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RESEARCH NOTE

Harvest delay, storage and physiological quality of soybean seeds[1]

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ABSTRACT – Soybean is one of the leading commodities in Brazilian agribusiness. Its cultivation is widespread in different seed-producing regions of the country, where it generates income and local development. In this context, the soybean seed is a fundamental input, as its quality strongly influences the success of the crop. However, the period after the achievement of physiological maturity is critical for the maintenance of seed quality. For that reason, this study aimed at evaluating the effect of harvest delay on both the initial and final qualities of soybean seeds. The research consisted of four harvest times, in which the initial quality was assessed through tests of first germination count, germination, accelerated aging and tetrazolium. After 120 days of storage, the germination and accelerated aging tests were once again conducted. The experiment complied with a completely randomized block design with eight replications. The rainfall was monitored during the pre-harvest phase. All variables experienced negative impacts due to the delay in harvesting. Also, the seeds suffered more damage as the delay progressed, and the variables germination and seed vigor decreased after the storage period.

Index terms: deterioration, storage, damage by moisture, *Glycine max*.

Atraso na colheita, armazenamento e qualidade fisiológica de sementes de soja

RESUMO – A cultura da soja está difundida nas diferentes regiões produtoras de grãos do Brasil, sendo uma das principais *commodities* do agronegócio nacional, gerando fonte de renda e desenvolvimento em diferentes regiões. Neste contexto, a semente de soja é um dos principais insumos da lavoura, apresentando forte relação entre sua qualidade e o sucesso do cultivo. Entretanto, o período após a maturidade fisiológica é crítico para a manutenção da qualidade fisiológica das sementes. Neste trabalho, objetivou-se avaliar o efeito do atraso de colheita nas qualidades inicial e final de sementes de soja. O experimento consistiu em quatro períodos de colheita, nos quais foi avaliada a qualidade inicial através de ensaios de primeira contagem de germinação, germinação, envelhecimento acelerado e pelo teste de tetrazólio. Após 120 dias de armazenamento, foram conduzidos testes de germinação e envelhecimento acelerado. Foi utilizado delineamento em blocos ao acaso com oito repetições, e as chuvas na região foram monitoradas durante a fase pré-colheita. Observou-se influência negativa em todas as variáveis estudadas, ocasionadas pelo atraso na colheita. Também foi verificado um aumento na incidência de danos por umidade devido ao retardamento da colheita. Por último, notou-se um comportamento decrescente nas variáveis germinação e vigor, após período de armazenamento.

Termos para indexação: *Glycine max* L., retardamento de colheita, deterioração, vigor, dano por umidade.

Introduction

Soybean is one of the central commodities in contemporaneous agribusiness. In Brazil, it stands out among other cultures, with a producing area of about 35 million hectares in the 2018/ 2019 crop – approximately 60% of the total area used for summer cultures (Conab, 2019). On that account, the utilization of high quality seeds is important for

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the success of the crops. The high vigor of soybean seeds has a direct impact on yield components (number of pods per plant), which can lead to a productivity up to 15% higher than that obtained from low-vigor seeds (Tavares et al., 2013; Silva et al., 2016; Bagateli et al., 2019).

In order to have their high quality attested, seeds ought to present certain physiological and sanitary conditions, including superior levels of vigor and germination, physical and varietal purities, and they must also be free from any pathogens. These factors respond for the performance of seeds in the field, favoring the establishment of the plant populations required by the cultivar, a fundamental aspect for accomplishing the desired high productivity (França-Neto et al., 2010).

The seed production process has to be adjusted, so that to result in high-quality seeds that meet the necessities of farmers. It is worth remarking that pre-harvest procedures are highly critical because of the quantitative, qualitative, and sanitary susceptibility of the seeds to adverse climate conditions, such as rainfall and variations in temperature and relative air humidity (Daltro et al., 2010).

Delaying the harvest (once the physiological maturity has been achieved) negatively affects the quality of seeds (Pelúzio et al., 2008; Diniz et al., 2013). Conversely, collecting soybeans at their physiological maturation may not be the best alternative, as the high moisture content of the seeds could cause them to suffer latent injuries during the mechanical harvest. Thus, it is necessary to wait for the natural moisture decrease.

