



Enhanced Performance of Chitosan Film Containing Vinyl Imidazole-Hydroxyethyl Methacrylate Copolymers

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Abstract

Synthesis of vinyl imidazole (VI) and hydroxyethyl methacrylate (HEMA) copolymers in different molar ratios with high yield were synthesized using free radical polymerization. The obtained copolymers were incorporated into chitosan to prepare blend films with enhanced performance. Characterization of the produced films *via* ATR-FTIR, SEM and TGA in addition to the mechanical performance, swelling behavior and vapor water permeability were investigated. Incorporation of the copolymer in chitosan films decreased the hydrophilicity and water vapor permeability. The blend films showed enhanced thermal stability and elongation at break with slightly decline in tensile strength. The antibacterial performance of the blend films was evaluated using gram positive and gram negative bacteria inhibition zone test that showed enhanced performance of blend film containing high molar ratio of VI. The results revealed that the obtained blend films can be used in different biological application as in food packaging system.

Keywords: Vinyl imidazole; Hydroxyethyl methacrylate; Chitosan; Antibacterial film; Food packaging.

1. Introduction

Chitosan (Ch) is the deacetylated form of the chitin natural biopolymer [1], and primarily composed of 2-amino-2-deoxy-D-glucopyranos (D-glucosamine), units. It is regarded to be promising polysaccharide for biomedical uses [2]. This naturally occurring cationic polymer is an important raw ingredient for the preparation of graft copolymers due to its distinctive structure and characteristics. Excellent biocompatibility, biodegradability, and absence of allergic and inflammatory reactions are all displayed by chitosan and its derivatives [3–7]. Chitosan showed great and significant efficiencies as antibacterial, antifungal, antioxidant, anti-inflammatory and anti-tumor activities [4,8,9]. It has recently gained recognition as one of the most innovative biomaterials for vascular surgery, medication administration, tissue engineering, haemostatic use [10–12], the domains of food [1,13]

and treatment of wastewater [14–16]. Its chemical composition includes extremely reactive free function hydroxyl and amino groups that significantly facilitates its chemical alteration. Several function groups can be used in chitosan functionalization. Grafting, addition, coupling, cross linking, and other methods are examples of possible functionalization [17–19]. Polymer mixtures offer an innovative method for creating novel polymeric constituents with promising and preferred properties, without any requirement to synthesize totally new materials. Composites and copolymers of polysaccharides as chitosan with other natural or synthetic polymers are of great importance for biomedicine fields [20,21]. Poly(2-hydroxyethyl methacrylate), pHEMA, is hydrophilic, optically transparent, biocompatible, and non-degradable synthetic polymer [22–25]. The use of pHEMA in various biomedical applications has been developed

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such as soft contact lenses, artificial skin, artificial corneas, wound dressings, bone tissue regeneration, ocular and controlled drug delivery systems, catheters, breast augmentation and intrauterine inserts [26–28]. The pHEMA becomes swollen forming a hydrogel upon water absorption due to its hydrophilic pendant group, yet when dry, pHEMA is a rigid and brittle material. Transparent pHEMA-based hydrogels are highly permeable to small molecules and allow the flow of fluids and oxygen [29,30]. Different methods of polymerization and copolymerization can change the mechanical and optical characteristics, oxygen permeability, and water absorption of pHEMA-based hydrogels [26]. HEMA can be polymerized or copolymerized using thermal and photochemical polymerization techniques. Pure pHEMA is not antimicrobial against several gram-positive and gram-negative microorganisms, thus antimicrobial techniques can be used to perform antimicrobial medical devices to fight various pathogens [31,32]. Antimicrobial agents can be incorporated in several ways into pHEMA medical devices. Tetracycline, erythromycin, chloramphenicol, and ciprofloxacin are examples of antibiotics that can be used to use antimicrobial medical devices, such as contact lenses. Antibiotics action by directly eliminating or preventing the growth of microorganisms on the cornea and contactlenses [33,34]. N-vinyl imidazole is an important functional monomer because of its high biocompatibility and promising antibacterial action [35]. Furthermore, poly(N-vinyl imidazole) is a non-harmful polymer suitable for use in a range of biomedical applications such as medication and gene therapy [36], and delivery systems [37]. Macromolecular scientists are very concerned about N-vinyl imidazole copolymers because they can prepare new biopolymers with gigantic biological activity [38–42]. Moreover, N-vinyl imidazole played a crucial role in the synthesis of molecules with biological activity[43]. Grafting of N-vinyl imidazole was previously carried out onto different natural polymer as chitosan and carboxymethyl chitosan [44], to improve their antimicrobial activity [45]. This work aims to synthesize of copolymer based on N-vinyl imidazole (VI) and 2-hydroxyethyl methacrylate (HEMA) with different mole ratios followed by their mixed with chitosan to prepared

