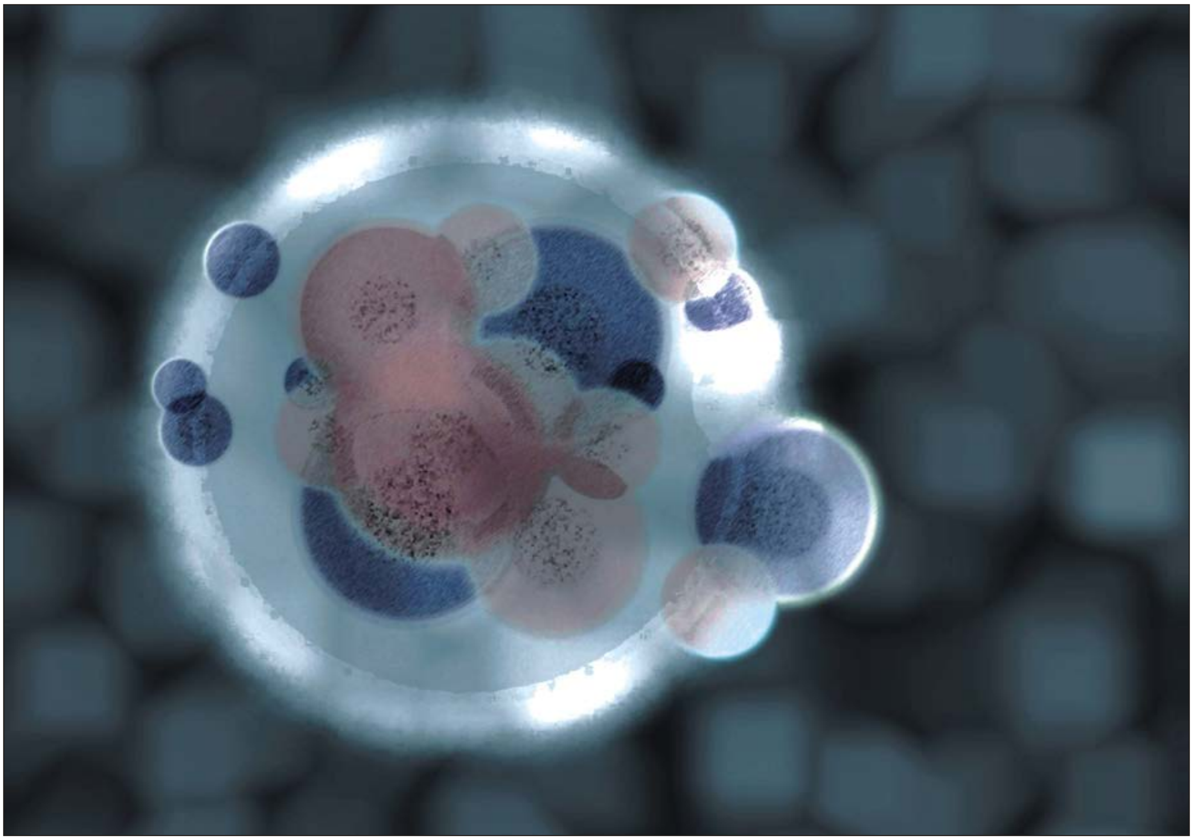

Journal of Biological Sciences

ISSN 1727-3048

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Journal of Biological Sciences 5 (2): 163-169, 2005
ISSN 1727-3048

Effect of Atonik Seed Treatment on Seedling Physiology of Cotton and Tomato

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Abstract: A lab experiment was conducted to evaluate the effect of Atonik as seed treatment on germination, establishment, growth and various biochemical parameters of cotton and tomato. Seed treatment of Atonik at 3 ppm in both cotton and tomato was found to be best in recording maximum germination, establishment and enzyme activity on concentration basis. The increase in seedling physiological parameters may be correlated with internal auxin content.

