Active chitosan—polyvinyl alcohol films with natural extracts

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1. Introduction

Packaging is an important factor in food industry and is dominated by petroleum-derived polymers. However, the amount of research involving the production and characterization of biodegradable films has increased substantially, mainly due to interest in minimizing the ecological impact caused by the use of synthetic packaging materials. Several biopolymers have been exploited to develop eco-friendly food packaging materials (Siracusa, Rocculi, Romani, & Rosa, 2008). A significant proportion of research on these films has been made using biopolymers from renewable sources, i.e. products or by-products derived from agriculture or from agro-industries. Usually, films based on biopolymers are highly sensitive to environmental conditions and generally present low mechanical resistance. As a result, several researchers have developed films based on mixtures of biopolymers and synthetic polymers (Chorpade, Gennadios, Hanna, & Weller, 1995).

Chitosan (Ch) is a very promising biopolymer because it is environmentally friendly due to its biodegradability and has good film forming properties. In the food industry, Ch films promise immense potential to be used as active packaging material due to its antimicrobial activity, non-toxicity and low permeability to oxygen (No, Meyers, Prinyawiwatkul, & Xu, 2007). Some synthetic polymers from non-renewable sources are also biodegradable, such as polyvinyl alcohol (PVA). PVA is a synthetic, water soluble polymer with excellent film forming, emulsifying, and adhesive properties. It also imparts good tensile strength (TS) and biodegrability and hence has been used in many biomaterial applications. PVA has also been approved for use in packaging meat and poultry products by the USDA (DeMerlis & Schonek, 2003). Ch contains free hydroxyl and amine groups, and is therefore miscible with PVA due to the formation of hydrogen bonds.

Polymer blending is one of the most effective methods to have new material with desired properties. Films formed by blending of polymers usually results in modified physical and mechanical properties compared to films made of individual components. Since synthetic polymers are easily obtained and have low production cost, blending of natural and synthetic polymers improves the cost—performance ratio of the resulting films. Blending of synthetic polymers, such as PVA (Bahrami, Kordestani, Mirzadeh, & Mansoori, 2003), polycaprolactone (Sarasam, Krishnaswamy, & Madihally, 2006) with Ch have been reported to improve mechanical properties of Ch films. In order to enhance utility of these films, addition of active agents is being investigated to improve food safety and quality. In terms of active agents that can be incorporated into films, plant extracts have received much attention as they contain high concentrations of phenolic compounds that possess strong antioxidant properties. Earlier studies in our laboratory have shown that mint extract (ME) and pomegranate peel extract (PE) have good antioxidant potential (Kanatt, Chander, & Sharma, 2007, 2010). These extracts effectively scavenged DPPH, hydroxyl and superoxide radical and their scavenging ability was comparable to the synthetic extracts (ME)/pomegranate peel extract (PE). The effect of these extracts on the physical, mechanical, antimicrobial and antioxidant properties of the films was studied. Increased protection against UV light was observed in the films containing the extracts. Addition of ME/PE improved the tensile strength of the films without affecting their puncture strength. Ch—PVA films incorporated with PE had the highest tensile strength (41.07 ± 0.88 MPa). Permeability characteristics of the films were not altered due to addition of extracts. ME/PE conferred antioxidant properties to Ch—PVA films as determined by DPPH radical scavenging activity. The films also exhibited antibacterial activity against Staphylococcus aureus and Bacillus cereus. PE containing films totally inhibited the growth of B. cereus and reduced the number of S. aureus by 2 log cycles. These results suggest that Ch—PVA film containing ME/PE can be used for development of active food packaging materials.

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antioxidant, butylated hydroxytoluene (BHT). ME/PE also had good reducing power and iron chelation capacity. Hence, the objective of this study was to develop composite active films from Ch and PVA incorporated with ME/PE. Optical, mechanical, barrier, antioxidant and antimicrobial properties of these films were also investigated.

2. Materials and methods

2.1. Materials

Commercial Ch from shrimp shells with a molecular weight of 1.86 × 10^5 Da and minimum deacetylation degree of 90% was purchased from Mahatani Chitosan Pvt. Ltd. (Veraval, India). 2, 2-diphenyl 1-picryl hydrazyl (DPPH) and catechin were purchased from Sigma Chemical Co. (St. Louis, MO). PVA (molecular weight 1.4 × 10^5 Da) and microbiological media were procured from HiMedia (Mumbai, India). All other reagents used were of analytical grade and procured from S. D. Fine Chemicals (Mumbai, India).

