***Original Research Article***

**EFFECT OF POMEGRANATE *(****Punica granatum****)* PEEL EXTRACT ON QUALITY OF MINCED CHICKEN**

**ABSTRACT**

Synthetic antioxidants are used to prevent lipid oxidation in meat and meat products. However, their use is restricted because of their possible health hazards. This has led to a growing interest in use of natural preservatives in meat and meat products. The study was undertaken to evaluate the effect of pomegranate *(Punica granatum)* peel extract on quality of minced chicken. Pomegranate peel aqueous (PPAE) and ethanol (PPEE) extracts were prepared separately by mixing the pomegranate peel powder with the distilled water and ethanol respectively in 15:1 (w/w). The minced chicken was divided into four groups. Meat from the first group was used without any treatment (TO), whereas, that from second and third group was treated by adding PPAE (T1) and PPEE (T2) respectively @ 200 ppm. The chicken from fourth group was treated by adding 100 ppm BHT (T3) and served as the positive control. The samples were aerobically packaged in LDPE pouches, labelled and stored under refrigeration (4±1°C). The analysis of samples for sensory, physico-chemical and microbiological attributes was conducted on 0th, 2nd, 4th, and 6th day of storage. PPAE and PPEE possessed the antioxidant activity comparable to the BHT. PPAE and PPEE improved the shelf life of the minced chicken up to six days compared to the four days that of the untreated samples at refrigeration (4±1°C) storage. Further studies are necessary to assess the potential of PPAE and PPEE for preservation of the meat and meat products on commercial level.

**1. INTRODUCTION**

Meat is an important source of high-quality protein, essential amino acids, and micronutrients such as selenium, iron, and vitamins A, B12, and folic acid (Biesalski, 2005; Lawrie & Ledward, 2006). Poultry meat is especially valued for its high polyunsaturated fatty acid content, making it a healthier option for consumers (Milicevic *et al*., 2015). However, the presence of unsaturated fats also makes chicken highly susceptible to lipid oxidation, leading to rancidity, off-flavors, and reduced shelf life (Dominguez *et al*., 2014). Although synthetic antioxidants like BHT and BHA are commonly used to delay lipid oxidation, their potential toxicological effects have raised consumer concerns and regulatory restrictions (Iqbal & Anwar, 2005; Naveena *et al*., 2008). As a result, interest of the scientists and meat industry personnel has grown significantly in natural antioxidants from plant sources. Pomegranate (*Punica granatum*), particularly its peel, is rich in polyphenolic compounds with proven antioxidant and antimicrobial activities (Zarfeshany *et al*., 2014; Akhtar *et al*., 2015). Pomegranate peel, a major byproduct of the pomegranate processing industry, contains higher concentrations of bioactive compounds than the pulp or seeds and has been used traditionally in herbal medicine (Saroj *et al*., 2020). Very limited data is available about the applications of pomegranate peel in preserving meat and meat products. Therefore, the study was undertaken to evaluate the potential of pomegranate peel extract as a natural preservative in minced chicken under refrigerated storage.

**2. MATERIALS AND METHODS**

**2.1 Materials**

The deskinned and deboned chicken obtained from healthy chicken (broiler birds) with an average of 2.0 kg live weight was procured hygienically on ice from approved meat shops in the local market. Borosil make glassware and Himedia make chemicals were used for conducting the experiments. Fresh healthy and mature pomegranate fruits of Ganesh variety were purchased from local market. Pouches made up of low-density polyethylene (LDPE) with a thickness of 55 were purchased from the local market and used for packaging of the samples of minced chicken.

**2.2 Methods**

**2.2.1 Sample Preparation:**

**Preparation of Pomegranate Peel Extracts (PPE):**

Pomegranate peel aqueous and ethanol extracts were prepared following the method of Zhenbin *et al*. (2011). Pomegranate peel powder was extracted using distilled water and ethanol separately in a 15:1 (w/w) ratio. The mixtures were stirred intermittently at room temperature for 12 hours, filtered through Whatman filter paper number 1, and the filtrates were dried at 50°C till the constant weight was attained. The resulting aqueous (PPAE) and ethanol (PPEE) extracts were packaged in LDPE pouches and stored under refrigeration (4±1°C) until further use.

**Preparation of chicken samples:**

The chicken was minced with the help of meat mincer by using 6 mm plate and divided into four groups i.e. untreated control (T0), treated with PPAE @ 200 ppm (T1), treated with PPEE @ 200 ppm (T2), and treated with BHT @ 100 ppm (T3), packaged in LDPE pouches and stored at refrigeration (4±1°C).

**2.2.2 Analytical methods:**

**Analysis of PPE:**

The PPE were analyzed for the following parameters.

