SHELF STABILITY OF MULTIGRAIN INCORPORATED BISCUITS ON STORAGE: ITS PHYSICO-CHEMICAL, TEXTURAL AND SENSORIAL STUDIES

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Abstract
The shelf stability of multigrain biscuits (MGB) containing 40% of multigrain premix (MGP) was carried out. The changes in physical, chemical, sensory characteristics when packed in polypropylene (PP) and metalized polyester (MP) during storage was monitored for the period of six months. The initial moisture content of biscuit was 3.38% and increased to 4.89 and 3.85% in biscuit packed in PP and stored room tempertature (RT) and 37°C respectively. The increase in moisture content of biscuits decreased the breaking strength of biscuits. The PV of fresh biscuit was 4.33 meqO_2/Kg fats and it increased to 15.74 and 19.37 meqO_2/Kg fat in biscuits packed in PP and stored at RT and 37°C. Thiobarbituric acid (TBA) of biscuits at an initial stage was 0.08 mgMA/kg and maximum increased to 1.18 mgMA/kg in PP packed biscuits and stored at 37° C. Free fatty acid (FFA) of biscuit increased from 0.53% oleic acid to 1.91, 2.16, 1.14 and 1.31% oleic acid in biscuits packed in PP, stored at RT, 37°C and packed in MP, stored at RT, 37°C respectively. Sensory studies indicated that MP packed biscuits was more stable than PP and can be stored up to 180 days at RT and 150 days at 37°C with acceptable sensory quality.

Key words: Multigrain biscuits, storage stability, packaging materials, peroxide value

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INTRODUCTION

The Indian market for ready to eat food products is witnessing exponential growth. Among ready to eat food products, the bakery segment is experiencing higher demand as evidenced by its two fold increase in production during the last five years (Kumar *et al*., 2015). Under the bakery segment, the biscuits, cookies are more popular among consumers due to the unique advantages like cheaper cost, easy to carry due to convenient pack size, longer shelf life, available in a variety of taste and texture etc. However, the biscuits available in the market are prepared from wheat flour, high sugar and fat are leading to a serious health concern. The refined wheat flour used in the preparation is known for inferior nutritional profile due to the removal of the bran, germ layers of wheat. Therefore, there is increased consumer demand for biscuits with high protein and dietary fibre along with other minor nutrients.

Storage stability of the product is defined as "the amountof time that a food product is considered acceptable for consumption when stored at the appropriate storage conditions" (Fisch, 2014). Biscuits are dry products and considered to have a relatively longer shelf life due to its low moisture content of 2-5 %. Due to its low moisture content, biscuits are stored at room temperature in flexible pouches with a high barrier to moisture and oxygen. The bakery products containing high fat are subject to a natural deterioration after a certain period of time from the date of production. The most common quality deterioration phenomena during biscuit storage are loss of crispiness and lipid oxidation due to moisture uptake and their high fat content. Lipid oxidation in biscuit is high due to the use of high fat in the preparation and is one of the major causes of chemical spoilage which results in rancidity, deterioration of nutritional quality, color, flavor and texture of biscuit (Antolovich *et al.*, 2002). The rancidity in biscuit is due to lipid oxidation during storage can result in an off-odor and off-flavor generation that renders products unpalatable and impact their shelf life (Smith *et al.*, 2004).

The use of alternate ingredients is gaining importance worldwide. However, incorporating any other nutrition rich ingredient to biscuit formulation leading to technological challenges concerning its texture, taste and shelf stability. Addition of protein or fiber rich ingredients will alter the water requirement of the flour during dough development and also hold more moisture in the finished goods. The increased moisture content in the finished products will affect its storage stability.

Multigrain premix is a combination of severeal nutritious grains mixed together to prepare homogenious mix which can be used to replace wheat flour. Therefore, the use of multigrain will give multiple health benefits along with improvement in functional and sensory properties of the end products. The different grains such as barley, sorghum, chickpea, dry pea and soya flour were mixed at 20% level and this premix was used to replace wheat flour at 40% level.

Therefore, the present study is planned to study the effect of incorporation of protein and dietary fiber rich multigrain premix to wheat flour at 40% level to make multigrain biscuits and its effect on storage stability. The use of different packaging material to pack the multigrain biscuits and different storage conditions were covered.

MATERIALS AND METHODS

2.1 Raw materials

The different flours used in MGP were barley (*Hordeum vulgare L.*), sorghum (*Sorghum vulgare*), chickpea (*Cicer arietinum*), whole dry pea (*Pisum sativum*), wheat flour and the ingredients for biscuit preparation like skimmed milk powder (Nandini brand, Karnataka Milk Federation, Mysore, India), sugar, Marvo brand bakery shortening (Bunge India Pvt. Ltd, Mumbai, India), vanilla essence (Bush Boake Allen Ltd, Chennai, India), sodium bicarbonate, ammonium bicarbonate (SD Fine Chemicals, India) and dextrose (Fisher Scientific, India) procured from local market in Mysore, Karnataka, India and defatted toasted soya flour (*Glycine max*) procured from the Sakthi Soya Company, Pollachi, Tamil Nadu, India was used for the study.

