21

Development of Calcium Enriched Biscuits using Egg Shell as Calcium Source

J. Munahirah and K. Premakumar

Department of Agronomy, Faculty of Agriculture, Eastern University, Vantharumoolai, Chenkalady, Sri Lanka

Abstract

A study was conducted to develop calcium enriched biscuits using egg shell as a calcium source. The ratio of egg shell powder to flour mixture was maintained as follows 600mg/100g flour mixture to, 700mg/100g flour mixture to, 800mg/100g flour mixture and 900mg/100g flour. The biscuits were subjected to organoleptic and nutritional analysis to evaluate the suitability of the products for consumption and self life. There were no significant changes observed in nutritional characteristics such as crude fat, protein and total soluble solids but differences were observed in the moisture, ash and calcium content. In the organoleptic evaluation, significant differences were found among the treatment. According to the organoleptic evaluation, the biscuits developed from the egg shell concentration of 800mg/100g flour mixture were selected as best based on crispyness, flavour, taste and overall eating quality.

Packaging and shelf-life studies of calcium enriched biscuits were conducted using four types of packing material namely glass bottle, plastic bottle, polyethylene and aluminium foil and stored at room temperature. Nutritional, microbiological, sensory evaluations

were conducted at weekly intervals to evaluate the quality of product during storage. Significant differences in nutritional qualities such as fat, protein and ash content and sensory evaluation criteria such as colour, crispyness, flavour, taste and overall eating quality were observed. The product packed in glass bottle did not exhibit significant deteriotive changes in nutritional composition compared to other packaging materials as shown by the results of the microbiological analysis done on the product.

Introduction

Sri Lankas low calcium intake has reached a crisis level. In Sri Lanka about 30% both males and females have reported low bone mass in suburban communities (Sribaddana, 2004). Low calcium intake could be one of the contributory factor for it. Throughout the life cycle, calcium is essential for proper growth and development and good health. Calcium requirement are high in adolescents and accumulate 37% of the total bone mass during the growth spurt (Key & Key, 1994). Inadequate dietary calcium is associated with a number of common chronic medical disorders worldwide, such as osteoporosis, cardiovascular disease, hypertension, stroke and ischemic heart disease, diabetes and hypertensive disorders of pregnancy, obesity and colon cancer. (FAO, 1997). According to the National Osteoporosis Foundation, 28 million people are affected by osteoporosis and that number may rise to 41 million by 2015.Low calcium intake is proven to be one of a number of factors contributing to Osteoporosis. Calcium is obtained primarily from natural foods, particularly dairy products. Calcium-fortified foods and calcium supplements are options to help optimise calcium intake. One of the most-common, economical forms of calcium is calcium carbonate. Schaafsma et al. (2000) has proposed that chicken egg shell powder might be an attractive source of calcium for human nutrition, especially as a source of calcium and strontium. The present study was undertaken to develop calcium enriched biscuits from various concentration of egg shell powder and based on the chemical and sensory analysis best egg shell concentration will chosen for further storage study.

Materials and Methods

Egg shells were collected, cleaned and boiled in water. After that they were dried and grounded in a grinder. Then they were sieved with a help of muslin cloth. Fine purified egg shell powder was obtained. Hydrogenated fat (50gram) and powdered sugar (50gram) were creamed together. Different amounts of egg shell powder were added to the one hundred grams of all-purpose flour to produce following mixture (900mg/100g mixture, 800mg/100g mixture, 700mg/100g mixture and 600mg/100g mixture). Every mixture was then sieved twice together with a quarter teaspoon of backing powder and it was added with above creamed paste. From this mixture, firm dough was prepared by mixing them manually. The dough was rolled out to 2.5 mm thickness and cut into rectangular slices having 2.5 cm in diameter, which were placed on greased aluminium trays and baked in a preheated oven at 150^0C for 4 minutes to produce biscuits.

Sensory Evaluation

The sensory evaluation was conducted by 9-points hedonic scale rating test, using untrained panel consisting of 20 staff and student members. The following Organoleptic parameters were considered in the sensory test, Colour, Crispyness, Taste, Flavour and Overall eating quality.

Chemical Evaluation

Chemical parameters such as Moisture, Ash, Crude fat, Crude protein, Total sugar and Calcium analyzed for freshly developed product. Moisture was determined by the method AOAC (1998). Total sugar was determined by the method Lyon and Eynon. Ash was determined by the method AOAC (1998). Crude fat was determined by the method AOAC (1998). Crude protein was determined by the method Pearson (1998). Calcium was determined by the method AOAC (1990).

