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Scientific Article

Physiological and sanitary quality of cockscomb seeds stored for different periods

Janine Farias Menegaes [1*], Geovana Facco Barbieri [1], Rogério Antônio Bellé [1], Ubirajara Russi Nunes [1]

Abstract

The objective was to evaluate the physiological and sanitary quality of cockscomb seeds stored for different periods. The experiment was conducted in a completely randomized design, with a 2x5 factorial scheme (two species of cockscomb: *Celosia argentea* L. and *Celosia cristata* L., and five seed lots with different storage periods: 0, 4, 8, 12 and 16 years, from the date of harvest) with four replicates. After harvesting each batch of seeds, in their respective years, they were stored with an average moisture content of 11% and average germination of 98% in cold chamber in packages of Kraft paper (15 ºC and 40% RH). It was observed that the average vigor (first count of germination) of the seeds of the two species of cockscomb was 88%, and the average germination of normal seedlings was 89 and 96% for the *C. argentea* and *C. cristata* species, respectively, with mean germination of 2.9 days for all storage periods. The relative frequency demonstrated homogeneity in the germination of the two species of cockscomb maintaining germinative viability in all periods of storage. It is concluded that the physiological potential and sanitary quality of the seeds of the *C. argentea* and *C. cristata* were preserved for periods ranging from zero to sixteen years of storage.
Keywords: *Celosia argentea* L., *Celosia cristata* L., cut flower, ornamental seeds, vigor.

Resumo

Qualidade fisiológica e sanitária de sementes de celosias armazenadas por diferentes períodos

O objetivo foi avaliar a qualidade fisiológica e sanitária de sementes de celosias armazenadas por diferentes períodos. O experimento foi conduzido em delineamento inteiramente casualizado, com esquema fatorial 2x5 (duas espécies de celosias: *Celosia argentea* L. e *Celosia cristata* L.; e cinco lotes de sementes com diferentes períodos de armazenamento: 0, 4, 8, 12 e 16 anos, a partir da data de colheita), com quatro repetições. Depois de colhido cada lote de sementes, em seus respectivos anos, foram armazenados com grau de umidade médio de 11% e germinação média de 98%, em embalagens de papel Kraft na câmera fria (15 ºC e 40% UR). Observou-se que o vigor (primeira contagem de germinação) médio das sementes das duas espécies de celosias foi de 88%, e a germinação de plântulas normais média foi de 89 e 96% para as espécies de *C. argentea* e *C. cristata*, respectivamente, com tempo médio de germinação de 2,9 dias para todos os períodos de armazenamento. A frequência relativa demonstrou homogeneidade na germinação das duas espécies de celosias mantendo a viabilidade germinativa em todos os períodos de armazenamento. Conclui-se que o potencial fisiológico e a qualidade sanitária das sementes das celosias *C. argentea* e *C. cristata* foram preservados por períodos que variaram de zero a dezesseis anos de armazenamento.
Palavras-chave: *Celosia argentea* L., *Celosia cristata* L., flor de corte, sementes ornamentais, vigor.

Introduction

The seed storage aims to ensure the completeness and the viability of the plant structures for extended periods, preserving their genetic, physiological, sanitary and physical qualities (Carvalho and Nakagawa, 2012). The conservation of seed quality, mainly, the physiological occurs by means of dissection (water removal) and the decrease of temperature, and this technique is used only in orthodox seeds (Angelovici et al., 2010; Oliveira et al., 2011). The presence and the action of phytopathogens and insects, the relative humidity and air temperature, the types of packages, the availability of oxygen and the period of storage, are also important factors for the conservation of seeds quality (Costa, 2009; Carvalho and Nakagawa, 2012).

