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Original Research Article

Evaluation the Effect of Using Gelatin Membrane Loaded with ZnO, Ag Nanoparticles, and Nisin on Various Parameters of Yogurt

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Abstract: The study aimed to investigate the effect of silver AgNPs and zinc nanoparticles ZnNPs loaded on edible gelatin covers at concentrations of 20 and 30%, alone or with the nisin compound at a concentration of 50%, on some chemical properties of Yogurt was kept for 20 days at 5 ± 2°C. Preserving yogurt results showed a significant decrease ($p<0.05$) in moisture content during the storage period for all treatments., as well as the T7 treatment which silver nanoparticles were added at a concentration of 30% with the compound nisin at a concentration of 50% was the most capable of retaining moisture. As for the pH levels, T4 and T1 At the end of the treatments' storage time, they significantly reduced to be between (3.93 - 4.56). It was also found that the fat percentages continued to rise and reached their highest values in conjunction with the decrease in moisture values and were between (3.37 - 3.76 %) for both the T10 and T1 treatments at the completion of the storage time. As for the values of fatty acids, they did not differ significantly on the first day for all treatments, and they increased significantly as the storage period continuation, with values ranging between (1.42 - 1.94%) for the treatments, T4 and T3. As the humidity values decreased as the storage period continuation, as the storage period came to end, the ash percentage increased and reached its greatest values. The highest ash percentage was for treatment T7, reaching 0.83%, which differed significantly from the comparison treatment not coated with gelatin, T1, at which the ash percentage reached 0.70%.

Keywords: Nanoparticles, Gelatin Membrane, ZnO NPs, AgNPs, Nisin, Yoghurt.

INTRODUCTION

Since the beginning of time, milk and dairy products have been an important component of the human diet (Isra'a and Mahdi 2022). Within the food business, the dairy industry holds significant economic importance. It has encountered many challenges in meeting the increasing demand from consumers for high-quality, naturally produced products with minimal processing. Thus, utilizing leading-edge and novel technology might be a fascinating replacement. ،for example, the application of nanotechnology to delivery systems and packaging. This technique provides an intriguing method for supplying food preservatives that improve the mechanical barrier and functional qualities of packaging. It has the potential to enhance the quality and safety of dairy products. (Adriano *et al.*, 2023). During the past several decades, the dairy industry has experienced a period of global expansion and consolidation, which has presented a number of commercial opportunities as well as a variety of challenges relating to safety, sustainability, and innovation. Milk, an essential component of the dairy industry, is especially prone to degradation due to its relatively high fat, protein, and sugar content. (Claeys *et al.*, 2013). The study and description of materials with dimensions ranging from 10 to 100 nanometers, as well as the determination of their chemical, physical, and mechanical properties, are the subjects of nanoscience. Through the investigation of phenomena related to their small sizes. Since it investigates materials at the atomic or particle scale that differ significantly from their characteristics, nanoscience is frequently referred to as the science of extremely small materials. Similar substances on a bigger scale (Hussain *et al.*, 2017).

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Bacteria and fungi capable of manufacture nanoparticles with antimicrobial characteristics may produce both biological and chemical nanoparticles (Hussein *et al*., 2020). An attractive substitute for active food packaging was described as casein and gelatin nanocomposite films including liposome-encapsulated nisin and halloysite nanoclay (Boelter، J.F.; and Brandelli،A. 2016). In order to reduce the usage of plastic materials that are not biodegradable and cause environmental pollution, biomaterials had to be used in food packaging. However, biomaterials, which consist of occurring naturally plant or animal components including proteins, sugars, and lipids, are environmentally friendly, renewable, and biodegradable (Nemet *et al*., 2010). Thin biological layers that meet food product criteria and are recognized by international food organizations as safe from a health perspective are known as edible films. Along with food, they are consumed (Senturk *et al*., 2018). Because of its functional characteristics, gelatin is widely employed in a variety of industrial applications. The study on consumer awareness has led to investments in the preparation of edible films, such as coating cheese and meat, in recent years. Because of its ability to include antioxidants and microbial inhibitors, it has been utilized alone or in combination with fatty substances, proteins, and gums to produce edible wrappers that are notable for enhancing food qualities and increasing their shelf life (Ramos *et al*., 2016).

Yogurt was chosen for this study because to its importance as a commonly consumed food and its potential to get contaminated with germs that cause food poisoning either directly or indirectly during the manufacture, processing, marketing, or storage periods.

