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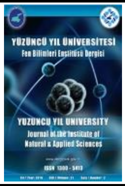

Research Article

Physicochemical and Mechanical Properties of Sodium Alginate Films Containing Thyme Essential Oil

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Abstract: In this study, it was aimed to develop edible film by the casting technique from sodium alginate (SA) (%2 w/v) using three different concentrations of thyme essential oil (TEO) (0.5%, 1%, 1.5% v/v). The physicochemical, mechanical, structural, and antioxidant properties of enriched SA films were also characterized. The highest film solubility (FS) was found as 91.78% in SA (control) films. The mechanical properties of films were found between 27.14 MPa and 50.06 MPa for tensile strength (TS) and between 13.88% and 32.02% for elongation at break (EAB). The highest total phenolic content (TPC) and antioxidant activity were found as 382.19 mg GAE/kg and 16.20% in SA film incorporated with TEO 1.5% respectively. As the concentration of TEO increased, the *L** value increased, leading the film colors to approach white. Additionally, it was observed that as the *b** value increased the film colors tented towards yellow. When the results of SEM analysis were evaluated, it was found that SA films were more homogeneous than other films. In general, it was concluded that TEO at a concentration of %1 (v/v) improves the physical and chemical properties of sodium alginate film compared to other groups.

Kekik Esansiyel Yağı İçeren Sodyum Aljinat Filmlerinin Fizikokimyasal ve Mekanik Özellikleri

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Anahtar Kelimeler
Film karakterizasyonu,
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Öz: Bu çalışmada dökme yöntemikullanılarak üç farklı konsantrasyonda kekik esansiyel yağı (KY) (%0.5, %1, %1.5 h/h) katkılı sodyum aljinat (%2 a/h) (SA) filmlerin yenilebilir film olarak geliştirilmesi amaçlanmıştır. Zenginleştirilmiş sodyum aljinat filmlerin fizikokimyasal, mekaniksel, yapısal ve antioksidan özellikleri karakterize edilmiştir. En yüksek film çözünürlüğü %91.78 olarak SA (kontrol) filmlerde bulunmuştur. Filmlerin kopma mukavemeti 27.14 MPa ve 50.06 MPa arasında, kopma anında uzama değerleri ise 13.88% ve 32.02% arasında bulunmuştur. En yüksek toplam fenolik içerik ve antioksidan aktivite sırasıyla %1.5 KY içeren SA filminde 382.19 mg GAE/kg ve %16.20 olarak bulunmuştur. KY'nin artan konsantrasyonlarına bağlı olarak *L** değeri arttı ve film renkleri beyaza yaklaştı. Ayrıca *b** değerinin artması ile film renkleri sarıya yaklaştığı görülmüştür.SEM analiz sonuçları değerlendirildiğinde SA filmlerin diğer filmlere göre daha homojen olduğu tespit edilmiştir. Genel olarak %1 (h/h) konsantrasyonunda KY'nin sodyum aljinat filminin fiziksel ve kimyasal özelliklerini diğer gruplara kıyasla daha iyi geliştirdiği sonucuna varılmıştır.

1. Introduction

Food packaging technology has been used for many years to prevent food from being affected by adverse environmental conditions and to transport them to consumers while preserving their quality. Recently, the development of coating technology has been a new direction in food technology in relation to film-forming materials (Mahcene et al., 2021).

Biodegradable edible films and coatings are one of the methods used to maintain quality, extend shelf life, contribute to the efficiency of packaging materials as an alternative to traditional petroleum-based food packaging for optimizing food quality (Aguirre-Joya et al., 2018). Edible films and coatings prepared with proteins, polysaccharides and lipids have become increasingly important in food preservation applications (Okcu et al., 2018). Edible films and coatings are materials obtained from natural sources (polysaccharides, proteins, lipids, enzymes, bacteriocins and other biologically edible components) that can be used together with food by applying them to the surface or inside of food in a thin layer (Dehghani et al., 2018; Bhagath & Manjula, 2019). They can also be prepared by combining these components with each other and using them. These natural components used to ensure that the film or coatings have different properties. Because the chemical structures of these components differ from each other. In general, polysaccharides are used to control the diffusion of oxygen and other gases, proteins are used to give mechanical strength to films, and lipids are used to reduce water transfer (Pavlath & Orts, 2009; Umaraw & Verma, 2015; Kurek et al., 2017; Okcu et al., 2018; Ulusoy et al., 2018).

