are most commonly used to enhance seed quality. GA_3, one of the plant growth promoting hormones, plays a vital role in growth on development of seed germination, stem elongation, flower development etc. Siadat *et al*. (2012) performed seed priming using GA_3 and KNO_3 in which GA_3 priming was found to be effective in the activation of antioxidant enzymes and improving the quality of aged seeds compared to other priming agents.

Research attempts on the effect of hormonal priming with GA_3 in improving the germination and seedling potential of deteriorated seed of sorghum were scanty. Hence the present investigation was

planned to study the extent of seed deterioration due to accelerated aging and standardizing the duration of priming of sorghum seed with GA_3 50 ppm for invigouration of such deteriorated seed.

MATERIAL AND METHODS

The present study was conducted with the seed of sorghum variety, NTJ-5, procured from Regional Agricultural Research Station, Nandyal in the Department of Seed Science and Technology, Advanced Post Graduate Centre, Lam, Guntur, Andhra Pradesh, India during 2019-20. Initially the seed was subjected to accelerated aging for 24, 48, 72 and 96 h. Accelerated aging was performed by placing the seeds in the incubator with high temperature (45ºC) and relative humidity (95%) (Tekrony, 2005). For each aging treatment, about 500 g of sorghum seeds were scattered within a vaccum container on wire screens; the floor of the container was covered by distilled water and the containers were placed in an incubator at a fixed temperature. In order to standardize the duration of seed priming with GA_3. The aged seed along with control was treated with GA_3 50 ppm using 1:3 seed weight to solution volume ratio (w/v) for 3, 6, 9 and 12 h and then air dried under shade till they reach to safe moisture content. The treated, untreated, aged and unaged seeds were tested for germination by between paper method using standard germination test in Factorial Completely Randomized Design (FCRD) with four replications of 100 seeds from each treatment. Data on germination and seedling vigour index were recorded as per the following formulae:

Germination (%):

On 10th day of germination test, the normal seedlings were counted and expressed as germination (%) as per the following formula:

Germination (%) =

$$\frac{\text{Number of normal seedlings}}{\text{Total number of seed sown}} \text{ X } 100$$

Seedling Length (cm)

The total distance from the tip of primary leaf to root tip of ten randomly selected seedlings from each replication of each treatment was measured with scale and their mean was expressed as seedling length in centimeter

Seedling vigour index

It was computed by adopting the following formula as suggested by Abdul-Baki and Anderson (1973) and was expressed as whole number.

Seedling Vigour Index =

Germination (%) × Seedling length (cm)

Data were analyzed by using SPSS (version 16.0) software after subjecting the data to appropriate transformations. The differences among the duration of seed priming and treatment means were compared by using Duncan's multiple range test at 5% level of probability.

RESULTS AND DISCUSSION

Analysis of variance of data (Table 1) disclosed that duration of seed priming and accelerated aging treatments had highly significant influence on all the seed quality parameters under study *viz.*, germination, seedling length and seedling vigour index. The interaction between aging treatments and duration of hormonal priming with 50 ppm exhibited highly significant influence on seedling length, a significant impact on seedling vigour index and non-significant effect on germination.

Accelerated aging showed a highly significant decrease in germination from 88.50% (initial) to

53.50% (96 h of aging) (Table 2). Seed priming of unaged seed with GA_3 showed highest mean germination (90.85%). Among different durations of accelerated aging, seed priming with GA_3 50 ppm showed highest mean germination (79.25%) with 24 h and lowest (58.20%) with 96 h of aging (Table 2). Significant increase in mean germination was recorded with increase in duration of seed priming from 0 h (72.71%) to 9 h (82.08%) and subsequently a decline in mean germination was noticed with further increase in duration of priming up to 12 h (73.83%). Highest germination (94.75%) was noticed with priming of unaged seed for 9 h while lowest (53.50%) was detected with unprimed seed that was subjected to 96 h of accelerated aging with an overall mean of 75.92%. Seed priming with GA_3 50 ppm for 9 h resulted in highest per cent increment in germination (27.57%) of 96 h accelerated aged seed (Fig. 1). Tabatabaei (2013) reported that priming of accelerated aged seeds of sesamum with GA_3 50 ppm for 24 h showed highest germination compared to aged seeds, which might be due to differential response of various crops to different durations of soaking. In contrary, Siadat *et al*. (2012) mentioned that seed priming with GA_3 100 ppm for 12 h improved the germination of aged seeds in maize. Ayaz *et al*. (2019) found that seed primed with GA_3 at 60 ppm for 18 h showed highest germination in bitter gourd.

