Modification of Chitosan towards a Biomaterial with Improved Physico-Chemical Properties

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ABSTRACT: Modification of chitosan (CTN), a natural biomaterial, leading to improved physical and chemical properties makes the polymer overcome the limitations on its application in biomedical fields. We have modified CTN using polyvinylpyrrolidone (PVP) in three ways, i.e., by blending (CTN : PVP), by graft copolymerization [poly(CTN-g-PVP)], and by free radical induced crosslinking [poly(CTN-cl-PVP)], varying the CTN to PVP ratio. The properties of all the modified and unmodified CTN studied included structural, thermal, mechanical, and water uptake capacity. Modification by blending imparted enhanced strength, toughness, glass transition temperatures, and thermal stability to the dry membranes. The equilibrium water content (EWC) of the membrane gels was investigated as a function of pH and ionic strength. Swelling ratio of the chemically modified membranes were found to be less sensitive to pH, with higher % EWC maintaining the gel integrity in both acidic and neutral media unlike the plain CTN. The hydrogen bonding and free volume of the films were found to play a major role in determining the properties of the type of modification. CTN membranes with enhanced water uptake capacities clubbed with optimum mechanical properties shall be a suitable choice for biomedical and controlled release applications. © 2006 Wiley Periodicals, Inc. J Appl Polym Sci 101: 63–69, 2006

Key words: chitosan; polyvinylpyrrolidone; graft copolymers; swelling ratio; mechanical properties

INTRODUCTION

Chitosan (CTN), a natural polyaminosaccharide, is widely used because of its favorable characteristics such as biocompatibility, biodegradability, renewability, and derivatisability.[1] In drug delivery, the requirement for a biomaterial is good mechanical strength and fast response time in a physiological environment. The mucoadhesive property is influenced by the surface charge, which should sustain the basic and neutral environments. For application as catheters, excellent tensile and mechanical properties are necessary. Although CTN has been widely used in several applications, its restricted dissolution characteristics, sensitivity to pH, and brittle nature of the film narrows down its applications in certain fields. Scope for modification of the polymer is enormous to suitably tailor-make the properties based on the type of application. Blending with another biocompatible polymer is a simple method of physical modification to tailor-make the properties of CTN as per requirement.[2–4] Blending CTN and polyvinylalcohol (PVA), a water-soluble

polymer, was done to improve the solution properties of CTN.[5] The thermo mechanical properties of CTN–starch and CTN–pullulan blends were reported by Lazaridou and Biliaderis,[6] wherein the relationship between the moisture and T_g of the films was studied. The swelling of CTN/cashew nut gum gels were studied by Paula et al.[7] and found that the carbohydrate groups on cashew nut play an important role.

Grafting is a promising approach of chemical modification wherein synthetic monomers with different functional groups can be attached to CTN, resulting in a wide spectrum of properties.[8,9] Effect of the monomer concentration, initiator percentage, reaction temperature, and time on the graft copolymerization of methacrylic acid onto carboxymethyl CTN to obtain optimum conditions of grafting was studied by Sun et al.[10] Chemical modification by grafting was done by Kurita et al.,[11] with mostly acid based membranes to improve water solubility, resulting in alteration of polymer bioactivities. Grafting of N,N-dimethyl-N-methacryloxyethyl-N-(3-sulfopropyl) ammonium onto CTN was done by Zhang et al.[12] and they arrived at the optimum reaction conditions to obtain a maximum of 50% grafting. Acrylamide and acrylic acid monomers were grafted onto chitin, using cerium (IV) as initiator, which resulted in improved hygroscopicity in comparison to chitin. It is evident that most of the graft copolymerization research work is focused on arriving at the optimum parameters to obtain efficient grafting percent-

age.[13] The possibility of generating free radicals on polymers to obtain crosslinking between two polymers is another suitable chemical modification method. Free radical-induced crosslinking in polymers by radiolysis has been studied by Sonntag[14] in polymethylmethacrylate and polyacrylic acid etc. He has reported that dose rate and polymer concentration play a major role in getting the crosslinked product. A comparative study on improvement in the properties and performance of CTN with respect to the modification would be of use to make appropriate choice of the modification type.[15]

The present study investigated the modification of CTN by blending with PVP (biocompatible hydrophilic polymer) and by graft copolymerization with *n*-vinyl 2-pyrrolidone.[16] We have also explored the chemical modification of CTN by free radical-induced crosslinking between the CTN and PVP homopolymers using cerium (IV). The modifications provide scope for variation in intra and intermolecular interactions through hydrogen bonding. The effects of concentration of PVP in the modified membranes on the swelling characteristics with respect to pH and ionic strength, mechanical, and thermal properties are reported.

