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Full Length Research Article

Biochemical Seed Quality as Affected by Different Storage Temperatures and Containers in Seed of Pigeonpea [*Cajanus cajan* (L.) Millsp.]

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ABSTRACT

Pigeonpea [*Cajanus cajan* (L.) Millsp.] is a rich source of nutrients and contains carbohydrates, proteins and lipids. Temperature and moisture content of seed during storage are most significant factors affecting the quality of seed. Present research was formulated to store two different seed lots (fresh and revalidated) using four different packaging containers (cloth bag, cloth bag containing drying beads, desiccator and desiccator containing drying beads) under two different temperature regimes i.e., ambient (~27.3°C) and cold room temperature (~15.5°C) to observe changes in seed quality. The effect was registered bimonthly in terms of total soluble sugars (TSS), total soluble proteins (TSP) and electrical conductance (EC). Fresh seed lot showed maximum TSS (46.77 mg/g), TSP (218.83 mg/g) and EC (0.63 µmhos cm^{-1} $seed^{-1}$) at moisture content ~8.42% when stored in desiccator containing drying beads under cold room conditions (~15.5°C temperature and ~55.5% relative humidity) after 12 months of storage. In revalidated seed lot, up to 10 months it showed TSS (43.44 mg/g), TSP (204.65 mg/g) and EC (0.72 µmhos cm^{-1} $seed^{-1}$) when stored in desiccator containing drying beads stored under cold room conditions while under ambient conditions (~27.3°C temperature and ~32.67% relative humidity), cloth bag showed maximum inactivation of TSS, more denaturation of proteins and high solute leakage.

***Key words*:** Fresh seed lot, Revalidated seed lot, Packaging containers, *Cajanus cajan* (L.), Storage temperature, Total soluble proteins, Total soluble sugars

Seed deterioration is the alterations occurring with time that increase the seeds exposure to external challenges and decreases the ability of the seed to survive. Loss of seed viability is related with various metabolic and biochemical alterations that result in loss of reduction in the energy metabolism, protein synthesis and degradation of DNA [1]. The biochemical processes occurring in seed during storage are directly influenced by moisture content, temperature and association of mycoflora [2]. Biochemical changes associated with seed deterioration are the increase in leakage of biomolecules [3], decrease in total soluble sugars and protein content [4] whereas amino acids and protease activity increases with seed deterioration. Value of electrical conductivity in storage environment grow linearly over time for packaging that allow exchange with environment that shows increase in breakdown and loss of integrity of cell membrane along storage time. However, availability of an adequate supply of high-quality seed is essential for a successful seed production program and the maintenance of a viable and productive agriculture. So, seed storability is important for maintaining seed quality from harvest till next sowing. Keeping in view an experiment was conducted during November 2015 to January 2017 at Seed Technology Section, Punjab Agricultural University Ludhiana to evaluate biochemical seed quality as affected by different storage temperatures and containers in fresh and revalidated seed lot of pigeonpea [*Cajanus cajan* (L.) Millsp.].

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MATERIALS AND METHODS

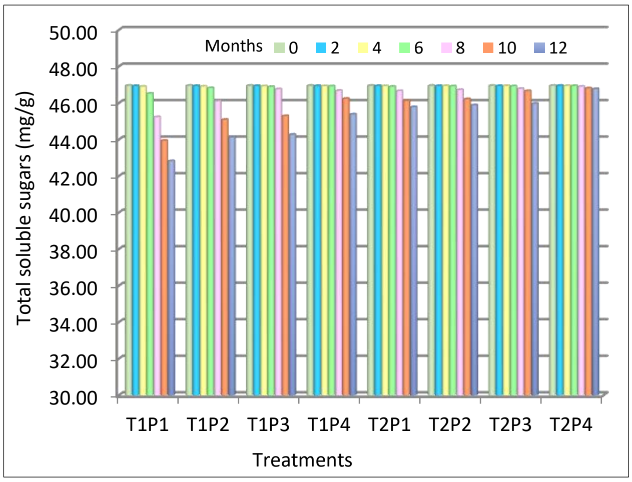
Fresh and revalidated seeds of pigeonpea [*Cajanus cajan* (L.) Millsp.] cv. PAU 881 were procured from Pulses Section, Department of Plant Breeding and Genetics and used for conducting different seed quality tests that included biochemical parameters such as total protein content, total soluble sugars and electrical conductance. It was conducted in