Data Analysis

The experiment was conducted using complete block design; the scores obtained from each study were statistically analyzed using analysis of variance and Duncan's multiple range tests for comparison of

means for colour, crispyness, flavour, taste and overall eating quality of biscuits. Significance was observed at P≤ 0.05 for each sensory criteria.

The software SAS was used for all analysis.

Evaluation of Chemical Characteristics of Stored Biscuits

Chemical parameters such as Moisture, Ash, Crude fat, Crude protein, Total sugar and Calcium analyzed for freshly developed product. Moisture was determined by the method AOAC (1998). Total sugar was determined by the method Lyon and Eynon. Ash was determined by the method AOAC (1998). Crude fat was determined by the method AOAC (1998). Crude protein was determined by the method Pearson (1998).Calcium was determined by the method AOAC (1990).

Microbial Evaluation of Stored Biscuits

Microbial evaluation was conducted at 7 days intervals for stored biscuits.

Results and Discussion

Experiment – 1 Product Development

Proximate Composition of Calcium Enriched Biscuits

Moisture percentage, Protein percentage, Fat percentage, Ash percentage, Total sugar percentage and calcium content of the freshly made biscuit are given in Table 1.

Organoleptic Evaluation of Calcium Enriched Biscuits

The results of DMRT test for the organoleptic evaluation of calcium enriched biscuits prepared by different concentration of egg shell powder/ flour mixture are shown in Table 2.

Colour

Colour is one of the important units in the sensory evaluation. An attractive colour leads to the food to make good demand. According to the panelist comments, no significant difference was observed between treatments code 337, 617 and 119. They had acceptable golden brown colour. To select the best product's colour, the mean score was used.

According to that, the biscuit prepared by concentration of egg shell powder800mg/100g flour mixture was selected as best.

Crispyness

It is clear that crispyness is related to fracture behaviour. Crispyness is characterized by multiple fractures accompanied by the emission of sound. The crispyness of many foods is readily lost due to redistribution of water and/or water uptake due to exposure to humid air. Clearly this point will be reached as a result of too high a moisture content after baking, before packing or as a result of pack performance (Roessler et al 1948). The biscuits made by the egg shell concentration of 800mg/100g flour mixture (sample code 617) were significantly differed from other product. Biscuits prepared by other egg shell concentrations were not significantly different. Biscuit prepared by concentration of egg shell powder800mg/100g flour mixture had the highest preference due to its long lasting crispyness nature. Visser (2000) revealed that the increasing moisture uptake reduced the crispyness behaviour in biscuits.

Flavour

Flavour plays a vital role as a sensory parameter in determining the acceptability of the new product. According to the panelists comments, all the samples contained very good flavour, particularly biscuits made by the egg shell concentration of 800mg/100g flour mixture had more acceptable flavour compared to the rest. In this product development no artificial flavour were added. Typically, calcium doesn't have an overwhelming effect on a fortified food's flavour, but in a bland system, certain flavours arise (Kanis and Passmore, 1989).

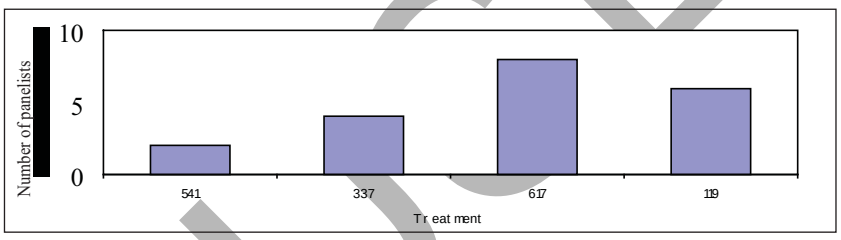
Taste

This is the primary factor, which determines the acceptability of any product, which has the highest market success of product, is concerned. Based on the panelists biscuits made by the egg shell concentration of 800mg/100g flour mixture (sample code 617) and 900mg/100g flour mixture (sample code 119) were not significantly different. These samples were significantly different from other two samples. To select the best sample based on the taste, mean score was used. According to

that, a biscuit made by the egg shell concentration of 800mg/100g flour mixture was selected as best.

