



# Water absorption and storage tolerance of soybean seeds with contrasting seed coat characteristics

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**ABSTRACT.** Seed coat characteristics may be related to seed quality and longevity, attracting the interest of breeding programs. This study aimed to evaluate the relationship between the water absorption parameters and storage tolerance of soybean seeds with contrasting seed coat characteristics. For this purpose, seeds of five soybean cultivars with different seed coat colors and lignin contents were used. Before storage, the seed coat lignin content, moisture content, water absorption rate in 11 hydration periods (1, 2, 3, 4, 5, 6, 7, 8, 24, 32, and 48 hours), hydration speed index, germination, viability, and seed vigor were determined. After six months of storage in a dry and cold chamber (at 11°C and 54% relative humidity [RH]) and uncontrolled environment (at 25°C and 71% RH), the seeds were evaluated for germination, viability, and vigor to quantify changes in their physiological quality during storage and relate them to the seeds water absorption parameters for the different cultivars. The seed coat lignin content is negatively correlated with the seeds water absorption parameters. Soybean cultivars have different storage tolerance levels. Seeds that absorb larger amounts of water have lower tolerance to deterioration related to temperature and relative humidity fluctuations that occur in storage under uncontrolled conditions, negatively affecting the physiological potential of seeds.

**Keywords:** deterioration; soaking; *Glycine max* (L.) Merrill; lignin; permeability.

Received on April 10, 2020.  
Accepted on July 2, 2020.

## Introduction

The seed coat protects the embryonic axis and reserve tissues against agents of climatic, biotic or mechanical nature, keeps the seed internal parts together, controls the hydration and gas exchange rate between the seed and environment, and regulates germination (Miller et al., 2010; Radchuk & Borisjuk, 2014; Marcos-Filho, 2015). Therefore, the quality of seeds is directly related to the seed coat characteristics, which are variable for different species and genotypes of the same species.

Soybean seeds can present permeable seed coats, which have as a main characteristic easy imbibition, and, semi-permeable seed coats, which restricts, in different levels, water imbibition by seeds. Seed coat semi-permeability is an important characteristic with regard to the seed susceptibility to deterioration in the field, and has an influence on storage potential, resistance to attack by pests and diseases, and mechanical and imbibition damage (Souza & Marcos-Filho, 2001; Henning, Maia, Mertz, Zimmer, & Oliveira, 2009; Mertz et al., 2009).

The chemical composition, structural arrangement and intercellular substances present in the seed coat layers will determine its degree of permeability (Qutob, Ma, Peterson, Bernards, & Gijzen, 2008; Vu, Velusamy, & Park, 2014). In this context, the influence of lignin on this characteristic has been highlighted by Kuchlan, Dadlani, and Samuel (2010) and Bellaloui, Smith, and Mengistu (2017). In addition, the high seed coat lignin content gives seeds lower permeability, making them less susceptible to deterioration (Marwanto & Marlinda, 2003).

Another characteristic associated with water absorption by seeds is the seed coat color. Ertekin and Kirdar (2010) and Santos, Póla, Barros, and Prete (2007) found that seeds with dark-pigmented coats have lower imbibition rates than seeds with yellow coats. According to Santos et al. (2007), this result is related to the lignin levels in the coat of these seeds.

The lower hydration rate of seeds can mitigate the effects of deterioration both in field pre-harvest conditions and during storage. This hypothesis is related to the hygroscopic character of seeds, in which their

water content is in balance with the air relative humidity (RH) under specific storage temperatures. Thus, knowing that the hygroscopic equilibrium point varies according to the genotype characteristics, it is considered that seeds that absorb water at lower rates, especially in the first hours of imbibition, may present greater tolerance to RH fluctuations that occur during the storage period, contributing to the maintenance of the physiological quality of seeds throughout this period, mainly under uncontrolled environmental conditions.

The storage period for soybean seeds varies from six to eight months and is a crucial stage in the production chain. In Brazil, producing regions are largely inserted in tropical and subtropical ecosystems, in which germination conservation and especially seed vigor are difficult to achieve when seeds are stored in environments without temperature and RH control (Zuchi, França-Neto, Sediyaama, Lacerda Filho, & Reis, 2013; Chandra et al., 2017). In addition, another finding is the difference in seed longevity among soybean cultivars, which may also be related to the seed coat characteristics.

