Influence of Different Maturity Stages on Tomato (*Solanum lycopersicum* L.) Seed Quality during Storage at -20°C

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**Abstract**

**Background:** Tomato is one of the most important vegetable crops in the world due to its dietary value. Harvesting and storing of physiologically matured tomato seeds is very important to secure good quality seeds for future use in a changing climate. The objective of the study was to assess the effect of maturity stages on seed quality of two tomato accessions during cold storage.

**Methods:** Seeds of two tomato accessions extracted at four maturity stages (*i.e.* initially ripe, half ripe, fully ripe and rotten) were stored at -20°C in a deep freezer for 12 months. The quality of stored seeds was measured by seed vigour and germination percentage at 3, 6, 9 and 12 months after storage.

**Conclusion:** The study showed significant differences in seed vigour and germination percentage at 3, 6, 9 and 12 months after storage (MAS) among the maturity stages. Seeds extracted at the initially ripe stage in GH 9305 had the least vigour at 3, 6, 9 and 12 MAS. The highest reduction in seed quality during the storage period was observed in seeds extracted at the rotten stage in both accessions. Seed vigour and germination were higher in GH 9207 than GH 9305. Storage of physiologically matured tomato seeds at freezing temperature is necessary to reduce seed ageing and to maintain high seed quality.

**Key words:** Maturity stage, Seed quality, Seed storage, Tomato.

**Introduction**

Tomato (*Solanum lycopersicum* L.) belongs to the solanaceae family. It is the second most important fruit or vegetable crop next to potato (*Solanum tuberosum* L.). Globally, approximately 182.3 million tons of tomato fruits are produced on 4.85 million ha each year (FAOSTAT, 2019). Out of the total tomato production, Asia accounts for 61.1% while Europe, America and Africa produces 13.5%, 13.4% and 11.8% respectively. Tomatoes are rich sources of vitamins and pro-vitamins (vitamin C, pro-vitamin A, β-carotene, folate), minerals such as potassium and secondary metabolites such as lycopene, flavonoids, phytoestrogens and polyphenols (Luthria et al., 2006). However, tomato yields are highly variable, thus the need to produce and conserve good quality seeds for optimum yields and nutritional security.

Seed quality comprises of several important seed attributes such as the genetic and chemical composition, physical condition, physiological germination and vigour, size, appearance and presence of seed borne pathogens, crop and varietal purity, weed and crop contaminants and moisture content. Maturity stage is one of the most important factors that influence the quality of seeds (Demir et al., 2008). Wang et al. (2008), Elias and Copeland (2001) indicated that harvesting too early may result in low yield and seed quality, because of the partial development of essential structures of seeds. Whereas, harvesting too late may increase the risk of shattering and decrease the quality of seeds due to ageing.

Seed storage is an essential step for the long term conservation of plant genetic resources. Pradhan and Badola (2012) indicated that maintaining seed viability for longer period is very essential to preserve the genetic integrity in stored samples. Seeds that have high initial viability withstand unfavourable storage conditions better than similar seeds of low initial viability. Tang et al. (2000) reported that the predictive ability of any seed quality test of seed deterioration in storage is based on the relationship that exists between the initial seed quality, seed longevity, seed moisture content and storage conditions of temperature, relative humidity and oxygen concentration. According to Copeland and McDonald (2001), several genetic factors of the seed such as hybrid vigour, susceptibility to seed damage and chemical composition can influence the seed vigour and ultimately, viability. Besides, inappropriate storage medium such as room temperature storage often results in low seed germination, seed deterioration and loss of viability, which are natural phenomenon during storage (Schmidt, 2002; Nasreen et al., 2000). Furthermore, if the seeds are not well dried to the appropriate moisture content before storage, the high moisture content may reduce the seed viability by promoting
fungal growth which could further result in decline of seed germination capacity (Romanas, 1991). However, not much information exists on the effect cold storage on tomato seed quality. The present study was undertaken to assess the effect of maturity stages on seed quality of two tomato accessions during cold storage.

**MATERIALS AND METHODS**

The study was carried out at the experimental site of CSIR-Plant Genetic Resources Research Institute, Bunso (N 06° 17.839, W 000° 27.595, Alt 198.3 m above sea level), Eastern region, Ghana from April, 2017 to December, 2018. Seeds of tomato (GH 9207 and GH 9305) obtained from the same institute were transplanted at 23 days after sowing. Tomato seedlings were spaced at 60cm x 60cm. The experiment was arranged in a randomized complete block design (RCBD) with three replications.

Agronomic practices undertaken during the experimental period include fertilizer application (NPK 15-15-15) at a rate of 400kg per hectare. Watering and weeding were carried out as and when required. Insect pests were controlled using K-optimal insecticide (Landa-cyhalothrin 15g/l + Acetamiprid 20g/l: EC) at a recommended rate of 2.7 ml/litre of water at two weeks interval.

