

Evaluation of Physiological Quality of Seeds of Improved Snap Bean Lines under Different Storage

ABSTRACT

Seed quality is a key factor to succeed in a yield generation and storage is an important activity to control the physiological seed quality, as it preserves seed viability while maintaining its robustness at a reasonable level between planting and harvest. On this basis, the goal of this study was to evaluate the physiological quality of snap bean seeds stored in different types of packages and storage conditions. Eight genotypes comprising six lines and two controls were analyzed. It was applied a completely randomized block design with four replicates in a factorial scheme (8×2×2×7). Seed storage was performed in permeable and impermeable packages under controlled and uncontrolled conditions. The physiological seed quality was evaluated by means of germination and vigor tests. The different types of package and storage conditions influenced the physiological seed quality. Under controlled storage conditions, the permeable and impermeable packages were more efficient in controlling the physiological quality of snap bean seeds.

Keywords: Phaseolus vulgaris L.; viability; vigor; germination; conservation.

1. INTRODUCTION

Snap beans is an annual vegetable widely sown in several Brazilian regions. It belongs to the same botanical species as the common bean plant (*Phaseolus vulgaris* L.), being consumed when the pods are still immature. Its tender pods are consumed in human food either in natural, processed, frozen or canned forms (Filgueira et al., 2013). It is an excellent source of nutrients, rich in vitamins, minerals, and antioxidant compounds (Blair et al., 2010), constituting an important food in the Brazilian diet.

It is estimated that global production is around 6.5 million tons/year (FAO, 2014), with China being the largest producer, followed by Indonesia and Turkey. Brazil is the sixth largest country in volume produced, with a production of 56 thousand tons/year (Miklas et al., 2003; CEASA, 2010). The Southeast region of Brazil produces approximately 37 thousand tons/year of snap beans, being the state of Rio de Janeiro responsible for 21% of this production (CEAGESP, 2014). The

average sales of snap beans in Rio de Janeiro, adding all the resale units of the Central Supply – CEASA, are around 600 tons/month (CEASA, 2010).

Good quality seed are an essential requirement for succeeding in establishing crops and obtaining high yields. The interaction among physiological, sanitary, genetic, and physical traits, which directly interfere with the performance potential in the field and during storage, is what determines the quality of the seeds (Marcos Filho, 1999). Their quality is not improved; however, by means of adequate storage, they can be kept to a minimum deterioration (Oliveira et al., 2018).

Seed quality is greatly influenced by the conditions in which it is stored between harvesting and sowing (Coelho, 2018). Delouche and Baskin (1973) argue that there are genetic factors, forms of handling, and storage conditions that influence the rate of seed deterioration. During the storage period, a number of factors influence the rate of seed deterioration, the most important ones being temperature and relative humidity (Smith and Berjak, 1995), in addition to the type of package, which will determine the deterioration rate and, consequently, the maintenance of the physiological quality of the seeds.

As stated by Popinigis (1985), the physiological quality is described by the germination capacity, vigor, and longevity of seeds and grains affected by genetic, physical, physiological and sanitary factors. This author claims that package and environmental aspects, including temperature and relative air humidity, have a direct impact on the physiological quality of canned products.

Storage packages can be categorized into three types: permeable, in which, depending on the variation in air humidity, there are variations in the humidity content of stored seeds; semi-permeable, in which, despite having some resistance to humidity exchange, there is no complete impediment; and impermeable, in which there is no influence of the external environment on the stored seeds (Popinigis, 1985; Silva et al., 2014).

Seed and grain conservation, particularly in hot and humid regions, continues to be a challenge, requiring studies to clarify their correct storage in order to extend their shelf life and quality maintenance (Silva et al., 2010).

Given the lack of technical/scientific information regarding the conservation of snap beans, this study intended to evaluate the physiological quality of snap bean seeds stored in different types of package and storage conditions.

2. MATERIAL AND METHODS

The snap bean genotypes in question were from the germplasm bank of the Genetics and Plant Breeding Program of the *Universidade Estadual do Norte Fluminense Darcy Ribeiro* – UENF located in the municipality of Campos dos Goytacazes, state of Rio de Janeiro, Brazil.

The seeds were obtained from March to September 2011 from the experimental station of the *Instituto Federal Fluminense – Campus de Bom Jesus do Itabapoana*, municipality of Bom Jesus do Itabapoana, state of Rio de Janeiro, Brazil, in association with UENF. Nevertheless, eight genotypes of indeterminate growth habit, that is, six lines and two controls, were analyzed (Table 1).

Table 1. Description of the six lines and two controls of snap beans used in the experiment concerning growth habit and origin in Bom Jesus de Itabapoana municipality, Rio de Janeiro state.

Genotypes	Growth Habit	Origin
(L5) UENF 7-4-1	Indeterminate	UENF
(L7) UENF 7-6-1	Indeterminate	UENF
(L11) UENF 7-12-1	Indeterminate	UENF
(L12) UENF 7-14-1	Indeterminate	UENF

(L13) UENF 7-20-1	Indeterminate	UENF
(L21) UENF 14-4-3	Indeterminate	UENF
(L1) TOP SEED Blue Line**	Indeterminate	Commercia
(L3) Parent 19 (UENF 1445)**	Indeterminate	UENF

** Controls

The experimental randomized block design with four replicates was adopted. The experimental plot comprised 12 plants, at a row spacing of 1 m and 0.50 m between plants. Sowing was performed in May 2011, placing two seeds per pit, and, around 15 days after the emergence, the plants were thinned, maintaining one plant per pit, and staked.

