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Modified atmospheres in paddy rice hermetic storage are biogenerated according to different mechanisms in Portugal and Mozambique

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Abstract

Hermetic storage (HS) relies on modified atmospheres (MA) to deliver a safe and environmentally friendly extended storage life technology for agricultural commodities. Here, we discussed the implications of the initial levels of the moisture and temperature in determining which mechanism drives the increased storage capacity of paddy rice using results from experiments with HS under two distinct agronomic and environmental contexts, namely Portugal and Mozambique. Collectively, our results showed its importance as drivers for the modification of the atmosphere content of paddy rice hermetically stored. At low insect infestation conditions of Portugal, the atmosphere was modified only under high moisture content, despite the temperature. In Portugal, when the flux of the grain surpassed the drying capacity, delayed, incomplete or ineffective drying occurred (moistures >14%); therefore, HS could be successfully used. Additionally, we showed that under tropical conditions of Mozambique, even at low moisture (ca. 12%), insect infestation was common, and temperature was high (24 to 36ºC), thus increasing insect activity which changed the atmosphere conditions due to high respiration. MAs are among the best solutions for stored products protection. Our overall results suggested that HS with biogenerated atmospheres was a successful strategy for stored paddy rice under different contexts. In both conditions, between 4 to 12 mo storage, insect populations were controlled under HS when compared with control storage in Portugal (totally supressed at 24ºC) and traditional storage in Mozambique (96% reduction) but driven by specific underlying mechanisms.

Keywords: Paddy rice, Modified atmospheres, Portugal, Mozambique, Insects

Introduction

Rice is important in the diet of populations of Mozambique and Portugal. In Mozambique, rice is mandatory for food security and one of the main cereals available, mainly in rural areas. This commodity is produced mainly by small farmers that use traditional unprofitable low-yield varieties. Moreover, additional losses are significant because of a deficient storage system (Guenha et al., 2014). For long-term storage, their facilities are not effective against the major pests of cereal crops, particularly under high temperature and relative humidity (RH) that are typical conditions of tropical regions, which result in increasing the respiration rate of living organism and insect pests' populations, contributing to quality and quality losses (*Phophi et al., 2020*). Portugal, outshines as the largest rice consumer per capita [~15 kg/person (INE, 2021)] and because rice is a seasonal crop, it needs to be stored and available all over the year. Good storage means no infestation and enough low water activity, below 0.6-0.7 a_w, to prevent grain and mould respiration. One of the sustainable alternatives to the practice of insecticides application is the use of hermetic storage (HS) technology, which is effective in providing long-term, chemical free and sustainable grain storage. Under this technology, biogenerated atmospheres might be achieved naturally from the conditions created by the respiration of living aerobics organisms, changes gas content and, consequently, reduced biotic activity. This mechanism is reported to be as efficient as conventional fumigants to control insects (Navarro, 2010). The present work aimed at comparing the effectiveness of biogenerated modified atmospheres in two different real settings of paddy rice in Mozambique and in Portugal.

Material and methods

Independent trials were conducted in the Sado Valley, Portugal and in Boane district, Mozambique.

Trials in Mozambique (Mz)

Two experiments were conducted in Umbeluzi Agricultural Station, Institute for Agrarian Research of Mozambique (IIAM), Boane District (as detailed in Guenha et al., 2014; Carvalho et al., 2019; Covele et al., 2020). The first experiment (Mz1) was carried out for six months from January to July 2013 and the second experiment (Mz2) was conducted for one year, from February 2016 to February 2017. For both trials, the rice variety used was ITA 312, one of the most cultivated in Mozambique, mainly in the southern region. During the experiments, data loggers HOBO UX100-011 and HOBO UX120-006 M (Onset Hobo® Data Loggers, MA, USA) were used to monitor temperature and relative humidity outside (Mz1) and inside the storage containers (Mz2), located in the middle of the containers (E2) and in the warehouse environment (Mz1 and Mz2). Moisture content was determined using ISO-712 (2009) Norma (Guenha et al., 2014; Covele et al., 2020).

