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# Comparative nutritional and phytochemical analysis of indigenous Pakistani Strawberry varieties along with shelf-life extension using modified atmospheric packaging technology

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The present study was conducted to check the nutritional characterization *i.e.,* proximate analysis, physicochemical properties (total soluble solids content, titratable acidity, texture and color), phytochemicals (total phenolic, flavonoids & anthocyanin) and antioxidant capacity using DPPH-2,20-diphenyl-1-picrylhydrazyl radical scavenging capacity of strawberry varieties (festival, chandler and korona) produced in Pakistan along with shelf life extension using modified atmosphere packaging (MAP) technology. Accordingly, samples were collected from farms and local market for analysis. Results showed statistically significant differences ($p< 0.05$) for proximate composition expect total ash and crude fat content. Higher moisture content documented in chandler variety as 91.12±7.28%, highest ash in korona variety as 1.4±0.17%, lowest protein in chandler as 0.95±0.13% and highest fat in festival variety as 1.13±0.31. Physicochemical analysis findings showed highest titratable acidity as 0.70±0.04 g citric acid/100g in chandler, highest °Brix as 8.41±0.11 in festival whereas, phytochemicals analysis revealed highest total phenolic and flavonoids in chandler as 926.17±53.08 mg GAE/100 g and 180.63±15.72 mg/100g, respectively, and lowest 864.44±44.16 mg GAE/100 g, 144.84 ± 12.11 mg/100g in variety korona, respectively. The chandler variety displayed highest antioxidant activity (82.09±6.34%) and anthocyanin content as 2.74 ± 0.09 mg/100g) however; non-significant differences were documented for color values. Furthermore, shelf life of strawberry samples extended by 09 days subjected modified atmospheric packaging under cold storage for 12 days. For this strawberry was categorized into 5 groups with different gas combination *i.e.,* $MAP_0$ (control group having 0.05% $CO_2$, 20.95% $O_2$ and 79% $N_2$), and 4 treatment groups $MAP_1$ (3% $CO_2$, 15% $O_2$ and 82% $N_2$), $MAP_2$ (5% $CO_2$, 10% $O_2$ and 85% $N_2$), $MAP_3$ (10% $CO_2$, 5% $O_2$ and 85% $N_2$), $MAP_4$ (15% $CO_2$, 3% $O_2$ and 82% $N_2$). Statistical differences ($p< 0.05$) were found between groups on different storage days (3$^{rd}$ , 6$^{th}$ , 9$^{th}$ , and 12$^{th}$ ) in physiochemical, phytochemical, sensory evaluation, and caloric parameters. In nutshell, the results provide valuable information on nutritional composition, phytochemical characteristics, and quality aspects of different strawberry varieties produced in Pakistan. This information is helpful for food processors in value addition, and modified atmosphere packaging technology for the reduction of post-harvest losses at different supply chain level.
**Keywords:** Strawberry, Varieties, Phytochemical, Antioxidant, Shelf life, Modified Atmospheric Packaging.

## INTRODUCTION

Strawberry (Fragaria ananassa, Duch.) one is the most widely consumed fruits in the world and its cultivation has been increasing in different regions of Pakistan, especially Punjab and Khyber Pakhtunkhwa (KPK). In Punjab, strawberry-growing areas include Nankana Sahib, Sheikhupura, Sargodha and Sialkot whereas in KPK, include Swat, Abbottabad, Mirpur, Mardan and Peshawar. Furthermore, it has been documented that strawberry cultivation has risen by 22% in recent years and is expected to increase due to its taste preference, consumer & processing acceptability, application in different food products, as well as health benefits upon its consumption (Mehmood *et al.,* 2018).

As a fruit of Mediterranean diet, strawberries are appreciated for their high nutritional compositions in terms of phytochemicals & flavonoids, sensory quality, unique aroma and sweet taste. The strawberry is popular in both forms, i.e. fresh and processed versions all over the world however, fresh strawberries are mainly consumed as a fruit in Pakistan, while processed strawberries are being used in products like jams, juices, nectars, beverages, smoothies, milk shakes, ice cream, yoghurts and desserts etc. Literature has already reported strawberry fruit has high nutritional value due to its

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micronutrient profile and phytochemicals diversity (Li *et al.,* 2020).

Strawberries are one of the excellent sources of bioactive substances, including phenolic compounds, total flavonoids, anthocyanins and antioxidants that are considered vital for the optimal human health. Anthocyanins are one of the major polyphenols present in strawberry and it is considered as rich sources of dietary polyphenols as provide over 1 mg of total polyphenols per serving of the fruit (Chatham *et al.,* 2020). Additionally, nutrient content of strawberry is affected by several factors like agricultural practices and climatic conditions that may differ overall quality and bioactive properties of fruit (Ganhão *et al.,* 2019).

Strawberry is one of the highly perishable fruit essentially harvested at fully ripe stage thus offer very limited (approximately 1- 2 days at room temperature) shelf life of the produce that demand the on spot consumption or subjected to processing. The higher metabolism rate ultimately promote degradation process resulting spoilage of fruit. Thus, due to limited shelf-life of the fruit, it is mainly consumed in the local areas and its access to far off areas is very limited thus it is quite difficult to access remote locations. The one and only method to reduce deterioration is temperature management as well as increasing fruit quality and the post-harvest life of strawberry fruits. Various preservation methods are used for increasing the shelf life of fruits. Among them fungicides are known to control many plant pathogenic fungi, but application of fungicides showed negative effect in our environment as well as reduce the number of soil fungi. Another method of preservation is freezing that is relatively quick process but results in detrimental loss of taste and vitamins B and C in freezing process. An active coating containing antifungal micro particles is also used in edible coating but still this process also required freezing and alone the process can result limited increase in shelf life of the product. Packaging technologies are also used for increasing shelf life of fruit and among them, modified atmosphere packaging (MAP) technology is one of the latest and best at protecting fresh food by reducing amount of oxygen present in a package (Jalali *et al.,* 2020; Zitouni *et al.,* 2020).

Considering the scenario, present research was carried out to examine nutritional characteristics of indigenous strawberry varieties (festival, chandler, and korona) grown in different regions of Pakistan by measuring nutritional compositions, physicochemical characteristics, phytochemical analyses, quantification of anthocyanins content, and antioxidant level as well as use of innovative packaging technology like modified atmospheric packaging technology to extend shelf life of strawberry fruit.

## MATERIALS AND METHODS

The present study was conducted at the Department of Food Science and Human Nutrition, University of Veterinary and Animal Sciences, Lahore. The chemicals and reagents for the study were obtained from a local trader in Lahore, Pakistan and Folin-Ciocalteu's phenol reagent (FCR), Gallic acid (anhydrous), methyl red, cyanidin 3-glucoside and sodium acetate 2,2-diphenyl-1-picrylhydrazyl (DPPH) were obtained from Merk, German. The strawberry varieties botanical confirmation was done by the Institute of Botany, Punjab University Lahore. Accordingly, fresh strawberry samples of three varieties i.e., "chandler", "Korona" and "festival" were collected from Sharqpur, Sheikhupura and commercial market of Lahore, respectively. All strawberry fruit samples were collected at their maturity stage, red colored stage, uniform in size, shape and free from any visible defects (Morris & Sistrunk 2018). For the sampling of different strawberry varieties, (≈2 kg) of each strawberry variety were picked and transported to the laboratory for processing and sample preparations, where overripe and unripe strawberries were discarded and whole strawberries of each variety processed for sample preparation and analysis. Furthermore, we divided each variety into three replicates (1.5kg = 3 x 0.5kg) and stored at 4°C for further processing. Likewise, strawberry variety "Chandler" was selected for the shelf-life study using the modified atmosphere packaging technology due to high moisture content as well as other nutritional parameters findings For shelf-life study using modified atmospheric packing technology, the strawberries samples were categorized into 5 groups with different gases combinations i.e., $MAP_0$ (control group having 0.05% $CO_2$, 20.95% $O_2$ and 79% $N_2$), and 4 treatments groups $MAP_1$ (3% $CO_2$, 15% $O_2$ and 82% $N_2$), $MAP_2$ (5% $CO_2$, 10% $O_2$ and 85% $N_2$), $MAP_3$ (10% $CO_2$, 5% $O_2$ and 85% $N_2$), $MAP_4$ (15% $CO_2$, 3% $O_2$ and 82% $N_2$). A total 25 MAP trays (5 trays of each group) prepared and kept at 4°C and stored for 12 days. The strawberry samples were subjected to a number of analyses for characterization and during MAP study trial with detail as below details as below.

