Annals of Plant and Soil Research 26(3): 387-394 (2024)
https://doi.org/10.47815/apsr.2024.10375

Enhancement of physiological and biochemical attributes of low vigour brinjal (S*olanum melongena* L.) seeds through various priming treatments

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Received, Febuary, 2024; Revised accepted, May, 2024

Abstract

Brinjal (Solanum melongena L.) seeds are known to undergo loss of vigour and viability. Hence, the present study was conducted on brinjal var. Punjab nagina to assess its seed viability and vigour. The freshly harvested seeds were divided into two groups. In order to study the effect of various priming treatments one group was converted to a low vigour by exposing the seeds to accelerated ageing, whereas the other group retained as high vigour. Both these groups were subjected to priming with KNO_3, KH_2PO_4, GA_3, and PEG. The seeds were then kept in moisture-proof packets and stored in a refrigerator for 12 months at 4^OC. The seeds were drawn after zero, three, six, nine, and twelve months of storage and total soluble sugars, total soluble proteins, α-amylase activity, starch content and total free amino acids were estimated. The biochemical attributes of seed quality declined as the storage duration increased in both high and low vigour seeds. Priming treatments significantly enhanced the physiological and biochemical attributes in both high and low vigour seeds although the degree of improvement varied. Even after being stored for one year, in the seeds primed with GA_3 (100 ppm) and KNO_3 (1%) the quality parameters were at par to or higher than the seeds that were not subjected to any storage. Hence, employing GA_3 and KNO_3 priming for 12 hours is suggested to enhance the quality of brinjal seeds and improve their storage longevity.

Keywords: Brinjal, GA_3, Germinability, Longevity, Priming, Quality parameters

INTRODUCTION

Brinjal (S*olanum melongena* L.) is a warm-season crop and grows well under tropical and subtropical conditions. High-quality seed is a pre-requisite for successful seedling establishment and an appropriate plant population in the field. Seed viability and vigour are two crucial characteristics that define seed quality. Factors such as genetic composition, environment during seed production, seed moisture, mechanical damage, pathogens, insect, pests, seed dressing chemicals and seed treatments affect subsequent field emergence. During prolonged storage, seed quality is known to decline (Shaban, 2013) leading to poor and delayed germination, slow rate of field establishment and ultimately leading to lower yield and hence economic loss to the farmers. In order to ameliorate deteriorative changes in the seeds, seed priming treatments are employed as they reduce germination time, enhance the uniform field emergence, increase germination percentage and improve seed vigour (Nawazetal.,2013) and enhance pre-germination metabolic process for rapid germination (Chen *et al.*, 2010). Seed priming involves controlled hydration in an appropriate priming solution for suitable duration. Priming induces imbibition and pre-germinative biochemical changes such as enzyme activation (Farooq *et al.*, 2010) and repairs any cellular and sub-cellular metabolic damage that may have occurred (Aswin *et al.*, 2019). To improve the growth of seedlings and the establishment of the crop, seed-based technologies such as innovative seed improvements and seed treatment methods like priming, pelleting, coating, synthetic seeds, etc. were employed (Shantharaja *et al.*, 2024). These methods enhance the growth kinetics and thus ensure better seed establishment (Sharma *et al.*, 2020).

A range of priming agents and techniques viz., hydro-priming (soaking seeds in pre-determined amounts of distilled water), osmo-priming (soaking seeds in osmotic solutions like PEG), hormonal priming (treating seeds with hormones like GA_3), halo-priming (soaking seeds in solution containing in organic salts) and solid matrix priming (mixing seeds with organic or inorganic solid materials and water indefinite proportions) (Venkata Subramanian & Umarani, 2007) have delivered promising results in several species such as in

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round gourd (Singh *et al.*, 2023), lettuce (Fessel *et al.*, 2002), sugarbeet (Costa & Villela, 2006), onion (Caseiro *et al.*, 2004), carrot (Pereira *et al.*, 2009), cauliflower (Kikuti and Filho, 2009), cucumber (Lima & Filho, 2010), chili (Fialho *et al.*, 2010), pepper (Albuquerque *et al.*, 2009), tomato (Venkatasubramanian & Umarani, 2007) and eggplant (Nascimento & Lima, 2008). However, it is important to standardize the priming treatments for seed species. In the present investigation, the Brinjal (*Solanum melongena* L.) var. *Punjab nagina* seeds were subjected to various priming treatments prior to storage. Physiological and biochemical studies were conducted at the end of the storage period to know the overall effect of priming on seed vigour.

