

## **Consequence of nitrogen gas flushing and laminate packaging on physico-chemical characteristics of microwave ready-to-eat mutton snacks**

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### **Abstract**

Emerging trends in ready-to-eat food market industry has given wide scope for development of a variety of innovative packaging materials and methods to extend shelf life and maintain quality. Laminates and nitrogen gas flushing are the most popular choice as a packing material and method respectively. Hence this study was conducted to evaluate the effect of nitrogen gas and laminate on shelf life of ready-to-eat mutton based snack. During storage, moisture and water activity increased and pH decreased irrespective of method of packaging. Statistically, there was significant ( $P<0.05$ ) difference in moisture content and water activity between treatments (Aerobically packaged snacks-APS and Modified atmosphere packaged snacks-MAPS), days of storage and their interaction. There was no significant ( $P>0.05$ ) difference between treatments i.e., modified atmosphere packaging and aerobic packaging. Initially TBA value decreased and thereafter increased throughout storage. In APS, redness decreases during the storage, however, in MAPS it decreases up to 15<sup>th</sup> day and thereafter increased. Yellowness also increased significantly irrespective of the packaging. Results of the present study indicated that physico-chemical characteristics for MAPS were superior to APS.

**Keywords:** Mutton based snacks; storage; nitrogen gas flushing; laminate packaging; physico-chemical characteristics

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### **Introduction**

It is of the essence to develop good quality, healthful and shelf stable meat products containing high protein and low profile of calorie, sodium and cholesterol. There is vital call for extend the shelf life of product so that it can be utilized for long time. Pszczola (2002) identified innovation in 'meat snacks' as an area with high potential. Due to the fast changing pace of life, urbanization and changing socio-economic conditions, snack foods are gaining popularity now-a-day. The reported growth of the market of meat snacks is due to their low carbohydrate, fat content and diverse

flavour selection (Bosse and Boland, 2008). It has become an integral part of the eating habits of the majority of the world's population.

The Indian snacks market worth around US\$ 3.5 billion is one of the largest snack markets in the Asia-Pacific region, with the organised segment taking half the market share and growing at around 20% per annum. The organized sector of the snack food market is growing at 15-20 percent a year while the growth rate of the unorganized sector of US\$ 1.56 billion is 7-8 per cent (SSTI, 2011). So there is imperative need to develop technology for manufacture, package snacks on a larger scale and to give good distribution solutions.

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Modified atmosphere packaging (MAP) is the removal and/or replacement of the atmosphere surrounding the product before sealing in vapor-barrier materials (McMillin et al., 1999). Nitrogen is colourless, odourless, tasteless, non-toxic and does not react with any other compound. By excluding oxygen and moisture, gas flushing with nitrogen prevents oxidation and rancidity within the food product and, as a result, helps improve shelf life and product quality. Nitrogen flushing treatment constitutes a promising option to extend the shelf life (Lloyd et al., 2009).

Moisture, light and oxygen are the enemies of many high-quality snack food products. In order to protect against moisture, light and oxygen in gas-flushed packs, a metalized high-barrier packaging film should be used. Plastic laminates and films provide properties such as high barrier to oxygen, moisture aroma/flavour or sealing property to a package even when its surface is contaminated with product. Moisture proof, durable and appealing confectionery packaging is the key to enhance the shelf life, brand image and marketability of confectionery products.

Snack food are extremely moisture sensitive, and can easily absorb moisture even at low relative humidity conditions, and at critical moisture content (CMC) levels, loss of crispness occurs, rendering the product unacceptable to the consumers. Previously reported quality changes in meat and dried meat products including changes in pH, colour, moisture content, water activity and lipids (Rhee and Pradahn, 1999; Rao et al., 1996; Lin & Lin, 2002; Modi et al., 2007; Gok et al., 2008).

In present study, efforts were focussed to reduce changes in physico-chemical properties of snack foods. Nitrogen gas replacing the air of packaged snacks which in turn prevents oxidation, bacterial growth and detrimental effect on physico-chemical properties while laminates resist the change in physico-chemical properties by providing high barrier to oxygen, moisture, aroma/flavour or sealing capacity to a package.

## Materials and Methods

### Source of raw materials

#### Spices, condiments and other ingredient

Spices, condiments, flours, salt, vegetable oil, sugar and monosodium glutamate were procured from the local market of Bareilly, India. Starch used in the experiment was obtained from SRL Pvt. Ltd., Mumbai, India.