The inadequacy of storage conditions provides the acceleration of deterioration and, consequently, the reduction of seeds quality, evinced during the germination and the initial development of seedlings (Costa, 2009; José

[1] Federal University of Santa Maria (UFSM), Crop Science Department, Santa Maria-RS, Brazil. *Corresponding author: janine_rs@hotmail.com
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et al., 2010). The velocity in which occurs this deterioration can be mitigated by the adequacy of initial quality of the lot, in relation to the degree of humidity, to the stage of seeds maturation in the moment of the crop, among others, thus, allowing a good development of seedlings in field (José et al., 2010; Sales et al., 2011).

In Brazil, the main cut flowers produced from seeds are: *Anthirrhinum majus* Linn, *Dianthus chinensis* x *barbatus*, *Celosia argenta* L. and *C. cristata* L., *Helianthus annus* L., *Eustoma grandiflorum* (Raf,) Shinners, *Callistephus chinensis* Nees L., among others (Paiva and Almeida, 2014). Facing this diversity, the success of flower propagated by seeds needs high physiological and sanitary quality, as vigor, viability and potential of storage of the used seeds (Khan et al., 2003), contributing positively, for the formation of the plants stand and, consequently, for the production of the floral stem of quality.

Among the main cut flowers produced by seeds, we highlight the cockscomb species, belonging to the Amaranthaceae family, known popularly as plumed cockscomb (*Celosia argentea* L.) due to the fact that its characteristic inflorescence is in form of feather and, the cockscomb species (*Celosia cristata* L.) in virtue of its inflorescence is fan-shaped. Both species present ornamental character, with intense flourishing and diversity of colors of their inflorescences (Bellé and Spannenberg, 1997; Lorenzi, 2013).

In this sense, the objective of this work was to evaluate the physiological and sanitary quality of cockscomb seeds (*Celosia argentea* L. and *Celosia cristata* L.) stored by different periods.

Material and Methods

The experiment was conducted in the period between September to December, 2016. We used the completely randomized design, in 2x5 factorial scheme (two species of cockscombs and five batch of seeds with different periods of storage), with four replications. The batch of seeds of cockscomb species (*Celosia argentea* L. and *Celosia cristata* L.) were obtained from the cultivation carried out in experimental area of the Floriculture Sector (29º43'S; 53º43'W and altitude of 95 m), in the respective years: 2000, 2004, 2008, 2012 and 2016, and they were collected in the month of March of each year before quoted. After harvested each batch of seeds, in their respective years, they were stored with average degree of humidity of 11% and average germination of 98%, in packages of Kraft paper in cold chamber (15 ºC and 40% RH), by the periods of 0, 4, 8, 12 and 16 years from the date of harvest.

The qualities of seeds of the two species were evaluated by the following tests: weight of one thousand seeds determined by the methodology described by the manual of Rules for Seeds Analysis (Brasil, 2009a); degree of humidity determined by the method of greenhouse 105±3 ºC (24 h), adapted by Brasil (2009a); germination pattern degree four replications of 100 seeds were sowed, in boxes of crystal polystyrene (gearbox), moistened with distilled water in the proportion of 2.5 times the mass of dry paper. The boxes were maintained in germinator type BOD (Box Organism Development), with photoperiod of 24 h and temperature of 20±2 ºC. The evaluations of germination were performed in the fourth and 14th days after the sowing (DAS) and, the results expressed in percentage of normal seedlings. The germination of normal seedlings (GER) was determined at the 14th DAS (Brasil, 2009a); first germination count (PCG), germination speed index (IVG) and mean time of germination in days (TMG): they were performed together with the standard test of germination. The PCG was determined in percentage of normal seedlings on the fourth DAS (Brasil, 2009a). The IVG and TMG were determined with daily evaluations until seven DAS, according to the methodology described by Maguire (1962) and Furbeck et al. (1993), respectively; length and mass of seedling: they were carried out with four replications of 20 seeds, sowed in two lines kept apart in superior third of gearbox and, maintained in the same condition of the standard test of germination. On the fourth DAS, it was measured the length of the aerial part and of the root of ten normal seedlings of each repetition. In the sequence, ten normal seedlings of each repetition were submitted to the drying in greenhouse of forced ventilation at 65±5 ºC for 48 h, for determination of dry mass (Nakagawa, 1999); relative frequency of germination determined by methodology of Labouriau and Valadares (1976), expressed in Equation 1.