Even in regions with ideal climate conditions for soybean cultivation, rainfall and variations in relative humidity and temperature within the span between the achievement of physiological maturity and the harvest might increase the percentage of damages due to moisture, characterized by integument wrinkling. This condition intensifies seed deterioration and facilitates the penetration of pathogens, as the inner tissues become more exposed. Therefore, it potentially reduces overall productivity (Marcandalli et al., 2011; Pádua et al., 2014; Tsukahara et al., 2016).

Then, the present work aimed at evaluating the deleterious influence of delaying harvest on the physiological quality of soybean seeds, assessed initially and after storage.

Material and Methods

The study was carried out during the 2017/ 2018 crop, in a seed-producing field of the cultivar NS 5959IPRO. This variety has an indeterminate growth habit and a 5.9 soybean maturity group, in the Brazilian state of *Rio Grande do Sul*.

The experiments covered five harvest times. The first one corresponded to the moment the plants reached the R7.8 stage – a period suitable for obtaining high-quality seeds (Fehr and Caviness, 1977). The others took place 7, 14, 21 and 28 after the stage R7.8 had been reached. In each case, the oven method at 105 ± 3 °C was used (Brasil, 2009), and moisture contents of 19.4%, 16.8%, 15.2%, 21.7% and 13.1% were verified from matching the first to fifth harvest time, respectively.

Data gathered from the National Institute of Meteorology website allowed to classify the local climate as Cfa, according to Köppen's classification. The precipitation levels during the pre-harvest period are showed in Figure 1.

The plants were harvested within two linear meters of each parcel every seven days to obtain the five harvest times.

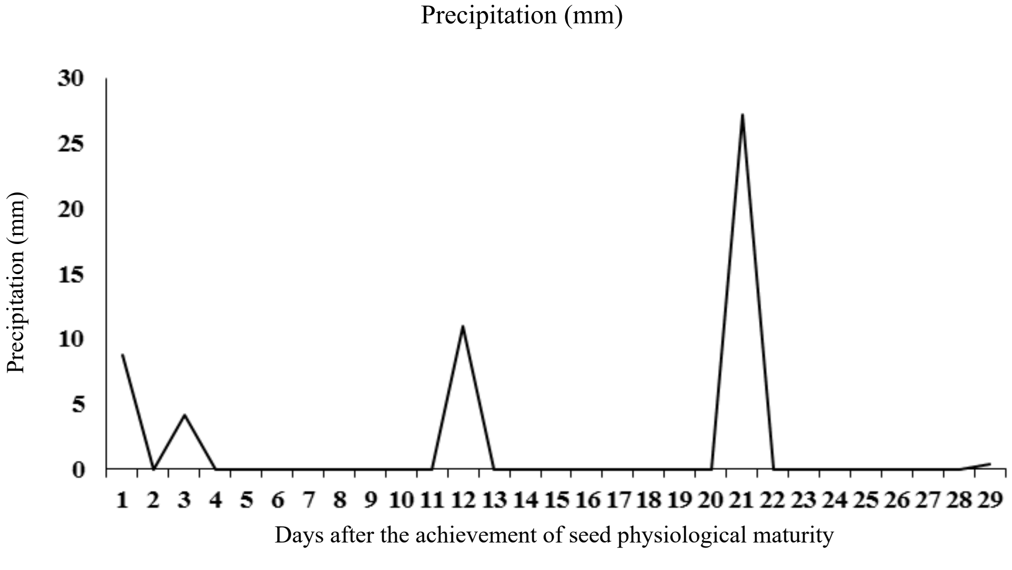

Figure 1. Precipitation (mm) during the field storage (adapted from INMET, 2019)

After each harvest, the plants were manually thrashed, and the seeds were dried to 12% moisture. Then, they were packed in paper bags and stored in a controlled environment at 15 ºC and 70% relative humidity from April to August 2018.

