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Review

Analysis of seed quality: A nonstop evolving activity

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The enhancement of the production processes and increasing demand for high-quality seeds, has led the seed quality control companies to technically improve their activities. Due to this demand, we searched to improve tests related to physiological seed quality, aiming to obtain analysis results that are more consistent with the findings related to field conditions, providing information about the possibilities of the seeds in producing vigorous and representative seedlings. The studies related to the tests for seed vigor evaluation are noteworthy, as they are auxiliaries to the germination test, which has its limitations, especially when regarded to the differentiation of the lots and a relative delaying in obtaining reliable results in short time periods. Therefore, the aim of this review was to describe the available information about seed quality analysis, reporting the possible incorporation of a new method for this analysis.

Key words: Rapid tests, viability, vigor.

INTRODUCTION

Evaluating the physiological potential of seeds is the main component in a program of quality control, considering that it supplies information that identifies and fixes problems during the production process, besides estimating seed performances in the field. When talking about tests of physiological seed quality for sowing and marketing purposes, we can focus on the germination test, while being conducted under ideal and artificial conditions, allows for obtaining the maximum germination percentage. However, this test has limitations, mainly, with respect to the differentiation of lots and the relative delay in obtaining results, which, over the years has stimulated the development of reliable and fast vigor tests speeding up decision making (Bertolin et al., 2011). Thus, the identification of vigor tests that supply a safe margin in regards to seed behavior in the field, have been a tireless search and a necessity, since the adverse environmental conditions impose uniformity between the germination test and the field results, thus establishing the need to identify tests that provide equivalent conditions to germination in the field, combined with all the adversities that may affect the performance of a

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cultivar. Therefore, during the last decades, the interest to develop appropriate techniques for better information about crops has been a central topic of research (Dell'aquila, 2009).

The use of seeds with high physiological potential is an important aspect that should be considered for increased productivity and, therefore, the quality control of seeds tends to be increasingly efficient, including tests that assess this aspect quickly, allowing accurate differentiation between seed lots (Fessel et al., 2010). Thus, this review aimed to describe the main information existing to date on the analysis of the physiological quality of seeds, as well as to report the possible incorporation of a new method for analysis of seed vigor.

QUALITY AND PHYSIOLOGICAL POTENTIAL OF SEEDS

The history of agriculture demonstrates that the first contacts between man and seed physiology were established from the moment the possibility of its use was discovered for propagating plants in the century LXXX a.C. (Krzyzanowski and Vieira, 1999). In this situation, besides causing profound positive changes in the habits of life, the beginning of the use of seeds for the establishment of cultures aimed at producing foods also became a source of concern (Peres, 2010). With the domestication of plant species, new challenges have emerged with the need to determine the most favorable seasons for sowing (Marcos Filho, 2005). For being an indispensable ingredient, due to its role in the agricultural chain and in human history, the seed both socially and economically, contributes to the quantitative and qualitative increase in productivity, noting that the use of high quality seeds is an important factor for the success of any culture (Gaspar and Nakagawa, 2002). The procedures adopted in the control of seed quality programs are based both on previous recommendations from research or practical experience, just like the survey data which allows the detection of problems, and the proposition of adequate solutions (Krzyzanowski and Vieira, 1999). Seed quality is basically determined by their physiological potential, which gathers information on viability and vigor of a seed lot, with the potential term translated as virtuality or set of skills to perform tasks and produce results (Marcos Filho, 2005). The test commonly used to determine the viability of the seed is germination, whose main aim is to obtain information about the value of seeds for sowing, as well as the comparison of quality of different lots (Lima et al., 2006).

Its conduction occurs under optimal conditions in order to provide maximum germination of the sample analyzed. These conditions refer to the availability of water, aeration and temperature (Marcos Filho et al., 1987; Brasil, 2009). The germination test is aimed towards at least two aspects: providing information on the potential of a lot to germinate under favorable environmental conditions and to provide high degree of standardization, with ample opportunity for repetition of the results with reasonable levels of tolerance, as long as the instructions established are followed (Marcos Filho, 1999; Brasil, 2009).

However, the germination test might overestimate the physiological potential of the seeds for not evaluating the physiological, biochemical, physical and cytological changes related to the process of deterioration, not allowing to differentiate in the field and in storing seed lots in regards to vigor (Abrantes et al., 2010). For this reason, research has performed studies to develop methods that allow the evaluation of vigor of the seed (Kikuti et al., 1999; Ávila et al., 2007; Ohlson et al. 2010). The vigor of a seed comprehends the set of properties, which determines the ability of emergence and the fast development of normal seedlings under a wide range of environmental conditions (Baalbaki et al., 2009). Thus, its basic aim is to identify properly which lots have greater potential to survive and generate good productivity under field conditions (Marcos Filho, 2005).