Mz1: To evaluate the efficacy of HS, super bags GrainPro (GrainPro Inc., Philippines) (SGB) were compared with traditional raffia bags, filled with 20 kg of paddy and replicated six times. Raffia bags were considered the control. At the end of 3 mo, three of the replications were opened, and the remaining three replications were opened at 6 mo. For each replication, samples were taken from several cardinal points at the top and bottom position as well as in the periphery and center of bags, thus mixed to obtain a composite sample of about 1 kg, using the methodology recommended by Webley (1985) and Mathur and Kongsdal (2003).

Mz2: Five storage containers were evaluated: 1) traditional Polypropylene bags (control - PP) filled with 50 kg of paddy grain; 2) Super Grain Bags (SGB) filled with 50 kg; 3) Polyethylene Drums (PD) filled with 210 kg; 4) Polyethylene Silo Tanks (PST) filled with 750 kg; and 5) Safe Grain Bags (GB), filled with 1000 kg; all with three replications per treatment. Grain samples and Storgard WB probe traps (Trece, Inc., Oklahoma, USA) were placed in the middle of all storage containers, in order to attract and collect insects. The probe traps were emptied in every sampling and replaced in the containers.

Samples were transported from the experimental site to the laboratory in airtight plastic packaging to preserve their integrity.

Trials in Portugal (PT)

Experiments were conducted in a paddy rice warehouse, in the Sado Valley region, Portugal. Three trials were carried out from December to November 2016: the 1st trial ran 4 mo from December to April (PT1); the 2nd trial ran 6 mo+ from December to July (PT2); and the 3rd trial ran 4 mo from July to November (PT3) (Carvalho et al., 2019). Experiments used two rice varieties: Ronaldo, *Oryza sativa sbsp. Japonica*, and Sírio, *Oryza sativa sbsp. Indica.* The relative humidity and temperature were monitored by Hobo® Data loggers, with the probe placed inside the bags. In all experiments, the two varieties were stored as paddy bulks and submitted to three different relative humidities: 67, 75 and 85% at three different average temperatures: 14 (PT1), 17 (PT2), and 24ºC (PT3). Moisture content was determined using ISO-712 (2009) Norma. At the end of the experiments, CheckpointII Portable O_2 and CO_2 Gas Analyzer was used to assess gas contents at the bottom and top of each bag, making for a total of six measurements per treatment, and the results registered. The gas content was expressed in terms of percentage by volume in air. For each treatment and variety, three replications were carried out.

GrainPro (GrainPro Inc., Philippines) (SGB) bags were used. The efficacy was measured using samples of 20 g infested with 1 wk old of ~20 unsexed *Sitophilus zeamais* Motschulsky, the dominant key-pest of stored rice in Portugal (Carvalho et al., 2013). These samples were placed inside paper bags and each bag was settled inside of each paddy bag. At the end of each trial, these samples were collected and brought to laboratory to evaluate mortality and population growth. *Sitophilus zeamais* was originally collected from Portuguese rice mills and reared in climatic chambers at 25±2ºC and 70± 2% RH.

Data analysis

Mozambique (Mz) - Microsoft Excel, Statistica 12 (*StatSoft, 2011*), and SPSS 20.0 software were used for data analysis: two-way factorial ANOVA analysis and Tukey's HSD mean separation test under 95% confidence limits and Pearson Product-Moment Correlation Coefficient (Guenha et al., 2013; Covele et al., 2020).

Portugal (PT) - R software (R Core Team, 2017) was used for data analysis: function *lm,* simple linear regressions, two-way ANOVA, interaction models, ANOVA models fitted with the R function *aov*, Tukey and Kruskal-Wallis with Fisher's least significant difference tests with functions HSD test and Kruskal from package 'agricolae' (Mendiburu, 2017).

Results

Temperature and relative humidity, moisture, and gas content

Mozambique (Mz) - Mz1: The average temperature was ~24°C [14 to 36ºC] and the mean RH was 67.0% (46 to 83%). The paddy was stored at ~12.0% moisture content (mc). After 3 and 6 mo of storage, mc increased slightly but with no significant differences and not limiting for insect development (Guenha et al., 2014).