$$Fr = ni / \sum_{i=1}^{k} ni \quad (1)$$

In which: Fr = relative frequency of germination; ni = number of seeds germinated by day; Σni = total number of germinated seeds observed in test of IGV; entropy: synchronization index of germination determined by the methodology of Labouriau and Valadares (1976), expressed in Equation 2.

$$E = \sum_{i=1}^{k} fi.\log 2.fr \quad (2)$$

In which: E = informational entropy (bits); fr = relative frequency of germination; log2 = logarithm in base 2; sanity test in filter paper: it was carried out through the incubation in substrate of paper (Blotter Test), in gearbox boxes, with four repetitions of 100 seeds.

The germination was inhibited by freezing method by 24 h and, subsequently, the seeds remained in BOD for five days with photoperiod of 12 h of light and temperature of 20 ± 2 ºC. They were evaluated with magnifying glass (stereoscope microscope) the percentages of infested seeds and the identification of phytopathogens at genus level (Brasil, 2009b).

The data expressed in percentage were transformed in arc-sine $\sqrt{x/100}$ and, the analysis of variance (ANOVA) and the comparison of averages by Scott-Knott test, at level of 5% of mistake, carried out with the help of the statistical program SISVAR (Ferreira, 2011).

Results and Discussion

In Figure 1A, we observed that the weight of one thousand seeds presented significant variation of 1.4% among the different periods of storage, and the general average was of 0.91 g and 0.74 g for *Celosia argentea* and *Celosia cristata,* respectively.

In relation to the humidity degree of cockscomb seeds of both species, initially with 11% of humidity and after the tested periods of storage, we verified that there was no statistical difference (Figure 1B). This result was expected in virtue of the adequate form of stocking. Marcos Filho (2015) reports that the humidity degree of seeds is a narrow characteristic associated to deterioration and, when stored inside the range of 10% to 13%, for orthodox seeds, as the cockscomb seeds, the conservation of the physiological potential can occur by long periods, as observed in this work. Hong and Ellis (1996) pointed that the orthodox seeds, characterize themselves for tolerating storage by long periods without occurrences of damages to its metabolism during the germination process.

The average first germination count (PCG) of seeds of both cockscomb species was of 88%, for all the periods of harvest and storage, we highlight that the batch of cockscomb seeds with eight years of storage (harvested in 2008) was the one that suffered the most deterioration in its physiological quality in relation to the other batches of seeds (Figure 1C).

