Original Research Article

Shelf Life of Spent Hen Meat Papad Stored at Ambient Temperature

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Abstract

India is the most popular country for the production of poultry eggs which lead to the problem with the disposal off spent hens. Utilization of such spent hen meat is a big challenge to modify into potential palatable value-added products. The present study was carried out to study the shelf life of spent hen meat papad stored at ambient temperature. The spent hen meat papad were prepared and packed by aerobic packaging and modified atmosphere (80 per cent nitrogen and 20 per cent carbon dioxide) packaging (MAP) methods in sterile Pet/Poly pouches and packed samples were stored at ambient temperature. The samples were analysed on 0 day, 30th day and 60th day. Significant (P < 0.05) difference in pH, Thiobarbituric acid, water activity and FFA was observed during storage period. During the storage period microbial count gradually increased but within acceptable level. In sensory scores the colour, flavour, texture, crispiness and over all acceptability were significantly higher in MAP sample. Sensory scores gradually decreased during storage periods. Based on the findings, it is concluded that the spent hen meat papad could be stored up to 60 days at ambient temperature by aerobic and modified atmosphere packaging without any noticeable deterioration in the quality and acceptability of the product.

Keywords

Shelf life, Spent hen, Meat papad, Storage

Introduction

India occupies the number one position in livestock population especially the poultry production around 851.81 million. Worldwide, the egg producer’s dispose of 2.6 billion spent hens after their laying cycle is finished. The availability of the culled and spent hens is also increased manifold parallel with the rapid development of poultry layer industry. Spent hens are those birds whose egg laying capacity is over. The spent hen meat is usually tough, less tender and has poor functional properties due to increased collagen content and linkage (Sarvadnya et al., 2018). The emerging livestock product technology provide a path to produce variety of comminuted products without adhering any hindrance associated with toughness of spent hen meat (Kondaiah, 2004). The utilization and consumption of spent hen meat by the consumers of India is much limited and
makes the poultry farmers difficult in disposal of spent hens. On the other hand, the imbalance between protein supply and exploding population necessitates the search for new protein sources. In such instances, the underutilized sources of animal origin like spent hen meat found to be the one of the potential sources in view of dual benefits of economy as well as nutritive value (Sreenivasa Rao et al., 2011). The annual poultry meat consumption has increased around 1.8 kg with the increasing number of non-vegetarians. Simultaneously the price of meat is also increasing which exerts a dire need to harvest every source of meat to reduce cost, maintain quality and meet the demand. Spent hen meat attracts the meat industry as a cheaper source. In view of difficult job to utilize the spent hen meat, the present study focused on Shelflife of spent hen meat papad stored at ambient temperature.

**Materials and Methods**

The process optimization of spent hen meat papad carried out at National Research centre on Meat. Spent hens obtained from a commercial poultry farm near by Hyderabad were utilized in this study. The birds were slaughtered and dressed, manually de-boned, packed in low density polyethylene (LDPE) bags and stored overnight at 4 ± 1°C in refrigerator. The ingredients of spice mix were procured from Hyderabad local market, cleaned and dried in oven at 50°C for 2 h; grounded and sieved through 100 mesh and the fine powder obtained was stored for subsequent use. The condiment mix contained onion, and garlic; prepared afresh in appropriate ratio as fine paste.

**Formulation for spent hen papad**

The following formulation was adopted for processing of emulsion for spent hen meat papad. The raw materials and main ingredients used for product are given in Table 1. Spice mix was prepared as per the formulation developed in the laboratory. Onion and garlic were used in the ratio 3:1 as condiments.

**Emulsion preparation**

The lean meat, fat and chicken by-products (heart + gizzard + skin) were minced separately through an 8-mm plate followed by a 4-mm plate of a meat mincer (model TC 22, RIO INOX, Sirman, Italy). Salt, sodium nitrite and sodium tripolyphosphate (STPP) were added to the minced spent hen meat and chopped for about 2 min with a bowl chopper (model TC 22, RIO INOX, Sirman, Italy). After addition of ice flakes it was chopped again for 1–2 min, SGH were incorporated slowly and chopping was continued till the SGH were completely dispersed in the batter. Condiments mix, spice mix and refined wheat flour as binder were added and chopping continued for 2 min to give a fine viscous emulsion. The temperature of the emulsion varied from 10 to 12 °C.