EXPERIMENTAL

Materials and methods

CTN 90% deacetylated, *N*-vinyl-2-pyrrolidone (VP), and polyvinylprrolidone (PVP) K 90 were purchased from Sigma Aldrich Company. Ceric ammonium nitrate (CAN) and all other chemicals, like acetic acid, of analytical grade were purchased from S.D. Fine Chemicals, India. *N*-Vinyl pyrrolidone was used after distillation under vacuum, and the others were used as obtained.

Preparation of blends of CTN and PVP

one percent (w/v) solution of CTN and PVP in 2% acetic acid was prepared by stirring at room temperature for 12–16 h. The films of the blends having weight ratios of CTN : PVP (80 : 20, 60 : 40, 50 : 50, 40 : 60, and 20 : 80) and also the homopolymers were prepared by solution casting in plastic trays and left overnight for drying at room temperature. The dried films were placed under vacuum for 72 h at 60°C for removing the traces of acetic acid and moisture in the films.

Preparation of poly(CTN-*g*-PVP) copolymer

Hundred milliliter of 1% (w/v) CTN solution in 2% acetic acid and 0.1 g of VP was taken in a four-necked reaction kettle equipped with a stirrer and reflux condenser. The solution was degassed with nitrogen and left for 30 min for stabilization at 60°C. CAN aqueous solution (0.1*M*) was added and the reaction was carried out for 3–4 h at 60°C under nitrogen atmosphere. The reaction mixture was precipitated using 0.1*N* NaOH solution and is poured into excess of water. The graft copolymer is filtered and purified by several washings with water to remove any unreacted monomer. Poly(CTN-*g*-PVP) copolymer obtained was dried under vacuum at 60°C for 24 h.

The graft copolymer yield was evaluated by calculating the percentage of grafting (% G).

$$\%G = \left(\frac{W_2 - W_1}{W_1} \right) \times 100$$

where, W_1 and W_2 are weights of CTN and grafted CTN, respectively. The percentage of grafting was found to be close to the theoretical ones and graft copolymer yields were found to be around 90%.

Preparation of poly(CTN-*cl*-PVP)

The preparation of crosslinked CTN with PVP is done according to the procedure described in the previuous section, using 0.1 g of PVP in place of VP.

Characterization

FTIR absorbance spectra (Perkin–Elmer Co, USA) were recorded after the samples were dried in vacuum. Tensile properties were determined using a universal testing machine (UTM AGS-10-KGN, M.S.Shimadzu, Japan). The tensile strength of the film is tested by fixing the film into the two gauges. The test specimen was 5×1 cm^2, with pulling speed 10 mm/min. The average values of maximum stress and maximum strain was taken after testing 10 pieces of each measurement. Glass transition temperatures of samples were studied using Rheometric Scientific Dynamic Mechanical Thermal Analyzer (DMTA IV model). Viscoelastic studies of samples were done on rectangular films with specimen dimension of 20×10 mm^2 in tensile mode at 1 Hz frequency with a heating rate of 10°C/min. The thermal stability of CTN and modified CTN samples was determined with thermo gravimetric analyzer (851 Mettler Toledo TGA instrument). To determine the swelling percentage, the appropriate size (1×0.5 cm^2) of the films was kept in pH 7.4 buffer solution at room temperature, until the equilibrium swelling percentage was attained (1 h). The wet weight of each film was determined by blotting the film with filter paper to remove surface water and then weighed immediately on an electronic balance. The swelling percentage of the films in the media was calculated as follows:
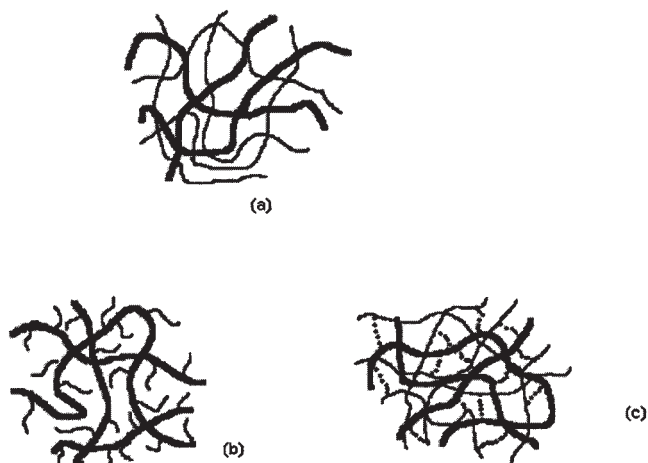
Figure 1 Schematic representation of (a) blends, (b) graft copolymers, and (c) crosslinked polymers.

