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# Quality changes and postharvest-losses of strawberry fruit packed in nano embedded biodegradable polyethylene films

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**Strawberries are highly perishable fruits with a short shelf life. This study aimed to investigate the impact of various low-density polyethylene (LDPE) packaging films on the quality changes and postharvest losses of strawberries. Four types of Low-density polyethylene (LDPE) films were used for packaging: conventional LDPE (CLDPE), biodegradable LDPE (BLDPE), biodegradable LDPE/nano-clay (BLDPE/NC), and biodegradable LDPE/nano-mineral (BLDPE/NM). The composition of the LDPE films influenced their oxygen transmission rate and water vapor transmission rate, resulting in improved barrier properties. Compared to CLDPE film, strawberries packed in BLDPE, BLDPE/NC, and BLDPE/NM films exhibited reduced color changes, lower decay indexes, and decreased microbial populations. Among the films, BLDPE and BLDPE/NC films showed the lowest weight loss during storage, while BLDPE/NM film had a slightly higher weight loss. The titratable acidity of the fruit showed similar trends among the different packaging films during storage. However, in contrast to the CLDPE, packaging with other films maintained firmness and had higher levels of soluble solids, vitamin C, total phenolics, total flavonoids, total anthocyanin, total antioxidant activity, and superoxide dismutase activity compared to those packed in CLDPE film. These findings suggest that the use of BLDPE and BLDPE/NC films, can effectively preserve the quality of strawberries by minimizing weight loss, maintaining firmness, and enhancing their nutritional and antioxidant properties during storage.**

**Keywords** Fruit decay, Nano-clay, Strawberry packaging, Shelf-life

Strawberries are highly valued for their rich content of anthocyanins, flavonoids, phenolic acids, vitamins, and minerals. Despite their nutritional benefits, strawberries have a short shelf-life and are highly perishable. Factors such as high respiration rates and microbial decay contribute to the rapid deterioration of strawberries[1]. Several methods, including heat treatment[2], ultraviolet (UV) irradiation[3], essential oil application[3], and coatings[4], have been explored to extend the shelf-life and maintain the quality of strawberries. However, packaging has emerged as a promising postharvest technology for preserving the quality and reducing postharvest losses of strawberries[5].

Packaging films play a crucial role in controlling water loss, microbial contamination, and the exchange of respiratory gases ($O_2$ and $CO_2$) during transportation and storage of fruit[6,7]. The levels of $O_2$ and $CO_2$within packages are critical factors that affect shelf-life and may trigger the deterioration of packaged fresh fruit[8]. Changes in the composition of $O_2$ and $CO_2$ significantly influence the respiration rate of the fruit inside the package[9]. The property of permeability of polymer films to gases can change the composition of gases inside the package[10]. Proper selection of packaging materials and technologies is vital for prolonging the postharvest life of horticultural crops. Low-density polyethylene (LDPE) films, a type of polymer film, have gained popularity in fresh produce packaging due to their affordability, reasonable physical properties (e.g., tensile and tear strength), and accessibility[11,12]. However, LDPE films have limitations in terms of gas permeability ($O_2$, $CO_2$, and water vapor), lack of antimicrobial activity[13], and environmental pollution caused by their slow degradation[10].

In recent years, modifying the structure of polymers has been explored to enhance their properties[6,14,15]. Biopolymers derived from compostable or biodegradable materials have gained significant attention in the packaging industry[16]. Biopolymer packaging can act as a barrier to solutes and gases, complementing other

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packaging methods and effectively preserving the quality and extending the postharvest life of crops[10]. Additionally, biopolymer films provide a suitable platform for incorporating various additives such as antimicrobial agents, antioxidants, and nutrients[17]. Biopolymers are produced by adding natural or synthetic compounds to synthetic polymers[18,19]. In addition to their role in the biodegradability of polymers, these compounds also improve and modify their properties, such as antimicrobial properties and gas permeability[13,16]. Poly (lactic acid) (PLA) is a synthetic biodegradable polymer produced from renewable plant resources and is widely used in the food packaging industry; however, its moderate water vapor barrier properties and poor $O_2$barrier properties are the most important limiting factors for this type of polymer[19].

Nano-clays have garnered considerable interest as a reinforcement compound for biopolymers, primarily due to their ability to improve the gas barrier properties of polymeric materials[20]. Nano-clays embedded in polymers create a longer "tortuous pathway" for gas molecules, enhancing the $O_2$barrier ability of the film[20]. Moreover, research has demonstrated the antimicrobial activity of nano-clay in polymer film structures, and inorganic clay have been utilized as carriers for inorganic biocides such as Cu, Fe, Mg, and Ag[20,21].

Packaging with polyethylene (PE) films holds promise for extending the shelf-life of strawberries by protecting and preserving the fruit from external factors, especially microorganisms. However, the influence of film structures and compounds used in the production of PE packaging on fruit quality maintenance remains a challenge. Therefore, this study aims to compare the quality changes and postharvest losses of strawberries packed with conventional LDPE films and bio-nanocomposite LDPE films.

## Materials and methods

### Fruit

Strawberry fruit (*Fragaria* × *ananassa* Duch. cv. Paros) were purchased and harvested early in the morning at commercially mature stage with color indexes L*= 45.54 ±0.63, a*= 48.71 ±0.63 and b*=35.05 ±1.16 at a local farm located in Sanandaj (35° 10 ×5.26´N, 46° 56 ×46.67´E and 1460 m above sea level), Iran. After harvest, strawberry fruit were immediately transported to the laboratory.

### Treatments and packaging

Fruit with similar size (˜10 g) and color and free of blemishes or disease were selected. Three types of LDPE films, i.e., (1) Experimental biodegradable LDPE (BLDPE), (2) biodegradable LDPE/nano-clay (BLDPE/NC) and (3) biodegradable LDPE/nano-mineral (BLDPE/NM) films (Baspar Pishrafteh Sharif Company, Iran) and conventional low-density polyethylene (CLDPE) film was used as a control. 18 bags (0.15 m × 0.15 m) from each film were prepared and about 0.1 kg fruit was packed in films randomly. All packages were stored at 4 °C with ≥ 80% relative humidity and quality characteristics and postharvest losses of fruit were evaluated at 0, 3, 6, 9, 12, 15 and 18 d. Three bags of each film type were sampled at each time point as 3 replications. The quality of the 3 set of fruit was immediately assessed after harvest (day 0).

### Characterization of e PE films

Thicknesses of the prepared films were measured with a micrometer (Diamond, China) with 0.01 mm sensitivity. Opacity of LDPE films was measured by a spectrophotometer (Unico UV-2100, USA) at 600 based on the following Eq[22].

$$Opacity\ \ = Abs_{600}/\ Film\ thickness$$

Tensile strengths of 10 identical specimens (about 0.05 m in length and 0.01 m in width) of each LDPE film were measured using ASTM with a texture analyzer (Santam, STM-1, Iran) with moving clamp speed of 0.08 m $s^{-1}$. The results were expressed in Newton (N).

The $O_2$ transmission rate (OTR) of the PE films was measured according to ISO 15105-1[23], based on differential-pressure methods. For this purpose, a set up was prepared and a volume control device (Custom made device, Iran) was placed in the setup. The sample area of 225 $cm^2$was tested under ambient conditions (25 °C and 50% relative humidity). The OTR was expressed as the mL ($m^2$. $day^{-1}$) using the following equation:

$$P_O = \ \ (OTR\ \times\ h)\,/\ \Delta_P$$

$P_O$ = oxygen permeability.
h = the film thickness (mm).
$\Delta_P$ = the water partial pressure difference between the inner and outer sides of the film, identified with the vapor pressure of water at 25 °C and 50% relative humidity, $\Delta P = 1.01 \times 10^5$ Pa.

The water vapor transmission rate (WVTR) of the PE films was measured according to ISO 15106-1[24], based on humidity detection sensor method. For this purpose, a set up was prepared and a humidity sensor (Custom made device, Iran) was placed in the set up. The WVTR of films with a size of 225 $cm^2$at 25 °C and 50% relative humidity was expressed as the g ($m^2$. $day^{-1}$) based on the following equation:

$$P_{wv} = \ \ (WVTR\ \times\ h)\,/\ \Delta P$$

$P_{WV}$ = water vapor permeability.
h = the film thickness (mm).
$\Delta_P$ = the water partial pressure difference between the inner and outer sides of the film, identified with the vapor pressure of water at 25 °C and 50% RH, $\Delta P = 1.58 \times 10^3$ Pa.