Key words: Atonik, seed treatment, growth and antioxidant enzymes

INTRODUCTION

Among the physiological constraints that limit the economic yield of cotton and tomato, poor germinability, dormancy, low seedling emergence and establishment are the most important because these lead to an uneven and sparse plant population[1]. Seedlings with high vigour will establish quickly and produce vigorous growth. Plant growth hormones are known to regulate the physiological process during seed germination[2], which are very crucial to increase the rate and percentage of germination. Atonik is an aromatic nitro phenolic compound, which consists of sodium ortho-nitrophenol, para-nitrophenol and sodium nitro-guaiacol as active ingredients, Atonik stimulates plant activity without causing malformation or toxicity to the plants and accelerates the plasma streaming of the cells by increase in the endogenous auxin level[3]. Phenolic acids are natural compounds of several plants, which can influence cell morphology, physiology and metabolism. It has also been observed that these compounds can affect seed germination. The influence of p-hydroxybenzoic acid on seed germination and vigour were studied in soybean seedlings, a significant reduction in the length, fresh and dry biomass of radicals and increase in protein content was observed as concentration of phenolic acid was increased[4].

In cotton, the effect of three phenolic acids viz., ferulic, vanillic and p-hydroxy benzoic acids on seed germination and seedling growth, was studied and it was observed that at 200, 100 and 50 mg L^{-1} germination was inhibited as dose increased[5]. These chemicals enhanced seedling growth and establishment parameters[6]. In onion, IAA 100 ppm recorded the highest germination percentage, vigour index and dry weight[7]. In germinating oily seeds, H_2O_2 production may also result from mobilization of stored reserves by β-oxidation in glyoxysomes[8]. Several deleterious effects of reactive oxygen species (ROS) on cellular processes have been described[9], but they can also be beneficial to cellular functioning as they are involved in molecular signalling[10]. ROS levels are therefore tightly controlled in the cells through the activity of enzymes such as superoxide dismutase (SOD), which eliminates O_2, catalase (CAT) and peroxidase (POX), which eliminate H_2O_2 and antioxidants such as ascorbic acid and glutathione. In sunflower, the seed germination rate is closely correlated with CAT activity[11]. Hence, the present study was designed to investigate the possible implication of seed treatment with nitrophenols (Atonik, a nitrophenolic compound) in cotton and tomato to increase the crop stand. The biochemical mechanism of action of Atonik on seedling physiological parameters was also studied in the present investigation.

MATERIALS AND METHODS

Seed materials of cotton variety MCU 12 collected from Cotton Breeding Station and tomato variety PKM1 obtained from Department of Vegetable Crops, Tamil Nadu Agricultural University, Coimbatore were used for the study. A laboratory experiment was undertaken to fix the optimum concentration of Atonik in cotton and tomato for seed treatment. The treatments involves six concentration of Atonik along with control, viz.,

T_1-Control
T_2-Seed treatment of Atonik with 1 ppm
T_3-Seed treatment of Atonik with 2 ppm

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T_4-Seed treatment of Atonik with 3 ppm
T_5-Seed treatment of Atonik with 4 ppm
T_6-Seed treatment of Atonik with 5 ppm
T_7-Seed treatment of Atonik with 6 ppm

The following observations were recorded in cotton and tomato:

Germination percentage: Standard germination tests were carried out by adopting the procedure detailed by ISTA[12] for cotton. Tomato germination test was performed using between paper towels medium, after 12 and 10 days the germination percentage was worked out following ISTA[13] rules for cotton and tomato, respectively.

Field emergence: For assessing tomato field emergence, 100 seeds in eight replications were dibbled in raised beds. On the 12th day, the seedlings were carefully removed, washed and the normal seedlings alone counted and expressed as percentage of germination in the field. For cotton, the number of healthy seedlings in the field was counted 14 days after sowing for every hundred seeds with eight replications and expressed as field emergence percentage.

Shoot and root length: On the 12th day after germination for cotton and 10th day for tomato, ten seedlings from each replication were carefully removed at random. Length of shoot was measured in 'cm' from the ground level to the tip of the longest leaf. Root length (cm) was measured from the base of the stem to the tip of the longest root. The average of eight replications was calculated and recorded.

