

- OTHMAN, S. H., EDWAL, S. A. M., RISYON, N. P., BASHA, R. K. AND A TALIB, 2017. Water sorption and water permeability properties of edible film made from potato peel waste. *Food Science and Technology (Campinas)*, **37** (1): 63-70. <http://dx.doi.org/10.1590/1678-457X.30216>.
- PANDISELVAM, R., PRAGALYAASHREE, M. M., KAILAPPAN, R., THIRUPATHI, V. AND P. KRISHNAKUMAR, 2014. Moisture dependent engineering properties of onion seeds. *Journal of Agricultural Engineering*, **51** (2): 36-43.
- SALEEM, M.A. AND M.S. SALEEM., 1982. Quantitative loss of some new varieties of pulses caused by *Callosobruchus maculatus* F. *Proceedings of Pakistan Congress of Zoology*, **3**, 82-93.
- TALJA, R. A., HELÉN, H., ROOS, Y. H. AND K. JOUPILA, 2007. Effect of various polyols and polyol contents on physical and mechanical properties of potato starch based films. *Carbohydrate Polymers*, **67**(3): 288-295. <http://dx.doi.org/10.1016/j.carbpol.2006.05.019>.
- THEERTHA, D. P., ALICE R.P.SUJEETHA, J., KAVITHA ABIRAMI, C.V. AND K. ALAGUSUNDARAM, 2014. Effect of moisture content on physical and gravimetric properties of black gram (vigna mungo L). *Int. J. of Advancements in Research & Technology*, **3**(3): 97-104.

Hermetic storage of dry soybean (Glycine max): creating an effective modified atmosphere using soaked grain as O₂ depletor

Hernán Taher¹, Ricardo Bartosik^{2,3,*}

¹ Doctoral Fellow, National Scientific and Technical Research Council (CONICET), Argentina - ² Researcher, National Institute of Agricultural Technology (INTA), Balcarce Research Station, Argentina - ³ Researcher, National Scientific and Technical Research Council (CONICET), Argentina - * corresponding author: bartosik.ricardo@inta.gob.ar
DOI 10.5073/jka.2018.463.141

Abstract

Hermetic storage of grains and oilseed has been proposed as a solution for reducing food losses in developing countries. However, to obtain full benefit of the hermetic storage it is required to achieve a low O₂ concentration (below 2%) or high CO₂ concentration (above 20%). The gas concentration inside the hermetic container is the result of the balance between the respiration and gas exchange rates with the outside (permeability and leakage). When the grain is dry, an insufficient modification in the internal atmosphere is achieved (exchange rate higher than respiration rate), allowing insect development and, hence, grain losses. This study focuses in creating an effective modified atmosphere during the hermetic storage of dry soybean using soaked grain as O₂ depletor. Three big bags with internal polyethylene liners of 70 µm thickness were filled with 590 kg of soybean (Glycine max, with 12.5% m.c.) and sealed. Gas concentration evolution was measured during 15 days (basal condition). Later, four plastic perforated bottles filled with of 4.3 kg of soaked soybean (44% m.c.) were inserted in each bag. The bags were re-sealed and gas concentration was measured during 45 days. Results indicated that the soaked soybean acted as an O₂ depletor, reducing the gas concentration to 1% in only 8 days, and maintained below 1% during 45 days. This research indicated that a small portion of soaked grain (0.4% dry matter (d.m.)) can be used to generate an effective modified atmosphere to prevent biological activity in the entire grain mass. This is a simple and inexpensive approach to reduce food losses under low cost hermetic storage.

Keywords: food losses; pest control; controlled atmosphere; respiration; gas leakage

Introduction

Hermetic storage of grains and oilseed has been proposed as a solution for reducing food losses in developing countries. However, to obtain full benefit of the hermetic storage it is required to achieve a low O₂ concentration (below 2%) or high CO₂ concentration (above 20%) (Navarro et al., 2012). The gas concentration inside the hermetic container is the result of the balance between the respiration and gas exchange rates with the outside (permeability and leakage). When the grain is dry, an insufficient modification in the internal atmosphere is achieved (exchange rate higher than respiration rate) (Abalone et al., 2011a, 2011b), allowing insect development and, hence, grain losses.