blend films. Studying the structural, mechanical, and biological properties of the prepared films (chitosan/VI-co-HEMA) will be investigated.

This work aims to synthesize N-vinyl imidazole (VI) / 2-hydroxyethyl methacrylate (HEMA) copolymer with different mole ratios followed by their blend with chitosan to prepared (Ch /VI-co-HEMA) films. The structural, mechanical, and biological properties of the prepared films (chitosan /VI-co-HEMA) were investigated. Evaluation of the blend films was carried out using ATR-FTIR, SEM, TGA, beside their swelling, porosity measurements, mechanical properties and water vapor permeability. The biological activity of prepared blend films against gram positive and gram negative pathogens was studied.

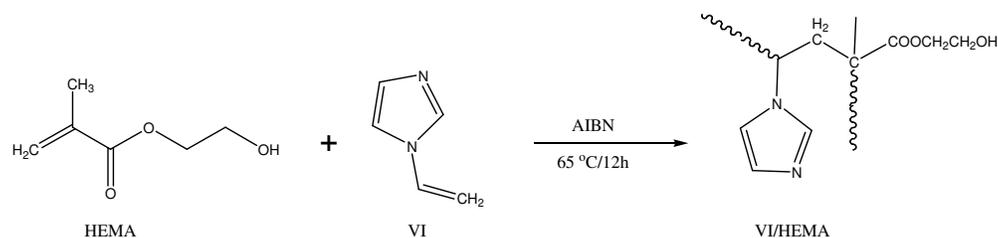
2. Experimental

2.1. Materials:

Vinyl imidazole and hydroxyethyl methacrylate monomers were delivered from Aldrich. Chitosan (medium molecular weight MW: 300kDa, Deacetylate 85%) was obtained from Aldrich. Azobis-isobutyronitrile (AIBN) was supplied by Sigma Aldrich and crystallized from ethanol to be ready for the experiments. Dioctyl phthalate was provided from Aldrich. Acetic acid, glycerol, and all other compounds were of analytical grade

2.2. Copolymerization process

Copolymerization processes were performed using N-vinyl imidazole (VI) and 2-hydroxyethyl methacrylate (HEMA) monomers to afford the copolymer VI/HEMA. 0.05 mole of the monomer mixture that dissolved in the least amount of freshly distilled THF, the reaction container was kept in an ultrasonic bath for about 5 minutes. Addition of AIBN as initiator (0.2% by weight of monomers) dissolved in 5 ml of freshly distilled THF was then carried out. The copolymerization reaction was maintained for 12h at 65 °C, then transferred to an ice bath, and about 20 ml of solvent (THF) was added. The copolymer was poured into the cyclohexane as non-solvent to be precipitated; the product was filtered and dried at 70 °C under vacuum. Three molar ratios were applied and the copolymerization procedure was performed using the components listed in Table 1. Poly(2-hydroxyethyl methacrylate), pHEMA, as homopolymer was also prepared. Scheme 1 represents the preparation of VI/HEMA copolymer.



Scheme 1. Preparation of VI/HEMA copolymer.