Shoot and root weight: On the 12th day after germination for cotton and 10th day for tomato, ten seedlings from each replication were carefully removed at random. The shoot and root were separated and dried in a hot air oven at 80°C for 24 h. The average of eight replications was calculated and expressed in mg.

Vigour index (VI) (based on shoot and root length): The vigour index of the seedling was calculated by multiplying the total length of the seedling and germination percentage and expressed as number[13].

Biochemical parameters: Total protein was extracted on the 12th day after sowing from the leaf samples of cotton and tomato and quantified by the method of Lowry *et al.*[14] and expressed as $\mu g\ g^{-1}$ of tissue. Total carbohydrate content was estimated from 12 day old seedlings by ethanol extraction method as suggested by Hedge and Hofreiter[15] expressed as $\mu g\ g^{-1}$ fresh weight. Total free amino acid content was estimated from ethanol extract of leaves by the reaction with ninhydrin solution[16] on the 12th day after sowing. It was expressed as $\mu g\ g^{-1}$ of fresh weight. Catalase activity was assayed on the 12th day after sowing. The procedure was adopted from Gopalachari[17] and expressed as mg $H_2O_2\ g^{-1}$. Peroxidase was assayed on the 12th day after sowing according to Perur[18] and expressed as change in optical density/g for five min. Polyphenol oxidase was quantified on 12th day after sowing and expressed as change in optical density/g for five min. The method described by Bateman and Daly[19] was followed.

RESULTS

Germination percentage: Germination percentage showed an increasing trend from 1 ppm (T_2) to 6 ppm (T_7). In both tomato and cotton maximum germination percentage (Table 1 and 2) was observed in 6 ppm (T_7) followed by 5 ppm (T_6). Treatments viz., T_4, T_5, T_6 and T_7 were on par with each other. The treatments recorded an increase of 13.2, 13.6, 15.2 and 15.5% over control for tomato. For cotton, the treatments viz., T_4, T_5, T_6 and T_7 were on par with each other. These treatments recorded an increase of 29.2, 29.9, 30.4 and 30.8% over control. However, there exists a significant difference between the other treatments.

Field emergence (%): Atonik treatments significantly increased the field emergence %. Field emergence increased with increasing concentration (Table 1 and 2). Maximum emergence was observed in 6 ppm (T_7) treatments in tomato and cotton. This treatment recorded an increase of 19.7% over control for tomato and 46.9% for cotton over control. The treatments viz., T_4, T_5, T_6 and T_7 were on par with each other for tomato and cotton.

Shoot length (cm): Results observed from the experiment showed that Atonik treatment increased the shoot length from 5.83 to 7.33 in control (T_1) and 6 ppm (T_7), respectively in tomato and from 7.73 to 12.70 in cotton, respectively (Table 1 and 2). Though there exists a significant difference among the treatments, T_5, T_6 and T_7 were on par with each other for tomato and cotton. These treatments increased the shoot length by 20.1, 20.3 and 20.4% over control for tomato and 38.60, 38.80 and 39.10% over control for cotton, respectively.

Root length (cm): Root length was found to be significantly increased by Atonik treatments. Maximum value for root length was observed in T_7 followed by T_6 in both tomato and cotton (Table 1 and 2). T_7 recorded a maximum value of 9.24 for tomato and 17.30 for cotton.