**Preparation of spent hen meat papad**

Spent hen meat papad were prepared by manually filling the emulsion into stainless steel, rectangular box (20 cm X 10 cm X 4 cm; L X W X H) smeared with little vegetable oil in the bottom. Emulsion was manually filled by pressing with moist fingers to give uniform smooth surface without any air gaps. The filled boxes were closed by tightening the clips and tied with thread. The filled boxes were cooked in a double jacketed stainless steel vessel fitted with electric coil in the bottom. The coil was submerged in hot water and the filled boxes were placed over the stand. Here the spent hen meat emulsion was cooked by the action of steam for 40 min without any pressure; core temperature (82°C)
of the cooked emulsion block was checked. Round Core (9 cm dia and 4.5 cm height) from block, slicing core (1.5 mm thickness) and slice dried in Micro oven.

**Packaging and Storage spent hen meat pappads**

The spent hen meat papad were packed by aerobic packaging and modified atmosphere (80 per cent nitrogen and 20 per cent carbon dioxide) packaging (MAP) methods in sterile Pet/Poly pouches and packed samples were stored at ambient temperature. The samples were analysed on 0 day, 30th day and 60th day.

**pH determination**

The pH of spent hen meat papad samples was determined by AOAC (1995) method. Five g of meat sample was homogenized with 45 ml of distilled water for one minute. The pH of the meat was recorded by immersing combined glass electrode and temperature probe of the digital pH meter (Model 420A + (Orion Research, Inc. Beverly, MA, USA) directly into suspension.

**Water activity (Aw)**

The water activity ($a_w$) of spent hen meat papad was determined with a Rotronic AG analyzer (Model Hygrolab 3, Grinddelstr 6, CH-8303 Bassersdorf, U.K.). The spent hen meat papad were cut into small square pieces and placed in water activity sample cups and the measurement was carried out at 25ºC.

**TBA number**

Thiobarbituric acid reactive substances (TBARS) assay was performed as described by Buege and Aust (1978), as modified by Lee et al., (1999). After drying, duplicate 0.5 g samples were taken from the papad. Papad samples (0.5 g) were then mixed with 2.5 ml of stock solution containing 0.375% thiobarbituric acid, 15% trichloro-acetic acid and 0.25 N HCl. The mixture was heated for 10 min in a boiling water bath (100 ºC) to develop pink color, cooled in tap water, and then centrifuged at 5,500 rpm for 25 min. The absorbance of the supernatant was measured spectrophotometrically at 532 nm against a blank that contained all the reagents minus the meat. The malonaldehyde (MDA) concentration was calculated using an extinction coefficient of 1.56×10$^{5}$ cm$^{-1}$ for the pink TBA-MDA pigment. The MDA concentration was converted to TBA number (mg MDA/kg meat sample) as follows:

\[ \text{TBA No (ppm)} = \frac{\text{Sample}_{532} \times 2.77}{7.7} \]

**Free fatty acid**

Estimation for the free fatty acid content of the papad was done by using modified method (AOAC, 1975). The free fatty acid of the papad was expressed in per cent oleic acid. 50 gm sample was taken on a watch glass and transferred to the blender. Half teaspoonful of anhydrous Sodium sulphate and about 137 ml of chloroform were added. After blending for about 2 min it was filtered through whatmann No.12 filter paper. Filtrate was collected in 250 ml stopper flask. Extraction was repeated by adding 50 ml chloroform and transfer to the flask. Volume was adjusted to 250 ml mark. 25 ml of filtrate was taken in a 125 ml flask, 10 drops of phenolphthalein indicator was added. Titration was done with 0.01N alcoholic potassium hydroxide to end point.

**Proximate composition**

Moisture, crude fat and protein of meat products were determined by standard procedures of Association of official Analytical Chemists (AOAC, 1995).
**Microbiological analysis**

The microbial load of papad was assessed by total viable count (TVC), coliform count, staphylococcal count, and Yeast and mould. It was done by spread plate technique. Duplicate 0.1 ml volume of inoculums of suitable dilutions were spread using sterile ‘L’ shaped spreader over the surface of pre-poured petridishes. The plates were incubated at 35 to 37°C for 48 hours. Counts were expressed as log10 cfu/g of sample.