1. **pH:** The pH of the PPAE and PPEE was determined by the method of AOAC (2012).
2. **Antioxidant Activity:** The antioxidant activity of PPAE, PPEE, and BHT was estimated by performing the DPPH (1,1-Diphenyl-2-picrylhydrazyl) radical scavenging assay suggested by Chaubey *et.al.,* (2021).

**Analysis of chicken samples:**

The aerobically packed chicken samples were examined at 0th, 2nd, 4th and 6th day of storage for the following parameters.

**Physico-chemical properties:**

1. pH: The pH was determined by the method of AOAC (2012).
2. Thiobarbituric Acid Reactive Substances (TBARS) value was estimated by the method suggested by Pikul *et al.* (1989).
3. Tyrosine value was estimated by the method described by Strange *et al.* (1977).

**Microbiological parameters:**

Total Plate Count (TPC), Psychrophilic count (PC), and Yeast and mold count (YMC) were determined by following the standard methods of APHA (1984).

**Sensory Analysis**:

The sensory analysis was carried out by semi-trained panelists. The samples were evaluated on the basis of various sensory attributes by using 5 point descriptive scale (Dzomba *et al.,* 2014)**.**

**2.3 Statistical analysis:**

The experiment was repeated three times, and the data generated during the study was analyzed by following the standard procedure (Snedecor and Cochran, 1989).

**3. RESULTS AND DISCUSSION**

**3.1 Properties of PPE:**

The results (Table 1) revealed that the pH of PPAE and PPEE was acidic. Both the extracts had an antioxidant activity comparable with that of BHT, the synthetic antioxidant. Similar results have been documented by Kumar and Neeraj (2018).

Table 1. Properties of PPE

|  |  |  |
| --- | --- | --- |
| Antioxidant | pH | Antioxidant activity  (% DPPH RSA) |
| PPAE | 3.605±0.12 | 88.940±0.85b |
| PPEE | 2.653±0.09 | 93.007±0.57a |
| BHT | NA | 91.115±0.31a |
| F value | 0.538NS | 10.904\*\* |

*Means ± (S.E.) bearing different superscripts between rows differ significantly. \* Significant value (p<0.05); \*\* Highly significant value (p<0.01); NS: Nonsignificant. n=6*

**3.2 Effect of PPE on sensory attributes of minced chicken:**

# The data on the effect of PPAE and PPEE on sensory attributes of minced chicken stored at refrigeration (4±1°C) is presented in Table 2.

The treatment of chicken with PPAE, PPEE and BHT did not affect the colour and appearance, odour and overall acceptability of chicken samples on day zero. However, the scores of all the treated samples for all three sensory parameters were significantly (p<0.01) higher than the untreated sample from second day onwards till end of the storage and that of PPEE treated samples were significantly (p<0.01) higher than all other samples on sixth day of storage. The untreated samples demonstrated the most rapid degradation of all the sensory parameters with advancement of storage periods, reaching to undesirable odour and acceptability by the fourth day. The results demonstrated that PPAE and PPEE preserved the chicken samples for longer period in consistent with BHT, the commercially used preservative as compared to the untreated samples.

The deterioration of odour may be ascribed to the lipid oxidation of chicken samples and increase in the bacterial load with advancement of storage periods leading to generation of sulphureous compounds in the samples (Khare *et al*, 2016). The preservative effect of PPAE and PPEE may be ascribed to its antioxidant activity demonstrated during the investigation. The results were in agreement with those of Abdel *et al.* (2016) recorded while studying the utilization of pomegranate peels for improving quality attributes of refrigerated beef burger, Sharma and Yadav (2020) recorded while studying the effect of pomegranate peel and bagasse powder and their extracts on quality of chicken patties, Nandnawre *et al.* (2021) while studying the effect of custard apple peel extract on quality of minced chicken, and ElBeltagy *et al* (2022 ) while evaluating the quality of cold stored (5±2°C) meat balls formulated with 0.5% (W/W) of nano-pomegranate peel.

