Beans quality during storage with different carnauba wax concentrations

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Abstract: The objective of this study was to evaluate the use of carnauba-based wax to preserve the quality of bean grains. Three lots of beans of 25 kg each were used. The first remained without carnauba wax (treatment 0), the second with carnauba wax diluted with water in a 1:1 proportion (treatment 1), and carnauba wax without dilution (treatment 2). Sensory analysis, color, cooking time, absorption capacity, and insect-plague infestation were performed during 8 months of storage. Carnauba wax does not compromise the sensorial quality and color of the beans. Thus, it can be used without interfering with the products acceptance by the consumer. Carnauba wax did not prevent the *hard-to-cook* damage, which the consumer associates with a longer period of cooking time. Moreover, it was not efficient in controlling the infestation of *Acanthoscelides obtectus* insects. The water absorption by beans was influenced by wax application, storage period, and hydration temperature.

Keywords: Acanthoscelides obtectus (say), color, cooking time, sensory analysis, water absorption

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1 Introduction

The quality of beans may be determined by consumption acceptability, according to certain characteristics such as coat color, hydration, and

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cooking time. These characteristics may be altered during storage because of inadequate conditions or insect presence (Resende et al., 2008).

An important damage that decreases grain acceptability is the hard-to-cook defect (Guzman-Mendez et al., 2014; Segura-Campos et al., 2014), which increases the time required to cook the beans and obtain the softening of the cotyledon. This process decreases the nutritive value of the beans owing to loss of vitamins, in addition to the loss of texture and flavor due to higher period of cooking. This defect is a consequence of a long storage period at high

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temperature (> 25 $^{\circ}$ C) and relative humidity (> 65%) (Ruiz-Ruiz et al., 2013).

Along with the hard-to-cook defect, inadequate storage allows the infestation of insects. Among the insect species that infest beans during storage, the *Acanthoscelides obtectus* (Say) is important to consider. This plague is found in tropical and temperate regions, and the insects are capable of opening holes in the grain, leading to the deterioration of the beans.

To control insects, residual insecticides and fumigation have been widely used for years and are highly efficient (Velasques et al., 2017). However, years of use has resulted in the selection of more resistant insects, and this increasingly difficult the control of the insect population with these methods. Thus, to mitigate this problem, new techniques to control insect population must be developed.

A currently used alternative solution is the use of carnauba wax. It is widely used in fruits, vegetables, cocoa and chocolate products, bakery products, coffee, tea, among other products (Freitas et al., 2019), to maintain or decrease the quality loss of the product during storage. The use of carnauba wax reduces water loss, provides a more attractive aspect to the product after drying and polishing (Mattos et al., 2017). It provides different permeability rates to O₂, CO₂, and water vapor as a function of the raw material properties, concentration, and thickness of the epidermis (Yousuf et al., 2018).

Being that stated, this study aimed to verify the effect of carnauba wax in the preservation of beans during post-harvest, assessing a sensorial analysis, color, cooking time, absorption capacity, and insect growth.

2 Material and methods

2.1 Raw material

Beans of the cultivar "BRSMG Majestoso," from the "carioca" group were used. They were manually harvested at the experimental field of the Agricultural Research Company of "Minas Gerais" state (Epamig), located at Oratórios city. Later, fumigants were applied to the grains, with the goal of avoiding infestation from the cultivation period.

The grains presented an initial moisture content of 0.1481 d.b. (dry basis). Three lots of 25 kg each were used. One of the lots was the control group (without use of carnauba wax); to the second lot, carnauba wax diluted with water at the proportion of 1:1 was applied; and the third lot, carnauba wax without dilution was used. The treatments were denoted as treatments 0, 1, and 2, respectively.

The application of the carnauba wax solutions was made through pulverization, with the grain mass homogenized to guarantee that application of the solution covered the surface of the entire lot of the beans. Afterwards, beans were spread at suspended yards for drying purposes. The moisture content of beans from treatments 1 and 2, after carnauba wax use were, respectively, 0.1547 and 0.1484 (d.b.).