Table 1: Effect of Atonik on seedling physiology of tomato Var. PKM 1

Treatments	Germination (%)	Field emergence (%)	Shoot length (cm)	Root length (cm)	Vigour index	Total protein content ($\mu g\ g^{-1}$)	Total carbohydrate content ($\mu g\ g^{-1}$)	Total free amino acid content ($\mu g\ g^{-1}$)	Catalase activity ($mg\ H_2O_2\ g^{-1}$)	Peroxidase activity (ΔOD 430 nm g^{-1} 5 min^{-1})	Polyphenol oxidase activity (ΔOD 495 nm g^{1} 5 min^{-1})	Shoot dry weight (mg $plant^{-1}$)	Root dry weight (mg $plant^{-1}$)	Total dry matter production (mg $plant^{-1}$)
T_1	85.330	(68.20)	69.00	(56.53)	5.83	7.16	1070.71	105.33	360.3200	92.8800	0.3910	0.413	0.2140	2.250
	0.673	2.92												
T_2	91.000	(73.93)	74.00	(59.83)	6.10	7.92	1274.31	140.34	400.2800	101.5200	0.4030	0.516	0.2870	2.950
	0.804	3.76												
T_3	94.330	(72.10)	78.33	(60.07)	6.80	8.33	1419.42	180.72	420.5200	108.3600	0.4120	0.573	0.3060	3.010
	0.802	3.83												
T_4	96.660	(83.07)	82.66	(66.27)	7.20	8.94	1545.60	216.82	480.3300	120.7200	0.4820	0.624	0.3100	3.230
	0.830	4.06												
T_5	97.000	(73.83)	82.33	(62.13)	7.30	9.13	1581.13	218.16	483.6600	123.6600	0.4860	0.642	0.3430	3.300
	0.832	4.13												
T_6	98.330	(77.56)	82.00	(63.03)	7.32	9.23	1599.36	224.66	486.4600	124.5400	0.4910	0.663	0.3420	3.340
	0.836	4.17												
T_7	98.660	(78.46)	82.66	(63.57)	7.33	9.24	1610.14	228.52	496.0000	128.5400	0.4930	0.694	0.3440	3.360
	0.841	4.20												
Mean	94.470	(75.30)	78.71	(61.63)	6.78	8.40	1431.99	185.06	443.1500	112.8800	0.4470	0.581	0.3020	3.040
	0.799	3.83												
CD (p=0.05)	4.776	2.68	0.132	0.164	48.060	10.69	11.66	10.20	0.0245	0.0112	0.0161	0.059	0.0152	0.212

Number in parentheses indicates log transformed value

Table 2: Effect of Atonik on seedling physiology of cotton Var. MCU 12

Treatments	Germination (%)	Field emergence (%)	Shoot length (cm)	Root length (cm)	Vigour index	Total protein content ($\mu g\ g^{-1}$)	Total carbohydrate content ($\mu g\ g^{-1}$)	Total free amino acid content ($\mu g\ g^{-1}$)	Catalase activity ($mg\ H_2O_2\ g^{-1}$)	Peroxidase activity (ΔOD 430 nm g^{-1} 5 min^{-1})	Polyphenol oxidase activity (ΔOD 495 nm g^{1} 5 min^{-1})	Shoot dry weight (mg $plant^{-1}$)	Root dry weight (mg $plant^{-1}$)	Total dry matter production (mg $plant^{-1}$)
T_1	72.33	(58.70)	60.330	(51.30)	7.73	9.85	1195.44	505.42	410.3300	200.0000	0.3100	0.385	0.214	62.160
	7.32	69.46												
T_2	76.66	(61.60)	67.660	(55.73)	9.00	11.25	1539.16	570.16	500.3300	240.3300	0.4220	0.502	0.326	70.120
	9.24	79.32												
T_3	81.00	(61.83)	72.330	(56.40)	10.15	14.00	1956.15	610.33	532.3300	275.6600	0.4820	0.614	0.436	79.330
	10.30	89.63												
T_4	93.00	(76.27)	88.000	(70.87)	12.40	16.50	2687.70	645.00	580.0000	312.4800	0.5140	0.824	0.554	86.590
	12.24	98.79												
T_5	94.00	(70.93)	88.000	(66.10)	12.60	17.05	2787.10	650.66	592.6600	320.0000	0.5260	0.841	0.572	87.230
	12.41	99.63												
T_6	94.33	(73.00)	88.330	(67.70)	12.65	17.25	2810.62	650.66	593.0000	320.3300	0.5320	0.862	0.574	87.590
	12.43	100.02												
T_7	94.66	(73.50)	88.660	(68.03)	12.70	17.30	2820.00	660.00	594.3300	325.6600	0.5400	0.874	0.586	87.920
	12.51	100.43												
Mean	86.57	(68.04)	79.040	(62.30)	10.95	14.63	2239.73	606.94	537.4500	282.2000	0.4710	0.692	0.458	80.130
	10.92	91.04												
CD (p=0.05)	3.67	2.78	0.578	0.761	114.95	33.41	29.38	15.06	0.0251	0.0241	0.0238	0.148	0.101	0.314

Number in parentheses indicates log transformed value

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Whereas control (T_1) showed a value of 7.16 and 9.85 for tomato and cotton and it was 22.6 and 43.0% decrease over the best treatment, respectively. There exists a significant difference among the treatments.