The packaging material for the study viz. polypropylene (PP) of 75 µ thickness and metalized polyester (MP) of 75 µ thickness packaging materials for packing of biscuits were procured from National Packaging Company, Bengaluru, India.

2.2 Preparation of MGP and MGB

The MGP is prepared by combining barley, sorghum, chickpea, whole dry pea and defatted soya flour each at 20% level. All the four grains are separately milled using ultra-centrifugal mill (Retsch ZM 200, Germany) using 200 µm sieve. The four milled grains and defatted toasted soya flour are combined in the ratio of 1:1 and were mixed in a Hobart mixer (Model N50, Hobart GmbH, Offenburg, Germany) for 10 min and passed through a sieve of 200 µm size to get a homogenous mixture. The MGPs were packed in an airtight container and stored at 4 °C for further use (Kumar *et al*., 2015).

Biscuits were prepared according to the AACC (2000) method 10-52. The ingredients used in biscuits preparation were wheat flour 60 g, MGP 40 g pulverised sugar 35 g; bakery fat 20 g; ammonium bicarbonate 1 g, salt 0.5 g; sodium bicarbonate 0.5 g; skimmed milk powder 2 g, dextrose 2 g and vanilla essence 1 ml.

The method of preparation was as follows: Mixed the wheat flour and MGP and sieved through 200 micron sieve for homogenous mixing. Pulverised sugar, bakery shortening, skimmed milk powder, dextrose and vanilla essence were creamed in Hobart mixer (Model N50, Hobart GmbH, Offenburg, Germany) with a flat blade, for 2 min at 61 rpm first, then 3 min at 125 rpm. The cream was mixed with water containing dissolved sodium chloride, sodium bicarbonate; ammonium bicarbonate was separately incorporated and mixed for 5 min at 125 rpm until homogenous cream obtained. Finally, sifted wheat flour was added and mixed at 61 rpm for 2 min. The dough was sheeted to 5 mm thickness using a metal frame and cut into a round shape of 55 mm diameter using a circular cutter. The baking was carried out at 200°C for 10 min. The biscuits were cooled and stored MP and PP pouches containing 12 biscuits each in the pack.

2.3 Evaluation of biscuit quality

2.3.1 Textural characteristics

The force required to break the biscuits were determined by texture analyser (TA- HD plus, Stable Micro Systems, Surrey, U.K) using triple beam snap (three-point break) techniques as per method defined by Gains (1991). The peak force from the resulting curve considered as the breaking strength of biscuit. The mean value of three independent determinations was reported.

2.3.2 Chemical characteristics:

The moisture, water activity of the cookies were measured according to AACC (2000) methods and Peroxide value (PV), free fatty acids were estimated by the standard method of AOCS (1990), whereas thiobarbituric acid value was determined using Tarledgis *et al*.(1960) method.

2.3.3 Sensory evaluation

Sensory quality of biscuit was evaluated by twenty panelists of age between 25 years to 50 years, including both male and female, who had earlier experience in sensory evaluation of bakery products. They were further trained in four sessions, including two hours of training in each session. Biscuits were evaluated in triplicates by each panelist for taste, texture, and overall acceptability on a 9-point hedonic scale (1 = dislike extremely, 5 = neither like nor dislike and 9 = like extremely) according to the method of Larmond (1997). The samples were identified by code number and presented in a random order to the panellist.

2.4 Statistical anlysis

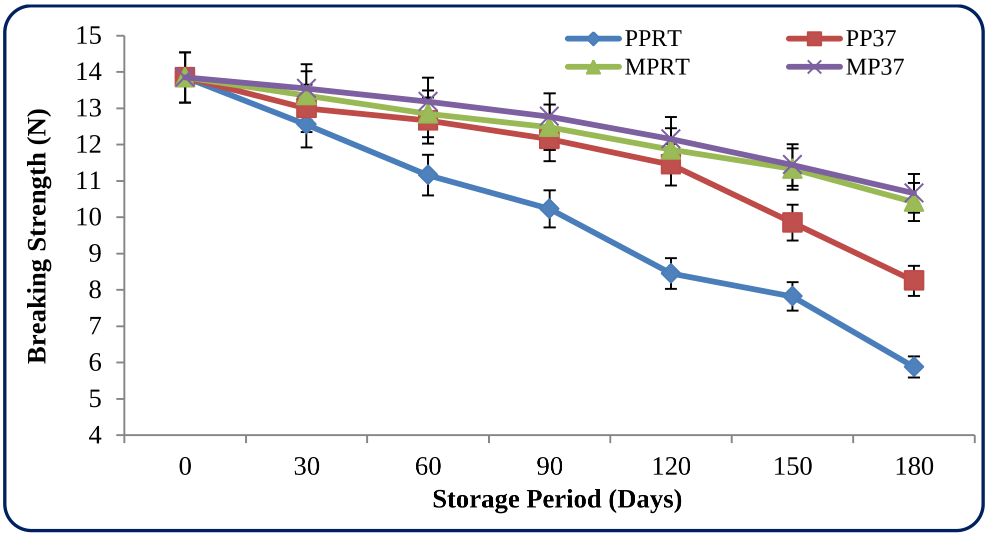
The experiements were carried out in triplicates, and the mean of three independent determination was presented. Regression analysis for the correlation was carried out, and the correlation coefficient was established using the software Curve Expert 1.3 (Hyams, 2003).

