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Hermetic storage for control of common bean weevil, *Acanthoscelides obtectus* (Say)

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ABSTRACT

This study evaluated hermetic storage as a method of controlling *Acanthoscelides obtectus* (Coleoptera: Chrysomelidae: Bruchinae) in stored beans. Recently harvested "vermelhinho" cultivar of the common red bean was used, which had already been infested by *A. obtectus* in the field. Beans with a moisture content of 15.0% wet basis were stored in silo bags (3 kg), plastic bottles (1.5 L), or non-hermetic glass containers (3 L) (control) for 120 days. The packages were stored in an acclimatized chamber at 25 °C with a relative humidity of 70 ± 5%. At time intervals of 0, 30, 60, 90, and 120 days, three packages of each treatment were opened, and analyses were performed to assess the infestation percentage by insect pests, moisture content, density, electrical conductivity, germination percentage, and cooking time. There was no increase in infestation by *A. obtectus* in the grains stored in the silo bags and plastic bottles during the 120 days of storage; however, there was a significant increase in infestation in the grains in non-hermetic storage (control). The quality of the beans correlated with infestation; it was not altered in the hermetic storage systems and decreased in the control sample. Hermetic storage of common beans is an effective tool in the control of *A. obtectus*.

1. Introduction

The common bean *Phaseolus vulgaris* (L.) is one of the most commonly used vegetables in human nutrition worldwide and is one of the main sources of protein, particularly in developing countries (Jones et al., 2011; Lopes et al., 2015). However, attack by bruchids (Coleoptera: Chrysomelidae: Bruchinae) during storage compromises the quality and commercial value of beans. The bruchid *Acanthoscelides obtectus* (Say) is one of the major insect pests affecting the common bean (Hagstrum and Subramanyam, 2009; Mutungi et al., 2015).These insects cause damage by reducing the mass and/or volume, reducing the physiological quality and germination capacity, and increasing the temperature and water content of the grains (Padín et al., 2002; Faroni and Sousa, 2006).

Control of insects is accomplished through the use of protective insecticides, pyrethroids, organophosphates, and aluminum phosphide fumigant (Corrêa et al., 2011; Pimentel et al., 2012). However, the continuous and indiscriminate use of insecticides for the treatment of grains has been questioned around the world. There is a growing social concern regarding residual pesticide levels in foods and the risks that they pose to consumers' health. Moreover, there is also the risk that insect pests will develop resistance to these insecticides (Guedes et al., 2006, 2009). Thus, there arises the need for studies on alternative protection and conservation techniques of stored products. The use of hermetic storage plays an important role in integrated pest management (IPM) because the system emphasizes the treatment of products without leaving pesticide residues (Carvalho et al., 2012; Navarro, 2012a).

The hermetic storage of grains has been used since ancient times in an attempt to preserve grains (Adler et al., 2000). The technology is based on the creation of storage environments that are unfavorable to pests by means of one of the following methods: vacuum hermetic fumigation, gas hermetic fumigation, or biogenerated modified atmosphere (Anankware et al., 2012; Navarro, 2012b). Countries such as Australia, Brazil, and Argentina have adopted a system of hermetic storage using polyethylene silo bags. The silo bag is manufactured in high-density polyethylene in three layers: two black internal layers and one white external layer made of titanium dioxide. Once the product is wrapped and the bag is closed, the level of oxygen within the bag drastically falls as a result

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of grain, insect, and fungus respiration. On the other hand, the level of carbon dioxide increases (Hell et al., 2014; Njoroge et al., 2014). Consequently, insects stop feeding, becoming inactive and eventually dying of asphyxiation (Moreno-Martinez et al., 2000) or desiccation (Murdock et al., 2012).

In rural areas of developing countries, hermetic storage is usually the only efficient method of safe grain storage. This system does not pose significant costs to the farmer because recycled or reused packaging can be used for storage (Quezada et al., 2006). In Brazil, beans are normally stored in polyethylene terephthalate (PET) bottles. In addition to a lower cost, the use of recycled packaging allows farmers to securely store the product and maintain it during the off-season and/or during periods of falling prices (Freitas et al., 2011). Although hermetic packages are widely used, there is a paucity of information in the literature on the hermetic storage of the common bean in silo bags. It is important to note that it was recently reported that the proliferation of *A. obtectus* did not occur in beans stored in hermetic Purdue Improved Crop Storage (PICS) bags over a period of six months (Mutungi et al., 2015). Additionally, Jones et al. (2011) reported evidence that hermetic technologies can be effective against key plagues of the common bean, incurring advantages in net earnings. Thus, the objective of this study was to evaluate the hermetic storage of the common bean in silo bags and plastic bottles for the control of *A. obtectus*.