### Chemicals

Chemicals were of analytical grade and obtained from standard firms (Qualigens, Hi Media, Polypharm, SRL etc.).

### Packaging material

Low density polyethylene (LDPE) films (250 gauge) in natural colour were procured from the manufacturer (M/S Hitkari Industries Ltd., New Delhi) for packing of the materials for storage studies. Aluminium foil-LDPE laminate packaging covers were purchased from local market of Bareilly, Uttar Pradesh, India.

### Mutton based snacks

Meat from the leg portion of sheep was obtained from experimental abattoir of Livestock Products Technology Division, IVRI, Bareilly. All separable fat, fascia and connective tissue were trimmed off and meat was packed in low density polyethylene (150 gauge) bags, and frozen at -18 to -20°C till further use. The meat was thawed before it was processed for preparing meat based snacks. Mutton based snacks were developed by heating the dry batter through microwave.

### Analytical procedures

#### Moisture

Moisture content was determined according to the method of Association of Official Analytical Chemists (AOAC, 1995). The ground sample (approx. 2 gm) was transferred to pre-weighed flat bottom stainless steel dish, which was again weighed. This was transferred to a hot air oven at 101±1°C and kept for 16-18 h. Dried sample was then placed in desiccator having fused CaCl<sub>2</sub> as desiccant. After 1 h, the dish was weighed. Moisture content was calculated by applying the following formula:

$$\text{Moisture (\%)} = \frac{W_2 - W_3}{W_2 - W_1} \times 100$$

W<sub>1</sub> = weight of empty dish

W<sub>2</sub> = weight of dish + sample

W<sub>3</sub> = weight of dish + dried sample

### Water activity

Water activity was measured with the help of a water activity meter (Hygrolab 3, Rotronics, Switzerland). Ground sample was taken in the sample container of the water activity meter and placed inside the meter, closed the upper lid and pressed the button. Reading was recorded in 'quick mode' and noted after the beep sound.

### pH

The pH of the meat based snacks was determined by using the method described by Strange et al. (1977). 10 g of sample (after grinding in the home mixer) was blended with 50 ml of distilled water for 1 min using an Ultra Turrax tissue homogenizer (Model T25, Janke and Kenkel, IKA Labor Technik, Germany). The pH of the homogenate was recorded by immersing a

combined glass electrode of a digital pH meter (Eutech Instruments, pH 510, Merck).

#### **Thiobarbituric acid reacting substances (TBARS) number**

The TBARS number of samples was determined by using the distillation method described by Tarladgis et al. (1960). To 10 g of ground sample, 50 ml of distilled water was added and homogenized for 2 min in an Ultra Turrax tissue homogenizer (Model T25, Janke and Kenkel, IKA Labor Technik, Germany). The slurry was quantitatively transferred to a 500 ml Kjeldahl flask. It was rinsed with 45 ml of distilled water and washing was transferred to the flask to which 5 ml of 6N HCl was added. Few drops of liquid paraffin were added to prevent frothing and few glass beads were dropped into the flask to prevent bumping during heating. Distillation was on high heat and 50 ml distillate was collected in a graduated stoppered glass cylinder.

The distillate was thoroughly mixed and 5 ml of distillate was pipetted (duplicate) into 20 ml glass stoppered test tubes. Five ml of TBA (0.02M 2-thiobarbituric acid in 90% glacial acetic acid) was added to each test tube. The contents were mixed well and immersed in boiling water bath for 30 min. The blank consisting of 5 ml of distilled water and 5 ml of TBA reagent was similarly prepared. The tubes were cooled for 10 min under tap water and optical density (O.D) was recorded using spectrophotometer at 538 nm. The O.D. was multiplied by the factor 7.8 and TBARS value was expressed as mg malonaldehyde/ kg of sample as suggested by Koniecko (1979).

#### **Colour**

The colour of the meat based snacks was measured using a Lovibond Tintometer (Model F, Greenwich, UK). Samples were ground for 1 min in the home mixer, taken in the sample holder and secured against the viewing aperture. The sample colour was matched by adjusting the red (a) and yellow (b) units, while keeping the blue unit fixed at 0.1. The corresponding colour units were recorded. The hue and chroma values were determined by using the formulae,  $(\tan^{-1}) b/a$  (Little, 1975) and  $(a^2+b^2)^{1/2}$  (Froehlich et al., 1983), where a = red unit, b = yellow unit.

