

## Optimizing duration of hydro-priming for seed quality enhancement in cotton

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**ABSTRACT** Seeds of five varieties of cotton were primed for different durations, dried and their subsequent seed quality parameters like germination percent, speed of germination, emergence percent, vigour index and activity of hydrolyzing enzymes were analyzed according to time duration for hydro-priming. The values of hydro-priming time accounted for 92% (30°C for 3 h), 78% (30°C for 6h), and 65% (30°C for 8h) of germination following priming. Seed priming is based upon seed water relations, it was hypothesized that the sensitivity of germination to reduced water potential before priming might be mechanistically related to, and therefore predictive of, priming duration responsiveness. This technique assures the superior performance and has found commercial applications. Seed hydration is a process, whereby seeds are hydrated and then re-dried to permit routine handling. This process results in increased germination rate, more uniform emergence, germination under a broader range of environments and improved seedling vigour and growth. However, there is insufficient information for the duration of priming during which the seed quality can be enhanced effectively. Therefore, response of different priming durations on different seed vigour and related parameters were evaluated in five commercial genotypes of cotton *viz.* three Bt (NCS 857, MRC 7017, MRC 7031) and two non Bt (LHH 144-American type and PAU 626H-desi type). The pre-sowing seed priming durations were slow hydration at room temperature for 3h, 6h and 8h. The pre-sowing seed hydration for 3h was observed to be most effective in promoting the different seed vigour parameters like germination percentage, speed of germination, emergence percent and vigour index. Amylase activity was also observed to be maximum during initial 48h of germination. Therefore, it was concluded that for getting maximum seed germination and seed vigour in cotton, the seed may be hydrated for 3h prior to sowing.

**Keywords:** Cotton, seed priming duration, seed quality enhancement

Seed priming has a central objective to improve seed performance under very specific regimes. Inadequate moisture and other abiotic stresses during cropping season result in poor germination and poor plant stand, ultimately leading to poor yield and sometimes crop failure. Rapid and uniform field emergence is extremely important for high yield and better quality. Percent seedling emergence has direct impact on yield, whereas, slow emergence results in weaker seedlings which are more prone to diseases. It improves the physiological performance of the seed that further enhance their performance. The objective of seed hydration technology is to increase the percentage and rate of germination, expand the range of temperatures over which the seed will germinate and increase the uniformity of

stand establishment. Hydro-priming involves soaking of seeds in water for a precise duration followed by re-drying. To accomplish these objectives, seeds must be hydrated in some way at a moisture level sufficient to initiate the early events of germination (Phase II of imbibition) but not sufficient to permit radical protrusion (Phase III) [1]. But there is no literature available, indicating the exact duration of hydro-priming for cotton seed. Therefore, present investigations were deliberated at Seed Technology Research farm of Punjab Agricultural University, Ludhiana during 2010, 2011 and 2012 to standardize the time duration of hydro-priming for enhancing physiological seed quality and registering invigoration at molecular level in five cotton varieties which were Bt as well as non Bt cotton.

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## MATERIALS AND METHODS

Five commercial cotton varieties *viz.* three Bt (NCS 857, MRC 7017 and MRC 7031) and two non Bt (LHH 144-American and PAU 626H-desi type) were used for the present studies. Seeds were subjected to slow hydration for different time durations followed by air drying at room temperature *i.e.* 3h ( $T_1$ ); 6h ( $T_2$ ); 8h ( $T_3$ ) along with control ( $T_0$ ). Hydration was performed using moist blotter papers (seed: water 1:1). The observations were recorded on germination percent, speed of germination, emergence percent, vigour index I and vigour index II. The germination was tested with "Between the Paper" method as per modified ISTA seed testing rules [2] in quadruplicate of 100 seeds each at 30°C in a germinator. For determining seedling dry weight, 10 normal seedlings, selected at random, were dried at 110°C for 17h and weighed. Seed vigour index I and vigour index II were calculated as per formulae [3]. Speed of germination was computed using 50 seeds in four replicates in Petri dishes by following TP method. Daily observations of emerged seedlings were recorded till the final count day. The speed of germination was calculated as per formula  $\Sigma nt / \Sigma n$  [4]. For computing field emergence, treated (hydro-primed) seeds were taken to the field immediately for sowing. Hundred plants in each treatment, spaced at 90 cm x 60 cm in field were sown. The recommended package of practices was followed for raising the successful crop. The data was recorded with respect to field emergence percent.

Hydro-primed seeds for different durations, after putting for germination, were drawn at different intervals *i.e.* after 0h, 12h, 24h and 48h and were subjected to estimation of hydrolyzing enzymes. For assay of amylases (a and b), 1g of cotyledonary tissue was homogenized in 0.2 percent calcium acetate (pH 6.0) and centrifuged. The supernatant was used for determination of amylase activity by the standard method [5].

