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Effects of packaging materials on weight loss, moisture content and quality of wheat flour infested by *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae)

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Abstract
The study investigated the effects of packaging materials on the storage losses of wheat flour due to infestation by *Tribolium casterneum* with a view to ensuring good storage and consumption of quality wheat flour. The ability of seven packaging materials: black low density polythene bag (10µm), white low density polythene bag (10µm and 20µm), blue high density polythene bag (25µm), brown paper bag (70g), white paper bag (80g) and laminated film (10µm) to control weight loss, moisture content and quality changes such as colour and fungal growth were evaluated for the period of 91 days under infestation with *Tribolium casterneum*. Laminated plastic (10µm) and white paper bag (80g) controlled all the defects assessed. Highest weight loss (22.67%) was recorded in black low density polythene (10µm) followed by white low density polythene (10µm) (13.80%). Blue high density polythene (25µm) allowed the highest gain in moisture content while caking, colour change and fungal growth were observed with white low density polythene (20µm) and blue high density polythene (25µm).

Keywords: *Tribolium castaneum*, fungal growth, moisture

Introduction

Insect pests of flour include *Sitophilius granaries, S. oryzae, S. zeamais, Tribolium confusum* and *Tribolium castaneum*. Red flour beetle has long been considered an economic pest that can be found just about anywhere in homes flour mills and grocery stores (Bhadriraju and Jennifer, 1999) [1]. Dunkel (1988) [2] reported that during feeding and reproduction, the metabolic activity of insects could cause an increase in moisture level and temperature of grains, making it suitable for fungal growth. According to Onwueme (1978) [3] storage environment of yam flour must be dry to prevent the growth of mould as microbial contamination of flour leads to loss of quality. The criteria for quality in packaged food products include availability of wholesome, clean, unadulterated food items with minimal losses during transport, hygienic conditions, and a reasonable price for the consumer. Purvis (1983) [4] and Scott (1984) [5] had shown that polythene bags could be used to create modified atmosphere and such bags are called physiological packaging. The procedure is based on the selective permeability of some polymer membrane to the respiratory gases (O_2, CO_2). McFarlane (1970) [6] observed that insect mortality in airtight pouches most likely occurs from oxygen depletion and carbon (iv) oxide build up. Highland (1984) [7] revealed that losses generally occur because insects either bore through package materials or even enter the package through existing holes either caused by imperfect seals or over wraps, mechanical damage or damage resulting from previous insect infestation. Both red and confused flour beetles are found in many parts of the world. Red flour beetles inhabit warmer climates, these beetles primarily occur in flour mills, feed mills, warehouses, retail grocery stores, boxcars, semolina mills, and bakeries (Trematerra *et al*., 2007) [8]. They also occur in empty cargo containers, farm grain bins, farm storages, grain elevators, peanut shelling plants, residences, and pet stores (Hagstrum and Subramanyam, 2009) [9]. To improve resistance to insect attack, many companies have implemented package-testing programs (Mullen and Mowery, 2000) [10]. The packaging of products is the last line of defense for processors against insect infestation of their finished products (Hou *et al*., 2004) [11]. Polymers with various thicknesses have different permeability (Marouf and Momen, 2004) [12]. This study investigated different packaging materials of low thicknesses in controlling weight loss, moisture content, colour change and fungal growth of wheat flour due to infestation by *T. casterneum*.

Materials and methods

Collection of materials

The wheat flour used for this study was a product of Supreme flour mill, Lagos, Nigeria. This was obtained from Oba market, Akure, Ondo State, Nigeria.

Packaging materials used were sourced for locally from their manufacturers. These include: brown paper bag (70g), white paper bag (80g), laminated film (10µm), black low density polythene bag (10µm), white low density polythene bag (10µm and 20µm) and blue high density polythene bag (25µm)

Adult beetles of *Tribolium castaneum* maintained on wheat flour at ambient temperature (28-32ºC) and 72 ±10% relative humidity were obtained from Storage Research Laboratory, Federal University of Technology, Akure, Ondo State, Nigeria.