***Nutritional composition of strawberry varieties:*** The proximate analysis (moisture, ash, crude fiber, crude protein, crude fat, and nitrogen free extract) of strawberry varieties samples were performed according to methods of the Association of Official Analytical Chemists (AOAC)2000 with a slight modification as described by (Ganhão *et al.,* 2019). The mineral level of samples determined using atomic absorption spectrometer (HITACHI Z-8230 polarized Zeeman) by following the protocol as described by (Trejo-Téllez *et al.,* 2014)**.** An iodine redox titration method was used for the determination of vitamin C concentration in different strawberry fruit samples by using the procedure as described by (Igwemmar *et al.,* 2013).

***Physicochemical analysis of strawberry:*** The titratable acidity (TA) of different strawberry varieties was measured according to the International Standards, based on a potentiometric titration method with a standard volumetric solution of sodium hydroxide by following the protocol as described by (Husain, 2021). Three measurements per sample group were taken. The total brix values of samples was measured by following the procedure as reported by (Agulheiro-Santos *et al.*, 2022) using refractometer (Abbe Refractometer - AR2008. A replicate sample tested three times, and results were reported as °Brix. The texture of different cultivars strawberry samples was analyzed as method described by (Cornuault *et al.*, 2018) with a slight modification. Cohesiveness, adhesiveness, and hardness of strawberry samples determined using texture analyzer (FRTS-100N-I Imada, Japan). Similarly, cohesiveness of samples was measured as breaking hydrogen bonds and deformed degree before damage, while hardness was determined using force penetration technique. A sample's adhesiveness is the amount of forces required to avoid attractive forces.

For color measurement of strawberry samples, a tristimulus colorimeter with a data processor using 8-mm diameter measurement area was used for accessing strawberry fruits' exterior color. In CIE color space, the instrument was calibrated by varying green to red (-60 to +60) and blue to yellow (-60 to +60) on a $D_{65}$. The L* values describe brightness of samples (0 for black to 100 for white) whereas, to compute tonality, hue (h°) was used: $h^{\circ} = \tan^{-1}\frac{b^*}{a^*}$

***Phytochemicals determination of strawberry varieties*:** The anthocyanins level in different strawberry cultivars samples were determined by using the protocol as described by Dzhanfezova *et al.,* (2020). In this context, 1g strawberry samples was added to a mixture of ethanol and citric acid (80:20) 1.0 mol/L. Afterwards, the slurry was passed through |whatman #1 filter paper used for filtering extract and residue and filter paper are rinsed with solvent of extraction to complete a volume of 50 mL followed by placing the extract in light-protected polyethylene bottles kept at 20°C to store extract. The collected extract was placed in a spectrophotometer set at a wavelength of ƛ= 535 nm for the absorbance measurement followed by quantification of anthocyanins in strawberries and expressed as mg of CGE/100 g (cyanidin-3-glucoside equivalents) of dry weight in strawberry samples.

The total phenolic contents of different strawberry cultivars were determined by following the method as described by Dzhanfezova *et al.,* (2020). Accordingly, 5 g strawberry samples were placed in 250 ml volumetric flask followed by addition of 20% ethanol in 2% citric acid. After this, the flask was shaken for 1 hour at $25^0$C using orbital shaker. After shaking, the samples were centrifuged at 11000 rpm for 15 min. Then took 200 µL samples and added 100 µL FCR solution followed by incubation for 3 minutes at room temperature. Then added 800 µL solution of $Na_2CO_3$ (7.5%) and incubated for 2 hrs. at room temperature. the absorbance was measured with spectrophotometer at 765 nm. Gallic acid equivalent (mg GAE/g), which was calculated from the standard curve of Gallic acid concentrations in methanol, was used to indicate the concentration of phenol.

The flavonoids contents of different strawberry cultivars were determined by following the procedure as mentioned by Ahouagi *et al., (*2021). For analysis, first of all we mixed the 1g strawberry samples with 5 mL of distilled water in a volumetric flask (50ml). After this, added 0.3 mL of 5% $NaNO_3$ to it and placed it for 5 minutes followed by addition of 0.6 mL of 10% $AlCl_3$, then after 5 minutes, added 2 mL of 1 molar sodium hydroxide (NaOH) solution and then shake it well and then made volume up to 2.4 mL distilled water. After all, solution was mixed well in the volumetric flask. The total flavonoid content was determined by checking the absorbance of flavonoids contents at 510 nm using spectrophotometer and the values were presented as mg GAE/g (Gallic acid equivalent).

For the determination of antioxidant capacity of different strawberry varieties samples, the free radical scavenging activity of the supernatants obtained from the strawberry extract was evaluated from 2,2-diphenyl-1-picrylhydrazyl (DPPH) assay by following the procedure as described by Nowicka *et al.* (2019). For the analysis, 0.1 mM solution of DPPH in methanol was diluted in 3.5 mL of 0.5 mL supernatants in ethanol at concentrations between 0.1 and 1 mg/mL. The mixture was left at room temperature for 30 minutes after shaking and blank was created using ethanol. The spectrophotometer was used to test the different samples solution's absorbance at 517 nm and ability to scavenge free radicals was determined using the formula as mentioned below

$$DPPH\ radical\ scavenging\ ability\ (\%) = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100$$

***Shelf life extension using Modified atmosphere packaging technology:*** The strawberries samples were subjected to analysis like pH, $^0$Brix, color, fruit firmness, total flavonoids, total phenolic content and sensory evaluation were stored at refrigeration temperature were determined at different storage intervals 0, 3rd, 6th, 9th and 12th storage day. The pH of strawberry samples measured using a pH meter (Sartorius PP50, Goettingen, Germany) as described by Basak *et al. (*2022), color using hunter lab colorimeter (CR-400 colorimeter, Konica Minolta Sensing Co. Tokyo, Japan) as described by Hemmati *et al.*, (2021), Fruit firmness as method described by Castro *et al.* (2021), weight loss as method described by Nguyen et al. (2021). The total flavonoid contents determined by the procedure of Ahouagi *et al. (*2021), total phenolic content as per method of (Dzhanfezova *et al.,* 2020). Similarly, the sensory evaluation analysis of strawberry samples were measured considering taste, flavor, color and overall acceptability using 9-point hedonic scale (9 = Like Extremely, 8 = Like Very much, 7 = Like Moderately, 6 = Like Lightly, 5 = neither like nor dislike, 4 = Dislike slightly, 3 = Dislike Moderately, 2 = Dislike Very much, 1 = Dislike extremely) using the procedure of Gunness *et al.* *(*2009). The microbial stability of strawberry samples subjected to Modified atmosphere packaging were evaluated by determination of total plate count bacteria (TPC), yeasts and molds population at different storage days (0, 3rd 6th, 9th and 12th day). For the analysis, the plate count agar used to determine sample's microbial count by incubating at different temperatures and times using appropriate cultures. The total bacterial count incubated on plate agar for 48 hrs. at 30 °C by following guidelines for TPC as Zhang *et al.* (2012) along with yeast & molds on yeast-glucose chloramphenicol medium incubated at 25 °C for 5 days by following the procedure as Moreno *et al.* (2012) and the microbial counts are represented as log CFU/mL.

***Statistical Analysis:*** The collected data was analyzed using Statistical Package for Social Sciences (SPSS) version 20. The data values were expressed as means and standard deviation (Mean ± SD) and to check statistically significant differences between groups of strawberries varieties, analysis of variance (ANOVA) techniques was used whereas, Tucky test was used as a post hoc test for comparison of the Means of different parameters as per the guidelines described by (Koçak *et al. (*2018).

## RESULTS

***Nutritional Composition of strawberry varieties:*** Table 1 showed the results of proximate composition of different strawberry varieties (Festival, Chandler and Korona) that indicated moisture, crude protein, crude fiber and nitrogen free extract were affected significantly. However crude fat and ash content of strawberry samples differed non-significantly. Means for moisture content (%) of different varieties ranged from 88.41±6.93 in Festival to 91.12±7.28 in Chandler.