Amylase activity was expressed as  $\mu\text{g}$  of starch hydrolyzed  $\text{min}^{-1} \text{g}^{-1}$  tissue.

## RESULTS AND DISCUSSION

Seeds of five varieties of cotton when primed for various durations, dried, and their subsequent seed quality parameters were analyzed according to time durations of hydro-priming. Perusal of data established that among different durations, seed hydro-priming for 3h indicated significant superiority in all seed quality parameters *i.e.* germination percent, speed of germination, vigour index I, vigour index II and activity of hydrolyzing enzymes. (Table 1, 2 and fig. 1). Seeds of all varieties under study (NCS 857, MRC 7017, MRC 7031, LHH 144 and PAU 626H) when hydro-primed for 3h indicated enhanced germination percentage (84, 80, 82, 85 and 92%, respectively for all varieties). Similarly vigour index I (3461, 3224, 3222, 3722 and 3987, respectively), vigour index II (26.45, 26.0, 28.2, 28.8 and 28.34, respectively) and speed of emergence (10.9, 11.8, 11.8, 12.2 and 13.5, respectively) was also increased as indicated in Table 1. It was also observed that NCS 857 (84%), MRC 7017 (80%), MRC 7031 (82%) and LHH 144 (85.0%) were at par in terms of germination percent and PAU 626H (92%) showed maximum germination. It has been reported that varying durations of seed priming affected seed germination in maize [6]. Germination of seeds primed for longer periods, (6h and 8h) 74% and 70% in NCS 857 and MRC7017; 72 and 70% in MRC 7031; 74 and 71% in LHH 144 and 84 and 80% in PAU 626H respectively, for the varieties under study, exhibited a negative germination response relative to the control which was 69, 75, 73, 70 and 80 percent, respectively (Table 2). Though the germination percent was within minimum seed certification standards, but it was significantly reduced as compared to 3h hydro-priming duration.

This is very important from commercial point of view when the seeds with

**Table 1. Seed quality as influenced by hydro-priming for 3h in cotton. (Pool of 3 years)**

	84	78	3461	26.45	11.4
	80	70	3224	26	11.8
	82	76	3222	28.2	11.8
	85	78	3722	28.8	12.2
	92	84	3987	28.34	13.5
CD (p=0.05)	5.66	6.23	411.17	1.55	1.01

**Table 2. Effect of seed priming durations on various seed quality attributes in cotton (Pool of 3 years)**

Variety	PD (h)	G%	FE%	SE	VII	VIII
NCS 857 Bt2	0	69	55	8.8	3022	22
	3	84	78	11.4	3461	26.45
	6	74	66	10.4	3345	25.28
	8	70	55	10	3296	25.12
	CD (p=0.05)		10.90	17.45	1.71	295.95
MRC 7017	0	75	55	9.45	2998	20.22
	3	80	70	11.8	3224	26
	6	74	60	9.62	3026	24.12
	8	70	58	9.44	3022	24.24
	CD (p=0.05)		6.54	10.34	1.83	167.16
MRC 7031	0	73	56	9.8	2916	22.2
	3	82	70	11.8	3222	28.2
	6	72	65	10.6	2882	26.96
	8	70	55	10	2867	26.42
	CD (p=0.05)		8.46	11.51	1.43	267.43
LHH 144	0	70	55	10.5	3588	18.12
	3	85	78	12.2	3722	28.8
	6	74	63	11.4	3633	19.55
	8	71	60	11.6	3628	19.48
	CD (p=0.05)		10.95	15.75	1.12	89.96
PAU 626H	0	80	62	10.6	3722	18.96
	3	92	84	13.5	3987	28.34
	6	84	72	10.99	3788	20.34
	8	80	72	10.9	3766	20.34
	CD (p=0.05)		9.00	14.32	2.14	186.81

PD: Priming duration; G %: Germination %; FE%: Emergence %;  
SE: Speed of emergence; VI I: Vigour Index I; VI II: Vigour Index II.