The use of liners with gas barrier is an alternative to improve the effect of hermetic storage systems, since the O₂ entrance and CO₂ losses are strongly reduced. This creates an internal atmosphere with a higher modification level than standard liners, with potential conservation benefits (Cardoso et al., 2016). However, liners with gas barrier are expensive and it might not be affordable for family storage systems. Additionally, small perforations in the liner (as small as 1 mm) could eliminate the

benefit of the barrier (Abalone et al., 2011a), preventing the conformation of an effective modified atmosphere.

Injecting CO₂ or N₂ to create an effective atmosphere from the beginning of storage is a widely used technique, usually known as controlled atmosphere (CA) (Navarro, 2012). Carpaneto et al. (2016) implemented CA treatments injecting CO₂ in flexible liners storage systems (silo bags), and concluded that airtightness level is critical to maintain a lethal gas concentration. The lethal atmosphere can be lost in a few days after injection due to gas leaks through small perforations and would fail to achieve insect mortality. A survey made in the field reported that the airtightness level of silo bags is quite variable, due to perforations in the liners from wild animal activity or problems in the sealing (Cardoso et al., 2012). Thus, placing storage systems made of flexible liners in the field might result in perforations that could compromise the effect of the CA treatment.

One alternative is to incorporate an O₂ depletor in a storage system made of a standard liner to speed up the conformation of a lethal atmosphere, even storing dry grains, and also consume the O₂ that is entering by permeability through the linear or through the small perforations. Thus, this study focuses in creating an effective modified atmosphere during the hermetic storage of dry soybean in a big bag using soaked grain as O₂ depletor.

Methodology

1800 kg of healthy and fresh soybean with 12.5% moisture content (m.c.) was bought from a local grain elevator in August 2017. Soaked soybean was obtained by sinking dry soybean seeds in distilled water for 60 minutes. After removing the superficial water with towel paper, the m.c. of the soaked soybean was determined by the oven method (15 g of soybean at 104°C during 72 hs) (ASAE, 2007).

Respiration soaked soybean was characterized. Samples of 50 g of soaked soybean were placed in glass jars of 225 ml, sealed were incubated in a temperature control chamber at 21.8 °C during 7 days. Jars were opened, ventilated for one hour, re-sealed and stored again in the temperature chamber. CO₂ and O₂ concentrations were measured every 1.5 hours until 6 hours with a portable gas analyzer (Checkpoint, Dansensor, Denmark). Respiration rate in terms of O₂ consumption and CO₂ generation was computed according to the procedure described in Ochandio et al. (2017) (the respiration rate of dry soybean reported in this publication was used as reference).

Three big bags with internal polyethylene liners of 70 micrometers thickness and dimensions of 1.0 m x 1.0 m x 1.8 m were filled with 590 kg of 12.5% m.c. soybean, with a total exchange area for gas permeability of 4.07 m². After filling, the big bags were thermo-sealed with portable sealing equipment, and airtightness was tested by a pressure decay test (PDT). The PDT consisted of generating a negative pressure inside the big bag of 1200 Pa with a vacuum pump (Dosovac, DV 95, Argentina) and measuring the time at which the pressure dropped to half the initial value (Navarro, 1998). Following this, hermeticity was restored and CO₂ was injected until an average concentration of 60% CO₂ was achieved. CO₂ and O₂ concentrations were measured once a day for 10 days and permeability of CO₂ and O₂ was calculated. Additionally, samples of plastic liners were sent to the Science and Technology Polymers Laboratory (PLAPIQUI, CONICET-UNS, Bahía Blanca, Argentina) for O₂ standard permeability analysis.

