http://www.pjbs.org



ISSN 1028-8880

# Pakistan Journal of Biological Sciences

ANSIMet

Asian Network for Scientific Information 308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

# Improvement of Okra Seed Quality by Pre-soaking in H<sub>2</sub>O<sub>2</sub> Solution

T. M. T. Iqbal, M. M. Bahadur, M. A. Kabir, M. A. Hasan and D. A. N.Majumder Hajee Mohammad Danesh Science and Technology University, Dinajpur, Bangladesh

**Abstract:** The efficacy of H<sub>2</sub>O<sub>2</sub> solution in improving the seed quality of two Indian okra varieties (Anamika and Parbhani kanti) was investigated. The seeds were pre-soaked in different concentrations for six hours. Vigour index and % germination were estimated. Root and shoot lengths as well as dry matter contents of the normal seedlings were determined. Moreover, crop growth rate (CGR) at 10 and 20 days after sowing (DAS) were also worked-out. It has been experienced that, pre-soaking at 2.0% concentration significantly enhanced all the parameters studied. However, in some cases, 1.5 to 2.5% concentrations also gave identically the best results. Afterwards toxicity was clearly manifested through reduction of the studied parameters. Contrastly, pre-soaking in plain water resulted in poor values for all the parameters noted. Moreover, in terms of variety, Anamika was significantly better than Parbhani kanti.

Key words: Okra seed, H<sub>2</sub>O<sub>2</sub>, seed quality

### Introduction

Water imbibition is the first step in seed germination. But nursery bed/crop field may lack adequate moisture content for the same, so, poor and delayed germination occurs. To combat this, farmers pre-soak the seed in plain water for a few hours. But this may cause seed damage in more than one ways. Of them, the major one is that, excess water may be trapped in the area of embryonic axis, nodal zone and cotyledons. This leads to suffocation resulting in delayed and poor germination as well as weak seedling growth (Orphanos and Hydecker, 1968; Hydecker, 1977). So, aqueous solution of any chemical having capability to supply O2 at the embryonic axis during seed imbibition could be a worthwhile measure. As such, aqueous solutions of H2O2 at varying concentration(s) and imbibition period(s) have been found fruitful in various seeds (Copeland and McDonald, 1985). It improves germination percentage, reduces time to germinate and also increases early seedling growth e.g. in groundnut (Lai, 1988 and Rahman, et al., 1997), in pearl millet (Choi et al., 1989), with Vangueria infausta (Msanga and Maghembe, 1989), in cottonseed (Bordovsky et al., 1991) and with Tripsacum dactyloides (Kindiger, 1994). Lady's finger (Abelmoschus esculentus L.) is an important summer vegetable of Bangladesh. It is cultivated by direct dibbling of seed in the field during January to April, the driest period of any year. Hence, soaking its seed in aqueous solution of the above said H2O2 having accurate concentration(s) could be an appropriate answer. Unfortunately, information in this light is lacking. So, the present experiment was conducted to gain experiences about the fate of lady's finger seed presoaked with aqueous solution of H2O2.

# Materials and Methods

Two-factor pot experiment had 20 treatment combinations. There were 10 different concentrations of  $H_2O_2$  (0 = tap water, 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 3.5, 4.0 and 4.5%v/v in water). Moreover seeds of two okra varieties (Anamika of Namdhari and Parbhani kanti of Bonkim Seed Co. Ltd, India, respectively) grown extensively in the northern area of Bangladesh, were the test material. The study period was August-September, 1999. The experiment was conducted in randomized complete block design with three replications taking 100 seeds per observation. The seeds were pre-soaked for six hours with respective solution and were sown @ 20 seeds per earthen pot (30 × 25 × 15cm) filled with sandy loam soil. Watering was done as and when necessary. Seed germination was noted daily up to 10 days and then evaluated according to ISTA (1985) rules. From that observation, vigour index was computed (Maguire, 1962). In addition, seedling parameters for root and shoot lengths as well as their dry matter contents of 10-day old seedlings were also found-out

by drying them at 110°C for 8 hours. At this time, all the normal seedlings were taken into consideration. In addition, the crop growth rates at 10 and 20 DAS were estimated (Radford, 1967). Finally, the means were compared by the DMRT (Steel and Torie, 1960) and correlation coefficients were also estimated.