In the U.S.A. and in Canada, from 1976 to 1990, there was a significant increase in the use of vigor tests in laboratories of seed analysis. During this time the test of electrical conductivity was not mentioned as a vigor test although it began being mentioned as such in 1982 (Tekrony, 1983; Ferguson, 1993). This test, along with the accelerated aging and cold tests, were the aim of study by the committee of vigor of the Association of Official Seed Analysts, Inc (AOSA), which from 1983 to 1991 were considered the three most promising vigor tests (McDonald, 1993).

Searching the use of existing laboratory tests between members of ISTA (International Seed Testing Association), Hampton et al. (1992), recommended the test of electrical conductivity to evaluate the effect of pea seeds in Europe and New Zealand. With this the electrical conductivity and the accelerated aging tests became the only two recommended by the committee of vigor by ISTA (Hampton and Tekrony, 1995).

In Brazil, Krzyzanowski et al. (1991), assessing the situation of the use of vigor tests as routine among laboratory of seed analysis, concluded that, although fundamental, these tests have yet to evolve in a way that they can participate effectively in quality control programs in seed industries. In the particular case of the electrical conductivity test, its use is still very restricted to certain situations, especially to those directly related to research (Peres, 2010).

The opening of new agricultural frontiers and increased seed production in Brazil in recent years, has led seed companies to seek technical improvement of its activities primarily aimed at increasing productivity associated with an increase in product quality. Thus, both the ISTA and AOSA adopt procedures to evaluate the most adequate methodology for the inclusion in the Rules for Seed

Analysis, through benchmarking tests performed in different laboratories, under the coordination of specific committees, who evaluate purity, germination, and moisture content among other features.

To this end, a number of samples are sent to participating laboratories, accompanied by instructions that must be followed by analysts. Once in possession of the results, the Coordination Committee interprets them, verifying compatibility among the laboratories, detecting problems, diagnosing the situation and programing new test steps, until the level of standardization is satisfactory and allows the recommendation of the methodology. Therefore, it is possible to notice the occurrence of a careful process of standardization and quality control, which have the function of equating and determining the best method for monitoring vital seed (Krzyzanowski and Vieira, 1999).

The main purpose of seed analysis is to determine the quality of a seed lot and, consequently, its value for sowing (Brasil, 2009). The analysis is characterized by a detailed and critical test of a sample, with the aim of analyzing its quality in order for it to be used in research works, as well as in identifying causes and problems of quality.

In the United States (USA), various seed producing companies of big cultures have used vigor tests for the identification of lots that do not reach internal standards of quality, classification (ranking) of lots in different levels of physiological quality, evaluation of potential for the formation of regulator stocks (carry over), decision making in regards to commercialization, looking to commercialize, firstly, lots which attend to germination standards, but at the same time show lower vigor, and moreover, supply of information about the physiological quality of the lots to consumers (Frigeri, 2007).

Thus, the seed analysis is an important tool in quality control, particularly from the end of the maturation period, when the seeds reach physiological maturity. Also, there is an international consensus among researchers, technologists and producers of seeds about the importance of the determination of seed vigor and the necessity for evaluation. Information about vigor is still important for seeds of a higher commercial value, such as vegetables, which may have been pelleted (covered by films) and pre-conditioned physiologically, like in other countries. Moreover, for presenting a lesser quantity of stored reserves, they have a higher propensity to reduced vigor after physiological maturity (Kikuti et al., 2012). The cultivation of these species is done intensively and must be established using seeds that germinate rapidly and uniformly, and therefore with higher physiological quality. The demand for methods that are satisfactory in predicting seed quality, through the force of vegetable seeds serves as methodological updates from other cultures such as, hybrid corn seeds generating technological progress in search of quality in the determination of vigor tests (Peres, 2010).

Within this context, vigor tests are useful in the programs of seed production for the evaluation of physiological potential of lots with similar germination, allowing to differentiate lots based on potential of seedling emergence in the field, evaluation of storage potential, degree of deterioration, quality control post-maturity, and physiological quality, serving as a tool to aid in methods of selection during the improvement of plants, as well as to allow the effects of mechanical and thermal injuries, treatment with fungicides and other adverse factors pre and post harvest (Marcos Filho, 1999).

Often, seed lots with similar germination percentage can show different responses in the field and/or storage (Frigeri, 2007), being that the loss of germinating potential is an important indicator in loss of quality; however, it is the ultimate event of this process. Thus, the use of vigor tests is of extreme importance for monitoring the quality of seeds from maturity, because the fall of vigor precedes loss of viability (Dias and Marcos Filho, 1995).