2.2 Storage of beans treated with carnauba wax

After drying, the grains were stored in cotton bags, and every 30 days for 8 consecutive months, samples of 1 kg were retrieved to conduct analyses.

The temperature and relative humidity of the storage environment were measured daily at 9 a.m. by means of a mercury thermometer and a strand of hair hygrometer. The moisture content of the grain was determined by the gravimetric method, using an oven at $105 \ C \pm 3 \ C$ during 24 h, threefold ^{(Ministry of Agriculture, Livestock and Food Supply [MAPA], 2009).}

2.3 Sensorial analysis

The sensorial analysis used 350 g of grain from each treatment. The beans were put in vessels previously identified with 1 L of boiling water. Later, they were covered and only one small passage was kept for liberating vapor. The gas injectors were regulated for low flame to provide a uniform intensity of flames.

After the beans were cooked, the vessels were put on a table to cool down. Afterwards, the grain/broth proportion was standardized. To do so, a Petri dish with height of 1 cm was used. It was kept at horizontal position, placed in the vessel, and pushed down, making the broth to go up. When the broth did not reach the dish height, more water was added up until this trend was attained.

Furthermore, the broth was separated from grain, and the grains were weighted. Based upon their weight, the amount of salt and vegetal oil was calculated, and the sensorial analysis was accomplished. Samples were served in a dish of black bottom, coded, and were evaluated by 40 trained and selected experts, using the hedonic scale of 9 points (Associa ção Brasileira de Normas T écnicas [ABNT], 1998). This procedure was carried out throughout eight months of storage.

2.4 Colorimetric parameters

A *tristimulus* chromameter (MiniScan XE Plus 45/0-L, HunterLab, Reston, USA) was used to measure the grain color during storage. The Hunter-Lab scale and illuminant 10 9D65 were used. Using the L^* , a^* , and b^* coordinates, the colorimetric indices chroma (C^*) (Equation 1) (Subhashree et al., 2017), which defines intensity and purity of a color, and hue angle (h^*) (Equation 2) (Subhashree et al., 2017), which defines color (0 ° as red, 90 ° as yellow, 180 ° as green, and 270 ° as blue), were calculated. ΔE , which denotes the total color difference when compared to the initial conditions of storage (Equation 3), was also acquired (Subhashree et al., 2017).

$$C^* = (a^{*2} + b^{*2})^{1/2} \tag{1}$$

$$h^* = \tan^{-1} \frac{b^*}{a^*}$$
 (2)

$$\Delta E = (\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2})^{1/2}$$
(3)

The darkening index (*BI*) of grain (Equation 4) (Subhashree et al., 2017), was also calculated:

$$BI = [100(x-0.31)]/0.172 \tag{4}$$

In which:

$$x = \frac{a^* + 1.75L^*}{5.645L^* + a^* - 3.012b^*}$$
(5)

2.5 Cooking time

To determine cooking time, the samples were previously imbibed in water for 14 h before cooking. This test was made using the Mattson equipment. The beans were placed inside a metallic vessel with 1 L of boiling water, completing this volume from time to time to maintain the volume constant. The beans were considered cooked when a rod passed through them, and the cooking time was registered when most of the grains (52%) were cooked, i.e., when the 13th rod penetrated the bean.

2.6 Water absorption capacity

The determine the amount of water absorption, 20 g of beans from each treatment were imbibed in 80 mL of distilled water, attaining a mass proportion of 1:4, during 12 h at temperatures of 20 °C, 30 °C, 40 °C, and 50 °C. After the imbibition period, the samples were put in filter papers to remove superficial water, during 2 min. The percentage of water absorption was determined by Equation 6 (Coelho et al., 2008):

$$A_{g} = \frac{m_{e} - m_{i}}{m_{i}} \times 100 \tag{6}$$

where, A_g is the water absorption, %; m_e is the grain mass after imbibition, g; and m_i is the grain mass before imbibition, g.