Therefore, information on seed coat characteristics and their relationship with the quality and longevity of seeds has increasing importance for breeding programs in the development of new soybean cultivars. In addition, this information can be used in the planning of seed storage and marketing.

Thus, this study aimed to evaluate the relationship between the water absorption parameters and storage tolerance of soybean seeds with contrasting seed coat characteristics.

## Material and methods

The experiment was developed at the Brazilian Agricultural Research Company, National Soybean Research Center (Embrapa Soja), in the Chemistry and Physiology laboratories, Department of Seed and Grain Technology, municipality of Londrina, state of Paraná, Brazil.

Seeds of five soybean cultivars with contrasting seed coat color and lignin content characteristics were used: BRSMG 715A, DM 6563 IPRO, BRS 413 RR, BRS 1003 IPRO, and BMX Valente RR (Table 1).

Seeds were placed in paper packaging and stored for a period of six months in two environments: a dry and cold chamber (CC; under controlled temperature and RH conditions) and an uncontrolled environment (UE; under natural conditions). During the experiment, temperatures and RHs in both environments were monitored using a data logger equipment model HT-500 (Figure 1).

Assessments related to the seed coat characteristics and water absorption were carried out before storage, while evaluations regarding physiological performance were carried out both before storage (zero months) and at six months of storage. The following methodologies were used.

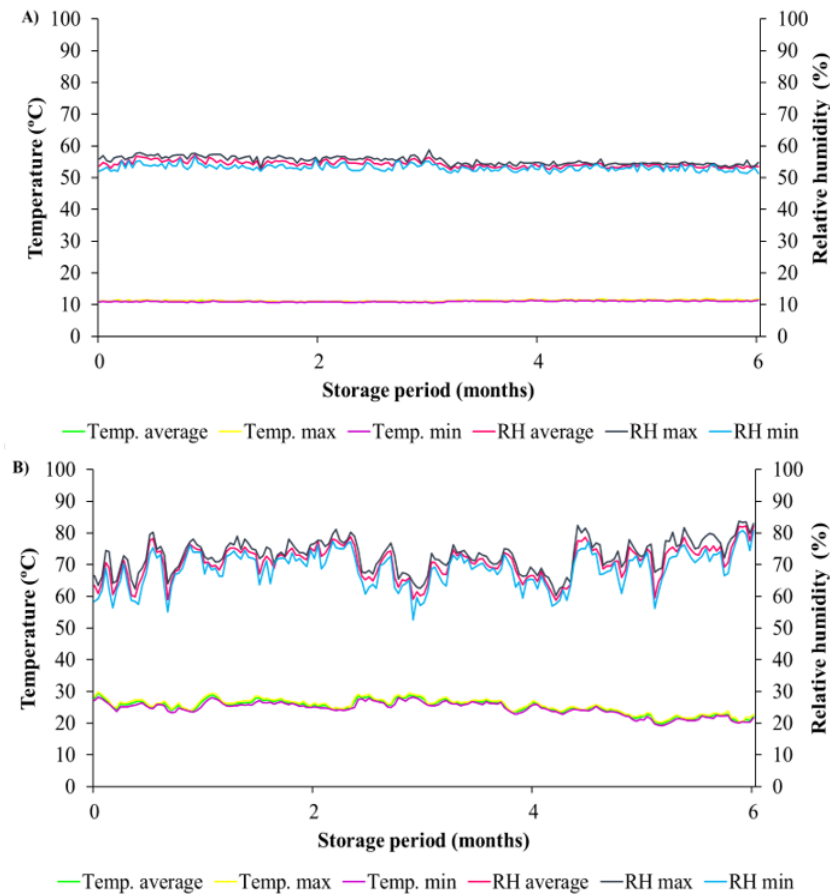
**Table 1.** Characterization of soybean cultivars used in terms of seed color and lignin content.

ID <sup>1</sup>	Cultivar	Seed coat	
		Color	Lignin content (%)
A	BRSMG 715A	Black	14.07 a
B	DM 6563 IPRO	Yellow	4.43 b
C	BRS 413 RR	Yellow	4.19 b
D	BRS 1003 IPRO	Yellow	3.80 c
E	BMX Valente RR	Yellow	3.29 d

<sup>1</sup>ID: acronym used to identify cultivars. Means followed by same letter in the column do not differ by Tukey's test at 5% probability. Coefficient of variation lignin content: 1.83%.