Vigour index: Maximum vigour index was recorded in T_7 (1610.14 and 2820.00 for tomato and cotton, respectively) (Table 1 and 2). There exists a significant difference among the treatments. The treatments viz., T_6 and T_7 were on par with each other for tomato and T_5, T_6 and T_7 for cotton. These treatments recorded an increase of 47.1, 49.3 and 50.3 for tomato and 33.1, 35.1 and 35.8% for cotton over control, respectively.

Total protein content (µg g^{-1}): Increase in total protein content was observed by Atonik treatments. Atonik treatment at 6 ppm recorded the highest protein content (228.52 and 660.00 for tomato and cotton, respectively) followed by T_5 (5 ppm) in tomato and T_4 and T_5 in cotton (Table 1 and 2). Whereas control had a lower content of 105.33 and 505.42 for tomato and cotton, respectively. There exists a significant difference among the treatments.

Total carbohydrate content (µg g^{-1}): Total carbohydrate increased from 360.32 to 496.00 for tomato and 410.33 to 594.33 for cotton (Table 1 and 2). The differences in total sugar content were significant among the treatments. Maximum carbohydrate content was observed in T_7 (6 ppm) followed by T_6 (5 ppm) in both tomato and cotton. The increase was 37.6 and 34.9% and 44.8 and 44.5% over control for tomato and cotton, respectively.

Total free amino acid content (µg g^{-1}): Total free amino acid followed the trend of total protein content (Table 1 and 2). The increase was 38.3, 34.0% and 30.5 and 28.7% over control for tomato and cotton, respectively. Mean values over treatments showed significant differences.

Catalase activity (mg H_2O_2 g^{-1}): It is inferred from the table that Atonik treatments increased the activity of catalase enzyme. Maximum enzyme activity was noticed in T_7 (6 ppm), followed by T_6 (5 ppm) (Table 1 and 2). The treatments viz., T_4, T_5, T_6 and T_7 and T_6 and T_7 were on par with each other for tomato and cotton, respectively and recorded an increase of 24.2, 25.5, 26.0% and 71.6 and 74.1% over control for tomato and cotton, respectively. There exists a significant difference among the treatments.

Peroxidase activity (ΔOD 430 nm g^{-1} 5 min^{-1}): The activity of peroxidase increased due to Atonik seed soaking. At 6 ppm (T_7) maximum activity was observed in both tomato and cotton (0.694 and 0.874, respectively) followed by T_6 (0.663 and 0.862) for tomato and cotton, respectively (Table 1 and 2). The increase in enzyme activity was 68.0 and 127.0% in tomato and cotton, respectively over control.

Polyphenol oxidase activity (ΔOD 495 nm g^{-1} 5 min^{-1}): Polyphenol oxidase enzyme activity showed an increasing trend from 1 to 6 ppm. T_7 (6 ppm), being a treatment of a higher concentration, proved its effectiveness by recording the highest activity of 0.344 and 0.586, which was 116.0 and 173.8% increase over control for tomato and cotton, respectively (Table 1 and 2). The differences in treatment were significant.

Shoot dry weight (mg $plant^{-1}$): Shoot dry weight followed the trend of shoot length. Maximum dry weight was observed in T_7 followed by T_6 (Table 1 and 2). The treatments T_6 and T_7 were on par with each other for tomato and cotton, respectively. The best treatment T_7 recorded an increase of 49.3 and 41.4% over control for tomato and cotton, respectively. This was followed by T_6 (48.4 and 40.9% for tomato and cotton, respectively over control). There exists a significant difference among the treatments.