After harvesting, the seeds were naturally dried until reaching humidity content of 12%. All plants were cultivation under the same conditions, and the cultural procedures were performed as recommended for the crop (Dourado and Fancelli, 2000).

The other phases of the experiment were conducted at the Agricultural Engineering Laboratory of the Center for Agricultural Sciences and Technologies (LEAG), located in UENF.

Two types of a package were used for seed storage: permeable (multiwall Kraft paper bag, size 20×10 cm) and impermeable (flexible multiwall aluminum – 12 µm polyester film (PET) + white ink + adhesive + 8 µm aluminum + adhesive + 15 g m² transparent polyethylene film). This last one was vacuum-sealed using the TEC MAQ sealer model AP-500 after the seeds were packed.

Storage conditions in cold chamber and uncontrolled environment were tested as well. The seeds were conditioned in a cold chamber, with humidity of approximately 12 to 13%, and, in the uncontrolled environment, they were exposed to normal conditions of temperature and humidity on the bench.

The experiment was performed in a completely randomized design with four replicates. It was applied the factorial scheme and the subdivided plots (8 × 2 × 2 × 7). Eight lines of snap beans and two types of package were used in each plot. The subplot included two environments, and the sub-subplot, seven storage periods (0, 30, 60, 90, 180, 360, and 450 days). After each period of storage, germination and seed vigor tests were conducted.

The germination and vigor tests were carried out following the Rules for Seed Analysis – (*Regras para Análise de Sementes*) RAS (Brazil, 2009). Four replicates of 50 seeds were placed on two sheets of germitest paper and covered with a new one. Rolls humidified with distilled water in the proportion of two and a half (2.5) parts of water for one (1) part of the paper weight were formed and left inside polyethylene bags to preserve their humidity. Subsequently, they were taken to a germinator, which was adjusted to 25°C. The evaluation of normal seedlings was conducted at five and nine days after the test set, and the results were given as a percentage. At the same time as the germination test, the percentage of normal seedlings obtained at the time of the first count of the germination test was also calculated. Results were given as a mean percentage of normal seedlings.

3. RESULTS AND DISCUSSION

After the analysis of data from Figure 1, it was found that there was variation in the water content of the seeds while using different types of package and controlled and uncontrolled environments. The impermeable packages showed almost no variation in the water content of the seeds in the evaluated environments (Figures 1B and 1D). It can be noted that the water content ranged from 12 to 14% among the lines evaluated when using impermeable packages in controlled and uncontrolled environments throughout the storage period (Figures 1B and 1D).

In contrast, the study found that permeable packages presented an exchange between the humidity content of the seeds and the environment under evaluation (Figures 1A and 1C). According to the study, L3 (TOP SEED) and L12 (UENF 7-14-1) exhibited a greater tendency to lose water content over the storage period than other lines, showing that, at 450 days, the loss in humidity content corresponded to approximately 9.8% (Figure 1A).

By using permeable packages under controlled conditions, the exchange between humidity of the seeds and the environment showed less oscillation than the conservation in uncontrolled environments and conditioned in the same type of package (Figure 1C).

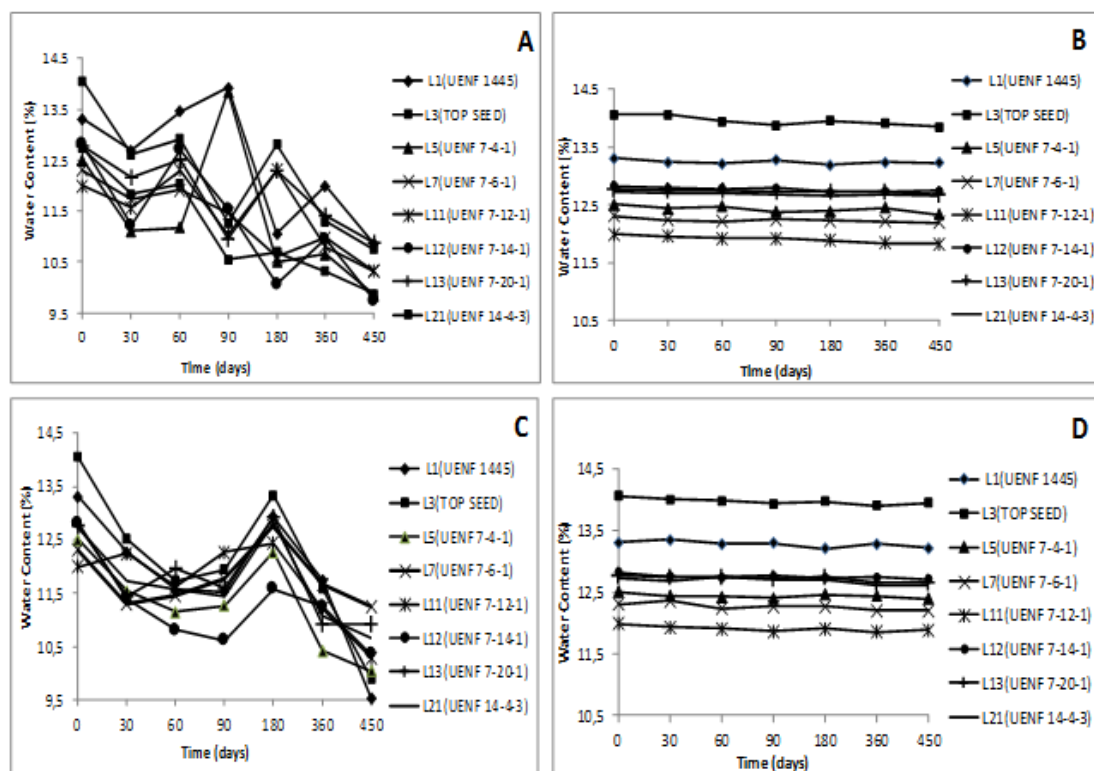


Figure 1. Variation of water content in the seeds of eight snap bean lines conditioned in different types of package and storage period (0, 30, 60, 90, 180, 360, and 450 days). (A) permeable package in an uncontrolled environment; (B) impermeable package in an uncontrolled environment; (C) permeable package in a cold chamber; (D) impermeable package in a cold chamber.