Later, the big bags were opened and internal atmosphere was allowed to equilibrate with ambient atmosphere, and big bags were sealed again. Gas concentration evolution was measured once a day for 15 days to obtain the basal O₂ and CO₂ concentration for dry soybeans. After this characterization, four plastic perforated bottles filled with 4.3 kg of soaked soybean (44% m.c.) were inserted in each bag (2.4 kg dry matter) (Figure 1). The soaked soybeans were placed inside plastic bottles to prevent the contact (and spoilage) with the dry soybean. Additionally, the bottle was perforated to allow the free gas exchange between the inside and outside. The bags were re-sealed and gas concentration measured during several days of storage with a portable gas analyzer.



Figure 1. Plastic bottle perforated and filled with soaked soybean.

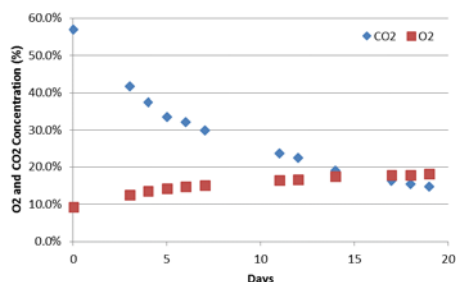


Figure 2. O₂ and CO₂ evolution inside a big-bag injected with CO₂ until 60% of concentration.

Results

Soybean respiration

The respiration rate of soaked soybean in comparison to dry soybean was about five thousand times greater in terms of O₂ consumption and eleven thousand times greater in terms of CO₂ generation (Table 1).

Table 1. Respiration rates of dry and soaked soybean.

Soybean moisture content (w.b.)	Respiration rate (mg/(kg d.m. d))		Source
	O ₂	CO ₂	
Dry (12.5 %)	1.465	0.617	Ochandio et al. (2017)
Soaked (44 %)	7418	6947	This study

The total O₂ consumption and CO₂ generation of the 590 kg of soybean with 12.5% m.c. (516 kg d.m.), were 1321 cc/day and 202 cc/day, respectively; and for the 4.3 kg of soaked soybean with 44% m.c. (2.4 kg d.m.) were 26870 cc/day and 9148 cc/day, respectively. This implies that the contribution to O₂ consumption of the soaked soybean was 20 times greater than the contribution of the dry soybean, while the contribution to CO₂ generation of the soaked soybean was 45 times greater than the contribution of the dry soybean (Table 2).

Table 2. Daily O₂ consumption and CO₂ generation of the dry (516 kg d.m.) and soaked (2.4 kg d.m.) soybeans (cc/day).

Gas	Dry soybean (12.5% m.c.)		Soaked soybean (44% m.c.)		Total (cc/day)
	(cc/day)	% of total	(cc/day)	% of total	
O ₂ consumption	1321	4.7	26810	95.3	28131
CO ₂ generation	202	2.1	9148	97.9	9350

Pressure decay test and permeability of the big bags

Results of the PDT for the three big bags were higher than 5 minutes. According to Navarro (1998), an hermetic structure 95% full should have a PDT of 3 minutes to be suitable for CA treatments and 5 minutes for modified atmosphere storage. However, Carpaneto et al. (2016) speculated that the threshold set by Navarro (1998) might be too strict for flexible grain storage systems, since there is no head space and, theoretically, no leakage of gas occurs as result of pressure release. Nevertheless, the results of the PDT indicated that there were some small leakage, which should be considered to estimate the gas exchange between the inside and the outside of the big bag, besides the permeability through the plastic liner.

The effective permeability of the plastic liner (taking into consideration the permeability through the plastic liner plus the gas exchange occurred through small openings) was calculated. Figure 2 shows the O₂ and CO₂ evolution after injecting CO₂ gas in the big bag up to a concentration of 60% of CO₂ and 10% of O₂. The rate loss (cc/(m² d Δ[%])) was derived from this figure, and linear models

for O₂ and CO₂ effective permeability were obtained (Figure 3). The permeability to both gasses was similar, 6941.3 and 7545.2 cc / (m² day atm) for CO₂ and O₂, respectively. The result of the standard permeability test of the liner for O₂ (without perforations) was 1434 cc / (m² day atm), indicating the effect of small perforations on the effective permeability of the system, which could quintuplicate the permeability of the liner. Abalone et al. (2011a) studied the effect of perforation in the effective permeability of flexible plastic liners (silo bags) and concluded that perforations provide non-selective permeation of gases, d.m. and also caused a significant modification in the internal atmosphere (although the effective permeability value of liners with perforation was not reported).