MATERIAL & METHOD

Yogurt Manufacture

The yogurt was prepared according to the method described by Tamime، A. Y. and Robinson، R. K. (2007). A portion of the milk was pasteurized for ten minutes at 90°C, and then it cooled to 42±2 °C. With Lactobacillus delbrueckii subsp bulgaricus and Streptococcus salivarius subsp thermophiles making up the starter culture, incubatedat 42±2°C for 4 to 6 hours, or until the pH fell to 4.6 as a result of the coagulation process. Following the coagulation procedure, the yogurt was kept at 5 °C until the required tests could be carried out one, ten, and twenty days from the date of manufacture.

Prepare Gelatin Membrane Loaded with ZnO and Ag Nanoparticles and Nisin

In accordance with Carvalho and Grosso's (2004) method, gelatin film solutions were made by weighing 10 g of gelatin powder, dissolving it in 80 ml of distilled water, and blending all the ingredients with a hot plate magnetic stirrer at 60 °C for 15 minutes. Following this, the volume was added to 100 ml of distilled water, the pH was adjusted to 7, and glycerol was added at a rate of 30% by weight of dried gelatin.

The Properties of Yoghurt Coated with Gelatin Films Were Tested

Chemical tests of Yoghurt

Estimation of Moisture Percentage

An electric oven was used to dry 3 g of yogurt samples at a temperature between 95 and 105 °C until a steady weight was reached. The percentage of moisture was then computed as suggested by Ling (2008).

$$\%\text{Moisture} = \frac{\text{Weigh the sample before drying- Weigh the sample after drying}}{\text{Weigh the sample}} \times 100$$

Measurement of pH:

Samples of yogurt were evaluated for pH by using a pH meter, Weigh 3 g of the sample with 10 ml of distilled water, mix well in a ceramic mortar, then measure the exponent (Hool *et al*., 2004).

Fat Percentage Estimation

The percentage of fat in the yogurt samples was estimated according to the Gerber method, by weighing 3 g of each sample and adding Sulfuric acid concentrate (10 ml) with amyl alcohol (1 ml) to each, centrifuging it for three to five minutes in Gerber tubes, and then measuring the fat column. As a percentage of fat in samples of yogurt, (Min and Ellefson, 2010).

Free Fatty Acids Percentage Estimation

Free fatty acids were determined in accordance with recommendations provided in A.O.A.C. (2010), through the mixture of 30 milliliters of ethanol alcohol, 30 milliliters of diethyl ether solvent, and 1 milliliter of phenolphthalein indicator. After adding 0.1 N sodium hydroxide to neutralize the prior mixture until a pink color emerged, after thoroughly mixing 10 grams of yogurt into the neutralizing solution, strain the mixture. Up until the neutralization point is reached, the filtrate is calibrated using the prior base, which is recognized by the solution taking on a pink hue. The quantity of free fatty acids—which are reported as a percentage of the free fatty acids in the yogurt and computed as oleic acid—was determined by measuring the amount of base consumed after the yogurt was added. In view of the subsequent equation:

F F A = 2.82 × T \ W
T: It is the volume of a 0.1 N solution of sodium hydroxide solution required for titration, in milliliters
W: Sample weight in grams.

Ash Percentage Estimation
Ash Percentage

2 grams of samples from yogurt were taken out and put into a porcelain jar with a known weight. For about six hours, or until white ash was produced, the samples were burned in an oven set to 550°C. After transferring the ash to a drying container to cool, the weight was measured. The direct burning method described in A.O.A.C. (2010) was used to calculate the ash percentage using the following equation:
Ash % = (The weight of the empty jar - weight of the jar with the ash contained) / (the original model's weight)

Statistical Analysis

The data were statistically analyzed through the experimental system within the prepared statistical program (SAS‹ 2012) and by using the complete randomized design system CRD as the averages were chosen according to the Dancan test (Dancan‹ 1955).