**Table 1. Proximate Composition of different indigenous strawberry varieties (Chandler, Festival and Korona) available in Pakistan**

| Parameters | Varieties | Mean± SD | *p*-value |
|---|---|---|---|
| Moisture (%) | Chandler | 91.12 ± 7.28$^{a}$ | 0.034 |
| | Festival | 88.41± 6.93$^{b}$ | |
| | Korona | 89.06± 6.36$^{ab}$ | |
| Ash content (%) | Chandler | 1.11± 0.26$^{a}$ | 0.360 |
| | Festival | 1.23± 0.30$^{a}$ | |
| | Korona | 1.41± 0.17$^{a}$ | |
| Crude fiber (%) | Chandler | 2.32± 0.07$^{b}$ | |
| | Festival | 2.02± 0.11$^{b}$ | 0.002 |
| | Korona | 2.75 ± 0.19$^{a}$ | |
| Crude protein (%) | Chandler | 0.95± 0.13$^{b}$ | |
| | Festival | 1.17± 0.13$^{ab}$ | 0.013 |
| | Korona | 1.54± 0.22$^{a}$ | |
| Crude fat (%) | Chandler | 0.90± 0.26$^{a}$ | |
| | Festival | 1.01± 0.24$^{a}$ | 0.562 |
| | Korona | 1.13± 0.31$^{a}$ | |
| Nitrogen free extract (NFE) (%) | Chandler | 85.84± 6.81$^{a}$ | |
| | Festival | 82.98± 6.57$^{ab}$ | 0.029 |
| | Korona | 82.23± 6.51$^{b}$ | |

Data expressed as "mean ± standard deviation". Superscript letters ($^{a,\ b}$) shows the statistically differences. Different letters indicate significant differences (P < 0.05, Tukey test) between strawberry varieties.

**Table 2. Physicochemical analysis of different selected strawberry varieties**

| Physicochemical Properties | | Chandler | Festival | Korona |
|---|---|---|---|---|
| Titratable acidity (g citric acid/100 g) | | 0.70±0.01$^{a}$ | 0.41±0.03$^{b}$ | 0.44±0.04$^{b}$ |
| Soluble solid content (°Brix) | | 7.81±0.61$^{a}$ | 8.41±0.68$^{a}$ | 4.79±0.38$^{b}$ |
| Surface Color | | | | |
| L* | | 47.1±3.77$^{a}$ | 44.0±3.52$^{b}$ | 41.4±3.26$^{c}$ |
| a* | | 34.1±2.73$^{a}$ | 34.4±2.75$^{a}$ | 31.8±2.56$^{b}$ |
| b* | | 27.8±2.52$^{a}$ | 21.1±1.90$^{b}$ | 30.4±2.70$^{a}$ |
| $h^0$ | | 40.4±3.61$^{a}$ | 40.8±3.68$^{a}$ | 41.7±3.67$^{a}$ |
| Internal zone (interior) | | | | |
| L* | | 31.4±2.45$^{c}$ | 37.1±3.81$^{a}$ | 34.4±2.71$^{b}$ |
| a* | | 23.4±1.74$^{a}$ | 22.7±1.61$^{a}$ | 19.4±1.43$^{a}$ |
| b* | | 16.4±1.34$^{b}$ | 17.1±1.26$^{b}$ | 20.9±1.58$^{a}$ |
| $h^0$ | | 38.1±3.74$^{b}$ | 37.4±3.71$^{b}$ | 44.4±3.96$^{a}$ |
| Texture | | | | |
| Superficial zone (surface) | Hardness (N) | 1.11±0.19$^{a}$ | 0.51±0.17$^{b}$ | 1.14±0.41$^{a}$ |
| | Adhesiveness(N/s | 1.70±0.48$^{a}$ | 0.97±0.11$^{b}$ | 1.77±0.49$^{a}$ |
| Internal zone (interior) | Hardness (N) | 0.940±0.20$^{a}$ | 0.41±0.140$^{b}$ | 0.99±0.12$^{a}$ |
| | Adhesiveness (N/s) | 1.49±0.48$^{a}$ | 0.75±0.419$^{b}$ | 1.71±0.40$^{a}$ |

Data are expressed as "mean ± standard deviation". Superscript letters ($^{a,b}$) shows the statistically differences. Different letters indicate significant differences (P < 0.05, Tukey test) between strawberry varieties.

For ash content (%) of strawberry varieties, highest value was reported as 1.41±0.17 in Korona followed by 1.23±0.30 in Festival and lowest as in 1.11±0.26 in Chandler. For crude protein (%) highest content was documented as 1.54±0.22 in Korona variety and lowest as 0.95±0.13 in Chandler. The highest NFE content was reported in Chandler and lowest in Korona.

***Physiochemical analysis:*** The organic and non-volatile acids are found in fruits which correlate with pH values of product that ultimately affects the titratable acidity of the fruits. During the study, the strawberry varieties were compared for titratable acidity and results showed statistically significant differences ($p < 0.05$) for different strawberry samples. Results in (Table 2) indicated that Korona cultivar documented lowest value of titratable acidity and Chandler showed highest value. The Brix of strawberry varieties also indicated significant differences with highest Brix means documented in Chandler (7.81 ± 0.49) and lowest in Korona *i.e.* 4.79±0.34. Furthermore, non -significant differences were found in surface zone color of different strawberry samples that showed typical red color of fruits. Highest L* value reported in Chandler as 47.1±3.77 whereas, lowest L* value documented in Korona as 41.4±3.26. Similarly, b* values of different varieties like Chandler, Festival and Korona were as 27.8±2.52, 21.1±1.90 and 30.4±2.70 respectively. Results (Table 2) regarding texture values of strawberry fruits variety Festival showed lowest value of firmness (0.511±0.17) N and internal zone (0.411±0.140) N.

***Phytochemical quality of strawberry varieties:*** Fruits are one of the most important sources of phytochemicals and are considered vital for optimal human health taken from human diets and their health benefits are mainly attributed to higher antioxidant activity. The strawberry samples were subjected to various phytochemical analysis including total phenolic contents (TPC), total flavonoids content (TFC) and anthocyanins (AC) contents. Results (Table 3) of the phytochemical parameters revealed that strawberry variety Festival showed the lowest value of TPC as 864.44 ± 44.16 mg GAE/ 100g compared to Chandler and Korona. Similarly, total flavonoids content showed the significant difference among strawberry varieties that were ranged from 144.84 ± 10.11 to 180.63 ± 12.72 mg/100g, whereas strawberry variety Korona reported lowest value of flavonoids and Chandler reported highest flavonoids content among tested strawberry varieties samples. Likewise, quantitative analysis of anthocyanins among strawberry varieties showed statistically significant difference ($p < 0.05$). Means for the parameter showed that strawberry variety Chandler have highest anthocyanin contents (2.74 ± 0.09) mg/100g) compared to Festival (1.98 ± 0.05 mg/100g) and Korona (1.50 ± 0.04 mg/100g) varieties, correspondingly. Literature also suggested that higher contents of phytochemicals in strawberry fruit are thought to be a sign of naturally occurring bioactive substances that are linked to promote human health upon its consumption.

***Antioxidant capacity and vitamin C level:*** The antioxidant phytochemicals in strawberry fruit include vitamin C, carotenoids, phenolic compounds (anthocyanins (ACN) that can influence the overall antioxidant capacity of strawberry fruit. The results (Fig. 1) showed antioxidant capacity of different tested strawberry varieties using DPPH method that showed statistically significant differences among varieties.