The physiological quality of the seeds was evaluated at two distinct times: at the beginning of storage (after the last harvest time), and 120 days later. In each case, the following tests were performed:

Germination: it followed the methodology recommended by Rules for Seed Testing (Brasil, 2009). Four replications of 50 seeds were sown in towel papers, which were moistened at the ratio of 2.5 times the dry paper weight. Next, the paper rolls were placed inside a germinator set at a constant temperature of 25 ºC. The percentage of normal seedlings emerged was accounted on the eighth day after sowing.

First germination count: it was carried out together with the germination test. The assessment was done after five days of sowing, considering the percentage of normal seedlings emerged.

Accelerated aging: plastic gerboxes were used as individual compartments, each one filled with 40 mL of distilled water. The seeds were placed inside them, in a single layer, and then kept at 41 ºC for 48 hours (Marcos-Filho, 2015). After that, they were subjected to germination test, and the percentage of normal seedlings obtained on the fifth day was calculated (Brasil, 2009).

Tetrazolium test: it was performed according to the methodology proposed by França-Neto et al. (1999). The percentages of viability, vigor, and moisture-related damages in the seeds were calculated.

The precipitation data occurred during the experiment were obtained from an automated station of the National Institute of Meteorology (INMET, 2019). The experiments complied with a randomized block design with eight replications. The resulting data underwent variance analysis and, once significance was noticed, they were subjected to regression analysis. The software WinStat version 1.0 (Machado and Conceição, 2003) handled all the statistical procedures.

Results and Discussion

The variables showed significant differences among the harvest times. In general, harvest delay had an impact on seed quality, both in the beginning, as well as after 120 days of storage.

According to Figure 2, a downward linear trend for first germination count (FGC) was verified, with a determination coefficient of 88.89. Right after the physiological maturity had been reached, the first germination count was equal to 85%. This value decreased by five percentage points (pp), fourteen days later, and by 20 pp, after 21 days. Similar behavior was observed in the variable germination (Figure 2), which also showed a linear decrease with a high determination coefficient. At the first harvest time, seed germination scored 85%, even achieving 90% after seven days in the R7.8 stage. Nevertheless, fourteen days afterward, there was a 10-pp drop. It is worth