Results and Discussion

- Characteristics of yogurt covered in gelatinous films
- The chemical composition of yoghurt

Moisture Percentage

The results in Table (1) for the average storage periods indicated that the humidity value differed significantly for all storage periods, and the lowest was the humidity rate at 20 days, which reached 81.56%, and the best was the ability to retain moisture at the end of the first day, which amounted to 87.43%. Likewise, with regard to the humidity rate for all treatments, it was obtained Variation in values between treatments. Treatment T5 coated with gelatin supplemented with nisin was the most capable of retaining moisture, reaching 88.01%. The results also showed that uncoated cheese samples covered in layers of gelatin or gelatin which AgNPs, ZnNPs and and adding nisin, the moisture content significantly decreased ($P < 0.05$) from the period beginning of the experiment and the 20 days of storage at a temperature of 5 ± 2°C. Moisture percentages were for the treatments including the first day, T1, T2, T3, T4, T5, T6, T7, T8, T9, T10, and T11, were 78.00, 89.30, 88.53, 87.63, 90.95, 88.66, 89.43, 87.06, 87.06, 88.70, and 88.50%, respectively. It is clear from the results that treatment T5 was the best treatment in terms of ability to retain moisture for the first and tenth days, and T7 was the highest in terms of ability to retain moisture after the storage time has ended. This indicates the ability of the gelatin membrane supported with 50 mg nisin or supported with nisin and nanosilver to retain high moisture due to its ability to form a cohesive component to prevent moisture loss Compared to the rest of the treatments, The gradual decrease remained until the lowest moisture content was reached when the storage came to end the treatments' period. T1, T2, T3, T4, T5, T6, T7, T8, T9, T10 and T11, 73.03, 81.76, 83.90., 82.33,85.36,84.26,85.53,79.60,79.93,81.83,79.66%. Upon comparing the treatments, it was noticed that the uncoated control sample T1 had the lowest moisture content, at 73.03%. While the best treatment in terms of its ability to retain moisture was the T7 treatment, which reached 85.53%, coated with gelatin and nanosilver at a concentration of 30 mg with nisin.

The findings agreed with what was reported by Al-Jubouri (2021), who indicated that, in comparison to the control sample, gelatin films containing nanoparticles could preserve dairy products' moisture content until the end of the storage period. The cause is because these membranes' capacity to hold onto moisture is impacted by increased crosslinking caused by chemical, physical, and enzymatic treatments. This was confirmed by Bourtoom (2009) who stated that the decrease in moisture content was 19.6% for uncoated cheese samples compared to the coated cheese, which reached 23.4%.

Table 1: The impact of various treatments on the moisture content of yogurt that is both uncoated and coated with gelatin films, and is kept at a temperature of 5± 2 °C for 20 days

Treatment	1 day	10 day	20 day	Treatment mean
Control 1	78.00 hi	74.33 i	73.03 i	75.12 d
Control 2	89.30 a-c	83.73 b-g	81.76 eh	84.93 bc
Ag 20 mg	88.53 a-c	84.10 b-g	83.90 b-g	85.51 a-c
Ag 30 mg	87.63 a-e	84.46 b-g	82.33 d-h	84.81 bc
Nis 50 mg	90.95 a	87.73 a-d	85.36 a-g	88.01 a
Ag 20+50	88.66 a-c	84.93 b-g	84.26 b-g	85.95 a-c
Ag 30 +50	89.43 ab	87.40 a-e	85.53 a-f	87.45 ab
Zn 20 mg	87.06 a-e	84.76 b-g	79.60 gh	83.81 c
Zn 30 mg	87.06 a-e	81.76 e-h	79.93 f-h	82.92 c

Treatment	1 day	10 day	20 day	Treatment mean
Zn 20 +50	88.70 a-c	83.46 c-h	81.83 e-h	84.66 bc
Zn 30 + 50	88.50 a-c	85.93 a-g	79.66 gh	84.46 bc
Period mean		87.43 a	84.60 b	81.56 c

* The table numbers represent the average for duplicates.
* Significant differences at the 0.05% level are shown by the different letters in a single column
T1 = control sample T2 = Yogurt and membrane only T3 = membrane + AgNPs 20 mg T4 = membrane + AgNPs 30 mg. T5= membrane + Nis 50 mg. T6=Membrane + AgNPs 20 mg + Nis 50 mg T7=Membrane + AgNPs 30 mg + Nis 50 mg T8=Membrane + ZnNPs 20 mg T9= Membrane + ZnNPs 30 mg. T10= ZnNPs 20 mg + Nis 50 mg T11= ZnNPs 30 mg + Nis 50 mg.

pH Values

In Table (2), the results for the average storage periods indicated that the pH value differed significantly for all periods, and the highest value was at the end of the first day, when it reached 4.64, and the lowest at the finish of the time limit, when it reached. 4.38 Likewise, with regard to the average of all treatments, there was a discrepancy in the values between the treatments. The treatment T6, coated with gelatin supplemented with nanosilver and nisin, and T4 recorded the highest value, reaching 4.59, while the lowest values were for the control treatment, which was 4.23.