Overall Eating Quality

According to the panelists, biscuits made by the egg shell concentration of 800mg/100g flour mixture (sample code 617) and 900mg/100g flour mixture (sample code 119) were not significantly different. To select the best sample based on the overall eating quality, mean score was used. Biscuits made by the egg shell concentration of 800mg/100g flour mixture (sample code 617) received highest scores in the colour, crispyness, flavour, taste and overall eating quality. Next was biscuit made by the egg shell concentration of 900mg/100g flour



mixture (sample code 119). Biscuits made by the egg shell concentration of 600mg/100g flour mixture (sample code 541) and 700mg/100g flour mixture (sample code337) had received relatively low scores.

Figure 4.1 Number of panelists for the preference of treatments.

Figure 4.1 indicates that the biscuits prepared by the egg shell concentration of 800mg/100g flour mixture preferred by most of the panalists, followed by biscuits prepared by the egg shell concentration of 900mg/100g flour mixture.

Experiments 2 –Storage Study of Biscuits

4.2.1 Proximate Composition of Calcium Enriched Biscuits

Biscuits made by the egg shell concentration of 800mg/100g flour mixture were stored for one month in different type of packaging materials, vz plastic bottle, polyethylene, glass bottle and Aluminium foil. The changes observed in proximate composition of calcium enriched biscuits at weekly intervals are given in figures 2, 3, 4, 5 and 6.

4.2.1.1 Moisture

The changes in moisture percentage of different treatments along with the storage period are given in figure 4. 2

Biscuits are very hygroscopic. They typically have an equilibrium relative humidity (e.r.h.) of about 30% so in most cases must be protected from the atmosphere to prevent or at least delay, moisture pick up. Moisture content of the biscuits fluctuated slightly. Manley (1986) reported that moisture content in baked goods vary from 4 to 7%. This fluctuation was high in biscuits stored in plastic bottle. The moisture gain of the biscuits occurred due to water vapour transmission through the lid of the plastic bottle. Because gas transmission rate was high in plastic bottle (Oswine, 1983). Meanwhile fluctuation was low in biscuits stored in glass bottle (Manely, 1986).

4.2.1.2 Fat

The changes in fat percentage of different treatments along with the storage period are given in figure 4.3

Shortening, fat or oil contribute to the tenderization of baked products through inhibition of gluten development and starch gelatinization. They do this through a waterproofing effect, possibly due to the complexing with the carbohydrate and/or protein. Fat can help leaven a product due to incorporation of air (Brooker, 1998). Development of off flavours, unpleasant flavours developed as a result of oxidation, particularly fats. This is known as oxidative rancidity. Development of this type of rancidity is particularly related to the presence of moisture, certain metal ions and certain wavelengths of light. Rarely, there may be some hydrolic rancidity associated with fats and moisture especially in alkaline condition. Berger (1970) reported that moisture uptake and gas were exchange causes of off odour development in biscuits. Fat percentage was initially same in all packaging materials. But the changes were high in biscuits stored in plastic bottle. Decreasing trend was observed in biscuits stored in all packaging materials and this reduction was due to the oxidation of fatty acids with the atmospheric oxygen and moisture uptake. The reduction was highly observed in biscuits stored in plastic bottle. But fluctuation was low in biscuits stored in glass bottle

4.2.1.3 Protein

The changes in protein percentage of different treatments along with the storage period are given in figure 4.4

A number of studies have shown the relationship between protein content and texture. (Primo *et al*., 1962). Juliano *et al.,* 1965 demonstrated that increased protein content within a flour resulted in firmer cooked bread. The various flour proteins present in wheat (i.e. gluten and gliadian) can undergo changes such as protein cross-linking, protein-carbohydrate interactions and protein denaturation during processing. Teo *et al*., (2000) reported that the modification of the protein component, rather than the starch, was primarily responsible for rheological changes associated with aging of flour. Calcium may react with protein, particularly with heat and cause sedimentation and gelation (Lawson *et al*., 1974). Protein of the biscuits was reduced with storage periods in all packaging materials. But the changes were low in biscuits stored in glass bottle, while the reduction was high in biscuits stored in plastic bottle. There was no significant difference observed in protein during the storage in different packaging materials.

4.2.1.4 Ash

The changes in ash percentage of different treatments along with the storage period are given in figure 4.5

Ash content of the biscuits was reduced with storage periods in all packaging materials. But the changes were low in biscuits stored in glass bottle, while the reduction was high in biscuits stored in plastic bottle. There was no significant difference observed in ash percentage during the storage in different packaging materials.