## Microbiological evaluations

For each replicate, 10 g combined sample of all fruit under sterile conditions was taken and added to 0.09 L sterile peptone water (0.1% w/v) to obtain microbial count. Then, serial dilutions ($10^{-1}$ to $10^{-3}$) were made. Total aerobic mesophilic bacteria (TAMB) were counted by pour plate method on plate count agar (PCA, Merck, Germany) and total yeasts + molds (TYM) were counted by surface plate method on a potato dextrose agar (PDA, Merck, Germany). Samples were prepared in triplicate. Plates of TAMB and TYM were held for 2 d at 30 °C and 25 °C, respectively[25].

## Fruit quality

Fruit weight was recorded at harvest day ($W_1$) and at all time-points ($W_2$) and the results were presented as percentages of weight losses (WL) by following equation:

$$WL\ \% \ = \ (W_1 - \ W_2)\ /\ W_1 \times 100$$

Eight fruit were used to measure flesh firmness on the two opposite sides of the fruit using a texture analyzer (Santam, STM-1, Iran) with a 0.008 m-diameter flat probe. The instrument was moved at 0.002 m. $s^{-1}$ to 0.008 m fruit depth and determined the maximum force of flesh penetration; the results were expressed in newton (N).

Two slices from opposite sides of all fruit in each replicate were taken and juiced. Soluble solid content (SSC) in the extracted fruit juice was measured with a digital brix meter (Atago, Japan). 3 mL extracted fruit juice was diluted to 30 mL with distilled water and titrated with 0.1 N NaOH to a pH of 8.2 (Metrohom 827, Switzerland) to determine titratable acidity (TA). TA was expressed as the percentage of citric acid using the following equation:

$$TA\ \% \ = 100\ \times\ (V\ \times\ N\ \times\ Meq)\ /Y$$

where V is the volume of NaOH used (L), N is NaOH normality, Y is bulk volume (L) and Meq is milliequivalents of citric acid, (0.064).

## Vitamin C

Approximately 1/8 th segments from two sides of the fruit were frozen in liquid nitrogen, ground and stored at −70 °C. Frozen samples were used to measure vitamin C, total phenol, total flavonoid, total antioxidant activity and total anthocyanin concentrations.

Vitamin C was extracted from 0.5 g frozen sample in 1 mL ice cold metaphosphoric acid (5% w/v) and homogenates were centrifuged (Universal PIT 320 R Co., Iran) at 13,700 × g (15 min at 4 °C) for 15 min. Briefly, 100 µL supernatant was added to 500 µL metaphosphoric acid (10% w/v), then 300 µL 2, 6-dichlorophenolindophenol (DCPIP, (0.03%, w/v)) and 300 µL citrate buffer (pH 4.2) were added. The mixture was incubated for 30 min and absorbance was measured by spectrophotometer (Unico UV-2100, USA) at 510 nm. All steps were carried out at 4 °C[26]. Concentrations were obtained using aqueous ascorbic acid standard.

## Total phenolic (TP) and total flavonoid (TF)

About 0.5 g frozen samples was homogenized in 2 mL 80% methanol- 1% HCl – 19% distilled water for 12 h at 4 °C. After centrifugation (Universal PIT 320 R Co., Iran) at 13,700 × g for 15 min at 4 °C, supernatant was used for TP and TF measurements as described previously[26]. For TP measurement, 20 µL prepared extract, 750 µL Folin–Ciocalteu reagent (1:10 v/v), 250 µL distilled water, and 800 µL $Na_2CO_3$ 7.5% (w/v) were mixed. The mixed solution was held at room temperature for,1 h and absorption was measured using spectrophotometer (Unico UV-2100, USA) at 765 nm. Data were showed as mg of gallic acid $kg^{-1}$ on fresh weight (FW).

For TF, 200 µL prepared sample was added to a solution of 60 µL 5% (w/v) $NaNO_2$ and 1280 µL distilled water. After 5 min, 60 µL 10% (w/v) $AlCl_3$ was mixed. After another 6 min, 400 µL 1 M NaOH was added to the mixture and absorbance at 510 nm was measured (Unico UV-2100, USA). The data was shown as mg of catechin $kg^{-1}$of FW[27].

## Total anthocyanin concentrations (TAC)

Approximately 0.3 g frozen tissue was diluted with buffers of pH 1.0 or 4.5. Homogenates were then centrifuged at 13,700 ×g for 15 min. The absorbance of supernatants was measured with spectrophotometer (Unico UV-2100, USA) at 510 nm and 700 nm in both buffers. Then, TAC was expressed as mg of pelargonidin-3-glucoside equivalent per kg FW using the following Eq[28]:

$$A\ =\ (A_{510} - \ A_{700})_{pH\ 1.0} - \ (A_{510} - \ A_{700})_{pH\ 4.5}$$

$$TAC\ =\ (A\ \times\ MW\ \times\ DF\ \times\ 1000)\ /\epsilon$$

where MW is the molecular weight of pelargonidin-3-glucoside (443.39 g $mol^{-1}$), DF is a dilution factor, and Ɛ is the coefficient of molar absorptivity (15,600).

## Total antioxidant activity (TAA)

TAA in strawberry friut was measured by the scavenging of 2, 2-diphenyl-1-picryhydrazyl (DPPH) free radicals using a spectrophotometer (Unico UV-2100, USA). Briefly, DPPH in methanol was prepared at a concentration of 0.24 g $L^{-1}$. Methanol extract (200 µL) was mixed with 500 µL DPPH solution and absorbance was measured at 515 nm. TAA was calculated using the following Eq. 2[5]:

$$TAA\% = 100 \times (Abs_{Control} - Abs_{Sample})/Abs_{Control}$$

where Abs $_{control}$ and Abs $_{sample}$ are blank and sample absorptions, respectively.

### Superoxide dismutase enzyme (SOD)

The activity of superoxide dismutase (SOD) was measured as described by Rasouli & Koushesh Saba[26] and protein concentration in each sample was determined using bovine serum albumin (BSA) as a standard.

### Color

Color factors L* (lightness), a* (green to red) and b* (blue to yellow) were determined with a colorimeter (TES 135 A, Taiwan) and expressed by CIE Lab System. Two measurements were performed on opposite sides of each fruit. Total color changes (ΔE) were obtained using the following equation. Results were reported as mean ± SE.

$$\Delta E = \left[(\Delta L*)^2 + (\Delta a*)^2 + (\Delta b*)^2\right]^{1/2}$$

where ΔL*, Δa* and Δb* were the changes of L*, a* and b* of the stored fruit at any given time compared to fresh samples on harvest day.

### Decay

Strawberry fruit decay symptoms were manifested as surface microbial decay, brown spots and unmarketable appearance area on each fruit surface by a five visual point scale, as follows: 0 = symptoms were not observed; 1 = less than 1/4 symptoms; 3 = 1/4–1/2 symptoms; 4 = 1/2–3/4 symptoms and 5 = more than 3/4 symptoms. Results were determined by the following equation:

$$Decay\ index\ (DI) = \sum (Decay\ scale \times quantity)\ /\ (5 \times total\ fruit\ number) \times 100$$

### Data analysis

Analysis of variance (ANOVA) was performed with MSTAT-C. Variation sources were different packaging materials, storage times (d) and treatments and data were expressed as mean ± standard error (SE) ($n = 3$). Least significant difference (LSD) test at $P \leq 0.05$ was used for mean comparison. The correlation matrix and principal component analysis (PCA) were performed with GraphPad Prism (version: 9.5.1) software.

## Results

### Barrier properties of PE films

The results showed that the highest tensile strength was observed in BLDPE, BLDPE/NC, BLDPE/NM and CLDPE, respectively (Table 1.). The barrier properties of PE films were evaluated by measuring the OTR and WVTR (Table 1.). The results demonstrated that the films containing nano-fillers exhibited reduced OTR compared to conventional PE films. On the other hand, the biodegradable PE showed a decline in WVTR, while films containing nano-clay and nano-minerals exhibited higher WVTR compared to conventional PE films (Table 1.).

### Microbiological evaluations

The TAMB of friut in all packaging materials gradually increased during storage, with the CLDPE packaging showing a higher rate of increase compared to other packaging films (Fig. 1A). The YAM count in CLDPE packaging was also higher than in other materials at all time-points (Fig. 1B). After 18 d of storage, the friut packed with BLDPE/NC packaging film exhibited the lowest TYM counts (2.52 Log CFU g$^{-1}$). While in this time, the lowest amount of TAMB was observed in BLDPE/NC and BLDPE/NM.

