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Physiological quality of stored soybeans (*Glycine max* L.)

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ABSTRACT

Objective: to evaluate the physiological quality of 11 soybean (*Glycine max* L.) varieties stored under natural conditions in the state of Yucatán, Mexico.

Design/Methodology/Approach: seeds of 11 soybean varieties were stored for 11 months in sealed polythene bags. At the start of storage, the seeds were evaluated for germination, electrical conductivity, and 100 seeds were weighed. From months 3 to 11, the germination and electrical conductivity of each sample were assessed monthly. The initial and final weights of 100 seeds were obtained. The data obtained were analyzed in a completely randomized scheme with factorial arrangement.

Results: differences were observed ($p \leq 0.05$) in storage tolerance between the varieties, however, the loss of physiological quality was noticeable beginning at the third month of storage. The Tamesi and H-100 varieties showed the seed's greatest loss in germination and weight, and the highest value of electrical conductivity.

Limitations/Implications: soybeans are produced in the Yucatán Peninsula, but limitations to conserving the seeds are high temperatures and relative humidity which can be common in the state.

Conclusions: soybeans exhibit problems in maintaining their physiological quality in storage under natural conditions, and these problems are also reflected in the loss of seed weight.

Keywords: germination, electrical conductivity, natural storage.

INTRODUCTION

Seeds are the main input for food production. These must meet certain characteristics in quality in order to express their potential in the field (Doria, 2010). Starting in the sowing stage, quality should be safeguarded with adequate technical management. However, postharvest handling is essential mainly to conserve the physiological qualities that allow for successful subsequent cultivation cycles (Balesdent *et al*., 2018). Seed storage is an indispensable step to maintain viability and germination, as well as to reduce seed aging to the minimum. The seeds' response to storage conditions differs among species, varieties, and even plots (Matsue *et al*., 2005).

The seeds of oleaginous species are challenging to preserve because they are composed of large quantities of oil, which contribute to an increase in the formation of free radicals that consequently accelerate the seeds' deterioration (Rahmati and Shaban, 2014). Nagel and Börner (2010) report that oleaginous seeds demonstrated a short storage duration under room temperature conditions (20.3±2.3 °C and 50.5±6.3% relative humidity), as in the case of

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sunflower (*Helliathus annus* L.), which in five years showed a reduction in germination to 50%, and in 10 years to less than 10%. In this sense, seeds such as soy present what some authors have called a "short shelf life" (Walter *et al*., 2005). Storage in tropical regions complicates the preservation of oleaginous seed quality (Kausar *et al*., 2009) due to the high temperatures and relative humidity that predominate in these regions. After analyzing 182 soybean accessions (*Glycine max* L.) stored for 34 years under controlled temperature and humidity conditions, Desheva *et al*. (2017) observed differences in seed viability between them. Shelar *et al*. (2008) found decreased germination in soybeans stored in room temperature conditions compared to seeds stored in liquid nitrogen, in addition to finding differences in the germination response between varieties in terms of tolerance to storage. Due to all of the above, this study evaluated the physiological quality of eleven soy varieties stored in room temperature conditions in Yucatán, Mexico.

MATERIALS AND METHODOLOGY

Genetic Material

The experiment took place in the facilities of INIFAP's Mocochá Experimental Field, located on Kilometer 25 of the old Mérida-Motul highway in the municipality of Mocochá. Eleven varieties of soy were used [Mariana, Luziania, Vernal, Tamesí, Otoño, Huasteca 100 (H 100), Huasteca 200 (H 200), Huasteca 300 (H 300), Huasteca 400 (H 400), Huasteca 600 (H 600), and Huasteca 700 (H 700)], produced in the 2018 spring-summer cycle and stored for eleven months under natural regional conditions (≥30 °C, ≥90% relative humidity) (Figure 1).

The harvested seeds were stored in laminated, hermetically-sealed bags with one-kilogram capacity. All the bags were labeled according to its corresponding variety and placed in room temperature and ambient humidity storage. Before storing the seeds, the germination, electrical conductivity, moisture content and weight of 100 seeds were evaluated. Moisture content was measured using a portable John Deere SW08120 moisture tester, and using the laminated bags, all varieties were kept at 12% moisture during storage. The first sampling was carried out after the third month of storage to quantify the physiological quality, followed by monthly repetitions.