The results also indicate that the pH levels of yogurt samples that have had gelatin added or coated to it contain silver, zinc nanoparticles, and nisin compared to the uncoated yogurt sample at the end of the initial day of storage for treatments T1, T2, T3, T4, T5, T6, T7, T8, and T9. T10 and T11 4.67, 4.64, 4.62, 4.61, 4.65, 4.64, 4.67, 4.64, 4.62, 4.62, 4.65, respectively. The T7 treatment coated with gelatin supplemented with nanosilver and nisin at a concentration of 50 mg recorded the highest values, which did not differ significantly from the control treatment T1. This explains the synergistic role of nanosilver and nisin together in influencing the effectiveness of microorganisms because they have an important role in converting the sugar lactose into lactic acid. Thus lowering the pH value

The pH levels gradually reduced throughout the period of storage, reaching lowest levels after 20 days. which ranging from 4.59 - 4.47 for samples T4 and T3 coated with gelatin supported with nanosilver, compared to the control treatments T2 and T1, which reached 4.39 and 4.23 respectively.

Table 2: The impact of various treatments on pH levels of yogurt that is both uncoated and coated with gelatin films, and is kept at a temperature of 5± 2 °C for 20 days

Treatment 1 day	10 day	20 day	Treatment mean
Control14.67 a	4.08 g	3.93 h	4.23 d
Control24.64 ab	4.34 f	4.19 g	4.39 c
Ag 20 mg4.62 ab	4.49 ed	4.32 f	4.47 b
Ag 30 mg4.61 ab	4.59 a-d	4.56 a-d	4.59 a
Nis 50 mg 4.65 ab	4.57 a-d	4.49 c-e	4.57 a
Ag 20+50 4.64 ab	4.59 a-d	4.54 a-d	4.59 a
Ag 30 +50 4.67 a	4.59 a-d	4.33 f	4.53 ab
Zn 20 mg4.64 ab	4.56 a-d	4.37 ef	4.52 ab
Zn 30 mg4.62 a-d	4.58 a-d	4.50 b-e	4.57 a
Zn 20 +50 4.62 a-d	4.57 a-d	4.48 ed	4.55 a
Zn 30 + 50 4.65 ab	4.59 a-d	4.51 b-d	4.58 a
Period mean	4.64 a	4.50 b	4.38 c

* The table numbers represent the average for duplicates.
* Significant differences at the 0.05% level are shown by the different letters in a single column.
T1 = control sample T2 = yoghurt and membrane only T3 = membrane + AgNPs 20 mg T4 = membrane + AgNPs 30 mg. T5= membrane + Nis 50 mg. T6=Membrane + AgNPs 20 mg + Nis 50 mg T7=Membrane + AgNPs 30 mg + Nis 50 mg T8=Membrane + ZnNPs 20 mg T9= Membrane + ZnNPs 30 mg. T10= ZnNPs 20 mg + Nis 50 mg T11= ZnNPs 30 mg + Nis 50 mg.

The acidity of all yogurts treatments increased significantly, while the pH decreased significantly in yogurts during storage due to the bacteria present in the yogurt that production of lactic acid The results agreed with what was mentioned by Hoda S *et al*., (2021). The results also agreed with the findings of Al-Jubouri (2021), It was stated by someone that there was a notable fall in the pH value of the yoghurt samples. At the conclusion of the storage period, the samples coated with gelatin films containing chitosan particles and titanium nanoparticles showed significant improvements compared to the control samples that were neither coated or solely covered with gelatin.