### Fruit quality

The WL of friut in all packaging materials increased during storage. The highest WL was observed in friut packed in BLDPE/NM, followed by CLDPE film. On the last day of storage, the WL in BLDPE and BLDPE/NC packaging materials was 0.62% and 0.82%, respectively, compared to 1.48% and 5.53% in conventional LDPE and antimicrobial packaging materials, respectively (Fig. 1C).

| **PE films** | **Thickness (mm)** | **Tensile testing (*N*)** | **Opacity** | **OTR (mL (m$^{-2}$ d)$^{-1}$)** | **WVTR (mL (m$^{-2}$ d)$^{-1}$)** | **Additive** |
|---|---|---|---|---|---|---|
| CLDPE | 0.01 | 2.14 ± 0.11 d | 3.098 ± 0.014 a | 1957.4 ± 66 a | 15.3 ± 0.8 bc | - |
| BLDPE | 0.07 | 8.31 ± 0.11 a | 0.956 ± 0.004 c | 1856.9 ± 31 b | 12.1 ± 1.4 c | Biodegradable |
| BLDPE/NC | 0.05 | 6.47 ± 0.08 b | 1.064 ± 0.003 c | 735.5 ± 69 c | 17.52 ± 1.0 ab | Biodegradable, nano-clay |
| BLDPE/NM | 0.04 | 4.59 ± 0.10 c | 1.914 ± 0.011 b | 959.9 ± 30 d | 21.36 ± 1.2 a | Biodegradable, nano-mineral |
| LSD 0.05 | | **1.079** | **0.280** | **27.41** | **4.08** | |

**Table 1**. Characterizations of PE films were used for packaging of strawberry fruit. Values represent means ± standard error ($n = 5$).

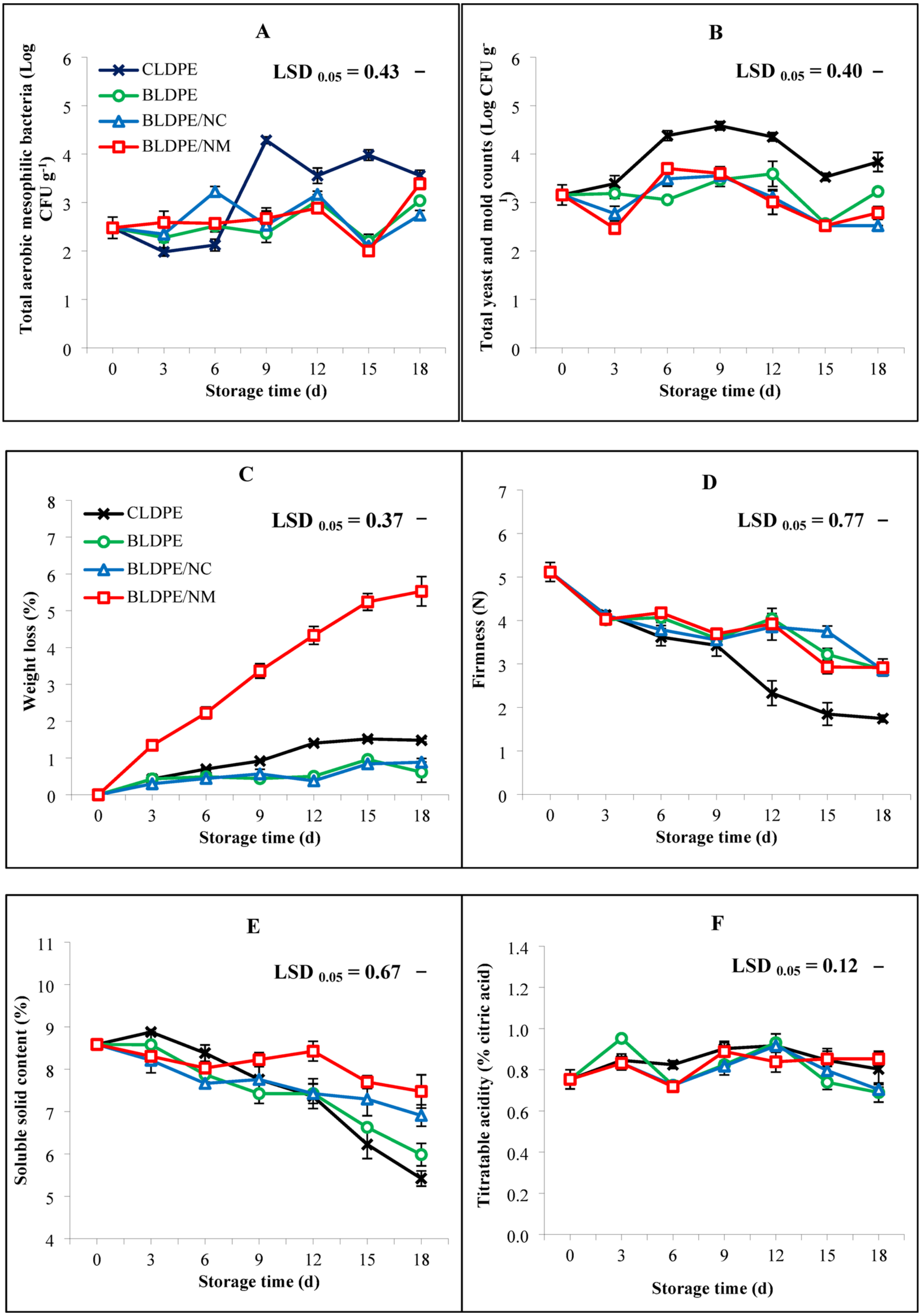


**Fig. 1**. Total aerobic mesophilic bacteria (**A**), total yeast + mold counts (**B**), WL (**C**), firmness (**D**), SSC (**E**) and TA (**F**) in strawberry fruit packed with conventional LDPE (CLDPE), biodegradable LDPE (BLDPE), biodegradable LDPE/nano-clay (BLDPE/NC) and biodegradable LDPE/nano-mineral (BLDPE/NM) films. Fruit was stored at 4 °C for up to 18 d. Data represent the mean ± standard error (*n* = 3).

The firmness of friut in all packaging materials decreased during storage, with no significant differences ($P \leq 0.05$) observed among the different packaging films until the 9 d. Afterward, the rate of decrease in firmness was higher in CLDPE packaging compared to other films (Fig. 1D).

During storage, the SSC content of friut decreased in all packaging films. After 9 d of storage, the SSC content of friut packed with BLDPE/NM film was greater than those in other packaging materials (Fig. 1E). The TA of all packaging materials showed relatively stable trends, with no significant differences ($P \leq 0.05$) between the packaging films during the storage period (Fig. 1F).

### Vitamin C

As shown in Fig. 2A, vitamin C content showed no significant changes ($P \leq 0.05$) in friut packed in CLDPE, while it increased in those packed in BLDPE, BLDPE/NC, and BLDPE/NM films.

### TP, TF and TAC

The TP levels of fruit in all packaging films exhibited a similar trend, initially increasing and then decreasing, with peak values at 6 d for CLDPE, BLDPE/NC, and BLDPE/NM films, and at 9 d for BLDPE films. The lowest TP was observed in friut packed in CLDPE film after 18 d of storage (Fig. 2B). The trend of TF followed a similar pattern, increasing initially and then decreasing over storage (Fig. 2C). Fruit packed in CLDPE film consistently had lower TF compared to other packaging films. The TAC of friut in BLDPE/NM packaging increased until 3 and 6 d of storage for CLDPE, BLDPE, and BLDPE/NC films, but gradually decreased thereafter. The lowest TAC was detected in friut packed with CLDPE film at the end of the storage (Fig. 2D).

### TAA and SOD

The TAA of friut in all packaging materials decreased during the first 6 d of storage. After the 6 d, the antioxidant activity of friut packed in CLDPE film increased, but it remained significantly lower than that at harvest time. TAA in other packaging films increased after 6 d, but there were no significant ($P \leq 0.05$) changes compared to harvest time (Fig. 2E). SOD activity initially decreased at 3 d of storage but gradually increased thereafter. The rate of increase was lower in friut packed with CLDPE film compared to other packaging films (Fig. 2F).