The use of packages is essential to avoid the fluctuation in the degree of humidity of the seeds in conservation, their use reduces their metabolic activity and, hence, the deterioration, extending the maintenance of their physiological quality (Bragantini, 2005). According to De Sena et al., (2018) the type of package has a direct influence on the conservation of the physiological nature of the seeds.

On the basis of the findings, it was evidenced that the humidity content of the seeds stored in the permeable packages had more influence from the conditions of the storage site than the ones stored in the impermeable packages. De Sena et al. (2018) found similar results in their studies as well. This was expected considering that this type of package does not prevent the water vapor exchange between the seeds and the environment where these seeds are stored, differently from the impermeable ones, which do not enable exchanges.

All these results prove that permeable packages do not offer resistance to seeds, which poses a high risk to their physiological quality when submitted to this type of storage. Silva et al. (2010) and Cardoso et al. (2012) reported similar results while studying the physiological potential of seeds in different cultures.

Figure 2 shows the results for the vigor and germination of seeds stored in permeable packages under conditions of uncontrolled environment. The data revealed that, throughout the storage period, a reduction in the vigor occurred for all the evaluated lines (Figure 2). Nevertheless, it appeared that the loss of vigor of the seeds proved to be more severe for lines L5 (UENF 7-4-1), L12 (UENF 7-14-1), L21 (UENF 14-4-3), and L13 (UENF 7-20-1), which had less than 60% vigor during the storage period of 450 days (Figure 2). L3 (Top Seed Blue line), L1 (UENF 1445), L7 (UENF 7-6-1), and L11 (UENF 7-12) were the lines that showed the lowest loss of vigor among the treatments evaluated, being this vigor above 75% at 450 days of storage. Alves and Lin (2003), Skowronski et al. (2007), and Silva et al. (2014) also found a reduction of vigor in bean from the six months of storage onwards.

As regards germination percentage, L1 (UENF 1445), L7 (UENF 7-6-1), L11 (UENF 7-12-1), L5 (7-4-1), L12 (7-14-1), and L13 (UENF 7-20-1) were found to have reduced germination capacity over the storage period. It should be emphasized that all lines showed a germination percentage above 75%, with the exception of line L13 (UENF 7-20-1), which showed a reduction of around 60% in the 450-day storage period (Figure 2).

Regarding the lines evaluated, L3 (Top Seed Blue line) and L21 (UENF 14-4-3) kept the germination percentage close to 100% during the storage period between the 450 days (Figure 2).

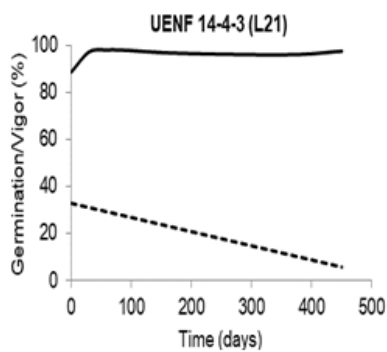
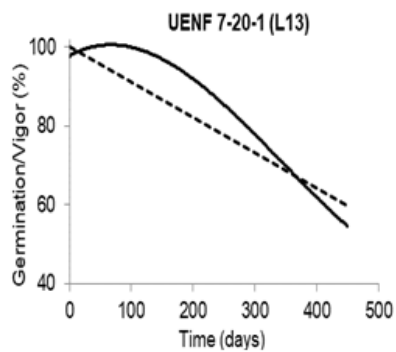
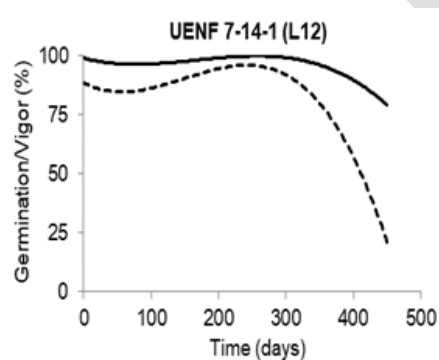
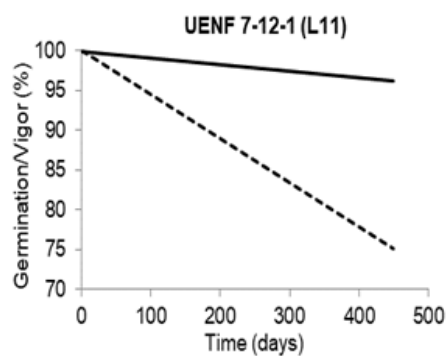
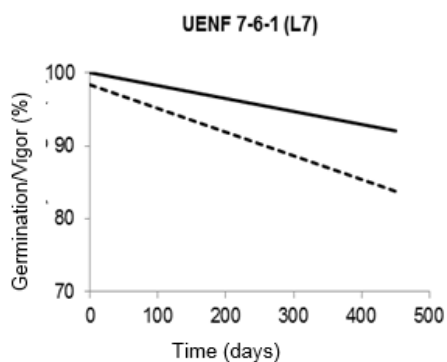
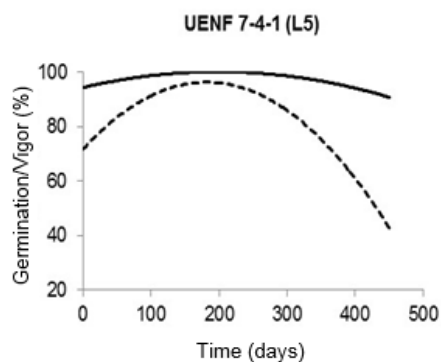
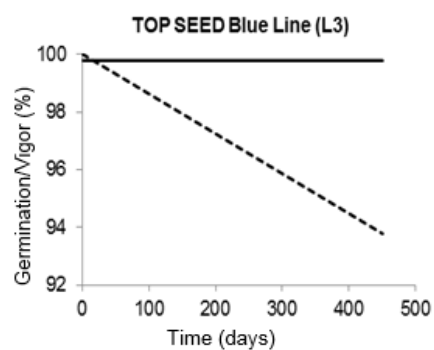
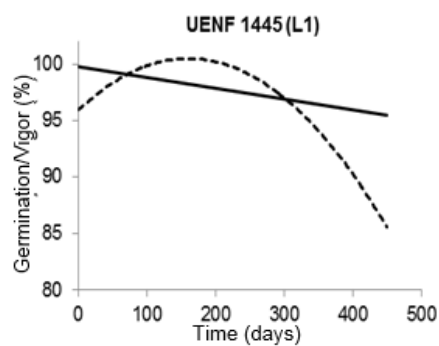


