ACCELERATED AGING AND CONTROLLED DETERIORATION FOR THE DETERMINATION OF THE PHYSIOLOGICAL POTENTIAL OF ONION SEEDS

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ABSTRACT: International research on vegetable seed vigor is not at the same level attained for grain crops species. This study was conducted to identify reliable procedures for the accelerated aging and controlled deterioration tests to rank onion (*Allium cepa* L.) seed lots according to their physiological potential. Six seed lots of the cultivars Aurora and Petroline were evaluated in the laboratory for germination, first count, seedling vigor classification, traditional and saturated salt accelerated aging (41ºC / 48 and 72 h), controlled deterioration (24% of water / 45ºC / 24 h) and seedling emergence tests. Seed moisture content after the saturated salt accelerated aging test was lower and uniform, which is considered an important advantage in comparison to the traditional procedure. The saturated salt accelerated aging (41ºC / 48 and 72 h) and controlled deterioration (moisture content adjusted to 24% / 45ºC / 24 h) tests were the best procedures to assess the physiological potential of onion seeds, and are indicated for use in quality control programs.
Key words: *Allium cepa*, seed analysis, vigor

ENVELHECIMENTO ACELERADO E DETERIORAÇÃO CONTROLADA NA DETERMINAÇÃO DO POTENCIAL FISIOLÓGICO DE SEMENTES DE CEBOLA

RESUMO: Estudos sobre testes para avaliação do potencial fisiológico de sementes de hortaliças têm sido menos freqüentes que os conduzidos com as de grandes culturas, tanto no Brasil como no exterior. O presente trabalho teve como objetivo avaliar a eficiência de diferentes procedimentos para a condução dos testes de envelhecimento acelerado e de deterioração controlada na determinação do potencial fisiológico de sementes de cebola. Seis lotes de sementes dos cultivares Aurora e Petroline foram submetidos aos testes de germinação, primeira contagem de germinação, classificação do vigor de plântulas, envelhecimento acelerado tradicional e com uso de solução saturada de NaCl (41ºC/48 e 72 horas), deterioração controlada (24% de água/45ºC/24 horas) e emergência de plântulas em casa de vegetação. O grau de umidade das sementes expostas à solução salina, menor e mais uniforme após os períodos de envelhecimento, constitui importante vantagem na utilização desse procedimento, em relação ao tradicionalmente utilizado para a condução desse teste. Os testes de envelhecimento acelerado com solução saturada de NaCl (41ºC/72 horas) e de deterioração controlada (24% de água/45ºC/24 horas) foram considerados eficientes para avaliação do potencial fisiológico de sementes de cebola, sendo indicados para utilização em programas de controle de qualidade.
Palavras-chave: *Allium cepa*, análise, vigor

INTRODUCTION

The germination test is routinely utilized to evaluate seed physiological potential, in laboratories; however, it frequently produces results that overestimate seed performance under less favorable environmental conditions. The use of vigor tests has been useful to identify consistent differences in the performance of seed lots under a wide range of environmental conditions.

Some tests are considered efficient to evaluate seed vigor, especially for grain crop species, but vegetable seeds have deserved less attention from researchers, in spite of their high market prices. As a consequence, studies in attempt to identify the efficiency of different vigor tests should be encouraged especially for those species possessing relatively small-sized seeds with less expressive amounts of stored reserves and prone to deterioration after physiological maturity. Onion seeds possess characteristics which fit the above description, since usually lose viability and vigor faster than those of other crops (George, 1985). For this reason, Thomazelli et al. (1990) and Stumpf (1993) emphasized the importance of testing seed vigor to monitor the deterioration process of onion seeds.

Among the relatively few studies dealing with seed vigor tests for onion seeds in Brazil are those on accelerated aging (Lima, 1993; Piana et al., 1995), controlled deterioration (Lima, 1993), cold and electrical con-

ductivity tests (Lima, 1993; Piana et al., 1995; Torres, 1998) but information documented in literature is not enough to establish standard procedures.