## Results and Discussions

Effect of H2O2 pre-soaking on germination was significant (Tables 1 and 2). The highest germination % (88.33) was obtained at 2.0 % solution (Table 1), which was statistically at par with that of 1.5% conc. (88.16). Again, the least germination % (72.00) was resulted with 4.5% conc. That was again statistically similar with the result from 4.0 % (74.00). Even those two values were also inferior to the control treatment/plain water soaking (77.00 %). These indicated that, concentrations > 2.5% exerted toxic effects on germination. Again, varietal difference was also significant (Table 1); Anamika gave the highest germination % (84.60) than Parbhani kanti (76.90 %). Moreover, the interaction effect of H2O2 and variety was also significant (Table 2). The maximum germination % (91.66) was obtained in Anamika treated with 1.5% solution. However, that gave statistically identical results with conc. up to 2.5%. On the contrary, the lowest germination % (69.00) was obtained in Parbhani kanti treated with 4.5 % H<sub>2</sub>O<sub>2</sub> and that value was statistically similar with those from 3.5 to 4.5% solutions. In combined effect, toxicity of the chemical was manifested at conc. > 2.5 % with both the varieties. Furthermore, the variety Anamika always showed better performance than its counterpart Parbhani kanti irrespective of concentration. Rahman et al. (1997) also obtained better germination in groundnut seed at 1.5 % concentration as well as toxic effects at >2.5 % concentrations of H2O2 pre-soaked for six hours. In addition, they also found varietal differences.

Effect of  ${\rm H_2O_2}$  presoaking on vigour index:  ${\rm H_2O_2}$  treatment significantly influenced vigour index (Table 1 and 2). The highest score (15.48) was obtained in seeds treated with 2 %  ${\rm H_2O_2}$  (Table 1). However, it was statistically identical to those of 1.5 % (14.59) and 2.5 % solutions (14.58). On the other hand, the lowest value (12.18) obtained with 4.5 % was insignificant among the rest six treatments. Vigour index was also significantly different among the varieties. The highest index (14.36) was obtained in Anamika while the lowest (12.84) in Prabhani kranti just like the % of germination, both the varieties captured their previous positions in the case of vigour index too. Moreover, there was significant interaction between variety and  ${\rm H_2O_2}$  conc. (Table 2). The top most index (16.85) was obtained in Anamika treated with 2.0 % solution. But the results from 1.5 to 2.5% solutions were statistically

Iqbal et al.: H2O2 and okra seed quality

Table 1: Effect of H<sub>2</sub>O<sub>2</sub> pre-soaking of seed on germination, vigour and early growth of two Indian okra varieties

Treatment Combination	Germination (%)	Vigor index	Shoot Length (cm)	Root length (cm)	Shoot dry	Root dry matter	Crop growth rate (CGR)	
					matter (mg/plant)	(mg/plant)	10 DAS	20 DAS
H <sub>2</sub> O <sub>2</sub> conc.								
0.00	77.00e	12.32c	12.63de	12.49e	57.50f	19.63f	2.55cd	7.75a
0.50	81.66cd	13.32bc	15.34bcd	14.09de	77.25cde	26.90 de	2.61bcd	7.74c
1.00	84.17bc	13.73bc	16.13bc	16.16bcd	87.00bcd	30.29bcd	2.70bcd	9.07b
1.50	88.16a	14.59ab	16.82b	17.28bc	97.50b	34.33b	3.20ab	10.37ab
2.00	88.33a	15.48a	19.86a	20.15a	120.17a	42.66a	3.28a	10.70a
2.50	85.33b	14.58ab	17.21b	17.45b	94.75bc	33.33bc	3.26a	10.51a
3.00	80.00d	13.70bc	15.71bc	16.25bcd	91.00bc	31.44bc	3.01 bc	9.07bc
3.50	76.83e	13.31bc	15.02bcd	14.64cde	82.00bcd	28.64bcd	2.81bcd	8.77bc
4.00	74.00f	12.82c	13.30cd	12.70e	70.00def	24.23def	2.59bcd	8.08c
4.50	72.00f	12.18c	10.09e	11.97e	60.50ef	20.74ef	2.28d	7.96c
CV(%)	2.89	9.30	14.62	13.72	17.31	19.00	16.69	14.63
Variety								
Anamika	84.60a	14.36a	16.13a	16.06a	93.50a	32.37a	3.14a	9.64a
Parbhani kanti	76.90b	12.84	14.29b	14.58ь	74.03b	26.07b	2.57b	8.62b
CV(%)	2.89	9.30	14.62	13.72	17.31	19.00	16.69	14.63

