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CONDITIONING AND SAFE STORAGE OF COWPEA SEEDS USING PLASTIC HERMETIC BAGS

Dalia H. Saleh^{1*}, T.Z. Fouda², M.M. Elkholy¹ and A.H. El Metwalli²

1. Agric. Eng. Res. Inst., Agric. Res. Cent., Egypt

2. Agric. Eng. Dept., Fac. Agric., Tanta Univ., Egypt

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ABSTRACT: Most important feature of the storage systems is to preserve the integrity of the grain for a given period with minimal loss in quality and quantity. The main objective of this study aimed to choose the proper conditions which ensure safe storage process using infrared heating at the optimum radiation intensity of 882.67 W/m² and exposure time of 15 min and the ultraviolet radiation at radiation intensity of 3.538 mW/cm² and exposure time of 40 min on storability of cowpea seeds using different types of plastic hermetic bags in comparison with polypropylene (pp) woven bags. The changes in cowpea seeds quality during storage of pre-treated cowpea seeds in terms of moisture content, protein content, total microbial count and insect detection were also determined. The results show that, the moisture content of seeds stored in pp woven bags increased in contrast with both types of studied hermetic bags, oxygen concentration for hermetic bags decreased during storage period and carbon dioxide increased unlike pp woven bag, The rate of seeds infection with microorganisms and weevils was lower comparing with the samples stored in both types of hermetic bags and crude protein content of seeds stored in hermetic bags reduced at a very slight rate in comparison with that stored in woven bag.

Key words: Hermetic storage, cowpea seeds, seeds quality, infra-red treatment, ultra-violet treatment, protein content, total microbial count and insect detection.

INTRODUCTION

The quality of stored grain depends on four important factors: initial conditions of the grain, environmental conditions during the period of storage, biotic factors, such as (insects - rodents - microorganisms) and various treatments applied on the grain during the storage period **Rajendran (2003)**. Stored grains and legumes are subject to insect infestation and deterioration from molds and bacteria. At the developed countries the average minimum overall losses from biological degradation is 10%, while in developing countries that estimate may be up to 20%. Stored food products are highly prevalent in developing countries, especially among smallholder farmers **World Bank and FAO (2011)**. Among sub-Saharan African countries and other developing countries, these losses

come as a result of inadequate use of highly improved post-harvest technologies during storage. Faced with such devastating losses, many farmers do not want to risk their cowpeas. Instead, they sell them at harvest time when prices are the lowest. Mapping up strategies to reduce these losses will ensure food security, lead to rapid economic growth, and improve nutrition on the continent **Affognon et al. (2015) and Keatts (2016)**.

Hermetic storage (HS) technology is a non-chemical-based system of storage. Its basic principle is the generation of oxygen depleted, carbon dioxide and or nitrogen enriched interstitial atmosphere caused by either the natural respiratory activities of living organisms in the bulk, or enhanced and accelerated by artificial means in an air tight storage structure **Jonfia et al. (2008)**. About 30% of cowpea stored

* Corresponding author: Tel. :+201212168615

E-mail address: eng_dalia_hasan@yahoo.com

on-farm was in potentially hermetic containers, but much of that also was treated with insecticide because farmers did not trust the efficacy of hermetic storage alone. Data collected in 2010 in Niger and Burkina Faso indicated that over 70% of farm-stored cowpea was in hermetic containers and from 7% to 38% of farm-stored cowpea was in Purdue Improved Cowpea Storage (PICS) bags **Moussa *et al.* (2010)**. The effect of hermetic and non-hermetic storage of cowpea in plastic containers in the tropics was studied the cowpeas were stored in hermetic and non-hermetic containers over a period of 12 weeks. The parameters evaluated were the moisture content, insect infestation, usable proportion, and 1000 grain mass in both hermetic and non-hermetic systems. The results showed that the moisture content in the hermetic containers increased slightly from 11.7 to 11.9% compared to a sharp increase from 11.7 to 17.2% in the non-hermetic plastic containers. From the fourth week to the twelfth week, the number of live insects drastically reduced to zero in the hermetic system. In the case of the non-hermetic containers, the population of live insects/100 g of grains increased from 5 on zero week to 71 on the twelfth week. Also, the mass of 1000 grains reduced from 156.50 g on week zero to 145.21 g in the non-hermetically stored grains, while the hermetically stored grains recorded a decrease to 148.95 g. Finally, the usable proportion of grains in the hermetic system declined from 98.55 to 94.80% after 12 weeks of storage as compared to the drop to 85.69% seen in the non-hermetic system **Aboagye *et al.* (2017)**.