### Color

The changes in color parameters L* and total color changes (ΔE) during storage are shown in Fig. 3. There were no significant differences ($P \leq 0.05$) in L* values of friut among all packaging materials until the 9 d of storage. Subsequently, L* values decreased in CLDPE packaging, while they remained relatively stable in other packaging materials until the end of the storage period (Fig. 3A). The ΔE values of friut in all packaging materials increased storage. No significant differences ($P \leq 0.05$) were observed among fruit ΔE values for all packaging materials until the 12 d, but thereafter, ΔE was significantly higher in CLDPE packaging (Fig. 3B).

### Decay

The first signs of decay were observed in friut packed with CLDPE film after 6 d, and the decay rapidly increased until the end of the storage period. In comparison, friut packed in BLDPE, BLDPE/NC, and BLDPE/NM films exhibited significantly lower decay rates, with average decay values of 4%, 0, and 6% respectively, compared to 54% in CLDPE packaging (Fig. 4).

### Correlation and PCA

The analysis revealed strong correlations among various parameters. The highest positive correlations were found between DI and ΔE ($R = 0.83^{***}$), firmness and SSC ($R = 0.73^{***}$), and SSC and TAC ($R = 0.63^{***}$). Conversely, the most significant negative correlations were observed between DI and L* ($R = -0.72^{***}$), ΔE and firmness ($R = -0.72^{***}$), ΔE and SSC ($R = -0.65^{***}$), DI and firmness ($R = -0.63^{***}$), and DI and TAC ($R = -0.60^{***}$) (Fig. 5).

The PCA conducted on various parameters related to the quality and microbial load of strawberries packed in PE films revealed significant insights into the relationships among these variables. The PCA identified key components that explain a substantial portion of the variance in the dataset. Notably, the first two principal components (PC1 and PC2) accounted for 54.92% and 21.19% of the total variance, respectively, highlighting their importance in understanding the underlying structure of the data (Fig. 6).

## Discussion

The mechanical properties (including percent elongation, tensile strength, bursting strength, and tear strength) and barrier properties (OTR and WVTR) of LDPE films play an important role in delivering a quality product to the consumer[12]. Tensile strength is a crucial characteristic for packaging materials, as it is a key measure of film strength. Consistent with this, research by Xing et al.[29] and Chen et al.[30]has shown that the incorporation of nano-minerals and nano-clays increases the tensile strength of packaging films. This mechanical strengthening has been attributed to the formation of hydrogen and ionic bonds between nanomaterials and polymer particles[31]. However, studies indicate that an optimal concentration exists for each nano-filler, and that higher concentrations can impair the tensile properties of the resulting polymer films[32].

The barrier properties of packaging films are crucial in preventing the permeation of gases and water vapor, as these can affect the quality and shelf-life of packaged products[19]. In current study, the results indicated that the films containing nano-fillers exhibited reduced OTR compared to conventional PE films. This reduction in OTR suggests that the presence of nano-fillers contributed to improved $O_2$ barrier properties. The decreased OTR in films with nano-fillers can be attributed to the tortuous path created by the nano-fillers within the LDPE matrix. The presence of nano-fillers hinders the diffusion of $O_2$molecules, making it more difficult for them to pass through the film. This phenomenon has been reported in the literature, supporting the findings of the

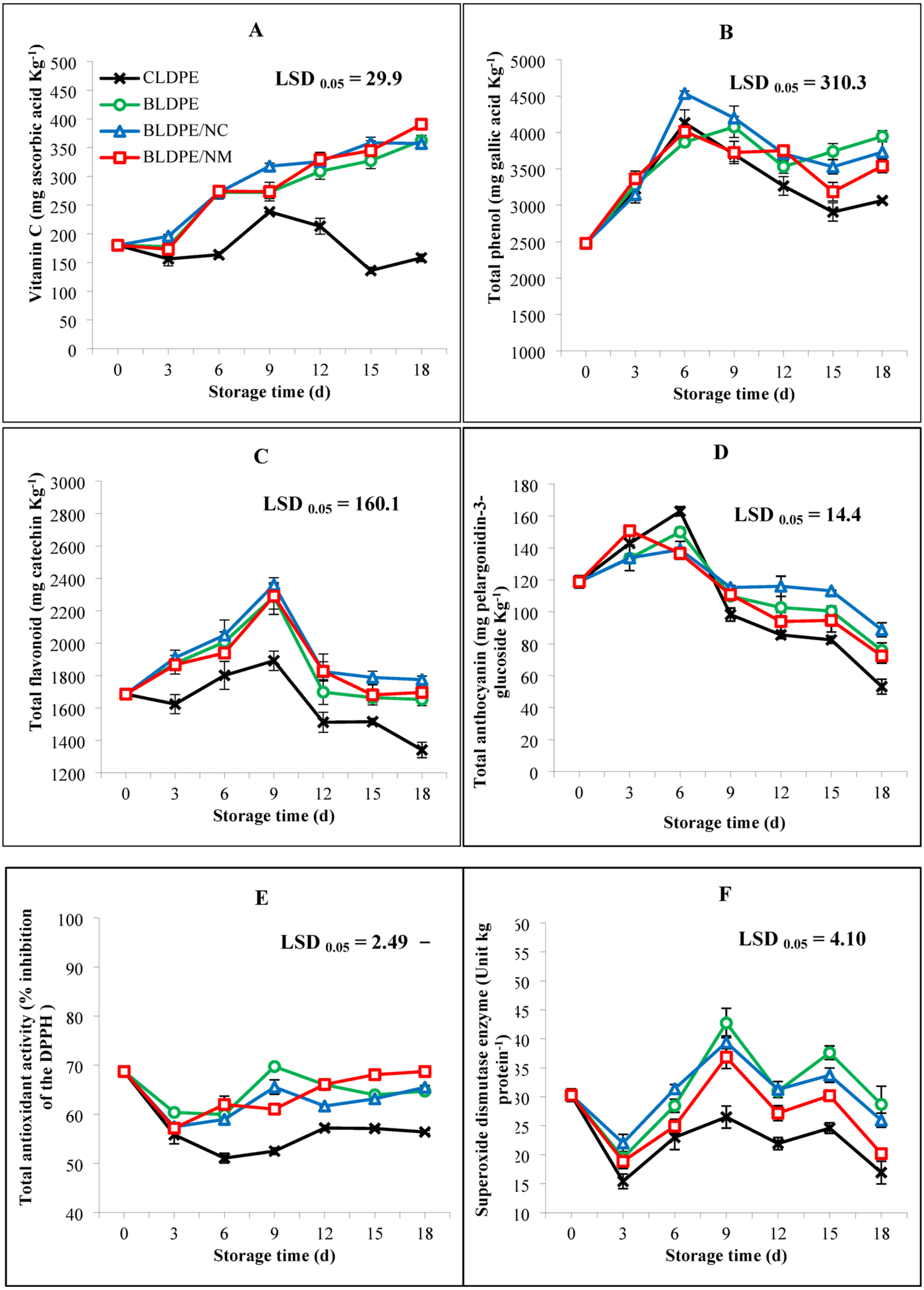


**Fig. 2**. Vitamin C (**A**), TP (**B**), TF (**C**), TAC (**D**), TAA (**E**) SOD activity (**F**) in strawberry fruit packed with conventional LDPE (CLDPE), biodegradable LDPE (BLDPE), biodegradable LDPE/nano-clay (BLDPE/NC) and biodegradable LDPE/nano-mineral (BLDPE/NM) films. Fruit was stored at 4 °C for up to 18 d. Data represent the mean ± standard error (*n* = 3).

