Journal of Food Science and Technology (Iran)

Homepage:www.fsct.modares.ir

[DOR: 20.1001.1.20088787.1401.19.132.15.8]

[DOI: 10.22034/FSCT.19.132.199]

Scientific Research

Effect of sorbitol concentration on physical and mechanical properties of bio-based film of quince seed mucilage

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ARTICLE INFO

Article History:

Received 2018/ 05/ 21
Accepted 2020/ 04/ 14

Keywords:

Bio-film,
Quince seed,
Sorbitol,
Physical and mechanical properties.

DOI: 10.22034/FSCT.19.132.199
DOR: 20.1001.1.20088787.1401.19.132.15.8

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ABSTRACT

Novel films based on the incorporation of sorbitol (20, 35, and 50% w/w) on quince seed mucilage were manufactured and physical, mechanical properties were investigated. Based on the results, by increasing sorbitol concentration the water vapor permeability (WVP), tensile strength (TS), water solubility and moisture content of quince seed mucilage films increased significantly and surface hydrophobicity declined. Moreover, by increasing sorbitol concentration, L^* b^* and a^* value of films decreased, increased, increased, respectively. The results revealed that incorporation of sorbitol in the biopolymer solution respected to glycerol (at the same concentration) led to obtaining a film with better barrier property and lower affinity to water. The WVP of the films increased with increasing storage relative humidity. Microscopic images showed the crystalline structure of film with sorbitol meanwhile a smooth, continuous structure with some cracks was observed in the film containing glycerol. Concluding, sorbitol suggested to incorporate in quince seed mucilage based films to obtaining advanced bio-films with low affinity to water for packaging of food products.

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1. Introduction

Packaging of food products can play an effective role in maintaining their quality. Due to the existing concerns regarding increasing the level of quality and health of food products and reducing waste from the packaging industry, the tendency to use biodegradable films has increased. By being placed on the surface of food systems, biofilms can be considered a good barrier against oxygen, carbon dioxide, aromatic compounds and fat] 1.[On the other hand, biofilms can be used as carriers for nutritious compounds and flavor enhancers]2[.

Quince fruit (*Cydonia oblonga Miller*) from family*Rosaceae*whose tree is cultivated in East Asia, South Africa and Central Europe]2[. After Turkey, China, Uzbekistan and Morocco, Iran is considered one of the most important fruit producing countries (FAO, 2014). Seed mucilage to be extractable under mild conditions in water]3[It is a complex of cellulose parts with hydrolyzable polysaccharides. The most important water-soluble polysaccharide of seed mucilage is acetyl-(4-oxymethyldiglucuronic acid)-di-xylan.] 4 .[One of the health features of seed mucilage is the destruction of cancer cells [5], helping digestive problems] 6[, activating growth factors and accelerating skin healing] 7[Cited. Recently, research has been done on the use of seed mucilage to produce film and edible coatings]8 ،9[. Biopolymers (polysaccharides, proteins, etc.) are used in the production of edible films and coatings along with additives such as softeners. [10]. Glycerol and sorbitol are common polyol softeners in the production of biofilms based on polysaccharides [11،12،13،14،15،16 [Regarding how plasticizers work in film production, we can mention the reduction of the friction force between polymer chains (lubricant theory), the breaking of inter-polymer bonds (gel theory) and also the increase of free volume and space between chains.] 15[. The flexibility of biofilms is largely dependent on the softener's chemical structure, molecular weight, and functional groups]9 .[The effect of plasticizers on the properties of the film obtained from new biopolymers is of interest to researchers. Due to the fact that the effect of sorbitol softener on grain mucilage film properties has not been investigated, the main purpose of this research was to investigate the effect of sorbitol concentration on the physicochemical properties of grain mucilage film and compare it with glycerol softener.

2- Materials and methods

The seeds used in this research were purchased from a market in Gilan, Iran. Glycerol (Sigma-Aldrich, USA) and sorbitol (Titrachem, Iran) were used for film production.