VIGOR TESTS OF SEEDS

Although vigor tests possess different technologies, they are intended to detect significant distinctions in the physiological potential of seed lots with similar germination among themselves (Lima et al., 2006; Dutra and Medeiros Filho, 2008), classifying them into different levels of vigor, especially proportional to the response of emergence of seedlings in the field (Marcos Filho, 1999). Thus, the selection of vigor tests must meet specific goals, making it important to identify the characteristics evaluated by tests and its relation to the responses of seeds by specific situations such as performance after drying, storage potential, response to mechanical damage and weather conditions.

According to McDonald (1993), vigor tests can be classified as physical, physiological, biochemical and of stress resistance. The physical tests assess morphological or physical characteristics of the seed, which may be associated with force, such as size, density, and color of seeds X-ray tests. Tests characterized as physiological are based on specific physiological activities which have their manifestation based on vigor, such as first germination count, index of speed of germination or emergence of seedlings. However, biochemical tests evaluate changes in metabolism related to the vigor of seeds; among these are the tests of tetrazolium and of electrical conductivity. Finally, stress resistance tests, which analyze the behavior of seeds when exposed to unfavorable environmental conditions, with emphasis for the accelerated aging tests, controlled deterioration, cold, germination at low temperature and submersion in water (Marcos Filho, 2005)

Seed vigor, according to what was defined by the

International Seed Testing Association (Ista, 1995), is the degree index of physiological deterioration and/or mechanical integrity of a seed lot of high germination, representing a broad ability to establish in the environment. This vigor definition is similar to the one formulated by the Association of Official Seed Analysts (Aosa, 1983).

Vigor tests contribute in detecting these information and, consequently, are useful for decision making on the destination of a seed lot. Among these tests of vigor available, it is worth to emphasize the test of electrical conductivity, which is a fast and objective test of vigor, which can be conducted easily by various laboratories of seed analysis with minimal spending on equipment and training of employees (Marcos Filho et al., 2009).

The results of the vigor tests are comparative, since it is not possible to quantify them since all the characteristics are not measurable. In fact, the results of 60% of normal seedlings in the accelerated aging tests, of cold, first count of germination, among others, mean nothing if not compared with what was obtained for another sample of the same species and cultivar. Thus, expressions like 70% vigor are incorrect and should not be used (Ista, 1999). The impossibility of quantifying vigor generates difficulty both for the understanding of its meaning, and for comparing information obtained in different tests. With this, research has sought to translate and establish indexes that contribute to interpretation and utilization of results obtained regarding the seed vigor (Peres, 2010).

The evaluation of the physiological potential of seeds is fundamental as a base for the processes of production, distribution and commercialization of seed lots. Thus, production companies and laboratories of seed analysis should use tests that provide reproducible results that are also reliable and that indicate with certainty the quality of a seed lot, mainly in respect to vigor (Frigeri, 2007). It is noteworthy that a seed lot is made of the same species from a defined, identified and homogenous quantity of seeds with similar physical and physiological attributes. However, lots, even from the same production area, may have imperfect homogeneity in terms of germination, which is related to variations in topography and soil fertility, or even, can be influenced by crop interspersed with rainy days (Peske et al., 2006).

Vigor analyses allow racking of lots, thus allowing commercialization according to the local conditions of cultivation. Thus, lots of higher vigor may be destined to regions with bigger environmental limitations during sowing season (Peske et al., 2006). Various vigor tests are available and differ regarding to technology, time and ease of execution, being that the most studied ones are those related to initial events of the deterioration sequence (Marcos Filho, 1999), such as degradation of cellular membranes and the reduction of respiratory activity, which allow separating seed lots in regards to vigor (Abrantes et al., 2010).

The test of first count of germination is based on the principle that the samples that present higher percentages of normal seedlings during the first count, established by the Rules of Seed Analysis (Brasil, 2009), for each culture, will be the most vigorous which correlates with the index of germination speed. However, there can be a better answer than the latter reinforcing so, the affirmation that this test is of great interest to evaluate seed vigor, taking into account its practicality and execution time.

To use the germination test itself for running both tests mentioned above, all what we need is follow the norms of rules of seed analysis (Brasil, 2009), where the uniformity and speed of seedling emergence are the most important components within the current concept of seed vigor, considering the evaluation of seedling growth a logical and specific vigor test, as well as to evaluate the length of normal seedlings (Aosa,1983).

The evaluation of dry matter and length of seedlings are related to germination speed, taking into account that lots that show more vigorous seeds will originate seedlings with higher rates of development and gain of biomass due to having greater adaptability, since they use their reserves of the tissues of storage for the differentiation of the tissues, and consequently, promoting the growth and development of the embryonic stem of the seedling (Dan et al., 1987).