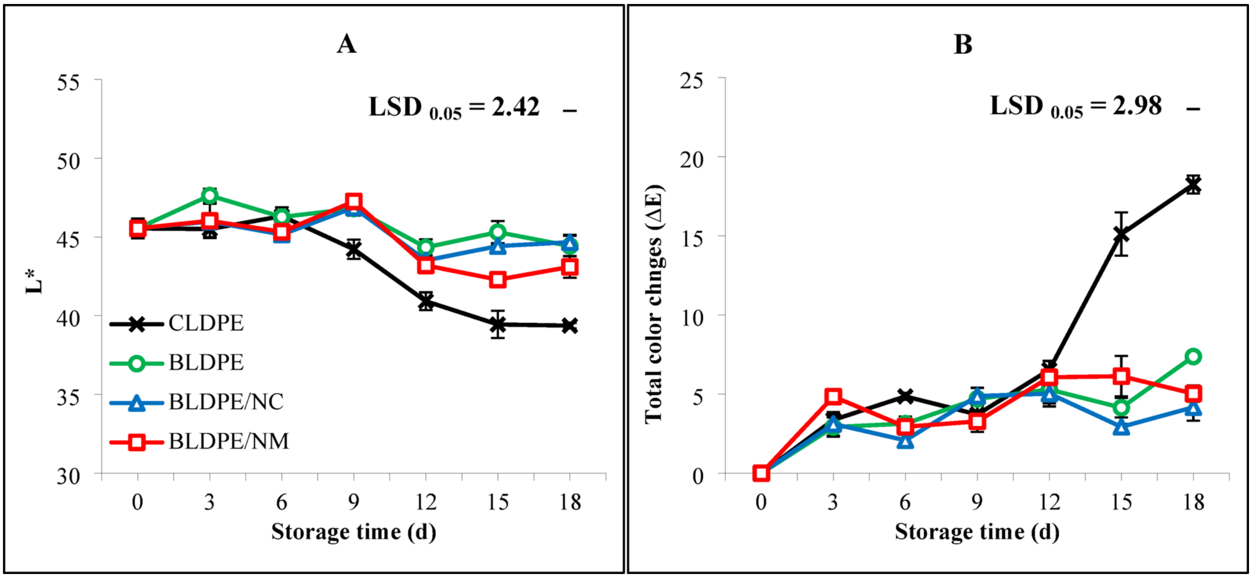


**Fig. 3**. L* (**A**), ΔE (**B**) and Decay in strawberry fruit packed with conventional LDPE (CLDPE), biodegradable LDPE (BLDPE), biodegradable LDPE/nano-clay (BLDPE/NC) and biodegradable LDPE/nano-mineral (BLDPE/NM) films. Fruit was stored at 4 °C for up to 18 d. Data represent the mean ± standard error (*n* = 3).

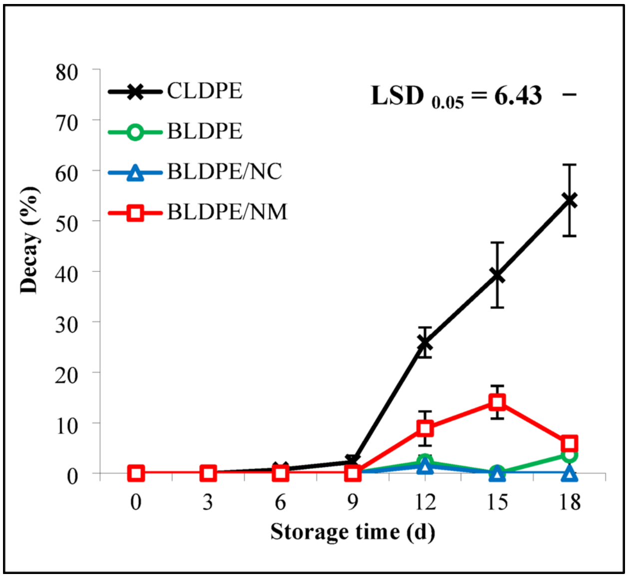
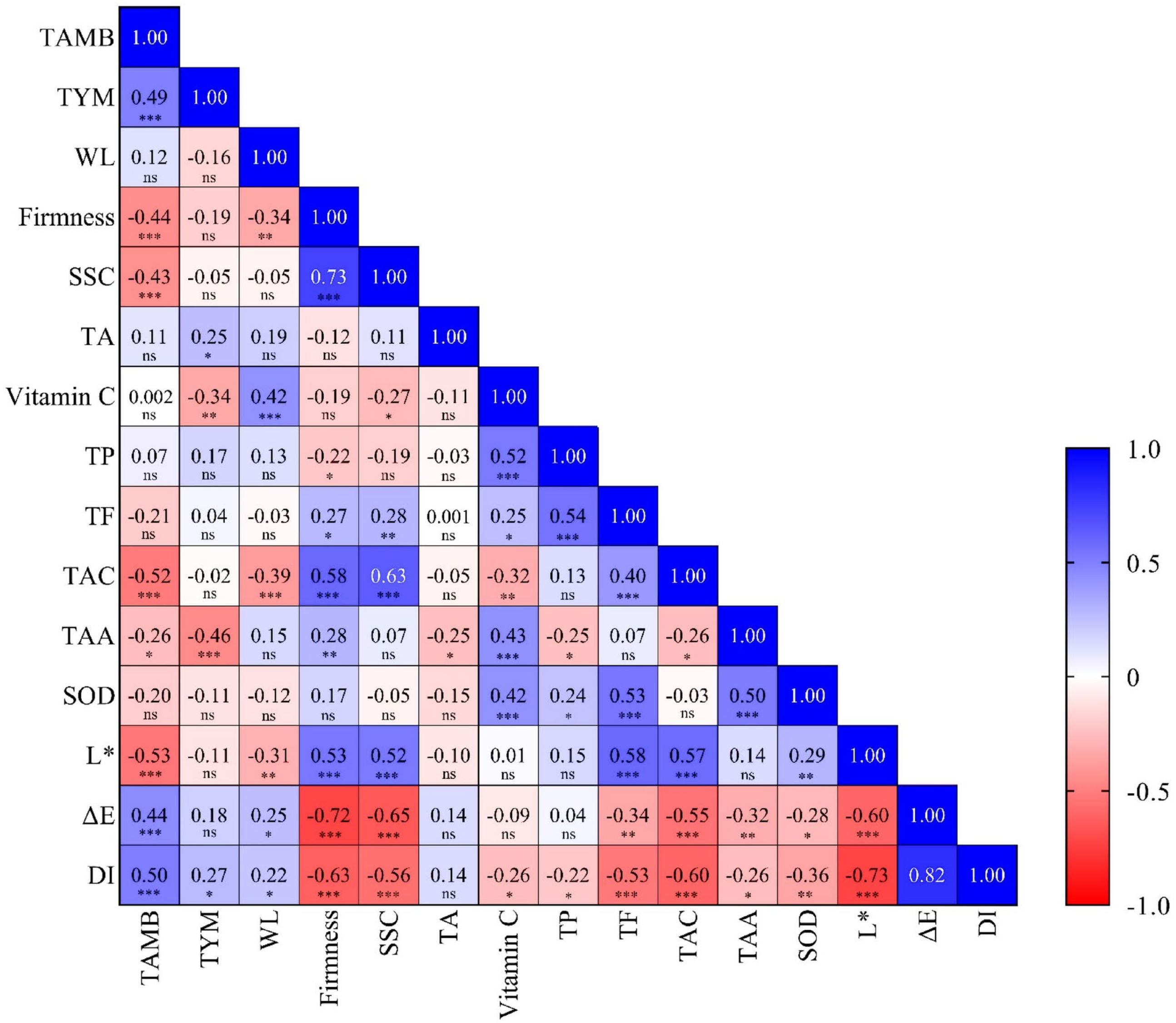


**Fig. 4**. Decay in strawberry fruit packed with conventional LDPE (CLDPE), biodegradable LDPE (BLDPE), biodegradable LDPE/nano-clay (BLDPE/NC) and biodegradable LDPE/nano-mineral (BLDPE/NM) films. Fruit was stored at 4 °C for up to18 d. Data represent the mean ± standard error (*n* = 3).

study[33]. By impeding the $O_2$ permeation, the films effectively reduce the exposure of packaged products, such as strawberries, to $O_2$, which helps minimize oxidative reactions and preserve the freshness and quality of the food.