The current study aims at testing and evaluating the effect of cowpea seeds pre-treatment with infra-red and ultraviolet and storage in different types of hermetic bags on safe storage and quality prevention of the stored seeds.

MATERIALS AND METHODS

The experiment work was carried out through season 2019 in the Rice Mechanization Centre at Meet El-Deeba, Kafr El-Sheikh Governorate, Egypt to study the effect of hermetic bags (three layers bags and seven layers bags) and Polypropylene woven bag on storage of pre-treated and non-treated cowpea seeds.

Two different types of barriers films were developed for the experimental work. The developed films were formed into a shape of bags with capacity of 20 kg/bag. The produced bags were filled by non- treated cowpea seeds at initial moisture content of (10.95%) w.b% and pre-treated cowpea seeds at the optimum intensity and exposure time of 882.67W/m² and exposure time of 15 min for the infrared heating at the radiation intensity and radiation intensity of 3.538mW/cm² and an exposure time of 40 min for the ultraviolet treatment, these values were selected based on pre-experiments.

The filled bags were stored in a proper storage room at three groups (three and seven layers-woven bags). Each group contain three different types of seeds (Non treated and treated seeds with IR and treated with UV). Measurements for carbon dioxide, oxygen gas, temperature and relative humidity inside the stored bags of each group were taken monthly. Also, quality evaluation tests for the stored seeds including moisture content, protein, total microbial count and insect detection were measured for all the examined treatments.

Measuring Procedure

Temperature and RH

Stainless probe with thermocouple type K was used for temperature measurement inside and outside the bags. Also, humidity content probe was used to measure the humidity content in the bulk of Cowpea seeds inside and outside the hermetic bags.

Moisture content

The moisture content for cowpea seeds was determined by the standard oven method at 105°C for 24 hours (**AOAC, 1990**).

The moisture content was calculated at wet basis (w.b. %) as following:

$$M_{wb} = [(m_w - m_d) / m_w] \times 100$$

Where:

M_{wb} : Moisture content, %.

m_w : Wet mass, g.

m_d : Dry mass, g.

O₂ and CO₂ concentrations

Were monitored every month using CO₂ and O₂ sensor (VI GAZ "Gas analysis- model Box 121, (VI GAZ Company, France).

Insect detection (insect/kg)

The cowpea seeds were sieved and the weevils were identified according to (AOAC, 2000).

Total microbial colony count (microbiological analysis), cfu/g

Total microbial count activity was determined following the methodology; about 25 g from the samples was transferred in to a stomacher bag (Seward, London, UK), and homogenized with 225 ml of sterile saline peptone water (SPW:1g/1 peptone, 8.5g/l sodium chloride) for 3 min. A 10- fold serial dilution was made from each sample and used for quantitative microbiological examinations after treatment with Serial dilutions of sterile saline peptone water with samples was prepared, and duplicate of 1 ml samples of appropriate dilutions were poured on agar plates. Total bacterial count (TBC) was enumerated on plate count agar (Merck, 1.05463) at 30°C for 48 h. All plates were examined for typical colony types and morphological characteristics associated to each culture medium.

Total protein content

The total nitrogen was determined by using micro kjeldahel method (AOAC, 1990). Total protein was calculated by multiplying the total nitrogen by 5.57.

RESULTS AND DISCUSSION

Ambient Air Temperature and Relative Humidity

Fig. 1 illustrated the recorded data of air temperature and relative humidity during the storage process. The storage process for cowpea seeds started from March to October 2020. As shown in Fig. 1, over the studied storage time, the highest and lowest ambient temperature were 29.7°C and 18.2°C with an average of 23.95°C. Meanwhile, the highest and lowest ambient relative humidity were 66% and 56.5% with an average of 61.3%.