<sup>\*</sup> values with same letter donot differ significantly at 5% level.

Table 2: Combined effect of Variety and H<sub>2</sub>O<sub>2</sub> conc. on germination, vigour and early growth of two okra varieties.

Treatment Combination	Germination (%)	Vigor index	Shoot Length (cm)	Root	Shoot dry	Root dry matter	Crop growth rate (CGR)	
				length (cm)	matter (mg/plant)	(mg/plant)	10 DAS	20 DAS
Anamika								
0.00	81.66fgh	13.01cdef	12.70efg	13.28efg	63.00fgh	21.15ghi	2.83bcde	8.11 cde
0.50	86.00cde	13.82bcdef	15.85bcdef	14.78cdefg	89.00bcdef	30.71bcdefg	3.01bc	8.07 cde
1.00	88.33abcd	14.42bcd	17.07bcde	16.38cdef	97.00cde	33.65bcd	3.02bcd	9.53bc
1.50	91.66a	15.31abc	21.30ab	18.12bcd	115.00abc	40.59b	3.53ab	10.27ab
2.00	90.33ab	16.85ab	22.05a	21.05a	127.00a	44.68a	3.64a	11.51a
2.50	90.66abc	15.53ab	20.30ab	19.15bc	118.00ab	39.33b	3.61a	11.13a
3.00	84.00efg	14.27bcd	16.75bcdef	17.63bc	105.00bc	36.59bcd	3.13bc	10.18bc
3.50	81.33fgh	13.93bcde	16.05bcdef	16.12cdef	99.00bcde	34.39bcd	3.01bc	9.18bcde
4.00	78.00hij	13.51bcde	13.15efg	12.68fg	78.00defgh	26.67cdefghi	2.67bcde	8.35bcde
4.50	75.00jk	12.97cdef	10.13g	12.43fg	65.00fgh	21.89fghi	2.53cde	8.25cde
Parbhani kan	ti							
0.00	72.33ke	11.63ef	12.55fg	11.70g	52.00h	18.11i	2.26de	7.58e
0.50	77.33hij	12.81def	14.83cdef	13.40defg	65.50fgh	23.08fghi	2.41 de	7.40e
1.00	80.00ghi	13.03cdef	15.19bcdef	15.94cdef	77.00efgh	26.93cdefghi	2.57cde	9.21bcd
1.50	84.66def	13.87bcde	15.33bcdef	17.17bcde	90.00bcdef	32.08bcdef	2.87abcde	9.87ab.cd
2.00	86.33bcde	14.10bcde	17.68bcd	19.75b	113.33ab	40.64ab	2.91abcd	9.88ab.cd
2.50	81.00fgh	13.63bcdef	15.19bcdef	16.15bcde	82.50cdefg	29.32cdefgh.	2.90ab.cd	9.88ab.cd
3.00	76.00ijk	13.12bcdef	14.67 cdef	14.87cdefg	77.00efgh	26.29defghi	2.76abcde	8.36bcde
3.50	72.33kl	12.69def	13.98cdefg	13.06fg	65.00fgh	22.89fghi	2.50cde	8.35bcde
4.00	70.00 I	12.12def	13.44defg	12.72fg	62.00fgh	21.79fghi	2.50cde	7.80de
4.50	69.00 I	11.39f	10.06g	11.50g	56.00gh	19.58hi	2.02e	7.67e
CV(%)	2.89	9.30	14.62	13.72	17.31	19.00	16.69	14.63

<sup>\*</sup> values with same letter donot differ significantly at 5% level.