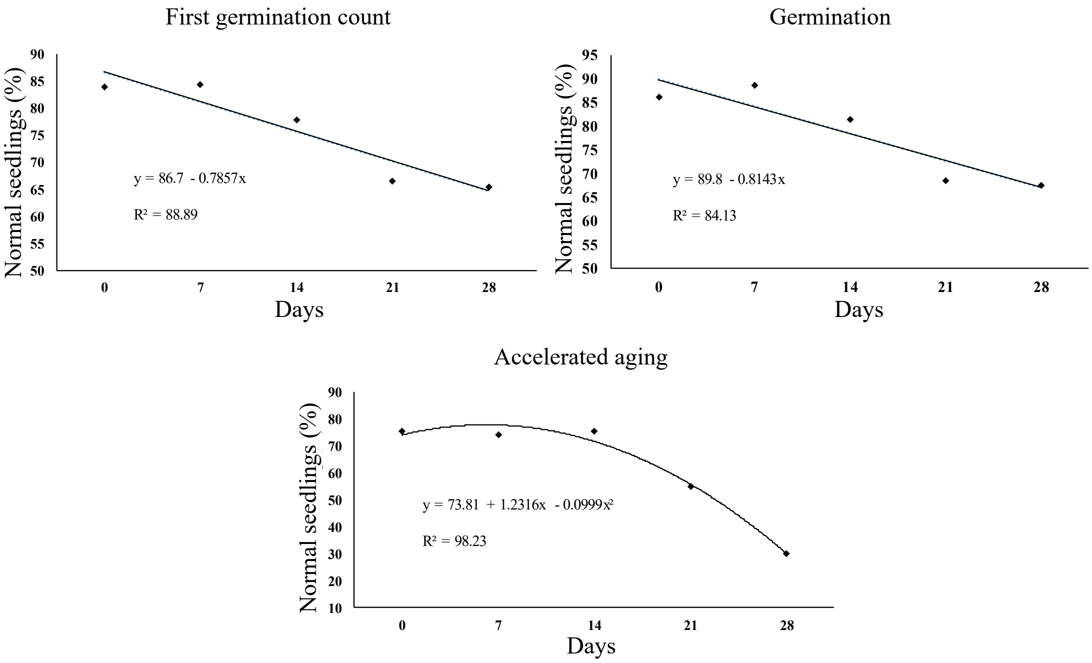

Figure 2. Initial quality of soybean seeds (assessed by first germination count, germination and accelerated aging, in %) at five harvest times (0, 7, 14, 21 and 28 days after reaching physiological maturity).

noticing that delaying the harvest for 21 or 28 days produced seeds with germination rates below 70%.

In the period between the physiological maturity and harvest, soybean seeds are openly exposed to adverse weather conditions. Thus, they are likely to suffer moisture-related damages, especially in regions where the climate gets too hot and dry during maturation. Variations and unevenness within the plant population throughout this phase are the main causes for harvest delay – and the longer the seeds remain in the field, the more severe their impairments (França-Neto et al., 2005).

For that reason, it is possible to affirm that the storage conditions (even the pre-harvest ones) influenced the decline of seed physiological quality. This can be justified by the fact that seeds are constituted by hygroscopic material and have intense metabolic activity. Thus, the joint effect of high moisture content in the seeds and hot temperatures intensifies the respiration processes, thus consuming seed reserves and, consequently, reducing their quality (Marcos-Filho, 2015). It is important to remark that variations in the water content of seeds might be related to the permeability and amount of lignin in the integument. So, genotypes with less lignin and higher permeability tend to be more sensitive to damages due to moisture (Huth et al., 2016).

The initial quality of the seeds is of great importance for their storage. To obtain superior quality seeds, it is advisable to perform an early harvest, followed by the drying operation of the seeds, so as to attenuate the influence of abiotic environmental factors over their quality. However, the high moisture content observed during the physiological maturity of soybean makes the mechanization of harvest unfeasible, as it could cause significant seed damage. Therefore, the best alternative is to harvest the seeds at around 18% moisture level (Peske et al., 2012).

In this sense, the vigor of seeds is closely related to the maturation environment, as humidity and temperature fluctuations might have deleterious effects upon this attribute. Such conditions accelerate respiration and consume energy that otherwise would be vital for seedling development. Besides, unfavorable ambient factors could trigger the formation of toxic compounds, which tend to buildup and cause degradation of plasmatic membranes of the seeds, therefore reducing their vigor (Aumonde et al., 2017).

Figure 2 contains information on the initial vigor, as assessed by the accelerated aging test. In this case, the curve displayed a quadratic behavior with a high coefficient of determination. Prior to fourteen days in the R7.8 stage, there was a tendency to stabilization, with vigor values higher than 70%. However, as the harvest delay progressed to 21 days, seed vigor diminished by some 15% and, eventually, it scored less than 40%, after field storage for 28 days.

According to the results of the tetrazolium test (Figure 3), the field storage duration influenced the performance of the variables analyzed. Seed vigor showed a cubic behavior, with values above 90% for harvest times inferior to fourteen days