The methods for evaluating vigor can be classified directly when performed in the field or even under laboratory conditions, which simulate adverse factors of the field; or indirect when performed in laboratory evaluating the physical, physiological and biochemical characteristics that express the quality of seeds (Ferreira and Borghetti, 2004). In general, the low vigor of seeds is associated to reductions in speed and lack of uniformity of emergence, as well as in the reduction of initial size of the seedlings, in the accumulation of dry matter, in the leaf area and, consequently, in the rate of cultivation growth (Schuch et al., 2000; Machado, 2002; Höfs et al., 2004; Kolchinski et al., 2005). The cause of failure or reduction in speed of emergence is frequently attributed to low vigor associated to the deterioration process of the seeds (Rossetto et al., 1997).

As previously reported, various vigor tests are available and differ in methodology, time and ease of implementation, and the most studied are those related to early events of sequence deterioration (Delouche and Baskin, 1973),. since the vigor and deterioration of the seed are physiologically linked, being reciprocal aspects of quality, where deterioration has a negative connotation, while vigor has a positive connotation, as they are inversely proportional (Delouche, 2002). The seed does not start the deterioration process before reaching physiological maturity, since before this period it is not an independent unit of the parent plant yet. However, unfavorable environment conditions during maturation may determine the formation of seeds with poor physiological potential (Marcos Filho, 2005).

The tests that evaluate the initial events of deterioration the test of electrical conductivity, as previously reported, was proposed by Matthews and Bradnock (1967) in order to estimate vigor of pea seeds. This test measures the amount of electrolytes released by the seeds during soaking, which is directly related to the integrity of cell membranes (Matthews and Powell, 1981). Among the various procedures used in the determination of seed vigor, one of the alternatives that have shown promising results is the measuring of respiratory activity under laboratory conditions (Mendes et al., 2009; Aumonde et al., 2012; Marini et al., 2012), since breathing is the first metabolic activity that goes along with rehydration of the seed and the increase in this metabolic process varies from negligible values to high levels shortly after the start of imbibition (Popinigis, 1977; Ferreira and Borghetti, 2004). In this process, the oxidation of organic substances in a cellular system with gradual energy release occurs through a series of reactions, with molecular oxygen as a final electron acceptor. The respiratory substrates may be carbohydrates like starch, sucrose, fructose, glucose and other sugars or even lipids, especially triglycerides, organic acids and proteins (Taiz and Zeiger 2009; Marenco and Lopes, 2007).,

The increase in respiratory activity of the seed can be evaluated by the amount of carbon dioxide (CO_2) eliminated, by the amount of oxygen (O_2) absorbed or by the respiratory quotient (QR). The respiratory rate of the seed is influenced by its moisture content, temperature, and membrane permeability by oxygen and light strain (Popinigis, 1977). Respiration implicates in the loss of dry matter and in gas exchange, being methods that are used based on the determination of these characteristics. However, the measuring of dry matter variation requires large amount of material, besides being considered an analysis that is somewhat time-consuming to obtain the result, given that the plant material must be completely dried in an oven (Marenco and Lopes, 2007).

The methods based on gas exchange are more sensitive, require less material and are not destructive, and can be the gage measuring O_2 consumption, for example, Warburg respirometer and Clark electrode (potentiometry), in measuring liberated CO_2, using physical methods as the analyzer of infrared gas (IRGA), or physiochemical which are based on CO_2 retention on a base and quantification by titration calorimetry or conductivimetry (Maestri et al., 1998). Among the different forms of verification of seed physiological quality in the breathing process special attention is due to the high ratio between this phenomenon and seed quality (Mendes et al., 2009; Aumonde et al., 2012). Given that the greatest current interest, when evaluating the physiological quality of seeds, is to obtain reliable results in a relatively short period of time, it is expected that this review agility allows fast decision making during different stages of seed production, especially between maturation phase and future seeding (Dias and Marcos Filho, 1996).

Tests for the rapid assessment of viability or vigor represent important components in the control of seed quality programs and allow to speed up the obtaining of information by discarding lots of inferior quality during the reception at the processing unit and rationed management (Marcos Filho, 2005).

Within this context and having in mind that the analysis of physiological seed quality should be seen as a dynamic activity, that shows constant evolution, both by improving the resources available for its evaluation and the incorporation of new methods (Novembre, 2001), it becomes of extreme importance to prove the efficiency of new analysis, such as the application of the breathing test in order to obtain results with the aim of separating seed lots in regards to vigor.

Conflict of Interest

The authors have not declared any conflict of interest.

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