2.7 Insect growth

The efficiency of carnauba wax to control insect growth was verified by infestation tests. 1 kg of beans, taken from each treatment, were placed in plastic pots with capacity of 2 L. These pots had perforated lids shut with organza, to assure oxygen entrance and forbid Fifteen insect exit. adult insects from the Acanthocelides obtectus (Say) species were placed inside each pot, and later stored at sites with temperature and relative humidity adequate for insect development. Three pots from each treatment were infested, and analyses of insect contact with the grain were made at 45, 90, 135, 180, and 225 days of storage. The percentage of infestation was evaluated according to previous methodology (MAPA, 2009).

2.8 Statistical analysis

The experiment was made according to a split-plot arrangement. The main plots were the treatments, and the split-plots were the storage periods. A randomized complete block design was employed, with the number of replicates varying according to the analysis made.

The experimental data was submitted to variance and regression analyses. Models were chosen based on the significance of regression coefficients using the "t" test, adopting a 5% level of probability, based on the determination coefficient and biological phenomenon.

3 Results and discussion

3.1 Storage of beans treated with carnauba wax

Figure 1 presents the values of temperature and relative humidity of air per month, and the moisture content of beans during storage.



(a) Temperature and relative humidity of the air

Figure 1 Mean values of temperature and relative humidity of the air (a) and moisture content of beans during storage(b)

variations Small were observed in the psychrometric properties of the air that surrounds the storage facility, characterized in Figure 1a by temperature (average of $26.8 \, \text{C}$) and relative humidity (average of 74%).

It can be observed in Figure 1b that the moisture content of the beans decreased throughout storage. However, for treatment 0, the moisture content presented slight oscillations from the beginning of storage until 6 months of storage. Beans with carnauba wax 1:1 (treatment 1) variations occurred throughout storage. This trend may be explained by the amount of water used to produce the carnauba wax solution, which may be present at the beans surface, increasing moisture content of these beans during the first 5 months of storage. Later, it decreased close to the values of beans from treatment 2, which decreased the moisture content in a continuous and smooth manner.

This trend happened because carnauba wax is a product with low solubility in water, generates a barrier effect on gas exchange, mainly water vapor, CO₂, and O₂, and thus, interferes in the mechanisms of water sorption between the product and air of the storage environment.

The highest value of moisture content was 0.1547 (d.b.), for treatment 1, while the lowest value was 0.1106 (d.b.), for treatment 2. The alterations in moisture content are explained by the ability to desorb or absorb water, i.e., hygroscopicity, and to reach an equilibrium state between moisture content of the product and the conditions of the environment.

3.2 Sensorial analysis

Figure 2 presents the results from the sensorial analysis.

It was observed that in all treatments, most of the experts classified the grains at the highest levels of acceptability of the hedonic scale (Figure 2). Treatment

2 received the highest grades among the three treatments, with a higher percentage of expert values at higher levels of acceptability. However, the Dunnett test indicated that there were no significant differences between the three treatments regarding sensorial acceptability, at a level of 5% of probability. Thus, the obtained result indicates that it is possible to use the carnauba wax to maintain the quality of beans during storage, without losing the sensorial aspects which are important to commercialization.



Figure 2 Sensorial analysis of the beans submitted to treatments 0, 1, and 2, stored for 8 months

3.3 Colorimetric parameters

The effects of the treatments used and storage on the chroma, hue angle, color difference, and darkening index of beans, are presented in Figures 3 and 4.



Figure 3 Chroma and hue angle of beans as a function of carnauba wax concentration and/or storage time Note: *significant at 5% of probability; **significant at 1% of probability



Figure 4 Color difference and darkening index of beans as a function of carnauba wax concentration and/or storage time Note: *significant at 5% of probability; **significant at 1% of probability

From Figure 3a, it is observed that the chroma values increased linearly with storage and quadratically with wax concentration. From the fitted model, it is verified that the maximum point is located near to a concentration of 59%, and afterwards it diminishes. Among the observed data, the highest value was 21.17 at 240 days of storage for beans from treatment 1.