# Table 2: Effect of PPE on sensory attributes of minced chicken at refrigeration (4±1°C) storage.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Treat-  ment | Sensory score | | | | F value |
| Day 0 | Day 2 | Day 4 | Day 6 |
| Appearance & Colour | | | |
| T0 | 4.78±0.09A | 3.94±0.15Bb | 3.89±0.15Bb | 1.89±0.11Cc | 89.039\*\* |
| T1 | 4.67±0.11A | 4.31±0.11Bab | 4.11±0.08Bab | 3.61±0.11Cab | 18.055\*\* |
| T2 | 4.55±0.11A | 4.22±0.10Aab | 4.53±0.12Aa | 3.86±0.10Ba | 9.115\*\* |
| T3 | 4.64±0.10A | 4.59±0.11Aa | 4.33±0.11Aa | 3.33±0.14Bb | 27.374\*\* |
| F value | 0.778NS | 5.101\* | 5.403\*\* | 58.010\*\* |  |
| Odour | | | | | |
| T0 | 4.69±0.09A | 3.81±0.12Bb | 2.64±0.12Cb | 1.50±0.12Db | 147.478\*\* |
| T1 | 4.67±0.11A | 4.44±0.12Aa | 4.31±0.11Aa | 3.67±0.11Ba | 14.545\*\* |
| T2 | 4.58±0.10A | 4.58±0.10Aa | 4.44±0.11Aa | 3.83±0.07Ba | 13.406\*\* |
| T3 | 4.67±0.11A | 4.39±0.10Aa | 4.36±0.11Aa | 3.50±0.12Ba | 22.040\*\* |
| F value | 0.223NS | 9.666\*\* | 60.145\*\* | 100.632\*\* |  |
| Overall acceptability | | | | | |
| T0 | 4.78±0.08A | 3.56±0.11Bb | 2.39±0.10Cb | 1.50±0.12Db | 179.331\*\* |
| T1 | 4.61±0.11A | 4.39±0.10Aba | 4.25±0.12Ba | 3.33±0.10Ca | 30.459\*\* |
| T2 | 4.44±0.11A | 4.56±0.11Aa | 4.39±0.11Aa | 3.67±0.10Ba | 17.000\*\* |
| T3 | 4.69±0.10A | 4.56±0.12Aa | 4.36±0.11Aa | 3.44±0.12Ba | 24.473\*\* |
| F value | 2.083NS | 18.563\*\* | 76.996\*\* | 81.967\*\* |  |

*Means ±(S.E.) bearing different superscripts (between column capital letters and between rows small letters) differ significantly. \* Significant value (p<0.05); \*\* Highly significant value (p<0.01); NS: Nonsignificant. n=6, T0: Control, T1:200ppm, PPAE; T2: 200ppm PPEE, T3: 200ppm BHT*

**3.3 Effect of PPE on physico-chemical properties of minced chicken:**

# The data on the effect of PPAE and PPEE on physico-chemical properties of minced chicken stored at refrigeration (4±1°C) is presented in Table 3.

The pH, TBARS and Tyrosine values of all the chicken samples increased with advancement of the storage periods, where, the increment in pH was nonsignificant. The increase was rapid in case of the untreated samples compared to the treated samples. The results demonstrated that PPAE, PPEE and BHT slowed down the rate of pH rise of the chicken samples as compared to the untreated chicken samples. TBARS and Tyrosine Values of untreated samples crossed the threshold limit of the spoilage on fourth day, while those of treated samples were within the threshold limit of spoilage till end of the storage in which PPEE performed better than the PPAE and BHT. This could be attributed to the antioxidant and antibacterial activity of the extract.

Similar results have been documented by Devtakal *et al.* (2014) for the ground goat meat and nuggets treated with pomegranate peel extract and by Ghimire *et al.* (2022) for pomegranate peel extract treated ground buffalo meat.

# Table 3: Effect of PPE on physico-chemical properties of minced chicken at refrigeration (4±1°C) storage

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Treatment | 0 Day | 2 Day | 4 Day | 6 Day | F value |
| TBARS (mg Malanoaldehyde/kg) | | | |  |
| T0 | 0.23±0.04B | 0.42±0.06Ba | 0.88±0.02Aa | 1.02±0.03Aa | 7.486\*\* |
| T1 | 0.21±0.02C | 0.35±0.04ABab | 0.47±0.04Bb | 0.64±0.08Ab | 12.524\*\* |
| T2 | 0.25±0.08B | 0.23±0.04Bb | 0.38±0.06Bb | 0.58±0.05Ab | 8.060\*\* |
| T3 | 0.24±0.04B | 0.38±0.06Bab | 0.86±0.01Aa | 0.72±0.01Aa | 11.262\*\* |
| F value | 0.129NS | 2.782\* | 14.346\*\* | 1.585\* |  |
| Tyrosine value (mg/100g) | | | | | |
| T0 | 14.80±1.37B | 17.03±0.43B | 18.02±1.91B | 23.67±0.82Aa | 8.938\*\* |
| T1 | 13.94±0.42B | 15.11±1.50B | 16.78±2.06AB | 20.58±0.45Ab | 4.886\*\* |
| T2 | 13.06±0.38B | 15.11±0.76B | 15.51±1.60B | 20.25±0.16Ab | 11.169\*\* |
| T3 | 14.43±1.05B | 15.08±0.86B | 16.63±0.60B | 23.33±0.50Aa | 27.156\*\* |
| F value | 0.687NS | 0.991NS | 0.390NS | 11.167\*\* |  |
| pH | | | | | |
| T0 | 5.85±0.85 | 5.92±0.10 | 6.23±0.14 | 6.32±0.31 | 1.556NS |
| T1 | 5.83±0.13 | 5.87±0.32 | 5.89±0.16 | 5.91±0.38 | 0.016 NS |
| T2 | 5.81±0.19 | 5.85±0.85 | 5.88±0.10 | 5.92±0.10 | 0.138 NS |
| T3 | 5.82±0.10 | 5.75±0.37 | 5.86±0.17 | 5.88±0.18 | 0.063 NS |
| F value | 0.017 NS | 0.077 NS | 1.469 NS | 0.620 NS |  |