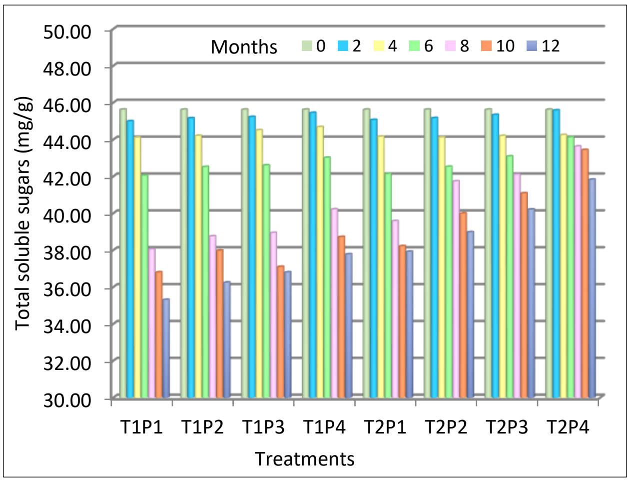
Seed Technology Section of Punjab Agricultural University, Ludhiana. Both fresh and revalidated seeds were packed and stored under different temperature regimes. Different packing materials were cloth bag, cloth bag containing drying beads, desiccator and desiccator containing drying beads. The different temperatures were ambient temperature (~27.3°C) and cold room temperature (~15.5°C). Total soluble sugars content was estimated using method of Dubois *et al.* [5]. The total soluble proteins was estimated using the method of Lowry *et al.* [6] while for measuring the electrical conductivity, three replicates of 50 seeds each were soaked in 50 ml of deionized water at 25°C for 17h. The seed leachate was collected and the conductivity was measured with the help of digital conductivity meter along with deionized water as control [7].

RESULTS AND DISCUSSION

The initial data of fresh (Table 1) and revalidated seed lot (Table 2) of total soluble sugars was noted before the start of storage period. In fresh seed lot control was 46.95 mg/g while for revalidated seed lot it was 45.63 mg/g [8] reported 31.00 mg/g total soluble sugars in mature seeds of pigeon pea. During storage, chemical and metabolic modifications arise which includes inactivation of total soluble sugars. Soluble carbohydrate content decreases with storage of seed that limits availability of respiratory substrates for germination [9-10]. In the present studies after 6 months, in fresh seed total soluble sugars was maintained (46.94 mg/g) when stored in desiccator containing drying beads under cold room temperature conditions (~15.5°C) while under similar conditions in revalidated seed lot it showed 44.12 mg/g (Fig 1-2). Whereas it was declined (42.05 mg/g) in revalidated seed lot when seeds were stored in cloth bag under ambient temperature conditions (~27.3°C). After 8 months of storage, in revalidated seed lot total soluble sugars was found more when seeds were packed in desiccator containing drying beads (43.63 mg/g) while under similar conditions in fresh seed lot it showed maximum TSS (46.90 mg/g) under cold room conditions. Fresh seeds under ambient conditions showed decline in TSS (45.23 mg/g) when packed in cloth bag while under similar conditions in revalidated seed lot it was 38.05 mg/g. After 10 months of storage, maximum TSS i.e., 46.81 mg/g was observed in fresh seed lot when packed in desiccator containing drying beads under cold room temperature conditions while under similar temperature conditions, it also showed 46.66 mg/g when packed in desiccator.

Under cold room temperature conditions, in revalidated seed lot when seeds were packed in desiccator containing drying beads, it showed 43.44 mg/g of total soluble sugars. While maximum inactivation of total soluble sugars was found in revalidated seed lot when seeds were stored in cloth bag (36.80 mg/g) which was followed by seeds packed in cloth bag containing drying beads (37.28 mg/g) under similar temperature conditions. In fresh seed lot maximum inactivation of TSS was found when seeds were packed in cloth bag under ambient temperature conditions which was followed by cloth bag containing drying beads, desiccator i.e., 43.94 mg/g, 45.09 mg/g, 45.29 mg/g. After 12 months, fresh seeds when packed in desiccator containing drying beads indicated maximum total soluble sugars i.e., 46.77 mg/g, than all other packings as in desiccator (45.97 mg/g), cloth bag containing drying beads (45.88 mg/g) and cloth bag (45.78 mg/g) respectively, when kept under cold room conditions while under similar temperature conditions in revalidated seed lot when seeds were packed in desiccator containing drying beads showed more content of total soluble sugars i.e., 41.83 mg/g. But revalidated seeds when packed in cloth bag showed maximum inactivation of TSS (35.61 mg/g) when stored under ambient temperature as compared to seeds stored in same packing under cold room temperature conditions (37.92 mg/g). So, it was observed that maximum inactivation of total soluble sugars was observed when seeds were kept in cloth bag. It was recognized that increase in moisture content and storage temperature lead to biochemical changes. In black gram, reported that there was decrease in content of total soluble sugars with increase in storage period [11].