Figure 2. Mean percentage of vigor and germination of seeds of eight snap bean lines stored in a permeable package under conditions of uncontrolled environment over seven storage periods. (0, 30, 60, 90, 180, 360, and 450 days).

Figure 3 illustrates that the seed vigor in the impermeable package under uncontrolled conditions provided a distinct response along storage. After 400 days of storage, a more significant decrease in seed vigor was seen (Figure 3).

It is noteworthy that all treatments evaluated reported over 65% vigor, with the exception of lines L5 (UENF 7-4-1), L7 (UENF 7-6-1), L12 (UENF 7-14-1), and L21 (UENF 14-4-3) that had less than 30% vigor at 450 days of storage (Figure 3).

Nevertheless, lines L3 (Top Seed Blue line), L1 (UENF 1445), L11 (UENF 7-12-1), L5 (UENF 7-4-1), L12 (UENF 7-14-1), and L21 (UENF 14-4-3), in impermeable packages under uncontrolled conditions (Figure 3), showed a percentage of vigor similar to the ones found when they were conditioned in permeable packages and uncontrolled environments. As expected, this indicates that the impermeable packages did not hinder the humidity exchange between the seeds and the external environment.

Furthermore, by analyzing Figure 3, all treatments evaluated demonstrated germination over 90% at 450 days of storage (Figure 3). Considering the results, the impermeable packages, under controlled environments, were efficient in storing the seeds.

De Sena et al., (2018) and Corlett et al. (2007) stated that storing seeds in impermeable packages enables a longer conservation period, since it guarantees the maintenance of an appropriate humidity level, leading to a lower risk of loss of physiological quality by deterioration. In this research, however, it should be inferred that impermeable packages were more efficient in controlling germination when comparing to vigor, with a divergence in the results obtained for vigor in lines L5 (UENF 7-4-1), L7 (UENF 7-6-1), L12 (UENF 7-14-1), and L21 (UENF 14-4-3) (Figure 3).