2-1- Seed mucilage extraction to

To extract the mucilage of seeds, the method of Jokio et al. (2014) was used with some changes] 8[. Based on this, a certain amount of seeds was weighed and immersed in distilled water at 25°C for 2 hours (1:25 ratio of water to seeds) and stirred frequently. The solution was filtered and the extraction process was repeated. The impurities of the extracted material were separated by centrifugation (6000 rpm) and the resulting mucilage was dried in an oven at a temperature of 40 ± 5 degrees Celsius for 20 hours.

2-2- Biofilm production based on seed mucilage

A 1% solution was prepared from dried grain mucilage. Sorbitol was added as a softener at a concentration of 25, 35 and 50% (based on the weight of dried grain mucilage powder). According to the authors' preliminary assessment of the effect of glycerol concentration on the physical and mechanical properties of biofilms based on grain mucilage, in this research, this grain mucilage containing 35% glycerol was considered as a control. The resulting film solutions were stirred for 8 minutes. Then they were placed in a centrifuge (5000 rpm) for 15 minutes. To remove air bubbles, keep the solutions in an ultrasonic bath for 1 hour Finally, they were dried at 40±5 degrees Celsius for 20 hours] 8[.

2-3- Evaluation of the properties of biofilms based on seed mucilage

2-3-1- Solubility in water of biofilms based on grain mucilage

The water solubility of the film was measured according to the method of Blogger et al. (2013) with some modifications] 17[. The samples of the films in sizes of 3 x 3 cm were kept in a

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desiccator containing phosphorus pentoxide until reaching a constant dry weight and weight (initial weight of the film) and kept in containers containing 50 ml of distilled water at room temperature for 4 hours. Further, after removing the film pieces from the solutions and absorbing the surface moisture, they were placed in a desiccator (the weight of the film after immersion and drying W_d^f W_d^f) and the solubility percentage in water of the produced films was calculated using the following equation.

% solubility=

Film weight after immersion and drying - initial film weight/Initial film weight ×100

2-3-2- The thickness of biofilms based on seed mucilage

The thickness of the films was measured with a manual digital micrometer (Mitutoyo, Mizonokuchi, Japan) with an accuracy of 0.001 mm at 12 points of the film randomly.

2-3-3- Moisture absorption of biological films based on seed mucilage

Films cut in 3 dimensions×3 cm were weighed (initial weight). Then they were placed in the oven with a temperature of 150 degrees Celsius until reaching a constant weight (final weight). The difference of the final weight compared to the initial weight was calculated and reported as the moisture absorption percentage of the sample] 18.[

2-3-4 - Water vapor permeability of biofilms based on seed mucilage

Permeability to water vapor was determined by measuring the weight changes of biofilms at a temperature of 25 degrees Celsius according to the ASTM standard method.] 19[. In this method, films were placed on the surface of cups and kept in a desiccator containing a saturated solution of sodium chloride (75% relative humidity) and water (100% relative humidity). The weight of each cup was measured regularly until a constant weight was reached. The rate of permeability to water vapor (WVTR) of the film was calculated from the slope of the straight line divided by the surface of the film according to equation 2. To determine the value of permeability, the rate of permeability was divided by the pressure difference between the two surfaces of the film (in kilopascals) (equation 3).

WVTR=Y/Y

$WVP=WVLR/\Delta P \times L_{film}$

In the above equation, L is the average thickness of the film in millimetersΔP ΔP is the partial pressure difference.

2-3-5- Mechanical properties of biofilms based on seed mucilage

Tensile strength (TS), elongation to break (EB) and Young's modulus (YM) of the film were performed by Instron Universal Testing machine (Model 200, Hiwa Engineering Co., Iran) according to ASTM standard method.]20[. Each of the films cut in dimensions of 2 x 5 cm were placed between the clamps of the device. The machine speed was set to 10 mm/min. Mechanics tests were performed in 8 repetitions.