Bulk Temperature and Relative Humidity Inside The Storage Bags

Temperature is considered a critical factor for seed storage. During the storage period the values of temperature and RH inside the bags are shown in Fig. 2. The recorded bulk temperature of the non- irradiated cowpea seeds ranged from 19.6 to 31.8, 19.6 to 29.6 and from 19.1 to 29.5°C with an average of 27.4, 25.85 and 25.9°C for the pp woven bags, three and seven layers hermetic bags, respectively. The recorded relative humidity inside the non-irradiated bags ranged from 57.9% to 61% with an average 59.45 % for woven bags, and ranged from 56.2% to 61.4% with the average 58.8% for the three layers bags while it ranged from 53% to 58.6% with an average of 55.8% at the seven layers bags. Meanwhile the bulk temperature of irradiated seeds with infra- red pre-treatment ranged from 18.9 to 31.2, 18.3 to 28.9 and 18.4 to 28.5°C with an average of 26.86, 25.11 and 24.9°C for the pp woven bags, three layers hermetic bags and the seven layers hermetic bags respectively. However, the relative humidity ranged from 59.1% to 61.7% with an average 60.4% for woven bags, from 55.3% to 58.8% with an average of 57.05% for three layers bags and from 53% to 57.6% with an average 55.32% for seven layers bags. On the other hand, the recorded bulk temperature of cowpea seeds irradiated with ultraviolet pre-treatment ranged from 18.5 to 30.9, 18.2 to 28.8 and from 18 to 28.5°C with an average 26.4, 25.025 and 24.56°C for the pp woven bags, three layers bags and the seven layers bags respectively. Meanwhile, the minimum relative humidity of 58.5% and maximum of 61.5% with an average 60% was recorded for the woven bags, from 54.3% to 56.9% with an average 55.6% for three layers bags and from 51.1% to 55.5% with an average 53.3% for seven layers bags.

Moisture Content of Stored Seeds

The change in moisture contents of cowpea seeds during storage process depends on the initial moisture content, the relative humidity surrounding the seeds, growth of insects, microorganisms and respiration rate of seeds, where both of seeds respiration rate and insects released water. As shown in the Fig. 3, moisture