2.2. Methods

2.2.1. Preparation of mint extract and pomegranate peel extract

Mint (Mentha spicata L.) and pomegranate (Punica granatum) were purchased from the local market. The arils and peel of pomegranate were separated. The pomegranate peel/mint leaves (100 g) were refluxed with distilled water (1000 ml) for 1 h. The extract was cooled and filtered through cheesecloth and the residue was again refluxed for an additional hour. The extracts were centrifuged at 4500 g for 20 min. The supernatant obtained was concentrated in a rotary evaporator (Buchi Rotavapor, Flawil, Switzerland). The concentrates were lyophilized to form powders (ME/PE) which were stored at 4 °C until further use.

2.2.2. Film preparation

The polymer films were prepared by the casting method. Ch solution (1% w/v) was prepared by dissolving Ch in 1% aqueous acetic acid solution at room temperature with stirring (500 rpm). The PVA solution (5% w/v) was prepared in hot distilled water. The solutions of PVA and Ch were carefully mixed at various ratios (1:1, 1:2 & 2:1). Glycerol (0.1% v/v of film forming solution) was added to the film forming solution at a concentration of 0.1% (Table 1). All the film forming mixtures were blended by stirring on a magnetic stirrer (200 rpm) at room temperature for 15 min and degassed. The films were prepared by casting an amount of 150 ml on teflon plates (15 cm × 15 cm) and drying at 50 °C in a ventilated oven at 50% relative humidity (RH) to obtain films of uniform thickness. The dried films were peeled from the casting surface. Film characteristics were determined after all sample films were preconditioned in a constant temperature humidity chamber (Model FX 1077, Jeio Tech Co., Ltd., India) set at 23 °C with 50% RH for 24 h.

2.2.3. Film thickness measurements

Film thickness was measured using a hand-held micrometer (Mitutoyo No. 7327, Tokyo, Japan). Measurements were taken at five different locations of each film sample and the average film thickness was calculated.

2.2.4. Optical properties

Each film specimen was cut into a rectangular piece and placed directly in a UV–Vis spectrophotometer (Model UV-1601, Shimadzu Co., Kyoto, Japan) test cell and measurements were performed using air as the reference. A spectrum of each film was obtained at wavelengths between 200 and 800 nm. The results have been expressed as percentage transmittance. The measurements were done in triplicate and the average of three spectra was calculated. The transparency at 600 nm (T600) was obtained using the following equation (Han & Floros, 1997).

\[ T600 = -\log\%T/b \]  

where %T is percentage transmittance and b is the film thickness (mm). The opacity of the films was calculated by the following equation according to the method described by Gontard and Guilbert (1994).

\[ \text{Opacity} = \frac{\text{absorbance at } 500 \text{ nm} \times \text{film thickness (mm)}}{2} \]

2.2.5. Mechanical properties

Tensile strength (TS) and puncture strength were measured using Texture Analyzer TA-HD plus (Stable Micro Systems, Surrey, UK) in accordance with ASTM D882-91 method (ASTM, 1991). For TS, the films were cut into strips of 25 mm wide and 100 mm long. Initial grip separation and cross-head speed were set at 50 mm and 5.0 mm/s, respectively. TS values were expressed in MPa and calculated by dividing the maximum stress (N) by cross-sectional area (m²) of the specimen. Puncture strength was evaluated using a needle probe of 2 mm in diameter at a constant rate of 1 mm/s and the results were expressed in terms of N. For both, tensile or puncture tests, each reported value corresponded to at least five determinations.

2.2.6. Water vapor transmission rate (WVTR) and oxygen permeability (OP)

WVTR tests were carried out using an automatic water vapor permeability testing machine L80-5000 (PBI Dansensor, Denmark) at 37 °C and 10/15% RH. The measuring range of the instrument was 0.03–10,000 g/m²/day. WVTR of films was measured using aluminum sample cards (reduction to 5% area). OP of the film was estimated with automated oxygen permeability testing machine OPT-5000 (PBI Dansensor, Denmark) at 23 °C and 0% RH. The measuring range of the instrument was 0.1–10,000 ml/m²/day. Sample was placed in the sample holder having an exposed testing area of 50 cm². WVTR and OP of each sample were averaged from three separate tests.

