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Original article

Traditional method of storing cowpea grains and seeds for insect-pest control

Método tradicional de almacenamiento de granos y semillas de caupi para el control de insecto-plaga

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ABSTRACT: This work aimed to evaluate the efficiency of traditional method of storage of cowpea grains and seeds for the control of the *Callosobruchus maculatus* (Fabr.) (Coleoptera: Bruchidae). The storage method consists of packing grains and seeds, mixed with threshing residue, in hermetically sealed polyethylene bottles or barrels. In the experiment, four treatments were evaluated: (i.) Perforated flask containing grains, (ii.) Non-perforated flask containing grains, (iii.) Perforated flask containing a mixture of grains and threshing residues and (iv.) Non-perforated bottle containing a mixture of grains and threshing residues, the latter representing the traditional method (control). For each treatment, 4 replicates were used. For each pot, 20 adult individuals of the insect-pest were inserted. After induced infestation, the pots were stored in a place protected from rain and sun, simulating the reality of farmers' storage. At 35 and 70 days after the induced infestation, the following variables were counted for each pot: number of live insects, number of dead insects, number of holes caused by insects in the grains and percentage of grains attacked. The use of threshing residue was not efficient for the control. On the other hand, the absence of gas exchange in the pot decreased the multiplication of the insect pest, indicating the importance of using hermetic barrels ourvacuum packaging for storage and commercialization.

Key words: hermetic barrels, Amazon, *Callosobruchus maculatus*, threshing residue, *Vigna unguiculata*.

RESUMEN: Este trabajo tuvo como objetivo evaluar la eficiencia del método tradicional de almacenamiento de granos y semillas de caupí para el control del gorgojo, *Callosobruchus maculatus* (Fabr.) (Coleoptera: Bruchidae). El método de almacenamiento consiste en empaquetar granos y semillas mezclados con los residuos resultantes del trillaje en botellas o barriles de polietileno sellados herméticamente. En el experimento se evaluaron cuatro tratamientos: (i.) frasco perforado que contiene granos, (ii.) frasco no perforado que contiene granos, (iii.) frasco perforado que contiene una mezcla de granos y residuos del trillaje y (iv.) frasco no perforado que contiene una mezcla de granos y residuos del trillaje; este último, representando el método tradicional (control). Para cada tratamiento se utilizaron 4 réplicas y para cada frasco se insertaron 20 individuos adultos del insecto plaga. Después de la infestación inducida, los frascos se almacenaron en un lugar protegido de la lluvia y el sol, simulando la realidad del almacenamiento de los agricultores. A los 35 y 70 días después de la infestación inducida, se evaluaron las siguientes variables para cada frasco: número de insectos vivos, número de insectos muertos, número de agujeros causados por insectos en los granos y porcentaje de granos atacados. El uso de residuos resultantes del trillaje no fue eficiente para el control. Por otro lado, la ausencia de intercambio de gases en los frascos redujo la multiplicación de insectos, lo que indica la importancia de utilizar barriles herméticos o bolsas de vacío para el almacenamiento y la comercialización.

Palabras clave: amazonia, *Callosobruchus maculatus*, *Vigna unguiculata*.

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INTRODUCTION

Callosobruchus maculatus (Fabr.) (Coleoptera: Chrysomelidae: Bruchinae) is an important pest of tropical and subtropical legumes that has been distributed worldwide through the seed and grain trade (1). There are records of the pest in all continents of the world (2), and it is even found in distant regions with traditional communities, such as the Juruá valley in the Amazon (Acre, Brazil).

The Juruá valley mesoregion, in the southwestern Amazon, presents a great diversity of traditional cultivars such as common bean (*Phaseolus vulgaris* L.) and cowpea (*Vigna unguiculata* (L.) Walp.), recognized as a conservation center (Figure 1 A) (3). There, common beans and cowpea are produced by traditional farmers and serve as a source of protein for the local population.

Some authors have mentioned that these cultivars are produced by traditional farmers in distant communities using low external input production systems (3) (Figure 1B). The non-use of pesticides is a characteristic that adds value to the production, which has potential for organic and origin ("*terroir*") certification. These farmers, through the use of traditional cultivars ("*criollos*"), play a predominant role in the conservation of genetic resources, with the storage of seeds and grains relevant to ensure the food sovereignty of traditional populations, which also contribute to the conservation of natural vegetation located in protected areas and indigenous lands.

Some farmers in the Juruá valley (Acre, Brazil) store in hermetically sealed tanks or polyethylene bottles (PET), the mixture of beans or seeds, common bean and cowpea, with the residues from the manual threshing process, after harvesting and drying them in the sun (4) (Figure 1 C and D). This process consists of manually separating the beans by beating the dried pods with a wooden stick, a method commonly performed on family properties in this region. This practice has even been mentioned by Professor Dr. Ana Maria Primavesi, who indicated that for better preservation of bean beans it is preferable to store them immediately after threshing, with residues from the process (5).

Traditional agricultural practices for storing grains and seeds are important to reduce the use of synthetic pesticides, reduce health risks and be more environmentally friendly (6). Research on these traditional storage practices can contribute to food security and the preservation of genetic resources (7).