Table 1: Weights ratio of monomers used in copolymerization

Monomer Composition VI/HEMA (Molar ratio)	Mole %	Weight (g)	Codes of copolymerblends
75/25	0.0375:0.0125	3.528:1.626	VI/HEMA 75:25
50/50	0.0250:0.0250	2.352:3.252	VI/HEMA 50:50
25/75	0.01250:0.0250	1.176:4.878	VI/HEMA 25:75
100/0	0.05	6.5	p(HEMA)

2.3. Preparation of the blend films

Chitosan solution was made by solubilizing (2g) of chitosan powder in 100 ml 2% aqueous acetic acid overnight while stirring till complete dissolution. The copolymers were separately dissolved in 10 % acetic acid aqueous solution under vigorous stirring at 60 °C for a day to obtain clear solution. The soluble copolymers were added to 20 ml chitosan solution in separate containers keeping the ratio of the copolymer to be 10% of the chitosan weight under continuous stirring to assure complete mixing of the polymer mixture. Glycerol (5% respects to chitosan weight) and an equal weight of dioctyl phthalate were added to each solution as plasticizers. After complete mixing of the solutions, they poured onto 10 cm diameter petri dish and let them dry in an oven at 50 °C for a day.

2.4. Characterization techniques

2.4.1. Infrared (FTIR) spectroscopy

The reactive function groups of the chitosan-copolymer blend films were assessed using an Attenuated Total Reflectance Fourier Transform Infrared spectrophotometer (ART-FTIR, Shimadzu

8400, Japan) within spectral range between 400 and 4000 cm^{-1} .

2.4.2. Scanning electron microscopy

The surface and cross-section topography of the prepared films were explored using high resolution scanning electron microscope (HR-SEM, QUANTA FEG 250 ESEM). Prior to examination, the prepared films were covered with gold vapor via Sputter Coater (S150A) Edwards.

2.4.3. Mechanical properties of prepared films

Mechanical properties (tensile strength and elongation at break) for the blend films of chitosan and VI/HEMA copolymers were tested using Zwick/Roell Z010, type X force P, S/N: 760608, Germany. The polymer films were dried and cut into dumbbell shape to evaluate its mechanical performance. The tests were carried out at a crosshead speed of 2 mm/min at 25°C.

2.4.4. Swelling behavior of chitosan films

The swelling of chitosan film with and without VI/HEMA copolymer was obtained by immersion 5x5 cm^2 for 3 samples of each film in distilled water

for a day at room temperature and weighted wet, then the films were dried at 100°C until constant weight[46]. The swelling was assessed by the equation (1):

$$Ws = (W_{\text{wet}} - W_{\text{dry}} / W_{\text{dry}}) \times 100 \quad (1)$$

Where W_{wet} and W_{dry} are the weight of swelled and dried films samples, respectively

2.4.5. Porosity of chitosan films

The porosity of the blended chitosan films was explored using dry-wet weight process[47]. The weight of the films in the wet state was assessed after removal the excess adsorbed water. The wet film was dried at 60°C for a day and its weight in dry state dry was documented. The porosity of Films was assessed as follow.

$$\varepsilon (\%) = [(W_{\text{wet}} - W_{\text{dry}}) / d_w \cdot A \cdot h] \times 100 \quad (2)$$

Where ε is the porosity of the film, d_w (g/cm³) is the density of distilled water (0.998), A (cm²) is the film surface area in the wet state and h (cm) is the film thickness in the wet state.

2.4.6. Thermal gravimetric analysis

Thermal stability of the blend films (TGA) was performed using Perkin-Elmer instrument (SDTQ600, TA Instrument, USA) in nitrogen gas as an inert atmosphere in temperature range from 40°C to 500°C with heating rate of 10 °C/min.

2.4.7. Water vapor permeability

For evaluation the water vapor permeability, a certain amount of pure water was hosted in vials, then the vials were fixed tightly with the blended film samples and weighted. The vials weight was monitored overtime interval and water vapor permeability rate was assessed via the following equation

$$WVP = \Delta w \cdot L / \Delta t \cdot A \cdot P \quad (3)$$

Where ($\Delta w / \Delta t$) is the flux assessed as weight loss of the vials per time and measured from the slope of the weight differences of the vials, to the nearest 0.0001 g, versus time (h), A (m²) is the active surface area of the sheet, L (mm) is the sheet thickness and P : (kPa) is the water vapor pressure differential calculated as 4.245 KPa at 30 °C [48].