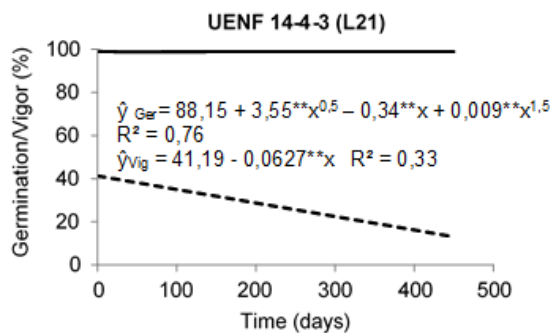
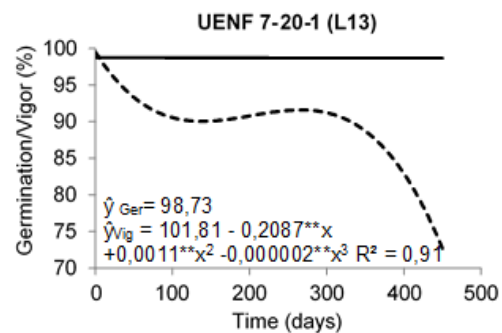
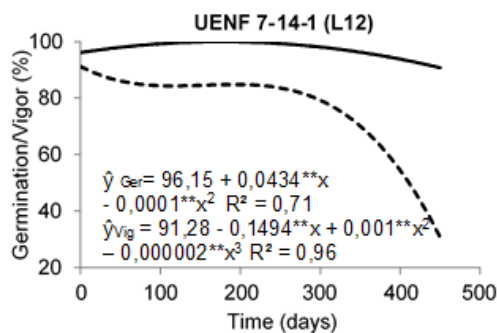
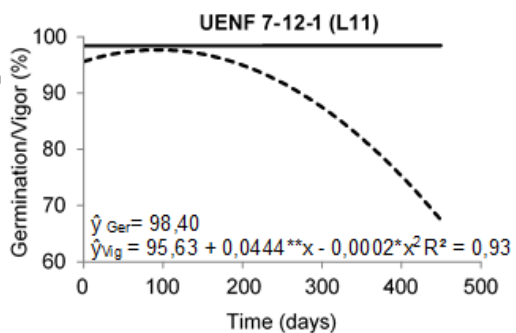
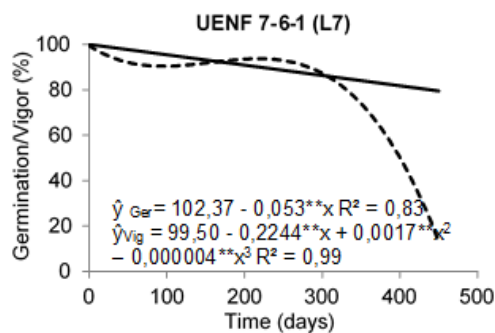
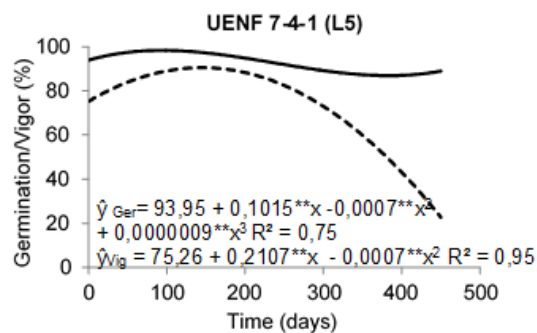
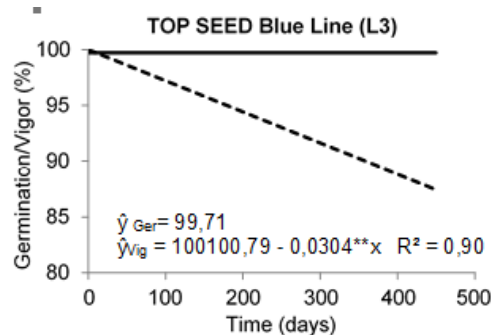
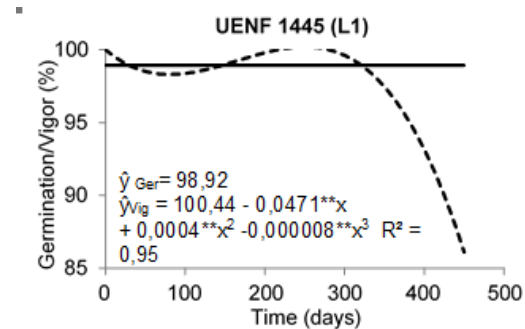


Figure 3. Mean percentage of vigor and germination of seeds of eight snap bean lines stored in an impermeable package under conditions of uncontrolled environment over seven storage periods. (0, 30, 60, 90, 180, 360, and 450 days).

Figure 4 depicts the vigor and germination percentages evaluated in permeable packages under controlled conditions. Clearly, all lines showed vigor above 70%, apart from line L21 (UENF 14-4-3), which showed vigor below 20%. Regarding the germination, it was higher than 90% for all lines evaluated. Furthermore, Figure 4 demonstrates that the germination of lines L11 (UENF 7-12-1), L13 (UENF 7-20-1), and L21 (UENF 14-4-3) was practically constant during the storage period.

Analyzing Figure 4, this study suggested that the seeds presented better physiological quality for storage under controlled conditions than when submitted to uncontrolled conditions. That is in agreement with Figueirêdo et al. (1982), who pointed out that the physiological quality of stored seeds is better when they are kept in environments where the temperature and relative air humidity are under control. Similar results were found by Zucareli et al., (2015) in their study about carioca beans seed.

