

# Development and Characterization of Gelatin Films Enriched With Olive Leaves Extract for Fresh Apple Packaging

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**Annotation:** To minimize microbial growth, food preservation must employ a variety of physical and chemical techniques. As an alternative to traditional synthetic materials that can be harmful to the consumer's health, the packaging materials could be derived from biological sources, such plants. The study was aimed to development and characterization of gelatin films enriched with olive leaves extract for fresh Apple packaging. In October 2024, olive leaves were gathered from olive trees in Kirkuk, Iraq. After being gathered, the leaves were cleaned with tap water and allowed to dry for 30 days at room temperature (between 25 and 30 °C). Distilled water was used to dissolve the gelatin powder at 60 °C and a 3% (w/v) concentration. The leftover extracts from olive leaves were then dissolved at 5, 10, and 20 µg/ml in the gelatin film-forming solution. The findings showed that the highest percentage of thickness was in the 8% olive leaves treatment in the first week of apple storage ( $0.21 \pm 0.04$ ), and even after the fourth week of treatment and for the 8%

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olive leaves film ( $0.18 \pm 0.04$ ) compared to the control treatment and the rest of the treatments. gelatin films containing olive leaves had tensile strengths ranging from  $55.52 \pm 3.89$  in T1 at the first week to  $23.11 \pm 5.12$  in T3 at the fourth week MPa. In terms of elongation rate, it was  $5.37 \pm 0.31$  in T3 during the first week in the extract-supported film, which was more than the elongation rate of control treatment  $4.21 \pm 0.09$ . In every week and transaction, it outperformed the control group. the edible film made with gelatin without olive leaves has a significantly ( $P < 0.05$ ) different WVT than the edible film made with olive leaves at varying concentrations. It appears that the highest percentage of WVT was in the control treatment in the first week of apple storage ( $0.08 \pm 0.005$ ), and even after the fourth week of treatment, 8% olive leaves film ( $0.05 \pm 0.003$ ) less than control treatment and the rest of the treatments. It is concluded that adding olive leaves extract to gelatin Edible Films led to enhancing the chemical and mechanical properties of the edible film.

**Keywords:** Gelatin, Edible films, Olive, Apple packaging.

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## Introduction

The development of biodegradable and packaging films to replace conventional plastics has garnered significant interest from the food industry in recent years due to the public's growing concern about environmental pollution and food safety and quality [1,2]. Food, packaging, and the internal and external environments can interact through active packaging to enhance sensory qualities and food safety while guaranteeing food quality [3,4]. Traditional food packaging primarily focuses on barrier capabilities, but active food packaging that performs better in terms of antioxidant and anti-ultraviolet properties has garnered a lot of interest [5,6]. The shelf life of fresh-cut fruits can be extended and postharvest quality maintained with the help of this active packaging material [7]. While olive fruits and olive oil are vital parts of a healthy daily diet for a large portion of the world's population, scientists and companies in the food, packaging, and supplementation industries are interested in olive leaves because of their high content of polyphenolic compounds [8]. Olive leaves are becoming more and more popular as tea and dietary supplements due to their believed antibacterial, antifungal, antioxidant, hypoglycemic, anti-hypertensive, hypocholesterolemia, and anti-inflammatory properties [9, 10]. The use of olive leaf extract in biodegradable polymers is a significant component of its use in packaging materials. Given the global problem of garbage accumulation and the challenges of recycling, this is especially important. Researchers are working to find ways to lessen the negative environmental effects of plastic trash, and one of their top priorities is the creation of eco-friendly packaging materials. This article highlights the potential of plant-derived compounds as natural preservatives with antioxidant and antibacterial capabilities by discussing various

commercial packaging modified using olive leaf extract. These changes show that it is feasible to use plant-based extracts to improve the functionality and sustainability of packaging materials [11,12,13]. Although casein addition [15] could result in a stiffer gelatin film, the addition of olive oil [14] to the gelatin matrix provides it with exceptional light and moisture barrier qualities. The antimicrobial chemicals are added to gelatin films to improve their qualities. After adding ethanol-propolis extract and/or essential oils, respectively, numerous studies have shown that gelatin films have an inhibitory effect on a variety of food spoilage microorganisms [16,17].