2.2.7. Antioxidant potential of films

Films (5 cm²) were placed in conical flasks containing 10 ml of distilled water. These flasks were then continuously shaken in orbital shaker (100 rpm) set at three different temperatures (37 °C, 28 °C and 15 °C). Three sets of each film were taken for each temperature. The antioxidant activity of the films was monitored by taking aliquots of the supernatant obtained from each flask at different time intervals and analyzed for total phenolics content and DPPH radical scavenging activity.

2.2.7.1. Estimation of total phenolics

The total phenolic content of the extracts was estimated by the Folin–Ciocalteu method of Singleton and Rossi (1965). Briefly, the diluted sample was mixed...
with Folin–Ciocalteu reagent and kept at room temperature for 5 min. Sodium bicarbonate solution (6%, 0.75 ml) was added to the mixture and further incubated for 90 min. Absorbance of the solution was measured at 725 nm. The total phenolic acid content was expressed as catechin equivalents.

2.2.7.2. DPPH radical scavenging activity. The DPPH radical scavenging activity was determined by the method of Yamaguchi, Takamura, Matoba, & Terao (1998). The diluted sample (1 ml) was added to DPPH in ethanol (1 ml) and vortexed vigorously. After 20-min incubation in dark at ambient temperature, the absorbance was measured at 517 nm. Percent DPPH scavenging activity was calculated as follows

\[
\text{[(Control absorbance } - \text{ Extract absorbance)}/\text{(Control absorbance) } \times 100}
\]

(3)

2.2.8. Antimicrobial activity

The test organisms used were Escherichia coli JM109, Pseudomonas fluorescens ost5 (16s rRNA gene sequence submitted to Genbank, Accession no. DQ439976) – a laboratory isolate, Bacillus cereus MTCC 470 and Staphylococcus aureus ATCC 6538P. The test organisms were grown in nutrient broth for 18 h at 37 °C. The cells were washed and suspended in sterile phosphate buffer saline (pH 7.2). The cells were further diluted to get concentration of \(10^5–10^6\) cfu/ml. Film samples (diameter of 1.5 cm) were transferred aseptically in tubes and aliquot of samples were immediately removed, diluted with sterile saline and plated on plate count agar. The tubes were then incubated at 37 °C for 24 h and the colonies were counted.

2.2.9. Statistical analysis

All experiments were carried out in triplicate and the average values with standard errors were reported. Analysis of variance was conducted and differences between variables were tested for significance by one-way ANOVA with Tukey’s post test using graphpad instat version 3.05 for Windows 95 (GraphPad Software, San Diego, CA, USA). A statistical difference at \(p < 0.05\) was considered to be significant.

3. Results and discussion

3.1. Appearance and film thickness

The composite films formed from Ch and PVA were visually homogeneous with no brittle areas or bubbles and could be easily peeled from the casting plates. Ch and PVA were miscible in all blends used in this study and this was attributed to formation of strong inter-molecular hydrogen bonds between Ch and PVA molecules (Kumar et al., 2010). Film thickness is a very important parameter in determining film’s physical properties. The films had an average thickness of 70 ± 2 μm. Ratio of Ch/PVA and addition of ME/PE did not change significantly \((p > 0.05)\) the average thickness of the films. The concentration of ME/PE used in this study was only 0.1% and therefore there was no effect on the film thickness. Benavides, Villalobos-Carvajal, and Reyes (2011) reported that the addition of oregano essential oil (1.5%) in the film forming emulsion led to an increase in the thickness of the film. Visually, the films containing ME had slight green color while those containing PE had a yellow color. Similar results have been reported by Siripatrawan and Harte (2010) who found that the incorporation of green tea extract into Ch films gave color to the film. Gómez-Estaca, Giménez, Montero, and Gómez-Guilén (2009) reported that the appearance of the gelatin films was affected by the addition of oregano or rosemary extracts.

3.2. Optical properties

One of the desired characteristics of packaging material is that it should protect food from the effects of light, especially UV radiation. To determine the light transmission properties of the films, they were scanned at wavelengths ranging from 200 to 800 nm and the percentage light transmittance was recorded (Fig. 1). Film samples exhibited low light transmission in UV light, especially at wavelengths of 280 nm. Films with ME/PE had lower percent transmittance compared with control film (without extract) suggesting that composite Ch–PVA films with ME/PE have good barrier to ultraviolet light, a powerful lipid-oxidizing agent in food systems. At 400 nm, the control films had transmittance values of around 10–20%, whereas, in ME/PE supplemented films the transmittance values further reduced suggesting that films with added extracts improved the light barrier properties. These results are in agreement with the previous findings of Gómez-Guilén, Ilh, Bifani, Silva, and Montero (2007) who reported that there was improvement in light barrier properties when antioxidant extract from murta was added to gelatin-based edible films. The effects of ME/PE on film opacity and transparency are shown in Table 2. Films in which the PVA content was greater were more transparent (lower opacity value). Incorporation of ME/PE reduced the transparency only marginally probably because the concentration of the natural extracts used was only 0.1%. Siripatrawan and Harte (2010) reported that Ch films without green tea extract were more transparent than those incorporated with extract. It has been reported that addition of phayom wood extract into the edible hydroxypropyl methyl cellulose films resulted in a decrease in their transparency (Jutaporn, Suphitchaya, & Thawien, 2011).