The most commonly used natural polymers (such as starch/modified starch, chitin/chitosan, pectin, galactomannans, cellulose/modified cellulose, alginate, carrageenan, gums (gelatin, xanthan, etc.), and pullulan) in the formulation of edible packaging include polysaccharides (Aguirre-Joya et al., 2018). Polysaccharides are abundant in nature, non-toxic, cost-effective, can act as stabilizers, are often edible, easily accessible, and possess numerous hydrogen bonds and hydroxyl groups. These properties support film formation, making them widely used in various applications (Pop et al., 2019; Mohamed et al., 2020).

Alginate, found in the outer cell wall of brown algae, is an anionic polysaccharide. Sodium alginate, known for its physical properties, is non-toxic and environmentally stable. It can gel, create films, and interact with various molecules (Gaowa et al., 2023). Alginates have been used for many years in the food, beverage, pharmaceutical, textile, printing and pharmaceutical industries as a thickener, stabilizer, emulsifier, chelating and encapsulating agent, colloidal stabilizer, gels, films and membrane. This polymer is low-cost, biocompatible and biodegradable (Gokbulut & Ozturk, 2018; Senturk Parreidt et al., 2018).

Alginate-based edible films and coatings are used in various applications including fruits, vegetables, red and white poultry meats, seafood, cheeses, etc. It provides reduction of dehydration, control of respiration, improvement of the appearance of the product, improvement of its mechanical properties, preservation of its qualities and increase of its shelf life. The most commonly used alginate salt in films or coatings is sodium alginate. Sodium alginate is included in the GRAS (generally safe) category by the FDA (Food and Drug Administration) (Senturk Parreidt et al., 2018; Anonymous, 2021).

Essential oils are added to the film forming solutions for various purposes, such as improving the chemical and physical structure of films, observing microbial activity. In this way, it is ensured that the prepared films have the desired properties (such as low water vapor permeability, high elongation at break, high antioxidant activity). Thyme essential oil consists of over 60 components, the majority of which possess significant antioxidant and antimicrobial properties (Sarıcaoglu & Turhan, 2020).The use of thyme essential oil in edible film and coating applications prepared from many different sources is available in literature studies (Emiroğlu et al., 2010; Kavoosi et al., 2013; Sarengaowa et al., 2018; Mahcene et al., 2019; Tabassum & Khan, 2020; Sarıcaoglu & Turhan, 2020; Chen et al., 2021; Shakerardekani et al., 2021; Venturini Antunes et al., 2023). However, literature information on the characterization analyses of sodium alginate films with thyme essential oil added at different concentrations is limited.

The main objective of this study was to develop sodium alginate (SA) films enriched with thyme essantial oils (TEO) at different ratios and evaluate the effects of TEO incorporation SA based films. Characterization analyses of the films (solubility, thickness, light transmittance, elongation at break and tensile strength, color, total amount of phenolic substance, antioxidant activity and scanning electron microscope imaging (SEM) were performed in each prepared film group. This study is expected to serve as an example for characterization analyses of sodium alginate films with added thyme oil at concentrations of 0.5%, 1%, and 1.5% for future studies. The study aims to determine the optimal concentration of thyme essential oil, making the film formulation solution suitable as a coating material for a specific food product.