MATERIALS AND METHODS

The freshly harvested seeds which had a germination percent of >80% (high vigour seeds) were selected for the studies. One seed lot was subjected to accelerated aging. The germination percentage was observed to be less than 65% and thus cited as low vigour seeds (Kaue *et al.*, 2023). In this study, both low and high vigour brinjal seeds were primed in solutions of KNO_3(1%), KH_2PO_4 (0.1M), PEG 6000 (30%), GA_3 (100 ppm), and distilled water by allowing them to imbibe the respective priming solution for 12 h (Kaur *et al.*, 2023; Singh *et al.*, 2023). The study on round gourd seeds belonging to the same family viz., Cucurbitaceae revealed that priming with KNO_3 was an effective treatment to ensure the germinability of the seeds with hard seed coats (Singh *et al.*, 2023). Primed seed lots were dried back to their pre-primed moisture, packed in moisture-proof aluminum bags, and placed in a refrigerator at 4^oC for 12 months. The seed lots were drawn at 3-month intervals (viz., 0, 3, 6, 9, and 12 months, respectively). The contents of total soluble sugars (Dubois *et al.*, 1956), total starch (Dubois *et al.*, 1956), total soluble proteins (Lowry *et al.*, 1951), total free amino acids (Lee & Takahashi 1956), and activity of α-amylase (Murata *et al.*, 1968) were estimated from both primed and unprimed seeds at all the stages. For statistical analysis, data were subjected to a two-way analysis of variance (ANOVA) by SAS software followed by Tukey's test to compute the individual comparison among storage durations and priming treatments.

RESULTS AND DISCUSSION

Seed priming has been demonstrated to improve the seed quality. This is attributed to the initiation of biochemical changes which include imbibition, hydrolysis of metabolic inhibitors, and enzyme activation in primed seeds (Kuppusamy and Ranganathan, 2014). Priming triggers few or all pre-germinative metabolic processes which persist in the seed even after redrying leading to fast reimbibition and quick revival of germination metabolism. In the present study, immediately after priming (0 months), under all the priming treatments, the total soluble sugars, total soluble proteins and α-amylase activity increased as compared to control in both high and low vigour seeds but the extent of increase varied between the two vigour levels and the priming treatments. While the starch content and total free amino acid content decreased in both the high and low vigour seeds immediately after priming (0 months) and the extent of the decrease varied.

Table 1a: Effect of pre-storage seed priming and storage duration on the amount of total soluble sugars (mg/g DW) in high vigour brinjal seeds

Priming treatments	Storage durations					
	0 months	3 months	6 months	9 months	12 months	Mean
1% KNO_3	$13.36±0.36^{a}$	$12.33±0.31^{a}$	$10.53±0.60^{b}$	$10.28±0.43^{bc}$	$10.03±0.57^{bcd}$	11.30^{A}
0.1M KH_2PO_4	$10.00±0.19^{bcd}$	$9.39±0.26^{b-e}$	$8.94±0.27^{b-g}$	$8.28±0.16^{e-h}$	$7.62±0.12^{f-i}$	8.84^{C}
30%PEG6000	$10.34±0.45^{bc}$	$9.65±0.24^{b-e}$	$8.79±0.38^{c-g}$	$8.24±0.38^{e-h}$	$7.69±0.40^{f-i}$	8.94^{BC}
$GA_3$100ppm	$10.30±0.51^{bc}$	$9.82±0.26^{b-e}$	$9.27±0.14^{b-f}$	$9.02±0.15^{b-g}$	$8.77±0.19^{c-g}$	9.43^{B}
H_2O	$9.18±0.07^{b-f}$	$8.80±0.18^{c-g}$	$7.46±0.08^{g-j}$	$6.68±0.24^{h-k}$	$5.90±0.10^{jk}$	7.60^{D}
CONTROL	$9.11±0.15^{b-g}$	$8.54±0.21^{d-g}$	$7.68±1.62^{f-i}$	$6.48±0.66^{ijk}$	$5.28±0.48^{k}$	7.42^{D}
MEAN	10.38^{A}	9.75^{B}	8.78^{C}	8.16^{D}	7.55^{E}	