2-3-6- The contact angle of biofilms based on seed mucilage

PG-X goniometer (Thwing-Albert Instrument Co., USA) was used to measure the contact angle of biofilms. 5 microliters of deionizer water was poured on the surface of the film measuring 5x5 cm and the tangent angle of the water drop with the surface of the film was determined at a temperature of 25 degrees Celsius.

2-3-7- The structure of biofilms based on seed mucilage

The structure of biofilms containing 35% softener (sorbitol and glycerol) were evaluated using scanning electron microscope images (Philips, Eindhoven, Netherlands). Cut film and gold powder in BAL-TEC SCD005 with sprayerBAL-TEC AG, Balzers, Liechtenstein))The dispersion and its structure were examined with a scanning electron microscope under high vacuum at a voltage of 20 kV.

2-3-8- Infrared spectroscopy with Fourier transform of biofilms based on seed mucilage

The effect of sorbitol concentration on film structure using Fourier transform infrared spectroscopy (FTIR) using a Bruker Equinox 55

FTIR spectrometer**(**Ettlingen, Germany**)** in the range of cm^{-1}500-4000 were investigated.

2-3-9- Color properties of biofilms based on seed mucilage

The color properties of the films were determined using image processing analysis. For this purpose, Genius Color Page HR6X Slim scanner was used. The produced films were placed in the scanner and the image was scanned with 32-bit RGB with a resolution of 300 dpi. Then, using the J Image software, the color indicators of the produced films are included*L ، *a and*b were determined.

2-4- Statistical analyses

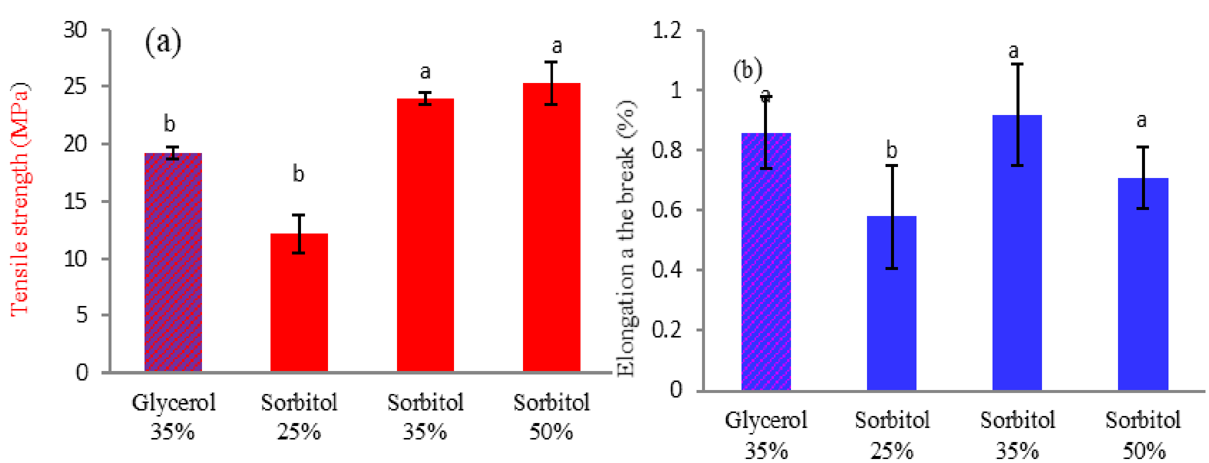
Statistical analysis was performed using completely randomized design with analysis of variance (ANOVA) and using SAS software (Version 9.1). In order to compare the average values of the properties of the films, LSD multi-range tests were used at the statistical level of 95%.

3. Results and Discussion

The biofilm based on grain mucilage without the presence of softener had no continuous structure and was also very fragile. Therefore, it was not possible to evaluate its physical and mechanical properties.