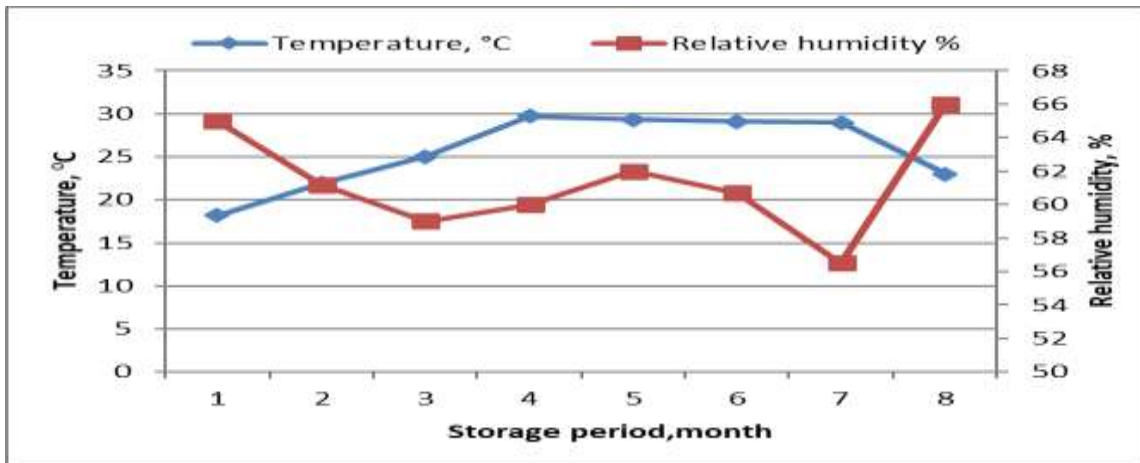


Fig.1. Ambient air temperature and relative humidity during the storage period

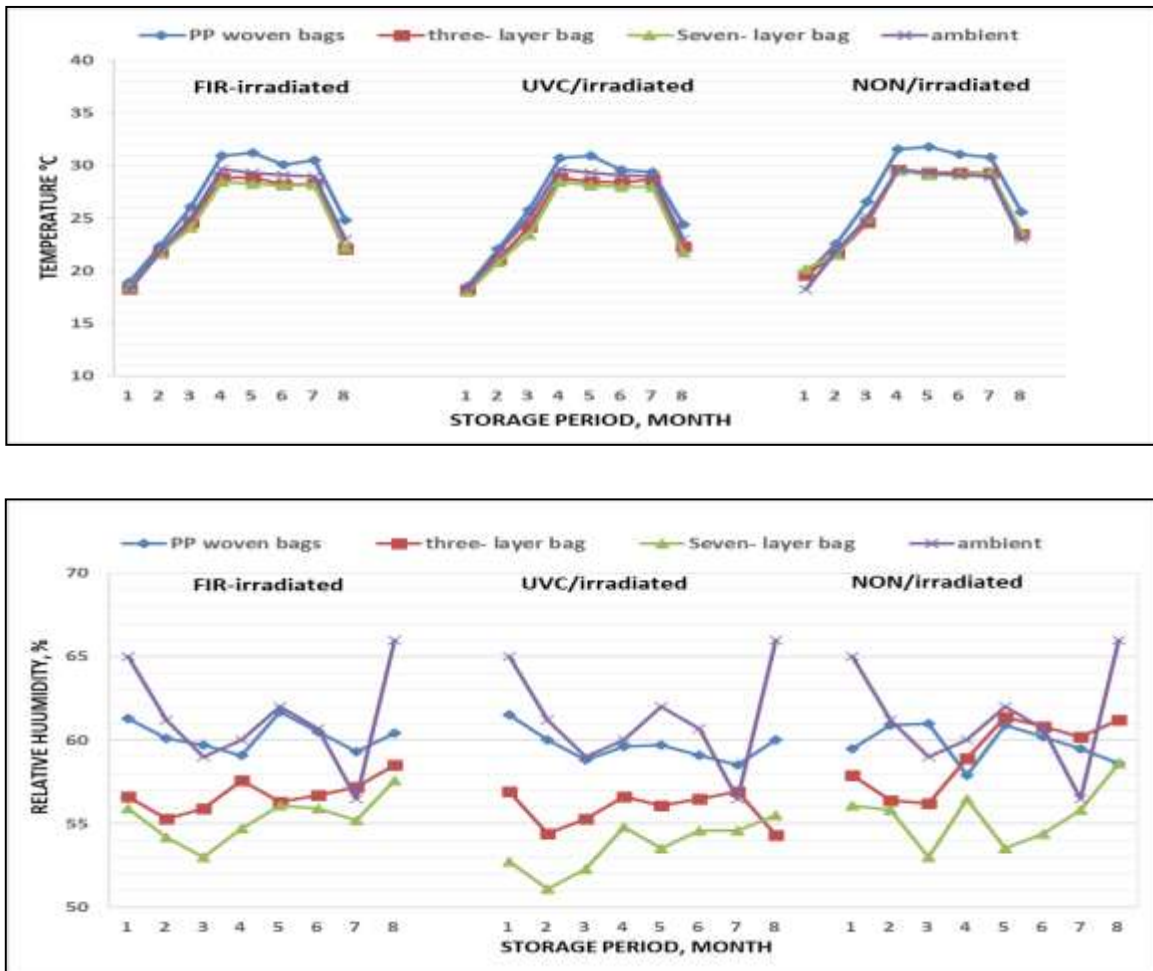


Fig.2. Bulk temperature and relative humidity inside the storage bags

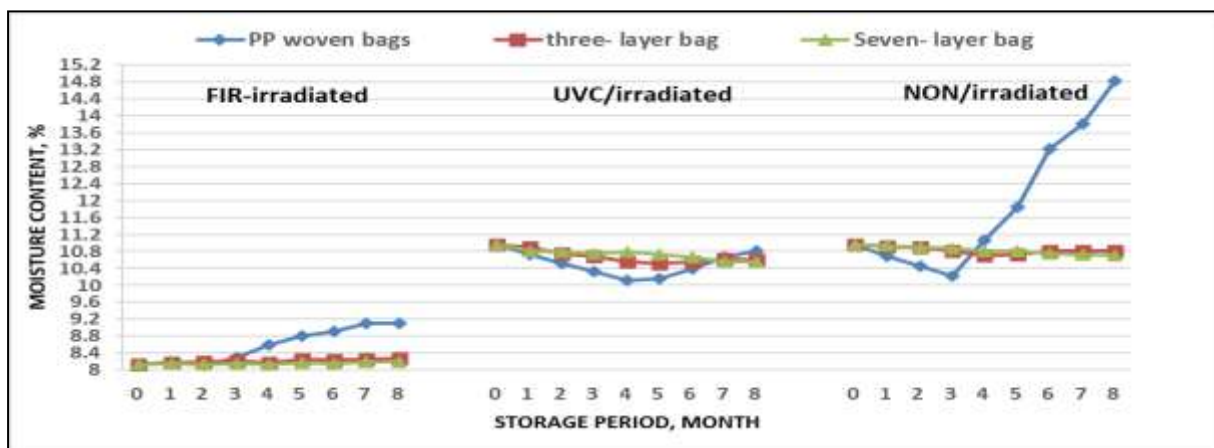


Fig. (3) Moisture content of stored seeds

content of the non-irradiated seeds, relatively decreased from 10.95% to 10.21% W.b% at the first three months of storage, then starts to increase to 14.82% for the pp w treatments due to the water oven bag. However, the three and the seven layers hermetic bags recorded very low changes in seeds moisture content for all studied sealing effect of the plastic hermetic bags. The results also show that, there is relationship between deterioration rates, insect infestation level, molds and yeast attack, storability and moisture content of seeds.

Generally, the contaminated non treated seeds showed higher value of moisture content and bulk temperature compared with the treated seeds due to insect respiration and this is consistent with Embaby and Abdel-Galile (2006).