2. Materials and Methods

SA (Sigma-Aldrich W201502), glycerol (Sigma-Aldrich 104092), Tween 80 (Sigma-Aldrich 9005-65-6), were used for film production. TEO (*Thymus vulgaris*) was supplied from the shop that sells spices (Doğan Baharatçılık, Turkey). Ultra-pure distilled water was used for analyses. All other chemicals were supplied from Merck (Germany)

2.1. Film preparation

The films were prepared by modifying the method used by Peretto et al. (2014). The ratios of SA, glycerol, and Tween 80 used in the production of films were determined through preliminary trials. The TEO ratios used in the production of the film were prepared based on the study conducted by Tabassum & Khan (2020), with changes according to the results of the preliminary trial. SA solution (2%, w/v) was dissolved in distilled water at 70 ℃ for 15 min with a magnetic stirrer until homogeneity. Glycerol 2% (v/v) was added to the solution at the same temperature as a plasticizer and mixed for another 15 min. When the film forming solution cooled to 40℃, 0.15% (v/v) Tween 80 was added and mixed. To prevent TEO from being affected by temperature, the solution was cooled to room temperature (20-25 ℃). 0%, 0.5%, 1% and 1.5% (v/v) TEO were added to the solution at the desired concentrations and mixed for a while. The prepared solutions were homogenized with ultra-turrax by mixing at 13400 rpm for 4 min (Peng & Li, 2014). After the procedure, the foaming was removed by slightly shaking the solutions for a while. The film forming solutions were poured into petri dishes, 25 mL each, and left to dry at room temperature uncovered. The dried films were kept in a desiccator at room temperature until analysis.

2.2. Film characterizations

2.2.1. Film solubility and thickness

The film solubility (%) and thickness (mm) of the films were determined by using the method of Ahmad et al. (2012). The film samples were prepared by cutting 2 x 3 cm^2 in size and placed in petri dishes and left for 7 days in the desiccator. The film samples were mixed with 80 mL of distilled water at room temperature for 24 h. The mixture was filtered through filter paper. The remaining particules of solid film were held in the oven for 24 h at 105℃. After drying, the film samples were weighed and the film solubility values were determined by taking into account the ratios of the initial and final weights of the film samples. The film's thickness was measured using a digital micrometer (Mitutoyo Digimatic Micrometer Absolute Digimatic 2, Japan) for 10 randomly selected points.

2.2.2. Tensile strength and elongation at break values

According to the standard ASTM method D 882-88 (2001) the tensile strength (TS, MPa) and elongation at break (EAB, %) values of films were determined using a TA. XT-plus Texture Analyser (Stable Micro Systems, UK). The films with measured thicknesses were cut into dimensions of 7 × 2 cm^2 and stored in a desiccator at a relative humidity of 50 ± 5% for 48 hours at 25 ± 0.5 °C. TS and EAB values of films were detected under 5 kg load with 50 mm/min speed. At least three replications were conducted per experiment.

2.2.3. Color, light transmission and opacity of the films

The color values of the films (CIE L^*, a^*, and b^*) were determined according to the method used by Özdestan & Uren (2009) using Hunter colorimetry (Colorfex HunterLab, USA). After the colorimetry was calibrated with black and white plates the color measurement of the films was applied in daylight. The sample chamber was closed with a white plate and applied 10 times. According to the method applied by Peng & Li (2014), the light transmittance and opacity values were determined in the film samples. The film samples cut into 1 x 4 cm2 sizes were placed in a quartz cuvette and the transmittance values of the film sample were measured at wavelengths of 200-800 nm using a UV-Vis spectrophotometer (Agilent Technologies Cary 60 UV–Vis, USA). The opacity value of the film samples was calculated by the ratio of the absorbance at 600 nm to the thickness.

2.2.4. Total phenolic content of the films

When determining the total phenolic content (TPC) of film samples, modifications were made to the method originally used by Xu & Chang (2007). The sample extraction process was carried out according to the method applied by Boulekbache-Makhlouf et al. (2013). 3 mL of pure water, 0.25 mL of Folin-Ciocalteu reagent and 0.75 mL of 7% (w/v) Na^2CO^3 were added to 50 µL of extract taken from film samples and mixed. And it was kept at room temperature for 8 min. Then 0.95 mL of pure water was added to them and kept in the dark for 2 h. Absorbances were measured at 760 nm by UV–Vis spectrophotometer and the measurement results were given as gallic acid equivalent (µg of GAE/g sample).