The accelerated aging test have been developed to estimate the relative storability of seeds (Delouche & Baskin, 1973). This test has been used to evaluate onion seed performance during storage, providing indications that the temperature and the aging period would be 42ºC and 48 hours (Stumpf, 1993) or 40ºC/48 hours (Caneppele, 1994). The International Seed Testing Association (ISTA, 1995) suggests a combination of 41ºC/72 hours.

Some studies have also verified the efficiency of this test to evaluate field seedling emergence potential. Research conducted with onion seeds (Lima, 1993; Piana et al., 1995) and other vegetable crop seeds, such as carrot (Spinola et al., 1998) and broccoli (Mello et al., 1999) has shown that, among other studied tests, the accelerated aging test provided the closest relation to seedling emergence.

However, for most vegetables and other species that produce relatively small seeds, the traditional procedure used for the accelerated aging test have limitations such as a non-uniform water absorption among seed samples. This situation may lead to different levels of deterioration, thus compromising the standardization of germination results after aging.

In that respect, Powell (1995) reported that after 24 hours of accelerated aging (100% relative humidity at 45ºC), onion seeds showed a marked variation in seed moisture content, ranging from 11.8 to 24.0% among seed samples; germination after the aging period was inversely proportional to seed moisture content. Thus, Marcos Filho (1999) recommends variations in seed moisture content up to 3 to 4% after aging as tolerable. These indications are based on research conducted for major crop seeds, since information concerned to vegetable crop seeds is less available.

The SSAA (Saturated Salt Accelerated Aging), was proposed by Jianhua & McDonald (1996) working with seeds of *Impatiens wallerana* Hook. They verified that the use of a NaCl solution was efficient to control the water uptake by seeds, to evaluate vigor and to reduce water content variation among seed samples submitted to this test.

The efficiency of this alternative to detect different levels of seed physiological potential among seed lots was also verified for green pepper (Panobianco & Marcos Filho, 1998), carrot (Rodo et al., 2000), cucumber (Bhering et al., 2000) and tomato (Panobianco & Marcos Filho, 2001). On the other hand, results of Ribeiro & Carvalho (2001) did not show the same efficiency when testing seed vigor of lettuce, broccoli and carrot.

The controlled deterioration test was also developed as an alternative for vigor testing in vegetable seeds, in order to provide greater precision in the control of the high relative humidity and temperature to which seeds remain exposed during the accelerated aging test. Thus, while during accelerated aging the water uptake by seeds from each sample occurs at different speeds, in the controlled deterioration test the initial seed moisture content is adjusted to the same level, before the exposure to a high temperature in a water bath (Matthews, 1980).

First studies to evaluate vegetable seed lots (lettuce, beet, brassicas, onion and carrot) of poor filed emergence and low storability (Matthews, 1980; Powell & Matthews, 1984), indicated that the controlled deterioration test should be successfully utilized to evaluate seed vigor. This was also observed for onion (Lima, 1993), green pepper (Panobianco & Marcos Filho, 1998), broccoli (Mendonça et al., 2000) and tomato seeds (Panobianco & Marcos Filho, 2001).

Therefore, the objective of the present work was to evaluate the efficiency of different procedures for conducting the accelerated aging and controlled deterioration tests to determine the physiological potential of onion seeds, allowing them to be used in quality control programs.

MATERIAL AND METHODS

This study was carried out in Piracicaba, SP, Brazil from April to October, 2000. Six onion seed lots of cultivars Aurora and Petroline produced, respectively, in the 96/97 and 98/99 cropping seasons in Bagé, RS, Brazil, were utilized, being submitted to moisture content determination in an oven at 105±3ºC, during 24 hours, according to recommendations of the Rules for Testing Seeds (Brasil, 1992), with two replicates of approximately 3.0 g of seeds per lot, expressing results as mean percentage for each lot (wet basis). Germination was evaluated at 20ºC, with four replicates of 50 seeds for each lot, distributed in plastic boxes (11.0 x 11.0 x 3.5 cm) on two sheets of paper (blotter type) moistened with an amount of water equivalent to 2.5 times the weight of the dry paper; normal seedlings were recorded at six and 12 days after sowing, also according to criteria established in the Rules for Testing Seeds (Brasil, 1992). Results were expressed as mean percentage of normal seedlings, for each lot.