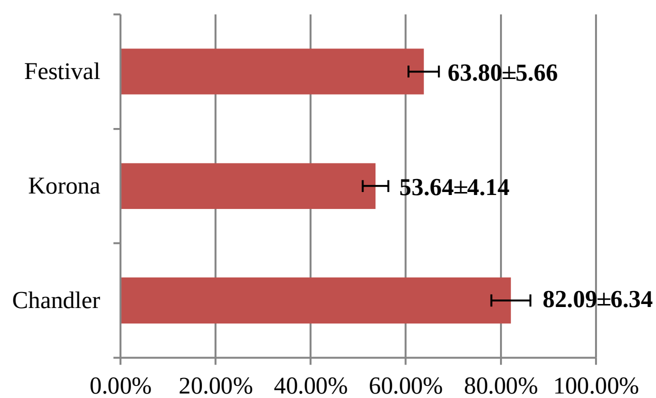


**Figure 1. Mean ± SD, Evaluation of Antioxidant Capacity of strawberry varieties.**

**Table 3. Phytochemicals characterization of different indigenous strawberry varieties**

| Parameters | Varieties | Mean ± Std. Deviation | P-Value |
|---|---|---|---|
| Total phenolic Contents (mg GAE/100 g) | Chandler | 926.17 ± 53.08[a] | 0.031 |
| | Korona | 871.55 ± 48.48[b] | |
| | Festival | 864.44 ± 44.16[b] | |
| Total flavonoids (mg/100g) | Chandler | 180.63 ± 12.72[a] | 0.024 |
| | Korona | 144.84 ± 10.11[c] | |
| | Festival | 163.62 ± 11.42[b] | |
| Anthocyanins (mg/100g) | Chandler | 2.74 ± 0.09[a] | 0.002 |
| | Korona | 1.50 ± 0.04[c] | |
| | Festival | 1.98 ± 0.05[b] | |

Data are expressed as “mean ± standard deviation”. Superscript letters ([a, b]) shows the statistically differences. Different letters indicate significant differences ($P < 0.05$, Tukey test) between strawberry varieties.

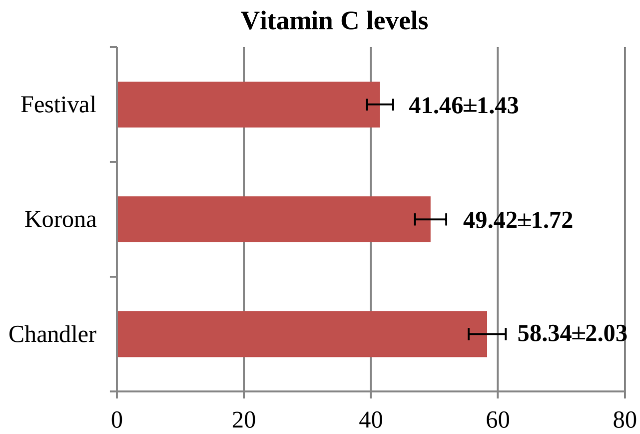


**Figure 2. Vitamin C analysis of different strawberry varieties expressed in mg/100g"**

The results showed that highest antioxidant capacity (Free radical scavenging activity %) was documented in strawberry variety Chandler and lowest in strawberry variety Korona. According to findings strawberry variety Korona showed lowest value (41.46 ±1.43 mg/100g) and highest values of vitamin C in Chandler as 58.34 ± 2.03 mg/100g) as indicated in (Fig. 2). Vitamin C enhances the immune system of living organism as it is natural killer of damaged cells and antimicrobial activity, lymphocyte proliferation and chemotaxis. Vitamin C also protects cells against reactive oxygen species produced during the respiratory burst and during inflammation by ensuring their redox integrity (Kawashima *et al.*, 2015).

***Minerals Analysis*:** The mineral contents of strawberry samples are shown in Table 4 which are expressed in mg/100g of dry sample of strawberry for different varieties. The findings about potassium quantification of strawberry varieties available in Pakistan showed significant results whereas, means for potassium level for Korona variety reported highest value as 254.68 ± 22.12 mg/100g and variety Chandler having the lowest value as 206.13 ± 20.11 mg/100g . Similarly, the iron contents analysis for different strawberry varieties showed significant results among varieties. Maximum iron value was documented in Chandler as 0.51 ± 0.03 mg/100 g whereas lowest iron level was reported in Korona as 0.43 ± 0.02 mg/100 g. Strawberry is a good source of magnesium and observed data also showed significant difference for magnesium level in strawberry varieties and The magnesium level ranged from 21.96 ± 1.76 to 16.48 ±1.01 mg/100g whereas Festival showed highest value for magnesium and Korona have lowest value. Strawberry is also good source of zinc and among tested varieties, the Festival had highest as 12.69 ±1.02 zinc content and Korona reported lowest values as 8.20 ±1.22 mg/100g.

**Table 4. Mineral contents of different indigenous strawberry varieties (Chandler, Festival and Korona).**

| Minerals | Varieties | Mean ± SD | P-Value |
|---|---|---|---|
| Potassium (mg/100g) | Chandler | 206.13 ± 13.11$^{c}$ | 0.027 |
| | Festival | 229.39 ± 14.09$^{b}$ | |
| | Korona | 254.68 ± 15.12$^{a}$ | |
| Iron (Fe) (mg/100g) | Chandler | 0.51 ± 0.03$^{a}$ | 0.003 |
| | Festival | 0.44± 0.03$^{b}$ | |
| | Korona | 0.43 ± 0.02$^{b}$ | |
| Magnesium (mg/100g) | Chandler | 19.19 ± 1.63$^{b}$ | 0.023 |
| | Festival | 21.96$^{a}$ ± 1.76 | |
| | Korona | 16.48 ±1.01$^{c}$ | |
| Zinc (mg/100g) | Chandler | 9.16 ± 0.56$^{b}$ | 0.024 |
| | Festival | 12.69 ±.61$^{a}$ | |
| | Korona | 8.20 ± 1.22$^{b}$ | |
| Calcium (mg/100g) | Chandler | 10.42± 0.67$^{c}$ | 0.024 |
| | Festival | 16.22 ± 0.83$^{a}$ | |
| | Korona | 13.88 ±0.62$^{b}$ | |
| Sodium (mg/100g) | Chandler | 0.69 ± 0.03$^{c}$ | 0.034 |
| | Festival | 1.02 ± 0.04$^{b}$ | |
| | Korona | 1.12 ± 0.02$^{a}$ | |
| Cupper (mg/100g) | Chandler | 0.13 ± 0.02$^{c}$ | 0.001 |
| | Festival | 0.19 ± 0.02$^{b}$ | |
| | Korona | 0.30 ± 0.03$^{a}$ | |
| Manganese (mg/100g) | Chandler | 0.25 ± 0.04$^{a}$ | 0.024 |
| | Festival | 0.17 ± 0.01$^{c}$ | |
| | Korona | 0.23 ± 0.01$^{ab}$ | |

Data are expressed as "mean ± standard deviation". Superscript letters ($^{a,b}$) shows the statistically differences. Different letters indicate significant differences ($P < 0.05$, Tukey test) between strawberry varieties.

***Shelf-life extension of strawberry through modified atmospheric packaging technology with different gases compositions***

***Physiochemical Properties:*** The pH of strawberry samples subjected to modified atmosphere packaging was increased slightly during storage, corresponding to a decrease in titratable acidity (TA) in all tested treatments. The MAP strawberries were not statistically different from each other up to 09 days after storage whereas, pH values of strawberries after 9 days of storage were significantly higher than 06 and 09 days (Table 5). The modified atmosphere packaging (MAP) had non-significant differences for total soluble solids determined on strawberries. According to the results, in control group $MAP_0$ (control group having 0.05% $CO_2$, 20.95% $O_2$ and 79% $N_2$), the total soluble solids were 7.64± 0.67 and 8.42±0.51% at 0 (baseline) and 12$^{th}$ day of storage respectively whereas, the total soluble solids for the strawberry samples subjected to $MAP_4$(15% $CO_2$, 3% $O_2$ and 82% $N_2$) treatment were 7.69± 0.28 and 7.35 ± 0.78%, respectively (Table 5). Similarly, the results for acidity of strawberry samples reported higher acidity values start (first day stored strawberries) of storage that were decreased with progression of storage. It has been observed that storage of the strawberry samples at 4°C can affect the acidity which is decreased in both control and treatments groups (Table 5).