*Means that are followed by different letters are significantly different (P≤0.05) according to Tukey's test; *Significant at P=0.05*

Immediately after priming, the maximum amount of total soluble sugars (TSS) was observed after treatment with 1% KNO_3 in both high (13.36 mg/g DW) and low vigour seeds (9.63 mg/gDW) while the amount was high in high vigour seeds. In general, with an increase in storage duration, the total soluble sugar content in the seeds decreased in all the primed seeds as well as non-primed seeds but the extent of the decrease varied and the amount of TSS was higher in the high vigour seeds than in the low vigour seeds (Table 1a,1b). Within, each storage duration the amount of total soluble sugars in low vigour seeds treated with 1% KNO_3 was either statistically at par or significantly more than the amount of total soluble sugars in respective high vigour control seeds (Table 1a, 1b).

Table 1b: Effect of pre-storage seed priming and storage duration on the amount of total soluble sugars (mg/g DW) in low vigour brinjal seeds

Priming treatments	Storage durations					
	0 months	3 months	6 months	9 months	12 months	Mean
1% KNO_3	9.63±1.62^{a}	8.77±0.66^{a-d}	8.61±0.48^{a-d}	8.33±0.31^{a-e}	7.55±0.33^{a-f}	9.94^{A}
0.1M KH_2PO_4	9.13±1.01^{ab}	8.40±0.57^{a-e}	7.04±0.20^{b-g}	7.00±0.10^{b-g}	6.95±0.16^{b-g}	8.32^{AB}
30%PEG6000	8.88±0.25^{abc}	7.62±0.21^{a-f}	6.44±0.21^{c-h}	6.34±0.26^{d-h}	6.24±0.76^{e-i}	8.02^{B}
$GA_3$100ppm	7.58±0.13^{a-f}	7.06±0.32^{b-g}	6.91±0.16^{b-h}	6.85±0.11^{b-h}	6.78±0.11^{b-h}	8.24^{B}
H_2O	5.73±0.27^{f-i}	5.66±0.19^{f-i}	5.06±0.14^{g-j}	4.46±0.11^{h-k}	3.86±0.12^{i-l}	6.28^{C}
CONTROL	3.16±0.13^{jkl}	2.57±0.19^{kl}	2.25±0.21^{kl}	2.03±0.19^{kl}	1.82±0.21^{l}	4.89^{D}
MEAN	7.35^{A}	6.68^{AB}	6.05^{BC}	5.83^{C}	5.71^{C}	

*Means that are followed by different letters are significantly different ($P \leq 0.05$) according to Tukey's test; *Significant at P=0.05*

Before storage, the minimum amount of starch content was recorded with GA_3 priming i.e. 4.35 mg/g DW and 8.27 mg/g DW in high and low vigour seeds respectively. Conversely, in general, high vigour seeds had lower starch content than low vigour seeds. At the end of each storage duration, the starch content in low vigour was higher than in the high vigour seeds (Table 2a, 2b).