*Means ± (S.E.) bearing different superscripts (between column capital letters and between rows small letters) differ significantly. \* Significant value (p<0.05); \*\* Highly significant value (p<0.01); NS: Nonsignificant. n=6, T0: Control, T1:200ppm PPAE; T2:200ppm PPEE, T3: 200ppm BHT*

**3.4 Effect of PPE on microbiological properties of minced chicken:**

# The data on the effect of PPAE and PPEE on microbiological properties of minced chicken stored at refrigeration (4±1°C) is presented in Table 4.

TPC and PSC of all the samples increased progressively with advancement of the storage periods, where, rise in treated samples was nonsignificant and untreated samples was significant (p<0.01). TPC of the untreated samples increased most rapidly crossing the threshold limit of meat spoilage on fourth day of storage, whereas, that of all treated samples was within the limit of spoilage till end of the storage. The yeast and mold count was detected on the last i.e. sixth day of the storage. It was significantly (p<0.01) higher in the untreated samples compared to the treated samples. Similar results have been recorded by Hafssa *et al.* (2015) for beef sausage containing pomegranate peels under refrigeration, Sharma, and Yadav (2020) for chicken patties treated with pomegranate peel and bagasse powder and their extracts and Dua *et al.* (2016) in fat rich meat treated with pomegranate rind extract.

# Table 4: Effect of PPE on microbiological properties of minced chicken at refrigeration (4±1°C) storage

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Treatment | 0 Day | 2 Day | 4 Day | 6 Day | F Value |
| **Total Plate count (log10 CFU/g)** | | | | | |
| T0 | 4.66±0.36B | 4.69±0.21B | 6.71±0.40Aa | 6.78±0.35Aa | 12.786\*\* |
| T1 | 4.28±0.26 | 4.32±0.31 | 4.48±0.30b | 4.56±0.38b | 0.206NS |
| T2 | 4.22±0.30 | 4.29±0.30 | 4.32±0.22b | 4.41±0.25b | 0.749NS |
| T3 | 4.32±0.25 | 4.39±0.25 | 4.48±0.30b | 4.59±0.30b | 0.176NS |
| F Value | 0.449NS | 0.515NS | 11.899\*\* | 12.280\*\* |  |
| **Psychrophilic count (log10 CFU/g)** | | | | | |
| T0 | 00D | 2.36±0.23Ca | 3.58±0.38Ba | 4.67±0.4Aa | 44.166\*\* |
| T1 | 00C | 00Cb | 2.48±0.29Bb | 3.28±0.21Ab | 92.429\*\* |
| T2 | 00C | 00Cb | 2.27±0.19Bb | 3.02±0.15Ab | 174.620\*\* |
| T3 | 00C | 00Cb | 2.66±0.35Bb | 3.36±0.22 Ab | 74.381\*\* |
| F Value |  | 102.132\*\* | 3.503\* | 7.947\*\* |  |
| **Yeast and Mold Count (log10 CFU/g)** | | | | | |
|  | 4 Day | | 6 Day | | F Value |
| T0 | 00B | | 1.08±0.03 Aa | | 1128.774\*\* |
| T1 | 00B | | 0.90±0.17 Aab | | 27.614\*\* |
| T2 | 00B | | 0.30±0.06 Ac | | 27.000\*\* |
| T3 | 00B | | 0.60±0.16 Abc | | 13.500\*\* |
| F Value |  | | 7.792\*\* | |  |

*Means ± (S.E.) bearing different superscripts (between column capital letters and between rows small letters) differ significantly. \* Significant value (p<0.05); \*\* Highly significant value (p<0.01); NS: Nonsignificant. n=6, T0: Control, T1:200ppm PPAE; T2:200ppm PPEE, T3: 200ppm BHT*

**4. CONCLUSIONS**

1. PPAE (200 ppm) and PPEE (200 ppm) possessed antioxidant activity, 88.940±0.85, and 93.007±0.57 % DPPH RSA respectively that was comparable with that of the synthetic antioxidant, BHT (100 ppm) 91.115±0.31% DPPH RSA.
2. Incorporation of PPAE and PPEE @ 200 ppm improved the shelf life of the minced chicken packed aerobically in LDPE pouches of 50 µ thickness up to six days compared to the four days that of the untreated samples at refrigeration (4±1°C) storage.
3. Further studies are necessary to assess the potential of PPAE and PPEE for preservation of the meat and meat products on commercial level.

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