### Seed coat lignin content

Determined with four replicates of 100 seeds for each cultivar, which were initially immersed in water for 12 hours. Then seed coats were removed and dried in an oven at 105°C for 24 hours. The dry matter mass obtained was crushed and homogenized. Subsequently, 300 mg of seed coat was weighed and submitted to centrifugation (3300 rpm for six min.) with different solutions (sodium and potassium phosphate, Triton X-100, 1.0 M NaCl, deionized water, and acetone) for obtaining the cell wall. After this process, tubes with samples were transferred to a vacuum desiccator and then placed in an oven at 60°C. After drying, samples were macerated and the protein-free material was obtained. Subsequently, the total lignin was quantified using the acetyl bromide method (Moreira-Vilar et al., 2014). Results were expressed as percentages.



**Figure 1.** Maximum, average, and minimum daily temperature (°C) and maximum, average, and minimum daily relative humidity (RH) (%) during the storage period of soybean seeds in the cold and dry chamber (A) and uncontrolled environment (B).

### Seed moisture content

Determined by the laboratory oven method at 105°C using four replicates for each treatment, according to methodology described in the Rules for Seed Analysis (Brasil, 2009). Results were expressed as percentages.

### Water absorption rate

Water absorption was quantified in 11 hydration periods (1, 2, 3, 4, 5, 6, 7, 8, 24, 32, and 48 hours), using four replicates with 25 seeds per treatment. Initially, each sample was weighed (initial mass) using a scale (accuracy of 0.0001 g) and placed between two germitest paper sheets moistened with distilled water inside a plastic box. Distilled water corresponding to 2.5 times the paper weight was used (Brasil, 2009). The plastic boxes were placed in a germinator chamber at 25°C. At predetermined times, weighing was performed to quantify the wet mass gain. At each weighing, seeds were removed from the boxes, placed on paper to remove the excess external water, weighed and then returned to the boxes between the moistened paper and germinator. At the end of 48 hours, seeds were removed from the boxes and weighed (final mass). From the initial (*MI*) and final (*MF*) masses of each sample, the mass gain (*GM*) in percentage of water absorbed in the seeds was determined for each soaking time using the following formula:  $GM = 100 \times (MF - MI) / MI$ .

### Hydration speed index

The hydration speed index (HSI) was determined based on the germination speed index formula (Maguire, 1962), with the replacement of germination data by the amount of water absorbed and evaluation days by hydration periods (Nakagawa, Cavariani, Martins, & Coimbra, 2007).

### Germination test

Performed with two subsamples of 50 seeds per replicate, totaling 400 seeds per treatment. Seeds were distributed in rolls of germitest paper, moistened with distilled water in the amount of 2.5 times the substrate weight. After assembly, rolls were placed in a germinator chamber at 25°C. After eight days, evaluations were

carried out according to the recommendations of the Rules for Seed Analysis (Brasil, 2009), and results were expressed as the percentages of normal seedlings.

### Tetrazolium test

Evaluation was carried out with 50 seeds per replicate, preconditioned on germitest paper moistened with distilled water for a period of 16 hours in a germinator chamber under constant temperature of 25°C. After this period, seeds were transferred to 50 mL plastic cups and completely submerged in tetrazolium solution (2,3,5 triphenyl tetrazolium chloride) at 0.075% concentration and kept at 40°C for approximately 150 and 240 min. for yellow and black seed coats, respectively. After the staining process, seeds were washed and kept submerged in water until evaluation. Subsequently, seeds were evaluated individually, sectioned longitudinally and symmetrically with the aid of a scalpel blade and classified according to criteria proposed by França-Neto and Krzyzanowski (2018). Viability was represented by the sum of percentages of seeds belonging to classes from 1 to 5, and vigor level by classes from 1 to 3. Results were expressed as percentages.

### Statistical analyses

For each storage environment, a completely randomized design with four replicates was adopted, with treatments applied in a  $5 \times 2$  factorial scheme (cultivars  $\times$  storage periods). Data for the water content and hydration speed index variables were obtained at the beginning of the experiment; therefore, the cultivar was the only source of variation in the analysis of variance (ANOVA). ANOVAs for germination, viability, and vigor data, which were obtained at the beginning and end of the experiment, considered the complete factorial model. Significant inter- and intra-factor means in the ANOVA ( $F$ -test) were compared using Tukey's multiple comparison test. Analyses were performed using the SAS/STAT software, version 9.4. Copyright© 2016 SAS Institute Inc.