Seedling growth declined greatly with rise in the duration of accelerated aging from initial (38.58 cm) to 96 h of aging (29.02 cm) (Table 3). Maximum mean seedling length (40.00 cm) was recorded upon priming of unaged seed with GA_3 50 ppm. Among different accelerated aging treatments and seed priming with GA_3 showed maximum (37.44 cm) and minimum (31.60 cm) mean seedling length with 24 h and 96 h of aging treatments. The mean seedling length had significantly increased with increase in duration of hormonal priming with GA_3 50 ppm from 0 h (34.54 cm) to 9 h (37.83 cm) and later declined with further increase in the duration up to 12 h (36.26 cm). The maximum (42.00 cm) and minimum (29.02 cm) seedling lengths were recorded with 9 h duration of hormonal priming of unaged seed with GA_3 50 ppm and unprimed seed that was subjected to 96 h of accelerated aging, respectively. The grand mean was observed as 36.20 cm. Seed priming with GA_3 50 ppm for 9 h contributed to maximum (16.68%) per cent increment in seedling length of 96 h accelerated aged seed (Fig. 2). The significant influence of interaction between aging treatments and duration of seed soaking in GA_3 on root growth was noticed by Siadat *et al.* (2011) in maize. Hormonal priming with GA_3 100 ppm for 12 h improved the seedling length in maize (Kumari *et al*., 2017). Improvement in the seedling length of naturally aged seeds of onion after seed priming with GA_3 50 ppm for 16 h was earlier reported by Brar *et al*. (2019).

Accelerated aging caused a highly significant decline in seedling vigour index from 3423 (initial) to 1553 with 96 h of accelerated aging (Table 4). Seedling vigour index showed the same trend as that of germination and seedling length. Therefore among all the treatments highest mean seedling vigour index (3636) was recorded upon seed priming of unaged seed with GA_3 50 ppm. Among different durations of accelerated aging treatments and seed priming with GA_3 showed highest (2971) mean seedling vigour index with 24 h and the lowest (1849) was observed with 96 h of aging. A significant improvement in seedling vigour index of sorghum seed was noticed after seed priming with GA_3 for different durations (up to 9 h). The mean seedling vigour index increased with extended duration of hormonal priming with GA_3 50 ppm from 0 h (2560) to 9 h (3129) and decreased subsequently with further increase in the duration up to 12 h (2714). Highest seedling vigour index (3979) was recorded with 9 h duration of seed priming of

Table 1. Analysis of variance for seed quality parameters of sorghum seed subjected to accelerated aging and seed priming with GA_3 50 ppm for different durations

Source	Degrees of freedom	Mean sum of squares		
		Germination (%)	Seedling length (cm)	Seedling vigour index
Duration of seed priming (D)	4	175.02**	40.90**	1,262,257.39**
Aging (A)	5	1616.93**	202.13**	9,580,483.07**
Duration × Aging (D × A)	20	8.34	2.30**	59,143.33*
Error	90	6.06	0.93	31,866.46

** Significant difference at 1% probability level; * Significant difference at 5% probability level

Table 2. Influence of accelerated aging and seed priming with GA_3 50 ppm for different durations on germination (%) of sorghum seed