$$\text{Equilibrium Water Content (EWC)}\% = \frac{Wt - Wi}{Wi}$$

where W_t is the weight of the sample at time t and W_i, is the weight of initial dry sample.

RESULTS AND DISCUSSION

Composition and structure of modified CTN membranes

The modified blend membranes form clear, transparent films in all their compositions, indicating their miscibility. Miscible binary blends of amorphous polymers have only one phase, and their properties normally reflect the weighted average properties possessed by their blend components. The CTN membranes modified by grafting and crosslinking also present good film forming nature, resulting in transparent and clear films. A schematic representation of the type of modification is given in Figure 1 and the chemistry of graft copolymerization has been illustrated in Figure 2.

Infrared spectroscopy

The structural changes in CTN due to modification were confirmed by FTIR spectroscopy as shown in Figure 3, in which the spectra for plain polymers and the modified ones are included for comparison. The main peaks of CTN at 3420 cm^{-1} are assigned to the O—H and the vibration of N—H; three strong peaks at 1030, 1080, and 1150 cm^{-1} are characteristic of the saccharide structure; and the peak at 1599 cm^{-1} shows a high degree of deacetylation, as seen in the figure.[10] The O—H stretching vibration bands for the blends are found to be shifted from 3435 to 3412 cm^{-1}, indicating the presence of intermolecular hydrogen bonding between the two homopolymers. The absorption band of C=O appears at 1680 cm^{-1} for the commercial K 90 homopolymer PVP. It appears at the same place for the blends but shifts to lower wave number (1660 cm^{-1}) in the graft and the crosslinked poly-
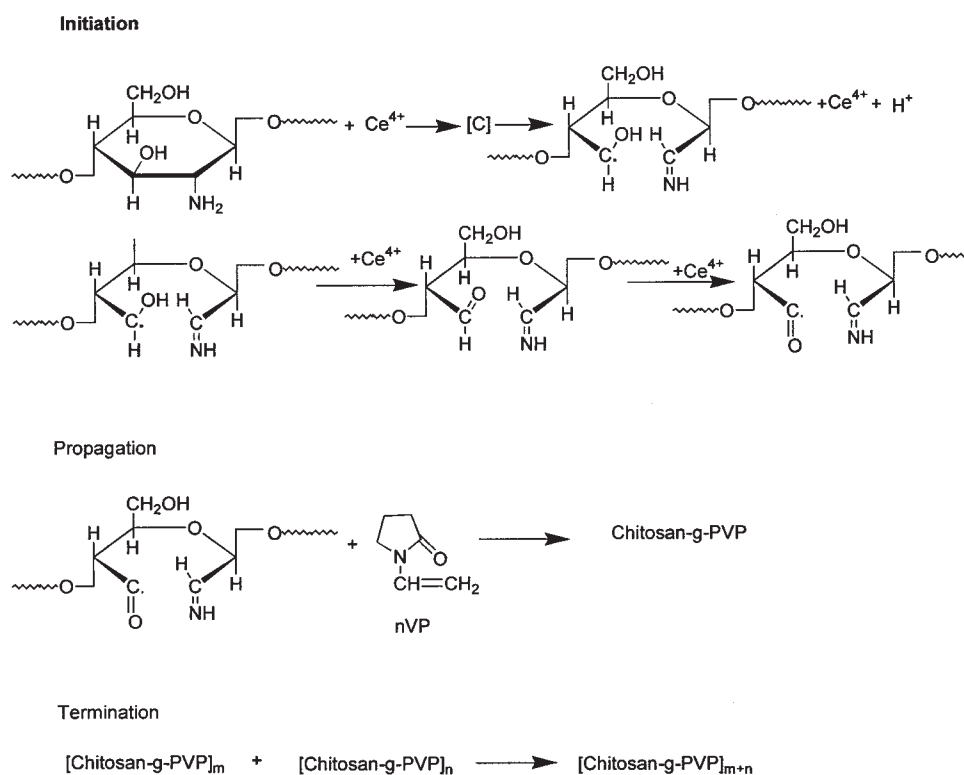

