CELLULOSE ACETATE/PEG/HYDROXYAPATITE BLENDED MEMBRANES FOR DIALYSIS OPERATION

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Abstract- The purpose of this research is to prepare cellulose acetate (CA) based membranes for hemodialysis. It is intended to blend hydroxyapatite (HA) with CA in the presence of polyethylene glycol (PEG). The blended membranes were tested for the effectiveness in dialysis application. Later contact angle and pure water flux of blended membranes were determined to investigate the alterations in CA/PEG matrix after HA addition. Surface morphology of CA/PEG/HA blended membranes was studied using SEM, AFM, FTIR. The results revealed effective and homogeneous blending of CA, PEG and HA. The blended membranes were also tested for Bovine serum albumin (BSA) rejection and urea clearance in order to view their application in dialysis process. The increase of HA weight % in doped solution improved BSA rejection and Urea clearance up to 96.8% and 55.2 % respectively. Prominently, addition of HA to CA/PEG matrix imparted better compaction, protein absorption and porosity which lead to membranes with enhanced BSA rejection, Urea clearance and hydrophilicity.

Keywords- CA/PEG/HA blended membranes, Hemodialysis, BSA rejection, Urea Clearance.

I. INTRODUCTION

Membrane technology is one the most widely used technique for separation, concentration and purification operations. Membranes processes are simple, compact, recyclable and are low energy operations. All these characteristics make membrane processes more advisable to be used in industries, medical and other fields for separation processes [1].
Dialysis is well known since 1861and was commonly used as laboratory operation for the distillation and purification of small amount of solutes [2]. For last 20- 30 years it is gaining recognition as lifesaving technique for the patients suffering with end stage renal disease. Dialysis in case of kidney patient removes excess uremic wastes and water from patient's blood and maintain level of certain ions and components at their optimal level in infected blood. For this purpose, a lot of polymeric materials are being used for the synthesis of dialysis membranes
In our previous work [3] we used cellulose actetate with Poly ethylene glycol (PEG) and glycerol as plasticizer and pore forming agent. The fabricated membranes were studied for pure water flux, urea selectivity and sugar selectivity. In this work our aim was to enhance the mechanical properties of already synthesized membranes using Hydroxyapatite (HA) an in organic nanoparticles. HA possess good bio-activity and bio-compatibility and is good adsorbent for protein. All these properties make this more applicable for dialysis membranes and membrane used for ultrafiltration. Fabrication of flat sheet membranes was done as described in our previous article and various characterization techniques including SEM, AFM, FTIR, XRD and EDS were carried out in order to study the effect of HA addition in already prepared composition.

Material:
Cellulose acetate (30,000 Da) by Sigma–Aldrich was used as basic polymer. Polyethylene glycol (PEG 400) by Panreac, Glycerol by Fisher Chemicals and self-synthesized Hydroxyapatite

[$Ca_5(PO_4)_3(OH)$] were used as additives. Acetic acid supplied by Sigma Aldrich with analytical purity ≈ 98% and distilled water were used as solvent and non-solvent agent respectively agent.

Preparation of casting solution
Cellulose acetate (CA) of 15% weight concentration was dissolved in mixtures of acetic acid and PEG of various ratios as shown in Table1. The basic composition (CA: PEG: Acetic acid: Distilled water) was used at it is illustrated in our previous research. HA particles in different weight % ages were loaded into the casting solutions. Other components of recipe were kept constant.
Changing numerical values of components else than HA particles shows adjustment for net 100 % of doped solution. Membrane without HA was also casted so as to observe the effect of absence of HA on the dialysis membrane properties and this membrane is named Mo.

The solution temperature was maintained at 70 °C. Solution was continuously stirred for 6 hours to confirm that the polymer and additives are completely dispersed and dissolved. After attaining complete homogeneity, the doped solution was cooled and poured into a storage bottle. The solution was then degassed in an ultrasonic bath for about two hours to eradicate any air bubbles present. The prepared casting solution was kept away from direct sunlight to slow down its aging process.

Table 1
Composition of casting solutions

Element	Weight percentage %			
	M_0	M_1	M_2	M_3
Cellulose acetate	15.0	15.0	15.0	15.0
Acetic acid	75.0	70.0	65.0	60.0
Polyethylene glycol	4.0	4.0	4.0	4.0
Glycerol	2.0	2.0	2.0	2.0
Hydroxyapatite	0	5.0	10.0	15.0
Distill water	4.0	4.0	4.0	4.0
Net total	100	100	100	100

Membrane fabrication

The doped solution was poured on the glass plate and was then spread over the glass plate using casting knife of thickness 250um. The glass plate with membrane spreaded over it was immersed in a coagulation bath with temperature up to 25 ^{o}C to complete the phase separation, where exchange between the solvent and water take place. The Loeb and Souriajian or Phase separation /inversion process is one of the most popular procedure used to produce asymmetric porous polymeric membranes [4]. During the process a doped solution is casted as a thin film. The casted film is then immersed into a non-solvent coagulant bath. The diffusional interchange of solvent and non- solvent occurs through the boundary between casting solution and non-solvent agent, resulting in phase-separation to form a membrane with a symmetric or asymmetric structure [5, 6].