3-1- Evaluating the effect of softener concentration on the mechanical properties of produced films

Tensile strength (TS), extensibility (EB) and elastic modulus (E) are among the useful parameters to describe the mechanical properties of the film.] 21.[The results of the mechanical properties of grain mucilage film in the presence of different concentrations of sorbitol are shown in Figure 1. As can be seen in Figure a1, by increasing the level of sorbitol from 25 to 50% (based on the weight of the dry mucilage powder), the tensile strength (TS) of the film increased from 14.14 to 25.32 MPa, which is in agreement with the results of other researchers regarding the effect of increasing the concentration The softener corresponded to the decrease in tensile strength] 22 And23[. Also, the comparison of the results of the biofilm containing sorbitol and glycerol in the same concentration showed that the tensile strength of the film in the presence of sorbitol was more than that of glycerol. The results of this research were consistent with the results of other researchers regarding the effect of glycerol and sorbitol on the mechanical properties of the biofilm.24-26[. As shown in Figure 1, the stretchability of the film increased with the increase of sorbitol concentration. It is also worth noting that flexibility (Fig**1**) and the elastic modulus of the produced films (Figure 1c) at the same concentration of sorbitol and glycerol did not have statistically significant differences.]25[. As shown in Figure c1, with increasing sorbitol concentration, the elastic modulus increased, which can be attributed to the higher molecular weight of sorbitol compared to glycerol.



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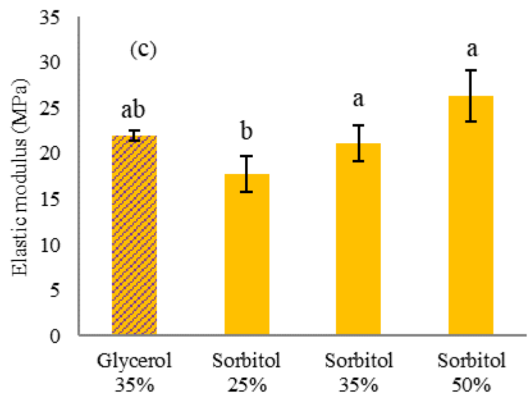

Fig 1 Effect of different concentrations of sorbitol on mechanical properties of quince seeds mucilage films, (a) tensile strength, (b) elongation at the break (c) modulus of Young; standard deviations are indicated by error bars, different letters on error bars represent statistical significances.

2-3-Evaluating the effect of softener concentration on color properties

The most important color properties of the film are the transparency index (0 to 100), the green to red index (−60 to +60) and the blue to yellow index (−60 to +60). As shown in Table 1, the brightness index of 83 Yianger production films had a relatively favorable transparency of the grain mucilage film. The results of this research showed that with the increase of softener concentrations, a significant decrease was observed between the brightness index of grain mucilage film at the level of 5% ($P < 0.05$, Table 1), which was consistent with the results of other researchers.]26 ‹23] According to the report of researchers, the presence of softeners in high concentration leads to an increase in the clear index of biofilm based on polysaccharides.28‹27[. On the other hand, with the increase in sorbitol concentration, the yellowness index of the produced films increased (Table 1).

The results of evaluating the color difference (ΔE) of the produced films in this research are given in Table 1. As can be seen, the color difference of the biofilms in the presence of sorbitol was greater compared to glycerol. The color difference of the produced films of grain mucilage was similar Other biofilms such as soy protein]29[, Chitosan/Hydroxypropyl Methyl Cellulose]30[and grain mucilage to]31[Was. It should be noted that the color difference of production films based on grain mucilage is more than that of some biological films such as albumin protein.]32]‹ Soy protein isolate] 33]‹ Alginate[18] and agar]34[Was. The brightness index of seed mucilage is greater in the presence of glycerol compared to sorbitol at the same concentration which can be considered due to the effect of glycerol molecular weight on film transparency and shine.