T_1: Ambient temperature, T_2: Cold room temperature, P_1: Cloth bag, P_2: Cloth bag containing drying beads, P_3: Desiccator, P_4: Desiccator containing drying beads

Fig 1 Effect of different temperatures and packaging on total soluble sugars (mg/g) in fresh seeds of pigeonpea during storage

Fig 2 Effect of different temperatures and packaging on total soluble sugars (mg/g) in revalidated seeds of pigeonpea during storage

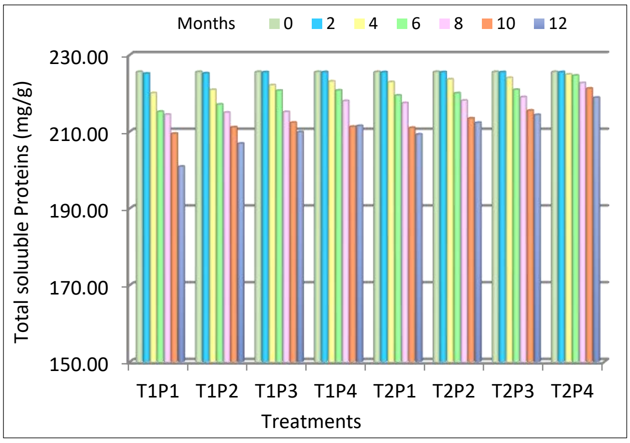
Table 1 Initial biochemical parameters of seed quality in fresh seeds of pigeonpea before storage

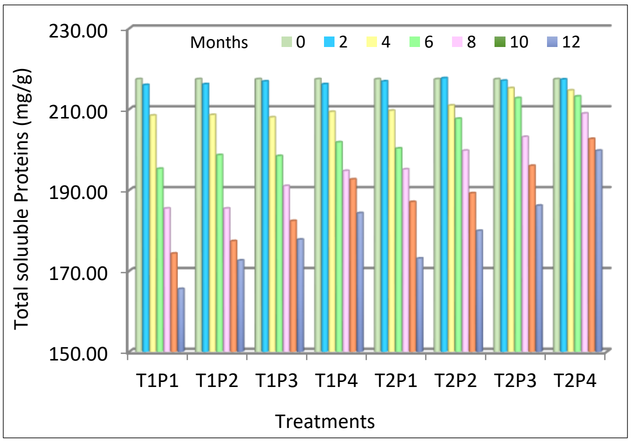
Total soluble sugars (mg/g)	Protein content (mg/g)	Electrical conductivity (μmhos cm^{-1} $seed^{-1}$)
46.95	225.54	0.55

Table 2 Initial biochemical parameters of seed quality in revalidated seeds of pigeonpea before storage

Total soluble sugars (mg/g)	Protein content (mg/g)	Electrical conductivity (μmhos cm^{-1} $seed^{-1}$)
45.63	217.44	0.58

The initial data of fresh (Table 1) and revalidated seed lot (Table 2) of total soluble proteins was taken before the start of storage period in which fresh seed lot showed 225.54 mg/g while revalidated seed lot showed 217.44 mg/g that showed quality of fresh seed lot was better than revalidated seed lot. Chemical and metabolic changes occur during storage period which includes denaturation of total soluble proteins. Till initial 6 months, in fresh seed lot total soluble proteins was maintained in all packings under both temperature regimes while in revalidated seed lot, it started decreases whereas maximum decrease was found when seeds were packed in cloth bag under ambient temperature conditions (195.26 mg/g) (Fig 3-4).



