Asian Research Journal of Agriculture

8(2): 1-7, 2018; Article no.ARJA.39149
ISSN: 2456-561X

Comparative Analysis of Germination Behavior of Three Species of *Solanum* at Different Storage Periods and Conditions

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Authors' contributions

This work was carried out in collaboration between all authors. Author Olosunde Adam designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors AS, AO and OM managed the literature searches. Author Oluwadare Ayooluwa managed the analyses of the study. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/ARJA/2018/39149
Editor(s):
(1) Afroz Alam, Department of Bioscience & Biotechnology, Banasthali University, Rajasthan, India.
Reviewers:
(1) Munyaneza Emmanuel, Institute of National Museums of Rwanda, Rwanda.
(2) V. R. Akabari, Junagadh Agricultural University, India.
Complete Peer review History: http://www.sciencedomain.org/review-history/23167

Original Research Article

Received 21st November 2017
Accepted 5th February 2018
Published 13th February 2018

ABSTRACT

Solanum constitutes the largest and most complex genera of the Solanaceae family, many of which are economically important. However, seed germination of the majority of the cultivated species of general *Solanum* is a major constraint to its production. The objective of this study was to investigate the germination behavior of three *Solanum* species under three storage environments at different periods. Seeds of three accessions of the *Solanum* species: *S. macrocarpon*, *S. nigrum* and *S. aethiopicum* were used in this study. The processed seeds of each accession were partitioned into three equal parts and packed in aluminium cans. Samples from the three accessions were kept separately in three storage environments, namely: ambient, short and medium term conditions in February 2015. The stored seed samples were drawn at quarterly intervals starting from May 2015 to February 2016 which constituted three storage periods and evaluated for germination. The laboratory experiment was conducted at Seed Testing Laboratory

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of the National Centre for Genetic Resources and Biotechnology (NACGRAB) Ibadan, Nigeria. The experiment was arranged in 3 x 3 x 3 factorial using completely randomized design (CRD) in three replications. Results of analysis of variance (ANOVA) revealed that effects of *Solanum* species, storage environments, storage periods and their interactions were highly significant (*P*=.01) on germination. The germination values of 18.67%, 41.83% and 44.17% were observed for seed samples stored under ambient, short term and medium term conditions respectively. Also, germination decreased as storage period increased with values of 44.2%, 38.5% and 16.2% for the first, second and third quarters respectively. In conclusion, the result of the interactive effect of storage environments and periods on germination revealed that a medium storage condition is the best environment to store *Solanum* species seeds. However, <50% germination value observed in all the storage periods indicates that further studies could be carried out on effective dormancy breaking method for these species to enhance their germination.

Keywords: Solanum; germination; storage; environments; periods.

1. INTRODUCTION

Solanum constitutes the largest and most complex genera of the Solanaceae family. It is composed of more than 1500 species many of which are economically important such as eggplant, *Solanum melongena* L. [1]. The other species of concern are hairy fruited pea-eggplant (*Solanum stramonifolium* Jacq) and turkey berry, also known as Thai eggplant (*Solanum torvum* Sw). *Solanum nigrum* (black nightshade), *Solanum macrocarpon* (eggplant) and *Solanum aethiopicum* (garden egg) are common vegetables in southern part of Nigeria. Although *S. nigrum* is considered as an important weed in many crops [2,3], this species and *S. macrocarpon* are important leaf vegetables for some people of southern part of Nigeria, especially for the rural dwellers. *S. aethiopicum* also serves as an important fruit vegetable for a large population of rural and urban dwellers of Nigeria.

Seed germination can be described as the initial emergence of the radicle from the seed coat. This process requires that the plant embryo leaves the quiescent state, mobilizes stored nutrients, overcomes the barrier of surrounding tissues, and resumes cell elongation, cell division, and development. The Association of Official Seed Analysts (AOSA) defined seed germination as 'the emergence and development from the seed embryo of those essential structures which, for the kind of seed in question, are indicative of the ability to produce a normal plant under favourable conditions' [4]. Seed germination, therefore, forms an important stage in any plant life cycle and determines the optimal plant density and crop uniformity.

Seed germination of a majority of the cultivated species of general *Solanum* is a major constraint to its production. The reports from earlier studies revealed that different germination rates have been described in different species including some accessions of *Solanum melongena* and related species [5,6]. Study of germination in these species would be of interest for their conservation and utilization in breeding programs.