Insect culture

Two hundred and fifty grammes (250g) of wheat flour were put into a transparent, two litre capacity, perforated and netted plastic container with screw-type lid to enhance the respiratory activities of *Tribolium castaneum*. Identified *Tribolium castaneum* from Storage Research Laboratory, Federal Universsity of Technology, Akure, were used for the culture. Fifty (50) beetles of mixed sexes were introduced into the container and monitored in an open laboratory where the temperature fluctuated between 28 and 32°C and relative humidity from 72 to 82%. The culture was monitored to obtain newly hatched adults and larvae of desired instar stages needed for the experiment in four to six weeks.

Sex determination of *Tribolium castaneum*

To determine the sex of *Tribolium castaneum,* adult beetles were first inactivated by placing them into Petri dishes over crushed ice for a few seconds and then examined under a binocular microscope for sex characters as described by Halstead (1963) [13]. The sex was confirmed by observing the tip of the abdomen which the adult starts to retract before fully recovering from the short chill. The tip of the abdomen is smoothly rounded in the male while the small ovipositor is clearly visible in the female.

Experimental procedure

Thirty grammes (30g) of wheat flour were weighed out into each of the packaging materials (12cm x 8cm) in triplicate along with the control. Five copulating pairs of newly hatched adult *Tribolium castaneum* were introduced into each packaging material in triplicate. Ten first instar larvae were introduced into another set of the packaging materials and sealed. Each packaging material of different treatment was stored in a two litre capacity, transparent, perforated and netted plastic container with screw-type lid. The experiment was conducted at ambient temperature (28-32ºC) and 72 to 82% relative humidity. Weight loss, quality change and change in moisture content were determined as follows:

Weight Change assessment

The simple method of using weighing balance was used for the weight change determination and the initial weight of the sample in each packaging material was determined and recorded (w_i). The final weight was also taken at the end of the experiment and recorded (w_f). The percentage weight change was determined as follows:

% Weight change = $\underline{W_i - W_f}$ X 100 W_i

Where: W_i = Initial weight, W_f =Final weight

Quality change assessment

The quality change assessment for caking, colour change and fungal growth was done by visual examination of the stored flour after 91 days of storage.

Moisture content determination

The percentage (%) water loss was determined using the method of the Association of Official Chemist (AOAC), (2005) (14). This was achieved by clean and dried porcelain dishes which were weighed using mettler balance and their respective weights were recorded. About two gram (2g) of the samples were weighed into respective porcelain dishes and transferred into a dessicator immediately after each weighing in order to prevent absorption of moisture from the atmosphere. The porcelain dishes containing the samples were transferred from the dessicator into the oven, maintained at 105°C and dried for about three hours (3 hours), after which they were removed, cooled in a dessicator and weighed. They were later returned to the oven and re-dried for further one hour, cooled and weighed. The procedure was repeated until a constant weight was obtained.

The loss in weight during drying was expressed as a percentage.

% Change in moisture content = Average loss in weight X 100

Average weight of sample before drying

Statistical analysis

All data collected were analyzed using the one-way analysis of variance (ANOVA) method (Steel and Torrie, 1980) (15) and where significant differences existed the means were separated by Duncan's Multiple Range Test (DMRT) technique (Duncan, 1955) [16].

Results

Table 1: Weight loss in infested packaged flour at 91 days

Type of packaging materials	Mean % weight loss in pack containing adult	Mean % weight loss in pack containing larvae	Mean % weight loss in pack without insect (control)
Brown PB (70g)	9.20±0.30e	9.15±0.05f	2.39±0.09f
White PB (80g)	1.23±0.03a	0.33±0.04a	0.98±0.11e
Laminated film (10µm)	0.90±0.10a	0.30±0.05a	0.07±0.01a
White LDP (10µm)	13.80±0.20f	0.48±0.02b	0.46±0.09d
White LDP (20µm)	5.47±0.04d	0.83±0.01c	0.52±0.02c
Black LDP (10µm)	22.67±0.01g	1.70±0.01e	0.49±0.01c
Blue HDP (25µm)	2.89±0.11b	1.30±0.01d	0.31±0.03b

Means followed by the same letter(s) in the same column are statistically the same $p < 0.05$: *Duncan*'s Multiple Range Test (DMRT) Legend: PB =Paper bag, LDP = Low density polythene, HDP =High density polythene

Table 2: Moisture content of infested packaged flour at 91 days of storage (Initial Moisture Content = 11.30)