Figure 2 Schematic illustration of graft copolymerization.
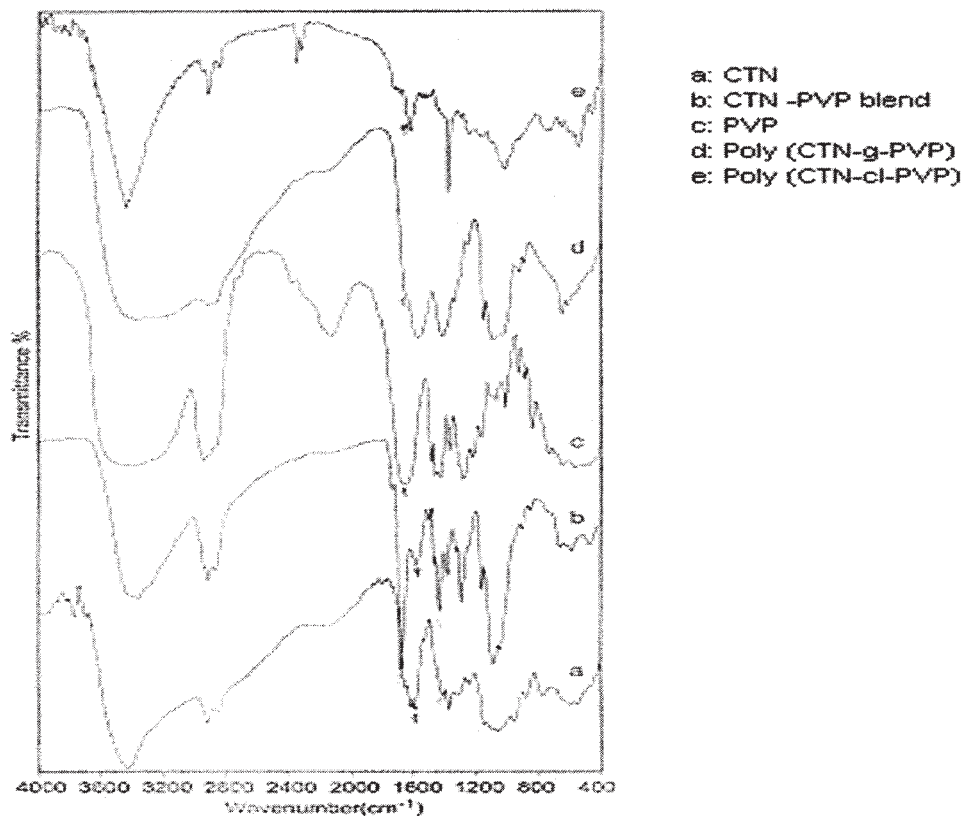
Figure 3 FTIR spectra of CTN and modified CTN polymers.

mers.[16] Thus, in the chemical modification, the covalent bond formation between PVP chains and CTN back bone is seen. The spectra of all the modified CTN samples confirm the incorporation of PVP, showing the C=O peak.