Table 2a: Effect of pre-storage seed priming and storage duration on the amount of starch (mg/gDW) in high vigour brinjal seeds

Priming treatments	Storage durations					
	0 months	3 months	6 months	9 months	12 months	Mean
1% KNO_3	5.96±0.04 $^{c-i}$	5.79±0.08^{e-i}	5.69±0.17 $^{f-i}$	5.45±0.18 $^{g-j}$	5.20±0.27^{i-l}	5.62^{C}
0.1M KH_2PO_4	7.68±0.05 ^{ab}	7.14±0.22^{abc}	7.24±0.18^{ab}	7.07±0.21^{a-d}	6.89±0.24^{a-f}	7.20^{AB}
30%PEG6000	5.87±0.17 $^{d-i}$	5.45±0.09 $^{g-j}$	5.36±0.21^{h-k}	5.11±0.15 $^{i-l}$	4.85±0.34 $^{i-l}$	5.32^{C}
$GA_3$100ppm	4.35±0.15^{jkl}	4.34±0.29^{jkl}	4.32±0.06 ^{jkl}	4.21±0.05 ^{kl}	4.01±0.11 ^{l}	4.25^{D}
H_2O	6.95±0.41^{a-e}	6.90±0.26 $^{a-f}$	6.86±0.08 $^{a-f}$	6.67±0.15 $^{b-g}$	6.83±0.40^{a-f}	6.85^{B}
CONTROL	7.90±0.01^{a}	7.51±0.21^{ab}	7.44±0.17^{ab}	7.00±0.30^{a-e}	6.56±0.52 $^{b-h}$	7.28^{A}
MEAN	6.45^{A}	6.19 ^{AB}	6.15^{AB}	5.91^{BC}	5.73^{C}	

*Means that are followed by different letters are significantly different ($P \leq 0.05$) according to Tukey's test; *Significant at P=0.05*

Table 2b: Effect of pre-storage seed priming and storage duration on the amount of starch (mg/gDW) in low vigour brinjal seeds

Priming treatments	Storage durations					
	0 months	3 months	6 months	9 months	12 months	Mean
1% KNO_3	8.07±0.03^{g-j}	7.90±0.13^{hij}	7.87±0.07^{ij}	7.67±0.19^{j}	7.47±0.37^{j}	6.71^{D}
0.1M KH_2PO_4	10.94±0.34^{cde}	10.65±0.03^{de}	10.10±0.02^{de}	10.18±0.35^{de}	9.74±0.64^{e-h}	8.76^{C}
30%PEG6000	10.90±0.45^{cde}	10.64±0.23^{de}	10.21±0.14^{de}	9.81±0.34^{efg}	9.72±0.23^{e-i}	7.79^{C}
$GA_3$100ppm	8.27±0.14^{f-j}	8.15±0.34^{g-j}	8.07±0.23^{g-j}	7.91±0.20^{hij}	7.76±0.24^{j}	6.14^{D}
H_2O	12.55±0.10^{c}	11.69±0.19^{cd}	10.79±0.44^{cde}	10.58±0.81^{de}	10.36±0.06^{de}	9.02^{B}
CONTROL	17.64±0.06^{a}	17.36±0.10^{a}	16.96±0.23^{a}	15.82±0.46^{ab}	14.68±0.14^{b}	11.89^{A}
MEAN	11.39^{A}	11.06^{AB}	10.67^{BC}	10.32^{CD}	9.96^{D}	

*Means that are followed by different letters are significantly different ($P \leq 0.05$) according to Tukey's test; *Significant at P=0.05*

Overall, the priming treatments resulted in an increase in TSS content and concurrently lower starch content in both high and vigour seeds over their respective control seeds (unprimed) though the extent of increase varied significantly. The maximum amount of TSS was recorded with 1% KNO_3 priming while the lowest starch content was recorded with GA_3 priming. This can be explained in terms of increased activity of α-amylase with priming treatments (Table 3a, 3b) that catalyze the conversion of starch into sugars. Within each storage duration the synergistic effect of seed vigour level and priming treatments on starch content was significant. This may be attributed to enhanced activity of α-amylase due to priming. The activity of α-amylase was lower in low vigour seeds (Table 3b) hence the seeds had higher starch content (Table 2b) and low total soluble sugar content (Table 1b). Priming of low vigour seeds with GA_3lowered the amount of starch content that was statistically at par with the starch content in high vigour seeds stored for zero, three, six, nine, and twelve months respectively (Table 2a, 2b). In low vigour seeds, priming treatments increased the TSS content and α-amylase activity, concomitantly decreasing the starch content compared to the control, indicating that priming treatments significantly improved the quality of low vigour seeds. With increased storage duration, the amount of total soluble sugars and α-amylase activity decreased while starch content increased with the best performance observed with KNO_3 and GA_3, and the improvement gained through these priming treatments was retained for up to 12 months.