There is a significant interaction between hue angle and storage time (Figure 3b), denoted by a decrease in hue angle values throughout storage. The mean values of hue angle at 0 and 240 days of storage were 56.50 and 50.64, respectively. Thus, there is a difference between the initial and final stages of storage, altering the tonality of the brown color.

Based on Figure 4a, the color difference increased linearly with storage, while it decreased with the wax concentration applied on beans. Considering the initial conditions of storage, the highest color difference was 10.19, at 210 days of storage, in the beans from treatment 0.

The application of carnauba wax, which has a dark brownish color, contributed to the darkening of beans, and it increased with storage (Figure 4b). The model of darkening index satisfactorily represented the influence of storage and treatment on beans from the "carioca" group. This conclusion was made using the "t" test at 1% of probability and an elevated determination coefficient. Darkening of the bean tegument during post-harvest is an undesirable characteristic for commercialization, which leads to economic losses to the bean producers, distributors, and exporters (Elsadr et al., 2011).

A combination of environment and genetics leads to chemical alterations within the grain, leading to the darkening effect. It is accelerated when beans are exposed to light, high temperature, moisture, and storage period (Yousif, 2014). Rios et al. (2002) concluded that darkening of beans during storage could be a consequence of an increase in activity of the polyphenol oxidase enzyme, associated with the peroxidase enzyme and the increase of phenolic compounds. They also verified that the "carioca" cultivar presented lower enzymatic activity during storage, and therefore, it obtained the lowest darkening index. Previous research attributed the darkening of beans from the "carioca" group, stored under noncontrolled conditions, to temperature and light (Brackmann et al., 2002). In beans from cultivars with

colorful integument, including the "carioca" group, possess a high tannins content, which may be oxidized by catechol oxidase under presence of oxygen (Luh and Phiithakpol, 1972).

3.4 Cooking time

From Figure 5, it is possible to verify the effect of storage over the cooking time of beans.



Figure 5 Cooking time of beans as a function of storage time

Note: *significant at 1% of probability

The linear model satisfactorily represented storage time against cooking time, presenting elevated values of determination coefficient and significance level. This behavior was also reported (Resende et al., 2008). Cooking times of 18.7 min were observed for beans from treatment 0 at the beginning of storage and 58.4 min for beans from treatment 2, after 240 days of storage. The increase in cooking time is an undesirable characteristic for bean quality.

Based on studies with fruits (Malgarim et al., 2007), the use of carnauba wax resulted in a metabolism reduction of the product, leading to lower loss of water. Thus, it was expected that the application of carnauba wax would lead to a decrease in the cooking time of beans, when compared to the control beans, during storage. However, this trend was not observed, because hardening of cotyledon was observed, which is characteristic of a damage known as hard–to–cook. The resistance of beans to softening during cooking can be attributed to several mechanisms such as lipid oxidation, formation of insoluble pectate, and lignification of the middle lamella (Liu, 1995). Higher resistance of beans to softening though cooking is due to the shell that acts as a barrier between the cotyledon and maceration water (Castellano et al., 1995). Beans absorbs water during maceration process, but do not achieve the adequate softening level, even after a reasonable cooking period.

The behavior of different genotypes of beans from the commercial "carioca" group was evaluated (Lemos et al., 2004). They reported cooking times ranging from 15 to 25 min for beans recently harvested. They concluded that seedlings in different periods affect the hydration characteristics and cooking time.

The effect of water temperature over cooking capacity was evaluated and tested the hypothesis that hydration could be indicative of reduced cooking time for beans (Coelho et al., 2008). These authors studied five different cultivars and concluded that is impossible to standardize the previous conditions to cooking time without a more detailed characterization of each cultivar.