**Sensory evaluation**

The sensory panel consisted of experienced scientists and highly skilled labour. The panelists were explained the nature of the experiments without disclosing the identity of the samples and were asked to rate them on eight point descriptive scale on the sensory evaluation proforma for different attributes. They were requested to record their preference on 8 point hedonic scale (8=extremely desirable, 1=extremely undesirable) (Keeton, 1983) for appearance, flavour, texture, crispiness and acceptability. Water was provided to rinse the mouth between tasting the samples.

**Statistical analysis**

All data were analysed using SPSS(version 10.0 for Windows, SPSS, Chicago, Ill., USA). Three trials were conducted and the measurements were One way ANOVA was applied for proximate composition. However, in case of storage data two-way ANOVA was performed. Significant means were separated using the least significance difference (LSD) test (P<0.05).

**Results and Discussion**

The present study was carried out to study the shelf life of spent hen meat papad stored at ambient temperature. The spent hen meat papad were prepared and packed by aerobic packaging and modified atmosphere (80 per cent nitrogen and 20 per cent carbon dioxide) packaging (MAP) methods in sterile Pet/Poly pouches and packed samples were stored at ambient temperature. The results thus obtained were interpreted and discussed in detail for storage periods with statistical inferences.

**Proximate composition**

The proximate composition of spent hen papad given in the table -2. It is a meat product that has good nutritive value i.e 48.72±1.03 protein.

**Physicochemical characteristics**

The Physicochemical characteristics of spent hen papad given in the table -3. There was no significant difference in the pH values of meats between the two packaging systems at the start of storage; however, significant differences in pH were found after 30, and 60 days of storage. The pH of aerobic packaged samples decreased from 6.18 to 5.82 and that of the MAP samples from 6.18 to 5.89 during the storage at ambient temperature.

The pH levels decreased gradually during the entire period of storage and significant (P < 0.05). The pH scores decreased gradually during the entire period of storage and the present findings agreed with the results of Modi et al., (2007) during the storage of dehydrated Kebab mix; of Bennani et al., (2000) for kaddid, a salted, dried mutton; of Rubio et al., (2007) for a dry cured Spanish sausage, salchichon and Mishra et al., (2015) for dehydrated chicken meat rings. The decreasing trend in pH value was attributed to the chemical activity as hydrolytic rancidity increases free fatty acid level but not to the microbial activity.
The pH values of the aerobic and MAP were comparable to each other on 0 day whereas on days 30 and 60 of storage the MAP had a significantly (P < 0.01) higher value than the aerobic. Precooking of meat also increases the pH value in dried meat products due to change in net charge of proteins during denaturization (Babu et al., 1994; Kharb and Ahlawat, 2010). Water activity decreased significantly (P < 0.05) during the entire period of storage. Dried products usually have aw below 0.7 (Lewicki, 2004).

The formation of secondary lipid oxidation products, such as malonaldehyde can be reported in terms of TBARS numbers. The TBARS value increased significantly (P < 0.05) during storage as compared to the initial value. However, there was significantly (P < 0.05) lower value of TBARS were observed in the MAP than the aerobic during the entire period of storage. The initial high 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 the mixing of various meat constituents and prooxidants. Missra et al., (2015) reported a similar trend in TBARS values during storage of dehydrated chicken meat rings, which was attributed to the reactions of malonyldialdehyde with proteins. There was no rancid flavor detected by sensory panelists of this present study, as the threshold value for rancidity in meat was reported as 1-2 mg malondialdehyde/kg (Alkass et al., 2013). The free fatty acid content of papad remained comparable up to day 30 of storage in both packaging and then increased significantly (P < 0.05) in aerobic packaging on 60th day of storage. However, there were significantly (P < 0.05) lower values of free fatty acid in the MAP than the aerobic packaging on 60th day of storage. Lipase action in meat products during storage causes a gradual increase in FFA values in dehydrated meat products (Modi et al., 2007; Chukwu and Imodiboh, 2009).

Microbiological characteristics

The mean values for different microbiological parameters are presented in Table -4. During the whole storage period, TPC for the aerobic packaging was higher than that of the MAP. No coli forms and Staphylococcus aureus were detected throughout the storage study. Yeast and molds were not detected on day 0 of ambient storage in both aerobic and MAP products but they increased significantly (P < 0.05) during storage. In the both packaging, there was significant (P < 0.05) increase in yeast and molds with subsequent storage interval.