Table 1 Color parameters plasticized films and Contact angle

Type of films	Thickness (mm)	L^*	a^*	b^*	ΔE	Contact angle (degree)
Glycerol 35%	$65 \pm 3^{c*}$	93.59 ± 0.98^{a}	0.85 ± 0.12^{d}	8.81 ± 0.46^{d}	7.83 ± 0.31^{d}	46.16 ± 1.02^{c}
Sorbitol 25%	57 ± 4^{d}	91.66 ± 0.85^{b}	1.22 ± 0.28^{c}	11.16 ± 0.25^{c}	10.84 ± 0.31^{c}	53.52 ± 2.69^{a}
Sorbitol 35%	68 ± 3^{b}	89.02 ± 0.70^{c}	1.67 ± 0.37^{b}	14.11 ± 0.32^{a}	14.77 ± 0.27^{b}	49.13 ± 0.12^{b}
Sorbitol 50%	72 ± 2^{a}	83.95 ± 0.25^{d}	1.80 ± 0.45^{a}	12.22 ± 0.85^{b}	16.84 ± 0.55^{a}	45.12 ± 2.85^{d}

*Values for each film are means ± standard deviations. Values with different superscript letters in a given column are significantly different ($p < 0.05$) according to LSD test.

3-3- Evaluating the effect of softener concentration on the contact angle of produced films

The hydrophobicity of biofilms can be evaluated by determining the contact angle. contact angle Films can vary from 0 degrees (full spreading of the water drop on the film surface) to 180 degrees (slight absorption of moisture on the film surface). The results of evaluating the contact angle of the produced film in the presence of different concentrations of softener are shown in Table 1. As can be seen, with the increase of sorbitol concentration, the contact angle of the film decreased from 52.53 to 45.12 degrees. Also, at a concentration of 35%, glycerol softener had a smaller water contact angle compared to sorbitol (Table 1), which can be attributed to the greater hydrophilicity of glycerol compared to sorbitol. The results of the contact angle obtained from the grain mucilage film in the presence of glycerol in this research with the report of Joki et al. (2013) matched]35[.

Fig 2 Quince seed mucilage films with 35% sorbitol as a plasticizer

3-4- Evaluation of the effect of softener concentration on water solubility of produced films

The results of evaluating the water solubility of the produced films were shown in Figure 3. As can be seen, the solubility in water of grain mucilage film containing 25% sorbitol (18.7±0.75%) compared to 35% (37±2.29) and 50% sorbitol (51.82±2.75%)**)** It was less. Based on the obtained results, the biofilm containing 35% sorbitol as a non-volatile hexa-alcohol had less solubility in water compared to the film containing glycerol in the same concentration, which is in agreement with the results. Fakhuri Et al (2012) that the water solubility of the film containing sorbitol was reduced compared to glycerol] 37]. These characteristics have caused sorbitol to be considered a suitable softener with the aim of producing more water-resistant films. Comparison of the results of the solubility of grain mucilage films in this research with other hydrocolloids as a substitute for synthetic polymers indicates that the grain mucilage film is more water resistant than gelatin-based films with a solubility percentage of 63.81.36 [and alginate with a solubility percentage of 99.5]18 [Was.

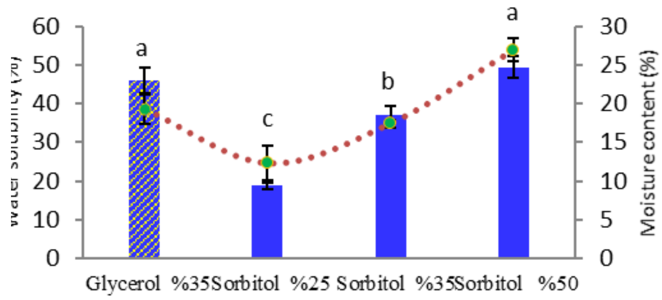

Fig 3 Effect of different concentrations of sorbitol on water solubility and moisture content of quince seeds mucilage films

5-3-Evaluating the effect of softener concentration on the humidity of production films

By increasing the amount of sorbitol softener, the moisture content of the grain mucilage film increased significantly (P < 0.05).**Shape**3). According to the results obtained from this research, the moisture percentage of the film in the presence of 35% glycerol was higher compared to the same concentration of sorbitol. The reason for this can be attributed to the hygroscopic property and greater hydrophilicity of glycerol [23 ،38 ، 39، 40 And41].