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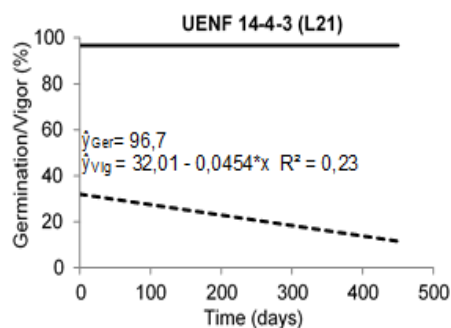
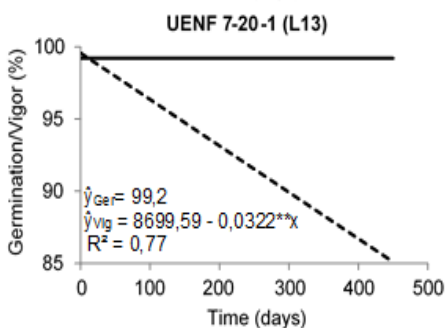
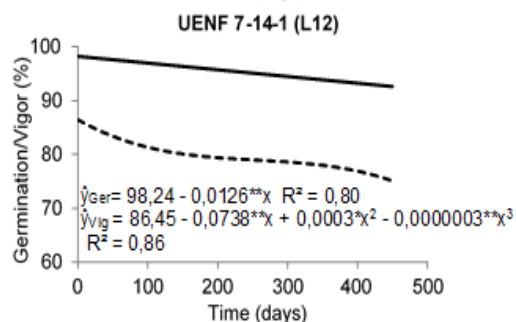
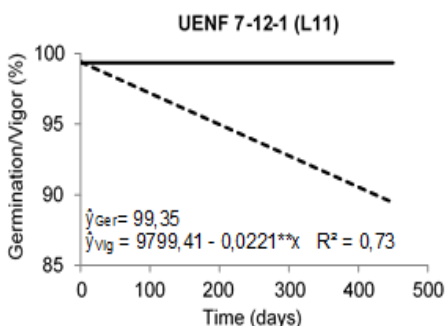
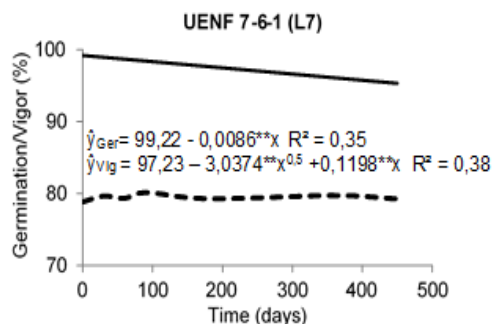
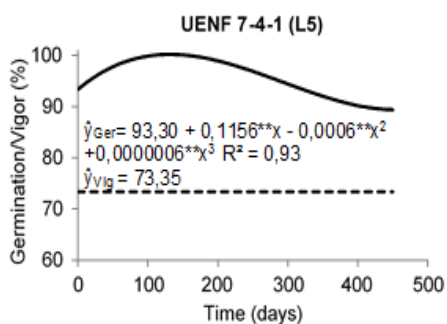
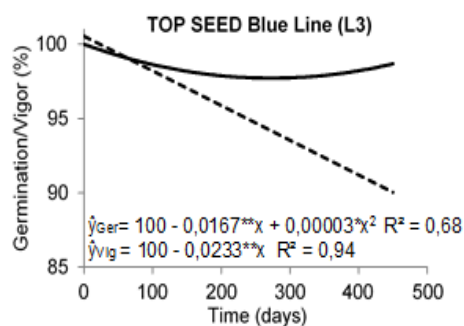
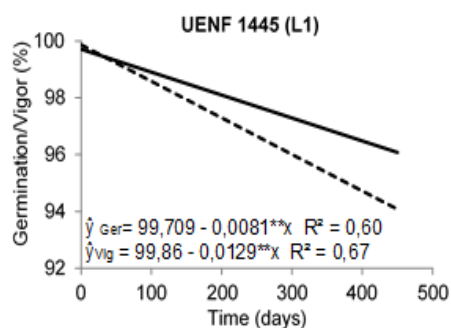


Figure 4. Mean percentage of vigor and germination of seeds of eight snap bean lines stored in a permeable package under conditions of controlled environment (cold chamber) over seven storage periods. (0, 30, 60, 90, 180, 360, and 450 days).

Concerning Figure 5, the vigor percentage was found to be higher than 85% for L1 (UENF 1445), L3 (Top Seed Blue line), L7 (UENF 7-6-1), L11 (UENF 7-12-1), L12 (UENF 7-14-1), and L13 (UENF 7-20-1), all but lines L21 (UENF 14-4-3) and L5 (UENF 7-4-1), which were less than 30% in vigor at 450 days after storage. There was, on the other hand, a stabilization of the germination percentage for all evaluated lines, but only for L5 (UENF 7-4-1). It is important to note that the germination percentage was higher than 98% for all the evaluated lines at 450 days after storage.

When considering Figures 4 and 5, L1 (UENF 1445), L3 (Top Seed Blue line), L11 (UENF 7-12-1), and L13 (UENF 7-20-1) had a similar percentage of vigor and over 85% by using storage in permeable and impermeable packages under controlled conditions. However, the germination percentage also coincided for the L11 (UENF 7-12-1) and L13 (UENF 7-20-1) lines, which stayed constant under the same storage conditions.