Root dry weight (mg $plant^{-1}$): Atonik seed treatment significantly increased the root dry weight over control. Among the treatments, T_7 (6 ppm) recorded the maximum root dry weight in both tomato and cotton (0.841 and 12.51, respectively) followed by T_6 (5 ppm) (0.836 and 12.43%, respectively). Control recorded minimum root dry weight (0.673 and 7.32%) for tomato and cotton, respectively. All the treatments showed significant differences (Table 1 and 2).

Total dry matter production (mg $plant^{-1}$): Total dry matter production was highly influenced by Atonik treatment. All the treatments showed significant differences over control (Table 1 and 2). Among treatments, T_7 (6 ppm) recorded the highest total dry matter production in tomato and cotton (4.20 and 100.43%, respectively) followed by 6 ppm (4.17 and 100.02%, respectively). This recorded an increase of 43.8, 44.5, 42.8 and 43.9% over control for tomato and cotton, respectively.

DISCUSSION

Perusal of previous literature reveals that, if a seed acquires primary and secondary dormancy, seed soaking in plant growth regulators will relieve the dormancy[2]. In the present study it was revealed that Atonik 3 ppm concentration was optimum for seed soaking, since an early germination and uniform establishment of cotton and tomato seedlings could be observed. As suggested by the previous reports that phenolic compounds exhibited an inhibitory effect on seed germination[20], no such deleterious effect was noticed upto six ppm (T_7) in both cotton and tomato.

The seedling growth and vigour, as measured through root length, shoot length and germination %, were also high in seeds treatment with Atonik at 6 ppm (T_7). The same treatment surpassed other treatments in all biochemical (protein, total free amino acids and sugar) and enzyme assays (catalase, peroxidase and polyphenol oxidase).

The increase in the above said parameters may be attributed to increase in endogenous auxin level by the phenolic compound like Atonik[21,22]. Growth promoted by IAA may be assigned to the increased cell wall plasticity by its action. Alterations in the amount and type of RNA and protein synthesis in Atonik treated tissues, to increase the activity and/or amount of RNA polymerases and to enhance selectively the activity of other enzymes may suggest enzyme induction. The increased cellulase, in this manner, may have involved in cell wall loosening during auxin induced growth[23]. Tomaszewski and Thimann[24] have reported the synergistic interaction of IAA and phenols in oat and pea seedling physiology. The results presented here are in line with these findings. Growth promoting activity of phenols has also been reported by Datta *et al.*[20], Toyal and Sharma[25].

During seed germination, seed protein plays an important role both as food reserve and as hydrolysing enzyme, which are involved in breaking down of starch[26]. Co-incidentally, the seed protein content was also the highest with Atonik six ppm treatment[27,28]. The increase in total protein content and total free amino acids may be attributed to increase in amino acid synthesis of aspartic and glutamic acids[29].

A number of antioxidants and metal chelating agents have been shown to play a significant role in counteracting free radical reactions[30]. Compounds like phenols are able to reduce the damage caused by reactive oxygen species[31]. Improvement of seedling growth rate by Atonik seed treatment was clearly associated with higher catalase, peroxidase and polyphenol oxidase activities. As already suggested by Bailly *et al.*[32] the high antioxidant enzyme activity might be due to its enhanced synthesis during germination. Seedling growth rate is related to the efficiency of H_2O_2 elimination, since higher the antioxidant enzyme activity, faster the seedling development. This was observed in Atonik treatment, as activities of all the antioxidant enzymes viz., catalase, peroxidase and polyphenol oxidase were more. However, H_2O_2 together with other ROS may have many cellular targets, such as proteins and DNA to denature[9]. Alternatively, catalase also plays a key role in H_2O_2 removal during fatty acid β-oxidation in glyoxysomes[33]. Therefore, high catalase activity in Atonik treatment could be associated with better mobilization of lipid reserves and faster seedling development. Finally, the regulation of H_2O_2 homeostasis by catalase, peroxidase and polyphenol oxidase could be important in terms of the control of H_2O_2 dependent signalling pathway, which has been shown to involve many gene expressions including that of lipoxygenase[34].