***Phytochemicals properties:*** The results (Table 6) showed the effect of modified atmosphere packaging technology on phytochemicals attributes *i.e.*, total phenolic content, total flavonoid content, anthocyanin content and antioxidant activity of strawberry samples at different storage intervals that were affected significantly. The results showed that TPC, TFC, AC and antioxidant decreased with increase of storage period. Means for total phenolic compounds expressed in mg

**Table 5. Effect of modified atmospheric packaging with different gases compositions on Physiochemical Properties of strawberries**

| Parameters | Storage day | $MAP_0$ | $MAP_1$ | $MAP_2$ | $MAP_3$ | $MAP_4$ |
|---|---|---|---|---|---|---|
| pH | 0 | 3.22±0.51$^{c}$ | 3.28±0.46$^{b}$ | 3.21±0.46$^{b}$ | 3.22±0.37$^{a}$ | 3.22±0.46$^{a}$ |
| | 3$^{rd}$ | 3.29±0.33$^{cA}$ | 3.27±0.31$^{bA}$ | 3.22±0.39$^{bB}$ | 3.23±0.51$^{aB}$ | 3.21±0.27$^{aB}$ |
| | 6$^{th}$ | 3.36±0.42$^{bA}$ | 3.32±0.48$^{bB}$ | 3.24±0.74$^{bC}$ | 3.23±0.51$^{aC}$ | 3.24±0.42$^{aC}$ |
| | 9$^{th}$ | 3.49±0.04$^{aA}$ | 3.41±0.38$^{aA}$ | 3.24±0.03$^{bB}$ | 3.25±0.51$^{aB}$ | 3.24±0.35$^{B}$ |
| | 12$^{th}$ | 3.58±0.47$^{aA}$ | 3.43±0.48$^{aA}$ | 3.38±0.26$^{aB}$ | 3.24±0.12$^{aC}$ | 3.25±0.42$^{aC}$ |
| Total Soluble Solids (%) | 0 | 7.64±0.67$^{c}$ | 7.67±0.73$^{b}$ | 7.71±0.63$^{b}$ | 7.63±0.41$^{a}$ | 7.69±0.28$^{a}$ |
| | 3$^{rd}$ | 7.43±0.77$^{cA}$ | 7.62±0.82$^{A}$ | 7.33±0.52$^{cA}$ | 7.21±0.45$^{bA}$ | 7.61±0.18$^{aA}$ |
| | 6$^{th}$ | 8.06±0.28$^{bA}$ | 7.24±0.93$^{bB}$ | 7.21±0.18$^{cB}$ | 7.33±0.29$^{bB}$ | 7.52±0.16$^{aB}$ |
| | 9$^{th}$ | 8.17±0.36$^{bA}$ | 8.07±0.17$^{aA}$ | 7.47±0.54$^{cB}$ | 7.42±0.14$^{bB}$ | 7.46±0.23$^{bB}$ |
| | 12$^{th}$ | 8.42±0.51$^{aA}$ | 8.05±0.11$^{aA}$ | 7.95±0.02$^{aA}$ | 7.62±0.71$^{aB}$ | 7.35±0.78$^{bB}$ |
| Titratable Acidity (g citric acid/100 g) | 0 | 0.70±0.16$^{a}$ | 0.78±0.62$^{a}$ | 0.76±0.29$^{a}$ | 0.73±0.21$^{a}$ | 0.72±0.79$^{a}$ |
| | 3$^{rd}$ | 0.68±0.13$^{aA}$ | 0.69±0.21$^{bA}$ | 0.71±0.26$^{aA}$ | 0.72±0.64$^{aA}$ | 0.72±0.56$^{aA}$ |
| | 6$^{th}$ | 0.65±0.92$^{bB}$ | 0.68±0.53$^{bB}$ | 0.67±0.19$^{bB}$ | 0.68±0.76$^{bB}$ | 0.71±0.42$^{aA}$ |
| | 9$^{th}$ | 0.62±0.21$^{bB}$ | 0.65±0.87$^{bB}$ | 0.66±0.72$^{bB}$ | 0.64±0.26$^{cB}$ | 0.69±0.03$^{aA}$ |
| | 12$^{th}$ | 0.60±0.23$^{bB}$ | 0.61±0.42$^{cB}$ | 0.63±0.41$^{bB}$ | 0.62±0.71$^{cB}$ | 0.68±0.63$^{aA}$ |

Data are expressed as "mean ± standard deviation". Superscript letters shows the statistically differences. Means that have no significant differences receive same letters. ($P < 0.05$, Tukey test), whereas TSS= Total soluble solids, TA= Titratable acidity

**Table 6. Effect of modified atmospheric packaging with different gases compositions on phytochemicals properties of strawberry**

| Parameters | Day | $MAP_0$ | $MAP_1$ | $MAP_2$ | $MAP_3$ | $MAP_4$ |
|---|---|---|---|---|---|---|
| Total phenolic content (mg GAE/100 g) | 0 | 926.17±53.08$^{a}$ | 927.22±64.34$^{a}$ | 932.15±63.11$^{a}$ | 928.33±66.32$^{a}$ | 929.36±73.18$^{a}$ |
| | 3$^{rd}$ | 703.28± 49.37$^{bD}$ | 764.26±16.34$^{bC}$ | 804.34±42.34$^{bC}$ | 873.53±41.04$^{bB}$ | 922.24±44.03$^{aA}$ |
| | 6$^{th}$ | 643.19± 45.21$^{cD}$ | 727.34±34.26$^{cC}$ | 753.65±35.76$^{cC}$ | 846.36±41.41$^{bB}$ | 895.44±40.03$^{bA}$ |
| | 9$^{th}$ | 571.56± 39.43$^{dC}$ | 698.71±31.09$^{dB}$ | 732.48±31.08$^{cB}$ | 793.27±38.02$^{dA}$ | 871.25±41.02$^{bA}$ |
| | 12$^{th}$ | 417.39±36.84$^{eD}$ | 582.54±27.08$^{eC}$ | 689.81±34.05$^{dB}$ | 722.73±41.03$^{dB}$ | 852.43±46.23$^{bA}$ |
| Total flavonoid content (mg/100g) | 0 | 180.63±12.72$^{a}$ | 174.63±14.43$^{a}$ | 184.43±11.49$^{a}$ | 179.21±9.23$^{a}$ | 187.68±10.29$^{a}$ |
| | 3$^{rd}$ | 152.74±8.02$^{bC}$ | 157.32±7.26$^{bC}$ | 162.46±9.03$^{bB}$ | 164.71±8.14$^{abB}$ | 182.64±9.22$^{aA}$ |
| | 6$^{th}$ | 116.67±7.34$^{cD}$ | 124.67±6.05$^{cD}$ | 158.76±6.01$^{bB}$ | 163.54±7.14$^{abB}$ | 179.54±7.33$^{aA}$ |
| | 9$^{th}$ | 96.76 ± 6.09$^{dC}$ | 117.52± 6.36$^{dC}$ | 124.39 ±7.11$^{cC}$ | 148.02±6.29$^{bcB}$ | 165.83±6.37$^{bA}$ |
| | 12$^{th}$ | 84.64±5.02$^{eD}$ | 94.82±6.21$^{eC}$ | 119.43±7.19$^{cC}$ | 137.39±7.32$^{dB}$ | 158.93±7.39$^{bcA}$ |
| DPPH free radical scavenging activity %) | 0 | 82.09±6.38$^{a}$ | 81.03±2.17$^{a}$ | 83.28±4.88$^{a}$ | 82.26±4.17$^{a}$ | 83.66±5.51$^{a}$ |
| | 3$^{rd}$ | 72.97±4.31$^{bB}$ | 71.04±4.22$^{bB}$ | 74.18±3.10$^{bB}$ | 78.24±4.29$^{aA}$ | 81.08±2.91$^{aA}$ |
| | 6$^{th}$ | 63.49±4.07$^{cC}$ | 64.27±3.91$^{cC}$ | 66.46±2.13$^{cB}$ | 69.14±4.05$^{bB}$ | 77.61±2.02$^{bA}$ |
| | 9$^{th}$ | 51.23±3.01$^{dC}$ | 53.28±4.03$^{dC}$ | 61.86±3.06$^{cB}$ | 65.43±3.31$^{bB}$ | 72.28±3.04$^{bA}$ |
| | 12$^{th}$ | 42.76±3.21$^{eD}$ | 47.15±0.04$^{eC}$ | 48.43±2.01$^{dC}$ | 54.09±2.02$^{cB}$ | 68.54±4.01$^{cA}$ |
| Anthocyanins (mg/100g) | 0 | 20.74±2.09$^{a}$ | 21.65±2.09$^{a}$ | 20.31±2.47$^{a}$ | 21.61±2.33$^{a}$ | 20.23±2.08$^{a}$ |
| | 3$^{rd}$ | 16.82±1.03$^{bB}$ | 18.32±1.01$^{aA}$ | 19.29±1.08$^{aA}$ | 19.43±0.86$^{aA}$ | 19.25±1.45$^{aA}$ |
| | 6$^{th}$ | 15.74±0.42$^{bB}$ | 16.54±2.13$^{abB}$ | 17.72±1.52$^{abA}$ | 18.75±1.84$^{aA}$ | 18.54±0.83$^{aA}$ |
| | 9$^{th}$ | 11.74±1.42$^{cC}$ | 12.74±1.04$^{cC}$ | 13.63±1.01$^{bC}$ | 16.32±1.65$^{bB}$ | 18.43±0.76$^{aA}$ |
| | 12$^{th}$ | 10.23±1.82$^{cC}$ | 10.65±0.42$^{cC}$ | 11.54±0.53$^{cC}$ | 14.76±0.64$^{bB}$ | 16.84±0.92$^{bA}$ |