So, the study was aimed to development and characterization of gelatin films enriched with olive leaves extract for fresh Apple packaging.

## **Materials & methods**

### **Olive leaves extract**

In October 2024, olive leaves were gathered from olive trees in Kirkuk, Iraq. After being gathered, the leaves were cleaned with tap water and allowed to dry for 30 days at room temperature (between 25 and 30 °C). After that, the leaves were pulverized into a fine powder (0.1 to 0.6 mm). Then, in a beaker, 100 mL of ethanol and 10 g of powdered olive leaves were mixed. Sonication was applied to the combination for two hours while the solvent surface remained at room temperature. Following that, the leftovers underwent another extraction process after being dissolved in ethanol. A rotary evaporator set at 50 °C was used to concentrate the resultant extracts after they had been filtered using vacuum filtration and Whatman filter paper. A freeze-dryer was then used to dry the crude extract. OLE's extraction yield was approximately 9.8% [18, 19].

### **Preparation of gelatin-based films**

Distilled water was used to dissolve the gelatin powder at 60 °C and a 3% (w/v) concentration. The leftover extracts from olive leaves were then dissolved at 5, 10, and 20 µg/ml in the gelatin film-forming solution. The gelatin film-forming solution was plasticized with 15% v/v glycerol. Following that, 25 milliliters of each gelatin film-forming solution were poured onto 13.5-cm-diameter polystyrene Petri dishes. These dishes were then dried for 48 hours at 25 °C and 50% relative humidity (RH) in a vented climatic chamber (Binder, Tuttlingen, Germany). Four varieties of gelatin-based films were ultimately produced: CF: olive leaf film at 2%, 4%, and 8%; control gelatin-based film.

## **Characterization of films**

### **Film thickness**

The film's thickness was measured using a digital thickness gauge. Each film sample was measured four times at various locations. When calculating and considering mechanical properties, the mean value was utilized.

### **Mechanical properties**

A texture analyzer (Godalming, UK) was used to assess the tensile strength (TS, MPa) and elongation (%) of film samples in compliance with standard method ISO 527-3. A standardized precision cutter (USA) was used to size rectangular film samples (2.5 × 8 cm) in order to produce a tensile test piece with exact and parallel sides along the length. After that, the test apparatus's extension grips were used to stretch the film samples uniaxially at a cross-head speed of 50 mm/min until they broke. The associated stress-strain curves were employed to calculate the maximum load and the ultimate extension at break, which were then utilized to calculate TS and EAB. The measurements were made at room temperature with a relative humidity of 40 ± 5%. For every formulation, five samples were examined.

### **Water vapor permeability (WVP)**

The approach outlined by Sobral and Habitante [20] was used to determine WVP. Cells were

placed in desiccators with distilled water at 22 °C after films were adhered to the cell entrance (permeation area = 15.9 cm<sup>2</sup>) containing silica gel. After that, it was weighed every hour for eight hours. WVP testing was conducted on three films. The movie's WVP was determined using Eq.

$$WVP (g\ m - 1s - 1Pa - 1) = w \times l \times A - 1 \times t - 1 \times (P_2 - P_1) - 1$$

where *l* is the thickness of the film (m) and *w* is the cup's weight gain (g); *A* represents the film's exposed area (m<sup>2</sup>), and *t* represents the gain time (s). The vapor pressure differential throughout the film is (*P*<sub>2</sub>–*P*<sub>1</sub>).

### Statistical analysis

A one-way analysis of variance (ANOVA) was performed using the statistical package for the social sciences (SPSS) software for Windows™ (Chicago, IL, USA). Duncan's multiple range test was used to compare the measured parameters for different gelatin-based films. Statistical significance was established for the mean differences at the 95% (*p* < 0.05) confidence level.