Physiological Quality

Samples from each of the varieties were taken randomly and their germination percentages were determined. Four repetitions with 25 seeds were prepared following the procedure stated by ISTA guidelines (2005). The seeds were placed on paper towels pre-soaked in distilled water, uniformly distributed (25 seeds per repetition), and these were rolled up and placed vertically in a germination chamber at 25 °C±1. The first count was made four days after sowing and the second was made at seven days. The number of normal seedlings was counted and the result was reported as a percentage. For electrical conductivity (EC), the methodology proposed by Moreno was used (1996), consisting in two repetitions of 50 seeds that were weighed with an approximation of 0.01 g between samples; the seeds were placed in beakers with a diameter of 80 mm±5 mm containing 250 mL of deionized water at 25 °C. The beakers containing the seeds were kept at 25 °C±1 for 24 h within the germination chamber with controlled temperature of 25 °C±1. At the end, the seeds were separated from the water and this was where the electrical conductivity

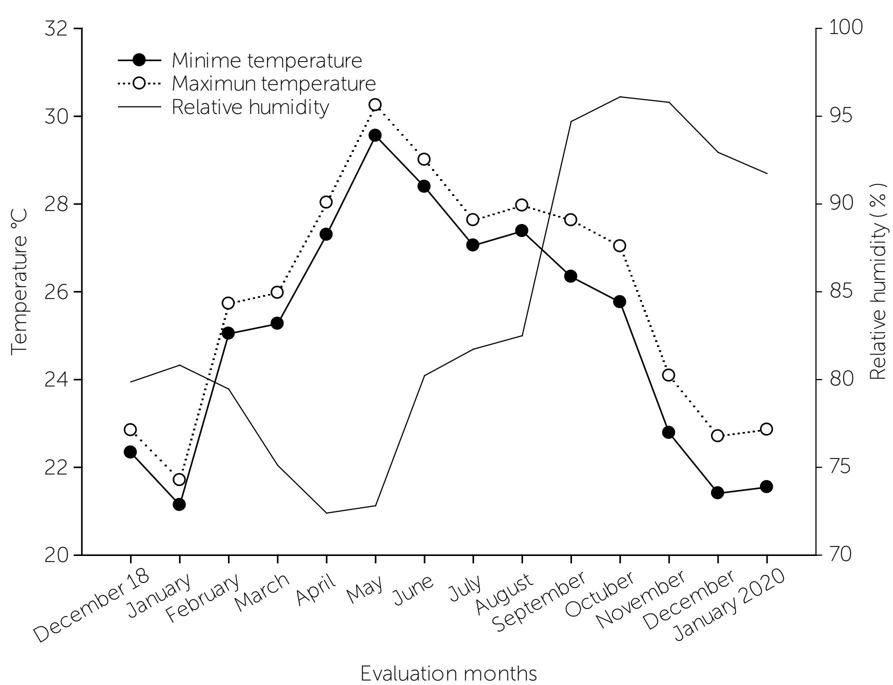

Figure 1. Maximum, minimum temperature and relative humidity conditions present during the study. Data obtained from https://datos.gob.mx/busca/dataset/red-nacional-de-estaciones-agrometeorologicas-automatizadas-inifap

was then measured with an OAKTON Mod. CON 150 electrical conductivity meter; the results for seeds were registered in $\mu Scm^{-1} g^{-1}$.

Weight of 100 Seeds

Ten samples of 100 seeds of each variety were weighed and their weight was registered at the start of storage and at the end (after 11 months). To ascertain whether the experiment was valid, it was corroborated with the methodology proposed by Moreno (1996). The results were reported in g.

Statistical Analysis

All the assessments (EC, germination, and weight of 100 seeds) were evaluated in a completely randomized factorial design, composed of two factors. In the case of EC and germination, the factors were 11 varieties and 11 months of storage; in the case of the weight of 100 seeds, the factors were varieties, and initial and final storage weight. Only the germination data was modified using arc sine, and both cases were subjected to variance analysis using the SAS 9.1® statistical package. Means comparison was done with the Tukey test ($p \leq 0.05$) and the results were expressed in the original unit.