Harvesting of fruits was done at four physiological maturity stages (Tetteh *et al.*, 2018: Initially ripe = green with light red spot, Half ripe = light red/orange, Fully ripe = red, Rotten = red with decomposing texture) for seed extraction at 90 days after sowing. Before seed extraction, fruits were rinsed with water to remove unwanted materials. Seeds were extracted manually and air dried to attain a lower moisture content of 7%. After drying, manual cleaning of seeds was done to remove inert materials and infested seeds. Seeds were placed on silica gel to attain constant moisture content before storage.

Hundred seeds of tomato from each treatment were counted into plastic bags of dimension 8cm x 12cm and sealed with cellotape to make it air-tight. This was done to avoid contact with the seeds during sampling for seed quality test. Bagged seeds for all the treatments were put into aluminum foil pouches and sealed for storage at -20°C in a deep freezer.

Seed vigour and germination test were carried out before storage and at 3 months interval up to a period of 12 months under field conditions using seed boxes filled with sterilized topsoil. For each treatment, 50 seeds were used and the experiment was replicated four times. The randomized complete block design (RCBD) was used. The first count (seed vigour) and final count (germination percentage) were established on the 5th and 14th day respectively. The following formula was used to calculate seed germination (ISTA, 1999):

\[ \text{Seed germination} (\%) = \frac{\text{Number of germinated seeds}}{\text{Total seeds sown}} \times 100 \]

Statistical analyses was conducted using the SPSS Statistics 21 (IBM, Chicago, IL, USA). Data was subjected to two-way ANOVA and when the treatment means were significant, Tukey’s HSD test was conducted to identify differences among treatments.

**RESULTS AND DISCUSSION**

The effect of maturity stage on seed vigour of two tomato accessions (GH 9207 and GH 9305) before storage and at 3, 6, 9 and 12 months after storage were studied in the experiment (Table 1). Maturity stage had significant (p<0.001) effect on seed vigour before storage and at 3, 6, 9 and 12 MAS. Both accessions differed significantly at 3, 6, 9 and 12 MAS (months after storage). Seeds extracted at the initially ripe stage in GH 9305 had the least vigour during storage. Seed vigour of the two tomato accessions extracted at the initial ripe stage decreased with the increase in duration of storage. The highest reduction was observed at 9 and 12 months after storage in GH 9305. Rajjou and Debeaunon (2008) indicated that when seeds deteriorate during storage, they lose vigour, become more sensitive to stress during germination and ultimately become unable to germinate. However, seed aging during storage is an inevitable phenomenon, but the degree and speed of decline in seed quality depend strongly, beside storage conditions, on plant species stored and initial seed quality (Sudhakaran, 2020; Balešević-Tubić *et al.*, 2005; Elias and Copeland, 1994) as well as on seed genetic traits (Malenčić *et al.*, 2003). Milošević *et al.* (1996) reported that seed longevity is genetically determined and that significant differences exist among cultivars of the same crop in their ability of quality maintenance during storage. The observed reduction in seed vigour of the initial ripe seeds at storage could be attributed to inadequate food reserves in both accessions. This could have a direct effect on the performance of seeds planted to regenerate a crop (TeKrony and Egli, 1991). Additionally, Caddick (2007) reported that seeds with low vigour will show stunted growth and abnormalities in the developing shoot and root system and subsequently affect crop establishment. Studies on other crop species found a positive correlation between seed vigour and yield including lettuce (Smith *et al.*, 1973), cauliflower (Finch-Savage and McKee, 1990), peas and tomato (Basra, 1995). Thus, seeds extracted at the initial ripe stage and stored for longer periods of time may affect the crop growth and yield.

Maturity stage had significant (p<0.001) effect on germination percentage before storage and at 3, 6, 9 and 12 MAS (Table 2). Seeds extracted at the fully ripe stage, half ripe and rotten stages in both accessions were not significantly different from each other. Seeds extracted at the initially ripe stage in GH 9207 had a higher germination percentage than GH 9305. Both accessions differed significantly with GH 9207 recording the highest germination percentage BS and at 3, 6, 9 and 12 MAS. Seeds extracted at the rotten stage showed the highest reduction in germination during the storage period. Perhaps, the
A reduction in germination at the rotten stage during the storage period could be attributed to microbe infestation before storage. The process of deterioration involves several physiological and structural changes within the seed. Structural changes involve membrane permeability, proteins, sugars, nucleic acids, fatty acids and volatile substances, while physiological processes involve enzyme activity, respiratory competence, lipid peroxidation and physiological repair mechanisms (Chhabra and Singh, 2019; Qun et al., 2007; Walters, 1998). According to Vertucci and Roos (1990), optimum protocols for seed storage must take into account the chemical composition of the seed, the physiological status of the seed and the physical status of water within the seed. In the present study, the slight decrease in germination percentage in the half ripe and fully ripe could be attributed to the attainment of maximum dry mass at maturity before storage. Baruah et al. (1996) and Doijoe (1988) observed high germination from seeds extracted from red ripe tomato fruits which agrees with our findings. Similarly, Belleti and Quagliotti (1991) indicated that, the highest percentage of seed germination can be obtained from full (red or yellow) coloured fruit rather than from green fruits. Thus, the quality of seeds during the storage period is strongly influenced by the quality of the initial seed (before storage), seed moisture content, temperature and humidity.