2.5. Antibacterial properties

The antibacterial performance of blended chitosan film against three strains of Gram-positive bacteria (*Bacillus Subtilis*, *Staphylococcus aureus* and *Streptococcus faecalis*) and three strains of Gram-negative bacteria (*Escherichia coli*, *Neisseria gonorrhoeae* and *Pseudomonas aeruginosa*) were investigated using disc-diffusion methods in comparison with the reference drugs ampicillin and Amphotericin B as antibacterial and antifungal standards [15]. Agar diffusion plate examination was used to assess the antibacterial performance, the bacteria strains were cultivated on nutrient agar and incubated at 37 °C for a day. 100 µL of dilute bacterial cell suspension was extended onto Petri dishes and the films were adhered on the agar plate surface. The plates were incubated overnight at 37 °C. The inhibition zone thickness was measured.

3. Results and discussion

3.1 Characterization of the prepared copolymers and its chitosan/blend films

3.2.1. Fourier transforms infrared (FT-IR) analysis

The FT-IR technique is useful for detecting the reactive functional groups of materials as our copolymer; vinyl imidazole and hydroxyl ethyl methacrylate (See Figure 1). The spectra of the synthesized VI-HEMA copolymers showed the disappearance of -CH Alkenes stretching at 3025 cm⁻¹ and -C=C- at 1640 cm⁻¹ indicating the successful polymerization process. On the other hand, a new sharp peak for the -OH group, at 3397 cm⁻¹ for copolymer (VI-HEMA) 25:75, and the peak intensity with broadening of the peak at 3403 cm⁻¹ for 50:50 and 3401 cm⁻¹ for 75:25[49] indicating the formation of hydrogen bonding of the HEMA polymer. The peak at 1716 cm⁻¹ for VI-co-HEMA with high ratio of HEMA was related to carbonyl groups of acrylates, whereas the band at 1625 cm⁻¹ was corresponding to -C=N groups of imidazole.

The interactions between the molecules of the chitosan with VI-HEMA blends were also analyzed by FT-IR measurements and displayed in Figure 2. FT-IR spectrum of plain film exhibited a broad band at 3432 cm⁻¹ that can be correlated to stretching vibrations of -OH and -N-H groups found in chitosan and copolymers[50,51]. The band at 2825 cm⁻¹

corresponds to the asymmetric stretching vibrations of -CH groups in the both polymer chains. The bands at 1716 cm^{-1} correspond to symmetric stretching of carbonyl (-C=O) of acrylate group while the peak at 1638 related to -C=O stretching vibrations of acetamide group of unhydrolyzed part of chitosan.