Type of packaging materials	Pack containing adults		Pack containing larvae		Pack without insects	
	Final mean MC	Change in MC	Final mean MC	Change in MC	Final mean MC	Change in MC
Brown PB (70g)	11.36±0.01c	-0.06	11.68±0.02e	-0.38	11.27±0.02b	0.03
White PB (80g)	11.19±0.01b	0.11	11.49±0.01d	-0.19	11.19±0.02a	0.11
Laminated film (10µm)	11.36±0.01c	-0.06	11.31±0.02c	-0.01	11.28±0.02b	0.02
White LDP (10µm)	11.27±0.02c	0.03	10.77±0.02a	0.53	11.26±0.02b	0.04

White LDP (20µm)	11.92±0.02d	-0.62	11.94±0.02f	-0.64	11.31±0.02c	-0.01
Black LDP (10µm)	11.03±0.01a	0.27	10.04±0.01b	0.29	11.29±0.02bc	0.01
Blue HDP (25µm)	12.59±0.02e	-1.29	12.99±0.03g	-1.69	11.39±0.01d	-0.09

Means followed by the same letter(s) in the same column are statistically the same $p < 0.05: Duncan$'s Multiple Range Test (DMRT)
Legend: PB =Paper bag, LDP = Low density polythene, HDP =High density polythene, MC = Moisture content
+ve = decrease in moisture content, -ve = increase in moisture content

Table 3: Quality of packaged flour after *Tribolium castaneum* infestation for 91 days by visual assessment

Type of packaging materials		Parameters	
	Caking	Colour change	Fungal growth
Adult pack			
Brown PB (70g)	-	-	-
White PB (80g)	-	-	-
Laminated film (10µm)	-	-	-
White LDP (10µm)	-	-	-
White LDP (20µm)	*	* (Brown)	*
Black LDP (10µm)	-	-	-
Blue HDP (25µm)	**	** (Brown to black)	**
Larval pack			
Brown PB (70g)	-	-	-
White PB (80g)	-	-	-
Laminated film (10µm)	-	-	-
White LDP (10µm)	-	-	-
White LDP (20µm)	*	* (Brown)	*
Black LDP (10µm)	-	-	-
Blue HDP (25µm)	*	* (Brown)	*

Legend: * = low, ** = high, - = absen

The percentage weight loss in the wheat flour at 91 days of storage with adults of *Tribolium castaneum* (Table 1) varies with the different packaging materials. The percentage weight losses recorded in the packaged stored flour were significantly ($p<0.05$) different. Highest weight loss (22.67%) was recorded in black low density polythene (10µm), followed by white low density polythene (10µm) (13.80%) and minimum weight loss (0.90%) in laminated film (10µm). With the larvae of *Tribolium castaneum* (Table 1), percentage weight losses range from 0.30 to 9.15. The highest weight loss was recorded in brown paper bag (70g) and the lowest weight loss occurred in laminated film (10µm). The pack without insect (control pack) showed little or no weight loss (Table 1)

The final moisture content of infested wheat flour determined at 91 days of storage is shown in Table 2. This table also shows the respective percentage moisture content change recorded in the infested packaged flour at different treatment levels. The results obtained for the final moisture content were significantly ($p<0.05$) different (Table 2). The table further shows the percentage change in moisture content as discussed from initial (11.30) and final moisture content. The negative values depict increase in moisture content while the positive values depict decrease in moisture content. The results were significantly ($p<0.05$) different.

Table 3 shows the visual assessment of packaged flour after *Tribolium castaneum* infestation for 91 days. The wheat flour stored in brown paper bag showed a low level of colour change at 91 days of storage with the adult of *Tribolium castaneum.* The blue high density polythene (25µm) pack with adult *Tribolium castaneum* showed a high level of caking, colour change and a high level of fungal growth while adult pack of low density polythene (20µm) and larval pack of white low density polythene (20µm), and blue high density (25µm) packs with larvae showed a low level of caking, colour change and fungal growth. These defects were not observed in the remaining packaging materials.