**CONCLUSION**

The present study showed significant differences in seed vigour and germination percentage at 3, 6, 9 and 12 months after storage among the maturity stages. Seeds extracted at the initially ripe stage in GH 9305 had the least vigour at

| Accession | Maturity stage | Seed vigour | BS | 3 MAS | 6 MAS | 9 MAS | 12 MAS |
|-----------|----------------|-------------|----|-------|-------|-------|--------|
| GH 9207   | Initially ripe | 33.5 (4.0) c| 36.25 (0.96) d| 34.5 (3.9) b| 32.5 (4.4) c| 32.0 (4.3) c|
|           | Half ripe      | 46.8 (1.5) a| 46.00 (1.41) ab| 42.8 (1.5) a| 43.0 (0.8) ab| 42.5 (0.6) ab|
|           | Fully ripe     | 47.0 (2.2) a| 47.00 (0.82) a| 46.8 (1.3) a| 47.0 (1.4) a| 46.8 (1.3) a|
|           | Rotten         | 48.0 (0.8) a| 46.75 (0.96) a| 45.8 (1.7) a| 45.3 (2.4) ab| 44.8 (1.7) ab|
| GH 9305   | Initially ripe | 17.3 (2.9) d| 20.25 (1.50) e| 19.5 (1.3) c| 16.0 (3.4) d| 16.3 (2.6) d|
|           | Half ripe      | 39.5 (4.9) bc| 41.50 (2.89) c| 42.8 (1.7) a| 40.8 (1.7) b| 41.5 (1.3) b|
|           | Fully ripe     | 44.0 (3.6) ab| 45.75 (0.96) ab| 45.8 (1.5) a| 45.3 (1.9) ab| 45.8 (1.7) ab|
|           | Rotten         | 42.0 (2.7) ab| 43.00 (1.41) bc| 44.8 (1.7) a| 41.5 (1.3) ab| 42.0 (1.6) ab|

**ANOVA**

Maturity stage (MS) *** *** *** ***
Accession (A) *** *** *** ***
MS x A * *** *** *** ***

Each value is the mean of four replicates and the standard deviation is shown in parentheses. Two-way ANOVA: *p<0.05, **p<0.01, ***p<0.001. When significant interaction between maturity stage (MS) and Accession (A) was detected, Tukey’s HSD test was performed to identify significant differences among the 4 treatments. Values with different letters are significantly different at p<0.05.

Table 2: Effect of maturity stage on germination percentage of two tomato accessions (GH 9207 and GH 9305) before storage (BS) and at months after storage (MAS).

| Accession | Maturity stage | Germination (%) | BS | 3 MAS | 6 MAS | 9 MAS | 12 MAS |
|-----------|----------------|-----------------|----|-------|-------|-------|--------|
| GH 9207   | Initially ripe | 76.50 (5.74) b | 78.50 (2.52) b| 74.50 (3.40) c| 75.50 (5.26) b| 69.50 (1.90) b|
|           | Half ripe      | 98.50 (1.91) a | 96.00 (1.63) a| 95.00 (1.15) ab| 91.50 (4.12) a| 95.50 (1.90) a|
|           | Fully ripe     | 96.00 (2.82) a | 98.00 (1.63) a| 97.50 (1.00) a| 96.50 (3.42) a| 95.50 (1.90) a|
|           | Rotten         | 97.00 (2.00) a | 97.50 (1.91) a| 97.00 (1.15) a| 95.00 (3.46) a| 94.50 (1.90) a|
| GH 9305   | Initially ripe | 45.00 (5.29) c | 57.00 (1.15) c| 50.50 (4.12) d| 52.00 (7.30) c| 48.00 (4.32) c|
|           | Half ripe      | 94.50 (2.52) a | 93.00 (3.46) a| 91.00 (1.15) b| 90.50 (1.00) a| 91.50 (1.91) a|
|           | Fully ripe     | 98.50 (1.91) a | 96.50 (2.52) a| 97.50 (3.00) a| 97.00 (3.46) a| 94.00 (2.83) a|
|           | Rotten         | 95.50 (1.91) a | 94.00 (4.32) a| 94.00 (2.82) ab| 90.00 (1.63) a| 92.50 (1.00) a|

**ANOVA**

Maturity stage (MS) *** *** *** ***
Accession (A) *** *** *** ***
MS x A *** *** *** ***

Each value is the mean of four replicates and the standard deviation is shown in parentheses. Two-way ANOVA: **p<0.01, *** p<0.001. When significant interaction between maturity stage (MS) and Accession (A) was detected, Tukey’s HSD test was performed to identify significant differences among the 4 treatments. Values with different letters are significantly different at p<0.05.
The highest reduction in seed quality during the storage period was observed in seeds extracted at the rotten stage in both accessions. Seed vigour and germination were higher in GH 9207 than GH 9305. For long-term storage of tomato seeds at low temperature, it is necessary to extract seeds which are physiologically matured (half ripe and fully ripe) to reduce seed ageing and maintain high seed quality.

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