2.2.5. Antioxidant activity of the films

Some modifications have been made to the DPPH method used for analyzing the antioxidant activity of films by Brand-Williams et al. (1995). The sample extraction process was carried out according to the method applied by Boulekbache-Makhlouf et al. (2013). 1.98 mL of DPPH (60 µM) solution was mixed with 30 µL of the extract taken from film samples. Absorbance value of the solution was determined with UV–Vis spectrophotometer at 515 nm The inhibition (%) was calculated as follows (1);

$$inhibition\% = \frac{(Absorbance_{control}) - (Absorbance_{sample})}{(Absorbance_{control})} * 100 \qquad (1)$$

2.2.6. Scanning electron microscopy analysis

The surface morphology of the films was imaged by scanning electron microscopy (SEM) (Thermo Scientifc ApreoS, ABD) using at 5000× magnification.

2.2.7. Statistical analysis

SPSS Statistics 25.0 was used for statistical analysis of the experimental data. One-way analysis of variance (ANOVA) and Duncan's multiple interval tests were performed to evaluate the confidence of the scale at 95% ($P \leq 0.05$). The analyses were conducted in at least 3 parallels and 3 replicates.

3. Results and Discussion

3.1. Thickness of the films

The thickness of the films directly affects the film solubility, tensile strength, elongation at break and barrier properties of the films. Therefore, the fact that the film thickness is an important parameter contributes to its selection as a packaging material (Gutiérrez et al., 2015; Khodaei et al., 2020). Table 1 shows the average thickness values of SA and TEO/SA films. It has been shown that the pure SA (control) film thickness has the lowest value (0.087 mm) among all film formulations (Table 1). It is observed that the film thicknesses increased due to the increase in the concentration of thyme oil added to the film forming solution, and the sample with the highest thickness value was SA/TEO 1.5% (0.231 mm) film. In the study conducted by Jouki et al. (2014), films prepared from quince seed mucilage with added thyme essential oil at concentrations of 0%, 1%, 1.5%, and 2% (v/v) showed thicknesses of 0.063 mm for the control film. As the concentration of thyme essential oil increased, the thicknesses of the films also increased to 0.066 mm, 0.070 mm, and 0.076 mm, respectively. Parallel results were obtained to the findings of this study. The lipid phase in essential oils can affect the sequence of the polymer chain in such a way as to cause a loose and irregular structure, and it has been reported that this can lead to an increase in film thickness (Tongnuanchan et al., 2012).

3.2. Mechanical properties of the films

While TS gives information about the strength of the films against breaking, EAB gives information about the stretching capacity of the films before breaking (Liu et al., 2020) Table 1 shows the TS and EAB values of the developed films. According to the results, the TEO concentration used during film preparation had a remarkable effect on the TS and EAB values of SA based films. It was found that the significant differences were determined in TS values between TEO added films and SA (control) films ($P<0.05$). While the TS value of SA films was high at 50.06 MPa, the TS value of the films decreased from 37.29 to 27.14 MPa with the addition of 1.5% TEO. The essential oils added to the film forming solutions cause the degradation of the film matrix network and therefore a decrease in the TS values occurs (Hosseini et al., 2009). It has been observed that the EAB values increase with the increase of TEO concentration. The group with the highest EAB value was SA/TEO 1.5% film (32.02 %). In the study by Abdel Aziz et al. (2018), film formulation solutions were prepared using 1% (w/v) sodium alginate with different concentrations of cumin oil (0%, 1%, 2%, and 3%). For the control (SA) films, the tensile strength was measured at 17.35 MPa. At a concentration of 1% castor oil, this value increased to 48.15 MPa, but it decreased to an average of 30 MPa at concentrations of 2% and 3%. In the control (sodium alginate) films, the elongation at break was measured at 10.04%, and this value increased up to 15.86% with increasing concentrations of castor oil. Parallel results were obtained to the findings of this study. With the addition of TEO to the films, the EAB values increased and, depending on this situation, the films became more elastic and extensible. By adding essential oils to the film formulation, a heterogeneous structure is formed with matrix networks that do not show stability. This leads to an increase in the EAB values of the films and a decrease in the TS values (Benavides et al., 2012; Shojaee-Aliabadi et al., 2013; Moey et al., 2018).