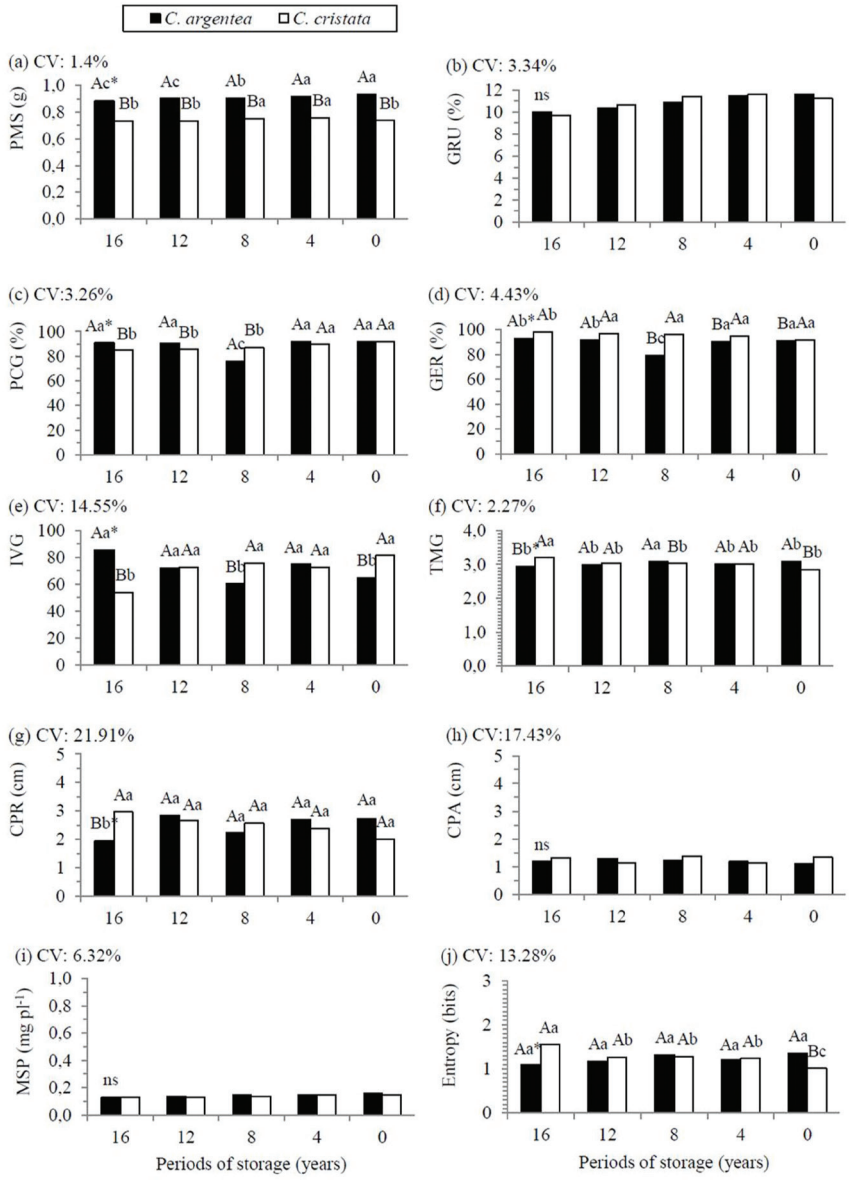

Figure 1. Weight of one thousand seeds (PMS), degree of humidity (GRU), first germination count (PCG), germination of normal seedlings (GER), germination speed index (IVG), mean time of germination (TMG) length of root seedling (CPR) and aerial part (CPA) entropy, seedling dry mass (MSP) of *Celosia argentea* L. and *Celosia cristata* L. as a function of different storage periods. * significant interaction and non-significant interaction of factors. Testing of averages not followed by the same letter, uppercase between species and lowercase between storage periods, differ by the Scott-Knott test (5% probability of error). CV: Coefficient of variation.

The germination of mean normal seedlings was of 89% and 96% for *C. argentea* and *C. cristata* species, respectively, for all the periods of storage (Figure 1D). Bellé and Spannenberg (1997) evaluated the germination of cockscomb seeds produced in Santa Maria, RS, with average of 72% to 95%, according to the time of harvest. Rocha and Demattê (2003) observed germination of 79% for seeds stored for one year of *Amaranthus tricolor* L., preserving its physiological quality.

The MAPA (Ministry of Agriculture, Livestock and Supply) regulates minimum germination value of 70% for seeds with physiological quality (Brasil, 2011). We observed that, in the present work, the first count and the germination of normal seedlings for both cockscomb species in all the periods of storage remained above the value required by MAPA, indicating efficiency in the way of storage of these seeds for maintenance of their physiological potential.