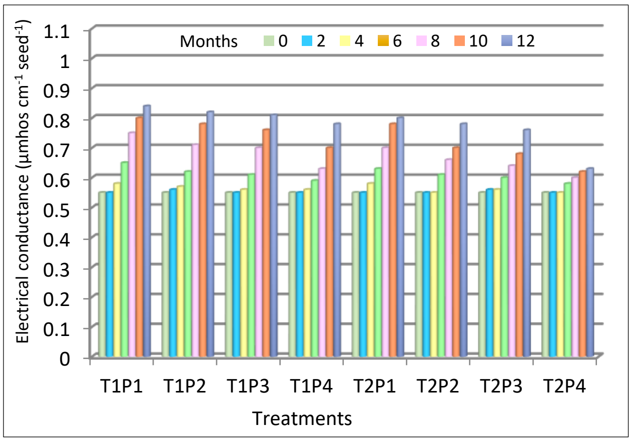


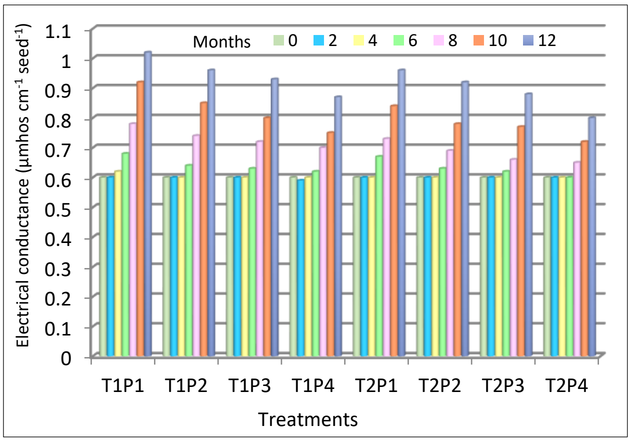
T_1: Ambient temperature, T_2: Cold room temperature, P_1: Cloth bag, P_2: Cloth bag containing drying beads, P_3: Desiccator, P_4: Desiccator containing drying beads

Fig 3 Effect of different temperatures and packaging on total soluble proteins (mg/g) in fresh seeds of pigeonpea during storage

Fig 4 Effect of different temperatures and packaging on total soluble proteins (mg/g) in revalidated seeds of pigeonpea during storage

After 8 months of storage, in revalidated seed lot maximum quantity of total soluble proteins (TSP) was found when seeds were packed in desiccator containing drying beads i.e., 208.95 mg/g while under similar conditions in fresh seed lot it showed 224.60 mg/g. Fresh seed were stored under ambient temperature conditions showed decline in total soluble proteins (214.42 mg/g) when packed in cloth bag while under similar conditions in revalidated seed lot it was 185.45 mg/g. After 10 months of storage, in fresh seed lot maximum total soluble proteins (TSP) was 221.22 mg/g under cold room temperature conditions in desiccator containing drying beads which was followed by desiccator 215.46 mg/g. In revalidated seed lot under cold room temperature conditions, when seeds were packed in desiccator containing drying beads 204.65 mg/g of total soluble proteins. While maximum denaturation of total soluble proteins was found in revalidated seed lot when seeds were stored in cloth bag i.e., 174.30 mg/g which was followed by seeds packed in cloth bag containing drying beads (177.38 mg/g) under similar temperature conditions. In fresh seeds, maximum decline was found under ambient temperature conditions when seeds were packed in cloth bag i.e., 209.45 mg/g. In fresh seed lot, after 12 months of storage, total soluble proteins was observed to maximum i.e., 219.83 mg/g when placed in desiccator containing drying beads under cold room condition while minimum i.e., 165.54 mg/g was found in revalidated seed lot when seeds were packed under ambient temperature conditions in cloth bag. Certain changes in physiological and biochemical characters of pigeonpea seed included deterioration of both germinability and seed viability. Syed *et al.* [12] reported that during storage conditions, due to pathogens deterioration takes place which results in decrease in total soluble proteins in pigeonpea, green gram, chickpea seeds. After storage of 9 months nearly 50% reduction in protein content was reported in pigeonpea seeds.