Oxygen Concentration Inside the Storage Bags

Oxygen concentration was influenced by the type of bags and seeds condition as shown in Fig. 4. For the pp woven bags filled with irradiated or not irradiated seeds the Oxygen level fluctuated between 19.8% and 19.3% all over the storage period. Meanwhile, the three layers bags filled with non-treated seeds showed a reduction of Oxygen concentration from 19.8% to 6.7% while, in the seven layers bags, the Oxygen concentration decreased from 19.8% to 5.5%. This means that oxygen concentration decreased by 66.16% and 72.22% for the non-treated seeds stored in three and seven layers bags respectively. For the bags filled with infra-

red treated seeds, the Oxygen concentration decreased from 19.8% to 7.5% for the three layers bags and from 19.8% to 7.3% for the seven layers bags respectively. Whilst the bags that filled with treated seeds with the ultraviolet radiation (UVC), the oxygen concentration decreased from 19.8% to 8.4% for the three layer bags and from 19.8% to 8.1% for the seven layers bags respectively.

The above mentioned results revealed that, depletion of oxygen concentration in the hermetic bags associated with lower metabolic activity of seeds and minimum rate of insects, bacterial and fungal growth, which consume the O_2 and release CO_2 required for safe storage of seeds (Njoroge *et al.*, 2014).

Carbon Dioxide Concentration Inside the Storage Bags

As shown in Fig. 5 the highest levels of carbon dioxide concentration inside the bags were recorded in seven layers bags which approached 24.8%, 20.9% and 20.5%, while they were 19.7%, 16.9% and 16.2% for three layers bags at the end of storage period for the non-irradiated, infrared irradiated and ultraviolet irradiated bags respectively. In contrast pp woven bags in all conditions of seeds (irradiated or non-irradiated) recorded very low levels of CO_2 which ranged from 0.1% to 0.7%. In general, Oxygen concentration was dropped and carbon dioxide increased in the hermetic bags, due to respiration of stored seeds that cause releasing carbon dioxide and depletion of oxygen inside the hermetic bags.