The germination first count test was performed with the germination test; the percentage of normal seedlings was recorded on the sixth day after sowing (Brasil, 1992).

Seedling vigor classification comprised the evaluation of normal seedlings in the germination test, as both tests were conducted concurrently. In the first evaluation, normal seedlings that were healthy, without defects and with well-developed cotyledons and root systems were classified as “strong” normal seedlings (Nakagawa,

1999). The other seedlings remained in the substrate for a second evaluation, during which normal seedlings were identified and computed either as "strong" or "weak". The results were expressed as mean percentage of "strong" seedlings for each lot, considering the two evaluations.

Accelerated aging was conducted in plastic boxes (11.0 x 11.0 x 3.5 cm) having a suspended aluminum screen inside, in which seeds, after being weighed (approximately 3.0 g), were distributed so as to form an uniform layer. Water was added (40 ml) to each plastic box. The boxes were covered and maintained in an incubator at 41ºC for 48 and 72 hours, after which seeds were submitted to the germination test. Evaluations were performed six days after sowing and the results expressed as mean percentage of normal seedlings for each lot. Seed moisture content was determined before and after the aging period, to evaluate the uniformity of test conditions. The same test was also carried out utilizing the procedure proposed by Jianhua and McDonald (1996), replacing the 40 mL water added to each individual compartment with the same amount of a NaCl saturated solution (40 g NaCl/100 mL water).

For the controlled deterioration test seeds were first moistened by the humid atmosphere method (Rossetto, 1995) until they reached 24% water. For this, 40 mL of water were added inside the plastic boxes (11.0 x 11.0 x 3.5 cm) and samples of approximately 3.0 g seeds were placed on the aluminum screen, distributed as a uniform layer. The boxes were covered and maintained in an incubator at 20ºC; seed moisture content was monitored by means of successive weighings, until the desired values were obtained. Later on, samples were packaged in hermetically sealed aluminized containers and maintained under cold storage at 5-8ºC for five days, to ensure uniform water distribution inside the seeds. After that period, seed samples were placed in a water bath at 45ºC for 24 hours (Powell, 1995). Following, the containers were quickly immersed in cold water to reduce the temperature and then the germination test started. Evaluations were performed six days after sowing and the results were expressed as mean percentage of normal seedlings for each lot.

Greenhouse seedling emergence was performed in four replicates of 100 seeds for each lot distributed in polystyrene trays with individual cells containing the commercial substrate Plantimax®. Trays were covered with expanded vermiculite as a protection against excessive evaporation, transferred to a greenhouse equipped with a micro-sprinkler system and maintained at 25ºC. Evaluations were performed 14 days after sowing, when emerged seedlings with a height equal to or above 1.0 cm were counted. Results were expressed as mean percentage of normal seedlings, for each lot.

The results of each test were statistically analyzed, separately for each cultivar, according to a completely randomized design, with six treatments (lots of different vigor levels) and four replicates. The Tukey test was used at 5% for multiple comparison of means, by using the "Sistema de Análise Estatística para Microcomputadores – SANEST" (Zonta & Machado, 1984).

RESULTS AND DISCUSSION

Table 1 shows that the results obtained for germination, first count, seedling vigor classification and greenhouse seedling emergence indicated the lowest performance of lots 1, 2 and 3 of 'Aurora" seeds; the seedling vigor classification and the seedling emergence tests also indicated the highest physiological potential for lot 4.

Differences between seed lots were also observed for 'Petroline' (Table 1). Lots 11, 12 and 14 were considered as higher in physiological potential by the germination, first count and seedling emergence tests, and lots 10, 13 and 15 were identified as those of lowest quality.

Results of the saturated salt accelerated aging (48 and 72 hours) and controlled deterioration (24% water) tests showed that lot 4 was superior in quality and lot 3 had inferior potential (Table 2), in a similar way as the seedling vigor classification and greenhouse seedling emergence tests (Table 1); the traditional accelerated aging test (48 and 72 hours) detected the poorest performance for lot 3 and an intermediate position for lots 1 and 2 (Table 2).