In this context, the objective of this work was to evaluate the efficiency of the traditional method of storing cowpea grains and seeds for the control of the weevil, *Callosobruchus maculatus* (Fabr.) (Coleoptera: Bruchidae).

MATERIALS AND METHODS

The research was conducted at the Federal University of Acre - Floresta Campus (UFAC), located at 7° 33'38"S and 72° 43'01"W, in the municipality of Cruzeiro do Sul, Brazil, from October 2015 to January 2016. In this region, located in the humid Brazilian tropics, where there are two seasons: the rainy season that extends from October to April, average relative humidity of 88 % and the dry season that extends from June to August, and average relative humidity of 75 %. The average annual precipitation is 2166 mm, the average annual temperature is 25.31 °C, the maximum temperature is 32.7 °C and the minimum temperature is 17.1 °C (8).

For the development of the following research, experimental models were created in laboratory conditions that simulated the barrels used by farmers to store beans or cowpeas; the assembly was carried out in opaque polyethylene jars of 9.5 cm in diameter and 15.5 cm in height (Figure 2 A).

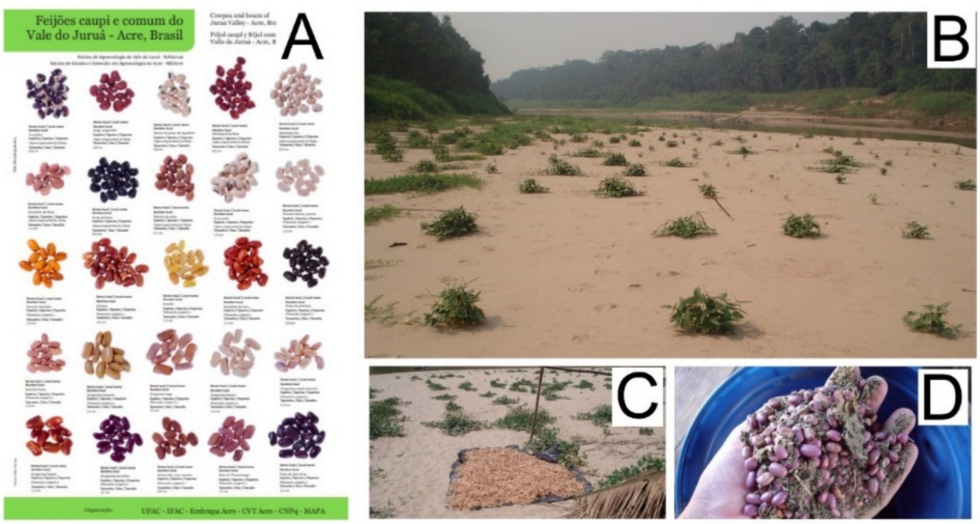

Figure 1. (A) poster produced by the Federal University of Acre on the diversity of common beans and cowpeas in Vale do Juruá; (B) traditional cowpea cultivation on the beach of the Juruá River; (C) cowpea pods drying in the sun; (D) polyethylene tubs used by farmers to store beans containing threshing residues

Four storage conditions of *V. unguiculata* pods of the traditional cultivar "Quarentão" were evaluated. Each storage condition corresponded to a treatment: (i.) perforated jar containing grains (CFSR), (ii.) non-perforated jar containing grains (SFSR), (iii.) perforated jar containing a mixture of grains and threshing residues (CFCR) and (iv.) non-perforated jar containing a mixture of grains and threshing residues (SFCR), control treatment. The perforated jars had 25 holes of 0.6 mm circumference each. For treatments containing residues, 60 grams of threshing residues were mixed with the grains. Threshing was performed manually with a wooden stick, simulating the farmer's practice (Figure 2 B and C).

To obtain the pest insects of the species *C. maculatus*, adult individuals were collected from discarded grain lot (variety "Quarentão") by local trader at the farmers' market, a marketing center for regional products in Cruzeiro do Sul (Acre, Brazil). Subsequently, these insects were transferred to polyethylene bottles with perforated lids containing 300 g of new grains (feeding substrate) from a batch without the presence of pest insects. The bottles were placed in a "Biochemical Oxygen Demand - B.O.D." chamber (model TE-401), regulated at a temperature of 25 °C. After oviposition, only the grains were kept in the bottles, without the presence of live or dead adult insects. After hatching, with the help of metal tweezers, the emerged insects were transferred to the bottles.

A completely randomized design was used, with four replicates (flasks) per treatment. All jars were filled with grains or the mixture of "grains + threshing residues" and 20 adult individuals of the species *C. maculatus*. After the induced infestation, the jars were stored in a place protected from rain and sun (controlled conditions), simulating the reality of farmers storing grains inside houses or in small sheds. The grains had a water content of 13 %.

At 35 and 70 days after the induced infestation, the variables: number of live insects, number of dead insects, number of holes caused by insects in the grains and percentage of attacked grains were evaluated, the latter using the formula: (total attacked grains x 100)/total grains).