Swelling studies

The modified CTN membranes find application as hydrogels, with the ability to swell in water and retain certain amount of water within its structure without dissolving. This property is very useful for application in controlled release. Therefore, all the modified and the unmodified membranes were subjected to swelling studies to monitor the effect of modification on their percentage equilibrium water content (% EWC).

pH effect

PVP normally dissolves in water, irrespective of the pH of the medium because of its hydrophilic nature, while CTN slowly dissolves in acidic pH, swells in neutral, and precipitates in basic medium. Therefore, swelling of CTN was found to be the highest in acidic medium because of the electrostatic repulsion of the NH_3^+ ions, causing the polymer to expand. The effect

of pH and ionic strength on the % EWC of CTN and the modified ones are shown in Figure 4.

Blends

On blending with PVP, dilution of NH_3^+ charges on CTN occurs, resulting in the screening of electrostatic repulsion of the charges. Therefore, a constant decrease in the percentage of water uptake with decrease in the weight ratio of CTN in the blends was observed in acidic pH. In neutral medium, CTN is not charged and it has been reported by Paula et al.[7] that addition
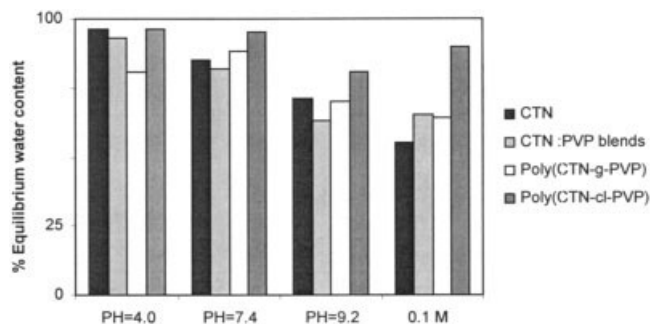

Figure 4 Effect of pH and the ionic strength (0.1M NaCl) on CTN and modified CTN films.

of a hydrophilic polymer increases the water uptake percentage. Accordingly, with increase in the weight ratio of PVP, an increase in the swelling ratio was found up to 50 : 50 in blends, but a further increase of PVP content decreased the % EWC. Because, beyond 50 : 50 compositions, the presence of higher content of water soluble PVP in the film does not permit the gel integrity, as it tends to dissolve and cause a decrease in % EWC. The area of the film increases by more than double its dry film size, with degree of swelling being highest for blend ratio close to one. In basic medium, % EWC was found to be less in comparison to neutral and acidic media. A decrease in % EWC is noticed because of the coiling of CTN polymer chains in basic pH.

Graft copolymers

The poly(CTN-g-PVP) films were found to be less pH sensitive than the blends. In general, the % EWC of the copolymers was found to decrease with increase in percentage of grafting in all the three pH's tested, with higher values in acidic pH. The grafted chains of PVP screen the free amino groups of CTN, which are responsible for its hydrophilic nature. In neutral pH, the presence of the grafted hydrophilic PVP chains help in increased water uptake capacity. All the grafted films were found to retain the gel integrity unlike the blends. Lower sorption capacities were noticed for the films in basic medium.

Crosslinked polymers

According to Sonntag,[14] free radicals when formed on two polymers can combine to form a bimolecular terminated reaction, yielding a crosslinked polymer with increased molecular yield. In case the free radicals are present on the same polymer chain, crosslinking between these radicals leads to loop formation and no increase in the molecular weight is noticed. Chain scission reaction is another important phenomenon that occurs in carbohydrates because of homolytic β-fragmentation reaction of the radicals formed next to the glycoside linkage. These chain scissions lead to low molecular weight polymer molecule formation. The poly (CTN-cl-PVP) presented the highest % EWC (~95) than CTN and other modifications in both acidic and neutral media. Crosslinking and intermolecular hydrogen bonding among the two polymers would have lead to the reduction in the EWC, which is not seen in the present case. Therefore, it is predicted that the free radicals formed during the reaction must have triggered chain scissions, forming low molecular weight polymers that act as plasticizers in the matrix, leading to increased chain segmental mobility and free volume in the membranes. The membranes

have shown highest % EWC in both acidic and neutral media and showed no variation with the percentage of crosslinking. In basic medium, the % EWC is slightly lowered similar to the other modifications.