Fat Percentage

The results in Table (3) for the average storage periods indicated that the percentage of fat recorded the maximum value at the end of the storage time, reaching 3.51%, which differed significantly from the first day after manufacturing, which amounted to 3.30%. As for the rate of all treatments, there was a discrepancy in the values between the treatments, and it was Treatment T9 coated with gelatin fortified with nano-zinc recorded the lowest value, reaching 3.30. % the lowest values were for the control treatment T1, which was 3.57%, and the rest of the values ranged between the two values. In Table (3), the results show the fat percentage of yogurt samples coated with gelatin films and added to each of AgNPs, ZnNPs and Nisin in the treatments under study T1, T2, T3, T4, T5, T6, T7, T8, T9, T10 and T11 that was kept for an extended period For 20 days, at 5 ± 2 °C 3.33, 3.33, 3.33, 3.26, 3.43, 3.20, 3.16, 3.36, 3.16, 3.26, and 3.43%, respectively. The results showed that at the end of the first day of manufacture, the fat percentage in each yogurt treatment was consistent and ranged between 3.16 to 3.43%. The reason for this slight difference in the fat percentage could be due to the difference in the percentage of moisture content between these treatments. It was also noted from the results that there was an increase in the percentage of fat in all treatments as the storage period progressed. The percentage of fat increased after 20 days to reach between 3.76-3.37% at the end of the storage period for treatments T1 and T10. The decrease in moisture content is what causes the fat percentage increase as the storage period goes on. The percentage of the other ingredients in the yogurt increases as the amount of humidity decreases.

Table 3: The impact of various treatments on fat content of yogurt that is both uncoated and coated with gelatin films, and is kept at a temperature of 5± 2 °C for 20 days

Treatment	1 day	10 day	20 day	Treatment mean
Control 1	3.33 d-g	3.63 ab	3.76 a	3.57 a
Control 2	3.33 d-g	3.50 b-e	3.53 a-d	3.45 a-d
Ag 20 mg	3.33 d-g	3.36 c-g	3.46 b-e	3.38 b-e
Ag 30 mg	3.26 e-g	3.30 d-g	3.53 a-d	3.36 c-e
Nis 50 mg	3.43 b-f	3.53 a-d	3.60 a-c	3.52 ab
Ag 20+50	3.20 g-f	3-46 b-e	3.60 a-c	3.42 b-e
Ag 30 +50	3.16 g	3.36 c-g	3.43 b-f	3.32 de
Zn 20 mg	3.36 c-g	3.46 b-e	3.46 b-e	3.43 b-e
Zn 30 mg	3.16 g	3.30 d-g	3.43 b-f	3.30 e
Zn 20 +50	3.26 e-g	3.37 c-g	3.36 c-g	3.33 de
Zn 30 + 50	3.43 b-f	3.50 b-e	3.50 b-e	3.47 a-c
Period mean	3.30 b	3.43 a	3.51 a	

*The table numbers represent the average for duplicates.

*Significant differences at the 0.05% level are shown by the different letters in a single column.

T1= control sample T2 = yoghurt and membrane only T3 = membrane + AgNPs 20 mg T4 = membrane + AgNPs 30 mg. T5= membrane + Nis 50 mg. T6=Membrane + AgNPs 20 mg + Nis 50 mg T7=Membrane + AgNPs 30 mg + Nis 50 mg T8=Membrane + ZnNPs 20 mg T9= Membrane + ZnNPs 30 mg. T10= ZnNPs 20 mg + Nis 50 mg T11= ZnNPs 30 mg + Nis 50 mg.

These results align with the discoveries made by Ahmed (2020). Analysis revealed that the presence of silver and zinc nanoparticles in gelatin films used to cover soft cheese resulted in an increase in the fat content towards the end of the cheese storage period. The research findings aligned with the arguments put forth by Hoda S *et al*., (2021). It showed that all yogurt samples' percentages of dry matter, protein, fat, and ash increased slightly but significantly over the period of storage, mainly as a result of whey interaction brought on by cold storage. El-Shibiny *et al*., (2018) reported similar results about changes in yogurt's chemical structure over storage and the decrease in moisture content during the course of the storage period may have contributed to the rise in fat content (Zheng *et al*., 2018).

Free Fatty Acids Percentage

Free fatty acid values are an indicator of the shelf life of dairy products, especially yogurt. Table (4) shows the average storage periods on the percentage of free fatty acids. They differed significantly for all periods. The highest value for the percentage of free fatty acids was at 20 days, reaching 1.31% compared to the average values on the first day, which amounted to 0.87%. As for the treatments rate, was recorded. T4 coated with gelatin reinforced with nanosilver had the lowest value, reaching 0.98%. The highest value was recorded for the control treatment T1, which was 1.50%. The rest of the values ranged between T4 and T1.