#### **Statistical analysis**

The data generated from various trials under experiment were analyzed by statistical method of one way-ANOVA using SPSS software package as per the procedure of Snedecor and Cochran (1995) and the significant differences ( $P<0.05$ ) in the means were compared by using Duncan's Multiple Comparison Test (Duncan, 1955).

#### **Experimental design**

The experiment was designed to evaluate the change in physico-chemical properties of shelf stable microwave ready-to-eat mutton based snacks during 60 d of storage and compared the physico-chemical properties of snacks packaged in aluminium foil-LDPE laminate with nitrogen gas flushing (MAPS-modified atmosphere packaged snacks) and in low density polyethylene (LDPE) films (250 gauge) without nitrogen gas flushing (APS - aerobically packaged snacks).

#### **Results and Discussion**

The changes in physico-chemical qualities and colour of snacks exposed to aerobically modified atmosphere during storage are given in Table 1 & 2 respectively.

#### **Moisture content**

Moisture content plays a significant effect on the texture of ready-to-eat snacks as it directly affects the crispiness that is a key factor for their acceptance. Statistically, there was significant ( $P<0.05$ ) difference in moisture content between treatments (APS and MAPS), days of storage and their interaction. Thus, method of packaging and time of storage significantly affected the moisture content of ready-to-eat snacks.

Moisture percentage of MAPS was significantly lower than APS at particular days of storage, however, in APS moisture content increases significantly but in MAPS it increases insignificantly. This might be due to difference in packaging material used in APS and MAPS. The gradual increase of moisture and water activity of all samples could be due to moisture absorption by the food from the environment that gradually permeated through packaging materials and also from the respiration of the growing microorganisms (Sawaya et al., 1995). Paine (1969) had mentioned that aluminium foil had less water vapour transmission rate as compared to polyethylene.

Modi et al. (2007) also observed a significant increase in water activity and moisture content of dehydrated chicken *kebab* stored in metallic polyester pouches at ambient temperature.

#### **Water activity**

Water activity is an important factor affecting the stability of dry and dehydrated products during storage. The crispness intensity and overall hedonic texture of dry snack food products are function of water activity (Katz and Labuza, 1981). There was a significant ( $P<0.05$ ) difference in water activity between treatments (APS and MAPS) and also between days of storage, which might probably be due to the higher water vapour permeability of LDPE films and consequent absorption of moisture from atmosphere. In

**Table 1: Changes in the physico-chemical qualities of aerobically packaged and modified atmosphere packaged snacks during storage at ambient temperature (Mean± S.E)**

Particulars	Days of storage				
	0	15	30	45	60
			Moisture %		
APS	4.63±0.04 <sup>b</sup>	5.09±0.14 <sup>bA</sup>	5.73±0.12 <sup>aA</sup>	5.71±0.13 <sup>aA</sup>	5.63±0.27 <sup>aA</sup>
MAPS	4.63±0.04	4.70±0.16 <sup>B</sup>	4.70±0.12 <sup>B</sup>	4.89±0.08 <sup>B</sup>	4.80±0.07 <sup>B</sup>
			Water activity		
APS	0.27±0.006 <sup>c</sup>	0.36±0.010 <sup>bA</sup>	0.39±0.004 <sup>aA</sup>	0.40±0.005 <sup>aA</sup>	0.41±0.007 <sup>aA</sup>
MAPS	0.27±0.006 <sup>c</sup>	0.29±0.005 <sup>cB</sup>	0.31±0.010 <sup>bB</sup>	0.34±0.008 <sup>aB</sup>	0.35±0.009 <sup>aB</sup>
			pH		
APS	6.42±0.01 <sup>a</sup>	5.10±0.06 <sup>c</sup>	5.01±0.02 <sup>dB</sup>	5.26±0.01 <sup>b</sup>	5.28±0.00 <sup>b</sup>
MAPS	6.42±0.01 <sup>a</sup>	5.14±0.01 <sup>d</sup>	5.14±0.01 <sup>dA</sup>	5.21±0.01 <sup>c</sup>	5.27±0.01 <sup>b</sup>
			TBA (mg malonaldehyde/kg )		
APS	1.06±0.08 <sup>a</sup>	0.60±0.03 <sup>bA</sup>	1.03±0.06 <sup>aA</sup>	1.01±0.03 <sup>a</sup>	1.02±0.01 <sup>a</sup>
MAPS	1.06±0.08 <sup>a</sup>	0.47±0.01 <sup>cB</sup>	0.85±0.02 <sup>bB</sup>	0.99±0.03 <sup>a</sup>	0.97±0.01 <sup>a</sup>