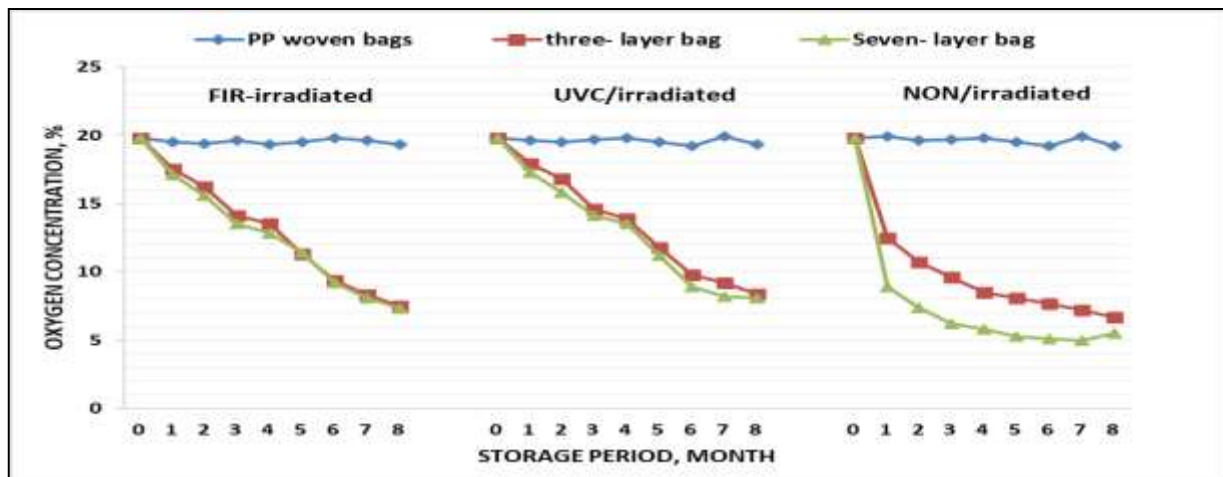


Fig.4. Oxygen concentration inside different types of storage bags

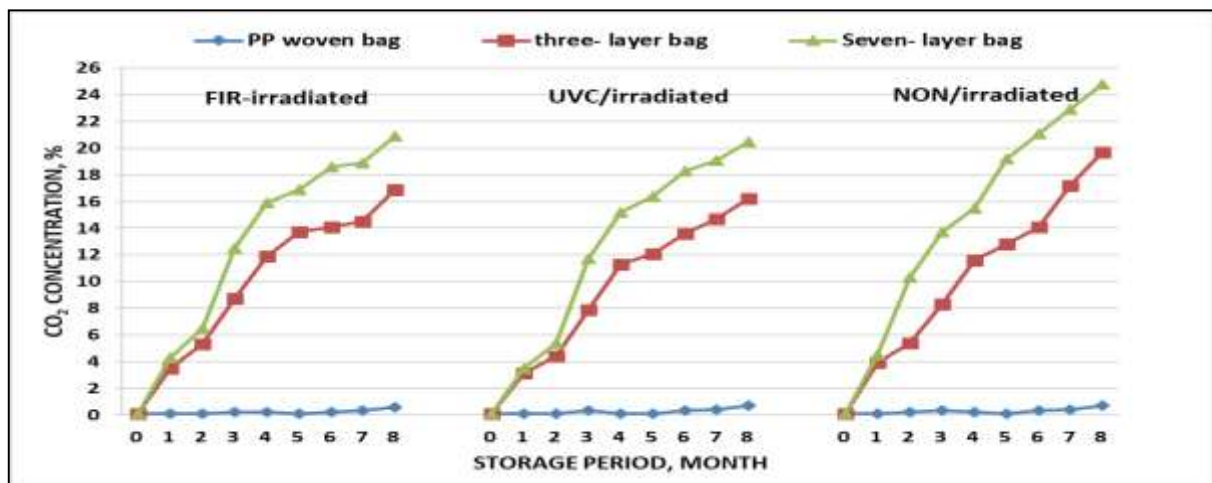


Fig.5. Carbon dioxide concentration inside different types of storage bags

Insect Detection of Cowpea Seeds During Storage

Insect detection of cowpea seeds (insect/Kg) during storage process inside different types of bags illustrated in Table 1. It was noticed that the non-treated and treated seeds stored on the pp woven bags contained live populations of cowpea weevil insect (*Callosobruchus maculatus*). The number of weevil insects/kg were 81, 13 and 11 for the non-irradiated, infra-red irradiated and ultraviolet irradiated treatments. However, the hermetic bags when emptied, the weevils were found dead.

This means that, in case of using the hermetic bags, the produced carbon dioxide from seeds respiration approached levels not proper for continuous growth of insects.

Total Microbial Count during Storage Period

Total microbial count (log cfu/mg) was influenced by the type of bags and seeds condition (treated – non treated) as shown in Fig. 6. The total microbial count of seeds that not irradiated increased from 4.7 to 5.68 (Log cfu/g) for pp woven bags whereas the three layers bags and the seven layers bags recorded total microbial count of 3.1 and 2.8 (Log cfu/g), respectively.

Table 1. Insect detection during storage period of bags

Insect detection (Insect/Kg)									
Storage period, month	Type of bag								
	PP Woven bag			3 layer bag			7 layer bag		
	N.IR	FIR	UVC	N.IR	FIR	UVC	N.IR	FIR	UVC
0	4	0	0	4	0	0	4	0	0
1	3	0	0	5	0	0	6	0	0
2	7	2	3	3	1(dead)	2(dead)	2(dead)	0	0
3	9	3	5	4	0	0	3(dead)	3(dead)	0
4	8	3	4	2(dead)	2(dead)	0	2(dead)	0	1(dead)
5	12	2	7	3(dead)	0	0	3(dead)	0	0
6	32	4	5	7(dead)	0	0	5(dead)	0	0
7	58	7	8	5(dead)	0	0	1(dead)	0	0
8	81	13	11	6(dead)	0	0	0	0	0

N.IR = Non irradiated, FIR = Far infra-red irradiated, UVC = Ultraviolet C irradiated