4.2.1.5 Total Sugar

The changes in total sugar percentage of different treatments along with the storage period are given in Table 4.3

Total sugar in all treatments have approximately same with storage period, because the sugars are not used as substrates for respiration and no any hydrolysis occurred. Changes were low in plastic bottle.

4.2.1.6 Calcium Content

The changes in calcium percentage of different treatments along with the storage period are given in Table 4.4

There were no any changes observed in calcium content of the biscuits. Hasling *et al.,* (1990) reported that calcium has good heat stability and storage stability. Parfitt, (1990) reported that calcium is fairly stable and heat or oxidation won't change the calcium content.

4.2.2 Organoleptic Evaluation

Results of DMRT for the Organoleptic Evaluation of Biscuits Stored in Different Packaging Materials at 14th Day of Storage are shown in Table 4.5

Significant difference was observed in the sensory score of the biscuits stored in different packaging materials. Biscuits stored in glass bottle had highest score in quality attributes (colour, crispyness, flavour, taste and overall eating quality).At 7th days of storage, sensory scores for the quality attributes were same as the initial scores among biscuits stored in different packaging materials. At 14th day of storage, sensory scores such as colour, taste and overall eating quality of the biscuits stored in glass bottle and aluminum foils were not significantly different at $p>0.05$. Crispyness and flavour of the biscuits stored in glass bottle were significantly different at $p<0.05$

Results of DMRT for the Organoleptic Evaluation of Biscuits Stored in Different Packaging Materials at 28th Day of Storage are shown in Table 4.6

At 28th day of storage analysis showed a significant difference among biscuits stored in different packaging materials. Quality attributes (colour, crispyness, flavour, taste and overall eating quality) of the biscuits stored in glass bottle were significantly differed with other packaging materials. According to the sensory scores, glass bottle was selected as best packaging materials. Subsequently Aluminium foil then polyethylene and Plastic bottle ranked in order.

4.2.3 Microbial Characteristics Evaluation

The results of the microbial study for 0,7th, 14th, 21st and 28th day of storage given in Table 4.7

There were no microbial colonies found during the storage periods (0,7th, 14th and 21st days). However, at the end of the storage periods (28th day), a small amount of colonies observed in biscuits stored in plastic bottle and polthyne pack. They were stained and identified as *Fusarium* spp and *Cuvularia* spp.

Conclusion

The finding of this study revealed that biscuits made by the egg shell concentration of 800mg/100g flour mixture were superior in both organoleptically and chemically than biscuits made by the egg shell concentration of 600mg/100g flour mixture, 700mg/100g flour mixture and 900mg/100g flour mixture. Biscuits made by the egg shell concentration of 900mg/100g were chosen as best by the panelists next to biscuits made by the egg shell concentration of 800mg/100g flour mixture.

Biscuits stored in glass bottle were preferred both organoleptically and chemically than biscuits stored in other packaging materials (Aluminium foil, polyethylene and plastic bottle). The moisture percentage was increased with storage period. During storage fat hydrolysis increased so that fat percentage was reduced. Protein denaturation and changes were low so that protein percentage was reduced. Ash percentage was reduced with storage but the changes were low. In total sugar percentage and calcium content there wasn't any changes occurred, because of the storage stability of these parameters were high. The changes in quality attributes (colour, crispness, flavour, taste and overall eating quality) were low in biscuits stored in glass bottle. But the changes were high in biscuits stored in plastic bottle.

Annexture

Table 1. Proximate composition of calcium enriched biscuits

Component	541	337	617	119
Moisture	4 ± 0.1	3.9 ± 0.1	3.8 ± 0.1	3.8 ± 0.1
Protein	22.14 ± 0.05	22.12 ± 0.05	22.12 ± 0.053	22.12 ± 0.053
Fat	18.75 ± 0.05	18.75 ± 0.05	18.75 ± 0.05	18.75 ± 0.05
Total sugar	45.93 ± 0.115	46 ± 0.115	46 ± 0.115	46 ± 0.115
Ash	2.4 ± 0.1	2.4 ± 0.1	2.45 ± 0.1	2.5 ± 0.1
Calcium	0.061 ± 0.01	0.071 ± 0.01	0.081 ± 0.001	0.091 ± 0.001

Values are means of three replicates± Standard Deviation of the replicates determinations.

Table 2. Organoleptic Evaluation of Calcium Enriched Biscuits Prepared by using Different Concentration of Egg shell powder/flour mixture.