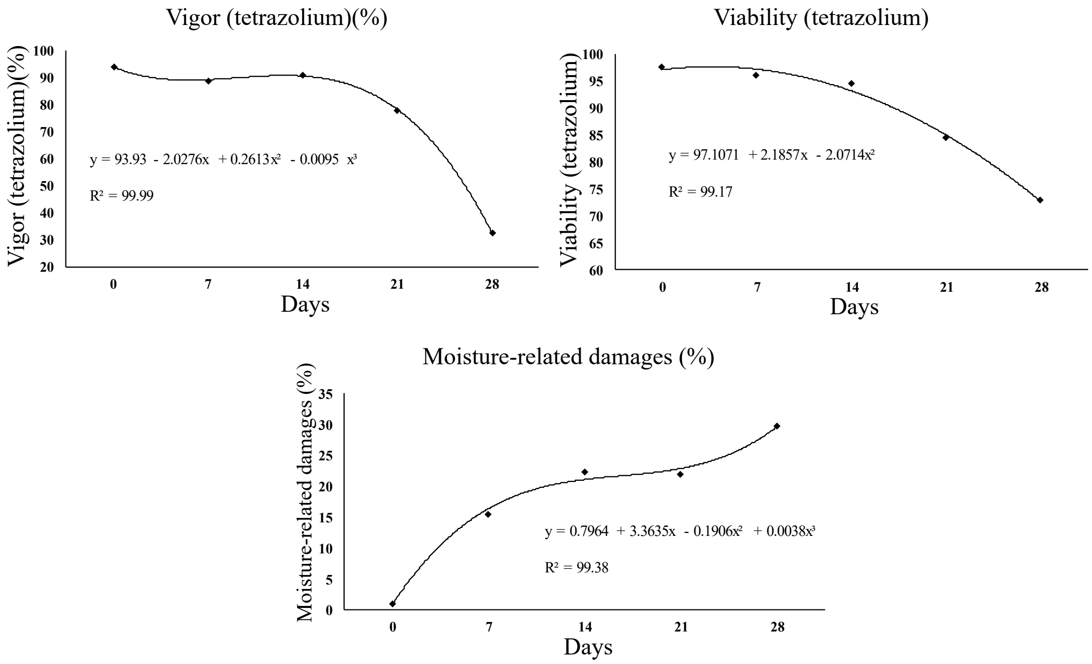

Figure 3. Evaluation of the initial quality of soybean seeds (assessed via the tetrazolium test, in %) at five harvest times (0, 7, 14, 21 and 28 days after reaching physiological maturity).

in the R7.8 stage. After this period, nevertheless, this variable decreased significantly, reaching approximately 75% and 30%, after 21 and 28 days of harvest delay, respectively.

In this context, Diniz et al. (2013) studied the effect of three harvest-delay times on eight soybean cultivars. They concluded that the field emergence of seedlings from seeds harvested after 30 days was inferior to that of seeds picked at 0 or 15 days after they had reached maturity. Such an outcome supports the findings of the present work, as the harvest delay negatively affected the physiological quality of the seeds. In general, field conditions are not ideal for seed storage, especially those rich in both oils and proteins, such as soybeans. When stored under high humidity conditions, they are propense to lipid peroxidation, which is likely to reduce membrane selectivity, thus leading to the excessive leakage of electrolytes.

The viability evaluated by the tetrazolium test (Figure 3) showed a quadratic behavior. This variable scored above 90% when the field storage lasted up to fourteen days. By the 21st day (fourth harvest time), a sharp decrease in seed viability was noticed, with values inferior to 85%. The lowest performance, however, was observed after 28 days (fifth harvest time), when this variable level stayed under 75%. The deterioration tends to intensify as the storage progresses, generally due to psychrometric conditions related to field storage, either in bulk or in packages.

The curve of moisture damages evaluated by the tetrazolium test (Figure 3) evidenced a cubic behavior. This factor (measured at the R7.8 stage) manifested the lowest values at the first harvest time, but it progressively increased as the delay extended. Ultimately, by the 28th day, about 30% of the seeds exhibited some damage caused by moisture.

Figure 1 presents the local rain incidence during the days the seeds remained stored in the field. Higher indices were observed 1, 12 and 21 days after the stage R7.8 had been reached. However, the effect of precipitation over the physiological quality of soybean seeds is probably associated with the volume and timing, as the rainfall around the beginning of the R7.8 stage did not impair seed quality considerably – although it did increase the damage due to moisture. This fact can be explained by the high water content of the seeds when near the maturity point, which could have acted by softening the water variation within the seed. This likely reduced the intensity of the damages and, consequently, the loss of quality.