3.3. Mechanical properties

A food packaging film is required to maintain film integrity in order to withstand the stress that occurs during shipping, handling and storage. The mechanical properties of the studied films are summarized in Fig. 2. The films in which the PVA concentration was more (C1P2) had higher TS values (30.86 ± 1.01 MPa). PVA fraction thus contributed to increase in TS and a greater force was required
to rupture them. Ch membranes blended with PVA have been reported to have good mechanical properties for medical products and for controlled delivery of drugs (Wang, Du, & Fan, 2005). The tensile strength of the films was affected by the addition of ME/PE. In the presence of ME/PE, there was an increase in TS of the films especially in films in which the PVA concentration was high. Ch–PVA films incorporated with PE (C1P2P) had the highest TS of 41.07 ± 0.88 MPa. Phenolic compounds contain number of OH groups which form hydrogen bonds (Arcan & Yemenicigil, 2011) with chitosan and therefore there was an increase in tensile strength on addition of ME/PE. This observation agrees with the findings of Siripatrawan and Harte (2010) who also reported the improvement in TS of Ch films incorporated with green tea extract and attributed it to the interaction between Ch matrix and polyphenolic compounds from green tea extract. Similarly, Sivarooban, Hettiarachchy, and Johnson (2008) observed that the incorporation of grape seed extract significantly increased TS of soy protein isolate films. It was also seen that the TS of Ch–PVA films incorporated with ME/PE was higher as compared to the reported TS values of other biopolymer films, such as alginate films incorporated with ginseng extract (Norajit, Kim, & Ryu, 2010), gelatin films containing seaweed extract (Rattaya, Benjakul, & Prodpran, 2009).

Another important mechanical property of packaging materials is the puncture resistance (Lange, Mokdad, & Wyser, 2002). In many applications, packaging can be damaged by penetration, leading to reduced barrier properties and loss of package integrity. In Fig. 2, the puncture strength of composite Ch–PVA films has been shown. Results indicated that puncture strength of films with and without ME/PE were not significantly different (p > 0.05).

### 3.4. Barrier properties

The determination of the barrier properties of a polymer is crucial to estimate and predict the product-package shelf-life. The specific barrier requirement of the package system is related to the product characteristics and the intended end-use application. Proper barrier to moisture and oxygen in food systems can increase quality of the product. Water vapor and oxygen are two of the main permeants studied in food packaging applications, because they may transfer from the internal or external environment through the polymer package wall, resulting in a continuous change in product quality and shelf-life.

WVTR is a measure of the ease with which the moisture penetrates and passes through a material and has great influence on the food shelf-life. The results of WVTR analyses of films are shown in Fig. 3. The WVTR of the films decreased significantly (p < 0.05) when Ch content in the film increased. The WVTR of C1P2 was 2860 ± 30.82 g/m²/day while that of C2P1 was lower at 2582 ± 30.97 g/m²/day. Similar results have been reported by Li, Peng, Yue, and Xie (2006) who found that that an increase in chitosan concentration resulted in decreased WVTR of konjac glucomannan–chitosan films. Pelissari, Grossmann, Yamashita, and Pineda (2009) reported that on increasing the content of chitosan in cassava starch–chitosan film there was a decrease in WVTR due to the formation of hydrogen bonds between the NH₂ present in Ch and the OH⁻ of cassava starch thus reducing the availability of the hydrophilic groups. The WVTR of composite films depends on the hydrophilic–hydrophobic ratio of the film constituents. Highly polar polymers exhibit high degrees of hydrogen bonding, resulting in elevated WVTR values (Kester & Fennema 1986). One of the reasons for Ch addition in edible films is its relatively more