The increase in dry matter in Atonik treatment was probably due to better uptake and accumulation of the mineral nutrients[35]. Auxin is known to promote cell elongation and protein synthesis[36]. The increase in shoot length and root length in Atonik treated plants has led to the overall assimilation and redistribution of food materials within the plant[37] and hence, resulted in high seedling biomass production. As Atonik may induce IAA biosynthesis in plant[22], thus, increased growth is a consequence of increased dry matter accumulation.

Among the seed treatments imposed, Atonik 3 ppm (T_4) was found to be economically sound, since higher concentrations upto 6 ppm, were on par with T_4. Therefore, it may be concluded that seed treatment of Atonik at three ppm for cotton and tomato may be administered to improve germinability, seedling growth and to get a better crop stand.

REFERENCES

1. De Castro, R.D., A.A.M. Van Lammeren, S.P.C. Groot, R.J. Bino and H.W.M. Hilhorst, 2000. Cell division and subsequent radicle protrusion in tomato seeds are inhibited by osmotic stress but DNA synthesis and formation of microtubular cytoskeleton are not. Plant Physiol., 122: 327-335.
2. Khan, A.A., 1977. The Physiology and Biochemistry of Seed Dormancy and Germination. Amsterdam, Elsevier Scientific Publications Company, pp: 77-175.
3. Datta, K., S. Premsagar, R.C. Hasija and R.L. Kapoor, 1986. Effect of Atonik, miraculan and phenols on growth and yield of pearl millet. Ann. Biol., 2: 9-14.
4. Sehuab, S.R.P., A.L. Braccini, O. Ferrarese Filho, C. Scapim and M.C.L. Braccini, 2001. Physiological seed quality, seedling lipid and protein content of soybean in response to different concentrations of p-hydroxy benzoic acid. Seed Technol., 23: 126-137.
5. Wang, Pu and Zhao Xiuqui, 2001. Effect of allelochemicals on cotton seed germination and seedling growth. J. China Agri. Uni., 6: 26-31.