Treatment	Colour	Crispyness	Flavour	Taste	Overall eating quality
541	6.15^{b}	5.9^{b}	5.7^{bc}	6.25^{b}	6.05^{b}
337	7.15^{a}	4.5^{b}	5.3^{c}	5.25^{c}	5.2^{c}
617	7.35^{a}	7.85^{a}	6.8^{a}	7.6^{a}	7.9^{a}
119	7.5^{a}	7.6^{b}	6.5^{bc}	7.4^{a}	7.6^{a}

(Values are means of the 20 samples; means with the same letters in each columns are not significantly differed at 5%probability level)

Scale: 9 = Like Extremely 1 = Dislike Extremely

Treatment code Treatment

541 Concentration of egg shell powder is 600mg/100g flour mixture

337 Concentration of egg shell powder is 700mg/100g flour mixture

617 Concentration of egg shell powder is 800mg/100g flour mixture

119 Concentration of egg shell powder is 900mg/100g flour mixture

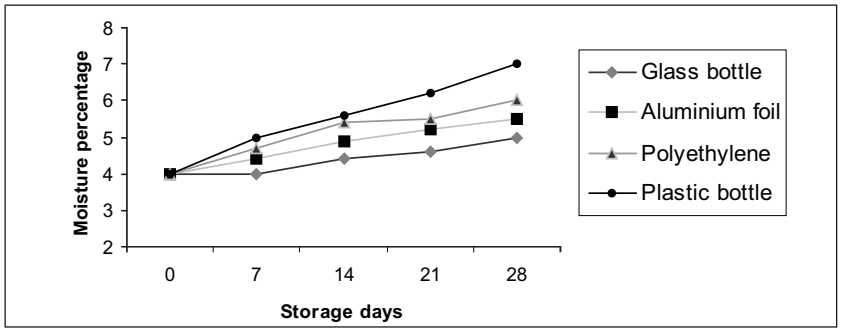


Figure 4.2 - Changes in moisture percentage of biscuits stored in different type of packaging materials.

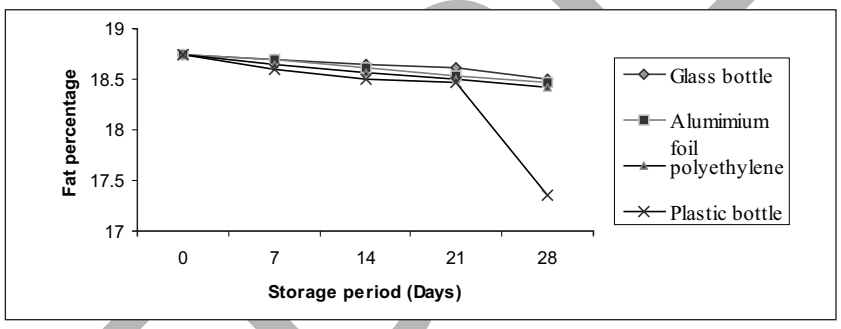


Figure 4.3 - Changes in fat percentage of biscuits stored in different type of packaging materials

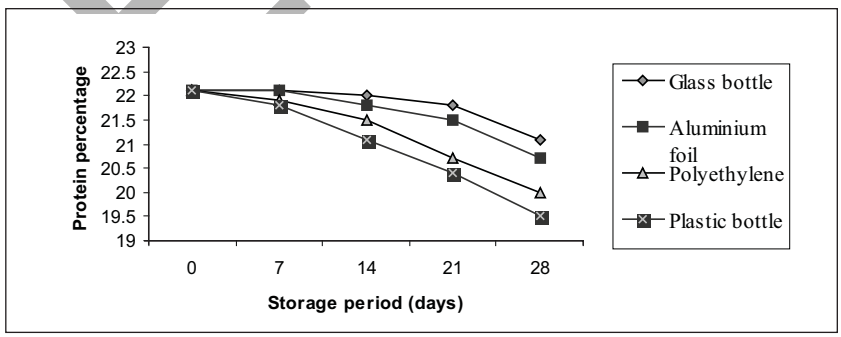


Figure 4.4 - Changes in Protein percentage of biscuits stored in different type of packaging materials