### Table 2

<table>
<thead>
<tr>
<th>Sample</th>
<th>Transparency</th>
<th>Opacity</th>
</tr>
</thead>
<tbody>
<tr>
<td>C1P1</td>
<td>7.61 ± 0.23a</td>
<td>0.17 ± 0.01b</td>
</tr>
<tr>
<td>C1P2</td>
<td>9.13 ± 0.54c</td>
<td>0.08 ± 0.02d</td>
</tr>
<tr>
<td>C2P1</td>
<td>6.71 ± 0.36f</td>
<td>0.16 ± 0.01e</td>
</tr>
<tr>
<td>C1P1M</td>
<td>7.62 ± 0.27e</td>
<td>0.14 ± 0.03f</td>
</tr>
<tr>
<td>C1P2M</td>
<td>8.38 ± 0.41a</td>
<td>0.11 ± 0.02b</td>
</tr>
<tr>
<td>C2P1M</td>
<td>7.00 ± 0.10f</td>
<td>0.16 ± 0.02a</td>
</tr>
<tr>
<td>C1P1P</td>
<td>8.71 ± 0.31c</td>
<td>0.11 ± 0.01b</td>
</tr>
<tr>
<td>C1P2P</td>
<td>8.59 ± 0.22a</td>
<td>0.09 ± 0.02b</td>
</tr>
<tr>
<td>C2P1P</td>
<td>7.94 ± 0.18a</td>
<td>0.16 ± 0.03a</td>
</tr>
</tbody>
</table>

* *Different superscript letters in each column are significantly different (p < 0.05).*
hydrophobic nature that could provide higher moisture barrier and water resistance (Mathew & Abraham, 2008). Addition of ME/PE did not affect the WVTR of the films. Since a main function of a food packaging is often to avoid or at least to decrease moisture transfer between the food and the surrounding atmosphere, or between two components of a heterogeneous food product, WVTR should be as low as possible (Gontard, Guilbert, & Cuq, 1992).

Gas permeability of packaging materials is of great importance for food preservation. Barrier to oxygen in a packaging system can increase food product shelf-life and also improve the food quality (Brody, Bugusu, Han, Sand, & Mchugh, 2008). The OP values showed that all the films were impermeable to oxygen. Ch could form intermolecular hydrogen bonds with PVA, which limits the intermolecular chain mobility, contributing to the decrease of OP. Moreover, Butler, Vergano, Testin, Bunn and Wiles (1996) have reported that the Ch films are highly impermeable to oxygen. This property of Ch film has been used extensively in modified atmosphere packaging of fruits and vegetables (Tharanathan, Srinivasa, & Ramesh, 2002). The OP values of Ch–PVA films are better than those of synthetic plastics such as LDPE and HDPE (Brody & Marsh, 2008).

Fig. 3. Water vapor permeability of Ch–PVA films incorporated with ME/PE. Bars indicate standard deviation (n = 3).

Fig. 4. Total phenolic content of Ch–PVA films incorporated with ME/PE. Results are mean of three independent experiments.
In general, polysaccharide films are expected to be good oxygen barriers, due to their tightly packed and ordered hydrogen-bonded network structure and low solubility.

### 3.5. Antioxidant activity

Antioxidant packaging is a major category of active packaging and very promising technique for extending food product shelf-life. Enriching films with antioxidants allows nutritional and aesthetic quality aspects to be enhanced without affecting the integrity of the food product (Guilbert, Contard, & Gorris, 1996). Food packed in antioxidant biodegradable packaging may be stored at different temperatures depending on its composition and hence the release of phenolics and their DPPH radical scavenging activity from the Ch–PVA films containing natural extracts was monitored at different temperatures.

#### 3.5.1. Total phenolics

Phenolic compounds are secondary plant metabolites. They are effective hydrogen donors and possess ideal structural properties for scavenging free radical and thus have good antioxidant potential. When the composite films were immersed in distilled water to study the release of the bioactive components, being polar the films started to hydrate, causing relaxation of the polymer chain with consequent swelling. The films lost their structural integrity and released the active components (Flores, Conte, Campos, Gerschenson, & Nobile, 2007). The control films (without any added extract) contained no phenolics (data not shown). The total phenolic content of films containing ME was greater than in the films with PE (Fig. 4). Furthermore, the amount of phenolics released from the films differed considerably according to testing temperature. At higher temperature (37 °C), the phenolics released from the film was maximum and very little phenolics were released from films kept at 15 °C. The total phenolic content of films directly correlated with the antioxidant activity measured by DPPH radical scavenging activity. Arcan and Yemenicioglu (2011) also reported that the incorporation of phenolic compounds such as catechin, gallic acid, p-hydroxy benzoic acid and ferulic acid in zein films rendered antioxidant potential to the films, since a considerable portion of the phenolic compound in the films exists in soluble form.