3-6- Evaluation of the effect of softener concentration on the permeability to water vapor of produced films

Permeability to water vapor of biological packages can affect the rate of browning, lipid oxidation, vitamin structure change, enzyme activity, microbial growth and tissue destruction. Permeability to water vapor is an important parameter that depends on the solubility, the rate of moisture penetration into the film substrate and the partial pressure of water vapor [42].The results of evaluating the effect of softener on water vapor permeability of the film in the presence of relative humidity of 50 and 70% are shown in Figure 4. As can be seen, by increasing the concentration of sorbitol in the film solution, the permeability to water vapor of the produced films increased significantly in the investigated relative humidities.**(**5/0 P **<)** In this respect, it is consistent with the results of other researchers regarding the increase in permeability to water

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vapor of the hydrocolloid film with an increase in the concentration of sorbitol.11،13،23،41،43 ،44Also, based on the results obtained from this research, with increasing humidity from 50 to 70%, the permeability to water vapor of films containing glycerol from $^{11-}10 \times 36$ **until the** $^{11-}$**10** × 9/54 grams. meters per square meter The increase in pascal seconds showed that it was similar to the change in permeability to water vapor of films containing sorbitol (Figure 4). These results indicated the sensitivity of the grain mucilage film to changes in the relative humidity of the environment. Accordingly, in higher relative humidity, the formation of hydrogen bonds between the film and polyol with water absorption increases the ratio of polysaccharide-water and polyol-water bonds, and as a result, water diffusion. in the film bed and finally the percentage of moisture increased. It should be noted that increasing the permeability to water vapor is not considered desirable in most edible films.]45[.

The water vapor permeability of films containing glycerol and sorbitol at a concentration of 35% in a relative humidity of 50% was better than 36.01 and 24.7 g/m2 pascal seconds.**Shape**4) It can be said that glycerol as a hydrophilic and small compound can enter between the polymer chains and increase by reducing the intermolecular force and molecular movement between the polymer substrate, and this increased mobility naturally leads to an increase in the free volume and partial movements and accelerating the migration of molecules. water vapor. The results obtained from this research were consistent with the results of other researchers regarding the favorable inhibitory effect of sorbitol compared to glycerol in biofilm.37 ، 13 And23[.

3-7- Evaluation of the structure of the produced film using scanning electron microscopy (SEM)

Electron microscope images can better express the relationship between water vapor transfer, mechanical and structural properties of the film. The cross-sectional images of grain mucilage film in the presence of sorbitol and glycerol at the same concentration are shown in Figure 5. As can be seen, the structure of the film in the presence of glycerol is uniform with pores and cracks**(**Figure C5, d4 (More evident compared to sorbitol**(**Figure a5, b4**)** had. Also, the surface of the film containing sorbitol is very uneven, which can probably be related to the crystallization of sorbitol during drying. On the other hand, the higher molecular weight and hydrogen-hydroxyl bonds of sorbitol compared to glycerol can lead to limiting the diffusion of this softener in the film substrate and accumulation on the surface. It should be noted that this surface accumulation can be a confirmation of the decrease in permeability to water vapor and solubility in water, as well as the increase in the tensile strength of the film.]46[. On the other hand, these images show that the compatibility of glycerol with the seed mucilage film even at higher magnification (Figure b5, d4), no phase separation was visible, but cracks were visible in the presence of glycerol softener, which can be a reason for increased permeability to water vapor and reduce its tensile strength]47[.