The precipitation on the 12th and 21st days after the R7.8 stage had been established had a strong influence on seed quality. In Figures 1, 2 and 3, it became evident that, specifically after these rainfalls, there was a severe decline in the variables linked to physiological quality, remarkably in vigor (measured via tetrazolium) and accelerated aging. Conversely, when using artificial rainfall, Castro et al. (2016) did not find any influence of this procedure on the quality of seeds. They point out, however, that such an outcome could be justified by the fact that the artificial rain might not have been applied at volume or duration intense enough to damage the seeds.

In a study on the behavior of fully mature soybean seeds under field conditions, Tsukahara et al. (2016) verified that the seed moisture is directly impacted by the number of days with precipitation levels equal to or above 3 mm, within the span between the beginning of the R8.2 stage and the harvest. They also concluded that the most substantial productivity losses happen in environments with high rainfall frequency, hot temperatures, and intense sunlight incidence.

Moisture damages are common in soybean seeds due to variations in the water content caused by rainfalls or oscillations in the relative air humidity. This phenomenon is explained by the lack of an integument layer composed by hourglass-shaped hypodermic cells, in the region opposite to the hilum. These cells could attenuate the expansions and contractions induced by moisture changes (Marcos-Filho, 2015).

Genotype and environmental factors operating throughout the development of the crop, and during formation, harvest, processing, and storage of the seeds have a decisive role in their storage potential. Soybean seeds subjected to adverse circumstances in any of these phases are prone to experience cytological, physiological and biochemical alterations, which might curb the quality of the final lots. The degree of each of these deficiencies is unfixed, depending on genetic features intrinsic to each cultivar (Gris et al., 2010).

Delaying harvest had a significant negative consequence for the storage potential of soybean seeds (Figure 4). The longer they stayed in the field, the lower their physiological quality after 120 days of storage. According to Peske et al. (2012), among many other aspects acting upon the storage potential, the initial quality has prominence, and it is highly compromised when the harvest is delayed. Efficient storage is only possible by associating the use of high-vigor seeds with the maintenance of an adequate environment. These conditions favor seed longevity and the preservation of their physiological quality.

The seed germination had a linear decrease (Figure 4). Soybean seeds harvested at the beginning of the R7.8 stage (day zero) exhibited germination above 90%. Nevertheless, 21 days afterward, those values dropped to about 80%, evidencing an increase by some 15 pp in the abnormal seedlings and dead seeds due to harvest delay.

As for the vigor assessed by accelerated aging (Figure 4), there was a maximum point, in which the most vigorous seeds were identified, corresponding to the first harvest time (at physiological maturity). After that, the number of emerged seedlings plateaued from the 7th to the 21st day, implying that

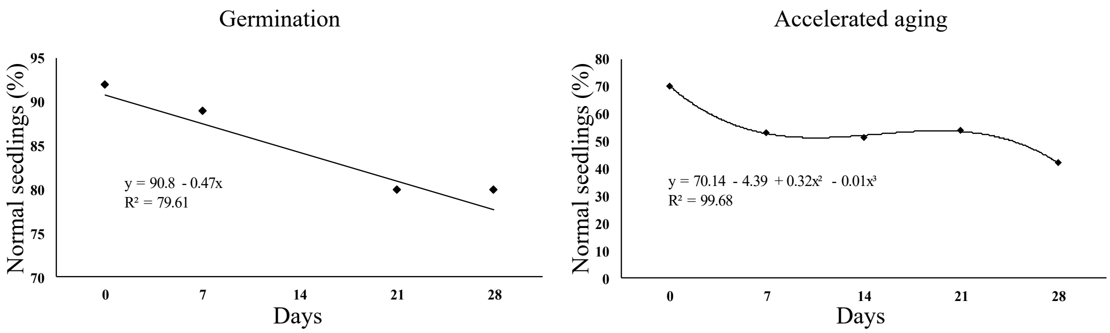

Figure 4. Physiological quality of soybean seeds stored by 120 days (assessed via germination and accelerated aging tests, in %), considering five harvest times (0, 7, 14, 21 and 28 days after reaching physiological maturity).

no significant difference was noticed by delaying harvest for these periods – although the variable scored around 50%, 20 pp below the results found when seeds were collected the closest to the R7.8 stage. Eventually, the minimum point verified at the harvest on the 28th day, with the vigor reaching around 40%, which is 30 pp less than what was obtained when the harvest was carried out at the best timing.