Data are expressed as "mean ± standard deviation". Superscript letters ($^{a,b}$) shows the statistically differences. Means that have no significant differences receive same letters ($p < 0.05$, Tukey test), whereas TPC= total phenolic compounds, TFC = total flavonoids compounds; $MAP_0$ (control group having 0.05% $CO_2$, 20.95% $O_2$ and 79% $N_2$), $MAP_1$ (3% $CO_2$, 15% $O_2$ and 82% $N_2$), $MAP_2$ (5% $CO_2$, 10% $O_2$ and 85% $N_2$), $MAP_3$ (10% $CO_2$, 5% $O_2$ and 85% $N_2$), $MAP_4$ (15% $CO_2$, 3% $O_2$ and 82% $N_2$).

GAE/ 100g decreased in control group ($MAP_0$) reported a decrease in values from 926.17±53.08 (0 day) to 417.39±36.84 mg GAE/ 100g after 12th day of storage. Similarly, TPC values differed significantly among different MAP treatment compare to control group and the means for TPC in $MAP_4$ treatments showed that TPC values were slightly decreased from 929.36±73.18 to 852.43±46.23 mg GAE/ 100g at 0 and 12th day of storage. Likewise, the total flavonoids content expressed in mg/100g showed significant difference among control and treatment groups. Means for total flavonoids showed that the values in control ($MAP_0$) group were decrease from 180.63±12.72 to 84.64±5.02 and in $MAP_4$ values decrease from 187.68±10.29 to 158.93±7.39 mg/100g at 12th day of storage compared to start of trial (O day). The quantitative analysis of anthocyanins and antioxidants activity among different treatments of strawberry samples subjected to modified atmosphere packaging technology showed significant difference ($p < 0.05$). Means for antioxidants activity reported 48% decrease in control group and compared to 17% decreased in $MAP_4$ group indicating the potential of packaging technology for preserving the components that can help to maintain the antioxidant potential of the strawberry samples . similarly, anthocyanins content reduction was documented with progression of storage and among treatments maximum reduction was observed in control ($MAP_0$) followed by $MAP_1$ $MAP_2$, and $MAP_3$ at baseline 20.74±2.09, 21.65±2.09, 20.31±2.47, 21.61±2.33 to termination of storage, 10.23±1.82, 10.65±0.42, 11.54±0.53, 14.76±0.64 respectively (Table 6).

***Microbial and Sensory evaluation of strawberry*:** Microbial load, physical appearance and consumer acceptance are integral parts in shelf life studies of the processed food for accessing the quality and safety. For the microbial study of the strawberry samples subjected to modified atmosphere packaging technology, total plate count, yeast and molds findings are presented in (Table 7) and sensory evaluation findings in (Fig. 3). The results elaborated that quality changes of strawberry during storage occurred both in microbiological and physiological processes. At the start of trial (day 0), the total plate count value was 2.7 x $10^2$ cfu/g that was increased to $6.18\times10^8$ at the completion of storage whereas no yeast and mold were found in the start of the trial however, the values were reported in $MAP_0$, $MAP_1$ and $MAP_2$ at the completion of storage. Overall, the higher microbial load was reported in control group than MAP treated strawberry samples. In $MAP_0$ (control group) TPC values increased from $2.82\times10^2$ to $6.18\times10^8$, while in $MAP_4$ the TPC values decreased from $2.72\times10^2$ to $1.91\times10^2$ cfu/g. Yeast and Mold grown in control group were $2.41\times10^9$ and $2.52\times10^8$ at 12th day of storage, respectively while in MAP treated groups no yeast and mold were reported up to 9th day and slight growth was observed at 9th day for $MAP_0$, $MAP_1$ and $MAP_2$. For sensory evaluation, trained panelist arranged and accessed the taste, texture, appearance, color, aroma and overall acceptance through 9 hedonic scales of strawberries subjected to modified atmospheric packing technology. The results showed that MAP treated strawberries ($MAP_4$) have better consumer acceptability than control groups whereas storage has a detrimental effect on the strawberries. Overall, the strawberries subjected to MAP treatments were acceptable up to 9 days and control group ($MAP_0$) was acceptable only for 3 days.

***Color differences during storage days*:** Color change of strawberries during storage is an important quality parameter to determine their shelf life as well as consumer acceptability. Samples were analyzed during storage by instrumental color measurements. Results indicated that chromatic parameter

**Table 7. Microbiological evaluation of strawberry samples subjected to modified atmospheric packaging**

| Microorganisms | Storage days | MAP0 | MAP1 | MAP2 | MAP3 | MAP4 |
|---|---|---|---|---|---|---|
| Total Plate count | 0 | $2.82\times10^{2,\ d}$ | $2.85\times10^{2,\ d}$ | $2.75\times10^{2,\ cd}$ | $2.81\times10^{2,\ a}$ | $2.72\times10^{2,\ a}$ |
| (log cfu/g) | 3rd | $2.89\times10^{2,\ dA}$ | $2.73\times10^{2,\ dA}$ | $2.82\times10^{2,\ cdA}$ | $2.82\times10^{2,\ aA}$ | $2.71\times10^{2,\ aA}$ |
| | 6th | $3.31\times10^{3,\ cA}$ | $2.91\times10^{3,\ cA}$ | $3.6\times10^{2,\ cB}$ | $2.42\times10^{2,\ bB}$ | $2.25\times10^{2,\ abB}$ |
| | 9th | $4.12\times10^{5,\ bA}$ | $2.38\times10^{4,\ bB}$ | $2.31\times10^{3,\ bC}$ | $2.31\times10^{2,\ bD}$ | $2.13\times10^{2,\ abD}$ |
| | 12th | $6.18\times10^{8,\ aA}$ | $2.7\times10^{7,\ aB}$ | $2.7\times10^{4,\ aC}$ | $2.05\times10^{2,\ bcD}$ | $1.91\times10^{2,\ cE}$ |
| Yeast | 0 | ND | ND | ND | ND | ND |
| (log cfu/g) | 3rd | ND | ND | ND | ND | ND |
| | 6th | ND | ND | ND | ND | ND |
| | 9th | $1.62\times10^{5,\ bA}$ | $1.93\times10^{4,\ bB}$ | $1.12\times10^{4,\ bB}$ | ND | ND |
| | 12th | $2.41\times10^{9,\ aA}$ | $2.24\times10^{7,aB}$ | $2.11\times10^{7,\ aC}$ | $1.42\times10^{6,\ D}$ | $1.03\times10^{6,\ D}$ |
| Molds count | 0 | ND | ND | ND | ND | ND |
| (log cfu/g) | 3rd | ND | ND | ND | ND | ND |
| | 6th | ND | ND | ND | ND | ND |
| | 9th | $2.46\times10^{6,\ bA}$ | $2.05\times10^{4,\ bB}$ | $2.4\times10^{3,\ bC}$ | ND | ND |
| | 12th | $2.52\times10^{8,\ aA}$ | $2.21\times10^{6,\ aB}$ | $2.62\times10^{5,\ aC}$ | $1.45\times10^{5,\ D}$ | $1.32\times10^{5,\ D}$ |

Data are expressed as "mean ± standard deviation". Superscript letters ($^{a,b}$) shows the statistically differences. Means that have no significant differences receive same letters ($P < 0.05$, Tukey test), whereas ND=Not detected any growth