Table 3a: Effect of pre-storage seed priming and storage duration on the activity of α-amylase (µg maltose produced/ml/min) in high vigour brinjal seeds

Priming treatments	Storage durations					
	0 months	3 months	6 months	9 months	12 months	Mean
1% KNO_3	21.51±0.15^{a}	20.58±0.25^{ab}	18.57±0.53^{abc}	16.57±0.50 $^{c-f}$	14.74±0.28^{d-h}	18.39^{A}
0.1M KH_2PO_4	16.64±0.25^{c-f}	16.35±0.36^{c-f}	15.61±0.23^{c-g}	13.68±0.49^{f-i}	11.75±1.01^{hij}	14.81^{C}
30%PEG6000	17.87±0.21^{bcd}	14.73±0.11^{d-h}	14.04±0.48^{e-h}	13.33±0.28^{f-j}	12.63±0.12^{g-j}	14.52^{C}
$GA_3$100ppm	20.72±0.13 ^{ab}	19.00±0.19 ^{abc}	16.16±0.87 $^{c-f}$	15.01±0.95^{d-h}	13.87±0.95^{f-i}	16.95^{B}
H_2O	17.27±0.03^{b-e}	16.72±0.46 $^{c-f}$	14.85±0.28^{d-h}	13.72±0.64^{f-i}	10.24±0.72^{ij}	14.56^{C}
CONTROL	15.63±0.21^{c-g}	14.58±0.21^{d-h}	13.86±0.44^{e-h}	11.84±0.27^{hij}	9.82±0.20^{j}	13.15^{D}
MEAN	18.27^{A}	16.99^{B}	15.51^{C}	14.02^{D}	12.17^{E}	

*Means that are followed by different letters are significantly different (P≤0.05) according to Tukey's test; *Significant at P=0.05*

De novo synthesis of α-amylase was reported in prior studies during priming in rice seeds (Lee and Kim 2000). In high vigour seeds, immediately after harvesting the highest activity of α-amylase was recorded in seeds treated with 1% KNO_3 over other treatments, while in low vigour seeds the highest α-amylase activity was recorded in seeds soaked in GA_3 100ppm (18.14 µg maltose produced/ml/min) over other treatments (Table 3a, 3b). A similar trend was observed in the succeeding months of storage. These findings were corroborated by the increased amount of total soluble sugars with increased α-amylase activity after KNO_3 priming reported in tomato (Nawaz *et al.*, 2011) and marigold (Afzal et al., 2009) seeds.

Table 3b: Effect of pre-storage seed priming and storage duration on the activity of α-amylase (µg maltose produced/ml/min) in low vigour brinjal seeds

Priming treatments	Storage durations					
	0 months	3 months	6 months	9 months	12 months	Mean
1% KNO_3	16.08±0.15^{ab}	13.75±1.16^{bcd}	10.16±0.38^{e-i}	9.46±0.71^{f-j}	8.76±1.10^{f-k}	11.64^{B}
0.1M KH_2PO_4	11.42±0.05^{d-g}	11.31±0.13^{d-g}	10.87±0.50^{d-g}	8.72±0.38^{f-k}	6.56±0.34^{jkl}	9.77^{C}
30%PEG6000	11.47±0.10^{def}	10.54±0.28^{e-h}	10.62±0.76^{d-h}	9.55±0.34^{f-j}	8.47±0.10^{f-k}	10.13^{C}
$GA_3$100ppm	18.14±0.28^{a}	16.80±0.76^{ab}	14.98±0.67^{bc}	12.94±0.64^{cde}	10.91±0.16^{d-g}	14.75^{A}
H_2O	9.56±0.43^{f-j}	9.26±0.29^{g-k}	8.33±0.79^{g-k}	7.25±1.38^{i-l}	6.16±0.09^{kl}	8.11^{D}
CONTROL	7.61±0.53^{h-l}	6.18±0.82^{kl}	5.07±0.54^{l}	4.57±0.35^{l}	4.64±0.10^{l}	5.61^{E}
MEAN	12.38^{A}	11.31^{B}	10.00^{C}	8.75^{D}	7.58^{E}	