The water absorption pattern of each cultivar was analyzed based on the asymptotic nonlinear regression model proposed by Stevens (1951), following parameterization by Ritz, Baty, Streibig, and Gerhard (2015):  $f(x) = c + (d - c)[1 - \exp(-x/e)]$ . The parameters of this equation have direct biological interpretation, in addition to being more adequate to describe responses to stimuli of increasing intensity in samples from non-normal populations. In this study,  $f$  represents the relative increase in the water absorption of seeds after  $x$  hours of hydration, while parameters  $c$  and  $d$  represent the lower (water absorption by seeds due only to the initial contact with the moisture surface,  $x = 0$ ) and upper (volume of absorbed water,  $\lim_{x \rightarrow \infty} f(x)$ ) limits of  $f$ . Finally, parameter  $e$  determined the humidity growth step described by  $f$  as the imbibition time  $x$  increased. Therefore, according to the parameterization adopted, the higher the  $e$  value, the lower the water absorption rate of the cultivar within the studied period.

Estimates of  $f$  parameters were obtained using the 'drc' package in R (Ritz et al., 2015; R Core Team, 2018), applying the following restrictions:  $c \geq 0$  and  $d \leq 100$ . Subsequently, the 'lmtest' (Zeileis & Hothorn, 2002) and 'sandwich' packages (Zeileis, 2004) were used to adjust the estimates of standard errors, obtained by the 'drc' package regarding the presence of outliers. From these curves, the 20<sup>th</sup> and 50<sup>th</sup> percentile values and their 95% confidence intervals were obtained. To display these values, a figure was built using the HighLow command from the 'sgplot' procedure of the SAS/STAT system.

In addition to the water absorption curves, Pearson's correlation coefficients ( $r$ ) for water absorption rates in each of the 11 hydration periods and HSI, and the lignin content, vigor, and viability, which are variables that indicate physiological quality, were also calculated using R.

## Results and discussion

For the water content, there was no significant difference between cultivar, showing the water content uniformity of seeds of different cultivars (Table 2), which is essential for studies of this nature; according to Silva and Villela (2011), the initial water content of seeds interferes with the imbibition process.

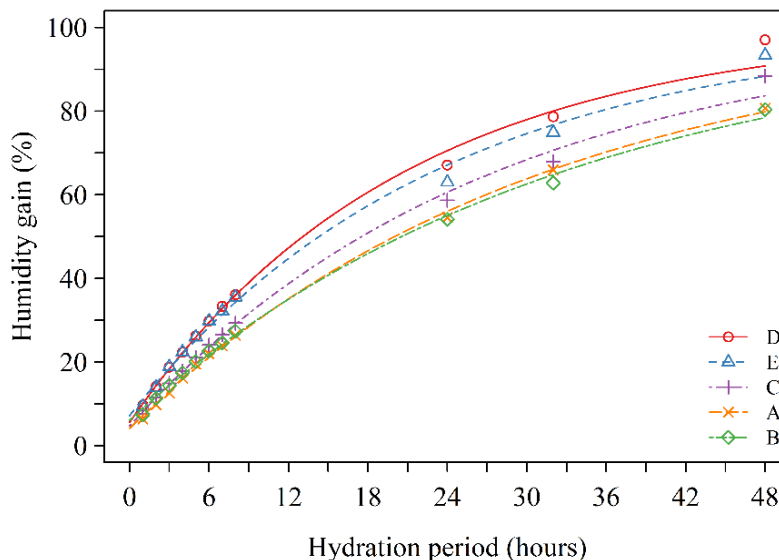
The water absorption curves for seeds of each cultivar suggests that easy water absorption is an intrinsic characteristic of cultivars (Figure 2). Similar results regarding differences in water absorption speed and total capacity among soybean cultivars were observed by Costa, Pires, Thomas, and Alberton (2002). However, these authors did not investigate the seed characteristics that may be related to behavior differences among cultivars.

The curves for the five cultivars can be separated into three groups that are not necessarily independent, but have more similar responses within the groups than between them (Figure 2). This distinction can also be assessed using the parameters of these curves (Table 3) and their medians (Figure 3).