Mz2: The variation of temperature was similar in all the containers and ranged between 19.0 to 29.0ºC with June the coolest period and February the hottest month. The relative humidity inside polypropylene bag showed fluctuations along the storage period which was not observed inside the HS. The paddy was stored at 11.0%. There was a significant difference in the mc of grain stored in polypropylene bag (PB), Super Grain Bag (SGB) and polyethylene silo tank (PST) after six months of storage. The Super Grain Bags recorded the lowest moisture content. No significant change was observed in the moisture content of the grain stored in the polyethylene drum (PD) over the storage period (Covele et al., 2020). Gas content was not analysed.

Portugal (PT) The relative humidity did not change inside the hermetic bags under all conditions. The temperature followed the environmental temperature (Fig. 1) and got an average of 14 (PT1), 17 (PT2), and 24ºC (PT3). Paddy mc varied 13% (under 67% RH), 14% (under 75% RH) and 15% (under 85% RH). Under 67% RH, there was no significant change in O_2 and CO_2 content, and no respiration was detected - a sign of no biological activity and good storage conditions. Depending on the increase in RH, more O_2 was consumed due to the increase in the respiration rate of the organisms, and consequently, an increase in CO_2 as well (Fig.2).

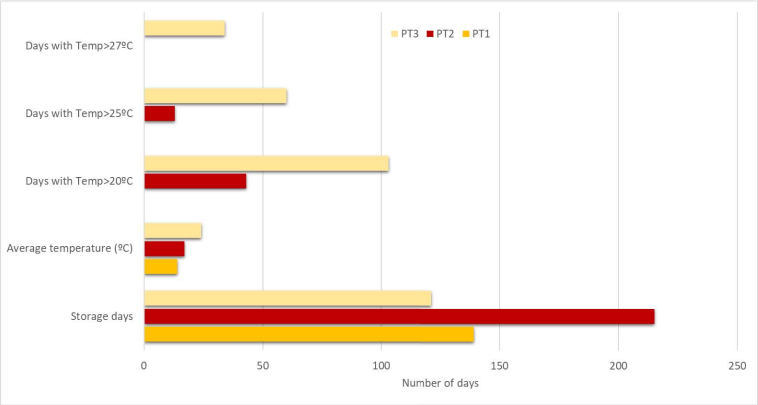


Fig. 1. PT: Average temperature and number of days with temperature greater than 20, 25 and 27ºC (data adapted from Carvalho et al., 2019).

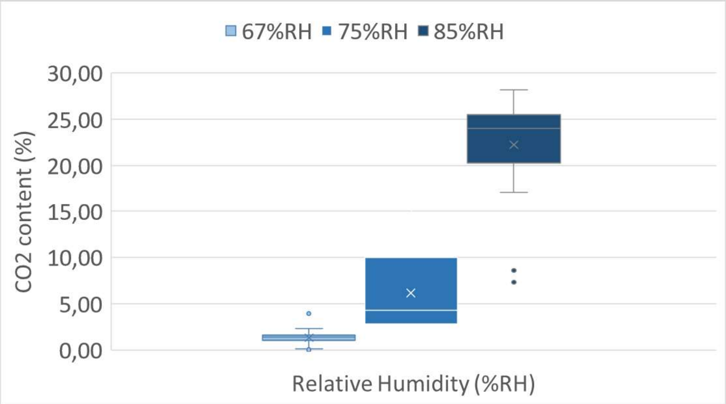
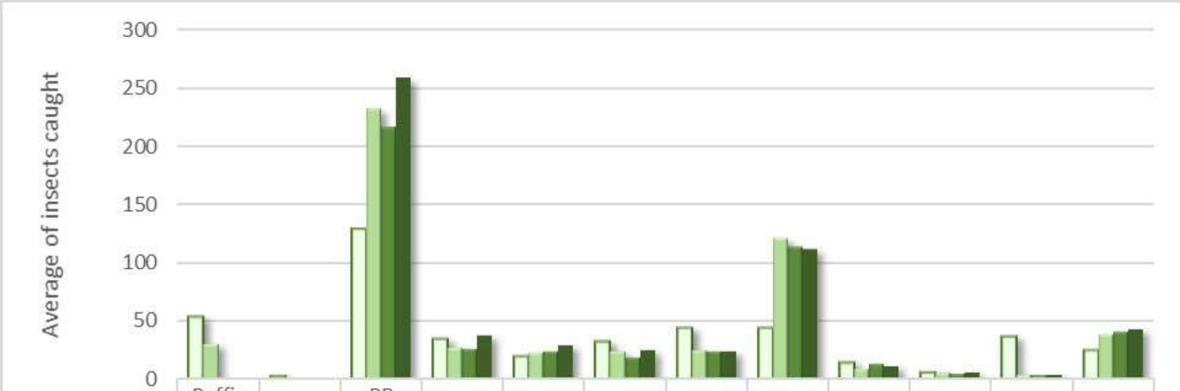