Treatments	Duration of seed priming with GA_3 50 ppm (h)					
	0	3	6	9	12	Mean
A_1	88.5	88.5	88.5	88.5	88.5	88.5
	(70.61)	(70.61)	(70.61)	(70.61)	(70.61)	(70.61)^{B}
A_2	75.25	76.75	81.75	85.5	77	79.25
	(60.16)	(61.15)	(64.70)	(67.61)	(61.33)	(62.99)^{C}
A_3	68.5	69	75.75	81	69.75	72.8
	(55.84)	(56.15)	(60.49)	(64.17)	(56.62)	(58.65)^{D}
A_4	61.75	62.5	68	74.5	63	65.95
	(51.78)	(52.22)	(55.54)	(59.66)	(52.52)	(54.34)^{E}
A_5	53.5	53.75	61	68.25	54.5	58.2
	(46.99)	(47.13)	(51.34)	(55.69)	(47.57)	(49.74)^{F}
A_6	88.75	89.5	91	94.75	90.25	90.85
	(70.39)	(71.11)	(72.55)	(76.77)	(71.79)	(72.52)^{*A}
Mean	72.71	73.33	77.67	82.08	73.83	75.92
	(59.30)^{e}	(59.73)^{d}	(62.54)^{b}	(65.75)^{*a}	(60.07)^{c}	(61.48)
S Em±	D		A		D × A	
	0.5		0.55		1.23	
CD (5%)	1.41		1.55		NS	
CV (%)	4					

*Values in the parenthesis indicate arc-sine transformed values

The values in the same column with the same alphabet are not significantly different as per DMRT

NS – Non-significant

Treatments:

A_1 – Control (unaged seed without priming) Seed priming with GA_3 50 ppm of

A_2 – Seed subjected to accelerated aging for 24 h; A_3 – Seed subjected to accelerated aging for 48 h

A_4 – Seed subjected to accelerated aging for 72 h; A_5 – Seed subjected to accelerated aging for 96 h

A_6 – Unaged seed

Table 3. Influence of accelerated aging and seed priming with GA_3 50 ppm for different durations on seedling length (cm) of sorghum

Treatments	Duration of seed priming with GA_3 50 ppm (h)					
	0	3	6	9	12	Mean
A_1	38.58	38.58	38.58	38.58	38.58	38.58^{B}
A_2	36.06	36.28	38.25	39.02	37.58	37.44^{C}
A_3	33.97	35.38	37.08	37.98	36.41	36.16^{D}
A_4	31.03	32.22	34.59	35.56	33.64	33.41^{E}
A_5	29.02	30.18	33.31	33.86	31.65	31.60^{F}
A_6	38.6	39.4	40.27	42	39.72	40.00^{A}
Mean	34.54^{e}	35.34^{d}	37.01^{b}	37.83^{a}	36.26^{c}	36.2
S Em ±	D		A		D × A	
	0.2		0.22		0.48	
CD (5%)	0.55		0.61		1.36	
CV (%)	2.67					

The values in the same column with the same alphabet are not significantly different as per DMRT ($P< 0.01$)

Table 4. Influence of accelerated aging and seed priming with GA_3 50 ppm for different durations on seedling vigour index of sorghum

Treatments	Duration of seed priming with GA_3 50 ppm (h)					
	0	3	6	9	12	Mean
A_1	3423	3423	3423	3423	3423	3423^{B}
A_2	2714	2784	3126	3336	2893	2971^{C}
A_3	2328	2441	2808	3077	2539	2639^{D}
A_4	1915	2014	2353	2650	2119	2210^{E}
A_5	1553	1622	2032	2311	1725	1849^{F}
A_6	3425	3526	3664	3979	3585	3636^{A}
Mean	2560^{e}	2635^{de}	2901^{b}	3129^{a}	2714^{cd}	2788
S Em ±	D		A		D × A	
	36.44		39.92		89.26	
CD (5%)	102.55		112.34		251.2	
CV (%)	6.4					

The values in the same column with the same alphabet are not significantly different as per DMRT ($P< 0.01$)

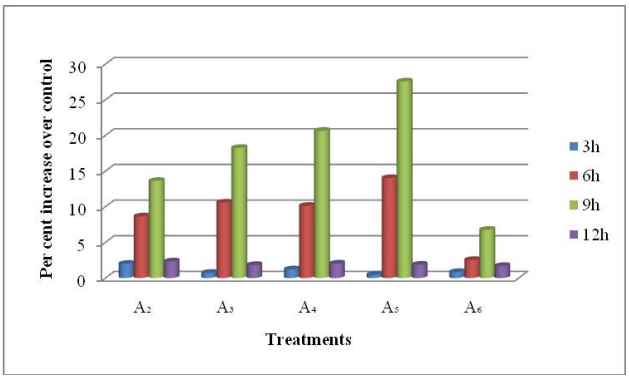

Fig. 1: Per cent improvement in germination of sorghum seed after seed priming with GA_3 50 ppm for different durations