Discussion

Weight loss in adult and larval packs can be mainly attributed to beetle feeding. This is consistent with the report of Emechute (1998) [17]. Since little moisture loss was recorded in some of the packs without the beetle of *Tribolium castaneum* (Table 1) , the weight loss observed in packs without *Tribolium castaneum* is probably due to drying and the permeability of films to the rate of transport in gram of water gained or lost per day (Theodore, 1982) [18]. This could also be incriminated for the change in moisture content observed in packaging materials under the different treatment levels (Table 2). The highest weight loss (22.67) observed in black low density polythene (10µm) of adult pack could be due to the ability of black materials to absorb heat faster leading to temperature rise in favour of proliferation of *Tribolium castaneum* which means increase in food consumption level (Shazali and Smith, 1986) [19]. The low percentage weight losses, 2.89 and 3.70 in high density polythene (25µm) could be attributed to inability of *Tribolium castaneum* to feed due to possibility of early mortality conferred upon by these packaging materials. The perforation that occurred in brown paper bag (70g) (adult and larval packs) and white low density polythene (10µm and 20µm) possibly enhanced the growth-development of the beetle, which invariably mean food is consumed for growth, and thus may be the cause for the weight change of the stored wheat flour. Laminated film (10µm) was found highly effective for controlling weight loss when compared with other packaging materials. Brown paper bag (70g) and black low density polythene (10µm) had poor control effect on the larvae of *Tribolium castaneum* and on weight losses (Table 1).

The gain in moisture content of the wheat flour stored in some packaging materials (Table 2) could primarily be attributed to feeding, reproduction and the metabolic activity of insects (Dunkel, 1988) [2] and hygroscopic nature of dried product, storage environment (temperature, relative humidity) as well as the nature of packaging material (Marouf and Momen, 2004, Nagi *et al.*, 2012) [12, 20] Relative humidity is another factor that could account for change in moisture content. If there is a difference in the relative humidity of the wheat flour stored inside the packaging materials and the immediate environment, the lower relative humidity tends to absorb moisture from higher relative humidity in attempt to reach equilibrium (Theodore, 1982) [18]. It was noted that the flour stored in blue high density polythene (25µm) and white low density polythene (20µm) showed the highest moisture content increase both in adult and larval packs (-1.29 and – 1.69) respectively. This may be due to the fact that, the package materials limit the exchange of the internally produced moisture due to relative humidity difference and physiological processes of the beetles (*Tribolium castaneum*). The white and black low density polythene (10µm) had a reduction in moisture content (Table 2) in the infested packaged flour. The beetle perforations on these two packages must have allowed the moisture produced to escape freely to the immediate environment, thus lead to decrease in moisture content. Considering the paper bags (brown and white paper bags) increase in moisture content was observed mainly in the larval pack of the infested packaged flour (Table 2). The reason for this might be due to the fact that larvae respired much more rapidly than the adult stage (Emekci *et al.*, 2002) (21) which means a high level of moisture production. However, the normal moisture content of a processed food should be maintained during storage. This is a main function of packaging for some foods. In this study, none of the packaging materials actually maintained the original moisture content of the stored wheat flour but laminated film pack (10µm) showed the minimum gain in moisture content of the stored wheat flour in both adult and larval packs (Table 2). This could be due to the impervious nature of aluminium foil in the laminate to air and water vapour (Nagi *et al.*, 2012) [20]. It can therefore be said that this package proves best a package materials for moisture control of wheat flour.

The caking, colour change and fungal growth observed (Table 3) after 91 days of storage could be as a result of combined effect of moisture and microbial proliferation in the product (Saltmarch and Labuza, 1980; Bothast *et al.*, 1991) [22, 23]. Packaging materials with high moisture permeability may predispose its content to high rate of deterioration (Butt *et al.*, 2003) [24]. Discolouration and caking occurred due to fungal pigment and severe fungal growth on the flour respectively. This is in conformity with the work of Odeyemi and Daramola (2000) [25]. This colour change could also be due to the production of hydroquinones into flour by *Tribolium castaneum* which is associated with cancer development (Christensen, 1974) [26].

Conclusion

The weight loss, moisture gain and change in quality that occurred in some of the packaging materials generally make flour unacceptable to the consumers as it poses serious health risk. It can be concluded that the packages in which these changes occurred should not be used for long term storage of flour especially under the conditions that favour mould growth such as high relative humidity.

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