In Nigeria, one of the national strategies for genetic resources conservation led to the establishment of The National Centre for Genetic Resources and Biotechnology (NACGRAB) located in Ibadan, Nigeria. To fulfil this mandate, NACGRAB has different storage chambers operating at different storage conditions and over the years, NACGRAB had been distributing seeds of *Solanum* species accessions from her collections to meet the requirements of researchers and other users. However, it has been observed that some species of *Solanum* seeds stored in NACGRAB storage chambers have a certain period of time with reduced ability to germinate. In this present study, a preliminary investigation was carried out on seed germination behavior of three species *Solanum* (*S. macrocarpon, S. nigrum and S. aethiopicum*) at different storage periods and conditions. The knowledge gained in this study would be useful in facilitating further laboratory studies towards enhancing germination of seeds of these species. The objective of this study, therefore, was to investigate the germination behavior of three *Solanum* species namely, *S. macrocarpon*, *S. nigrum*, *S. aethiopicum* under three storage environments at different periods.

2. MATERIALS AND METHODS

2.1 Genetic Materials, Seed Processing and Location of the Experiment

Seeds of three accessions of the *Solanum* species: *S. macrocarpon*, *S. nigrum* and *S. aethiopicum* sourced from the genebank of The National Centre for Genetic Resources and Biotechnology (NACGRAB), Moor Plantation, Ibadan were used in this study. Seed regeneration of these materials was carried during the late growing season of 2014 at the experimental farm of NACGRAB. Fruits of the three accessions were harvested at physiological maturity stage and seeds were extracted directly after harvesting. The extraction was done by hand to minimize mechanical damage. Seeds were air-dried in a seed drying machine at 35°C until the seeds reached about 10% moisture content. The laboratory experiment was conducted at Seed Testing Laboratory of The National Centre for Genetic Resources and Biotechnology (NACGRAB) Ibadan, Nigeria.

2.2 Seed Storage and Measurement Temperature and Relative Humidity

The seeds of each accession were partitioned into three equal parts and packed in aluminium cans. Samples from the three accessions were kept separately in three storage environments located in NACGRAB. The first environment was ambient which represents normal room conditions. The second storage environment was a short-term where genetic materials are kept for purpose of distribution and regeneration and the third was medium-term storage environments where genetic materials are kept as safety duplicate. The seed samples were stored in the environments in February 2015. The stored seed samples were drawn at every quarter between May 2015 and February 2016 constituting three storage periods. Electricity supply was ensured for at least twelve hours daily in both short and medium term storage environments. Temperature and relative humidity of both short and medium term storage environments were taken daily but the only temperature of the ambient environment was taken daily.

2.3 Experimental Design

The experiment was arranged in 3 x 3 x 3 factorial using completely randomized design (CRD) in three replication. The three factors were *Solanum* species, storage environments and storage periods. The stored seed samples were drawn at quarterly intervals starting from May 2015 to February 2016 which constituted three storage periods and evaluated for germination.

2.4 Standard Germination Test and Data Analysis

Standard germination test was assayed by placing one hundred seeds in a germination plastic container lined with four layers of tissue paper moistened with 15ml of distilled water. The containers were covered and placed in a germinating chamber at 25 ± 2°C. The seeds were kept moist every day for seven days thereafter germination percentages were determined at seven days after planting according to International Seed Testing Association (ISTA) rules [7].

Data on germination percentage were log transformed to ensure conformity to normality and subjected to analysis of variance (ANOVA) using Statistical Analysis Software, SAS Version 9.1 [8]. However, since ANOVA did not detect any significant difference between transformed and untransformed values, untransformed values were hereby presented. Pertinent means were thereafter separated by the use of the least significant difference (LSD) at 0.05 level of probability using SAS software.

3. RESULTS AND DISCUSSION

3.1 Storage Conditions and Variability among Factors

The mean temperature ranges in the three environments, and mean relative humidity ranges in the short and medium term storage environments during the period of study were presented in Table 1. Temperature under ambient conditions ranged from 23 to 29.5°C while that of the short-term storage environment ranged from 15.1 to 22.6°C. The lowest temperature value was observed under medium-term storage environment which ranged from -4.2 to 4.1°C (Table 1). The mean relative humidity ranges in short and medium-term storage environments were 26.9 to 50.7% and 42.7 to 72.1% respectively (Table 1). Results of analysis of variance (ANOVA) revealed that effects of species of *Solanum* (SPP), storage environment (ENV), storage period (STP) and their interactions were highly significant (P=.01) on germination (Table 2).