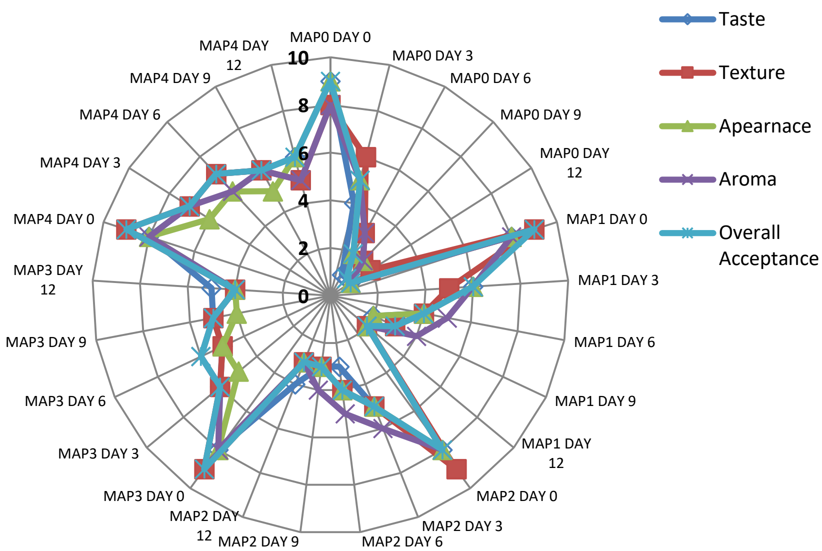


**Figure 3. Radar diagram displays the sensory evaluation parameters including taste, texture, appearance, aroma, and overall acceptance of strawberries on a hedonic scale (0-9) during 12 days of storage in all MAP groups**

**Table 8. Effect of modified atmospheric packaging with different gases compositions on color differences (ΔEL*a*b*) during storage days.**

| | | Days | | | | |
|---|---|---|---|---|---|---|
| | | 0 | 3rd | 6th | 9th | 12th |
| $MAP_4$ | L* | 55.22±3.21 | 53.47±2.94 | 52.08±2.38 | 51.18±2.36 | 51.43±1.92 |
| | a* | 37.09±2.13 | 36.98±2.08 | 36.03±2.11 | 34.28±2.19 | 32.34±1.13 |
| | b* | 21.45±2.17 | 21.37±1.52 | 20.29±1.57 | 19.33±1.65 | 18.24±1.06 |
| | ΔE* | ------ | $1.80±0.03^{cC}$ | $3.51±0.12^{bC}$ | $5.66±0.42^{aD}$ | $6.87±0.24^{aC}$ |
| $MAP_3$ | L* | 56.28±2.38 | 53.34±2.08 | 51.12±2.03 | 49.09±2.68 | 46.41±1.45 |
| | a* | 37.26±2.13 | 36.21±2.13 | 35.36±1.59 | 32.76±2.13 | 29.82±2.13 |
| | b* | 21.19±2.17 | 21.07±1.39 | 20.24±1.53 | 18.39±2.16 | 17.29±1.65 |
| | ΔE* | ------ | $3.09±0.07^{dB}$ | $5.58±0.26^{cB}$ | $8.93±0.45^{bC}$ | $12.96±0.61^{aB}$ |
| $MAP_2$ | L* | 55.43±2.38 | 52.41±1.45 | 50.49±46 | 47.34±2.38 | 40.42±2.38 |
| | a* | 37.43±2.13 | 35.61±2.18 | 33.67±2.13 | 30.29±2.08 | 24.56±2.06 |
| | b* | 20.45±2.23 | 20.09±1.87 | 19.82±1.84 | 19.76±2.17 | 16.35±1.14 |
| | ΔE* | ------ | $3.54±0.17^{dB}$ | $6.24±0.34^{cB}$ | $10.81±1.03^{bC}$ | $20.19±1.07^{aA}$ |
| $MAP_1$ | L* | 55.26±3.24 | 51.27±2.83 | 48.41±12.32 | 41.54±2.38 | ND |
| | a* | 36.25±1.03 | 35.22±2.87 | 34.85±2.13 | 29.23±2.13 | ND |
| | b* | 21.23±1.84 | 20.76±2.17 | 20.12±2.17 | 18.31±2.17 | ND |
| | ΔE* | ------ | $4.15±0.08^{cA}$ | $7.07±0.24^{bB}$ | $15.68±0.73^{aB}$ | ND |
| $MAP_0$ | L* | 56.42±2.38 | 52.52±2.16 | 42.31±1.52 | 31.34±1.21 | ND |
| | a* | 37.81±2.13 | 35.72±2.08 | 31.43±1.64 | 24.39±1.02 | ND |
| | b* | 21.85±2.17 | 20.85±1.05 | 19.07±2.17 | 16.04±0.89 | ND |
| | ΔE | ------ | $4.54±0.28^{cA}$ | $15.72±0.82^{bA}$ | $29.11±1.05^{aA}$ | ND |

Data are expressed as "mean ± standard deviation". Superscript letters ($^{a,b}$) on ΔE* values shows the statistically differences ($P < 0.05$, Tukey test), whereas ND=Not detected due to spoilage of strawberries.

(ΔE* values) changed with increasing storage time as value of ΔE increased with storage days as significant effect was documented by the different treatments of modified atmospheric packaging technology. The value of ΔE on 3rd day in $MAP_0$ (Control group) is 4.53 ± 0.28 is higher than $MAP_4$ as 0.77 ± 0.03 and theses values increased with storage from at 9th and 12th day of storage. ΔE values not detected in control while MAP treated group $MAP_4$ the values were as 3.01 ± 0.42 and 3.54 ± 0.24 at 9th and 12th day of storage.

## DISCUSSION

The nutritional analysis of different raw and processed foods helps to find out how much micronutrients, macronutrients, and phytochemicals are present in food that ultimately impacts consumer health. The present study was conducted to check nutritional characterization *i.e.*, proximate analysis, physicochemical properties, phytochemicals and antioxidant capacity of selected strawberry varieties produced in Pakistan as well as shelf life extension through modified atmospheric packaging technology by optimizing the different gases ($O_2$, $CO_2$ and $N_2$). The data showed significant and non-significant differences for different parameters. In proximate composition, moisture content, crude protein and crude fat findings are lien with the results reported by (Hossain *et al.*, 2016) who analyzed 4 different genotypes for strawberry. They further reported that ash content is consistent with the reports of (Khan *et al.*, 2010) and (Ganhão *et al.*, 2019). Overall, proximate composition of strawberry cultivars in Pakistan are similar reported worldwide and the information is useful for the development of healthy and nutritious foods using strawberry as raw and in processed form (Shahzad *et al.*, 2020). In physiochemical analysis titratable acidity, color and texture were measured. Titratable acidity results are lien with (Kaczmarska *et al.*, 2016) who documented titratable acidity as 0.84g/100 g for the chandler variety. Literature also suggest that fruity maturity, postharvest storage conditions as well as and environmental factors may influence titratable acidity of the strawberry fruits (Vallarino & Osorio 2019). The total soluble solid which is also known as Brix (°Bx), is a crucial factor indicating an aqueous solution's sugar content used for checking the maturity as well as ripening. In contrast, organic acids and sugar make majority of total soluble solids in strawberry fruit however, sugar content can determines sweetness (taste) of the product. Moreover, factors such as genetics can also effect total soluble solids and results for different varieties also showed this trend and the similar findings were also reported by (Zhang *et al.*, 2016).. Our findings are also similar to ones reported by Kaczmarska *et al.* (2016) and the attractive red color, sweet and fruity taste along with acceptable texture is major parameters that affects consumer acceptance for the fruits and fruits based processed products. Additionally, the observed surface color values were consistent with color values as documented by (Crecente-Campo *et al.* (2012) who reported superficial surface color values as 22.1 ± 8.9 for a* value, 27.8 ± 4.1 for b* value and for interior color value as 29.7 ± 7.8 for a* value flesh and 23.5 ± 7.0 for b* flesh of strawberry varieties. Although strawberry color varies is more prone towards its maturity and the post-harvest period (becoming darker and less bright) thus can affect quality attributes of strawberry like shelf life, quality as well as finally consumer acceptability (Parra-Palma, Morales-Quintana *et al.*, 2020). The firmness of fruit is an important quality parameter that needs to be measured to detect shelf life and consumer acceptance. After postharvest, if the strawberry has a softer texture that can lead to decreased consumer acceptance resulting lower quality and finally consumer rejection. Strawberry is a perishable fruit crop that loses moisture quickly resulting decreased moisture content if not stored properly. Our results regarding firmness of different cultivars of strawberry are in line with findings of (Lewers *et al.*, 2020) who observed 2.7 for surface and 2.6 for interior zone of "Chandler" variety produced in Queenstown, Maryland, USA. Furthermore due to their phenolic content, strawberries belong to a group of phytochemical-rich fruits that upon consumption can protect humans against several non-consumable diseases. A polyphenol compound that has been extensively studied in strawberry fruits is anthocyanin, which is responsible for strawberry red color and serves as primary criterion for assessing quality of strawberries. The phenolic compounds are potential source of antioxidants and can plays a protective role in human health. Findings of our study reported that strawberry variety Chandler has highest level of phenolic compounds contents and these results are consistent with findings of (Dzhanfezova *et al.*, 2020) who also reported similar compounds for strawberry variety korona grown in Denmark. According to (Guerrero-Chavez *et al.*, 2015) different fruit varieties have impact on phenolic contents quantification and berries including strawberry contain flavonoids. Whereas our observed values of strawberry samples are consistent with values reported by (Cervantes *et al.*, 2020) as 54.96±4.97 mg CAE/100 g. Anthocyanin is a naturally occurring glycosylated pigment that belongs to phenolic group responsible for health benefits including antidiabetic, anticancer, anti-inflammatory, antimicrobial effect and anti-obesity effects. Our results are in agreement with Zahid *et al.*, (2021) who recorded the anthocyanins as (3.47 mg/100 g) of same variety and Lalk *et al.*, (2020) in which they reported the value ranged from 19.6 to 20.2 mg/100 g of Pg-3-glc equivalent whereas, literature also stated that anthocyanin content in strawberry is mainly affected by location, climate and as well as weather conditions.