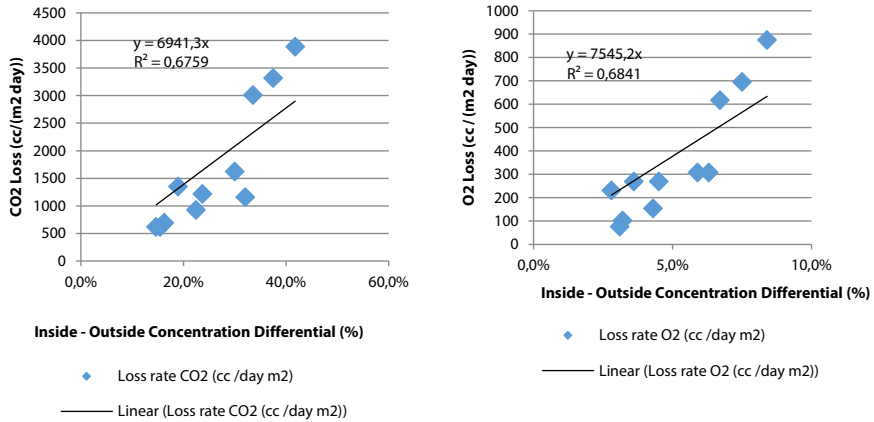


Figure 3. O₂ and CO₂ permeability rate (cc/(m² day)). Note: the intercept of the linear model was set to zero.

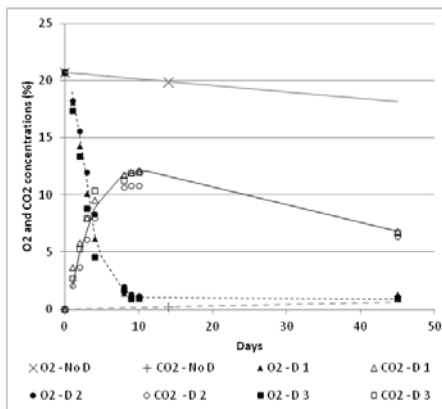


Figure 4. O₂ and CO₂ concentration inside the big bags **Figure 5.** Plastic bottle with soaked and spoiled soybean after 45 days of storage.

Gas concentration inside the big bag with and without O₂ depletor

Gasses concentration when dry soybean was stored in big bags did not change during 14 days of storage, being the O₂ and CO₂ concentrations of 19.9% and 0.2% respectively. This is because the respiration rate of dry soybean was low in comparison with the permeability of the big bag (Table 1). However, when the soaked grain was inserted inside the big bag, the respiration rate of the grain was high enough to create a substantial modification of the internal atmosphere (O₂ consumption and CO₂ generation increased from 20 to 45 times, respectively (Table 2)). The O₂ concentration dropped at a rate of 2.5 percentage points per day, reaching a concentration of 1% after 8 days and

maintaining such a low O₂ level until 45 days of storage, while the CO₂ concentration increased up to 12% at 8 days of storage and slowly decreased to 7% at 45 days (Figure 4).

After 45 days of storage the big bags were opened and the plastic bottles with the soaked and spoiled grain were removed. The grain inside the plastic bottles was completely spoiled, while the rest of the grain was without any visible damage (Figure 5). However, a strong smell to spoiled grain was detected and it was necessary to ventilate the grain to partially remove the odors.

Discussion

The key point to achieve an effective hermetic storage is to create an internal atmosphere with low O₂ concentration (below 2%) to prevent insect development and reduce microbial activity. For most insect species, 15 days of exposure to O₂ concentration below 2% would be enough to achieve 100% mortality.