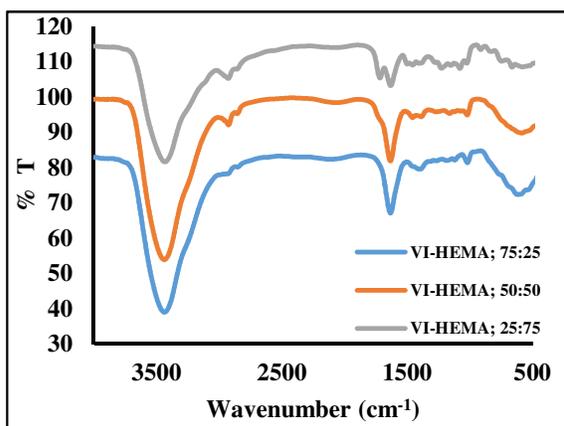


Figure 1. FTIR spectra of prepared copolymers of (VI) and HEMA

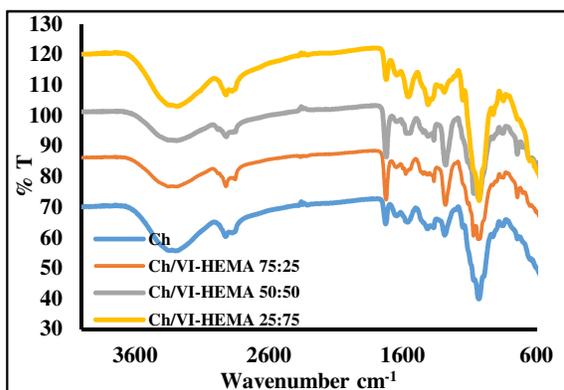


Figure 2. FTIR spectra of pure chitosan and blend chitosan with the copolymer

3.2.2. Thermal properties of the films

Thermal gravimetric analysis (TGA) of the films in inert atmosphere within the range 50 and 500 °C are displayed in Figure 3. The degradation temperatures and mass losses of the films at various temperature ranges are assessed. Chitosan film displayed three degradation steps; which matched with previous studies[52]. The first degradation step occurs within the range of 50 and 110 °C related to the evaporation of adsorbed water with mass loss of 5%. The second

degradation step ranging from 120 to 240 °C with mass loss of 23% that related to the evaporation of ammonia and deacetylation of chitin part of chitosan in addition to degradation of glycerol and other additives. The third degradation stage, took place within range 240 and 410 °C, was correlated to the pyrolysis of chitosan main chains with ash content about 28%. The TGA profiles of prepared blend films of chitosan with VI/HEMA have the same pattern as chitosan with more thermal stability relative to blank chitosan film. The thermal stability of blend film containing VI/HEMA with ratio of 75:25 was the more stable one that may be due to the presence of aromaticity of the imidazole ring with high thermal stability. The ash content of this blend film was about 31% after 500 °C. These results indicated that the blend film was one phase with relative high thermal stability.

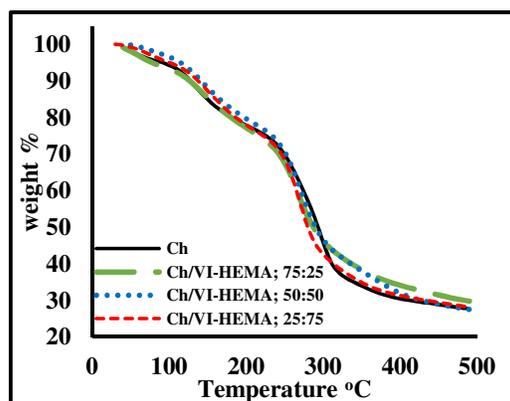


Figure 3. Thermal gravimetric analysis of the prepared chitosan blend films

3.2.3. Mechanical performance of the prepared chitosan blend films

As presented in Table 2; the incorporation of VI/HEMA copolymers into the chitosan matrix resulted in slight decrease in tensile strength with enhancement in elongation at break. The elongation at break was slightly increased as the flexibility of copolymer chains relative to the carbohydrate structure of chitosan. The tensile strength (TS) that is dependent on microstructure and inter molecular forces (as hydrogen bonding) between chains in the continuous phase showed slight decrease of chitosan film due to the presence of another polymer that weakened the hydrogen bonding of the chains[53].

Changing the ratios of vinyl imidazole and hydroxyethyl methacrylate may have resulted in reduction of TS; with the more stable reading related to the copolymer with high VI content. The young's modules showed the same behavior like the tensile strength. The results indicated that the blend films weren't affected largely by mixing the copolymer with chitosan matrix, and slight decrease in mechanical performance that didn't affect the film performance during different application.