3.3. Film solubility

The FS, which is an important feature of edible films, is one of the characteristics should be low in order to increase the integrity and water resistance of food in potential food applications (Ghasemlou et al., 2011). Table 1 shows the FS levels for SA (control) and SA/TEO films prepared at different TEO concentrations. The SA film prepared was very soluble in water (91.78%). A decrease in FS values occurred due to the addition of TEO to the film matrix. There was a 17% decrease in FS values compared to the SA/TEO 0.5% film with the SA (control) film; a 24% decrease in FS values compared to the SA/TEO 1% film with the SA (control) film and a 27% decrease compared to the SA/TEO 1.5% film with the SA (control) film. There was no significant difference between the FS values of SA/TEO 1% and SA/TEO 1.5% film samples ($P>0.05$). It is observed that TEO added to the film forming solution caused a decrease in film solubility due to their concentration.

Table 1. Thickness, mechanical properties, and FS values of SA and SA/TEO films

Types of films	Thickness (mm)	FS (%)	TS (MPa)	EAB (%)
SA	0.087 ± 0.001^{d}	91.78 ± 1.11^{a}	50.06 ± 6.59^{a}	13.88 ± 2.53^{c}
SA/TEO- 0.5%	0.142 ± 0.004^{c}	76.06 ± 1.70^{b}	37.29 ± 3.86^{ab}	24.56 ± 1.27^{b}
SA/TEO- 1%	0.206 ± 0.005^{b}	68.98 ± 1.88^{c}	29.34 ± 1.29^{b}	30.90 ± 2.98^{ab}
SA/TEO- 1.5%	0.231 ± 0.006^{a}	66.50 ± 4.06^{c}	27.14 ± 1.30^{b}	32.02 ± 5.77^{a}

SA: sodium alginate, SA/TEO-0.5%: sodium alginate/ 0.5% thyme essential oil, SA/TEO-1%: sodium alginate/ 1% thyme essential oil, SA/TEO-1.5%: sodium alginate/ 1.5% thyme essential oil.

3.4. Optical properties of the films

The color characteristics affect the appearance, suitability, marketability of the coated products and the consumer's purchasing decisions (Shahbazi, 2017). Table 2 shows the color values (CIE $L*$, $a*$, and $b*$) measured of the film samples. When taken from the point of view of $L*$ values, it was observed that the SA (control) and SA/TEO 0.5% films did not differ (P>0.05), but increased slightly in the SA/TEO 1.5% film group, when the concentration of TEO was the highest. With this increase in the $L*$ value, it was concluded that the color of the films was closer to white and clearer. When $a*$ (redness-greenness) values were compared, three other films other than the SA (control) film did not have significant differences (P>0.05). In addition, as the TEO concentration increased, the $b*$ value increased from 12.09 to 13.14. In other words, it was seen that the film colors yellow with an increase in $b*$ value. It was observed that the SA/TEO 1.5% film is more yellow and less opaque than other films with an increased concentration of TEO. When considering the a values, there was no significant difference among the first three groups (P > 0.05). However, there was a significant difference observed in the %1.5 KY-SA film group (P < 0.05). Regarding the a* values, except for the control (SA) film, the other three films did not show significant differences (P > 0.05). According to Yan et al. (2012), color differences between $a*$ and $b*$ values in films containing TEO may be caused by phenolic compounds that contribute to the yellowish color. Color changes in films can be related to the properties of the components added to the film matrix and the drying conditions of the films. In the study by Mahcene et al. (2019), a film formulation solution was prepared using 3% (w/v) sodium alginate, 2.5% (w/v) glycerol, and various essential oils. The color values (L*, a*, b*) for sodium alginate films were found to be 87.86, -0.348, and 10.34, respectively. Upon addition of various essential oils derived from plants, the L* value ranged from 85.83 to 88.68, the a* value ranged from -0.446 to -0.329, and the b* value ranged from 11.3 to 12.21.