Ionic strength effect

All the blends were subjected to the swelling studies in the presence of NaCl (0.1M) to study the effect of addition of counter ions. Paula et al.[7] reported that on addition of Na$^+$ counter ions to the medium, the % EWC of CTN decreases because of the screening of the electrostatic repulsion of NH$_3$$^+$, resulting in molecular contraction. Therefore, plain CTN has shown maximum decrease in swelling because of the counter ion addition in comparison to the modified ones as shown in Figure 4. It was noticed that with increase in the content of CTN, the % EWC decreases because of the contraction or coiling of CTN. The crosslinked films once again do not show any effect of the counter ions that would lead to decrease in the swelling property. Instead, they have shown an increase in % EWC similar to the observation in pH effect. The explanation is that the effect of screening on NH$_3$$^+$ ions of CTN by the counter ions that would shrink the polymer has been dominated by the increase in the available free volume of the matrix because of the scissioned polymer molecules, resulting in the enhanced sorption capacity.

Thermal properties

Dynamic mechanical and thermal analysis (DMTA)

Glass transition temperature of a polymer is the second order thermodynamic transition that can be detected in DMTA. The T_g depends on the physical and chemical nature of the polymer, and therefore, the type of modification such as crosslinking, branching, blending, molecular weight, and any other special interactions lead to a change in the polymer's thermal behavior. The variation in the storage modulus (E') of the CTN and the modified one are shown in Figure 5. A steep drop in storage modulus or the peak of tan δ is normally taken as the T_g of a polymer. We have taken the valley of E' as the T_g. The scope of intermolecular hydrogen bonding in the blends per unit volume might possibly be higher, resulting in higher T_g in comparison to unmodified CTN. Among the blends, the CTN : PVP ratio close to one have yielded higher T_g than the other ratios tested, because the hydrogen bonding between the two polymers might as well behave as crosslinks. The modification of CTN by grafting had shown lower T_g than the plain CTN and could be attributed to the limited scope for intermolecular interaction. The T_g was found to be the lowest
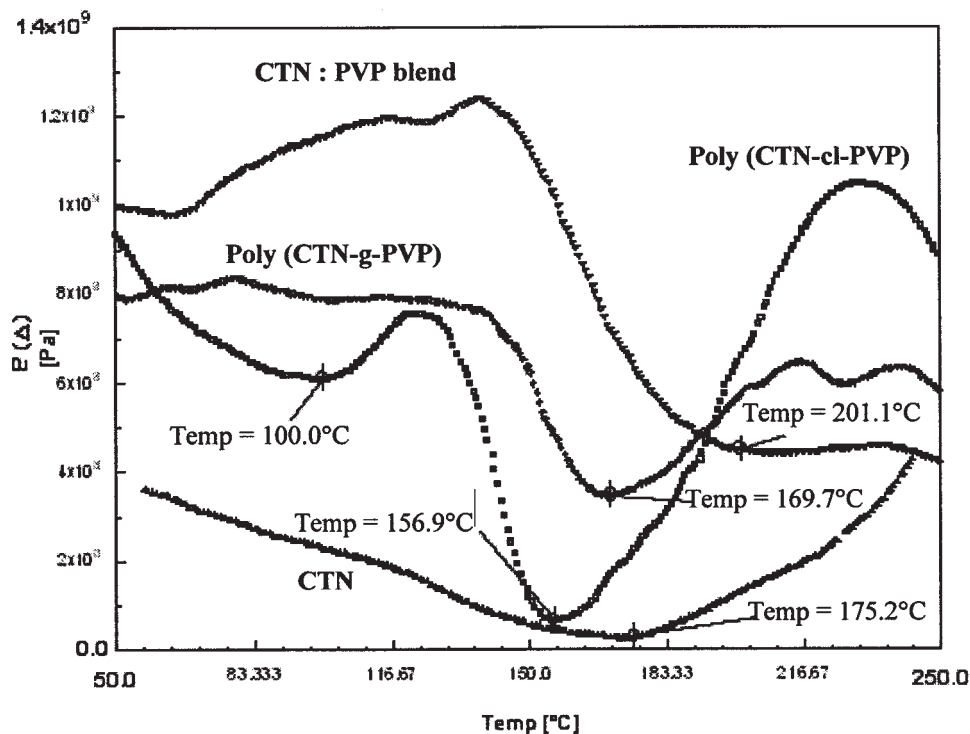
Figure 5 Storage Moduli of CTN and modified CTN versus temperature.