The germination speed index (IVG) seeks to measure the speed in which the seeds establish themselves as seedlings. For the *C. argentea* species, the batch that obtained the greatest IVG (85.6) was the one with 16 years of storage (harvested in 2000).Yet, for the *C. cristata* species, we observed IVG of 81.5, for the batch of seeds harvested in 2016 (zero year of storage) (Figure 1E). The two batches of seeds mentioned presented TMG of 2.9 days, with coefficient of variation of 2.27% (Figure 1F). Khan et al. (2003) working with cockscomb seeds (*C. argentea* e *C. cristata*), in Pakistan, observed average of 78% of vigor on 3rd DAS (days after sowing) and 100% of germination on 7th DAS, for both species.

In relation to the development of seedlings of cockscomb species, conferred by the length tests of the root and aerial parts and dry mass of seedlings, we verified that the average of the root length of the seedling was of 2.48 and 2.52 cm for the *C. argentea* and *C. cristata* species, respectively. For the biometrical parameters of length of the aerial part and dry mass of seedling, we did not observe statistical difference (Figures 1H and 1I), with general average of 1.20 cm and 0.15 mg seedling^{-1}, respectively, for the two species. Marcos Filho (2015) mentions that the process of deterioration of seeds depends of genetic heritage, since the rearrange of the membranes for the beginning of the three-phase germinative process, added to the degree of humidity and the period of storage, are evinced in the development of seedlings (length and dry mass).

In the Figure 1J, the entropy of synchronization of germination was close for both cockscomb species with average of 1.23 and 1.27 for *C. argentea* and *C. cristata*, respectively. Lopes and Franke (2011) classify this parameter as a measurer of organization of germinative system, in this case represented by the batches of seeds analyzed in the germination test. Nassif and Perez (2000) report that the lower is the entropy; the greater is the organization and synchrony of the system. The proximity of the entropy, in which occurs the synchronization of the germination for the different batches of seeds can be associated to the good physiological quality, resulting in an elevated percentage of germinative (Rollwagen and Carvalho, 2011), as it was observed in this work for the two species of cockscomb in all the periods of storage.

In the Figure 2, the relative frequency of germination of seeds was distributed homogeneously in the period of evaluation of seven DAS, having an average time of germination of 2.9 days for both cockscomb species in all the periods of storage, corroborating with the values of the Figures 1D and 1F. Lopes and Franke (2011) report that the main mode of frequency distribution, in this case symmetric, demonstrates homogeneity of physiological potential of the different batches of seeds that presented fast germination, a result from the genetic material of high vigor and with viability of storage for long periods. Marcos Filho (2015) reports that the conditions of storage, mainly, the degree of humidity, affect negatively the physiological quality of seeds, resulting in low percentage of germination. However, in this work, we observed that the conditions of storage of seeds of cockscomb species maintained their physiological potential, identified by the percentages of germination, entropy and relative frequency of germination (Figures 1D, 1E and 2).