On the other hand, the WVTR results showed contrasting trends. In the biodegradable PE films, WVTR declined compared to conventional PE films, indicating improved water vapor barrier properties. This reduction in WVTR suggests that the biodegradable PE films exhibited better resistance to water vapor permeation, which can help prevent moisture loss or gain in the packaged products. This feature is beneficial in preserving the moisture content and texture of the strawberries. However, in the films containing nano-clay and nano-minerals, WVTR was higher compared to conventional PE films. The increased WVTR in these films can be attributed to the absorption of water molecules by the nano-clay and nano-minerals. The presence of these nano-fillers might facilitate the absorption of water, leading to an increased transmission of water vapor through the film. This effect could be a potential drawback when it comes to preserving the moisture-sensitive products, as it may result in moisture loss or condensation inside the packaging[9,10].



| | TAMB | TYM | WL | Firmness | SSC | TA | Vitamin C | TP | TF | TAC | TAA | SOD | L* | ΔE | DI |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| TAMB | 1.00 | | | | | | | | | | | | | | |
| TYM | 0.49 *** | 1.00 | | | | | | | | | | | | | |
| WL | 0.12 ns | -0.16 ns | 1.00 | | | | | | | | | | | | |
| Firmness | -0.44 *** | -0.19 ns | -0.34 ** | 1.00 | | | | | | | | | | | |
| SSC | -0.43 *** | -0.05 ns | -0.05 ns | 0.73 *** | 1.00 | | | | | | | | | | |
| TA | 0.11 ns | 0.25 * | 0.19 ns | -0.12 ns | 0.11 ns | 1.00 | | | | | | | | | |
| Vitamin C | 0.002 ns | -0.34 ** | 0.42 *** | -0.19 ns | -0.27 * | -0.11 ns | 1.00 | | | | | | | | |
| TP | 0.07 ns | 0.17 ns | 0.13 ns | -0.22 * | -0.19 ns | -0.03 ns | 0.52 *** | 1.00 | | | | | | | |
| TF | -0.21 ns | 0.04 ns | -0.03 ns | 0.27 * | 0.28 ** | 0.001 ns | 0.25 * | 0.54 *** | 1.00 | | | | | | |
| TAC | -0.52 *** | -0.02 ns | -0.39 *** | 0.58 *** | 0.63 *** | -0.05 ns | -0.32 ** | 0.13 ns | 0.40 *** | 1.00 | | | | | |
| TAA | -0.26 * | -0.46 *** | 0.15 ns | 0.28 ** | 0.07 ns | -0.25 * | 0.43 *** | -0.25 * | 0.07 ns | -0.26 * | 1.00 | | | | |
| SOD | -0.20 ns | -0.11 ns | -0.12 ns | 0.17 ns | -0.05 ns | -0.15 ns | 0.42 *** | 0.24 * | 0.53 *** | -0.03 ns | 0.50 *** | 1.00 | | | |
| L* | -0.53 *** | -0.11 ns | -0.31 ** | 0.53 *** | 0.52 *** | -0.10 ns | 0.01 ns | 0.15 ns | 0.58 *** | 0.57 *** | 0.14 ns | 0.29 ** | 1.00 | | |
| ΔE | 0.44 *** | 0.18 ns | 0.25 * | -0.72 *** | -0.65 *** | 0.14 ns | -0.09 ns | 0.04 ns | -0.34 ** | -0.55 *** | -0.32 ** | -0.28 * | -0.60 *** | 1.00 | |
| DI | 0.50 *** | 0.27 * | 0.22 * | -0.63 *** | -0.56 *** | 0.14 ns | -0.26 * | -0.22 * | -0.53 *** | -0.60 *** | -0.26 * | -0.36 ** | -0.73 *** | 0.82 | 1.00 |

**Fig. 5**. Correlation heatmap among total aerobic mesophilic bacteria (TAMB), total yeast + mold counts (TYM), weight losses (WL), firmness, soluble solid content (SSC), titratable acidity (TA), Vitamin C, total phenolic (TP), total flavonoid (TF), total anthocyanin concentrations (TAC), total antioxidant activity (TAA), superoxide dismutase enzyme (SOD), total color changes (ΔE) and decay index (DI) of strawberry fruit packed in polyethylene films. The red (−1) and blue (+ 1) colors, respectively, exhibit the positive and negative correlations between different indexes. (ns p > =0.05; * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$).

Microorganisms, particularly fungal agents such as grey mold rot (*Botrytis cinerea* Pers.) and rhizopus rot (*Rhizopus stolonifer*Ehrenb.), play a significant role in causing quality changes and postharvest losses in strawberry friut[4]. Incorporating nano-clay and metal ions, such as silver (Ag), into polymeric packaging materials enhances their antimicrobial properties[9,20]. These agents can alter the membrane permeability, enzyme systems, and metabolism of microorganisms, leading to their death[34]. Furthermore, bio-nanocomposites can reduce microbial growth in packaging by absorbing water drops produced through vapor condensation inside the packaging and by altering the gas composition within the package[10,18]. The OTR and WVTR values of the PE films (Table 1.) support these findings.

WL in friut is directly associated with water loss and relative humidity inside the package. The temperature during film production, density of PE, additive compounds, film thickness, and nano-fillers, influence the barrier properties of PE films against gases[35]. Nano-fillers can act as physical and chemical barriers, slowing down the diffusion of $O_2$, $CO_2$, and water vapor through the polymeric matrix. Additionally, the barrier properties against gases can indirectly affect fruit respiration and alter the gas composition inside the package, resulting in reduced WL. The results also indicated that the LDPE structure effectively maintained the weight of strawberry friut, and the higher film thickness in BLDPE packaging materials and the presence of nano-clay in BLDPE/NC packaging materials increased the barrier properties against water vapor and gases, leading to lower WL in strawberry friut. The higher WL in BLDPE/NM packaging material correlated with its higher WVTR. In addition to the

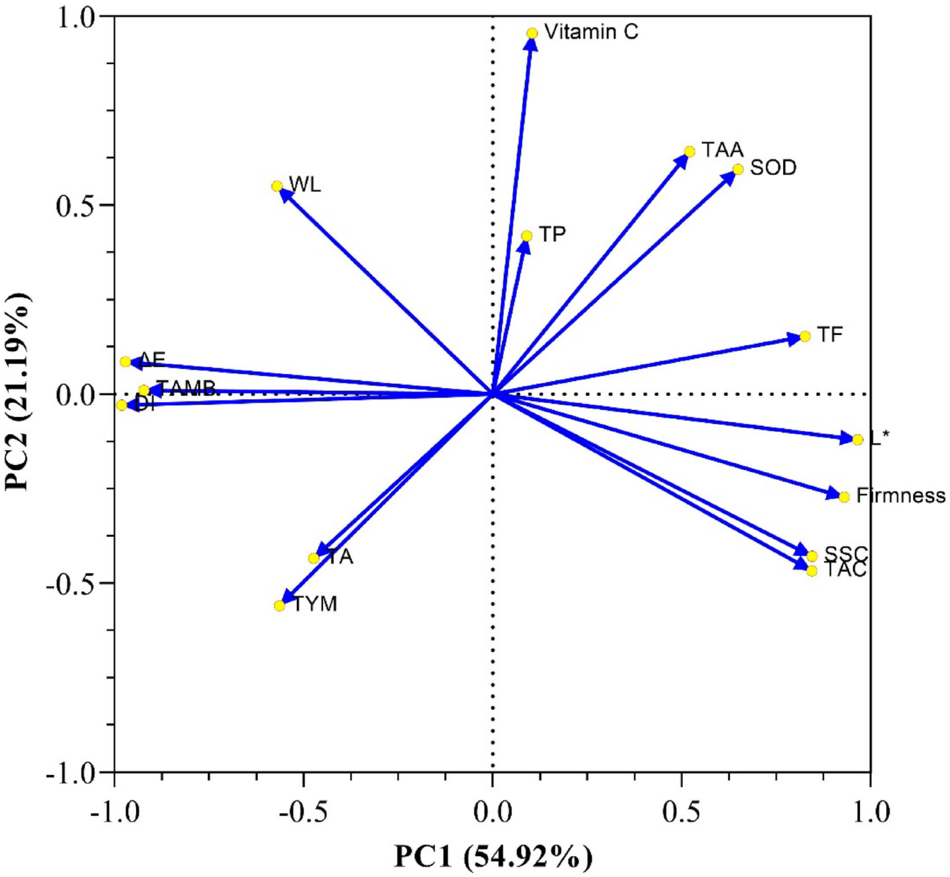


**Fig. 6**. Principal component analysis (PCA) of total aerobic mesophilic bacteria (TAMB), total yeast + mold counts (TYM), weight losses (WL), firmness, soluble solid content (SSC), titratable acidity (TA), Vitamin C, total phenolic (TP), total flavonoid (TF), total anthocyanin concentrations (TAC), total antioxidant activity (TAA), superoxide dismutase enzyme (SOD), total color changes (ΔE) and decay index (DI) of strawberry fruit packed in polyethylene films.

composition of the polymer layer, the WVTR is also affected by the thickness of the polymer layer, so that increasing the thickness of the polymer layer decreases the amount of WVTR[36,37].