Hermetic storage of dry products has some limitations due to the low respiration rate. In most cases, the respiration rate is about at the same magnitude order (or lower) than the permeability of the liners (or rigid hermetic structure), thus the system reach an equilibrium at a O₂ and CO₂ concentrations that is not effective for preventing insect or mold activity. Only if the insect or mold activity increases substantially, respiration rate surpass the permeability rate and an effective internal atmosphere is achieved. However, when this occurs, uncontrolled quality deterioration in the stored product is observed.

One possible solution for this problem is to incorporate liners with O₂ barrier, which extremely reduce the permeability to O₂ and allow achieving an effective atmosphere even when the respiration rate of the dry product is low. However, liners with O₂ barrier are not always available and also are more expensive, which prevent its use in low cost family storage systems. Additionally, regardless the permeability of the liner, there is gas exchange through small perforations or micro-failures in the sealing (most plastic liners are fragile and, during storage, wild and domestic animals can cause damage) substantially increasing the effective permeability of the liner. Thus, it is difficult to guarantee a low effective permeability during storage, even if O₂ barrier liners are used.

Other alternative to achieve an effective internal atmosphere is to incorporate O₂ depletor to consume the O₂ that is entering into the storage system. In this study, 2.4 kg (d.m.) of soaked soybean (44% m.c.) in 516 kg (d.m.) of dry soybean (12.5% m.c.) reduced the O₂ concentration to 1% in less than 10 days, and after 45 days of storage the O₂ concentration did not change. Such a low O₂ concentration is effective to control insects (Navarro et al., 2012) and would also reduce microbiological activity (Ochandio et al., 2017). The results of this study indicated that with a controlled loss of 0.46% of d.m. a safe storage condition can be achieved. However, even though the modification in the internal atmosphere was satisfactory, a strong smell to spoiled grain was detected after 45 days of storage inside the big bag. Using a chemical compound as O₂ depletor could be a better solution. In this case the chemical compound should react with the O₂ without generating toxic bi-products neither unpleasant odor. Taking as an example a big bag full with 590 kg of soybean, it would be required to consume 66080 cc of O₂ (86.45 g) to reach 0% concentration. Additionally, to maintain 0% concentration during 15 days it would be necessary to consume 6030 cc/day (7.9 g) extra of O₂ per day to compensate the permeability of the big bag. Thus, the O₂ depletor must be able to capture a total of 156530 cc (205 g) of CO₂ to reach and maintain 0% of O₂ during 15 days. If such chemical product is identified, effective hermetic storage of dry grains in big bags could be implemented using standard polyethylene liners.

Conclusions

The incorporation of 2.4 kg (d.m.) of soaked soybean in a big bag containing 590 kg of dry soybean (12.5% m.c.) reduced the O₂ concentration to 1% in less than 8 days. Additionally, the O₂ that permeated into the system was consumed by the soaked soybean at the same rate, maintaining the O₂ concentration below 1% after 45 days of storage.

A small portion of soaked grain (0.4% d.m.) could be used as O₂ depletor to create an effective modified atmosphere during storage of dry products in hermetic systems made of liners without O₂ barriers or with small perforations.

This is a simple and inexpensive approach to reduce food losses under low cost hermetic storage systems.

O₂ depletors made of chemical compounds could be investigated to obtain the same results as using soaked grain, but without generating unpleasant smell.

Acknowledgement

The authors are thankful to the National Institute of Agricultural Technology (INTA) for the financial support for this research through the projects PNAlyAV-1130023 and PNCyO-1123023.