During the whole storage period, TPC for the aerobic packaging was higher than for the MAP. Higher total plate counts were observed in aerobic packaging might be due to the higher oxygen levels in the product atmosphere and the absence of antimicrobial agents. These results are in agreement with that of Singh et al., (2009) and Missra et al., (2015) who also reported an increase in total plate counts in aerobically packed chicken snacks stored at ambient temperature. No coliforms were detected throughout the storage study. Das and Jayaraman (2003) had reported absence of coliforms during ambient temperature storage of dehydrated chicken pulav. The absence of yeast and mold count on day 0 of storage might be due to the low water activity at the initial stage. Singh et al., (2009) observed an increase in yeast and mold counts in aerobically packed chicken snacks stored at ambient temperature.

Sensory qualities

Mean sensory scores of products are presented in Table 5. There was a decreasing trend observed in appearance of the products during entire storage.
Table 1. Spent hen meat papad formulation

| Ingredients                  | (%) |
|-----------------------------|-----|
| Spent hen meat              | 65  |
| Skin heart and gizzard (SHG)| 15  |
| Maida                       | 3   |
| Spice mix                   | 1.5 |
| Condiments                  | 3.5 |
| Salt                        | 1.5 |
| Sod. tripolyphosphate       | 0.3 |
| Sugar                       | 0.3 |

Table 2. Proximate composition of spent hen papad

|                | Moisture | Protein | Fat    | Ash    |
|----------------|----------|---------|--------|--------|
| Control        | 10.3±0.02| 48.72±1.03| 10.36±0.02| 3.2±0.02|

Table 3. Physicochemical characteristic of spent hen meat pappads at ambient temperature

| Parameters     | Packaging methods | 0       | 30th day | 60th day |
|----------------|-------------------|---------|----------|----------|
| **Ph**         | aerobic           | 6.18±0.01<sup>a</sup> | 5.90±0.03<sup>b2</sup> | 5.82±0.04<sup>c2</sup> |
|                | MAP               | 6.18±0.01<sup>a</sup> | 6.05±0.07<sup>b1</sup> | 5.89±0.01<sup>b1</sup> |
| **TBA**        | aerobic           | 0.571±0.02<sup>c</sup> | 0.654±0.03<sup>b1</sup> | 0.777±0.05<sup>a1</sup> |
|                | MAP               | 0.571±0.02<sup>c</sup> | 0.591±0.01<sup>b2</sup> | 0.742±0.02<sup>a2</sup> |
| **aw**         | aerobic           | 0.5875±0.01 | 0.537±0.00 | 0.517±0.01 |
|                | MAP               | 0.5875±0.01 | 0.531±0.00 | 0.522±0.01 |
| **FFA (% oleic acid)** | aerobic | 1.37±0.02<sup>b</sup> | 1.47±0.01<sup>b</sup> | 1.67±0.02<sup>a2</sup> |
|                | MAP               | 1.37±0.02 | 1.41±0.18 | 1.48±0.01<sup>1</sup> |

*Means ± standard errors (SE) with different superscripts row-wise (letters) and column-wise (numbers) differ significantly (P < 0.05).

Table 4. Microbial qualities of spent hen meat pappads stored at ambient temperature (log cfu/g)

| Microbial quality | Packaging method | 0       | 30th day | 60th day |
|-------------------|------------------|---------|----------|----------|
| **Total plate count** | Aerobic          | 2.37±0.02<sup>c</sup> | 2.62±0.00<sup>b1</sup> | 2.91±0.01<sup>a1</sup> |
|                   | MAP              | 2.37±0.02<sup>c</sup> | 2.53±0.01<sup>b2</sup> | 2.68±0.02<sup>a2</sup> |
| **Yeast and mould** | Aerobic          | ND      | 2.49±0.12<sup>b1</sup> | 2.62±0.04<sup>a1</sup> |
|                   | MAP              | ND      | 2.20±0.05<sup>b2</sup> | 2.47±0.02<sup>a2</sup> |