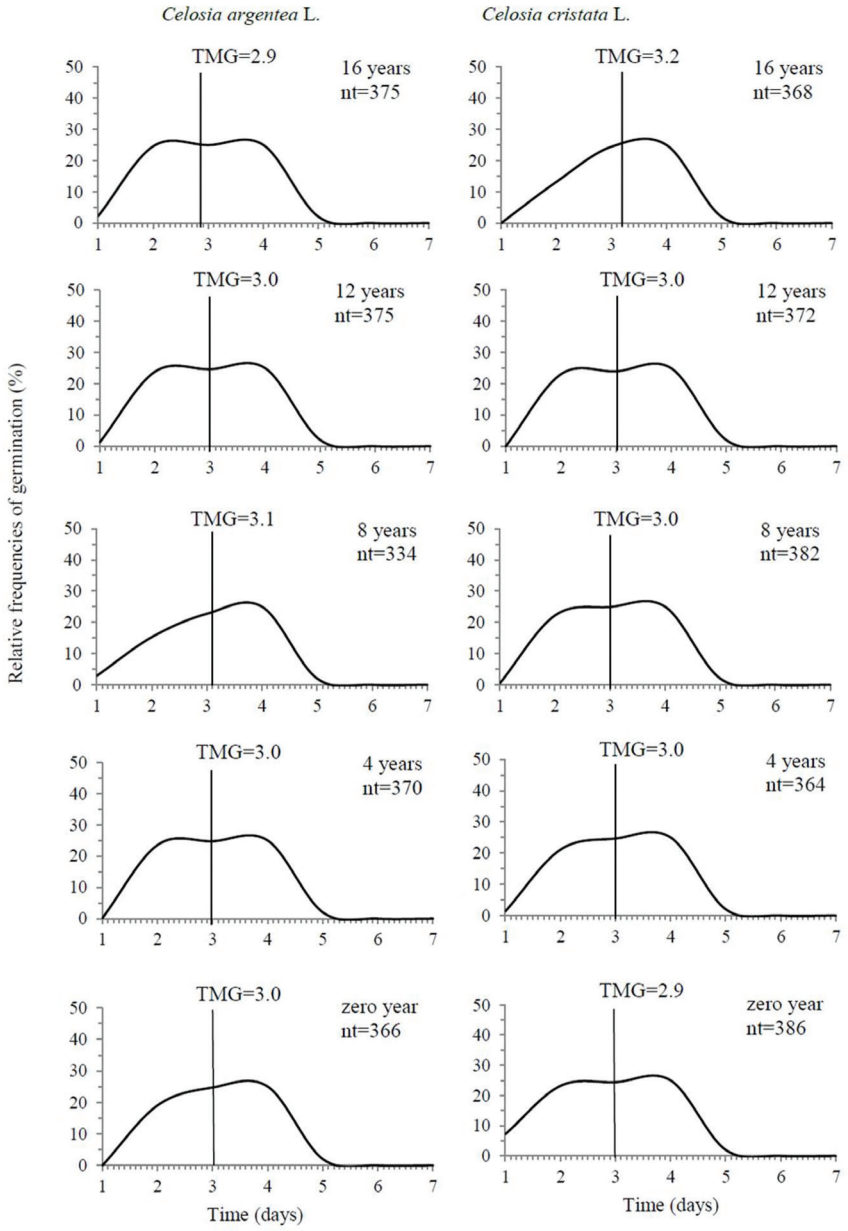

Figure 2. Relative frequencies (%) of the germination of *Celosia argentea* L. and *Celosia cristata* L. seeds as a function of time and different storage periods. TMG: mean germination time (in days), nt: total germinated seeds.

In Figure 3, we verified that the seeds of cockscomb species presented high incidence of phytopathogens in average of 45% for the period of 16 years of storage, in comparison to the other periods of storage. We observed that with the prolongation of the storage period, there was an increase of the percentage of incidence of phytopathogens in relation to the years of more recent storage, with average of 13%; 17%; 21% and 24% for the periods of 0, 4, 8 and 12 years, respectively.

The phytopathogens of greatest incidence identified in the seeds of cockscomb of the two species of this work were the ones of *Botrytis* sp., *Fusarium* sp. and *Penicillium* sp. genus. Among the phytopathogens, the ones of lowest percentage of infestation were the ones of *Botrytis* sp. genus, with general average of 2.2% of infestation on the seeds of the two species of cockscomb; however, the presence of these pathogens affects negatively the ornamental characteristic of the inflorescences, making it impossible their commercialization. Besides that, when the cultivation is destined for production of seeds, inflorescences attacked by these phytopathogens become unviable (Bellé and Spannenberg, 1997).

Pathogens of *Fusarium* sp. genus presented general average incidence of 14.2% on the cockscomb seeds. Phytopathogens of this genus characterize themselves by infestation at the end of the cycle, and they are more aggressive when the production is destined to the harvest of seeds. Yet, the pathogens of *Penicillium* sp. genus presented general average incidence of 7.4% on the seeds of cockscombs of both species. Machado (2000) mentions that the phytopathogens of *Penicillium* sp. genus characterize themselves as pathogens of storage and they contribute for the deterioration of the seeds, depreciating their physiological quality.