Table (4) shows the results of the comparison test with the control sample T1, which similarly show that all treatments have low fatty acid values and that at the end of the first day, no significant differences ($P<0.05$) were found between them. When the storage duration reached 10 days, there was a noticeable change in the free fatty acid value., which was at a rate of 1.00 and 1.20 for the T3 and T4 treatments coated with nanosilver only, which was less significant compared to the control treatments T1 and T2, 1.42 and 1.70, respectively .At the end of the storage period, the percentages of free fatty acids increased, reaching their highest values between 1.10-1.42 for the treatments T3 and T4, which showed significant differences with the control treatments T1 and T2, which recorded the highest percentages, reaching 531 and 1.94%.

Table 4: The impact of various treatments on free fatty acids content of yogurt that is both uncoated and coated with gelatin films, and is kept at a temperature of 5± 2 °C for 20 days

Treatment	1 day	10 day	20 day	Treatment mean
Control 1	0.87 j	1.70 b	1.94 a	1.50 a
Control 2	0.87 j	1.42 d	1.53 c	1.27 b
Ag 20 mg	0.87 j	1.20 ef	1.42 d	1.16 c
Ag 30 mg	0.85 j	1.00 i	1.10 gh	0.98 f
Nis 50 mg	0.88 j	1.01 hi	1.28 e	1.05 e
Ag 20+50	0.86 j	1.14 fg	1.21 ef	1.07 de
Ag 30 +50	0.87 j	1.08 g-I	1.12 fg	1.02 ef
Zn 20 mg	0.86 j	1.20 ef	1.28 e	1.11 cd
Zn 30 mg	0.88 j	1.08 g-i	1.15 fg	1.04 e
Zn 20 +50	0.87 j	1.15 fg	1.20 ef	1.07 de
Zn 30 + 50	0.87 j	1.12 fg	1.15 fg	1.05 e
Period mean	0.87 c	1.19 b	1.31 a	

*The table numbers represent the average for duplicates.

*Significant differences at the 0.05% level are shown by the different letters in a single column.

T1 = control sample T2 = yoghurt and membrane only T3 = membrane + AgNPs 20 mg T4 = membrane + AgNPs 30 mg. T5= membrane + Nis 50 mg. T6=Membrane + AgNPs 20 mg + Nis 50 mg T7=Membrane + AgNPs 30 mg + Nis 50 mg T8=Membrane + ZnNPs 20 mg T9= Membrane + ZnNPs 30 mg. T10= ZnNPs 20 mg + Nis 50 mg T11= ZnNPs 30 mg + Nis 50 mg.

The results mentioned above agreed with the results presented by Hamed *et al*., (2019), who reported that while creating functional yogurt improved with fish oil and olive oil, the percentage of free fatty acids gradually increases with a period of storage., as (yogurt whey protein) provides protection around polyunsaturated fatty acids and acts as a barrier that reduces Its susceptibility to oxidation, and the results were consistent with what was found by Soleimani *et al*., (2018) When comparing the xanthan gum-coated cheddar cheese samples to the uncoated control sample, there was less of an increase in the percentages of free fatty acids. The contribution of zinc and silver nanoparticles as well as the nisin component is the cause of the slight rise in the percentage of fatty acids in this, which inhibit microorganisms contaminating the samples and thus reduce the decomposition of its components, including fatty acids, compared to the two control treatments that recorded the highest decomposition values due to microbial contamination of those samples.

Ash Percentage

The results of the ash percentage in yogurt samples stored for 20 days at 5 ± 2°C are shown in Table (5). The ash percentage at the end of the storage period recorded the greatest value, according to the findings of the ash percentages during the storage periods, reaching 0.78%, which differed significantly from the average for the first day was 0.66%. As for the treatment rate, treatment T6, coated with gelatin enhanced with nanosilver at a concentration of 20 mg and nisin, recorded the highest ash percentage, reaching 0.76%. The lowest values were for treatment T3, reaching 0.67%, and the rest of the values ranged between the two values, which differed significantly. For the control treatments T1 and T2, the ash percentage was 0.65% and 0.68%, respectively.