3.5 Water absorption capacity

Figure 6 presents the observed and estimated values



(a) Function of storage (b) Water temperature

Figure 6 Percentage of water absorption of beans as a function of storage and water temperature

It can be observed, from Figure 6a, that the beans from treatment 0 presented a higher capacity of water absorption when compared to the remaining treatments, at the beginning of storage. This trend can be related to the use of carnauba wax. When storage period increased, absorption between treatments presented closer values. This fact may be because of the loss of carnauba wax effect due to environmental conditions. The carnauba wax is slightly soluble at room temperature (Freitas et al., 2019).

Higher water absorption capacity by beans has been related, in a direct form, to lesser cooking time, and this characteristic is very important for bean commercialization (Delfino and Canniatti-Brazaca, 2010). However, this relationship is not a rule, as stated by the present research and previous studies (Resende et al., 2008; Giuberti et al., 2015). The increase in water temperature caused an increment in the capacity of the beans to absorb water (Figure 6b). This result was also found by Coelho et al. (2008), who verified a significant increase in hydration capacity with the increment of water temperature for beans from the cultivars "pérola" and "rubi."

Esteves et al. (2002) indicated that polyphenols are responsible for bean hardening, which is caused by two mechanisms: polymerization of the bean skin or lignification of the cotyledons, both affecting the hydration capacity of the seeds; the first one makes water penetration difficult, and the second mechanism limits the hydration capacity.

A regression equation describing the variation of water absorption with storage could only be fitted for treatment 1 (Table 1). For the remaining treatments, regression equations were fitted with water temperature.

Table 1 Regression fitted to experimental values of water absorption capacity (Ag) of beans as a function of storage time (t) and water temperature (T), determination coefficient, and their respective significant levels

Identification	Fitted model	\mathbb{R}^2
Treatment 0	$\hat{A}_{g} = 102.389^{*} - 0.431^{*}T + 0.008^{*}T^{2}$	0.8685
Treatment 1	$\hat{A}_{g} = 94.075^{**} + 0.028^{**}t$	0.8338
	$\hat{A}_{g} = 99.246^{*} - 0.270^{*}T + 0.006^{*}T^{2}$	0.9761
Treatment 2	$\hat{A}_{g} = 99.024^{*} - 0.362^{*}T + 0.007^{*}T^{2}$	0.9992

Note: **significant at 1% of probability by the "t" test; *significant at 5% of probability by the "t" test.

3.6 Insect growth

Owing to the significant losses of beans during storage caused by insects, it is necessary to find alternatives to control the development and attack of insects. Studies used natural insecticides such as inert powder and essential oil from vegetal origin as effective ways of control (Cao et al., 2018; Tacoli et al., 2018; Campolo et al., 2014).

Similarly, it was expected that the application of carnauba wax on beans would form a pellicle that would prevent the oviposition of *Acanthoscelides obtectus* insects at the cracks in beans. This trend would

decrease the population of insects within the beans mass and thus eliminating the infestation. However, it was not observed a significant interaction between treatments and infestation levels; it was observed an exponential interaction with storage. Until the first 135 days of storage, the model could describe the phenomenon. Afterwards, the infestation level decreased in an abrupt manner, reaching zero at treatment 2 (Figure 7a), because the conditions of the stored beans were not adequate, i.e., their moisture content was elevated (Figure 7b). It was verified an increase in temperature and presence of fungi, which are proper conditions to beans and insect death.



Figure 7 Infestation level and moisture content of infested beans during storage

Note: **significant at 1% of probability by the "F" test

4 Conclusions

From the results obtained in the present work, it can be concluded that carnauba wax did not compromise the sensorial quality and color of the beans, and thus, it can be used without interfering with the product acceptance by the consumer. On the other hand, carnauba wax did not prevent the *hard-to-cook* damage of beans, which the consumer associates with a longer period of cooking time. Furthermore, regardless of the carnauba wax concentration, it was not efficient to control the infestation of *Acanthoscelides obtectus* insects. Water absorption by the beans was influenced by wax application, storage period, and hydration temperature.

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