## Results & Discussion

### Thickness of films

According to the edible film's thickness value (Table 1), the edible film made with gelatin without olive leaves has a significantly (*P*<0.05) different thickness than the edible film made with olive leaves at varying concentrations. Every recipe in this study has edible film that satisfies the Japanese Industrial Standard for thickness (0.25 mm). It appears that the highest percentage of thickness was in the 8% olive leaves treatment in the first week of apple storage (0.21±0.04), and even after the fourth week of treatment and for the 8% olive leaves film (0.18±0.04) compared to the control treatment and the rest of the treatments.

**Table 1: the thickness of gelatin films at different concentrations of olive leaves**

Properties	Treatments	Storage				
		W1	W2	W3	W4	<i>P</i> value
Thickness (μm)	Control	0.13±0.01 c	0.12±0.04 c	0.11±0.07 c	0.10±0.09 d	0.001
	T1	0.18±0.05 b	0.17±0.03 b	0.15±0.09 b	0.13±0.05 c	
	T2	0.19±0.03 b	0.18±0.03 b	0.16±0.06 b	0.15±0.01 b	
	T3	0.21±0.04 a	0.21±0.08 a	0.2±0.04 a	0.18±0.04 a	

W= week, different letters mean there is significant (*P*<0.05) different between groups in same week.

The thickness of gelatin-based films enhanced with varying concentrations of extracts from olive leaves is displayed in Table 1. The thickness of gelatin-based films is significantly affected (*p*<0.05) by the addition of extracts from olive leaves. However, by adding relatively large amounts (1 to 5% w/w, on protein basis) of phenolic chemicals, Le et al. [21] showed an increase in the thickness values of the horse mackerel gelatin films. In addition, these authors found that, when compared to the other phenolic substances that were evaluated, including catechin, gallic acid, caffeic acid, and ferulic acid, rutin was the most effective at increasing thickness. However, Nagarajan et al. [22] found that adding ethanolic extract from coconut husk (0.1 to 0.4% w/w) increased the thickness of tilapia skin gelatin-based films and decreased their tensile strength. According to these authors, when phenolics levels are high, hydrogen connections between phenolic functional groups and gelatin molecules can form, changing the integrity and length of the chain.

### Mechanical tests

In Table 2, gelatin films containing olive leaves had tensile strengths ranging from  $55.52 \pm 3.89$  in T1 at the first week to  $23.11 \pm 5.12$  in T3 at the fourth week MPa. In terms of elongation rate, it was  $5.37 \pm 0.31$  in T3 during the first week in the extract-supported film, which was more than the elongation rate of control treatment  $4.21 \pm 0.09$ . In every week and transaction, it outperformed the control group.

**Table 2: Mechanical tests of gelatin films at different concentrations of olive leaves**

Properties	Treatments	Storage				P value
		W1	W2	W3	W4	
<b>Mechanical: Tensile strength (MPa )</b>	Control	$61.8 \pm 5.14$ a	$58.95 \pm 4.45$ a	$55.31 \pm 4.25$ a	$52.94 \pm 3.14$ a	0.001
	T1	$55.52 \pm 3.89$ b	$52.11 \pm 6.34$ a	$50.85 \pm 3.31$ a	$48.31 \pm 5.79$ a	
	T2	$39.72 \pm 4.31$ c	$32.64 \pm 2.11$ b	$29.37 \pm 2.73$ b	$27.61 \pm 3.17$ b	
	T3	$31.31 \pm 7.21$ c	$27.31 \pm 4.04$ b	$25.41 \pm 4.63$ b	$23.11 \pm 5.12$ b	
<b>Elongation %</b>	Control	$4.21 \pm 0.09$ c	$4.01 \pm 0.12$ c	$3.41 \pm 0.13$ c	$3.05 \pm 0.17$ c	0.001
	T1	$4.56 \pm 0.11$ b	$4.25 \pm 0.19$ bc	$3.63 \pm 0.15$ c	$3.13 \pm 0.11$ c	
	T2	$4.71 \pm 0.15$ b	$4.39 \pm 0.07$ b	$4.03 \pm 0.13$ b	$3.52 \pm 0.17$ b	
	T3	$5.37 \pm 0.31$ a	$5.03 \pm 0.15$ a	$4.84 \pm 0.22$ a	$4.15 \pm 0.21$ a	