Table 1. Mean temperature (°C) and relative humidity (%) ranges in short and medium-term storage environments and mean temperature range under ambient conditions during the experiment

Storage environment	Temperature (°C)	Relative humidity (%)
Ambient	23 to 29.5	---
Short term	15.1 to 22.6	26.9 to 50.7
Medium	-4.2 to 4.1	42.7 to 72.1

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Table 2. Mean squares from the analysis of variance for laboratory germination test conducted on seeds of three *Solanum* species at Seed Testing Laboratory, NACGRAB, Ibadan

Source of variation	df	Germination (%)
Replication	2	39.41ns
Species (SPP)	2	21946.96**
Storage Environment (ENV)	2	3532.74**
Storage Period (STP)	2	11778.07**
SPP x ENV	4	2426.96**
STP x SPP	4	5141.63**
STP x ENV	4	586.07**
STP x SPP x ENV	8	1569.78**
Error	52	1971.26
Total	80	
R^2 (%)		0.96
CV		18.68
Mean		32.96

**, **, Significant at probability level of 0.05 and 0.01, respectively; ns = not significant*

3.2 Germination Performance as Influenced by *Solanum* Species, Storage Conditions and Periods

S. macrocarpon had germination percentage of 56.6%, *S. nigrum* had 26.2% and *S. aethiopicum* had 17.0% (Table 3). This result confirmed the fact that genetic constitution is a major factor in determining germination potential of seeds in any storage environment. The results support the findings of Olosunde et al. [9] where they found a significant difference for germination in two varieties of cowpea. The lowest mean germination percentage (23.0%) was recorded for seed samples stored under ambient conditions (Table 3). Adriana et al. [10] had similar conclusion where they stated that seeds stored in ambient conditions lose their viability and vigour very fast due to changes in temperature and relative humidity.

However, mean germination percentages for *Solanum* seeds stored under short-term (37.3%) and medium term (37.9%) conditions were not significantly different (Table 3). Also, germination of *Solanum* seeds was significantly influenced by storage period in this study. There was a significant decrease in germination percentage as duration in storage environment increased. Germination values of 44.2% were observed at first period (three months in storage) which decreased significantly to 38.5% and 16.2% at the second period (six months in storage) and third period (nine months in storage) respectively. Some authors also gave similar reports on other species of crops such as cowpea (Olosunde et al. [9], Brassica (*Brassica campestris*) [11] and *Rumex scutatus* [12].

Table 3. Influence of *Solanum* species, storage environments and storage periods on seed germination at NACGRAB, Ibadan

Factors	Seed germination (%)
A. Species	
S. macrocarpon	55.6a
S. nigrum	26.2b
S. *aethiopicum*	17.0c
LSD	2.9
B. Storage environment	
Ambient	23.6b
Short term	37.3a
Medium term	37.9a
LSD	3.4
C. Storage period	
First quarter	44.2a
Second quarter	38.5b
Third quarter	16.2c
LSD	3.4

Means with different letters within the column of the same factor are significantly different at P=0.05

3.3 Germination Performance as influenced by the Interactive Effect *Solanum* Species and Storage Conditions

The germination of seeds of *Solanum* species was significantly influenced by the interactive effect of the species and storage environment. This implies that germination of *Solanum* species varied from one storage environment to another. The germination percentage of *S. macrocarpon* stored under ambient (ENV1) environment was 39.1%, which was significantly lower than germination percentages of *S. macrocarpon* stored under short (ENV2) and medium (ENV3)

term storage conditions with germination values of 63.3 and 64.4% respectively (Table 4). Germination response of *S. nigrum* to the three storage environments was significantly different with germination values of 14.0, 29.6 and 35.1% for ambient, short and medium term storage conditions respectively (Table 4). There was no significant difference in germination percentages of *S. aethiopicum* stored under the three storage conditions with germination values of 17.8, 19.1 and 14.2% for ambient, short and medium term storage conditions respectively (Table 4). These results support the findings of Olosunde et al. [9] where they found out that germination of cowpea seeds varied from one storage environment to another.

Table 4. Mean germination of seeds of three *Solanum* species as affected by the interactive effect of species and storage conditions at NACGRAB, Ibadan

Species of *Solanum* (SPP)	Storage environment (ENV)	Germination (%)
SPP1	ENV1	39.1
SPP1	ENV2	63.3
SPP1	ENV3	64.4
SPP2	ENV1	14.0
SPP2	ENV2	29.6
SPP2	ENV3	35.1
SPP3	ENV1	17.8
SPP3	ENV2	19.1
SPP3	ENV3	14.2
LSD		14.9

SPP1= S. macrocarpon, SPP2= S. nigrum, SPP3= S. aethiopicum
ENV1= Ambient, ENV2=Short term, ENV3=Medium term