Treatments:

A_1 – Control (unaged seed without priming)

Seed priming with GA_3 50 ppm of

A_2 – Seed subjected to accelerated aging for 24 h

A_3 – Seed subjected to accelerated aging for 48 h

A_4 – Seed subjected to accelerated aging for 72 h

A_5 – Seed subjected to accelerated aging for 96 h

A_6 – Unaged seed

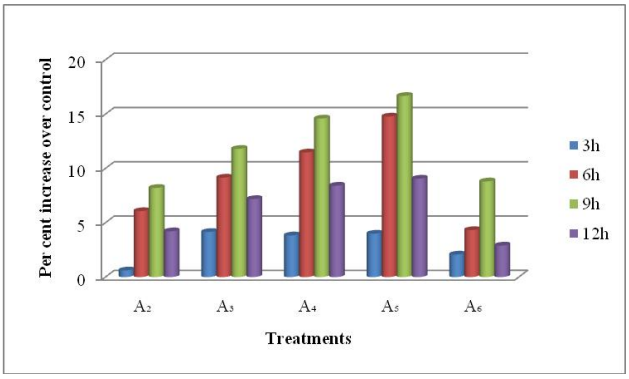

Fig. 2: Per cent increase in seedling length of sorghum after seed priming with GA_3 50 ppm for different durations

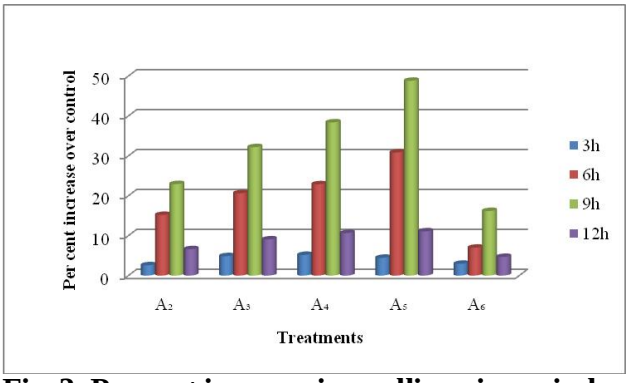

Fig. 3: Per cent increase in seedling vigour index of sorghum after seed priming with GA_3 50 ppm for different durations

unaged seed with GA_3 50 ppm and the least (1553) was observed with unprimed seed subjected to 96 h of accelerated aging. The overall mean of seedling vigour index was 2788. Seed priming of 96 h accelerated aged seed with GA_3 50 ppm for 9 h showed maximum per cent increment in seedling vigour index (48.81%) (Fig. 3). Kumari *et al*. (2017) found improvement in seedling vigour index upon priming of maize seed with GA_3 100 ppm for 12 h. Brar *et al*. (2019) found improvement in vigour of naturally aged onion seed upon priming with GA_3 50 ppm for 16 h.

Finch-Savage and Leubner-Metzger (2006) emphasized the role of gibberelline in increasing the growth potential of embryo and promoting germination and also stated that GA_3 is necessary to overcome the dormancy. The improvement in germination of aged seeds by GA_3 priming may be due to the stimulation of hydrolytic enzymes such as lyases and dehydrogenases that increases the starch metabolism thus increases in germination through cell division (Sarika *et al*., 2013). These enzymes mobilize the storage reserves from endosperm that stimulates germination and growth (Cirac *et al*., 2004). Higher metabolic activity in primed seed causes efficient food mobilization during early hours of germination, which leads to increased seedling length (Brar *et al*., 2019). The enhancement in seedling vigour index was due to increase in seed germination and seedling length by seed priming with GA_3.

CONCLUSION

Seed priming with GA_3 50 ppm for 9 h was found to be most effective in improving the germination, seedling growth and seedling vigou index of sorghum variety, NTJ-5, subjected to accelerated aging for different durations.

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