\*Mean±S.E. with different superscripts in a row (small letter) and column (capital letter) differ significantly (P<0.05); APS: aerobically packaged snacks, MAPS: modified atmosphere packaged snacks

**Table 2: Changes in the colour of aerobically packaged and modified atmosphere packaged snacks during storage at ambient temperature (Mean± S.E)**

Particulars	Days of storage				
	0	15	30	45	60
			Redness		
APS	2.43±0.07 <sup>a</sup>	2.30±0.07 <sup>abA</sup>	2.10±0.04 <sup>cA</sup>	2.12±0.03 <sup>cB</sup>	2.20±0.04 <sup>bcB</sup>
MAPS	2.43±0.07 <sup>a</sup>	2.22±0.02 <sup>cB</sup>	2.27±0.02 <sup>bcB</sup>	2.37±0.03 <sup>abA</sup>	2.37±0.04 <sup>abA</sup>
			Yellowness		
APS	4.93±0.20 <sup>c</sup>	4.87±0.17 <sup>c</sup>	5.83±0.06 <sup>ab</sup>	5.98±0.19 <sup>a</sup>	5.48±0.08 <sup>b</sup>
MAPS	4.93±0.20 <sup>b</sup>	4.77±0.21 <sup>b</sup>	5.97±0.11 <sup>a</sup>	5.82±0.21 <sup>a</sup>	5.50±0.08 <sup>a</sup>
			Hue		
APS	63.57±1.35 <sup>b</sup>	64.65±0.70 <sup>b</sup>	70.19±0.41 <sup>a</sup>	70.44±0.64 <sup>aA</sup>	68.70±0.34 <sup>a</sup>
MAPS	63.57±1.35 <sup>c</sup>	64.89±0.91 <sup>bc</sup>	69.17±0.40 <sup>a</sup>	67.77±0.64 <sup>aB</sup>	66.71±0.29 <sup>ab</sup>
			Chroma		
APS	5.51±0.17 <sup>cd</sup>	5.38±0.17 <sup>d</sup>	6.20±0.05 <sup>abB</sup>	6.35±0.18 <sup>a</sup>	5.91±0.08 <sup>bc</sup>
MAPS	5.51±0.17 <sup>b</sup>	5.26±0.20 <sup>b</sup>	6.38±0.10 <sup>aA</sup>	6.28±0.20 <sup>a</sup>	5.99±0.08 <sup>a</sup>

\*Mean±S.E. with different superscripts row wise (small letter) and column wise (capital letter) differ significantly (P<0.05); APS – aerobically packaged snacks, MAPS – modified atmosphere packaged snacks.

both groups there was significant increase in water activity up to 60th day of storage. Similar finding in water activity was recorded by Presswood (2012) in vacuum fried beef strips. An increase in the moisture content was also observed with an increase in the water activity for dried food products (Ozilgen, 2011). Most products follow a sigmoidal curve for loss of crispness as a function of water activity or water content (Roudaut et al., 1998). If the moisture content of the products increases, due to water absorption from the atmosphere or by mass transport from neighbouring components or phases, a loss of crispness is observed (Nicholls et al., 1995).

### pH

There was a sharp decrease in pH up to 30<sup>th</sup> day and after that gradual increase occurred up to 60<sup>th</sup> day in APS. However, for MAPS there was a sharp decrease in pH up to 30<sup>th</sup> day and then gradual increase was observed up to 60<sup>th</sup> day. ANOVA showed a significant (P<0.05) difference in pH between storage

days. Interaction between treatments and storage days was also significant. There was no significant (P>0.05) difference between treatments i.e., APS and MAPS had similar effect on pH. There was a sharp decrease in pH up to 30<sup>th</sup> day in both types of packaged snacks. The lowering of pH might be due to chemical activity as suggested by Modi et al. (2007). A decrease in pH followed by an increase had also been reported by Singh (2001) in intermediate moisture chicken meat in LDPE pouches and by Rubio et al. (2006) in vacuum packed dry cured beef.