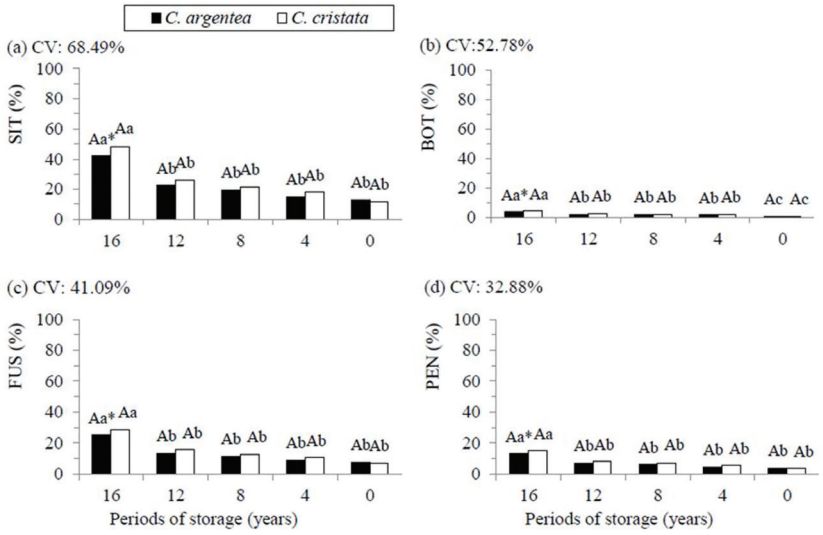

Figure 3. Total infested seeds (SIT) in the sanity test, *Botryti*s sp. (BOT), *Fusariu*m sp. (FUS) and *Penicillium* sp. (PEN) of *Celosia argentea* L. and *Celosia cristata* L. as a function of different storage periods. * significant interaction. Testing of averages not followed by the same letter, uppercase between species and lowercase between storage periods, differ by the Scott-Knott test (5% probability of error). CV: Coefficient of variation.

For Bellé and Spannenberg (1997) and Barreto et al. (2011), the greatest difficulty of the cultivation of floral stem from the seeds in the country is in virtue of the own production of seeds, because they characterize themselves as an activity highly specialized and of domain of international market. In the process of seeds import there is no information in relation to the sanitary quality, and this characterizes a problem that involves since the introduction of pathogens that do not exist in the country yet, as losses in virtue of diseases on seeds, saplings, plants in the field or even in post-harvest.

The longevity of seeds is a genetic characteristic; however, the conditions of inadequate storage can affect negatively their original characteristics (Costa, 2009). In general, we observed that the cockscomb species in all the periods of storage maintained intact their physiological and sanitary qualities. In which the germination of seeds has a relation inversely proportional with the degree of deterioration of the same, in which any change in conditions of storage can cause irreversible damage in the quality of seeds, specially, physiological, and maximizing this effect throughout the period of storage. Thus, the viability of storage of seeds through long periods, as observed in this work, helps the biological security of flora and maintenance of the germplasm bank focused on genetic improvement. Since the floriculture is an agroeconomic sector eager for innovation, a result of the constant investment in genetic improvement of ornamental flowers and plants.

Conclusions

The physiological and sanitary quality of the seeds lot of cockscomb (*Celosia argentea* L. and *Celosia cristata* L.) stored by different periods were preserved in conditions proposed in the present work, with viability of storage by long periods.

Author Contribution

JFM 0000-0001-6053-4221: planning, implementing of the experiment, collection and data interpretation, statistics and writing. **GFB** 0000-0003-0692-0206: planning, implementing of the experiment, collection and data interpretation and writing. **RAB** 0000-0001-6704-417X: planning, statistics and writing. **URN** 0000-0002-7124-9204: orientation, data interpretation and writing

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