Table 1 - Germination (G), first count in germination (GFC), seedling vigor classification (SVC) and greenhouse seedling emergence (GE) tests, in six onion seed lots of Aurora and Petroline cultivars.

	G	GFC	SVC	GE
Seed Lots	---------- % ----------			
	Aurora			
1	89 ab*	75 b	52 c	84 bc
2	80 bc	67 bc	51 cd	76 cd
3	76 c	57 c	42 d	69 d
4	93 a	90 a	85 a	92 a
5	91 a	85 a	70 b	90 ab
6	95 a	91 a	75 b	91 ab
CV%	5.3	4.3	4.8	4.4
Seed Lots	---------- % ----------			
	Petroline			
10	82 b*	71 c	59 b	70 d
11	94 a	91 a	74 a	92 ab
12	91 a	83 ab	60 b	85 abc
13	79 b	70 c	51 b	77 cd
14	94 a	91 a	77 a	93 a
15	85 b	79 bc	61 b	83 bc
CV%	3.5	4.7	6.1	4.4

*Mean comparisons within each column (Tukey test at 5%).

Table 2 - Traditional accelerated aging (TAA), saturated salt accelerated aging (SSAA) and controlled deterioration (CD) tests, in six onion seed lots of Aurora and Petroline cultivars.

	TAA		SSAA		CD
	48h	72h	48h	72h	24%
Seed Lots	---------- % ----------				
	Aurora				
1	67 c*	71 b	64 b	66 b	68 c
2	64 c	69 b	53 c	49 c	67 c
3	52 d	56 c	33 d	39 d	39 d
4	86 a	90 a	83 a	77 a	88 a
5	80 b	86 a	60 b	65 b	79 b
6	81 ab	87 a	64 b	69 b	81 b
CV%	3.2	4.7	3.5	2.7	1.9
Seed Lots	---------- % ----------				
	Petroline				
10	70 d*	61 b	74 b	64 d	69 c
11	80 bc	85 a	87 a	86 ab	87 a
12	77 c	61 b	77 b	79 bc	80 b
13	70 d	40 c	64 c	61 d	68 c
14	94 a	83 a	90 a	87 a	89 a
15	85 b	72 ab	78 b	72 c	79 b
CV%	2.6	7.8	4.4	3.8	2.5

* Mean comparisons within each column (Tukey test at 5%).

Seeds of all lots showed an uniform initial moisture content, with a maximum difference of 0.5% (6.8 to 7.3%). After moisturizing, the moisture content of seed samples for the controlled deterioration test ranged between 23.9 and 24.2%, thus indicating that seeds attained the desired and uniform moisture content. Variations of only 0.4% (10.9 to 11.3%) and 1.1% (10.2 to 11.3%) were verified after running the saturated salt accelerated aging test, for the 48 and 72 hour periods, respectively. However, for the traditional accelerated aging, the observed variations after aging were 1.5% (35.3 to 36.8%) for the 48 hour period and 5.8% (33.5 to 39.3%), for the 72 hour period; the later values exceeded the 3 to 4% limit recommended by Marcos Filho (1999).

With regard to cultivar Petroline (Table 2), higher seed vigor values were observed, in general, for lots 11 and 14, and smaller values for lots 10 and 13, for the traditional (72 hours) and saturated salt (48 and 72 hours) accelerated aging, and controlled deterioration (24% water) tests; this performance was also verified for the germination, first count and greenhouse seedling emergence tests (Table 2).

The saturated salt accelerated aging (72 hours) and controlled deterioration (24% water) tests exhibited greater compatibility with the results obtained for the greenhouse seedling emergence test (Table 2).

The initial seed moisture content among 'Petroline' lots showed a maximum difference of 0.3% (6.2 to 6.5%). The observed adjusted moisture content was also uniform for the controlled deterioration test, i.e., results within 24.3 and 24.7%; the adopted procedures were, therefore, efficient.