### TBARS value

Statistically there was significant (P<0.05) difference in TBARS between both treatments (APS and MAPS) and days of storage. TBARS value on 15<sup>th</sup> and 30<sup>th</sup> day was significantly lower for MAPS as compare to APS. This could be due to presence of nitrogen in MAPS which reduces the oxygen content and prevented oxidation of lipids. Comparatively high initial TBARS value observed might be due to the

mincing, mixing, cooking and drying steps involved in the preparation process, which resulted in extensive destruction of cellular structure, allowing mixing of various meat constituents and pro-oxidants (Rhee and Myers, 2003). Higher dry matter content in ready to eat meat based snacks could have also contributed to the higher initial TBARS values. Similar higher initial values for TBARS had been reported by Modi et al. (2007) in dehydrated chicken *kebab* mix and Mgbemere et al. (2011) in *kilishi*.

Singh et al. (2011) also reported a reduction in TBARS values in chicken snacks up to 18 days of storage in laminate pouches at ambient temperature. The reason for decrease of TBA value might be due the nature of the TBA analysis, because short-chain carbon products of lipid oxidation are not stable (Fernandez et al., 1997). The oxidation of these products results in organic alcohols and acids, which are not determined by the TBA test (Almandos et al., 1986). Additionally, malonaldehyde can react with amino acids thus decreasing TBA values (Gardner, 1979). However, the result was contrary to the findings of Sharma and Nanda (2002), Vasavada and Cornforth (2005) and Kharb and Ahlawat (2010) who observed an increase in TBARS values during storage of dehydrated products.

### Redness

Statistically there was a significant ( $P<0.05$ ) difference in redness values of treatments (APS and MAPS) and between days of storage. Their interaction was also significant. Decrease of redness in APS during storage might be due to oxidation reaction occurred in presence of oxygen. However, in MAPS initial decrease of redness might be due to oxidation reaction caused by residual oxygen. This indicated that concept and composition of packaging are the main factors for maintaining an oxygen-free atmosphere that will decrease possible oxidation during storage. The application of packages with low oxygen permeability enables minimizing changes in the colour of meat products (Ahvenainen, 1989; Arihara et al., 1993; Brewer & Novakofski, 1999; Kłossowska & Olkiewicz, 2000; Pikul, 2002; Jankiewicz, 2004). As the water content and humidity increase, the browning rate also starts to decrease. The dual role of water as solvent and reactant in the kinetics of the Maillard reaction was also pointed out by Van Boekel (2001). The rate of browning is high at relatively low water content, and this might explain the ease of browning in dried and concentrated foods.

### Yellowness

ANOVA shows that there was a significant ( $P<0.05$ ) difference in yellowness between days of storage. Yellowness increased significantly irrespective of the packaging. Yellowness increased significantly

irrespective of the packaging might be due to reduction of red colour during storage.

### Hue

Statistically there was a highly significant ( $P<0.01$ ) difference in hue value between days of storage. Hue value of both the packaged snacks increased significantly and on 60<sup>th</sup> day it decreased slightly. Hue is the colour as perceived by the eye (Potter and Hotchkiss, 1998) and increased hue could be due to the increase in yellowness.

### Chroma

Statistically there was a highly significant ( $P<0.01$ ) difference in chroma value between days of storage, however there was no significant difference between treatments (APS and MAPS). This indicated that there was no or very less effect of packaging on chroma value of developed snacks. Chroma is the purity, intensity, saturation or richness of colour (Potter and Hotchkiss, 1998) and an increase in the present study might be due to increased yellowness.

### Conclusion

Modified atmosphere packaging and aerobic packaging were compared and analysed by package the shelf stable microwave ready-to-eat mutton based snack. Modified atmosphere packaging lowered the moisture absorption, water activity and TBARS number. Aluminium foil-LDPE laminate shows more resistant to moisture absorption as compare to LDPE. Flushing of nitrogen in modified atmosphere packaging lowers the oxygen content and reduces the oxidation of lipids. Nitrogen flushing resists change in redness of mutton based snack by preventing oxidation. Modified atmosphere packaging is better than aerobic packaging of microwave ready-to-eat mutton based snack.

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