for the poly (CTN-*cl*-PVP) because of the occurrence of low molecular weight components that render plasticization effect to the modified film. The presence of an additional valley for the E' at ~100°C confirms the presence of the low molecular weight components supporting the swelling behavior described in the previous section.

Thermogravimetric analysis (TGA)

The TGA curves for the modified and unmodified polymers are shown in Figure 6. CTN has its maximum weight loss at 320°C while that of PVP (K90) is at 460°C. A single degradation peak for the blends of CTN/PVP was observed at 370°C, indicating that the blends were homogenous. The thermal stability of the 1:1 blend was higher than CTN. The graft copolymer shows degradation peak at 330°C, which is less than that of CTN, indicating a lower thermal stability than CTN. The crosslinked polymer modification reveals two stages of thermal degradation, one at 260°C for the low molecular weight compounds and the other over a range of 400–450°C for the crosslinked ones. The TGA behavior of poly(CTN-*cl*-PVP) reiterates the presence of low molecular weight components in them.

Mechanical properties

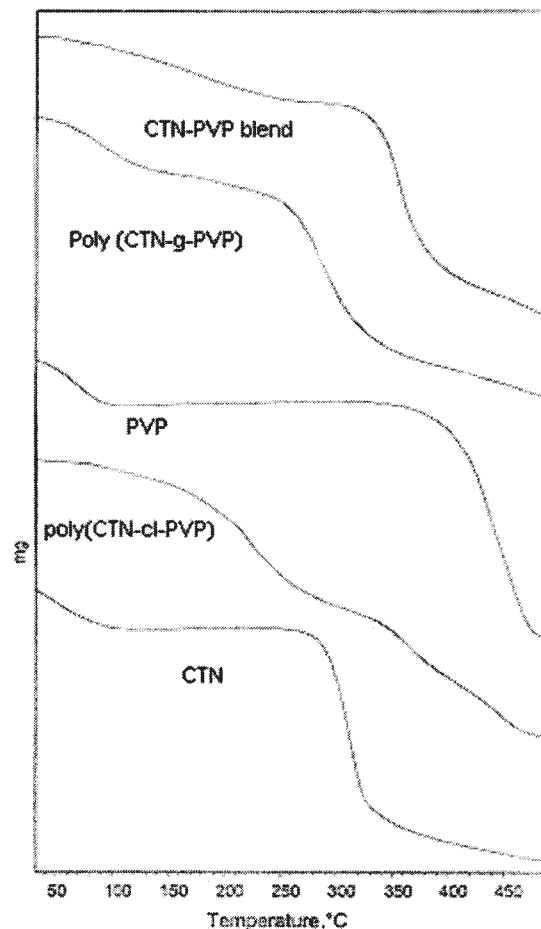Figure 7 shows the tensile strength and percentage elongation at break point for the unmodified and