Fig. 2. PT: Relation of CO_2 (%) and RH (%) at the end of the experiment, for paddy rice under 67, 75 and 85% RH. The vertical bars indicate the standard deviation of the replications (data adapted from Carvalho et al., 2019).

Survival of stored product insect pests

Mz1-Samples: Four species were identified over the storage period: *Rhyzopertha dominica* Fab., *Tribolium castaneum* Hb., *Sitophilus* spp., and *Sitotroga cerealella* Oliv. The results showed that HS inhibited the increase of insect populations by over 99% (Fig. 3).

Mz2-Samples: The same four species plus the fifth species, *Cryptolestes ferrugineus* (Stephens), were identified. The lesser grain borer was the most predominant species (71% incidence) followed by the rice/maize weevil (incidence of 17%). *Sitotroga cerealella* were the least predominant species and were not even detected in any hermetic container from the 6 to 9 mo of trials. There were differences between polypropylene bags and HS in the number of insects caught over the storage period. Inside the polypropylene bags, infestation increased by 48%, after 3 mo of storage, and 80% after 6 mo of storage when compared with hermetic devices. During the first 3 mo, SGB showed a lower effect when compared with other HS, but at 6 mo, the SGB reduced the number of insects registered in samples by about two-fold. At 12 mo storage, the initial infestation level in HS was reduced by from 57 to 72%, with an average of 66% reduction in levels, compared with polypropylene bags. Using probe traps, there were no significant differences between any of the devices after three months of storage. After 6 mo of storage, SGB, PD and PST were significantly more effective at reducing insect populations (Fig. 3) (Covele et al., 2020).



	MZ1		MZ2-samples					MZ2-probe traps				
	Raffia (control)	SGB	PP (control)	SGB	PD	PST	GB	PP	SGB	PD	PST	GB
3 months	54	3,5	129	35	20	33	45	45	15	7	37	26
6 months	31	1	233	28	23	25	26	122	10	7	4	39
9 months	0	0	217	27	25	19	25	114	14	5	4	41
12 months	0	0	259	38	30	26	25	112	12	7	4	43

PP = Polypropylene bag; SGB = Super Grain Bag; PD = Polyethylene Drum; PST = Polyethylene Silo Tank; GB = Safe Grain Bag.

Fig. 3. **Mz1-Samples:** Average number of insects caught in raffia and HS before and after storage (adapted from Guenha et al, 2014); **Mz2-Samples:** Average number of insects caught at different storage containers; **Mz2-Probe Traps:** Average number of insects caught at different storage containers (data adapted from Covele et al., 2020).

PT: The average number of insects was considered, subtracting the 20 parental weevil adults used initially (Fig. 4) to better understand if there was progeny during the trials. Respiration rate increased with the increase of relative humidity, followed by average temperature of the trials. Since the number of insects was very low, the authors considered that the main organisms responsible for biogenerated atmospheres might be fungi and paddy rice. Under 85% RH the efficacy of the treatment was achieved for insect control (Carvalho et al., 2019).