**Table 2.** Water content and hydration speed index (HSI) in seeds of five soybean cultivars with different seed coat colors and lignin contents.

Cultivar	Water content (%)	HSI
A	10.6 a*	13.41 a
B	10.8 a	14.50 a
C	10.5 a	15.25 ab
D	10.6 a	18.65 b
E	10.7 a	18.24 b

\*Means followed by the same letter in the column do not differ by Tukey's test ( $p \leq 0.05$ ).

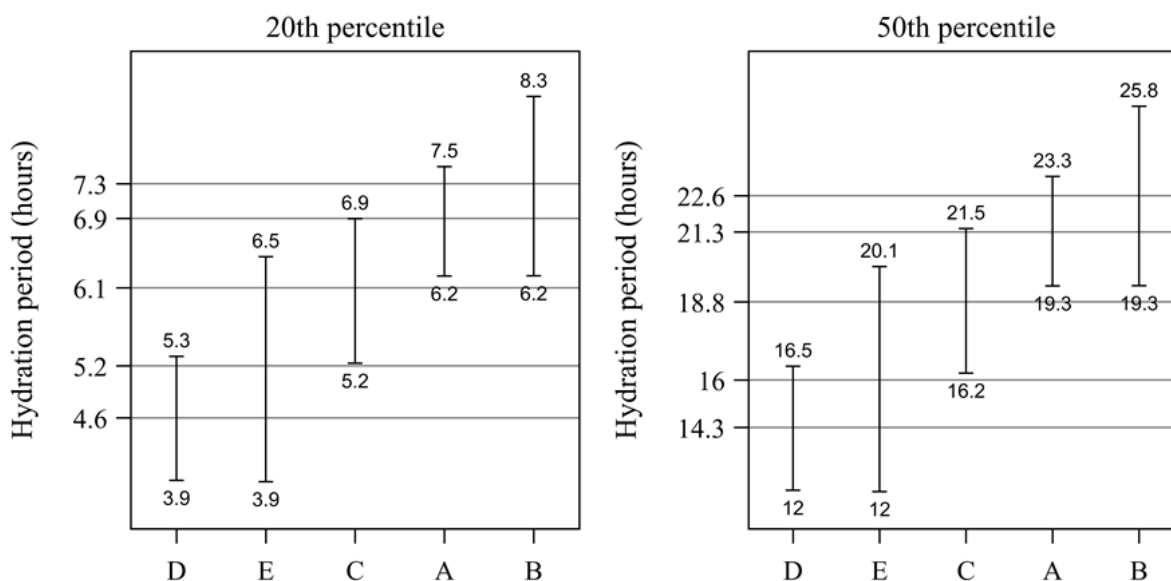


**Figure 2.** Water absorption curves of five soybean cultivars seeds submitted to 11 hydration periods (1, 2, 3, 4, 5, 6, 7, 8, 24, 32 and 48 hours).

**Table 3.** Estimates of parameters  $c$  and  $e$  and their standard errors ( $\hat{\sigma}_c$  and  $\hat{\sigma}_e$ ) for the water absorption curves of five soybean cultivars seeds, as a function of hydration periods.

Cultivar	$\hat{c}^1$	$\hat{\sigma}_c$	$\hat{e}^2$	$\hat{\sigma}_e$
A	4.2	0.4	30.8	1.9
B	6.1	0.5	32.6	2.6
C	4.7	0.5	27.2	2.3
D	5.6	0.8	20.6	1.8
E	7.2	1.2	23.1	2.9

<sup>1</sup>Lower limits of the curves (water absorption by seeds due only to the initial contact with the moisture surface). <sup>2</sup>Humidity growth steps in the curves as the imbibition time increased.



**Figure 3.** Estimates (gray lines) and 95% confidence intervals for the hydration periods required to increase the wet mass of seeds from five soybean cultivars by 20 and 50%.