RESULTS AND DISCUSSION

The biscuits were stored at room tempertature (RT) as well as accelerated controlled temperature of 37°C. Usually biscuits are stored at RT, where temperature will vary depending on the environmental conditions. However. In accelerated conditions, temperature is controlled and will get accurate results in less time. Storage stability of multigrain biscuit were investigated on the basis of changes in its breaking strength, colour, chemical parameters like moisture, peroxide value (PV), free fatty acid (FFA), Thiobarbituric acid value (TBA), and sensory parameters like taste, texture and OAA. Increase in moisture level and rancidity of flavor are the important deteriorative factors for biscuits during storage.

3.1 Breaking strength of biscuits

Breaking strength of biscuits evaluate the texture properties of biscuits. Biscuit texture parameters are important quality indices in the quality evaluation of biscuits (Conforti *et al.*, 2012). The breaking strength of the biscuits decreased during storage due to absorption of moisture (Fig. 1). The maximum decrease was observed with biscuits packed in PP pouches and decreased from 13.85 N at initial to 5.88 N stored at RT and 8.25 N stored at 37° C respectively at the end of six months. After 180 days of storage, biscuits were very soggy and lost its crisp nature. The biscuit packed in MP pouches and stored at both RT and 37° C were crisper even at the end of six months. A similar report of a decrease in breaking strength of roasted ground flax seed flour added biscuits during the storage period of 90 days wasstated by Rajiv *et al.* (2012b) and observed the slight loss of crispiness.

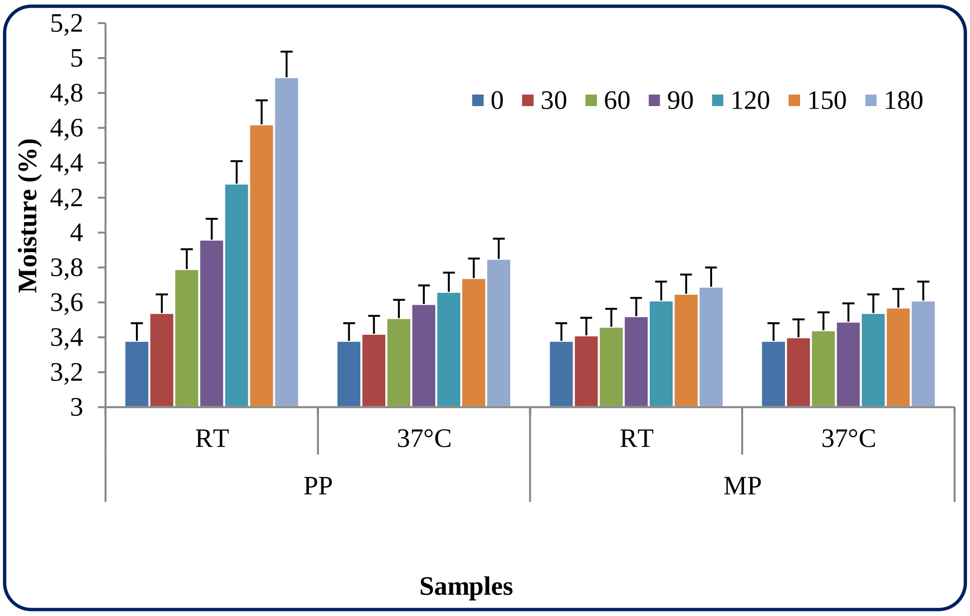


PPRT- Polypropylene room temperature; PP37- Polypropylene 37°C; MPRT- Metalized polyester room temperature; MP37-Metalized polyester 37°C

Figure 1 Changes in breaking strength of biscuits during storage

3.2 Moisture

Biscuits are very hygroscopic in nature, and these must be protected from the atmosphere to prevent or at least delay the moisture pick up during storage. Moisture is one of the important parameters to monitor during storage of biscuits as loss and gain of moisture is a serious problem in bakery products that can result in textural changes and may also help chemical and microbiological spoilage in low and intermediate moisture products. The changes in the moisture content of biscuits during the storage period have shown that the moisture content of biscuits increased in all samples irrespective of packaging and storage temperature (Fig. 2).