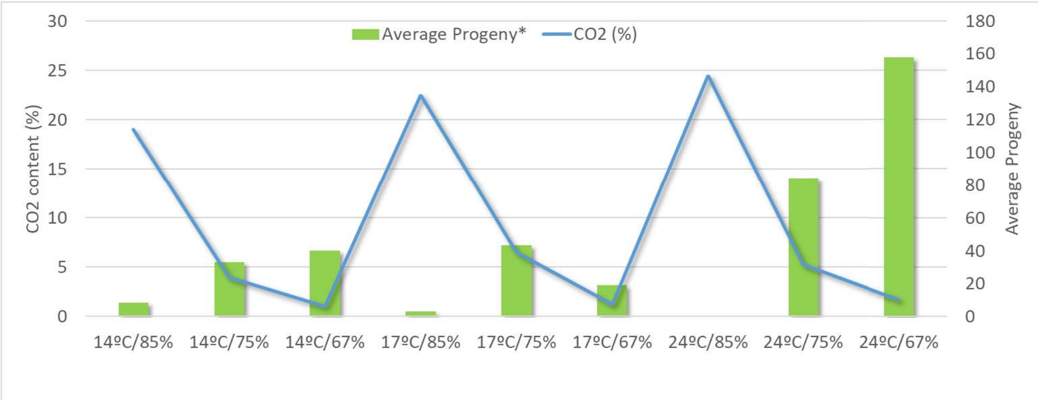


Fig. 4. PT: Average for the Progeny (*Progeny = alive adults + dead adults -20 parent adults) of *Sitophilus zeamais* under each temperature, relative humidity, and CO_2 condition (data adapted from Carvalho et al., 2019).

Discussion

Hermetic storage in temperate conditions is a successful practice for products with restricted infestations, in which modified atmospheres depend only on the relative humidity of the grain and time of storage, as we observed during the Portugal experiments. In contrast, in tropical environments, the grain to be stored typically has an initial high infestation. Under these conditions, the relative humidity and temperature are also high, so the respiration rates of insects, fungi and/or grain increase considerably during storage, reducing oxygen content to levels that make insect survival unsustainable and also reduce the activity of fungi and the activity of enzymes naturally present in the grains, thereby preserving the quality of the stored commodity (De Bruin et al., 2012), which matches the observations in the Mozambique studies. This suggests distinct underlying mechanisms are producing effective results. Therefore, our work emphasizes the implications of the levels of the moisture and temperature in determining the drivers of storage capacity of paddy rice using results from experiments with HS under two distinct agronomic and the contrasting environmental contexts of Portugal (temperate setting) and Mozambique (tropical setting). Collectively, we report that under the conditions of low insect infestation in Portugal, the atmosphere is modified only under high moisture content, despite the temperature and time of storage. In Portugal, when the harvested grain surpasses the drying capacity, then delayed, incomplete or ineffective drying occurs (moistures >14%); therefore, HS can be successfully used. It is mandatory for the grain to be free of insects so that MAs can act by maintaining the optimal conditions for product preservation. Under tropical conditions of Mozambique, even at low moisture (e.g., 12%), the common high insect infestation and high temperatures (24 to 36ºC) leads to increasing insect activity, which then changes the atmosphere conditions to decreased O_2 and increased CO_2 levels (due to high respiration) and also results in restrictive conditions that control infestation. In both conditions, from 4 to 12 mo storage, insect populations were controlled under HS when compared with control in Portugal (totally supressed at 24ºC) and Mozambican traditional storage (96% reduction). In conclusion, we could highlight that biogenerated MAs were among the best solutions for stored paddy rice protection under different contexts but driven by specific underlying mechanisms.

Despite the fact that some synthetic pesticides are effective in controlling pests, their use is being challenged by environmental sustainability concerns and, more importantly, will suffer additional limitations due to the development of insect resistance, food contamination, and safety issues (Silva et al., 2019). The use of HS stands as a sustainable alternative to insecticide application. The MAs in this controlled environment depend on the respiration rate of insects, microorganisms, and the cereal itself (Villers et al., 2006). Any stored grain that is dry and free of insects can take weeks to reduce oxygen levels without the injection of supplemental carbon dioxide.

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