RESULTS AND DISCUSSION

The analysis of simple factors presented differences ($p \leq 0.05$). The varieties that reported the least EC were H 200 and Luziania, while the highest reading was for H 100 with a difference of 16.34 and 16.05 $\mu Scm^{-1} g^{-1}$ of seed respectively. Storage time influenced the EC, increasing by 56.91 $\mu Scm^{-1} g^{-1}$ of seed from month zero to 11 months (Table 1). According to Moreno (1996), EC values above 30 and up to 43 $\mu Scm^{-1} g^{-1}$ of seed should not be used for early sowing due to faults in their sprouting, especially in adverse conditions.

Thus, the highest EC corresponded to the lowest germination response. The H 100 variety registered the lowest germination percentage while H 200 and Luziania maintained higher percentages, showing differences of 16.77 and 17.77 percentage points, respectively. During storage, the values with highest EC found coincided with a reduced germination response (Table 1). The difference between initial and final germination was 80.84%. Germination begins decreasing by close to 50% after the first three months of storage, when the EC was over 40 $\mu Scm^{-1} g^{-1}$ of seed.

Table 1. Response of simple factors evaluated in *Glycine max* L. seeds.

Factors / Variety	CE ($\mu Scm^{-1} g^{-1}$ of seed)	Germination (%)
Otoño	70,39 c	38,4 a
Mariana	69,47 c	36,55 abc
Luziania	62,33 d	42,37 a
Vernal	77,43 ab	37,1 ab
Tamesí	70,49 c	30,27 de
H100	78,38 a	24,6 e
H200	62,04 d	41,37 a
H300	70,53 c	30,15 de
H400	72,31 bc	30,8 cd
H600	73,56 abc	27,12 de
H700	69,47 c	31,22 bcd
dms	5,5	5,96
Months of storage		
0	38,33 g	86,09 a
3	53,48 f	47,182 b
4	61,99 e	48,45 b
5	67,92 d	29,09 c
6	71,36 d	22,45 de
7	72,15 d	24,45 cd
8	78,97 c	26,09 cd
9	79,48 c	29 c
10	87,17 b	18,45 e
11	95,24 a	5,25 f
dms	5,15	5,59

Means with the same letters are not statically different (Tukey 0.05). CE: Electrical conductivity. dms: minimum signifcan difference.

The deterioration of seeds begins from the moment of formation and it is a constant process, regulated by genetic and environmental aspects like temperature and relative humidity. On these last two depends the speed of deterioration, which will be reflected as low or null germination strength and loss of viability (Shelar *et al*., 2008).

The EC allows estimating the integrity of cellular membranes and the loss of cytoplasmic solutes, which are correlated with the rapid deterioration of seeds (Tajbakhsh, 2000). Soybeans are characterized by having high levels of oil, although these tend to oxidize, increasing the free fatty acids that accelerate aging (Motlagh and Shanba, 2014). This is why increases in EC and low germination were found in all the varieties during storage time.

Agronomic management also affects soybean quality. The implements used during harvest as well as seed moisture influence quality. This is why it is very important to ensure that the seed does not present fractures in the testa, as these cause a rapid loss of viability due to the entry of pathogens or detachment of the cotyledons (Bauer *et al*., 2003). When combining factors, differences could be observed in the response of EC and germination ($p \leq 0.05$). During storage, all varieties increased the EC; however, the response was not equal. While the Mariana variety increased by 12.58 $\mu Scm^{-1} g^{-1}$ of seed, the H 100 variety showed an increase of 79.08 $\mu Scm^{-1} g^{-1}$ of seed more than its initial value, which was 34.82 $\mu Scm^{-1} g^{-1}$ of seed (Figure 2a). These increases in EC are reflected in the germination percentage of all varieties during the eleven months of storage. This percentage gradually decreased until reaching percentages in the order of 1 to 5%. The H 200 variety was the most affected: germination was reduced from 95 to 2%, while for H 100 and H 300, the reduction was less drastic for 10 months until falling to 6.8% in the eleventh month (Figure 2b). None of the varieties withstood storage in natural conditions. Seed viability diminished rapidly, and this could be attributed to the temperatures in Yucatán that can reach higher than 30 °C, and a relative humidity that can rise above 90% (Figure 1). It has been demonstrated that these temperatures tend to damage the quality of seeds, reducing certain characteristics like sprouting vigor and speed, germination rate, and an increase in solute leaching (Doria, 2010). Sharma *et al*. (2013) observed that, with the passage of time in storage, lipid peroxidation increases considerably and antioxidant activity decreases. Unwelcome antioxidant activity in seeds causes damage to cellular membranes that lead to decrease in or loss of germination.