The opacity values of the films were interpreted based on values at 600 nm and were measured between 2.69 and 1.64 (Table 2). Opacity is the measure of a substance's ability to block the penetration of light. Transparent films allow consumers to easily see the food, which is desirable as it allows more light to penetrate through (Pereda et al., 2014). Thus, the aesthetic value of the product increases. But with more light penetration, foods can become more susceptible to spoilage. Opaque films exhibit light barrier properties and serve as a measure to protect foods from oxidation. It was determined that the opacity values of SA (control) films were higher than other film samples. Adding TEO at concentrations of 0.5% and 1% to films resulted in a decrease in opacity values, whereas TEO added at a concentration of 1.5% increased the opacity values of the films. According to Acosta et al. (2015) it has been reported that opacity is affected by intramolecular interactions and the distribution of components in the film matrix. In this study, it was concluded that interactions within the film matrix led to a decrease in light transmission. Transparency increased in SA/TEO 0.5% and SA/TEO 1% films, whereas transparency decreased in SA/TEO 1.5% films, approaching the transparency values of SA films. Similar results were observed in the study by Hammoudi et al. (2019). In this study, lemon essential oil was added to sodium alginate film solutions along with certain amounts of montmorillonite and Tween 80 (0.5%, 1%, 1.5%). The opacity values of the films were measured at 600 nm. The opacity value of sodium alginate films was found to be approximately 2.04 nm/mm. When montmorillonite and 0.5% and 1% (v/v) lemon essential oils were added, an increase in opacity was observed. However, with 1.5% (v/v) lemon oil, a decrease in opacity was observed.

Table 2. Colour parameters and opacity values of SA and SA/TEO films

Types of films	Color Parameters				$Opacity_{600\ nm}$
	$L*$	$a*$	$b*$	E	
SA	84.59±0.16^{b}	-1.51±0.01^{a}	12.09±0.14^{b}	2,64±0.75^{b}	2.69±0.09^{a}
SA/TEO- 0.5%	84.50±0.61^{b}	-1.59±0.03^{b}	13.85±0.55^{a}	3,04±0.73^{ab}	2.40±0.06^{a}
SA/TEO- 1%	84.94±0.71^{ab}	-1.60±0.03^{b}	14.22±0.97^{a}	3,71±0.23^{ab}	1.64±0.11^{b}
SA/TEO- 1.5%	85.78±0.18^{a}	-1.61±0.00^{b}	13.14±0.10^{ab}	4,12±0.41^{a}	2.29±0.38^{a}

SA: sodium alginate, SA/TEO-0.5%: sodium alginate/ 0.5% thyme essential oil, SA/TEO-1%: sodium alginate/ 1% thyme essential oil, SA/TEO-1.5%: sodium alginate/ 1.5% thyme essential oil

The light barrier capacity and visible light transmittance of the films are important for the packaging material. For the prevention and reduction of the effect of UV radiation, the visible light transmission of films is an important characteristic (Hu et al., 2020). Increased light transmittances in films can catalyze oxidation reactions in foods and thus cause undesirable reactions to begin. Table 3 shows the light transmission of SA (control) and SA/TEO films ranging from 200 to 800 nm. The average light transmittance values (%T) of the film samples in the visible region (350-800 nm) were measured as the highest SA (control) films. It is observed that a decrease in the light transmittance values occurs in films with the addition of TEO. As the concentration of TEO added to the film forming solution increased, light transmittance decreased. The samples exhibiting the lowest transmittance value among the groups were the SA/TEO 1.5% film samples. It is observed that the light transmittance values of the films are lower in the UV (200-280 nm) region compared to the values measured in the visible region. When the obtained results were considered statistically, it was found that there were significant differences in light transmittances between the film samples (P<0.05). The high light transmittance values of the control (SA) films were decreased with thyme oil added to the film formulation. This is important in terms of protecting foods from oxidation and being acceptable for the consumer. In general, it can be seen that SA (control) films are not very effective in terms of UV barrier, whereas SA films containing TEO have better UV barrier properties.