*Means that are followed by different letters are significantly different (P≤0.05) according to Tukey's test; *Significant at P=0.05*

Within each storage duration, the amount of protein content was higher in high vigour seeds than in low vigour seeds (Table 4a, 4b).

Table 4a: Effect of pre-storage seed priming and storage duration on the amount of protein (mg/gDW) in high vigour brinjal seeds

Priming treatments	Storage durations					
	0 months	3 months	6 months	9 months	12 months	Mean
1% KNO_3	$7.44\pm0.21^{b-e}$	$6.88\pm0.39^{c-f}$	$5.16\pm0.31^{h-k}$	$4.54\pm0.17^{i-m}$	$3.92\pm0.05^{k-n}$	5.59^{C}
0.1M KH_2PO_4	7.96 ± 0.24^{bc}	7.62 ± 0.24^{bcd}	7.17 ± 0.61^{cde}	$5.75\pm0.34^{f-i}$	$4.32\pm0.10^{j-n}$	6.56^{B}
30%PEG6000	8.64 ± 0.17^{ab}	7.86 ± 0.27^{bc}	$6.54\pm0.31^{d-g}$	$5.73\pm0.21^{f-i}$	$4.92\pm0.22^{i-l}$	6.73^{B}
$GA_3$100ppm	9.46 ± 0.17^{a}	8.49 ± 0.11^{ab}	8.15 ± 0.16^{bc}	$7.52\pm0.11^{b-e}$	$6.89\pm0.18^{c-f}$	8.10^{A}
H_2O	$6.25\pm0.23^{e-h}$	$5.81\pm0.29^{f-i}$	$4.56\pm0.25^{i-m}$	$4.10\pm0.17^{k-n}$	3.64 ± 0.17^{mn}	4.87^{D}
CONTROL	$5.41\pm0.11^{g-j}$	$4.18\pm0.19^{j-n}$	3.70 ± 0.11^{lmn}	3.38 ± 0.08^{mn}	3.07 ± 0.06^{n}	3.95^{E}
MEAN	7.53^{A}	6.81^{B}	5.88^{C}	5.17^{D}	4.46^{E}	

*Means that are followed by different letters are significantly different (P≤0.05) according to Tukey's test; *Significant at P=0.05*

Similar results of decreased protein content due to accelerated ageing have been reported in crops e.g., maize (Radha *et al.*, 2014) and radish (Jain *et al.*, 2006) seeds. The protein content decreases during ageing due to damage to protein structure due to high temperature and free radical attack (Zhang *et al.*, 2021). Priming led to an increase of total soluble protein content in both high and low vigour seeds stored for 12 months, the highest amount of protein content was recorded in seeds treated with GA_3 (100ppm) over other priming treatments (Table 4a,4b). Similar results of increased total soluble protein content with KNO_3 priming have been reported in tomato seeds (Pandita *et al.*, 2003), GA_3 priming in chickpea (Arun *et al.*, 2016), and groundnut seeds (Rouhi & Sepehri, 2020).

Table 4b: Effect of pre-storage seed priming and storage duration on the amount of protein (mg/gDW) in low vigour brinjal seeds