Table 3: Correlation coefficients\* for okra seedling parameters as affected by soaking in various concentrations of H<sub>2</sub>O<sub>2</sub>

Parameters	Vigour	Shoot length	Root length	Shoot dry matter	Root dry matter	Crop growth ra	ate
	index						
						10 DAS	20 DAS
Germination %	0.707	0.687	0.722	0.739	0.745	0.597	0.596
Vigor index		0.821	0.544	0.894	0.622	0.470	0.478
Shoot length			0.666	0.836	0.680	0.512	0.504
Root length				0.674	0.946	0.640	0.640
Shoot dry matter					0.753	0.537	0.464
Root dry matter						0.621	0.596
Crop growth rate	10 DAS						0.540

<sup>\*</sup>All the values were significant at 5% level of probability

similar among these three treatments. Again, the lowest vigour index (11.39) was observed in Prabhani kranti at  $4.5\,\%$  solution, which was insignificant with rest of the concentrations except at 2.0 % for the same variety. Rahman *et al.* (1997) obtained higher seedling vigour in groundnut seed at 1.5 % but adverse effects at concentrations  $> 2.5\,\%$  of  ${\rm H_2O_2}$  presoaked for six hours.

**Influence of H\_2O\_2 on shoot production:** Shoot production was also significantly influenced by the concentration imposed (Tables 1 and 2). The longest shoot (19.86cm) was produced

with 2.0% solution, which was significantly higher than rest of concentrations (Table 1). On the other hand, the shortest shoot (10.09cm) was produced with the highest concentration (4.5%). But that value was statistically alike the control one (12.63 cm). In combined effect (Table 2), the highest result was obtained with Anamika at 2.0% conc. (22.05cm). However, that value was statistically similar (21.30 and 20.30 cm) with the next lower and higher concentrations (1.5 and 2.0% respectively). In case of Parbhani kanti, shorter shoot was obtained than the Anamika irrespective of concentration. Till, 1.5 to 2.5% concentrations also gave

identically better results. Rahman et al. (1997) while working with seed of two groundnut varieties also attained nearly same results.

Furthermore, almost parallel results were obtained when shoot dry matter contents were evaluated. Such similar occurrence was quite expected.

Role of H2O2 on root production: Seedling root was also influenced by the solution used (Tables 1 and 2). The longest root (20.15cm) was produced with 2.0% solution (Table 1). That was significantly higher as obtained from the rest nine concentrations. On the other hand, the shortest root (11.97cm) was produced with 4.5% solution. But that was statistically similar with the two next higher concentration as well as the lowest and the control ones. In combined effect (Table 2), the deepest root (21.05cm) was also obtained at 2.0 % conc. for variety Anamika. However, that value was statistically unparalleled to all the rest concs. and varieties. Again, in case of Parbhani kanti, shorter root was obtained than the Anamika irrespective of concentration. But, 1.5 to 2.5% concentrations gave identically better results. Rahman et al. (1997) also obtained quite similar experiences from seed of two groundnut varieties.

Again, almost identical situations were manifested when root dry matter accumulation was evaluated. Such parallel results are also guite normal.

Crop growth rate (CGR): This trait also responded significantly to  $\rm H_2O_2$  concentration and variety (Tables 1 and 2). Both the two highest crop growth rates (3.28 and 10.51) at 10 and 20 DAS were obtained from 2.0% solution (Table 1). However, they were statistically identical with the results from 1.5 and 2.5% concentrations in both the situations. Almost similar trend was also manifested with Parvani kanti, though Anamika was still superior performer. In combined effect (Table 2), the topmost crop growth rate was also experienced from 2.0% concentration with Anamika variety.