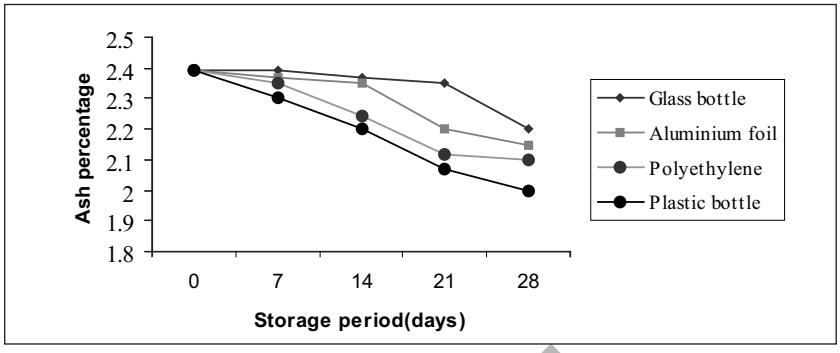


Figure 4.5 - Changes in ash percentage of biscuits stored in different type of packaging materials.

Table 4.3 Changes in Total sugar percentage of different treatments with storage period.

Storage days	T1	T2	T3	T4
0	45.9 ± 0.11	45.9 ± 0.11	45.9 ± 0.11	45.9 ± 0.11
7	45.9 ± 0.11	45.9 ± 0.11	45.9 ± 0.11	45.9 ± 0.11
14	45.9 ± 0.11	45.9 ± 0.11	45.9 ± 0.11	45.8± 0.11
21	45.9 ± 0.11	45.9 ± 0.11	45.9 ± 0.11	45.75 ± 0.11
28	45.9 ± 0.11	45.9 ± 0.11	45.9 ± 0.11	45.7 ± 0.11

The values are means of triplicates ± standard deviation.

Table 4.4 Changes in calcium percentage of different treatments with storage period.

Storage days	T1	T2	T3	T4
0	0.08±0.01	0.08±0.01	0.08±0.01	0.08±0.01
7	0.08±0.01	0.08±0.01	0.08±0.01	0.08±0.01
14	0.08±0.01	0.08±0.01	0.08±0.01	0.08±0.01
21	0.08±0.01	0.08±0.01	0.08±0.01	0.08±0.01
28	0.08±0.01	0.08±0.01	0.08±0.01	0.08±0.01

The values are means of triplicates ± standard deviation.

Table 4.5 Results of DMRT for the Organoleptic Evaluation of Biscuits Stored in Different Packaging Materials at 14th Day of Storage.

Biscuits	colour	Crispyness	Flavour	Taste	Overall eating quality
727	6.15^{b}	5.9^{b}	5.7^{bc}	6.25^{b}	6.05^{b}
427	7.15^{a}	4.5^{b}	5.3^{c}	5.25^{c}	5.2^{c}
389	7.35^{a}	7.85^{a}	6.8^{a}	7.6^{a}	7.9^{a}
875	7.5^{a}	7.6^{b}	6.5^{bc}	7.4^{a}	7.6^{a}

(Values are means of the 20 samples; means with the same letters in each columns are not significantly differed at 5%probability level)

Scale: 9 = Like Extremely 1 = Dislike Extremely

Table 4.6 Results of DMRT for the Organoleptic Evaluation of Biscuits Stored in Different Packaging Materials at 28th Day of Storage.

Biscuits	Colour	Crispyness	Flavour	Taste	Overall eating quality
727	5.85^{c}	5.5^{c}	5.25^{b}	5.05^{c}	5.25^{c}
427	6.3^{b}	6.2^{b}	5.35^{b}	5.35^{bc}	5.9^{b}
389	7.15^{a}	7.35^{a}	6.25^{a}	6.85^{a}	7.05^{a}
875	6.7^{b}	6.75^{b}	5.75^{ab}	6.0^{b}	6.35^{b}

(Values are means of the 20 samples; means with the same letters in each columns are not significantly differed at 5%probability level)

Scale: 9 = Like Extremely 1 = Dislike Extremely

Treatment code	Treatment
727	Plastic bottles
427	Polyethylene
389	Glass bottle
875	Aluminium foil

Table 4.7 Results of Microbial study

Treatment	Micro organism	Storage days			
		7	14	21	28
727	Fungus	–	–	–	+
	Bacteria	–	–	–	–
427	Fungus	–	–	–	+
	Bacteria	–	–	–	–
389	Fungus	–	–	–	–
	Bacteria	–	–	–	–
875	Fungus	–	–	–	–
	Bacteria	–	–	–	–

"+" denotes growth of micro organism and "–"denotes no growth of micro orga.

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