#### 3.5.2. DPPH radical scavenging activity

DPPH radical has been widely used to test the ability of compounds as free radical scavengers or hydrogen donors in order to evaluate the antioxidant activity. Fig. 5 shows the DPPH radical scavenging activity of the Ch–PVA films incorporated with ME/PE.
The control films (not containing ME/PE) did not show any radical scavenging activity (data not shown). Films containing ME had significantly higher ($p < 0.05$) antioxidant activity as compared to those containing PE (Fig. 5). Temperature was a major factor in the release of natural extracts from the Ch–PVA films. At higher temperature ($37 \, ^{\circ}\mathrm{C}$), ME/PE was released more from the films while at lower temperatures ($15 \, ^{\circ}\mathrm{C}$) the antioxidant activity of these films were significantly reduced due to the slow release of the extracts from the film. When the films were kept at $37 \, ^{\circ}\mathrm{C}$, the ME/PE was released from the film in the first half an hour and after that the release was rather constant as further incubation (24 h) did not significantly increase the antioxidant potential of these films. Similar trend has been reported by Mayachiew and Devahastin (2010) who found that the release of Indian gooseberry extract from Ch films was rapid during the initial phase (0–8 h) and then was rather constant in the second period (8–24 h).

3.6. Antimicrobial activity of films

The direct incorporation of antimicrobial additives in packaging films is a convenient methodology by which antimicrobial activity can be achieved. The antimicrobial activity of the Ch–PVA films against the selected microorganisms is presented in Fig. 6. All the Ch–PVA composite films showed significant ($p < 0.05$) antibacterial activity towards S. aureus and B. cereus. The exact mechanism for the antibacterial activity of Ch is not known. One of the reasons for the antimicrobial character of Ch is its positively charged amino group which interacts with negatively charged microbial cell membranes, leading to the leakage of proteinaceous and other intracellular constituents of the microorganisms (Shahidi, Arachchi, & Jeon, 1999).

All the films were ineffective against Gram negative E. coli and P. fluorescens. This could be due to the cell wall lipopolysaccharides of Gram-negative bacteria, which may prevent active components from reaching the cytoplasmic membrane (Ouattara, Simard, Holley, Piette, & Bégin, 1997). This finding matches with previous works in which Gram negative bacteria seemed to present higher resistance against Ch (Fernandez-Saiz, Lagaron, Hernandez-Muñoz, & Öcio, 2008). Concentration of PVA did not have any effect on the antimicrobial activity. Inherent antibacterial properties and film forming ability of Ch make it an ideal choice for use as a biodegradable antimicrobial packaging material that can be used to improve the shelf-life of perishable foods (Ouattara, Simard, Piette, Begin, & Holley, 2000; Rao, Kanatt, Chawla, & Sharma, 2010).

Incorporation of PE had significant effect on the antibacterial activity of Ch–PVA films. In case of PE containing films it was seen that the films totally inhibited the growth of B. cereus and reduced the number of S. aureus by 2 log cycles (Fig. 6). This enhanced antibacterial activity of PE containing films could be explained due to the inherent antibacterial activity of PE which has been reported (Kanatt et al., 2010). Other researchers have also reported the improvement in the antibacterial activity of Ch containing films by addition of natural extracts. Mathew and Abraham (2008) showed that the shelf-life of food has been extended by ferulic acid incorporated starch–chitosan blend films. Similarly, incorporation of garlic oil and nisin enhanced the antimicrobial activity of chitosan film (Pranoto, Rakshit, & Salokhe, 2005).

4. Conclusions

This study demonstrated that active Ch–PVA films can be made by incorporation of ME/PE. The extracts introduced excellent antioxidant activities to the films. The release of phenolics from the films was temperature dependent and maximum amount of phenolics were released from the films at $37 \, ^{\circ}\mathrm{C}$. The incorporation of these extracts into the Ch–PVA films also improved their tensile strength without significantly affecting their barrier properties. The films showed antibacterial activity against Gram-positive food pathogens. Active packaging is a promising system for the improvement of quality and shelf-life extension of food. Thus, this study demonstrates that the addition of natural extracts to bioactive biopolymers has great potential for being developed into functional packaging material for food and is a promising substitute for synthetic materials. Further studies on the use of these films in meat packaging are being carried out.

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