Correlation studies: From the correlation matrix (Table 3), it would be clear that, all the studied parameters were significantly associated. Those relationships are in the optimistic ways, because all of them possess only positive values. Of them, the highest value (0.946) was recorded with root length and root dry matter. On the contrary, the least but still significant figure was obtained between shoot dry matter and CGR at 20 days (0.464). Roberts and Osei-Bonsu (1988) claimed that, if care is taken to evaluate the relationships in biologically meaningful terms, it becomes evident that, most of the important attributes of seed vigour are indeed closely related.

The results of the present experiment suggested that presoaking in 1.5 to 2.5 %  $\rm H_2O_2$  solutions could increase seed % germination and vigour index in okra seed. The results further proved that, imbibition of slightly deteriorating okra seeds in  $\rm H_2O_2$  solution could be used in restoring its viability. In addition, seeds of the variety Anamika had more germination capacity, higher vigour index and early growth rate over the variety Prabhani kranti.

Bangladeshi farmers are poor. In addition, their level of technical knowledge is also very shallow. On the other hand, any technology that is scientifically sound must be economically viable and socially acceptable for ease of adoption. In this regard, pre-soaking seed in  $1.5 \cdot 2.5 \% \; H_2O_2$  solution is very cheap (cost involved for treating lady's finger seed/hectare will be about 50.00 Tk.). The chemical is available in any local medicine shop, non-toxic to human and animal and at the same time treatment procedure is also very simple. But before final recommendation, it is advisable to conduct further experiment for finding optimum-presoaking time, as it is also an important aspect to derive maximum benefit. Furthermore, field experiment should be done to assess yield advantages.

### References

- Bordovsky, J.P., W.M. Lyle and A.B. Onden, 1991. Cottonseed preparation for hydraulic planter. Agron. J., 83: 275-278.
- Choi, B.H., K.Y. Park and R.K. Park, 1989. Effects of presoaking seed and planting depth on dormancy breaking and seedling emergence of pearl millet (*Pennisetum* americannum) (L) Leeke) Korean J. Crops Sci., 34: 81-85.
- Copeland, L. O. and M. B. McDonald, 1985. Principles of Seed Science & Technology. Macmillan Publishing Co. USA.
- Heydecker, W., 1977. Stress and seed germination: an agronomic view. In the physiology and biochemistry of seed dormancy and germination, A. A. Khan, ed. Amsterdam: North Holland Publishing Company,
- ISTA (International Seed Testing Association), 1985. International rules for seed testing. Rules 1985. Seed Sci. and Tech., 13: 356-513.
- Kindiger, B., 1994. A method of enhance germination of eastern gamma grass (*Tripsacum dactyloides* L.) Maydica, 39: 53-56.
- Lai, T.B., 1988. Effect of seed presoaking with  $\rm H_2O_2$  on germination, growth and esterase isoenzymes in groundnut. Oil Crop of China, 3: 33-36.
- Maguire, J.D., 1962. Speed of germination-aid in selection and evaluation for seedling emergence and vigour. Crop Sci., 2: 176-197.
- Msango, H.P. and J.A. Maghembe, 1989. Physical scarification and hydrogen peroxide treatment improves germination of *Vigna infansta* seed. Forest ecology and Management, 28: 301-308.
- Orphanos, P.I. and W. Hydecker, 1968. On the nature of soaking injury of *Phaseolus vulgaris* seeds. J. Exp. Bot., 19: 770-784.
- Radford, P.J., 1967. Growth analysis formula- their use and abuse. Crop Sci., 7: 171-175.
- Rahman, M.M., M.M. Rahman and M.N. Islam, 1997. Effect of H<sub>2</sub>O<sub>2</sub> pre-soaking on germination and early growth of groundnut seeds. Bangla. J. Seed Sci. & Tech., 1: 63-68.
- Roberts, E.H. and K. Osei-Bonsu, 1988. Seed and Seedling vigour. pp. 898-910. In Summerfield, R.J. (ed.) World Crops: Cool Season Food Legumes. Kluwer Academic Publishers. London.
- Steel, R.C.D. and J.H. Torrie, 1960. 'Principles and Procedures of Statistics'. McGraw-Hill Book Co. Inc., New York, pp. 107-109.