Estimates of parameter  $e$ , which inversely proportionally determined the magnitude of the increase in seed water absorption, varied between  $20.6 \pm 1.8$  (cultivar D) and  $32.6 \pm 2.6$  (cultivar B) (Table 3). The significance of differences ( $d_{ij} = \hat{e}_i - \hat{e}_j$ ) between estimates of cultivars  $i$  and  $j$ , in which  $i, j \in \{A, B, C, D, E\}$  and  $i \neq j$  can be tested assuming that the statistic  $t_{c(ij)} = d_{ij}/\hat{\sigma}_{d_{ij}}$ , with  $\hat{\sigma}_{d_{ij}} = \sqrt{\hat{\sigma}^2_{\hat{e}_i} + \hat{\sigma}^2_{\hat{e}_j}}$  equal to the combined standard error of estimates  $\hat{\sigma}_{e_i}$  and  $\hat{\sigma}_{e_j}$ , can be approximated by distribution  $t$  with two degrees of freedom. Based on the results of tests of the hypothesis:  $H_0: t_c = 0$  vs  $H_1: t_c > 0$ , in which  $t_c = (e_i - e_j)/\sigma_{ij}$  is applied to these differences, it could be concluded that cultivar D presented a water absorption capacity significantly higher than those of cultivars A, B, and C; while cultivar E showed a water absorption capacity only significantly higher than those of cultivars A and B. Cultivars D and E also presented higher hydration speed index (HSI) than did cultivars A and B (Table 2). It is noteworthy that cultivars D and E are those with the lowest seed coat lignin contents (Table 1).

These results partially endorse the results shown in Figure 3, which estimates for each cultivar the hydration period necessary to increase the wet mass of seeds by 20 and 50%, and their respective confidence intervals. Considering the non-overlapping of intervals as evidence of significant difference, it could be concluded that cultivar D reached 20 or 50% the wet mass of seeds in a significantly shorter period than did cultivars A and B.

This difference in water absorption among cultivars, especially in the first hours of hydration, is possibly related to the seed coat permeability. According to McDonald Jr., Vertucci, and Roos (1988) and Marcos-Filho (2015), soybean seed coats regulate water absorption in the first hours of imbibition and, after this period, facilitates water transfer to internal parts of the seed.

Water absorption and HSI data correlated significantly and negatively with the seed coat lignin contents (Table 4). This association elucidates the results observed, since the highest water absorbed and HSI percentages were found in cultivars D and E, which had the lowest seed coat lignin contents (Table 1). Owing to lignin's hydrophobic character, it directly acts on the seed coat permeability (Zhao & Dixon, 2011), which supports the results found in this study.

Although the correlation between water absorption and lignin content was significant, it is noteworthy that the correlation coefficients ( $r$ ) were on average 0.52, which represents a moderate dependence (Mukaka, 2012). Thus, it is emphasized that in addition to the lignin content, there are other factors that interfere in the seed coat permeability and, consequently, in the water absorption capacity and rate.

In this context, the degree of permeability of seeds has also been related to the structure and compounds present in the seed coat, such as the thickness of the lacunous parenchyma (Cavariani, Toledo, Rodella, França-Neto, & Nakagawa, 2009); conformation of osteosclerids (Luan et al., 2017); presence of cuticle cracks (Ma, Cholewa, Mohamed, Peterson, & Gijzen, 2004; Ranathunge et al., 2010; Vu et al., 2014); pore number and shape (Chachalis & Smith, 2001); cutin, wax, and suberin levels (Shao, Meyer, Ma, Peterson, & Bernards, 2007; Bewley, Bradford, Hilhorst, & Nonogaki, 2013), among others.

Regarding the physiological potential of seeds, different behaviors of factors evaluated as a function of the storage environment were verified. In the dry and cold chamber, the isolated effect of the cultivar for viability and vigor and the isolated effect of the storage period for germination, viability, and vigor were verified.

For germination, during the storage of seeds in the dry and cold chamber, a reduction in the number of normal seedlings at six months of storage was observed (Table 5).

For the tetrazolium test, reductions of 4 and 9 percentage points were found for seed viability and vigor, respectively, after the storage period in the dry and cold chamber (Table 5). In this storage environment, there was an isolated effect of the cultivar, in which cultivars A and C showed higher viability and vigor values than did cultivar B.

In storage under uncontrolled temperature and RH conditions, significant interactions were observed between the cultivar and storage period for all variables related to the physiological quality of seeds. In the period prior to storage (0 months), no difference were observed among cultivars for all variables (Table 6), so differences observed after seed storage should not be attributed to their initial quality.