However, after the traditional accelerated aging test, variations in moisture content between seed lots were of 3.1% (34.9 to 38.0%) and 9.6% (31.0 to 40.6%), respectively, for the 48 and 72 hour periods; this value exceeded the limit considered as tolerable for conducting the test (3 to 4%, according to Marcos Filho, 1999). Therefore, as also verified by Powell (1995) with onion seeds, a striking variation occurred in moisture content after the traditional accelerated aging test; for this reason, the author has not considered the traditional accelerated aging test as reliable to detect differences in onion seed vigor.

The maximum variation in seed moisture content from 'Petroline' in the saturated salt accelerated aging test was 1.4% (11.2 to 12.6%) for the 48 hour period and 0.9% (11.2 to 12.1%) for the 72 hour period. The observations of Jianhua & McDonald (1996) were confirmed as the use of salt solutions induced seeds to absorb water at a lower speed, resulting in lower moisture content variation attained after the test. In addition, according to the same authors, since the relative humidity during the test is low, there is a considerable reduction of fungi development, which may be important sources of variation for the results; this advantage was verified in the present research (Figure 1).

Even though this work probably is the first to report the use of the saturated salt accelerated aging test with onion seeds, consistent results were verified for green pepper (Panobianco & Marcos Filho, 1998), carrot (Rodo et al., 2000), cucumber (Bhering et al., 2000) and tomato seeds (Panobianco & Marcos Filho, 2001). Therefore, considering both onion cultivars, studied here the saturated salt accelerated aging test for 72 hours, which showed the closest relation to seedling emergence test, deserves emphasis. Thus, this test can be considered as a promising alternative to evaluate the physiological potential of onion seeds, since it is conducted with the use of similar methodology and the same equipment that are similar to the traditional procedure. Likewise, the controlled deterioration test, with seed moisture content adjusted to 24%, was also considered as efficient; it is worth noting that, in spite of being more labor-intensive than the accelerated aging procedure, requiring the previous controlled standardization of the seed water content, it exhibits a desired sensitivity to detect differences in the physiological potential of onion seeds. Notwithstanding, both tests, which are based practically on the same principle, could be utilized in quality control programs for onion seeds.

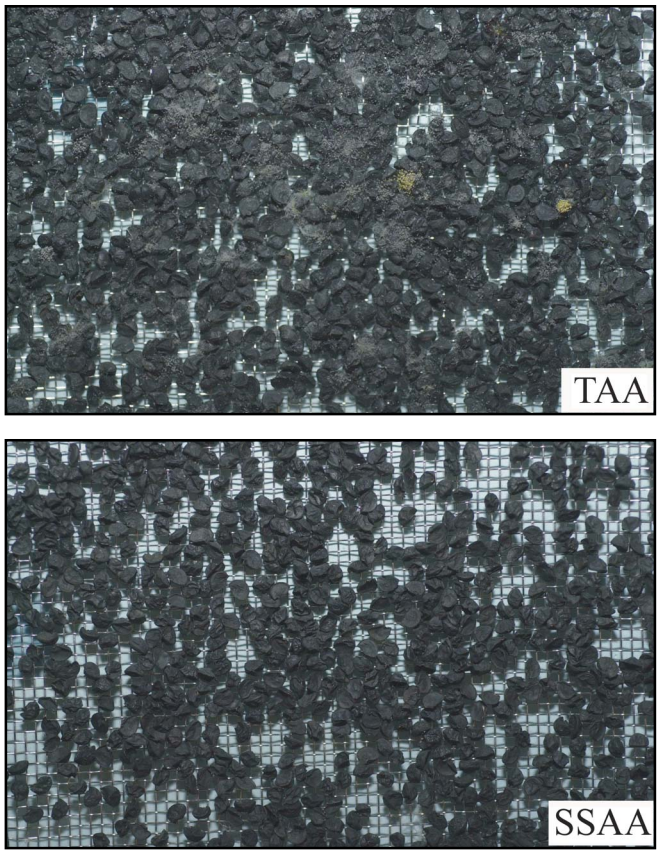

Figure 1 - Onion seed samples, cultivar Petroline, submited to traditional accelerated aging (TAA) and saturated salt accelerated aggings (SSAA) tests, 72 hours.

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