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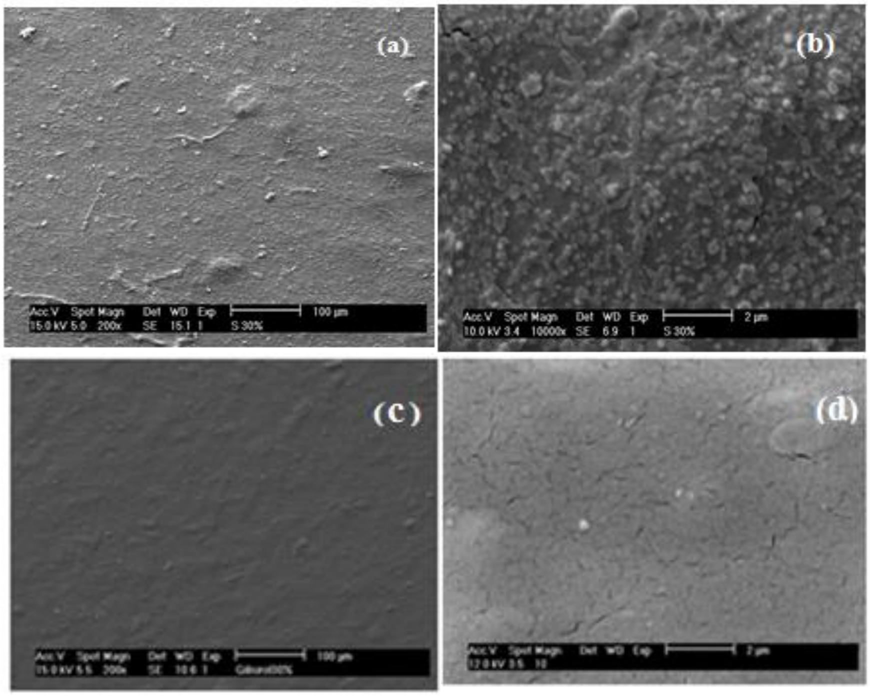

Fig 4 SEM micrographs of cross section for quince seed mucilage films containing 35% sorbitol (a, b) and glycerol (c, d)

3-8- Evaluation of the structure of the produced film using Fourier transform infrared spectroscopy

Infrared spectroscopy shows functional groups and film structure changes at the molecular level through detailed spectral analysis. Infrared spectroscopy of grain mucilage film in the presence of glycerol and sorbitol (35% concentration) is shown in Figure 6. The highest absorption in the cm wavenumber range^{-1}800 - 1200 as carbohydrate fingerprint, cm^{-1} **17/1618** For COO asymmetric stretching, cm^{-1} 85/2924 and cm^{-1} 74/2359 for symmetric vibration coupling$_2$CH and vibrational stretching of CH as well as cm^{-1} 17/3435 is observed for the hydroxyl group]8[. Grain mucilage film vibrates in the presence of glycerol in the range of cm wavenumber^{-1} 3100 to 3600 showed that it can probably be caused by the greater interaction of glycerol through the O-H group between the carbohydrate chains of the film compared to sorbitol. Based on this, the lower solubility of the film in the presence of sorbitol compared to glycerol can be justified. In the presence of sorbitol, the grain mucilage film has a higher absorption intensity at cm wavenumber^{-1}2869 showed that caused by the symmetric vibrational stretching of CH_2 which can be considered as indicating the binding of sorbitol with the film substrate. Also, minor differences in bonding and bonding intensity in grain mucilage film in absorption peak cm^{-1} 800-1200 FTIR visible.