Regarding germination, after six months storage under uncontrolled conditions, it was found that cultivar D had the lowest germination rate (75%; Table 6), differing from the others. Thus, under the conditions of this experiment, seeds of this cultivar could not be marketed after six months of storage, as they did not present the minimum 80% germination recommended by the national standards for the marketing of seeds of this species (Brasil, 2013).

**Table 4.** Pearson's correlation coefficients (*r*) between water absorption rate results in the 11 hydration periods and hydration speed index (HSI) with the lignin content and the physiological quality variables of seeds of five soybean cultivars evaluated after six months of storage in a dry and cold chamber (CC) and uncontrolled environment (UE).

Variables	LIGNIN	GER CC <sup>1</sup>	TZ VIA CC	TZ VIG CC	GER UE	TZ VIA UE	TZ VIG UE
A1	-0.57*	-0.50*	-0.02 <sup>ns</sup>	-0.08 <sup>ns</sup>	-0.43*	-0.45*	-0.47*
A2	-0.58*	-0.47*	-0.06 <sup>ns</sup>	-0.08 <sup>ns</sup>	-0.45*	-0.47*	-0.51*
A3	-0.57*	-0.50*	-0.03 <sup>ns</sup>	-0.05 <sup>ns</sup>	-0.43*	-0.46*	-0.50*
A4	-0.51*	-0.52*	0.00 <sup>ns</sup>	0.00 <sup>ns</sup>	-0.42 <sup>ns</sup>	-0.44*	-0.48*
A5	-0.48*	-0.50*	0.02 <sup>ns</sup>	0.02 <sup>ns</sup>	-0.45*	-0.40 <sup>ns</sup>	-0.47*
A6	-0.48*	-0.46*	0.02 <sup>ns</sup>	0.03 <sup>ns</sup>	-0.42 <sup>ns</sup>	-0.39 <sup>ns</sup>	-0.45*
A7	-0.49*	-0.43*	0.02 <sup>ns</sup>	0.06 <sup>ns</sup>	-0.49*	-0.40 <sup>ns</sup>	-0.49*
A8	-0.52*	-0.50*	0.01 <sup>ns</sup>	0.05 <sup>ns</sup>	-0.47*	-0.44*	-0.50*
A24	-0.51*	-0.36 <sup>ns</sup>	-0.09 <sup>ns</sup>	0.17 <sup>ns</sup>	-0.71*	-0.47*	-0.59*
A32	-0.40 <sup>ns</sup>	-0.45*	0.02 <sup>ns</sup>	0.23 <sup>ns</sup>	-0.57*	-0.36 <sup>ns</sup>	-0.49*
A48	-0.59*	-0.45*	-0.04 <sup>ns</sup>	0.13 <sup>ns</sup>	-0.55*	-0.41 <sup>ns</sup>	-0.49*
HSI	-0.56*	-0.50*	-0.02 <sup>ns</sup>	-0.01 <sup>ns</sup>	-0.47*	-0.46*	-0.51*

<sup>ns</sup>non-significant correlation and \*significant correlation at 5% probability. <sup>1</sup>Data on the germination (GER), viability (TZ VIA), and vigor (TZ VIG) of seeds after six months of storage in a dry and cold chamber (CC) and uncontrolled environment (UE). A1, A2, ..., A48: water absorption in the period of 1, 2, ..., 48 hours of hydration. HSI: hydration speed index.

**Table 5.** Germination (GER), viability (TZ VIA), and vigor (TZ VIG) by the tetrazolium test on soybean seeds, for the isolated effect of the storage period, the average of five cultivars, and for the isolated effect of the cultivar, the average of two storage periods under dry and cold chamber conditions.

Storage period (months)	GER (%)	TZ VIA (%)	TZ VIG (%)
0	93 a	93 a	88 a
6	86 b	89 b	79 b
Cultivar	TZ VIA (%)	TZ VIG (%)	
A	94 a	87 a	
B	88 b	78 b	
C	93 a	87 a	
D	90 ab	83 ab	
E	90 ab	83 ab	

Means followed by same letter in column do not differ by Tukey's test at 5% probability.

**Table 6.** Germination, viability, and vigor of soybean seeds by the tetrazolium test, according to the cultivar and storage period under uncontrolled environment conditions.