There is a slight difference in the ash percentage for the different treatments, as the treatments T1, T2, T3, T4, T5, T6, T7, T8, T9, T10 and T11 at the end of the first day were 0.60, 0.64, 0.62, 0.67, 0.65, 0.72, 0.62, 0.63, 0.66, 0.68, 0.72%, respectively. These results converged with the findings of Lynch *et al*., (1999) in that there was a difference in the ash percentage depending on the treatments, which could be attributed to the difference in the location and level of the samples taken or any other factors that could have an impact on the accuracy of the readings. In addition, the data showed that on the tenth day, there were significant differences within all treatments. A gradual increase in the ash percentage of yogurt samples was observed with increasing storage duration in the treatments T1, T2, T3, T4, T5, T6, T7, T8, T9, T10 and T11, as the highest percentage of ash reached 0.70, 0.74, 0.76, 0.81, 0.75, 0.80, 0.83, 0.76, 0.80, 0.81, 0.86%. straight. The highest percentage of ash was for treatment T7, which was 0.83%, while the lowest percentage was for treatment T5, which was 0.75%, which differed significantly from the control treatments T2 and T2. T1

Table 5: The impact of various treatments on Ash content of yogurt that is both uncoated and coated with gelatin films, and is kept at a temperature of 5± 2 °C for 20 days

Treatment	1 day	10 day	20 day	Treatment mean
Control 1	0.60 n	0.64 k-n	0.70 f-j	0.65 f
Control 2	0.64 k-n	0.66 j-n	0.74 e-h	0.68 ef
Ag 20 mg	0.62 l-n	0.64 k-n	0.76 b-e	0.67 ef
Ag 30 mg	0.67 j-m	0.73 e-I	0.81 abc	0.73 bc
Nis 50 mg	0.65 j-n	0.69 g-k	0.75 c-f	0.69 de
Ag 20+50	0.72 e-i	0.75 c-f	0.80 a-d	0.76 ab
Ag 30 +50	0.62 l-n	0.65 j-n	0.83 a	0.70 de
Zn 20 mg	0.63 k-n	0.64 k-n	0.76 b-e	0.68 ef
Zn 30 mg	0.66 j-n	0.68 h-l	0.80 a-d	0.71 cd
Zn 20 +50	0.68 h-l	0.72 e-i	0.81 ab	0.74 bc
Zn 30 + 50	0.72 e-i	0.73 e-i	0.86 a	0.77 a
Period mean	0.66 c	0.68 b	0.78 a	

* The table numbers represent the average for duplicates.
* Significant differences at the 0.05% level are shown by the different letters in a single column.
T1 = control sample T2 = yoghurt and membrane only T3 = membrane + AgNPs 20 mg T4 = membrane + AgNPs 30 mg. T5= membrane + Nis 50 mg. T6=Membrane + AgNPs 20 mg + Nis 50 mg T7=Membrane + AgNPs 30 mg + Nis 50 mg T8=Membrane + ZnNPs 20 mg T9= Membrane + ZnNPs 30 mg. T10= ZnNPs 20 mg + Nis 50 mg T11= ZnNPs 30 mg + Nis 50 mg.

These results were consistent with what reported Hoda S *et al*., (2021). When compared to the control yogurt (0.89%), the ash level of the yogurt treatments fortified with (40 and 60 mg/L milk) biosynthetic ZnO-NPs (1.09% and 1.18%) was significantly higher. Zinc is mostly attached to caseins and is not lost in the whey, according to prior studies by Kahraman and Ustunol (2012), which is compatible with the high ash content of yogurt treated with ZnO-NPs. The result was also consistent with what Dimitrelloo *et al*., (2015) reported about the increase in the ash percentage. As the storage period progresses, he pointed out that the treatments' decreased moisture content is what caused the ash content to rise during the storage time. It is clear from the results that the gelatin films covering the cheese contributed significantly to preserving the chemical composition of the cheese in terms of moisture, fat percentage and ash, which had slight gradual differences between them until the storage period's end, mainly affected by the barrier properties of the films, the most important of which is water vapor permeability.

CONCLUSION

The results showed the effect of zinc and silver nanoparticles with the nisin compound loaded on a gelatin membrane on the values of chemical tests for yoghurt samples. There was a clear decrease in moisture and pH levels for all parameters as the storage period increased, with a continuous increase in the levels of fat, free fatty acids and ash. The results also showed the importance of using nanotechnology in various fields of food and its applications, as it is one of the important and promising technologies in this field, and the importance of researching the preparation of various edible coatings loaded with nutritional supplements to harmonize with functional foods that provide health and therapeutic benefits to consumers, and studying the effect of edible coatings on their health and comparing them Its impact on the impact of industrial packaging materials and thus heading towards as safe foods and environmentally friendly packaging materials for the consumer.

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