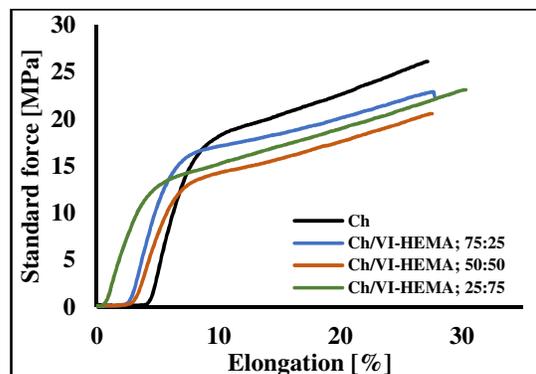


Figure 4. Mechanical properties of the prepared films

Table 2: Mechanical Performance of the prepared derivatives

Sample	Thickness (mm)	TS (MPa)	% E	YM (MPa)
Ch	0.12	26.1	27.1	0.22
Ch/(VI:HEMA) 75:25	0.15	22.9	27.6	0.11
Ch/(VI:HEMA) 50:50	0.14	20.5	27.5	0.14
Ch/(VI:HEMA) 25:75	0.15	23.1	30.2	0.15

3.2.4. Morphology of the prepared films

The surface and cross-section morphology of chitosan with and without the VI/HEMA copolymers were investigated using scanning electron microscope as indicated in figure 5. The film surface of pure chitosan and all blended films were appeared with some circular spots due to the presence of different component chitosan, VI-HEMA copolymer and the oily plasticizers; glycerol and dioctylphthalate (DOP). The later plasticizer has oily characteristics which resist its compatibility and miscibility with hydrophilic chitosan polymer. Blank chitosan film has more pores and larger circular spots than the other blend films indicating the presence of VI-HEMA copolymer slightly enhance homogeneity of the film and decrease the porosity. The film cross-section of blank chitosan has sponge like structure with more pores, while the blend films have more compact cross-section structure than blank chitosan with appearance of some macro voids. The more compacted cross section structure was the film containing high ration on HEMA this may be the

more hydrogen bonding between the chitosan and more functional groups of HEMA chains.

3.3. Swelling and water vapor permeability of the prepared films

Swelling is an indicator of the hydrophilicity (affinity) or hydrophobicity (resistance) of the film samples towards water, which is a significant factor for food packaging and bio-application because the possibility of contamination in the existence of water [46]. As can be seen in Table 3 Copolymers of vinyl imidazole and hydroxyethyl methacrylate decreased the water swelling performance of polysaccharide (chitosan) films. The increased of vinyl imidazole content in the copolymer improved the water resistance of film-forming solution by decreasing the swelling performance. For food packaging film, governing the barrier performance of films is required to prolong the shelf-life of food[50]. As shown in table 3, chitosan films showed relatively WVP value (1.17 g.mm/m² h kPa) of the single-component films. The addition of VI-HEMA copolymer to chitosan matrix relatively decreased the water vapor permeability and the decreased is higher

for the high content of vinyl imidazole moiety in the copolymer. The copolymer VI- HEMA with ratio (75:25) to chitosan film showed the least water vapor permeability and is considered the most promising polymer film with enhanced performance regarding the mechanical, thermal and vapor permeation properties.

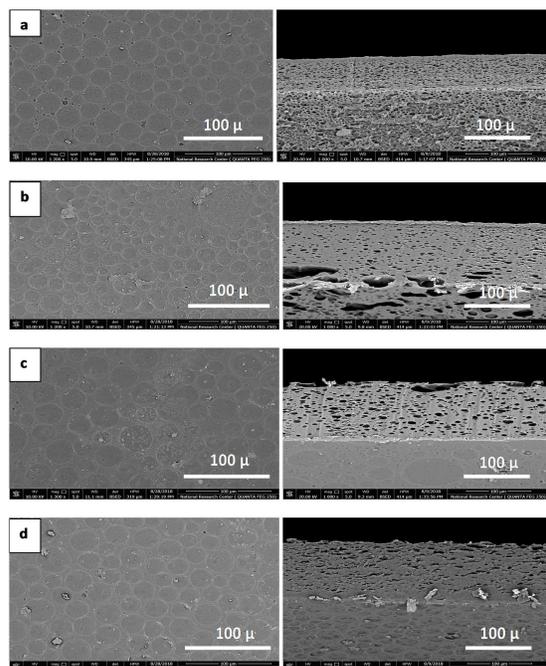


Figure 5. SEM of surface (left) and cross-section (right) of the blend films for a) Chitosan, b) Ch/VI-HEMA 75:25, c) Ch/VI-HEMA 50:50 and d) Ch/VI-HEMA 25:75

Table 3: Swelling and water vapor permeability of the blend films.