Cultivar	Germination (%)		Amplitude*
	Storage period (months)		
	0	6	
A	92 Aa	86 Ab	6
B	93 Aa	82 Ab	11
C	95 Aa	85 Ab	10
D	91 Aa	75 Bb	16
E	95 Aa	83 Ab	12
Cultivar	TZ Viability (%)		
	0	6	
A	96 Aa	91 Ab	5
B	91 Aa	79 Cb	12
C	94 Aa	86 ABb	8
D	92 Aa	78 Cb	14
E	94 Aa	81 BCb	13
Cultivar	TZ Vigor (%)		
	0	6	
A	90 Aa	83 Ab	7
B	85 Aa	66 BCb	19
C	91 Aa	80 Ab	11
D	86 Aa	60 Cb	26
E	90 Aa	68 Bb	22

Averages followed by same letter (lower case in the row and upper case in the column) do not differ by Tukey's test at 5% probability. \*Amplitude: difference between values observed at 0 and 6 months of storage within the same cultivar.

Comparing the storage periods under uncontrolled conditions, reductions in germination values were found in all cultivars (Table 6). Corroborating these results, El-Abady, El-Emam, Seadh, and Yousof (2012)

also observed reductions in the seed germination of soybean cultivars stored for a period of six months under uncontrolled temperature and RH conditions.

With regard to the tetrazolium test, at six months of storage under uncontrolled conditions, it was observed that seeds from cultivar A showed greater viability than did cultivars B, D, and E. Cultivars A and C demonstrated higher vigor values (Table 6). Regarding the effect of the storage period, reductions in viability and vigor were found by the tetrazolium test for all cultivars. However, although storage reduced in the physiological performance of all cultivars, it is important to note that these decreases were not homogeneous among cultivars, which can be seen in the variation percentages shown in Table 6. From these values, it was observed that differences were more accentuated for vigor, with variations from 7 (cultivar A) to 26 percentage points (cultivar D), which caused a discrepancy of 19 percentage points among cultivars, which certainly differentiates them regarding their storage tolerance.

In addition, under uncontrolled conditions, it is noteworthy that amplitudes observed for cultivar A, with black seed coats and higher lignin contents, were 6 for germination and 7 percentage points for vigor (Table 6), which are smaller than the average reductions verified between month zero and month six of storage in the dry and cold chamber (Table 5). Thus, the favorable performance of this cultivar is evidenced even in conditions not so adequate for the conservation of the physiological potential of seeds.

Regarding the relationship between the water absorption rates and physiological performance of seeds after storage, a significant and negative correlation was found between these characteristics, especially under uncontrolled conditions (Table 4). In this environment, corroborating the results of this analysis, greater reductions in physiological quality, especially in vigor were observed for cultivars D and E (Table 6), which showed the highest water absorption values (Table 2; Figure 2).

This association between water absorption characteristics and storage tolerance under uncontrolled conditions is possibly related to the RH and temperature conditions present in that environment and the hygroscopic character of seeds. In this environment, the RH variation over the storage period ranged from 52 to 84% (Figure 1), which changed the seed metabolism and contributed to their deterioration process in all cultivars. However, cultivars that absorbed larger amounts of water had this process intensified due to the lower resistance to RH fluctuations, resulting in greater reductions in physiological performance after the storage period.

Under uncontrolled storage condition, it was possible to observe differences in storage tolerance among cultivars that absorbed smaller amounts of water, with emphasis on the greater tolerance levels of cultivars A and C (Table 6).

Thus, it appears that water absorption and seed storage tolerance are not associated with a specific factor, but with a set of characteristics that may be related to the structure and chemical composition of the seed coat or embryo. Thus, further studies are necessary to identify and understand the other factors related to conserving the physiological potential of seeds.

## Conclusion

The lignin content in the seed coat is negatively correlated with the water absorption parameters of the seeds. Soybean cultivars have different storage tolerance levels.

Seeds that absorb larger amounts of water have lower tolerance to deterioration due to the temperature and relative humidity fluctuations that occur during storage under uncontrolled conditions, negatively affecting the physiological potential of the seeds.

## Acknowledgements

To the *Coordenação de Aperfeiçoamento de Pessoal de Nível Superior* – Capes, for granting the scholarship to the first author. To the *Universidade Estadual de Londrina* and the *Empresa Brasileira de Pesquisa Agropecuária* (Embrapa Soja) for the structure and financial support to develop this work.

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