PP-Polypropylene; MP- Metalized polyester; RT-Room temperature

Figure 2 Changes in the moisture content of biscuit during storage

The initial moisture content of biscuit was 3.38 % and it significantly increased to 4.89 and 3.85 % in biscuit packed in PP pouch and stored RT and 37° C respectively. But the increase in the moisture content of biscuits was not significant in biscuits packed in MP pouches and stored at both temperatures mainly due to the moisture barrier properties of MP pouches. The increase in moisture content in biscuit in PP pouches may be due to the absorption of moisture from the surrounding environment as PP pouches have low barrier to moisture and water vapour transmission whereas, increase in moisture content of biscuits packed in MP pouches could be due to water diffusion and redistribution within the porous structure of food products as explained by Romani *et al.* (2015), who reported increased moisture content of biscuits packed in MP pouches. Kumar *et al.* (2014) also observed a similar increase in the moisture content of oryznol fortified biscuits during storage and Nadarajah and Mahendran (2015) in 20 % defatted coconut flour incorporated biscuits packed in MP pouches.

3.3 Rancidity of biscuit during storage

Biscuits are very moisture sensitive products and can develop oxidative rancidity and hydrolytic rancidity at very low moisture content due to its high fat content (Kumar *et al.*, 2014). Biscuits were monitored for both types of rancidity during storage. In oxidative rancidity, lipid oxidation of unsaturated fatty acids into hydro peroxides and later forms aldehydes and ketones whereas, in hydrolytic rancidity, triglycerides of fats are converted into free fatty acids and glycerol, resulting in a distinctive soapy flavor in biscuits.

The extent of auto oxidation of fat extracted from biscuits was determined by peroxide and melonaldehyde content. Peroxide value is one of the most extensively and conventionally used chemical tests to determine oxidative rancidity and is a measure of the concentration of primary oxidation products like peroxides and hydro peroxides formed in the initial stages of lipid oxidation (Bhanger *et al.*, 2008). The main ingredients used in biscuits like wheat flour, sugar, fat are predominantly sensitive to oxygen reactions. The larger surface area of biscuits increases its exposure to oxygen and light which may lead to increased oxidation (Kumar *et al.*, 2014). The PV of biscuits stored in PP pouches was higher than MP pouches whereas, the increase in PV was higher in 37° C than RT samples as a function of storage. The PV of fresh biscuit was 4.33 meqO_2/Kg fats, and it increased to 15.74, and 19.37 meqO_2/Kg fat in biscuits packed in PP pouches and stored at RT and 37° C which corresponding value for MP pouch packed biscuits were 10.22 and 11.89 meqO_2/Kg fat respectively (Fig. 3a). Samples in PP pouches exhibited the highest PV at all stages of storage showing its high intensity of oxidation while MP pouch packed biscuit showed a slow rise in PV. A similar trend of increasing PV value of oryzanol fortified biscuit during storage was reported by Kumar *et al.* (2014) and rice bran extract added cookies was reported by Bhanger *et al.* (2008).

Melonaldehyde is a secondary or final product of auto oxidation of poly unsaturated fatty acids and measured using TBA test, which is recognized as a reliable estimator of lipid peroxidation (Botsoglou *et al.*, 1994). TBA test measures formation of saturated aldehydes, 2-enals and 2-dienals in the final stage of lipid oxidation by reaction with 2-thiobarbituric acid. TBA of biscuits at an initial stage was 0.08 mg (melonaldehyde) MA/kg and maximum increased to 1.18 mg MA/kg in biscuits packed in PP pouch and stored at 37° C (Fig. 3b). The increase in the rate of auto oxidation as measured by PV and TBA can be attributed to the breakage of long fatty acid chain into individual fatty acid moieties, decomposition of hydro peroxides and also increased lipid hydrolysis at elevated temperature (Romani *et al.*, 2015).

Free fatty acid content is also an indicator of the rancidity of foods, which shows the hydrolytic rancidity and measures formation of free fatty acids cleaved from triglycerides or phospholipids due to hydrolysis of triglycerides and may get promoted by its reaction with moisture (Frega *et al.*, 1999). Storage period and packaging material tangibly increased the

FFA of biscuit, and it increased from 0.53 % oleic acid to 1.91, 2.16, 1.14 and 1.31 % oleic acid in biscuits packed in PP pouch, stored at RT, 37° C and packed in MP pouch, stored at RT, 37° C respectively (Fig. 3c). Earlier Kumar *et al.* (2014) reported that the BIS specification for the maximum FFA limit as oleic acid in biscuits by mass is 1.2 %.