Film type	Swelling %	WVP (g.mm/m ² h.kPa)
Blank Chitosan	39.6	1.17
Ch/VI- HEMA; 25:75	33.9	1.17
Ch/VI- HEMA; 50:50	34.2	1.09
Ch/VI- HEMA; 75:25	34.0	1.07

3.4. Antibacterial performance

- Antimicrobial activity of Ch/p(VI/HEMA) copolymer blends

The in vitro antibacterial activity of the Chitosan with (VI-co-HEMA) blends with the various molar ratios of VI in the copolymer against three strains of Gram-positive bacteria (*Staphylococcus aureus*, *Bacillus Subtilis* and *Streptococcus faecalis*) and three strains of Gram-negative bacteria (*Neisseria gonorrhoeae*, *Escherichia coli* and *Pseudomonasaeruginosa*) were investigated using disc-diffusion methods in comparison with the reference drugs ampicillin and Amphotericin B as antibacterial and antifungal standards. The obtained results of antibacterial and antifungal activities are shown in Table 4 and Table 5, respectively. The given results of Table 4 clearly revealed that the investigated chitosan/copolymer blends in this study have relatively promising antibacterial efficiencies. This activity was found to be significant against the three Gram-positive strains other than the Gram-negative ones. Concerning the Gram-positive strains (*Bacillus Subtilis*, *Staphylococcus aureus* and *Streptococcus faecalis*), the antibacterial activity of chitosan/copolymer blends under investigation clearly showed that the lowest activity was related to the chitosan/homopolymer blend, Ch/p(HEMA), that has no vinyl imidazole content. This activity reached 35, 33 and 22% against the three Gram-positive strains respectively and these results are with respect to Ampicillin as reference standard antibacterial agent. The Ch/VI-HEMA blend that containing molar ratio of 25% vinyl imidazole in the copolymer exhibits an increase in the antibacterial effect against the tested Gram-positive strains to be 45, 61 and 44% when compared to the reference drug. The Ch/Copolymer blend, with vinyl imidazole content of 75% molar ratio, shows notable increase in the antibacterial efficiency against the same Gram-positive bacterial strains to become 65, 78 and 56%. On the other hand, results of antibacterial activity of the chitosan/homopolymer p(HEMA) and the copolymer p(HEMA)/VI blends, with its various molar ratios, show relatively lower efficiencies against the three Gram-negative bacterial strains (*Escherichia coli*, *Neisseria gonorrhoeae* and *Pseudomonas aeruginosa*). It is found that the Ch/(VI-HEMA) blends revealed antibacterial activity against these types of bacterial strains higher than that the Ch/p(HEMA) and this can be attributed to the absence of any imidazole contents in the blend.

The same Ch/p(HEMA) and Ch/(VI-HEMA) blends with the various molar ratios of VI in the copolymer were examined as antifungal agents against two types of fungi, *Candida albicans* and *Aspergillus flavus* using amphotericin B as a model drug. They have good antifungal performance towards the two types of fungi, *C. albicans* and *A. flavus*. The Ch/p(HEMA) and Ch/(VI-HEMA) blends showed significant antifungal activity against the *C. albicans* that ranges from 47% for Ch/p(HEMA) and for the other Ch/copolymer blends with the various vinyl imidazole contents were 63, 79 and 89% respectively. So, the increase of the molar ratio of vinyl imidazole in the copolymer enhances the antifungal activity of the Ch/copolymer blends against the *C. albicans*. The other tested fungi, *A. flavus* were affected by the same prepared chitosan/copolymer blends but with lower extent than *C. albicans*. Both antibacterial and antifungal activities of the chitosan/copolymer blends under investigation can be explained on the bases of the chemical structure, function groups and the percentages of the components present in these blends. The main component of the investigated blends is chitosan which a promising natural polymer with antibacterial properties and bactericidal activity. Furthermore, chitosan was an ideal compound for medical science due to its antibacterial properties, biocompatibility, and low toxicity [54,55]. The

positive charges present on the amino groups of chitosan interact electrostatically with the negatively charged species on the microbial cell membrane to give antimicrobial character [56].