Table 3. UV and visible light transmission values of SA and SA/TEO films

Types of films	Wavelenghts (nm)							
	200 nm	280 nm	300 nm	400 nm	500 nm	600 nm	700 nm	800 nm
SA	0.03±0.00^{a}	7.05±0.66^{a}	27.28±2.67^{a}	36.57±2.56^{a}	49.68±2.41^{a}	58.30±1.10^{a}	61.01±2.48^{a}	65.23±1.29^{a}
SA/TEO-0.5%	0.03±0.00^{b}	1.91±0.21^{b}	33.98±2.21^{a}	35.13±2.67^{a}	45.26±0.66^{b}	45.58±0.88^{b}	50.73±4.57^{a}	55.48±2.25^{ab}
SA/TEO-1%	0.03±0.00^{b}	0.08±0.00^{c}	33.10±2.38^{a}	35.13±4.26^{a}	45.18±0.42^{b}	45.98±7.79^{b}	49.85±1.02^{a}	51.70±6.07^{b}
SA/TEO-1.5%	0.02±0.00^{c}	0.02±0.00^{c}	26.00±8.29^{a}	29.69±10.54^{a}	21.95±0.77^{c}	29.55±5.64^{c}	34.97±8.16b	37.06±8.14^{c}

SA: sodium alginate, SA/TEO-0.5%: sodium alginate/ 0.5% thyme essential oil, SA/TEO-1%: sodium alginate/ 1% thyme essential oil, SA/TEO-1.5%: sodium alginate/ 1.5% thyme essential oil

3.5. Total phenolic content and antioxidant activity of the films

The mean TPC values of the film samples were measured between 86.21 mg/kg and 382.84 mg/kg (Table 4). When the results were considered statistically, it was determined that there were significant differences between the film samples in terms of TPC (P<0.05). It is observed that the total amount of phenolic content in the films increases as the concentration of TEO added to the film forming solution increases. The group with the highest total amount of phenolic content is the SA/TEO 1.5% group. The mean antioxidant activity values (inhibition%) of the film samples were measured between 0.53% and 16.20% (Table 4.). When the results were considered statistically, it was determined that there were significant differences between the film samples from the point of antioxidant activity (P<0.05). It is observed that the antioxidant activity values of the films increase due to the increase in the concentration of TEO added to the film forming solution, and the group with the highest antioxidant activity value has SA/TEO 1.5% film samples. There was no statistically significant difference between the SA film group and the SA/TEO 0.5% film group (P>0.05). The total TPC has free radical scavenging capabilities. Because of this, it also has an effect on antioxidant capacity. Therefore, it is expected that there is a correlation between TPC and antioxidant capacity. This situation has been the same for all film samples. TEO contains high phenolic components. Therefore, an increase in the total phenolic content and antioxidant activity of the films has been observed with increasing concentrations of TEO. In the study by Jouki et al. (2014), edible films were prepared from quince seed mucilage by adding thyme essential oil at concentrations of 0%, 1%, 1.5%, and 2% (v/v). Depending on the increasing concentrations of thyme oil (1%, 1.5%, and 2%), significant increases were observed both in antioxidant activity values and phenolic content. The findings in this study have shown parallels with the results obtained in the mentioned study.