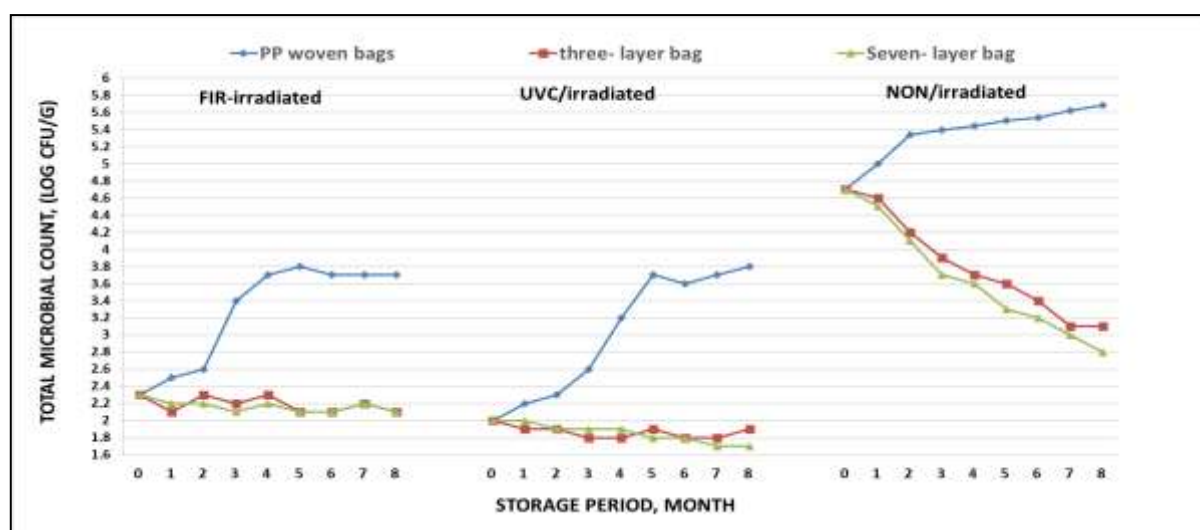


Fig.6. Total microbial count during storage period

For the FIR irradiated seeds stored in woven bags, the total microbial count increased from 2.3 to 3.7 (Log cfu/g) at the end of storage period. Meanwhile, it was decreased from 2.3 to 2.22 Log cfu/g for the three layers bags and from 2.3 to 2.1 Log cfu/g for seven layers bags.

The irradiated seeds with ultraviolet showed total microbial count 3.8 (Log cfu/g) for pp woven bags in comparison with 1.9 and 1.7 Log cfu/g for the three layers and the seven layers bags respectively.

The obtained results showed an increase in the infection of seeds stored in pp woven bags for the three conditions of seeds (not irradiated, FIR irradiated and UVC irradiated), whereas the rate of seeds infection was reduced in the hermetic bags (three layer bag and seven layer bag) for all conditions of seeds. The above-mentioned results agree with **Ahmad and Singh (1991)**. They reported that, hermetic storage makes slightly change of moisture content during the period of storage, and keep lower levels of CO₂ beside sealing the seeds from moisture absorption which results in safe storage of seeds.

Crude Protein Content of Cowpea

Protein content of cowpea influenced by seeds condition (Pre-treated and non-treated) and type of storage bags as shown in Fig. 7. For the non-irradiated woven bags, the crude protein decreased from 30.29% to 27.95%, while it was decreased from 30.29% to 28.75% and from 30.29% to 29.25% for the three layers bags and the seven layers bags respectively at the end of storage period. Meanwhile, the crude protein of the irradiated seeds with infra-red radiation decreased from 28.88% to 27.65%,

28.88% to 28.26% and 28.88% to 28.52% for the seeds stored in woven bags, three layers bags and seven layers bags respectively. For the irradiated seeds with ultraviolet it was decreased from 28.15% to 27.11%, from 28.15% to 27.92% and from 28.15% to 28.1% respectively.

The above mentioned results showed that the contaminated cowpea seeds recorded lower value of protein content. The microorganisms and insects feed on the basic components of seeds for its grow, *A.flavus* uses protein and carbohydrates for its growth and aflatoxin production as mentioned by (**Aziz and Mahrous 2004**).

Conclusion

Conditioning cowpea seeds with infra-red heating of 882.67W/m² and exposure time of 15 min or UVC radiation of 3.538 mW/cm² and an exposure time of 40 min and storage seeds in hermetic bags (three or seven layers) showed a safe storage results in terms of seeds quality and prevention of microorganisms and insects growth. In general, the UV pre-treatment and storage in 7 layers hermetic bag is recommended for safe storing cowpea seeds with keeping the final quality of seeds without deterioration.