3.4 Germination Performance as Influenced by the Interactive Effect *Solanum* species and Storage Conditions

Germination response of *Solanum* species also varied from one storage period to another. Germination of *S. macrocarpon* seeds (SPP1) at the third quarter (23.6%) was significantly lower than that of first (72.9%) and second (70.4%) quarters (Table 5). Similarly, germination of *S. nigrum* seeds (SPP2) at the third quarter (14.2%) was significantly lower than that of first (37.3%) and second (27.1%) quarters (Table 5). However, for *S. aethiopicum,* the germination values among the three quarters were not significantly different with values of 22.2, 18.0, and 10.9% for first, second and third quarter respectively. Olosunde et al. [13] gave the similar report that germination performance of *Amaranthus cruetus* varied from one storage period to another.

Table 5. Mean germination of seeds of three *Solanum* species as affected by the interactive effect of species and storage periods at NACGRAB, Ibadan

Species of *Solanum* (SPP)	Storage period (STP)	Germination (%)
SPP1	STP1	72.9
SPP1	STP2	70.4
SPP1	STP3	23.6
SPP2	STP1	37.3
SPP2	STP2	27.1
SPP2	STP3	14.2
SPP3	STP1	22.2
SPP3	STP2	18.0
SPP3	STP3	10.9
LSD		11.3

SPP1= S. macrocarpon, SPP2= S. nigrum, SPP3= S. aethiopicum
STP1= First quarter, STP2= Second quarter, STP3= Third quarter

3.5 Germination Performance as Influenced by the Interactive Effect Storage Conditions and Periods

The significant storage environment by period interaction implies that germination of *Solanum* seeds observed at each quarter varied with storage environment. Although, for the first quarter, the germination percentages among the three environments were not significant with respective germination values of 40.0%, 45.3% and 47.1% for ambient, short and medium term storage conditions. The reason for this non-significant differences might be as a result the early storage stage. However, the difference was pronounced at the second quarter with respective values of 27.6% 44.3% and 43.6% for ambient, short and medium storage environments. At the last quarter, which was nine months in storage, the lowest germination value was observed on *Solanum* seeds stored under the ambient environment (3.3%) which was significantly different from the values observed under short (22.2%) and medium (23.1%) term storage conditions (Table 6). These results suggest that in evaluating for *Solanum* species, environmental conditions of the storage chamber should be given prime consideration. However,

Table 6. Mean germination of seeds of three *Solanum* species as affected by the interactive effect of storage conditions and periods at NACGRAB, Ibadan

Storage period (STP)	Storage environment (ENV)	Germination (%)
STP1	ENV1	40.0
STP1	ENV2	45.3
STP1	ENV3	47.1
STP2	ENV1	27.6
STP2	ENV2	44.4
STP2	ENV3	43.6
STP3	ENV1	3.3
STP3	ENV2	22.2
STP3	ENV3	23.1
LSD		20.4

ENV1= Ambient, ENV2=Short term, ENV3=Medium term
STP1= First quarter, STP2= Second quarter, STP3= Third quarter

the non-significant values observed between the *Solanum* species seeds stored under short and medium term storage conditions across the storage periods in this present study could be attributed to the fluctuation in electricity supply, which could have masked the anticipated differences. Olosunde et al. [9] had earlier observed this in the storage experiment carried out on two cowpea varieties. This suggests that constant power supply to the cold-rooms for seed storage should not be compromised or probably the duration of power supply should be close to 24 hours per day if not 24/7. Furthermore, It was also observed that in all the storage periods, none of the environments was able to produce up to 50% germination. This finding is in agreement with the earlier report of Adebola and Afolayan, [14]; Taab and Andersson, [15] who reported low germination rates in different species of the genus *Solanum*. Ibrahim et al. [5] further stated that, seeds of a number of locally cultivated *Solanum* species are known to emerge slowly, and about 30 days could be needed to attain germination with percentage rates between 15 and 50% in S. *incanum,* S. *torvum,* S. *integrifolium,* S. *surattense,* S. *khasianum,* S. *sanitwongsei* and hybrids of S. *melongena* x S. *ingrifolium*.

4. CONCLUSION

In conclusion, there was differential germination response of *Solanum* species seeds to storage environments with lowest germination values recorded under ambient conditions. However, there was no significant difference between the germination of *Solanum* species seeds stored under medium and those in short-term storage despite the different storage conditions. This suggests that duration of power supply to the genebanks should be given priority in order to obtain anticipated differences between the cold environments. Furthermore, <50% germination values observed irrespective of the storage periods suggests that further studies could be carried out on effective dormancy breaking method for these species to enhance their germination.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sciencedomain.org/review-history/23167