6. Karan, S. and B.S. Afria, 1990. Seed germination, seedling growth, emergence and establishment responses to cotton cultivars as regulated by growth substances. Seed Res., 18: 25-30.
7. Kanaujia, V.P., C.P. Sachan and S.K. Tripathi, 2002. Effect of growth regulators and stratification on germination and vigour of onion seed. Seed Res., 30: 155-157.
8. Bewley, J.D. and B. Black, 1994. Seeds. Physiology of Development and Germination. 2nd Edn., New York, Plenum Press.
9. Scandalios, J.G., 1997. Oxidative Stress and the Molecular Biology of Antioxidant Defenses. New York, Cold Spring Harbor Laboratory Press, USA.
10. Inze, D. and M. Van Montagu, 1995. Oxidative stress in plants. Curr. Opin. Biotech., 6: 153-158.
11. Bailly, C., A. Benamar, F. Carl Ineau and D. Come, 1998. Free radical scavenging as affected by accelerated ageing and subsequent priming in sunflower seeds. Physiol. Plant., 104: 646-652.
12. ISTA., 1966. International seed rules for seed testing. Proc. Int. Seed Test Ass., 31: 1-152.
13. ISTA., 1976. International rules for seed testing. Seed Sci. Technol., 4: 3-49.
14. Lowry, O.H., N.H. Rosen brough, A.L. Farr and R.J. Randall, 1951. Protein measurement with folin-phenol reagent. J. Biol. Chem., 193: 265-275.
15. Hedge, J.E. and B.T. Hofreiter, 1962. In: Carbohydrates Chemistry, 17 vol. (Eds.) Whistler, R.L. and J.B. Be Miller. Academic Press, New York.
16. Sadasivam, S. and A. Manickam, 1996. Biochemical Methods. 2nd Edn. New Age International Publishers and Tamil Nadu Agricultural University, pp: 2.
17. Gopalachari, N.C., 1963. Changes in the activities of certain oxidising enzymes during germination and seedling development of *Phaseolus mungo* and *Sorghum vulgare*. Indian J. Expl. Biol., 1: 98-100.
18. Perur, N.G., 1962. Measurement of peroxidase activity in plant tissue. Curr. Sci., 31: 17-18.
19. Bateman, D.F and J.M. Daly, 1967. The respiratory pattern of *Rhizoctonia* infected bean hypocotyls in relation to lesion maturation. Phytopatholgy, 57: 127-131.
20. Datta, K.S., S. Kumar and K.K. Nanda, 1979. Effect of some diphenols and gibberellic acid on the growth and development of common millet. Ind. J. Agric. Sci., 49: 179-184.
21. Kafeli, V.I. and M. Kutacek, 1977. Phenolic substances and their possible role in plant growth regulation. Proc. 9th International Conf. Plant Growth Substances, (Ed. Pilet, P.E.), pp: 181-188.
22. Yadav, A.C., M.L. Pandita, Avtar Singh and G.P. Singh, 1987. Effect of cytozyme, Atonik, miraculan and mixatol on pea var. Bonneville. Haryana J. Agron., 8: 75-77.
23. Goren, R. and E. Tomer, 1971. Effect of seselin and coumarin on growth, indoleacetic acid oxidase and peroxidase with special reference to cucumber (*Cucumis sativa*) radicles. Plant Physiol., 47: 312-316.
24. Tomaszewski, M. and K.V. Thimann, 1966. Interaction of phenolic acids, metallic ions and chelating agents on auxin–induced growth. Plant Physiol., 41: 1443-1454.
25. Tayal, M.S and S.M. Sharma, 1985. Interaction of phenols and indoleacetic acid on germination and early seedling growth of *Cicer arietinum* L. Indian J. Plant Physiol., 28: 271-276.
26. Copeland, L.O., 1988. In: Principles of Seed Science and Technology. (Ed. Chhabra, S.S.), Delhi, Surjeet Publication, pp: 37-39.
27. Muntz, K., M.A. Belozersky, Y.E. Dunaevsky, A. Schlereth and J. Tiedemann, 2001. Stored proteinases and the initiation of storage protein mobilization in seeds during germination and seedling growth. J. Exp. Bot., 52: 1741-1752.
28. Vasudevan, S.N., K. Virupakshappa, S. Bhaskar and A. Seetharam, 2000. Seed quality of sunflower genotypes in relation to growth regulator application. Seed Res., 28: 1-4.
29. Hafez, S.A., S.A. Ismael and M.H. Mahmoud, 2000. Chemical changes of soybeans during germination. Egyptian J. Agri. Res., 78: 795-817.
30. Umarani, R., 1999. Studies on the physiological and biochemical basis of seed germination and deterioration in *Casuarina equisetifolia*. Ph.D (Ag.) Thesis (submitted), Tamil Nadu Agricultural University, India.
31. Pathak, G., 1980. Seed treatment for vigour viability and productivity of some oil yielding crops. Ph.D. Thesis, (submitted), Calcutta University, India.
32. Bailly, C., A. Benamar, F. Corbineau and D. Come, 2000. Antioxidant systems in sunflower (*Helianthus annuus* L.) seeds as affected by priming. Seed Sci. Res., 10: 35-42.
33. Olsen, L.J. and J.J. Harada, 1995. Peroxisomes and their assembly in higher plants. Annu. Rev. Plant Physiol. Plant Mol. Biol., 46: 123-146.
34. Desikan, R., S.A.H. Mackerness, J.T. Hancock and S.J. Neill, 2001. Regulation of the *Arabidopsis* transcriptome by oxidative stress. Plant Physiol., 127: 159-172.

35. Talwar, K.K. and H.P. Bhatnagar, 1978. Effect of growth regulators on fresh and dry matter, hemicellulose production and mineral uptake by seedlings in *Pinus caribaea.* Indian For., 104: 544.
36. Davis, P.J., 1973. Current theories of the mode of action of auxin. Bot. Rev., 39: 139-171.
37. Brian, P.W. and H.G. Hemming, 1955. The effect of GA on shoot growth of pea seedlings. Physiol. Plant, 8: 669-681.