*Means ± standard errors (SE) with different superscripts row-wise (letters) and column-wise (numbers) differ significantly (P < 0.05). ND = Not detected.
Table 5 Sensory qualities of spent hen meat pappads stored at ambient temperature

| Parameters                      | Packaging methods | 0               | 30th day         | 60th day         |
|---------------------------------|-------------------|-----------------|------------------|------------------|
| Appearance/colour               | Aerobic           | 7.29±0.10<sup>a</sup> | 7.12±0.05<sup>b</sup> | 6.88±0.09<sup>c</sup> |
|                                 | MAP               | 7.29±0.10<sup>a</sup> | 7.25±0.08<sup>a</sup> | 6.93±0.07<sup>b</sup> |
| Flavour                         | Aerobic           | 7.37±0.05<sup>a</sup> | 7.12±0.10<sup>b</sup> | 6.66±0.10<sup>c</sup> |
|                                 | MAP               | 7.37±0.05<sup>a</sup> | 7.25±0.10<sup>b</sup> | 6.88±0.09<sup>c</sup> |
| Texture                         | Aerobic           | 7.03±0.10<sup>a</sup> | 7.00±0.00<sup>a</sup> | 6.88±0.09<sup>b</sup> |
|                                 | MAP               | 7.03±0.10<sup>a</sup> | 7.00±0.00<sup>a</sup> | 6.93±0.07<sup>b</sup> |
| Crispiness                      | Aerobic           | 7.37±0.09<sup>a</sup> | 7.00±0.00<sup>b2</sup> | 6.88±0.09<sup>c2</sup> |
|                                 | MAP               | 7.37±0.11<sup>a</sup> | 7.25±0.16<sup>b1</sup> | 6.93±0.07<sup>c1</sup> |
| over all acceptability          | Aerobic           | 7.25±0.08<sup>a</sup> | 7.00±0.00<sup>b</sup> | 6.93±0.07<sup>c</sup> |
|                                 | MAP               | 7.25±0.08<sup>a</sup> | 7.12±0.00<sup>a</sup> | 7.00±0.00<sup>b</sup> |

*Means ± standard errors (SE) with different superscripts row-wise (letters) and column-wise (numbers) differ significantly (P < 0.05).

The score for appearance in the aerobic packaging decreased significantly (P < 0.01) in entire storage periods but within level. The score of MAP for appearance was almost stable up to day 30 but scores decreased significantly (P < 0.05) on 60 day. Das and Jayaraman (2003) reported a significant (P < 0.05) decrease in color of dehydrated chicken pulav during storage at ambient temperature and non significantly (P > 0.05) at chiller temperature. Furthermore Mishra et al., (2015) reported a significant (P < 0.05) decrease in color of dehydrated chicken meat rings at ambient temperature.

The score for flavour in the both packaged papad decreased significantly (P < 0.05) in entire storage periods but with within level. Flavour score for the MAP was higher than the aerobic during the entire period of storage. The progressive decrease in flavor scores could be correlated to an increase in TBARS number and free fatty acids in the meat products under aerobic conditions. Moreover Mishra et al., 2015 reported a significant (P < 0.05) decrease in flavour of dehydrated chicken meat rings at ambient temperature. In disparity Kharb et al., (2010) reported a non significant (P > 0.05) decrease in flavor scores for dehydrated spent hen meat mince in ambient temperature storage.

The texture score of the aerobic and MAP shows no significant difference up to day 30 of storage period. Singh et al., (2009) reported a nonsignificant (P > 0.05) decrease in the texture scores in snacks containing broiler spent hen meat, rice flour, and sodium caseinate. Smith et al., (1991) also reported no difference in mouth feel, taste, and texture of fermented beef snack during storage at room temperature (24 °C) for about 30 days. The crispiness score was significant decrease (P < 0.05) with progressive increase in period of storage in both packaging. Crispiness score for the MAP was higher than the aerobic during the entire period of storage.

The score for over all acceptability of aerobic packaging decreased significantly (P < 0.05) with progressive increase in period of storage periods. The score of MAP for over all acceptability was almost stable up to day 30 but scores decreased significantly (P < 0.05) on day 60. In Addition, over all acceptability score for the MAP was significantly (P < 0.05) higher than the aerobic during the 60 day of storage. The decrease in overall...
Acceptability could be due to increase in lipid oxidation, pigment oxidation, and degradation of proteins and fats in dehydrated chicken meat papad over the period of storage. Kharb et al., (2010) observed a non significant decrease in the acceptability of dehydrated chicken meat mince during storage. Das and Jayaraman (2003) reported a significant decrease in overall acceptability of dehydrated chicken pulav during storage at ambient temperature and nonsignificantly at chiller temperature. Furthermore Mishra et al., (2013) reported a significant (P < 0.05) decrease in overall acceptability of dehydrated chicken meat rings at ambient temperature.

Based on the results, it could be concluded that the product can be stored in aerobically and modified atmosphere packaged in Petpoly pouches for 60 days without much change in physicochemical, microbiological, and sensory properties.

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