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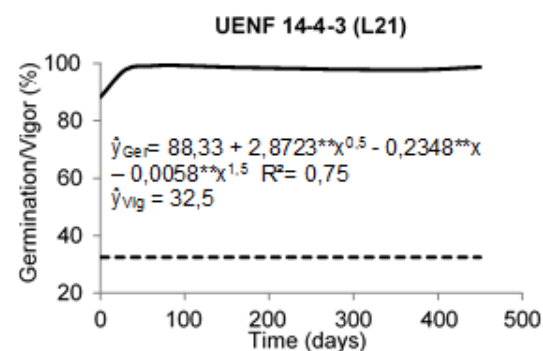
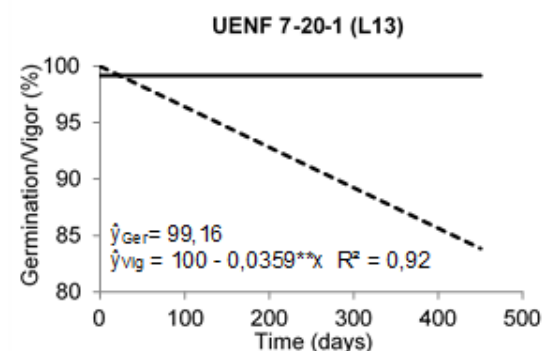
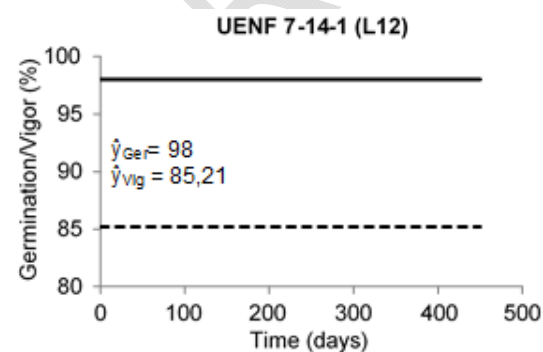
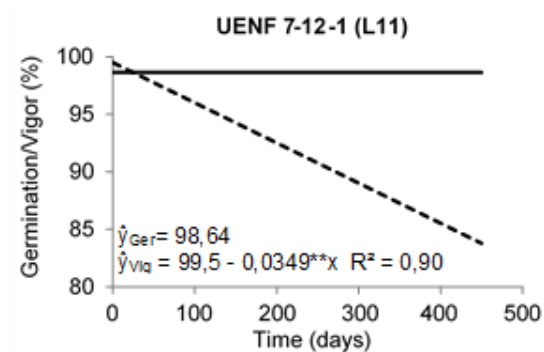
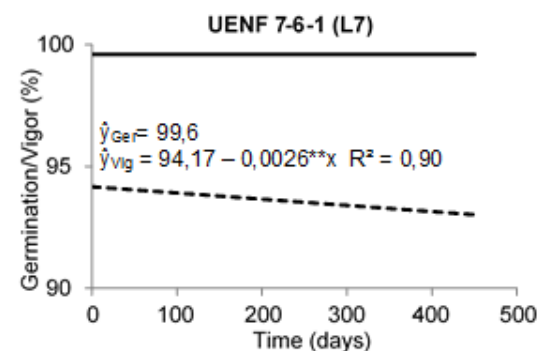
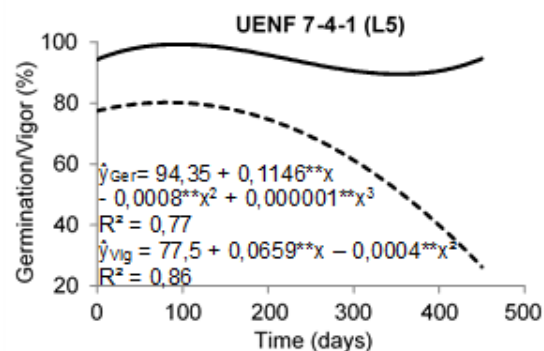
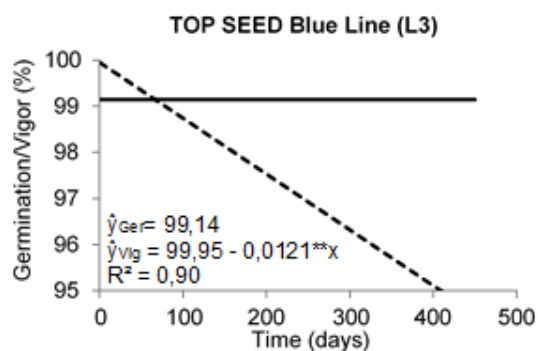
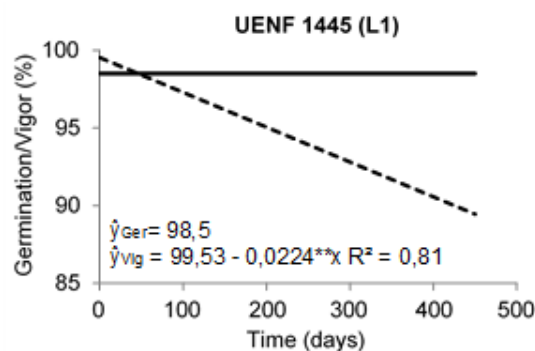


Figure 5. Mean percentage of vigor and germination of seeds of eight snap bean lines stored in an impermeable package under conditions of controlled environment (cold chamber) over seven storage periods (0, 30, 60, 90, 180, 360, and 450 days).

Under controlled environmental conditions, it is possible to keep the temperature low and constant, reducing oxygen and promoting unfavorable conditions for insect attack, as well as reducing the chemical reactions speed, thus improving the seed conservation (Alves and Lin, 2003; Marcos Filho, 2005; Zucareli et al., 2015). Therefore, it can be implied that higher percentages of vigor and germination rates can be achieved under these storage conditions.

4. CONCLUSION

The different types of package and storage conditions affected the physiological seed quality;

The evaluated lines presented variations in vigor and germination when submitted to different storage conditions;

The permeable and impermeable packages, while in controlled storage conditions, proved to be more efficient when it came to controlling the physiological quality of snap bean seeds.