Strawberry fruit is an excellent source of natural antioxidants and enzymes that can help to protect against free radicals generated in living organism through redox reactions under stressful conditions and as a byproduct of normal metabolism. Increasing levels of free radicals can cause oxidative stress, which can result in a range of biochemical and physiological reactions that can results as impairment of metabolism ultimately leading towards cell death. Consuming fruits and vegetables especially berries like strawberry, has been linked to lower incidence of diseases and reduced mortality rate of cancer, heart disease and a variety of other diseases. Our findings of antioxidants assays are in consistence with Wang *et al.*, (2019) in which they reported the values ranged from 37.50 ± 0.15 to 43.50 ± 0.24 for DPPH (% inhibition). Furthermore, minerals are necessary for the body's wellness and used for variety of purposes, including bone health, muscular, heart as well as brain function. They are also required for production of enzymes and hormones; maintain normal fluid balance inside our cells and to make hemoglobin.

In our study different minerals were analyzed *i.e.*, potassium, iron, zinc, calcium, cupper and manganese. The published literature suggest that mineral levels of strawberry cultivars are influenced by factors *i.e.*, environmental conditions, growth temperature, genetics, cultivation soil as well as harvesting time (Prasad *et al.*, 2022). Our findings regarding minerals levels for are significantly differing among tested varieties. The potassium mineral values are in line with findings of (Hossain *et al.*, 2016b) in which they compared potassium level of 4 varieties of strawberries. Similarly iron values of current study are consistent with (Prichko *et al.*, 2021) in which they reported iron level ranged from 0.85 to 1.34 mg/100 g. Similarly, magnesium results are in lieu with (González-Domínguez *et al.*, 2020) in which 5 varieties of strawberries were analyzed and reported magnesium level ranged from 142.5 ± 20.1 to 226.7 ± 26.9. The zinc values are in consistent with (Gondek *et al.*, 2020) in which they analyzed six strawberry varieties that were ranged from 9.5±0.9 to 13.4±2.0 (mg/ kg of dry matter ). Other minerals sodium, copper and manganese were also found in minute quantity. The levels of vitamin C are in consistent with (Görgüç *et al.*, 2019) in which they reported vitamin C ranged from 50.46 to 53.40 mg/100g and documented literature also discussed different levels of vitamin C are attributed to genotype, fertilizers type (organic vs inorganic), harvest time, maturity stage as well as temperature of environment used (Kilic *et al.*, 2021)

The effect of modified atmosphere packaging on microbiological and chemical parameters of strawberries has been evaluated in this work. For strawberry storage, modified atmosphere packaging treatments involving low level of oxygen ($O_2$) concentration has shown significant impact for increasing shelf life of strawberry. The treatment ($MAP_4$) showed more pronounced preservative effect on quality and shelf life parameters compared to control ($MAP_0$). Additionally, modified atmosphere packaging improved the visual quality of strawberries after cold storage whereas, strawberries sensory evaluation found that MAP storage negatively influenced their sensory attributes (taste, texture, aroma, and overall acceptance) despite the fact that MAP storage detected better taste, appearance, and freshness score for tested strawberries. The findings of our study are supported by one of the researchers group who conducted study to determine the impact of modified atmosphere packaging (MAP) with low oxygen & high carbon dioxide along with combination of ozone and edible film coatings on strawberry preservation. Based on their results, 2.5% oxygen and 15% carbon dioxide were optimum gas composition for strawberry MAP resulting increased shelf-life by 4 to 6 days compared to open-air storage. Additionally, strawberries reported to have extended shelf-life when treated with 4.3 mg m–3 ozone water dip and edible coating before packaging (Xiao *et al.*, 2004). Similarly, another study investigated MAP affects quality attributes of fully ripe strawberries using 'Senga Sengana' from organic production and 'Arosa' from conventional production. First fruits were pre-cooled to 2°C and placed in MAP (transmission rates: O 530 cc/m2/day and CO2 4200 cc/m2/day at 23°C); second MAP without pre-cooling; third pre-cooled and cold stored fruit without MAP and all fruits kept at 1°C for 5 days. Results indicated that MAP treatment did not affect firmness of 'Senga Sengana' while effect of MAP was primarily on reduction in bruised fruits along with non-significant differences for cultivar's total soluble solids and titratable acidity (Magazin *et al.*, 2013). Likewise , Dong et al. (2014) carried out study to access impact of strawberry packaging employing modified environment using 20 m PE film to enhance shelf life of strawberries in refrigerators. They determined the best gas condition for strawberries by taking into account respiration rate, firmness, decay grade, vitamin C content, total soluble solids, titratable acidity, sensory assessment and other quality indicators. The results showed that in 60% O2 + 1.5% CO2 gas conditions it was possible to preserve a low rot grade while still maintaining firmness, vitamin C content, total soluble solids, titratable acidity content, and sensory assessment that was not fast dropping. The effect of preservation was more noticeable at 5–1°C as well as modified environment packing could increase the strawberry's shelf life to 22 days. Furthermore, strawberries spoil quickly, and the flavor deteriorates with storage time. However, nothing is known regarding impact of storage strategies on the quality alterations. In this regard, Abu-Zahra (2017) conducted a study involving two storage methods like cold storage (CS) and Modified Atmosphere Packaging (MAP) to evaluate how they preserve quality. Findings showed that the storage method has an influence on maintaining its quality, and it proved that MAP is the appropriate storage method since it maintained strawberry quality, extended the storage term, and reduced weight loss.

***Conclusion*:** This study evaluated nutritional characterization, phytochemical composition, antioxidant capacity of three major strawberry varieties (Chandler, Korona and Festival) available in Pakistan along with shelf life extension through modified atmospheric packaging (MAP) technology. Results showed that all varieties have natural sources of health promoting properties due to phytochemical presence, antioxidant capacity and nutritional values whereas statistically significant differences ($p< 0.05$) for proximate composition were reported expect total ash and crude fat content. Also, strawberry variety Chandler has highest antioxidant effect based on total phenolic and anthocyanin contents and is superior to Korona and Festival considering phytochemicals and nutritional parameters. Additionally, modified atmosphere packaging technology under cold storage preserved strawberries shelf life by 9 days as indicted by microbial, quality and sensory evaluation findings. Overall, results provide valuable information on nutritional composition, phytochemical characteristics, and quality aspects of different strawberry varieties produced in Pakistan along with shelf life extension by packaging technology which are helpful for food processors in value addition, and modified atmosphere packaging technology for the reduction of post-harvest losses for strawberry supply chain actors.

***Conflict of interest*:** The Authors declare that there is no conflict of interest

***Authors Contribution*:** IYM executed the field research, laboratory analyses and written original draft, MS and AUK conceived the idea, supervised the research, reviewed and edited manuscript and IR Supervised, data analyzed, and reviewed the original draft

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