W= week, different letters mean there is significant ( $P < 0.05$ ) different between groups in same week.

One of the crucial aspects of a packing film that must protect the product's integrity from outside forces is its mechanical qualities [23]. The tensile strength (TS) and elongation findings of gelatin-based films enhanced with varying concentrations of olive leaves are displayed in Table 2. The enhanced films' mechanical properties showed a significant change ( $p < 0.05$ ). But according to Li et al. [24], adding natural antioxidants to the film significantly reduced its TS. According to these scientists, polyphenolic chemicals may decrease the protein–protein connections that maintain the protein network by forming covalent and hydrogen bonds with the amino and hydroxyl groups of the polypeptide in gelatin. However, it was noted that adding certain plant extracts raised the elongation values of gelatin-based films. This was explained by particular interactions between phenolic compounds and polypeptides. Indeed, the establishment of covalent cross-links may result in the creation of matrices that are more flexible and cohesive [25,26].

### Water vapor transmission (WVT)

According to the edible film's WVT value (Table 3), the edible film made with gelatin without olive leaves has a significantly ( $P < 0.05$ ) different WVT than the edible film made with olive leaves at varying concentrations. It appears that the highest percentage of WVT was in the control treatment in the first week of apple storage ( $0.08 \pm 0.005$ ), and even after the fourth week of treatment, 8% olive leaves film ( $0.05 \pm 0.003$ ) less than control treatment and the rest of the treatments.

**Table 3: Water vapor transmission of gelatin films at different concentrations of olive leaves**

Properties	Treatments	Storage				
		W1	W2	W3	W4	P value
Water vapor transmission (WVT) %	Control	0.08±0.005 a	0.08±0.004 a	0.07±0.003 a	0.07±0.006 a	0.001
	T1	0.08±0.005 b	0.07±0.004 b	0.07±0.004 a	0.06±0.005 ab	
	T2	0.07±0.003 b	0.07±0.003 b	0.06±0.007 b	0.06±0.008 ab	
	T3	0.07±0.006 a	0.06±0.008 c	0.06±0.004 b	0.05±0.003 b	

W= week, different letters mean there is significant ( $P<0.05$ ) different between groups in same week.

Additionally, Table 3 demonstrates that the WVP of gelatin-based films significantly dropped ( $p < 0.05$ ), primarily upon the addition of olive leaf extract in comparison to the control film. In actuality, films supplemented with the highest proportion of olive leaves had the lowest WVP. Similar findings were made by Wu et al. [27], who found that adding a green tea extract to the silver carp gelatin film reduced their WVP in comparison to the control film. Through hydrogen bonding and hydrophobic interactions, the phenolic compounds were able to incorporate into the network of gelatin molecules, influencing the secondary structure of the protein [28]. A denser and less permeable film structure was the outcome of a high degree of interactions [27]. Nonetheless, it was found that the WVP of gelatin films was raised by extremely high levels of enrichment with ethanolic extract from coconut husk (0.05–0.4% w/w) [29]. A non-uniform film matrix with numerous micropores could be the result of coagulation brought on by numerous interactions between gelatin molecules and phenolic chemicals. As a result, there was an increase in water migration across the films [22].

## Conclusions

It is concluded from the current study that adding olive leaves extract to gelatin Edible Films led to enhancing the chemical and mechanical properties of the edible film due to its properties and active compounds that contributed to improving the properties of gelatin Edible Film.

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