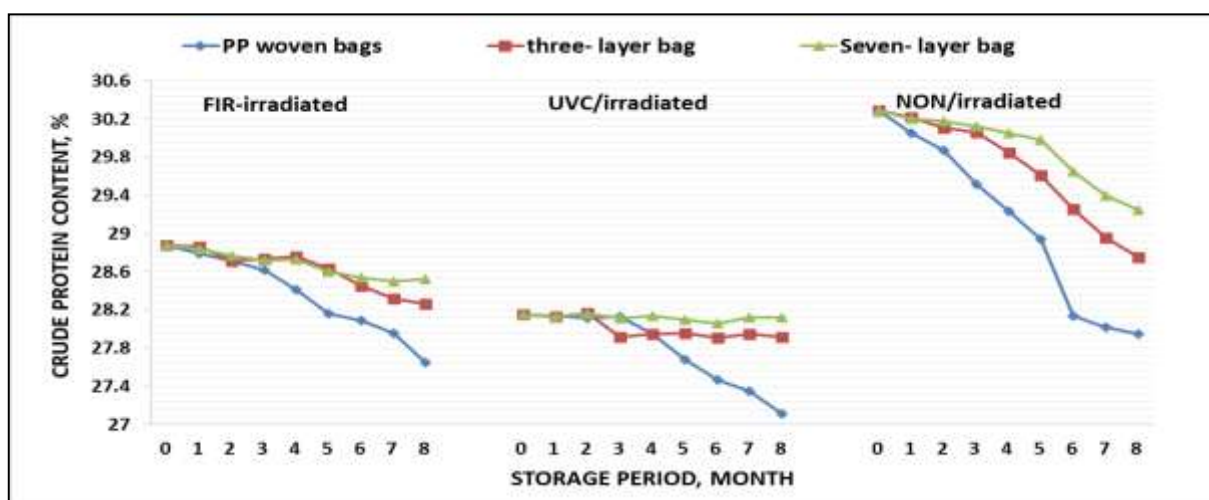


Fig.7. Crude protein content of cowpea

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التهنية والتخزين الآمن لبذور اللوبيا باستخدام الأجلة البلاستيكية المحكمة

داليا حسن صالح¹ - طارق زكي فودة² - محمد مصطفى الخولي¹ - عادل هلال المتولي²

1- معهد بحوث الهندسة الزراعية - مركز البحوث الزراعية - مصر

2- قسم الهندسة الزراعية - كلية الزراعة - جامعة طنطا - مصر

اللوبيا من أهم محاصيل البقوليات التي تتعرض لضرر كبير خلال التخزين وذلك لإحتوائها علي نسبة عالية من البروتين تتراوح بين (17%-30%)، ولذلك لابد من تخزينها عند محتوى رطوبي آمن يتراوح بين (8% - 10%) علي الأساس الرطب. تم إجراء التجارب خلال صيف 2019 بمركز ميكنة الأرز بميت الدبية محافظة كفر الشيخ ، حيث تواجه عملية تخزين البقوليات مشكلة كبيرة بسبب فقد الناتج عن الإصابة الحشرية والبكتيرية والفطرية بعد حصادها مباشرة وخلال فترة التخزين. تهدف هذه الدراسة إلى اختبار وتقييم تأثير طريقتين مختلفتين للمعاملة المبدئية قبل تخزين بذور اللوبيا (الأشعة تحت الحمراء والأشعة فوق البنفسجية) بغرض التعقيم قبل عملية التخزين، وتقييم وقت التخزين الآمن، والتغيرات في جودة اللوبيا أثناء التخزين تحت تأثير المتغيرات الأتية : أنواع مختلفة من الأجلة البلاستيكية النوعية (ثلاث طبقات - سبع طبقات - جوال بلاستيكي منسوج)، حبوب معاملة بالأشعة تحت الحمراء عند شدة إشعاع 882.67 وات/م² لمدة 15 دقيقة، حبوب معاملة بالأشعة فوق بنفسجيه عند شدة اشعاع 3.538 مللي وات/سم² لمدة أربعين دقيقة، حبوب غير معاملة. تم دراسة أثر هذه المعاملات على كل من: نسبة تركيز غاز الاكسجين وثنائي أكسيد الكربون داخل الاجولة خلال فترة التخزين، المحتوي الرطوبي للحبوب، محتوى الحبوب من البروتين، قياس درجة الحرارة والرطوبة النسبية للهواء خلال فترة التخزين، درجة الحرارة والرطوبة النسبية للهواء داخل الاجولة خلال فترة التخزين، حساب العد الميكروبي الكلي بعد عملية التعقيم للحبوب وخلال فترة التخزين ، الكشف عن الحشرات (حشرة / كجم). ومن أهم النتائج المتحصل عليها يمكن التوصية بالآتي:توصي الدراسة بتخزين اللوبيا بالأجلة المحكمة ذات السبع طبقات مع معاملتها معاملة مبدئية للحبوب قبل التخزين بالأشعة فوق بنفسجية عند شدة اشعاع 3.538 مللي وات / سم² لمدة أربعين دقيقة.

المحكمون:

1- ا.د. جمال الدين محمد نصر

2- ا.د. محمود عبد الرحمن الشاذلي

أستاذ الهندسة الزراعية المتفرغ - كلية الزراعة - جامعة القاهرة.

أستاذ الهندسة الزراعية المتفرغ - كلية الزراعة - جامعة الزقازيق.