REFERENCES

- Alves AC, Lin HS. Type of packaging, initial moisture and storage period in bean seeds. *Journal Science Agriculture, Piracicaba*. 2003; 4 (1): 21-26.
- Blair MW, Chaves A, Tofiño A, Calderón JF, Palacio JD. Extensive diversity and inter-genepool introgression in a world-wide collection of indeterminate snap bean accessions. *Theor Appl Genet*. 2010; 120(1):1381-1391.
- Bragantini, C. Some aspects of storage of seeds and bean grains. Documents 187. Santo Antônio de Goiás: Embrapa Rice and Beans, 2005.
- Cardoso RB, Binotti FFS, Cardoso ED. Physiological potential of crambe seeds as a function of packaging and storage. *Tropical Agriculture Research*. 2012 42 (3): 272-278.
- CEAGESP - Company of Warehouses and General Warehouses of São Paulo. 2014. Accessed in Aug / 2014. Available in: <<http://www.ceagesp.gov.br/>>
- CEASA: Prohort - Brazilian Program for Modernization of the Horticultural Market 2010. Accessed in Aug / 2014. Available in: <http://www.ceasa.gov.br/precos>.
- Coelho, EB. Influence of seed size on the physiological quality of soybean in different storage periods. Orientation Me. José Eduardo Barbosa de Souza; Goianésia: Faculdade Evangelica de Goianésia. 2018.
- Corlett FMF, Barros ACSA, Villela FA. Physiological quality of urucum seeds stored in different environments and packages. *Brazilian Journal of Seeds*. 2007; 29 (2): 148-158.
- Delouche JC, Baskin CC. Accelerated aging techniques for predicting the relative storability of seed lots. *Seed Science & Technology*. 1973;1 (2): 427-452.
- De Sena MA, Amaral MCA, Ribeiro SO, Castro Filho MN, AS Flag, Morai OM. Influence of the type of packaging on the physiological quality of seeds of lettuce and almeirão during the commercialization. *Cadernos de Agroecologia*. 2018; 13: (1): 1-6.
- Dourado D, Fancelli A. Bean production. Publisher Agropecuária. Rio Grande do Sul, Brazil, 2000.
- FAO – Food and Agriculture Organization Faostat. 2014. Accessed in Jan/2014. Available in: <<http://faostat.fao.org.>>.
- Page 1 New manual of olericultura: modern agro-technology in the production and commercialization of vegetables. Viçosa: UFV, 2013.
- Figueiredo F J C, Frazão D A C, Oliveira R P, Carvalho J E U (2005). Conservation of cowpea seeds. Belém: EMBRAPA-CPATU, pp.1-23.
- Marcos Filho, J. Seed physiology of cultivated plants. Piracicaba: Fealq. 2005.
- Marcos Filho J. Strength test: importance and use. In: Krzyzanowski, F.C .; Vieira, R.D .; France Neto, J.B. (Ed.). Seed vigor: concepts and tests. Londrina: Abrates, 1999.
- Miklas PN, Coyne DP, Grafton KF, Mutlu N, Reiser J, Lindgren DT, Singh SP. A major QTL for common bacterial blight resistance derives from the common bean great northern landrace cultivar Montana. *Euphytica*. 2003;131 (5): 137–146.

Oliveira FTG, Victory RZ, Posse SCP, Arantes SD, Schmildt O, Viana A, Malikouski RG, Barros BLA. Physiological quality of aroeira seeds under the storage conditions. Nucleus, 2018: 15 (2): 567-573.

Popinigis F. Seed physiology. 2.ed. Brasília: AGIPLAN, 1985.

Silva FS. Feasibility of storing seeds in different packages for small farms. Agro-environmental science journal. 2010: 8 (1): 45-56.

Silva MM. Physiological quality and storage of common bean seeds produced in northern Minas Gerais. Rev Agro ambien Online. 2014: 8 (1): 97-103.

IBGE Automatic Recovery System (SIDRA). 2006. Accessed in Oct / 2012. Available in: <http://www.sidra.ibge.gov.br/bda/tabela/listabl.asp?z=t&o=19&i=P&c=818>

Skowronski L, Giudice MPD, Borém A, Carne GES, DCFS Days, Pinto CMF, Vieira R F, Venzon M, Paula JRTJ, Mattos RN. Bean pod (*Phaseolus vulgaris* L.) in: Paula J R & Venzon M (Coord.) 101 Cultures: Manual of agricultural technologies. EPAMIG, Belo Horizonte, MG, 2007.

Smith MT, Berjak P. Deteriorative changes associated with the loss of viability of stored desiccations-tolerant and desiccation-sensitive seeds. In: G. Galili G & J.Kigel (eds). Seed development and germination. Marcel Dekker Inc., New York: 1995.

Zucareli C, Brzezinski CR, Abati J, Werner F, Júnior EUR, Nakagawa J. Physiological quality of carioca bean seeds stored in different environments. Brazilian Journal of Agricultural and Environmental Engineering. 2015: 19 (8): 803-809.

DEFINITIONS, ACRONYMS, ABBREVIATIONS

Term: Agronomic, post harvest.

Supplementary Table 1. Mean squares, averages and coefficients of experimental variation of germination of eight genotypes of beans in different boxes and environments, depending on the of the storage period

Source of variation	d.f.	Mean sum of squares	
		Vigor	Germination
Lin	7	59140.85**	230.20**
Pac	1	91.29	180.36**
Pac x Lin	7	632.85**	122.17**
Error A	45	31.62	11.08
Env	1	1405.00**	415.29**
Env x Lin	7	261.71**	138.91**

Env x Pac	1	136.72*	20.04
Env x Pac x Lin	7	364.73**	151.05**
Error B	45	19.81	5.91
Tim	6	11305.35**	520.26**
Tim x Lin	42	846.17**	107.61**
Tim x Pac	6	624.68**	84.04**
Tim x Env	6	2268.59**	188.80**
Tim x Env x Lin	42	326.29**	61.83**
Tim x Pac x Lin	42	416.51**	58.29**
Tim x Pac x Lin x Env	42	533.12**	142.62**
Error C	588	38.08	10.34
Average		79.96	97.22
Coefficient of variation %		7.47	3.19

Lin= Lines; Pac= package; Pac x Lin = package and lines interaction; Env= Environment; Env x Lin= environment and lines interaction; Env x Pac = Environment and package interaction; Env x Pac x Lin = Environment package and lines interaction; Tim= Time; Tim x Lin= time and lines interaction; Tim x Pac = time and package interaction; Tim x Env= time and environment interaction; Tim x Env x Lin; time environment and lines interaction; Tim x Pac x Lin =time package and lines interaction; Tim x Lin x Pac x Env= time lines package and environment interaction. ** The mean difference is significant at the .01 level by test F.; * The mean difference is significant at the .05 level by test F.