T_1: Ambient temperature, T_2: Cold room temperature, P_1: Cloth bag, P_2: Cloth bag containing drying beads, P_3: Desiccator, P_4: Desiccator containing drying beads

Fig 5 Effect of different temperatures and packagings on electrical conductivity (µmhos cm^{-1} seed^{-1}) in revalidated seeds of pigeonpea during storage

Fig 6 Effect of different temperatures and packagings on electrical conductivity (µmhos cm^{-1} seed^{-1}) in revalidated seeds of pigeonpea during storage

Electrical conductivity measures the quantity of electrolytes liberated during cell membrane reform during the seed imbibition process. The results of the electrical conductivity test can be affected by different temperatures and with different containers. Low vigour seeds have been shown to possess decrease membrane integrity as a result of storage deterioration and mechanical injury. The solutes leached into solution are mainly carbohydrates, fatty acids, amino acids, organic acids, proteins, phenolic compounds and ions [13]. Initially fresh seed lot showed less electrical conductance i.e., 0.55 µmhos cm^{-1} seed^{-1} than revalidated seed lot i.e., 0.58 µmhos cm^{-1} seed^{-1} which showed fresh seed lot more vigorous than revalidated seed lot. During initial months of storage, all the treatments vary non-significantly. After 6 months of storage both fresh and revalidated seed showed maintained vigour under both temperature regimes in all packing materials (Fig 5-6).

After 8 months of storage, maximum leakage of solutes was showed when revalidated seeds were packed in cloth bag under ambient conditions i.e., 0.78 µmhos cm^{-1} seed^{-1} lot which while in fresh seed under similar conditions maximum leakage was 0.75 µmhos cm^{-1} seed^{-1}. In fresh seed lot, minimum leakage showed when seeds were packed in desiccator containing drying beads i.e., 0.60 µmhos cm^{-1} seed^{-1} under cold room conditions whereas in revalidated seed lot minimum leakage was showed when seeds were packed in desiccator containing drying beads i.e., 0.65 µmhos cm^{-1} seed^{-1} under similar conditions. After 10 months of storage, maximum leakage of solutes was found when revalidated seeds packed under ambient conditions in cloth bag (0.92 µmhos cm^{-1} seed^{-1}) and was followed by cloth bag containing drying beads (0.85 µmhos cm^{-1} seed^{-1}). Membrane integrity was maintained in fresh seed lot under cold room temperature conditions when seeds were packed in desiccator containing drying beads (0.62 µmhos cm^{-1} seed^{-1}) which was followed by desiccator (0.68 µmhos cm^{-1} seed^{-1}). In revalidated seed lot, cell membrane integrity was maintained till 10 months i.e., 0.72 µmhos cm^{-1} seed^{-1} when seeds were stored under cold room temperature conditions in desiccator containing drying beads (0.72 µmhos cm^{-1} seed^{-1}). After 12 month of storage, maximum membrane integrity was found in fresh seed lot under cold room conditions when packed in desiccator containing drying beads (0.63 µmhos cm^{-1} seed^{-1}) while under similar conditions in revalidated seed lot minimum leakage was (0.80 µmhos cm^{-1} seed^{-1}). Maximum leakage (1.02 µmhos cm^{-1} seed^{-1}) was found when revalidated seeds were packed in cloth bag under ambient conditions while under similar conditions in fresh seed lot it was 0.84 µmhos cm^{-1} seed^{-1}. As reported, the electrical conductivity of onion seed is related to its membrane disintegration and finally loss of viability. Cucumber seeds with 12% initial moisture content were stored at 20°C up to 12 months and germination percentage remained high throughout the storage period, while vigour decreased and of electrolyte leakage increased from 6 months [14].

CONCLUSION

Based upon the observations it was concluded that during the storage period, metabolism of seed was greatly affected. If the fresh seeds of pigeonpea were stored at moisture content ~8.42%, when stored in a desiccator containing drying beads under cold room conditions (~15.5°C temperature and ~55.5% relative humidity), can maintain seed storability for more than 12 months in terms total soluble sugars, total soluble proteins and membrane integrity. Further it was also concluded that when revalidated seeds were packed in cloth bag under ambient conditions (~27.3°C temperature and ~32.67% relative humidity) lead to maximum deterioration in seed quality. The present results thus emphasis on the importance of seed moisture content, storage temperature, relative humidity, packaging material and storage period on the quality of pigeonpea seeds.

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