As long as the EC was lower than 60 $\mu Scm^{-1} g^{-1}$ of seed, germination was maintained at 50%; however, with more months in storage, the EC increased, causing a reduction in germination. Although the deterioration of the seeds in storage is an inevitable process, the pace at which it occurs depends on storage conditions, seed quality, variety, and the type of seed stored (Cruvinel *et al*., 2017). In this context, Manjarrez *et al*. (2017) reported the loss of physiological quality in canola seeds (*Brassica napus* L.) stored from one to five years, and recommended not storing them for more than a year owing to reduced germination. Pereira *et al*. (2017) reported that soybeans have tolerance to storage that depends on the state of seed development during harvest and the storage conditions. The same

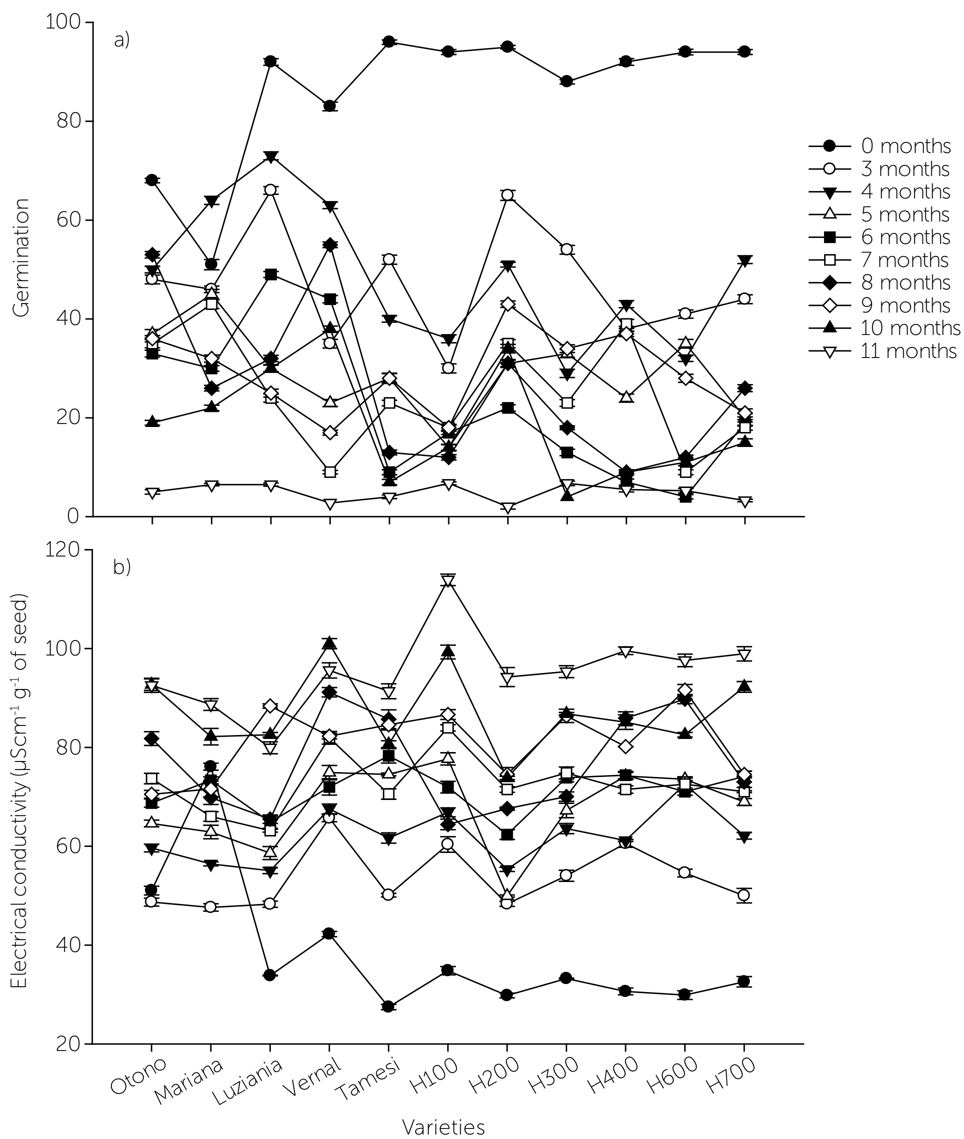

Figure 2. Response of eleven varieties of *Glycine max* L. stored for eleven months under room temperature conditions in Yucatán, Mexico. A) Germination, b) Electrical conductivity.