Priming treatments	Storage durations					
	0 months	3 months	6 months	9 months	12 months	Mean
1% KNO_3	6.61 ± 0.11^{a}	5.78 ± 0.46^{ab}	$4.87\pm0.66^{b-f}$	$3.93\pm0.34^{e-k}$	$3.00\pm0.04^{h-l}$	4.84^{A}
0.1M KH_2PO_4	$4.16\pm0.24^{b-i}$	$4.02\pm0.22^{d-j}$	$4.02\pm0.22^{d-j}$	$3.52\pm0.05^{f-l}$	$3.01\pm0.13^{g-l}$	3.75^{B}
30%PEG6000	5.70 ± 0.35^{abc}	$4.66\pm0.35^{b-g}$	$4.11\pm0.30^{c-i}$	$3.47\pm0.16^{f-l}$	$2.84\pm0.05^{i-l}$	4.16^{B}
$GA_3$100ppm	$5.61\pm0.08^{a-d}$	$5.33\pm1.05^{a-e}$	$5.01\pm0.33^{a-f}$	$4.79\pm0.26^{b-f}$	$4.57\pm0.27^{b-h}$	5.06^{A}
H_2O	$3.52\pm0.13^{f-l}$	$2.60\pm0.13^{i-l}$	$2.53\pm0.19^{i-l}$	$2.77\pm0.03^{i-l}$	2.43 ± 0.11^{jkl}	2.77^{C}
CONTROL	$2.65\pm0.14^{i-l}$	2.35 ± 0.05^{kl}	2.32 ± 0.06^{kl}	2.21 ± 0.10^{l}	2.10 ± 0.10^{l}	2.32^{C}
MEAN	4.71^{A}	4.12^{B}	3.81^{CB}	3.45^{CD}	2.99^{D}	

*Means that are followed by different letters are significantly different (P≤0.05) according to Tukey's test; *Significant at P=0.05*

Within each storage duration total free amino acid content was lower in high vigour seeds than in low vigour seeds (Table 5a, 5b). In general, with an increase in storage duration the amount of total free aminoacids in both high and low vigour seeds increased. The priming treatments decreased the amount of free amino acids in both high and low vigour seeds. In both high vigour and low vigour seeds prior to storage, priming with GA_3 (100ppm) resulted in least amount of free amino-acids as compared to other treatments. In both GA_3 primed high and low vigour seeds subjected to 12 months of storage, the free amino acid content increased (Table 5a, 5b).

Table 5a: Effect of pre-storage seed priming and storage duration on the total free aminoacids (mg/g DW) in high vigour brinjal seeds

Priming treatments	Storage durations					
	0 months	3 months	6 months	9 months	12 months	Mean
1% KNO_3	$1.74±0.08^{ijk}$	$1.82±0.10^{jk}$	$1.85±0.11^{ijk}$	$2.11±0.13^{h-k}$	$2.40±0.16^{f-j}$	1.98^{D}
0.1M KH_2PO_4	$2.42±0.09^{f-j}$	$2.52±0.10^{f-i}$	$2.71±0.13^{e-h}$	$3.10±0.16^{def}$	$3.49±0.19^{cd}$	2.85^{C}
30%PEG6000	$2.38±0.14^{f-j}$	$3.07±0.12^{def}$	$3.35±0.07^{cde}$	$3.71±0.06^{cd}$	$4.06±0.06^{bc}$	3.31^{B}
$GA_3$100ppm	$1.59±0.12^{k}$	$1.72±0.14^{jk}$	$2.05±0.17^{h-k}$	$2.31±0.01^{g-k}$	$2.56±0.06^{f-i}$	2.04^{D}
H_2O	$2.03±0.06^{h-k}$	$2.18±0.03^{h-k}$	$2.40±0.08^{f-j}$	$3.01±0.15^{d-g}$	$3.61±0.12^{cd}$	2.64^{C}
CONTROL	$3.11±0.11^{def}$	$3.54±0.33^{cd}$	$4.06±0.27^{bc}$	$4.64±0.23^{ab}$	$5.22±0.14^{a}$	4.11^{A}
MEAN	2.23^{E}	2.46^{D}	2.73^{C}	3.14^{B}	3.56^{A}	

*Means that are followed by different letters are significantly different (P≤0.05) according to Tukey's test; *Significant at P=0.05*

Seed priming resulted in a decrease in free amino acid content and a simultaneous increase in protein content indicating that seed priming improved the protein synthesis machinery at the sub-cellular basis resulting in the synthesis of more proteins using free amino acids. Thus, seed priming decreased the hydrolysis of proteins resulting in decreased free amino acid content in seeds. With all the priming treatments, protein content increased while free amino acid content decreased as compared to untreated seeds. Thus, improving the seed quality with increase in the storage duration, protein content decreased while free amino acid content increased with best performance observed with KNO_3 and GA_3, and seed quality was retained for up to 12 months with these treatments. Similar results of decreased free amino acid content with seed priming have been reported in sorghum (Zhang *et al.*, 2015) seeds.