2. Material and methods

The experiment was conducted in the Pre-Processing and Storage of Agricultural Products Sector of the Department of Agricultural Engineering, Universidade Federal de Viçosa (UFV). The "vermelhinho" cultivar of the common red bean was used, which was acquired from a bean producer in the municipality of Viçosa, MG, Brazil. The beans exhibited the following qualitative characteristics: infestation by insect pests = 1.0%, moisture content = 15.0% wet basis (w.b.), bulk density = 761.4 kg m^{-3}, electrical conductivity = 72.9 μS cm^{-1} g^{-1}, germination percentage = 100% and cooking time = 35.7 min. It is important to note that the beans had been recently harvested and were already naturally infested by *A. obtectus*, making artificial infestation unnecessary.

The beans were packed in polyethylene silo bags and polyethylene terephthalate (PET) bottles. The bags had a capacity for 3 kg and were made of the same 250-μm thick plastic used for the manufacturing of Silox TM (DuPont) silo bags. The bags were made of a three-layer plastic and were black on the inner side and white on the outer side with UV stabilizers. The plastic layers are a mixture of high dense (HDPE) and low dense polyethylene (LDPE). The plastic bottles were reused transparent soda bottles with a capacity of 1.5 L and thickness of 270 μm.

The bags were hermetically sealed with a multi-use sealing machine (hot bar 40/60 cm), and the bottles were properly sealed with a screw cap. For the control treatment, the grains were placed in transparent glass containers with a capacity of 3.0 L and closed with an organza fabric. The organza cloth was used to allow gas exchange between the ambient and inter-granular atmospheres, as well as to prevent the exit or entry of insects into the containers.

The packages were maintained in an acclimatized chamber at 25 °C with a relative humidity of 70 ± 5% for 120 days. At time intervals of 0, 30, 60, 90 and 120 days, three packages of each treatment were opened, and analyses were performed to assess the degree of infestation by insect pests, moisture content, density, electrical conductivity, germination percentage and cooking time.

To assess the percentage of grains damaged by insects, three samples of 100 beans were taken randomly from each package and immersed in water for 24 h. After this period, the beans were removed from the water, dried on filter paper, cut and examined individually. Grains considered infested were those containing young or adult insects and/or exit orifices of insect pests, according to recommendations of the Rules for Analysis of Seeds (BRASIL, 2009).

Moisture content was determined using the oven method according to the norms of the ASAE (2004), which prescribes the use of a forced air oven at 103 ± 1 °C for 72 h. Three replicates with 30 g of grain were used for each experimental unit. Results were expressed as a percent of wet basis. Determination of the bulk density was performed with the aid of a hectoliter balance with a capacity of one quart (250 mL) using clean grain. Five consecutive readings were performed for each sample, selecting the three closest readings (BRASIL, 2009).

Electrical conductivity of the solution containing the beans was measured using the Cup System or Mass Conductivity (Vieira et al., 2001). The tests were conducted with three replicates of 50 grains randomly taken from each treatment. The grains were weighed and placed in plastic cups (200 mL), to which 75 mL of deionized water was added. The grain samples were then maintained in a climatic chamber at 25 °C for 24 h. After this period, the electrical conductivity measurements of the solution containing the grains were performed.

The germination percentage was obtained in accordance with the Rules for Analyses of Seeds (BRASIL, 2009). The test was conducted in four replicates of 50 grains, and they were distributed on two sheets of standard filter paper for germination and moistened with distilled water at a ratio of 2.5 times the weight of the paper substrate at 25 °C. The final count of germinated grains was performed after nine days, and the data were expressed as the mean percent of germination.

Cooking time of the beans was determined using a Mattson cooker apparatus. The determination of the cooking time was based on the methodology adopted by Siqueira et al. (2013). Bean samples were first soaked in distilled water for 16 h at a temperature of 25 °C. After this period, whole grains were selected with the shell attached to the cotyledons, which were placed on the apparatus support plate with a pin on each grain. The device was placed in a metal container with boiling distilled water. Grains were considered cooked when the pin passed through the grain, and the cooking time of the sample was recorded when the 13th grain was penetrated.

A completely randomized design was used with subdivided blocks and three replicates. The blocks represented the storage conditions (silo bag, plastic bottle and control), and the sub-blocks contained the five storage periods (0, 30, 60, 90 and 120 days).

Values representing the degree of infestation by insect pests, moisture content, density, electrical conductivity, germination and cooking time were subjected to analysis of covariance using SAS software (PROC GLM, SAS Institute, 2011). Then, the data on the degree of infestation were submitted to a regression analysis as a function of time, using SigmaPlot software, version 13.1 (Systat Software, Inc., San Jose, CA, USA). A canonical correlation analysis (PROC CANCORR, SAS Institute, 2011) was used to investigate the association between the degree of infestation and the group of qualitative characteristics (water content, bulk density, electrical conductivity, percentage of germination and cooking time). Association between the percentage of grains damaged by insects and each of these characteristics was demonstrated by adjusting the linear regression using SigmaPlot, version 13.1 (Systat Software, Inc., San Jose, CA, USA).

3. Results

The infestation by *A. obtectus* significantly varied between the