authors indicate that soybeans stored at 35 °C and at 75% relative humidity demonstrate germination reduced to zero in just 90 days of storage. The storage of seeds in general is affected mainly by temperature and relative humidity (Mohammadi *et al.*, 2011), although the type of container used for storing will have an influence on maintaining germination and optimal health for use in the field (Puri *et al.*, 2017). The loss of weight associated with deterioration was evident, and differences were observed in all varieties (p≤0.05). The varieties of highest weight were Tamesí and H 100, and the varieties of lowest weight were H 600 and Otoño, with a difference of 3.73 g between the higher-weight and lower-weight varieties. The average weight at the start of sampling surpassed by one gram the average weight at the end of the eleven months of storage (Table 2).

Table 2. Weight response of 100 *Glycine max* L. seeds.

Factors / Varieties	Weight of 100 seeds (g)
Otoño	14,47 cd
Mariana	14,98 bc
Luziania	15,10 bc
Vernal	15,05 bc
Tamesi	17,91 a
H100	17,49 a
H200	14,56 bcd
H300	14,69 bcd
H400	15,31 b
H600	14,18 d
H700	15,19 bc
dms	0,76
Sampling during storage	
Initial	15,86 a
Final	14,86 b
dms	0,19

Means with equal letters are not statistically different (Tukey, 0.05). dms: Minimum significant difference.

The reduction in weight during the storage period was different in all the varieties, with Tamesí and H 100 as the most affected, showing a reduction of 2.54 and 4.14 g respectively. The reductions in the rest of the varieties were from 1.2 to 0.2 g (Figure 3). This response is the result of natural seed deterioration when subjected to tropical conditions in which respiration levels are altered, causing deterioration of membranes, protein denaturalization, and increases in free radicals, resulting also in reduced weight (Carbajal *et al.*, 2017).

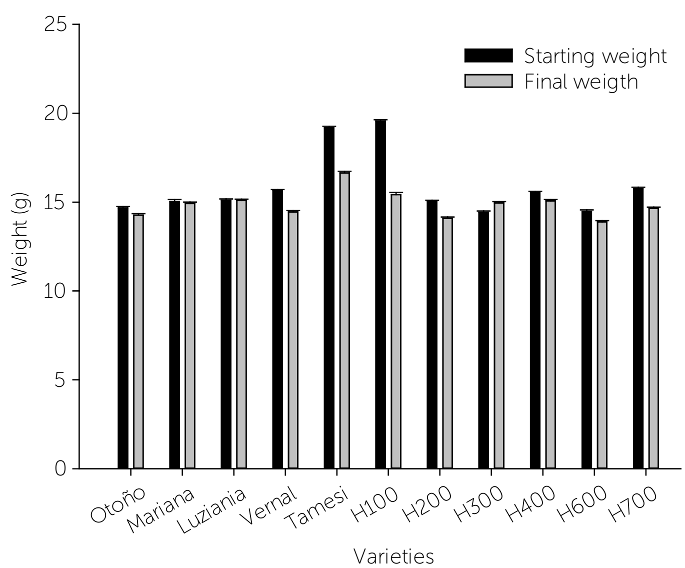

Figure 3. Weight loss in *Glycine max* L. varieties stored for eleven months.

The varieties that showed the greatest loss in weight correspond to those with the lowest germination percentage and highest conductivity found at the end of storage. These results agree with what Pérez-Camacho *et al.* (2008) reported, a reduction in seed weight attributed to the consumption of the seeds' reserves.

CONCLUSIONS

Soybeans have low tolerance to storage under natural conditions in the Yucatán Peninsula. Their physiological quality is affected at temperatures of approximately 30 °C, which is why it is not advisable to store this type of seed without the necessary temperature conditions. The deterioration response of soybeans was different among varieties, which could be worth considering within genetic improvement programs.

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