Fig. 3a: Changes in peroxide value

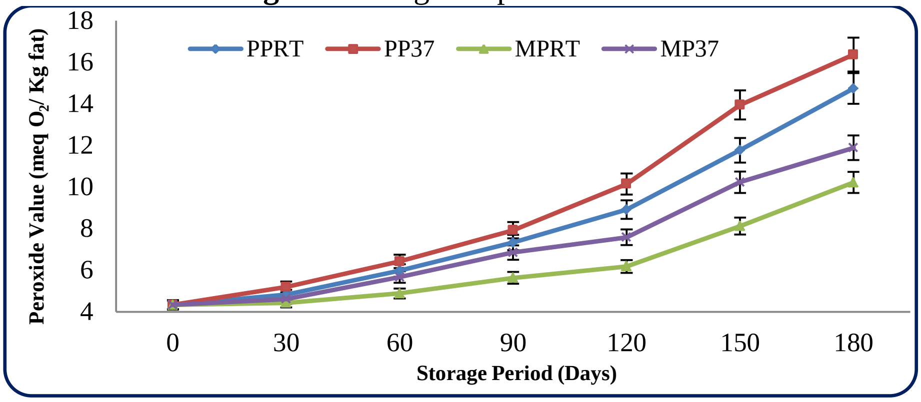


Fig. 3b: Changes in Thiobarbeturic acid

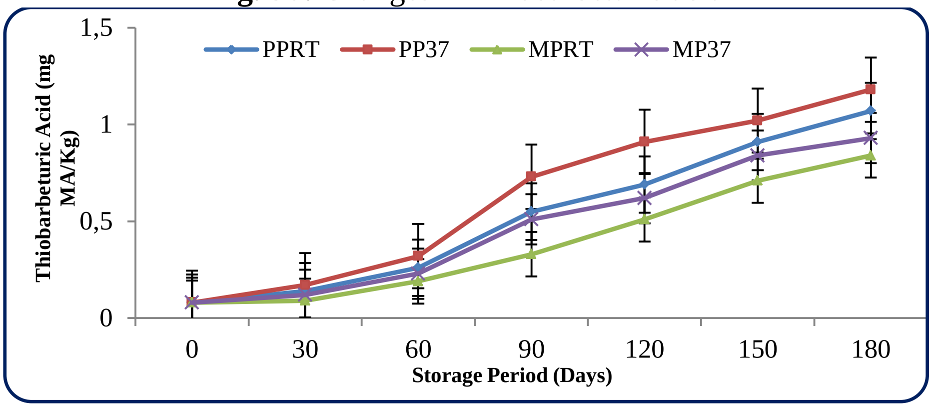
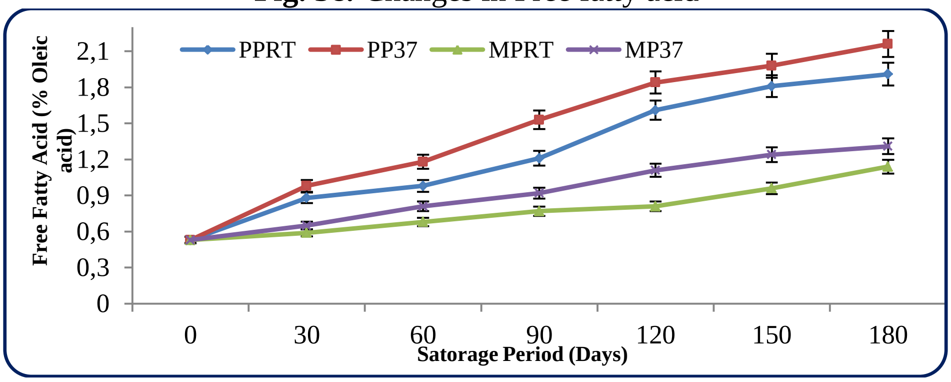


Fig. 3c: Changes in Free fatty acid



PPRT- Polypropylene room temperature; PP37- Polypropylene 37°C; MPRT- Metalized polyester room temperature; MP37-Metalized polyester 37°C

Figure 3 Changes in chemical characteristics of MGB during storage

The biscuit packed in MP pouch and stored at RT only had less than this limitation. Lipid hydrolysis of food products during storage is brought about by naturally occurring enzyme lipase (Clayton and Morrison, 1972), whereas in biscuits lipases must have destroyed by thermal treatment employed during baking, therefore formation of free fatty acids in biscuits during storage resulted from the decomposition of hydro peroxides (Thakur and Arya, 1990). The biscuits with high PV and TBA content showed to have higher FFA content suggesting thereby the involvement of lipid auto oxidation for an increase in FFA content (Khan *et al.*, 2014).

3.4 Correlation analysis

The storage studies of biscuits for lipid oxidation were subjected to correlation analysis to establish the best fit equations. Since the trend obtained for PV, FFA and TBA parameters were of the same pattern as per earlier discussion, to establish the correlation parameters amongst different parameters like PV, FFA and TBA values of biscuits during storage, regression analysis was carried out and the fit of equations were determined for the data with maximum correlation coefficient and minimum standard error and the best fit equations for the correlation studies have been represented in Fig. 4 (a,b,c). Correlation regression analysis was performed using the software curve expert 1.3 and linear fit equation established between PV vs. FFA, and polynomial fit equation established between PV vs. TBA and FFA vs. TBA and the same is represented in Table 1.