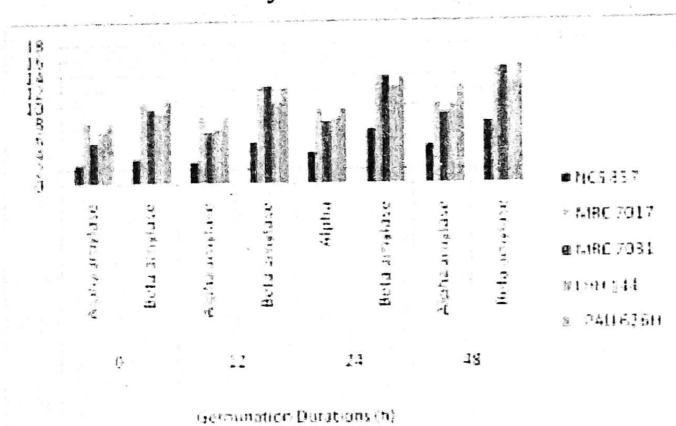
higher germination percent indicate higher vigour and fetch more premiums. Seed-water uptake measurements suggest that a reduction in the lag time of imbibition accounted for at least some germination-rate

enhancement in priming treatments duration [7]. Similar findings were also reported by Reddy and Khan [8] in Khirni, Natesh *et al.* [9] in chilli. Enhanced field emergence percent with hydro-priming treatment for 3h was

**Table 3. Activity of alpha and beta amylase as affected by seed priming treatment-T1 (moles min<sup>-1</sup>g<sup>-1</sup>) (Pool of 3 years).**

Varieties	Hours after putting for germination (h)							
	0		12		24		48	
	Alpha amylase	Beta amylase	Alpha amylase	Beta amylase	Alpha amylase	Beta amylase	Alpha amylase	Beta amylase
NCS 857	2.5	3.33	2.9	5.4	4.16	7.1	5	7.88
MRC 7017	7.9	10.5	8.75	12.51	9.58	13.72	10.25	15.01
MRC 7031	5.41	9.6	6.66	12.5	7.91	13.75	9.16	15.01
LHH 144	6.75	9.13	7.16	10.43	8.55	12.51	10.28	13.32
PAU 626H	7.8	10.88	8.75	12.41	9.58	13.68	12.55	15.25
CD (p=0.05)	2.78	3.82	2.97	3.81	2.78	3.57	3.45	3.88

### Enzyme Activity



**Fig. 1. Activity of alpha and beta amylase as affected by seed priming treatment-T1 (moles min<sup>-1</sup>g<sup>-1</sup>) (Pool of 3 years).**

indicated in variety PAU 626H (84%), followed by NCS 857 and LHH 144 (78.0%). Genotypes MRC 7031 and MRC 7017(70%) exhibited similar results in terms of emergence percent. Seed hydration for 3h enhances amylase synthesis that helps in conversion of reserve food material into amino acids and their translocation via phloem into and throughout the young root and shoots resulting in rapid rate of emergence. Similar findings were reported in previous studies [10-11] for enhanced emergence percentage with seed priming in okra using different durations. Hydro-priming treatments for 3h indicated similar results in terms of speed of germination *i.e.* 11.4 (NCS 857) and 11.8 in MRC 7017 and MRC 7031. However, PAU 626H registered

maximum speed of germination (13.5), followed by that in LHH 144 (12.2) when their seeds were hydro-primed for 3h. Higher speed of germination with hydro-primed seeds for 3h may be due to the increased metabolic activities during this duration for germination. An increase in speed of germination in pepper with gibberellins treatment for 3h was reported [12]. Also, it was reported that pre-sowing seed treatment in tomato, capsicum and onion seed with different concentrations of GA<sub>3</sub> enhanced rate of germination, hypocotyl length and field emergence compared to untreated seeds for different durations [13-14]. Vigour index determines the state of the health of seedlings, their biomass and ultimately the state of the productivity of the plant. Higher the vigour index, better will be the performance of the plant. The increased vigour index is also attributed to higher values recorded with respect to seedling length and seedling dry weight. Vigour index-I indicated that among the three seed priming durations, hydro-priming for 3h (3987) exhibited significantly higher value. Priming causes *de novo* synthesis of  $\alpha$ -amylase [15] increasing metabolic activities in seeds resulting into higher seed vigour. Similar findings were reported in *Mimusops hexandra* [8]. Amongst varieties, PAU 626H had maximum vigour index (3987) followed by that in LHH144 (3722). Similarly enhanced activity of hydrolyzing enzymes

was indicated in the cotton seeds hydro-primed for 3h (Fig. 1) during initial 48 hours of germination. Increased  $\alpha$ -amylase activity has been correlated with improved metabolic activities and higher seed vigour [16-17].

From the present studies, it was inferred that seed hydration treatment for 3h showed significantly improved seed quality parameters followed by 6h among the different seed priming treatments. Among the genotypes, PAU 626H exhibited maximum response towards the seed hydro-priming for 3h followed by LHH 144. Rest of the varieties, when hydro-primed for 3h were at par in terms of seed quality attributes, rest of the varieties NCS 857, MRC 7017, MRC 7031 were at par with each other in all seed quality parameters when hydro-primed for 3h. Therefore, it concluded that hydro-priming for 3h ensures rapid and uniform germination under abiotic stress (high temperature) and activity of hydrolyzing enzymes is eminent during initial 48h of its germination.

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