By contrasting the Figures 2 and 4, it became possible to verify some increment of both germination and accelerated aging after storage, which coincided with the longest harvest delays. The presence of damaging field fungi partly explains this result. The storage environment, thus not negatively affecting the testing, however, attenuated their pathogenicity. A similar result was found by Galli et al. (2007), who stored fungi-contaminated lots of two soybean cultivars in a cold chamber for six months. On that occasion, they also showed a lower incidence of the most common field fungi.

As verified in the present work, delaying the harvest has an unfavorable impact on the physiological quality of soybean seeds at the earliest stages, and it only tends to intensify as the storage progresses. This fact could be due to the higher metabolic activity of the tissues made vulnerable by moisture-related injuries. As a consequence, there is a reduction in both vigor and germination of seeds that had undergone long periods of field storage. Since it is clear that the water content of the seeds and the temperature throughout storage influence seed metabolism, an unwanted development might be averted by keeping the seeds in a place with cold temperature and low relative humidity (Peske et al., 2012; Marcos-Filho, 2015).

Figures 3 and 4 elucidate the close relation between the moisture damages and the accelerated aging after storage. In both cases, there was a trend to stabilization between the 7th and 21st days after the physiological maturity had been reached, indicating that the intensity of injuries had an isolated deleterious influence upon accelerated aging. Nonetheless, these injuries still represent a critical limiting aspect for the production of high vigor seeds.

It is worth remarking that environmental circumstances are directly linked to the incidence of damages due to moisture. That means that factors such as relative air humidity, temperature above 25 ºC, and rainfall during the pre-harvest phase may affect either the gain or loss of moisture by the seeds. These conditions cause damage to their integument and, consequently, decrease the quality and overall productivity (Marcandalli et al., 2011; Pádua et al., 2014). So, the deterioration triggered by harvest delays is a continuous and irreversible process, which bears a straight relation to field climatic conditions during the span between the beginning of maturity and the harvest.

Despite the clear correlation between environmental field conditions and soybean seed deterioration, the genetic component (that is, the intrinsic characteristics of each cultivar) must be considered. Lima et al. (2007) studied the delay of harvest in different cultivars, and they concluded that the genotypes that did not have the enzyme lipoxygenase (LOX+Linn) performed better in almost all quality tests. This fact should be better understood when comparing LOX + Linn, which holds one of the main substrates for lipoxygenase enzyme activity and, consequently, progress in the seed deterioration process.

Castro et al. (2016) evaluated soybean cultivars with different lignin contents, which were subjected to harvest delay under artificial rainfall. They verified a strict relation between the levels of this compound in seeds and the damages due to moisture. In this case, the variety with the most lignin in its composition displayed the lowest indices of moisture-related impairment and also the best physiological quality throughout storage. Likewise, Huth et al. (2016) proved that cultivars rich in lignin usually suffer less moisture-related damage. They additionally noticed that the varieties experienced different oxidative stress levels once they were damage by moisture.

Thereby, the environmental conditions played a decisive role during the period between the achievement of maturity and crop harvest. Such fact is evidenced by the sharp decrease in variables linked to physiological quality because of harvest postponement – that is, due to field deterioration, caused by rainfall after seed maturation. It is worth emphasizing that the intensity of this depreciation must be related to genetic characteristics. So, each cultivar is likely to respond in a particular way to harvest delay, even when under similar climatic conditions.

Even so, it is advisable to perform the harvest the closest to the maturation point as possible. In that condition, pods have enough moisture to undergo threshing, with minimum damage to the seeds. That way, it is possible to reduce deterioration in the field and produce seeds of superior quality.

Conclusions

Delaying harvest reduces the initial physiological quality of soybean seeds.

Moisture-related damages have a significant effect on the physiological quality of soybean seeds that have undergone harvest delay.

Delaying harvest (once the stage R7.8 has been achieved) results in physiological quality loss in soybean seeds after storage.

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