The linear fit model was found to be the best fit model to establish a correlation between PV vs FFA with a correlation coefficient of 0.9856, whereas polynomial fit was found to be the best fit to establish a correlation between PV vs. TBA and FFA vs. TBA with a correlation coefficient of 0.9945 and 0.9891 respectively. The above regression analysis reveals that there is a positive correlation exhibited between rancidity parameters of biscuit during storage.

3.5 Sensory acceptability of biscuit during storage

The sensory analysis of biscuits like taste, texture and OAA were evaluated for biscuits during storage and presented in Fig. 5 (a,bc). The sensory analysis of biscuits showed that there is a significant decrease in sensory parameters like taste, texture and overall acceptability during storage. The biscuits packed in PP pouch and stored at RT were soggy by the end of 120 days, and the taste was found to be rancid. However, the rancid flavor in biscuits with PP pouch and stored at 37° C were observed by the end of 90 days. The sensory score for biscuits taste at initial was 8.53, and it decreased to 6.1 on a 9-point hedonic scale (Fig. 5a). The biscuits packed in MP pouches and stored at 37° C had an equally acceptable taste to biscuit with MP pouch and stored at RT up to 90 days, but after it varied differently. The taste of biscuit stored at 37° C was acceptable up to 150 days and at 180 days showed maximum rancidity flavor in a biscuit as indicated by its decreased score for taste whereas RT stored biscuits had acceptable taste even at 180 days of storage. The texture of biscuit can be correlated to the breaking strength of biscuits. The sensory scores for texture parameters followed a similar trend like that of breaking strength of biscuits. The biscuits packed in PP pouches lost crispiness earlier than MP pouch packed biscuits. There was not much difference between MP pouch packed biscuit between RT and 37° C stored biscuits even at the end of six months. Initially, biscuits had an OAA of 8.41 and during storage biscuits with PP pouch stored at RT were acceptable up to 90 days, whereas at 37° C stored biscuits were acceptable only up to 60 days as indicated by its decreased OAA scores of 6.01 and 5.51 respectively. The OAA of 7.0 was taken as a cut off for acceptation of shelf-life of the biscuits. MP packed biscuits was more stable than PP pouch and can be stored up to 180 days at RT and 150 days at 37° C with acceptable sensory quality (Fig. 5c). Nadarajah and Mahendran (2015) reported that defatted coconut flour incorporated biscuits packed in MP pouches could be stored for a longer period due to its high barrier to moisture and water vapour.