Table 5b: Effect of pre-storage seed priming and storage duration on the total free aminoacids (mg/g DW) in low vigour brinjal seeds

Priming treatments	Storage durations					
	0 months	3 months	6 months	9 months	12 months	Mean
1% KNO_3	$3.01±0.14^{l-p}$	$3.43±0.21^{i-o}$	$3.99±0.24^{f-k}$	$4.53±0.25^{c-g}$	$5.07±0.29^{a-d}$	4.00^{C}
0.1M KH_2PO_4	$2.81±0.29^{nop}$	$3.03±0.11^{l-p}$	$3.69±0.06^{g-m}$	$3.92±0.12^{f-k}$	$4.14±0.08^{e-j}$	3.52^{D}
30%PEG6000	$3.32±0.08^{j-o}$	$3.57±0.13^{h-n}$	$4.08±0.10^{e-k}$	$4.38±0.24^{d-h}$	$4.68±0.23^{b-f}$	4.01^{C}
$GA_3$100ppm	$2.55±0.24^{op}$	$3.02±0.19^{l-p}$	$2.37±0.06^{p}$	$2.81±0.07^{m-p}$	$3.25±0.09^{k-p}$	2.80^{E}
H_2O	$3.74±0.14^{g-l}$	$4.21±0.01^{d-i}$	$4.41±0.22^{d-h}$	$4.73±0.17^{b-f}$	$5.04±0.15^{a-d}$	4.42^{B}
CONTROL	$4.74±0.21^{a-f}$	$4.90±0.18^{a-e}$	$5.38±0.13^{abc}$	$5.50±0.09^{ab}$	$5.62±0.11^{a}$	5.23^{A}
MEAN	3.36^{E}	3.69^{D}	3.99^{C}	4.31^{B}	4.63^{A}	

*Means that are followed by different letters are significantly different (P≤0.05) according to Tukey's test; *Significant at P=0.05*

CONCLUSION

Seed priming on brinjal seeds improved seed quality in both the high and low-vigour seeds subjected to storage for up to 12 months. The extent of improvement in terms of percentage increase in quality parameters i.e total soluble sugars, proteins, and activity of α-amylase, and percent decrease in total free amino acids and starch content was higher in low-vigour seeds with the priming treatments. Further, among the priming treatments, in GA_3 and KNO_3 primed seeds the quality in terms of biochemical parameters was retained even after 12 months of storage and it was equivalent to or higher than the seeds that were not subjected to any storage, i.e., these priming treatments were able to minimize deteriorative changes in the seeds during storage. Thus, based on the results of our present study, it is recommended that better improvement in seed quality with good storability can be obtained in brinjal seeds by subjecting the seeds to GA_3 and KNO_3 priming for 12 h, and these seeds can be safely dried back to original moisture content and stored to harness maximum efficacy of priming treatments and better storability.In conclusion, the research findings demonstrate that seed priming, particularly with GA_3 and KNO_3 treatments, significantly enhances the quality of brinjal seeds, particularly those with low vigour, even after storage for up to 12 months. These priming treatments not only improve biochemical parameters but also effectively minimize deteriorative changes during storage. Therefore, it is recommended to utilize GA_3 and KNO_3 priming for 12 hours to enhance seed quality and ensure better storability in brinjal seeds.

Author's contribution: JP conducted the experiments, collected and analyzed the data, and drafted the manuscript. SB did conceptualization, supervision, data curation, original draft writing, review, and editing. TS did data curation, editing, review, and original draft writing. RS conceived and designed the research. All authors equally contributed and reviewed the final draft of the manuscript.

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