The variables were obtained from manual counting by two people. For this, individually for each jar, the attacked grains, dead insects and live insects were separated manually. The live insects were closed in a polyethylene (PET) bottle during the counting process. The data of the evaluated variables were subjected to analysis of variance. Tukey's test was applied, with a 5 % probability, to compare treatment means. The analyses were performed with the help of R software (9).

RESULTS AND DISCUSSION

At 35 days after the induced infestation, there were no significant differences between treatments for the variables: number of dead insects, number of holes caused by insects in the grains and percentage of attacked grains. For the variable number of live insects, the SFCR treatment (control treatment) obtained a lower average than the CFSR treatment (Table 1). These results indicated that, for this evaluation period, storage conditions did not show differences when considering the damage caused to grains by insect pests.

Figure 2. (A) Polyethylene jars simulating barrels used by farmers; (B) bean pods before threshing; (C) residue from bean threshing

However, at 70 days after the induced infestation, jars without perforated lids (SFCR- Control and SFSR) showed smaller amounts of attacked grain and insect holes in pods compared to the treatments with perforated lids (CFCR and CFSR) (Table 1).

The absence of air ingress (jars without perforated lids) reduced damage to beans and the number of live insects, possibly due to the reduction in the concentration of gaseous oxygen and humidity inside the vessels. In this approach, another research work indicated that silo bags and PET bottles (airtight storage) were efficient in controlling *C. maculatus* and preserved the quality of cowpea grains for at least 120 days of storage (10). Some authors showed that oxygen levels below 4 % were effective in controlling these insect pests (11). For this reason, it is recommended that farmers use screw-capped barrels or polyethylene (PET) bottles, especially in hot and humid regions such as Amazonia. It should be noted that high temperatures (32.2 and 33.7 ºC) favored the population growth of the species (12).

Therefore, an alternative that can be applied in the field is the use of vacuum packaging in the post-harvest and marketing of grains, with the potential to add value to production. Currently, there is low-cost equipment that can meet the demand of small farmers; however, it is worth mentioning that this equipment depends on electricity (Figure 3). Other works recommend the use of vacuum hermetic storage for insect control during storage (7,13).

Table 1. Variables evaluated at 35 and 70 days after induced infestation

Treatments	Live insects (Un.)	Dead insects (Un.)	Holes (Un.)	Infested Grains (%)
		35 days after induced infestation		
CFCR	57.55 ab*	34.33 a	87.00 a	3.78 a
CFSR	93.66 a	51.00 a	92.00 a	3.36 a
SFCR	35.75 b	60.75 a	95.25 a	3.32 a
SFSR	69.25 ab	37.50 a	58.00 a	2.39 a
CV%	45.91	89.49	49.56	45.13
		70 days after induced infestation		
Treatments	**Live insects (Un.)**	**Dead insects (Un.)**	**Holes (Un.)**	**Infested grains (%)**
CFCR	153.66 b	1268.33 ab	3167.33 a	65.41 a
CFSR	1238.66 a	1963.66 a	4166.33 a	59.64 a
SFCR	349.25 b	551.50 b	801.75 b	25.31 b
SFSR	430.50 b	760.25 b	698.75 c	25.07 b
CV%	130.38	60.14	84.95	58.76

* Means followed by equal letters in columns do not differ according to Tukey's test at 5 % probability. Treatments: CFCR = perforated jar containing a mixture of grains and threshing residue; CFSR = perforated jar containing grains; SFCR - Control = unperforated jar containing a mixture of grains and threshing residue; SFSR = unperforated jar containing grains

The results suggest that the use of threshing residues did not provide benefits in reducing the damage caused to beans (Table 1). In another research work, with common beans, the threshing residue was not efficient in controlling another insect pest: *Zabrotes subfasciatus* (Coleoptera: Bruchidae) (14). In this context, for these distant communities of the Amazon, rural extensionists and researchers can think of new works focused on the use of botanical insecticides of Amazonian species, such as "Timbó" (*Derris amazonica* Killip) (15), mixed with threshing residues.

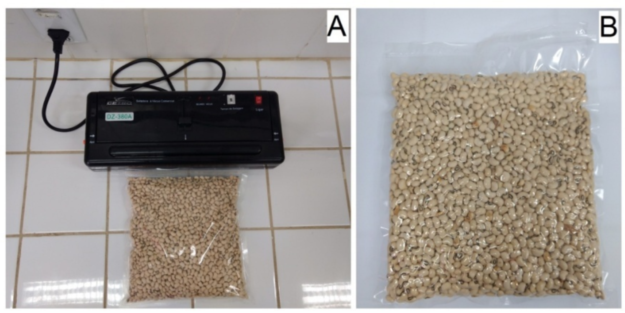

Figure 3. (A) Vacuum packing machine; (B) vacuum packing of 1 kg of cowpea (variety "Quarentão")

CONCLUSIONS

The use of hermetically sealed jars reduced the damage caused by *C. maculatus* on cowpea grains (variety "Quarentão"). Thus, the method of storage in hermetically sealed barrels seems interesting in distant communities. Also promising is the use of vacuum packaging or bags for the commercialization of grains produced in an organic production system (without the use of chemical insecticides).

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