Table 4. TPC and antioxidant activity values (DPPH) of SA and SA/TEO films

Types of films	TPC content (mg GAE/kg)	Antioxidant activity (%inhibition)
SA	86.1±9.07d	0.53±0.82c
SA/TEO- 0.5%	134.44±1.61c	4.27±1.87c
SA/TEO- 1%	264.64±3.97b	8.47±3.64b
SA/TEO- 1.5%	382.84±19.70a	16.20±0.12a

SA: sodium alginate, SA/TEO-0.5%: sodium alginate/ 0.5% thyme essential oil, SA/TEO-1%: sodium alginate/ 1% thyme essential oil, SA/TEO-1.5%: sodium alginate/ 1.5% thyme essential oil

3.6. SEM

Depending on the structure of the film matrix, changes may occur in the morphology of the film. The SA (control) films have a homogeneous and uniform structure, as shown in the surface images (Fig 1). The addition of TEO caused a heterogeneous appearance on the film surface areas. It is seen that SA/TEO 0.5% films are also close to a homogeneous appearance, but there are some air bubbles on their surfaces (Fig 1). While SA/TEO 1% films have a partially heterogeneous appearance, it is observed that SA/TEO 1.5% films have a heterogeneous appearance (Figure 1). It is observed that the emulsion stability cannot be maintained with the increase in the concentration of TEO and it causes a heterogeneous appearance on the film surfaces. The emulsion has a heterogeneous and rough surface due to the formation of two phases (oil-polysaccharide) in the film matrix. This rough structure occurs during the drying of the films by migrating the lipids to the film surface, causing irregularities to form on the surface. As reported by Jouki et al. (2014) the addition of TEO lipid droplets at concentrations of 1% and 1.5% caused a heterogeneous structure in the films by their retention in the film matrix containing polysaccharide-based continuity. The molecular bonding between TEO and SA may differ depending on the film forming solution. Lipid droplets vary depending on the chemical composition and physical properties of TEO, and therefore also affect the morphology of the film.

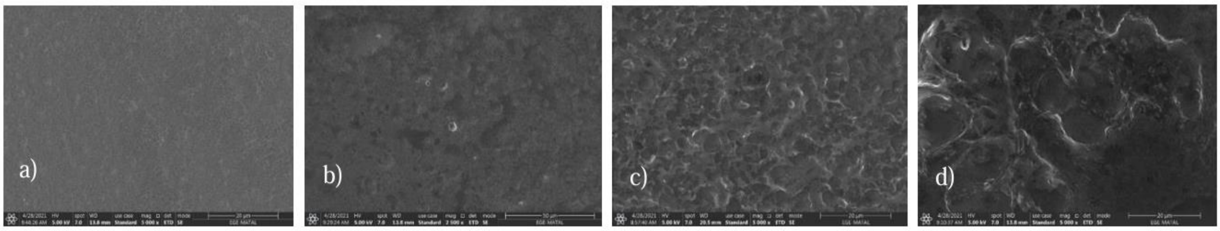

Figure 1. The surface SEM images of SA and SA/TEO films at 5000 magnifications (a) SA; b) SA/TEO 0.5%; c) SA/TEO 1%; d) SA/TEO 1.5%).

4. Conclusion

In this study, sodium alginate-based films with functional properties were enriched with thyme essential oil, and their physical and chemical properties were examined. The films prepared from sodium alginate SA (2% w/v) were aimed to be enhanced in their physical and chemical properties by adding thyme essential oil at different concentrations. TEO added to the SA based film forming solution caused the EAB and light transmittance properties of the films to improve, while the FS and TS properties of the films to decrease. Changes in the mechanical properties of films as a result of chemical bonds occurring in the structure with the addition of TEO show consistency with SEM images. According to the SEM images of the films, TEO has shown that it causes a heterogeneous structure in the film structure as a result of an increasing concentration. This situation supported that the emulsion structure was affected due to the increased concentration of TEO added. With TEO added to the films, the *b** value increased as a result of the occurrence of yellowness in the films and it has been observed that FT values have also increased. The components of carvacrol and thymol in TEO provide antioxidant properties to the essential oil (Dashipour et al., 2015). Therefore, an increase in the concentration of TEO has led to an increase in the antioxidant activity and TPC values of the films. In general, based on the results obtained, SA films with TEO addition are a promising alternative as edible packaging materials due to their low light transmittance, increased extensibility, improved antioxidant activity, and total phenolic content (TPC). For future studies, microbial analysis of SA/TEO films will open avenues for further investigation.

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