The most expected important mechanism for antimicrobial actions of chitosan may be due to the electrostatic contact between the polycationic chitosan and microbial cell surface [57]. The observed antimicrobial activity of the investigated chitosan/copolymer blends also can be due to the presence of n-vinyl imidazole units in the copolymer since polymers comprising the imidazole ring are biocompatible, biodegradable and exhibited antibacterial performance [58–61]. The imidazole offer not only antibacterial activity but they inhibit enzymes and kill fungal pathogens [62]. Mechanistically, the antibacterial activity of VI can be illustrated by the electrostatic interactions between the positively charged VI and negative charge of bacterial cell membranes as well as its hydrophobic interactions with the cytoplasmic membrane of bacteria [63]. So, the antimicrobial activity of the chitosan/copolymer, Ch/(VI-HEMA) exhibited higher antimicrobial activity than the Ch/pHEMA. At the same time the increase of VI molar ratios in the copolymer gave rise to enhancement of the antimicrobial potency of the investigated blends.

Table 4: Antibacterial activity of chitosan film with various molar ratios of HEMA/NVI copolymers blends

Sample	G +ve Bacteria				G -ve Bacteria	
	<i>B. Subtilis</i>	<i>S. aureus</i>	<i>S. faecalis</i>	<i>E. coli</i>	<i>N. gonorrhoeae</i>	<i>P. aeruginosa</i>
DMSO	0.0	0.0	0.0	0.0	0.0	0.0
Ampicillin (100 µg/mL)	20 ± 0.25	18 ± 0.22	18 ± 0.17	22 ± 0.28	20 ± 0.22	17 ± 0.19
Ch/p(HEMA)	7 ± 0.12	6 ± 0.34	4 ± 0.10	8 ± 0.31	5 ± 0.18	4 ± 0.17
Ch/VI-HEMA (25:75)	9 ± 0.33	11 ± 0.21	8 ± 0.16	10 ± 0.19	8 ± 0.23	7 ± 0.11
Ch/VI-HEMA (50:50)	12 ± 0.27	12 ± 0.19	9 ± 0.08	10 ± 0.22	11 ± 0.14	10 ± 0.09
Ch/VI-HEMA (75:25)	13 ± 0.18	14 ± 0.32	10 ± 0.23	11 ± 0.26	11 ± 0.23	11 ± 0.25

Table 5: Antifungal activity of chitosan film with various Molar ratios of HEMA/VI copolymers blends

Sample	<i>C.albicans</i>	<i>Asp.flavus</i>
DMSO	0.00	0.00
Amphotericin B (100µg/mL)	19 ± 0.20	17 ± 0.23
Ch/p(HEMA)	9 ± 0.18	6 ± 0.27
Ch/VI-HEMA (25:75)	12 ± 0.32	9 ± 0.21
Ch/VI-HEMA (50:50)	15 ± 0.25	11 ± 0.32
Ch/VI-HEMA (75:25)	17 ± 0.16	12 ± 0.22

4. Conclusion

The synthesis of vinyl imidazole-hydroxyethyl methacrylate copolymers was successfully carried out and blended with chitosan to obtain antibacterial films. The prepared films were well characterized using SEM, FTIR and TGA, in addition to swelling and porosity measurement. The obtained blend films displayed porous structure with enhanced thermal stability compared to blank chitosan film. The blend film containing VI/HEMA copolymer with high content of imidazole showed relative decrease in swelling performance and water vapor permeability which endorse these films in food packing application. The tensile strength was slightly decreased whereas the elongation at break was enhanced. The antibacterial activity of the blend films against gram positive, gram negative and fungi was enhanced by incorporation of the VI/HEMA copolymer with high ratio of vinyl imidazole.

Conflict of interest

The authors decline that there is no conflict of interest.

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