Firmness is a crucial quality attribute that influences consumer appeal and marketing of strawberry friut. The application of PE films incorporated with nano-silver (nano-Ag), nano-titanium dioxide (nano-$TiO_2$), and clay has been shown to delay fruit softening during storage[14,38]. The decrease in firmness during storage is primarily attributed to the solubilization of pectins by pectolytic enzymes such as polygalacturonase[39]. Additionally, WL, mechanical damage, and decay caused by microorganisms contribute to the reduction in strawberry firmness[40,41]. The gas composition inside the package, regulated by packaging materials, plays a key role in the respiration rate of friut during postharvest storage[42]. A low resistance to $O_2$ and water vapor transmission, as well as antimicrobial properties, can help maintain fruit firmness by reducing the activity of pectolytic enzymes and microbial growth.

SSC is an important quality parameter that affects the consumer acceptability of strawberry friut. The current study finding is consistent with previous research that reported a decrease in SSC of strawberries packed in PE films during storage[40,43]. The TA of strawberries affects fruit quality, and its content usually decreases gradually during storage[5]. Our findings showed that PE packaging could maintain TA. Sugars and acids are the main substrates in fruit respiration. The high physiological activity and respiration rate of strawberries facilitate sugar hydrolysis, leading to a decrease in SSC during postharvest storage[44]. The application of bio-compounds such as nano-clay and nano-minerals in the structure of polymers as fillers or reinforcements improves the barrier properties against gases[45]. These compounds help maintain a balance between the external atmosphere and the atmosphere inside the package, thus suppressing the hydrolysis of sugars by reducing physiological activity and respiration rate.

Strawberry is a rich source of vitamin C[46]. Generally, vitamin C content decreases during the storage period. However, there are reports of increased vitamin C in strawberries over storage[47,48]. Packaging with nano-PE has been shown to help maintain ascorbic acid (AA) during postharvest[49]. The mechanism by which the packaging material affected vitamin C in the current study is unclear, but there are two possible pathways: microbial load and biochemical reactions. It has been shown that postharvest decay can decrease the vitamin C content of friut[50]. Furthermore, changes in PE structure and composition can affect catabolic changes, such as respiration[35], which can induce oxidative stress. Vitamin C is an important antioxidant and can directly remove reactive oxygen species (ROS)[26]. Therefore, under higher oxidative conditions, it may be decreased. However, further studies are required to understand the effect of PE film packaging on vitamin C changes.

Phenolic compounds, including phenolic acids, flavonoids, and anthocyanins, are important bioactive compounds in strawberries. These compounds play crucial roles in various biological functions, such as protecting against cold, ROS, microbial attack, and maintaining fruit quality[51]. The concentration of phenolic compounds in strawberry friut can either be maintained or altered during storage. Phenolic compounds undergo oxidation by the polyphenol oxidase enzyme in the presence of $O_2$, leading to the production of brown or black pigments[26]. Biopolymers and nano-biopolymers are effective in preventing gas permeation, particularly $O_2$, through polymer films[10,34]. Polymers with superior $O_2$barrier properties can reduce the hydrolysis of phenolic compounds by the polyphenol oxidase enzyme. Furthermore, phenolic decline can be minimized by maintaining cell wall integrity, fruit firmness, vitamin C content, and antioxidant capacity[26]. Our findings indicate that BLDPE, BLDPE/NC, and BLDPE/NM packaging films effectively prevented phenol reduction, likely due to increased $O_2$ barrier properties and better preservation of fruit quality, including enhanced vitamin C content.

Strawberry friut have high levels of antioxidant activity, which can contribute to consumer health. TAA comprises antioxidant compounds, such as vitamins, phenols, flavonoids, anthocyanins, and antioxidant enzymes, including catalase (CAT), superoxide dismutase (SOD), ascorbate peroxidase (APX), glutathione reductase (GR), and guaiacol peroxidase (GPX)[28]. The higher TAA content in strawberry friut packed in BLDPE, BLDPE/NC, and BLDPE/NM films may be attributed to better preservation of antioxidant compounds, such as phenols and flavonoids, as well as increased vitamin C content compared to friut packed in CLDPE films. Similarly, Li et al.[52] reported higher antioxidant compounds and enzyme activity in strawberry friut packed in nano-$TiO_2$-LDPE compared to CLDPE film.

The color of strawberry is one of the main desirable features that might determine consumer acceptance of the product. In agreement with our findings, nano-PE prevented decrease of L* value in mushroom[15]. It has been reported that strawberry becomes darker during storage[53]. Color change in strawberry is generally due to the formation of brown or dark tissues induced by the decrease of anthocyanins and oxidative browning reactions[54]. Weaker color changes of friut in BLDPE, BLDPE/NC and BLDPE/NM packages than CLDPE package in the current study may be due to the preservation of phenolic compounds, especially anthocyanin.

The decay of strawberries during storage is a significant concern, as it directly affects their postharvest quality. In this study, our findings are consistent with the results reported by Yang et al.[55], who also observed higher decay levels in friut packed in CLDPE films compared to those in nano-packing films. The higher decay in CLDPE packaging can be attributed to factors such as its lower barrier properties, which allow for increased $O_2$permeation and subsequent oxidative reactions. Additionally, CLDPE films may offer less protection against mechanical damage during transportation and have limited control over moisture levels, which can further contribute to the decay of strawberry friut. Packaging materials play a crucial role in protecting food items from various environmental factors, including gases, light, mechanical damage, chemical contaminants, and microorganisms[10,35]. In this study, the use of BLDPE, BLDPE/NC, and BLDPE/NM films demonstrated their effectiveness in preserving the quality of strawberries by reducing decay (Fig. 7). These films act as barriers, preventing the entry of $O_2$ and water vapor that can accelerate decay processes. Moreover, they offer improved mechanical strength compared to CLDPE, which helps protect the friut from physical damage during handling and transportation.

The incorporation of biopolymers and nano-fillers in packaging materials has gained attention due to their potential for enhancing functionality and providing additional benefits. Biopolymers, such as BLDPE, offer a sustainable and renewable alternative to conventional plastics, reducing the reliance on non-renewable resources. Nano-biopolymers, such as BLDPE/NC and BLDPE/NM films, can act as carriers for antioxidant and antimicrobial agents, further enhancing the preservation of friut through their added functionalities[10].

The PCA aimed to reduce the dimensionality of the data while retaining as much variance as possible, thus allowing for a clearer interpretation of how different factors interact[56]. The analysis showed a notable correlation between the TAMB and the TYM. High microbial counts were associated with increased WL and DI, suggesting that microbial proliferation adversely affects the shelf-life and quality of strawberries. These findings align with previous studies indicating that microbial activity is a primary factor in the deterioration of fruit quality during storage[57]. Firmness, SSC, and TA emerged as critical quality indicators that were negatively impacted by microbial growth. Specifically, as TAMB and TYM increased, firmness decreased, which is consistent with the textural changes observed in friut during spoilage. Additionally, the decrease in SSC and the changes in TA suggest that microbial activity may lead to fermentation processes, further degrading the fruit's sensory attributes[58]. Interestingly, higher microbial loads were also associated with lower levels of important antioxidants (vitamin C, TP, TF, and TAC), indicating that microbial spoilage may compromise the nutritional value of strawberries. This relationship underscores the importance of maintaining low microbial counts to preserve not only the quality but also the health benefits of the fruit[59]. Furthermore, the TAA and SOD levels were assessed as indicators of the fruit's capacity to counteract oxidative stress. A decline in antioxidant activity correlated with increased decay index and microbial counts, reinforcing the notion that microbial spoilage adversely affects the fruit's ability to resist oxidative damage[60]. Finally, ΔE was highlighted in the PCA as a significant visual indicator of spoilage. As microbial activity increased, noticeable color changes occurred, suggesting that consumers may rely on visual cues to assess fruit quality[61]. This finding emphasizes the necessity for effective packaging solutions to extend the shelf-life of strawberries and maintain their aesthetic appeal.