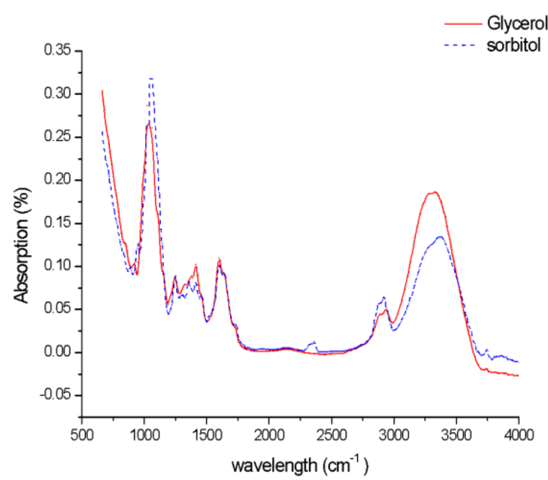

Fig 5 FTIR spectra of quince seed plasticized with addition of glycerol and sorbitol at the concentration of 35%

4 - Conclusion

The biofilm properties of grain mucilage were affected by different concentrations of sorbitol,

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and the efficiency of sorbitol as a softener in the production of grain mucilage film was higher than that of glycerol at the same concentration due to the reduction of WVP, moisture content, solubility in water, surface hydrophobicity percentage. In addition to the amazing increase in the tensile strength of the film without significantly affecting the tensile properties accepted The results of the electron microscope images showed that the increase in film hardness in the presence of sorbitol was the reason for the decrease in solubility and water vapor permeability of grain mucilage film. The use of grain mucilage containing sorbitol as a film-forming agent and also as an edible coating in peanuts as a protective agent against fat loss is investigated and will be published in the near future.

5- Resources

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[DOR: 20.1001.1.20088787.1401.19.132.15.8]

[DOI: 10.22034/FSCT.19.132.199]

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مقاله علمی_پژوهشی

تاثیر غلظت سوربیتول بر خواص فیزیکی و مکانیکی فیلم زیستی موسیلاژ دانه به

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اطلاعات مقاله

تاریخ های مقاله :

تاریخ دریافت: ۱۳۹۷ /۰۲/۳۱

تاریخ پذیرش: ۱۳۹۹/۰۱/۲۶

کلمات کلیدی:

فیلم زیستی،

موسیلاژ دانه به،

سوربیتول،

خواص فیزیکی و مکانیکی.

DOI: 10.22034/FSCT.19.132.199

DOR: 20.1001.1.20088787.1401.19.132.15.8

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چکیده

فیلم های زیستی بر پایه موسیلاژ دانه به در حضور سوربیتول (۲۰، ۳۵ و ۵۰ درصد وزنی/ وزنی) تولید و خواص فیزیکی و مکانیکی آن ها مورد بررسی قرار گرفت. نتایج به دست آمده از این تحقیق نشان داد که با افزایش غلظت سوربیتول، نفوذ پذیری نسبت به بخار آب، مقاومت کششی، حلالیت در آب و درصد رطوبت فیلم ها به طور معنی داری افزایش و آب‌گریزی سطحی کاهش یافت. هم چنین با افزایش غلظت سوربیتول اندیس روشنایی، قرمزی و زردی فیلم های تولیدی به ترتیب کاهش، افزایش و افزایش یافت. نفوذ پذیری نسبت به بخار آب فیلم ها با افزایش رطوبت نسبی محیط افزایش یافت. مقایسه خصوصیات فیلم موسیلاژ دانه به حاوی ۳۵ درصد گلیسرول (شاهد) با نرم کننده سوربیتول در غلظت مشابه نشان داد که خواص ممانعت کنندگی نسبت به بخار آب و خواص آب دوستی فیلم زیستی تولیدی در حضور نرم کننده سوربیتول بهتر بود. تصاویر میکروسکوپ الکترونی و طیف سنجی مادون قرمز نتایج به دست آمده را مورد تایید قرار داد. بر این اساس استفاده از سوربیتول در پلیمر موسیلاژ دانه به برای تولید فیلم های زیستی با آبدوستی کمتر جهت بسته بندی محصولات غذایی توصیه می گردد.