Figure 4a: PV vs. FFA

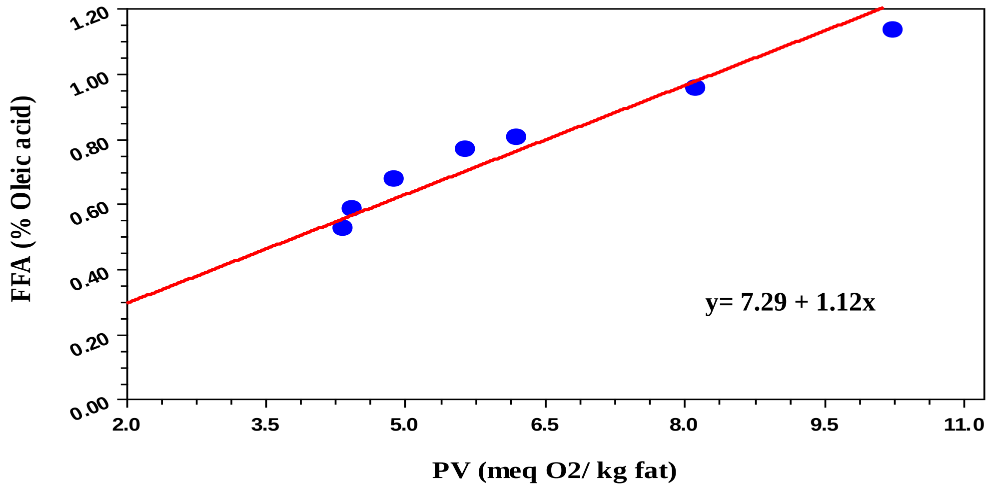


Figure 4b: PV vs. TBA

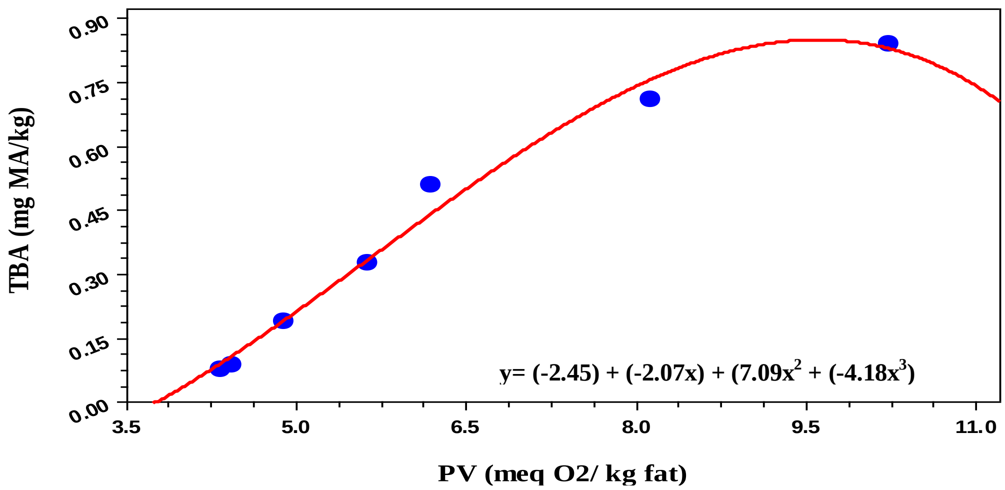


Figure 4c: FFA vs. TBA

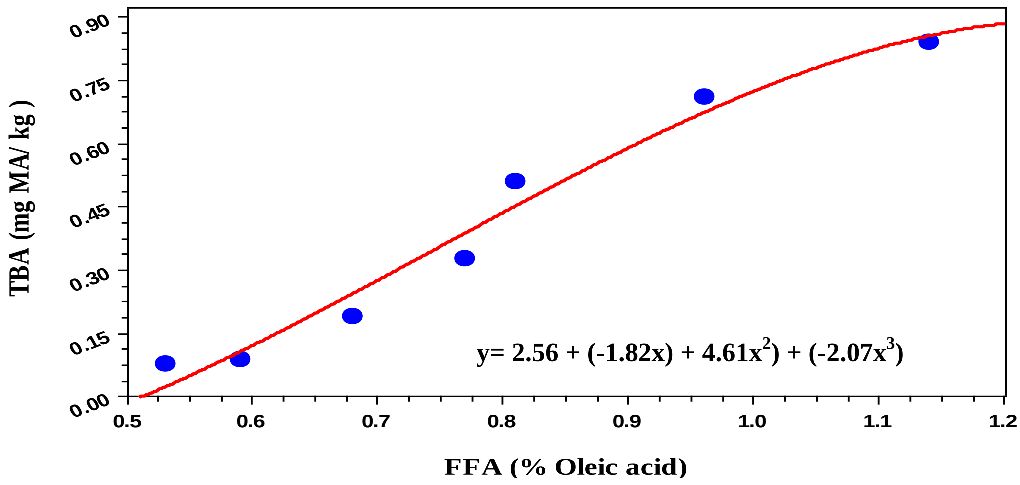


Fig. 4 Correlation regression analysis of PV, FFA and TBA

Figure 5a: Changes in taste scores during storage

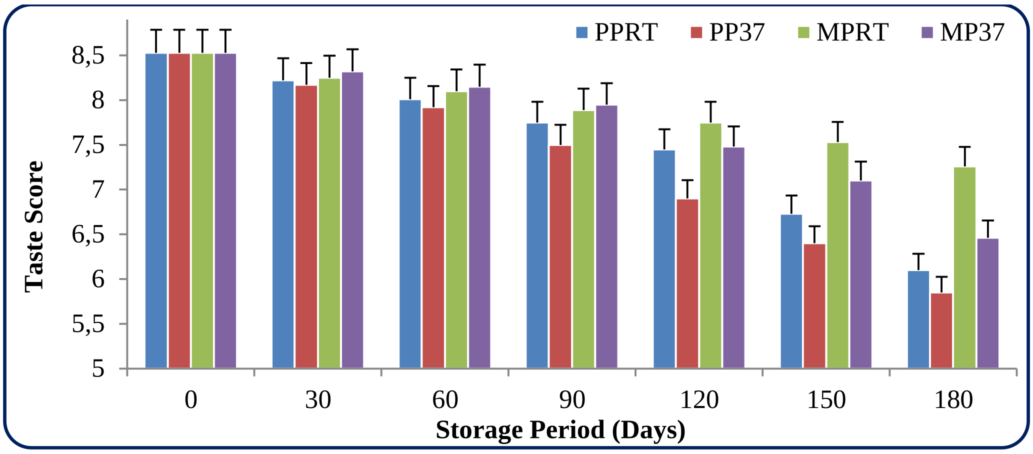


Figure 5b: Changes in Texture score during storage

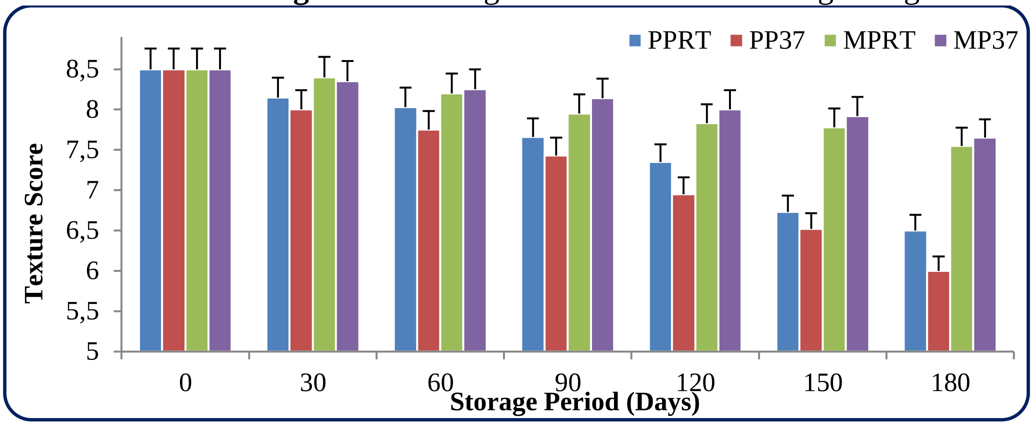
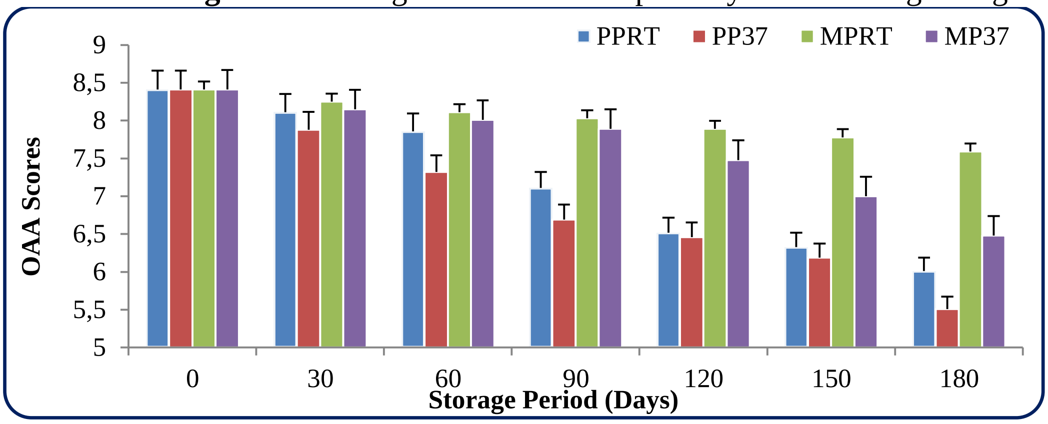


Figure 5c: Changes in Overall acceptability scores during storage



PPRT- Polypropylene room temperature; PP37- Polypropylene 37°C; MPRT- Metalized polyester room temperature; MP37-Metalized polyester 37°C

Fig. 5 Sensory evaluation of biscuit during storage

Table 1: Correlation regression analysis of PV, FFA and TBA

Parameters	Best established correlation fit	Correlation equation	Standard Error	Correlation coefficient (R)
PV vs. FFA	Linear fit	y = 7.29 + 1.12x	0.0621	0.9856
Pv vs. TBA	Polynomial fit	$y = (-2.45) + (-2.07x) + 7.09x^2 + (-4.18x^3)$	0.0434	0.9945
TBA vs.FFA	Polynomial fit	$y = 2.56 + (-1.82x) + 4.61x^2) + (-2.07x^3)$	0.0608	0.9891

PV: Peroxide value; FFA: Free fatty acid; TBA: Thiobarbituric acid

CONCLUSION

Use of suitable packaging material and storage condition will affect the shelf stability of products. The ideal packing material and storage condition can be determined by products chemical and physical alteration during storage. The significant increase in the moisture content of biscuits packed in PP pouches was observed. But the increase in the moisture content of biscuit was not significant in biscuit packed in MP pouches and stored at both temperatures. The breaking strength of the biscuits decreased during storage due to absorption of moisture, and the maximum decrease was observed with biscuits packed in PP pouches.

The PV, TBA and FFA contents of biscuits were found to increase during storage, and the extent of increase depended on packaging material and storage temperature used for the study. The highest increase in PV, TBA and FFA content of biscuits was observed in biscuits packed with PP and stored at 37°C followed by biscuits packed in PP pouch stored at room temperature, MP pouch stored at 37°C and MP pouch, stored at room temperature. Sensory studies of biscuits concluded that biscuits packed in MP pouches were more stable than PP pouches packed biscuits and can be stored up to 180 days at RT and 150 days at 37° C with acceptable sensory quality. Based on the physical, chemical characteristics of MGB, it can be concluded that biscuits can be stored up to 180 days at room temperature using metalized PP pouches without affecting any sensory attributes.

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