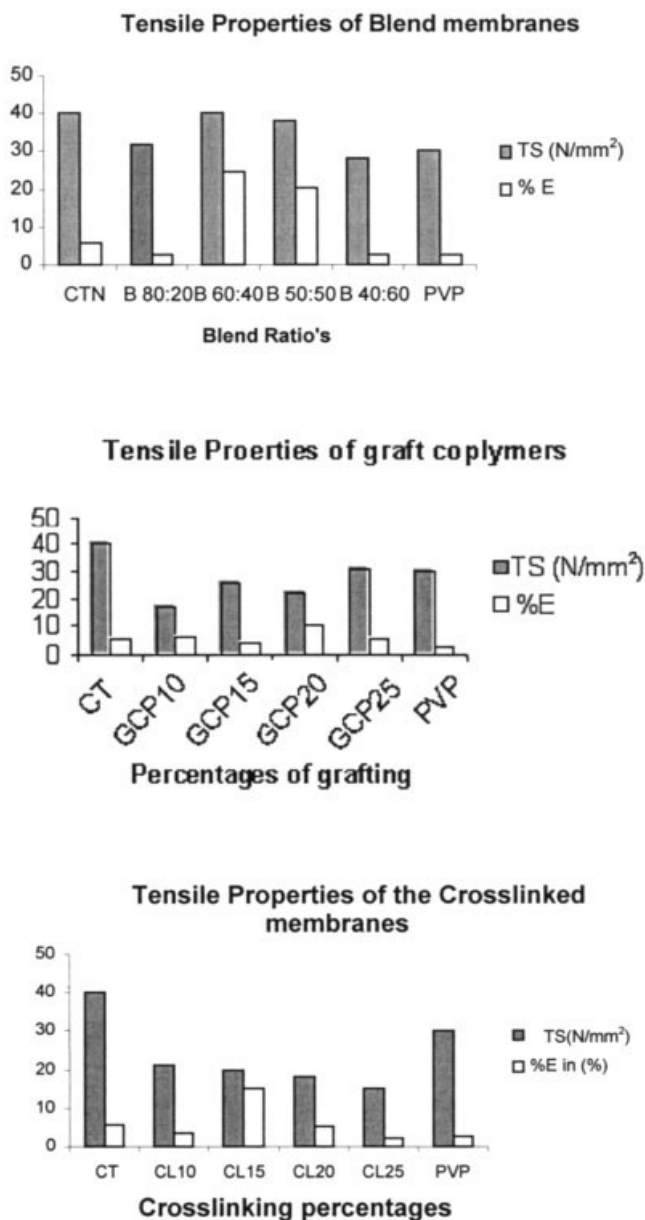**Figure 6** TGA curves of CTN, PVP, and modified CTN polymers.

Tensile Properties of Blend membranes

Tensile Proerties of graft coplymers

Tensile Properties of the Crosslinked membranes

Figure 7 Tensile properties of CTN and the modified CTN membranes. CT, chitosan; GCP, poly(CTN-*g*-PVP); CL, poly (CTN-*cl*-PVP); PVP, polyvinylpyrrolidone; TS, tensile strength; and %E, percentage elongation.

modified films. The tensile strength of the CTN and PVP homopolymers are 44 and 27 N/mm², respectively. The films are brittle and ductile with low % elongation of the order of 5. The tensile strength of the blend decreases except for 60 : 40 and 50 : 50 membranes. However, these blends show extraordinary improvement in their percentage elongation, imparting toughness to the films. The intermolecular hydrogen bonding may be optimum in these blends, leading to difficulty in stretching the film against the bonds, resulting in improved elongation property.

The tensile strength of poly(CTN-*g*-PVP) at break point and percentage elongation are found to be lower

than CTN. As discussed earlier, the graft chains provide increased chain segmental mobility to the rigid structure of CTN, causing a reduction in the tensile properties of the modified ones. As the percentage of grafting increased, the scope for intermolecular hydrogen bonding improves, with 25% graft copolymers presenting a tensile strength of ~35 N/mm². The crosslinked polymers have shown lower tensile properties in comparison to the other modifications. Fifteen percent crosslinking had shown the optimum strength and percentage elongation with 20 and ~15 N/mm², respectively. The tensile properties are in support to their thermal and swelling behavior.

CONCLUSIONS

Physical and chemical modification of CTN was studied by blending, grafting, and crosslinking with PVP. Modification of CTN by blending has shown enhanced tensile properties, % EWC, and thermal properties for the 1 : 1 ratio in comparison to the plain CTN. Grafting lead to improvement in swelling with a maximum for 15% graft copolymer, while the modification by crosslinking was found to be accompanied by chain scissions resulting in highest % EWC in comparison to CTN and other methods of modification. Although, the chemically modified dry films exhibited lower tensile and thermal properties, their wet films have displayed significant improvement in swelling properties associated with good gel integrity in physiological pH, which is necessary for application as hydrogel type of biomaterials in controlled release systems.

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