## Conclusion

This study's results indicate that the incorporation of nano-fillers in LDPE films can improve the $O_2$ barrier properties, reducing the OTR. However, the impact on water vapor barrier properties can vary depending on

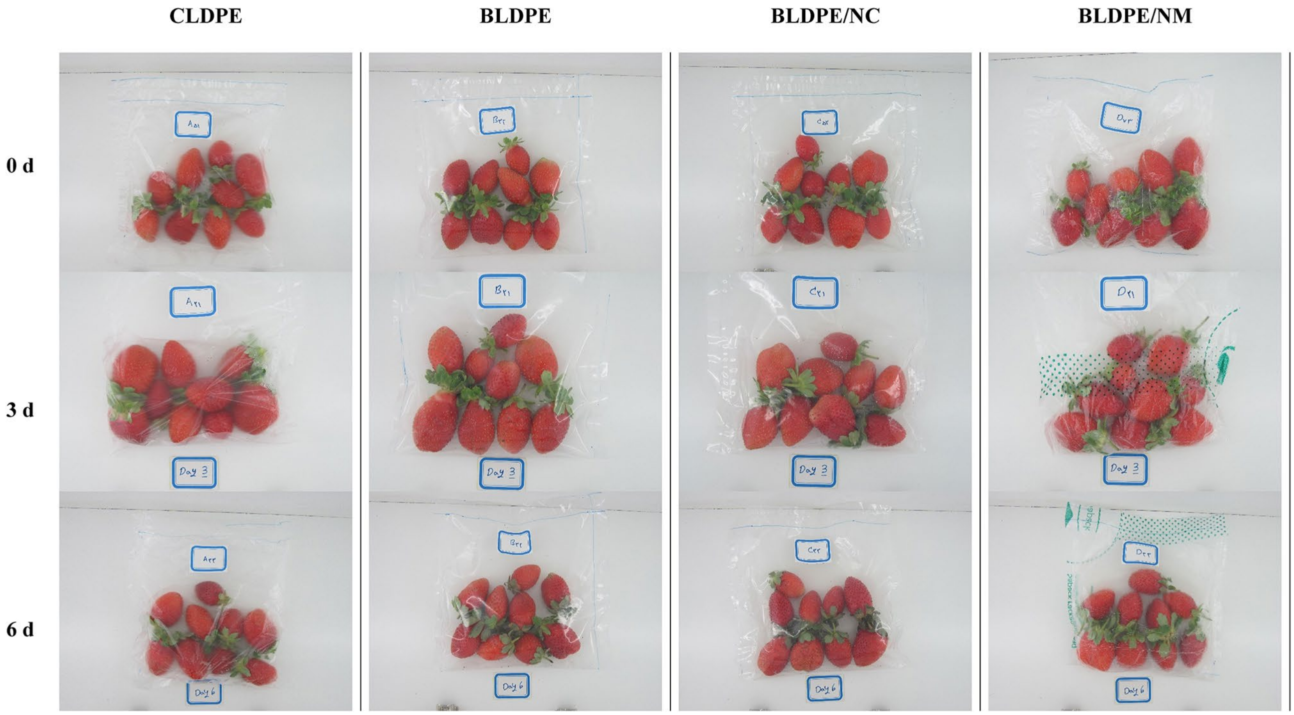


**Fig. 7**. Visual appearance of strawberry fruit packed with conventional LDPE (CLDPE), biodegradable LDPE (BLDPE), biodegradable LDPE/nano-clay (BLDPE/NC) and biodegradable LDPE/nano-mineral (BLDPE/NM) films. Fruit was stored at 4 °C for up to 18 d.

the specific nano-fillers used, with some films showing improved WVTR and others exhibiting higher WVTR due to the absorption of water molecules. These findings highlight the complexity of designing packaging films with optimal barrier properties and the need for careful selection and customization of materials to suit specific applications. Furthermore, the significant influence of different packaging materials was observed on the microbiological evaluations, fruit quality, vitamin C content, TP, TF, TAC, TAA, SOD activity, color, and decay of strawberries during storage. These findings highlight the importance of selecting appropriate packaging materials for strawberries to maintain their quality and extend their shelf-life during storage. Films incorporating nano-fillers, such as BLDPE/NC and BLDPE/NM, exhibited improved barrier properties and reduced decay, providing promising alternatives to conventional CLDPE films. The results from the PCA provide valuable insights into the interactions between microbial loads and various quality parameters of strawberries packed in PE films. These findings emphasize the importance of controlling microbial growth to maintain the quality, nutritional value, and visual appeal of strawberries during storage. Future research should explore innovative packaging techniques and preservation methods that can mitigate microbial spoilage and enhance the shelf-life of fresh produce. However, further research is warranted to optimize the packaging materials and develop strategies for enhancing the preservation of strawberries and other perishable friut.

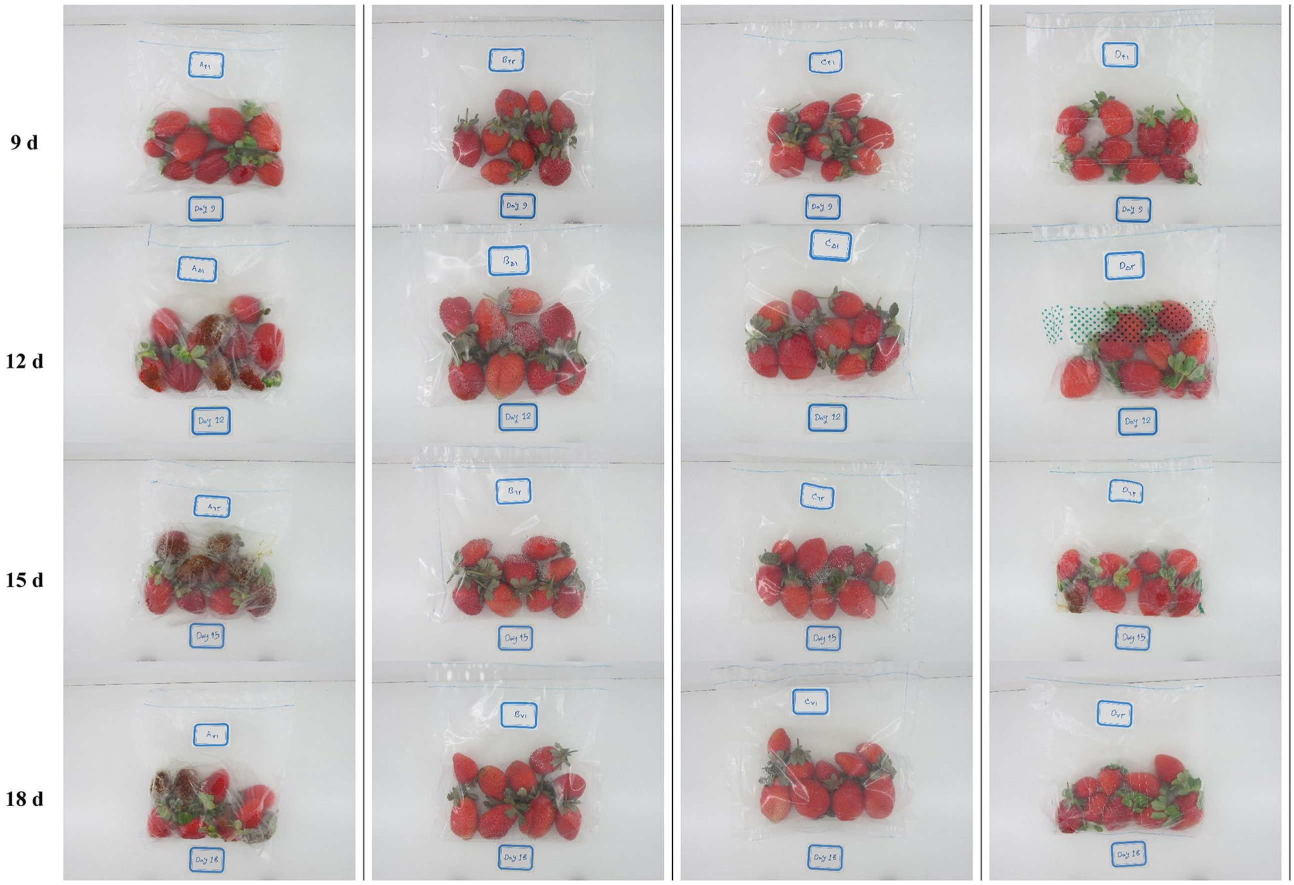


**Fig. 7.** (continued)

## Data availability

The datasets analyzed during the current study are available from the corresponding author on reasonable request.

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## Author contributions

MKS, MR: Conceptualization and designing the study, methodology.MKS: Supervision of the research, financial preparation. MR: Performing the experiment, collection of data.MKS and MR: Contribution to literature research and original draft preparation.MR, VF and MKS: Analyzing and interpretation of data and visualization. MKS and VF: Guiding all aspects of the research project and revising the manuscript.All authors read and approved the final manuscript.

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## Declarations

## Competing interests

The authors declare no competing interests.

## Additional information

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