References

- ABALONE, R., GASTÓN, A., BARTOSIK, R., CARDOSO, L. AND J. RODRÍGUEZ, 2011a: Gas concentration in the interstitial atmosphere of a wheat silo-bag. Part II: Model sensitivity and effect of grain storage conditions. *Journal of Stored Product Research*. 47, 276–283.
- ABALONE, R., GASTÓN, A., BARTOSIK, R., CARDOSO, L. AND J. RODRÍGUEZ, 2011b: Gas concentration in the interstitial atmosphere of a wheat silo-bag. Part I: Model development and validation. *Journal of Stored Product Research*. 47, 268–275.
- ASAE, 2007: ASAE D245.6 OCT2007 - Moisture relationships of plant-based agricultural products.
- CARDOSO, L., BARTOSIK, R., CAMPABADAL, C. AND D. DE LA TORRE, 2012: Air-Tightness Level in Hermetic Plastic Bags (Silo-Bags) for Different Storage Conditions, in: Navarro, S., Banks, H.J., Jayas, D.S., Bell, C.H., Noyes, R.T., Ferizli, A.G., Emekci, M., Isikber, A.A., and K. Alagusundaram, [Eds.], Proceedings of the 9th International Conference Controlled Atmospheres and Fumigation of Stored Products, October 15 to 19 of 2012, Antalya, Turkey..583–589.
- CARDOSO, L., BARTOSIK, R., CASTELLARI, C., ABADIA, B., DE LA TORRE, D. AND H. TAHER, 2016: Hermetic storage of wet corn in liners with and without EVOH barrier., in: Navarro, S., Jayas, D. and K. Alagusundaram (Eds.), Proceedings of the 10th International Conference of Controlled Atmospheres and Fumigation of Stored Products. CAF Permanent Committee, New Delhi, India, November 6-11 of 2016, pp. 117–128.
- CARPANETO, B., BARTOSIK, R., CARDOSO, L. AND P. MANETTI, 2016: Pest control treatments with phosphine and controlled atmospheres in silo bags with different airtightness conditions. *Journal of Stored Product Research*. Submitted, 143–151.
- NAVARRO, S., 2012: The use of modified and controlled atmospheres for the disinfection of stored products. *Journal of Pest Science*. (2004). 85, 301–322.
- NAVARRO, S., 1998: Pressure tests for gaseous applications in sealed storages: theory and practice, in: Zuxun, J., Quan, L., Yongsheng, L., Xianchang, T., and Lianghua, G. (Ed.), Proceedings of the 7th International Working Conference on Stored-Product Protection. IWCSPP, Beijing, China, pp. 385–390.
- NAVARRO, S., TIMLICK, B., DEMIANYK, C.J. AND N.D.G. WHITE, 2012: Controlled or Modified Atmospheres, in: Hagstrum, D., Phillips, T., Cuperus, G. (Eds.), *Stored Product Protection*. Kansas State University, Manhattan, Kansas, USA, pp. 191–202.
- OCHANDIO, D., BARTOSIK, R., GASTÓN, A., ABALONE, R., ARIAS BARRETO, A. AND A. YOMMI, 2017: Respiration rate of soybean seeds (*Glycine max*) in hermetic storage. Enviado para su revisión a *J. Stored Prod. Res.*

Biocidal efficacy of nitrogen (anoxic atmosphere) applied in operational condition to stored hazelnuts against pest insects at different stages of development.

Francesca Lampugnani*, Guglielmo Cassani, Luciano Süs, Dario Zanoni, Federico Ceriani

Via isonzo 20 – 20089 Rozzano (MI) – Italy

*Corresponding author: francesca.lampugnani@agrobly.com

DOI 10.5073/jka.2018.463.142

Abstract

Recently, a test was conducted in Italy for the evaluation of the biocidal efficacy of Nitrogen saturation (anoxic conditions). One application was carried out in a controlled atmosphere cell of a logistic center specialized in receiving, storing and shipping foodstuffs. The cell, circa 3682 m³ volume, with capacity of 752 big bags of fresh shelled hazelnuts on 4 height levels was saturated with Nitrogen (99,9%) and maintained at 15-18°C for 21 days. Five test species of insects *Plodia interpunctella*, *Cadra cautella*, *Corcyra cephalonica*, *Tribolium confusum*, *Oryzaephilus surinamensis* were observed at different development stages (egg, larva, adult). The target species were sorted in special biotest and inserted in the big bags to simulate an infestation. At the end of the exposure period the biotests were collected and analyzed. The treatment resulted sufficient to achieve a total control on eggs of Lepidoptera test species only. This result confirmed and integrates the available information in literature that showed the need of a longer minimum exposure period for total control of common stored pest insects.