The fabricated membrane was then shifted to another container containing glycerol for post treatment to eliminate the extra acetic acid from the membrane [7]. The membrane was then placed in distilled water and was then dried and characterized using different analytical techniques for required features.

Results and Discussion

Scanning Electron Microscopy: Scanning Electron Microscopy is an important analytical technique to study the surface properties of fabricated membranes. Scanning Electron Microscopy with model name JSM 6409A, JEOL, Japan was used to get micrographs of synthesized membranes. SEM images describe weather the surface is porous or dense and also tells about the shapes of pores as well. In Fig 2 it is shown that already present PEG in the composition act as porogen and facilitates the pore formation but addition of hydroxyl apatite effects the pore radius and distribution of pores throughout the membrane. The particle size of HA dictate pore radius of pores formed in membranes during phase inversion method [8]. Gradual increase of HA weight percentage not only increases the number of pores on membrane but also reduces the pores diameter. It heightens uniform distribution all over the membrane and also result in macrovoids formation in vertical direction as shown in Fig 3. HA % up to certain limits improves the required properties but increment in weight % beyond optimum level disturb the characteristics of synthesized membranes.

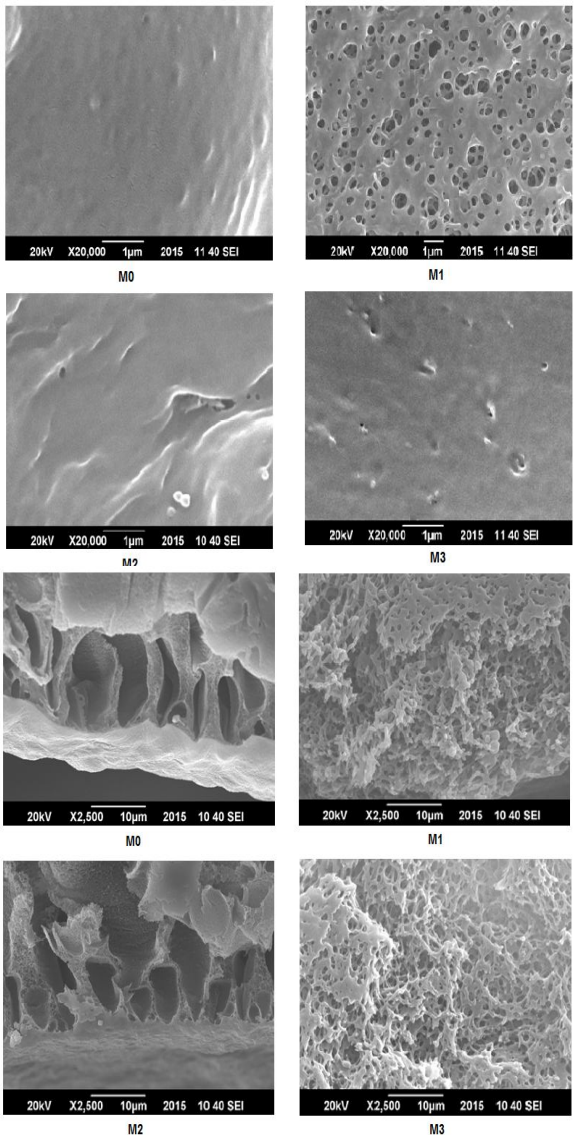


Fig 2 SEM images showing the surface morphology and Cross-section of CA and CA/HA blended membranes.

Atomic Force Microscopy: Atomic Force Microscopy is a well-recognized and useful tool for the surface topography examination of membranes. Thus, the influence of the HA additive on the PEG modified CA membranes was inspected under AFM in tapping mode [9]. The root mean square values for surface roughness of the membranes were measured by AFM. AFM JSPM-5200 Japan was used for characterization of pure and blended membranes. Three-dimensional AFM images of the top surfaces of all of the membranes with scanning area of 6 x 6 μm^2 are shown in Figure 4. The roughness parameters that describe the surface like arithmetic mean roughness (Ra) and the square average roughness (Rq) were analyzed for all of the membranes by AFM analysis software and are tabulated in Table 2. The light areas in images relate to height, whereas the dark regions

indicate depression. From the results, we concluded that the incorporation of HA at various concentrations one way or another improved the surface roughness with respect to the pure CA membrane except at the 15 weight % HA loading in doped solution. The apparent clarification for this behavior was the existence of HA in the CA modified composition, which spread homogeneously but generated better features.

Table 2
Mean roughness values of prepared membranes

Synthesized membranes	R_q (nm)	R_a (nm)
M0	87.7	69.8
M1	53.6	43.0
M2	48.3	39.2
M3	36.4	26.6

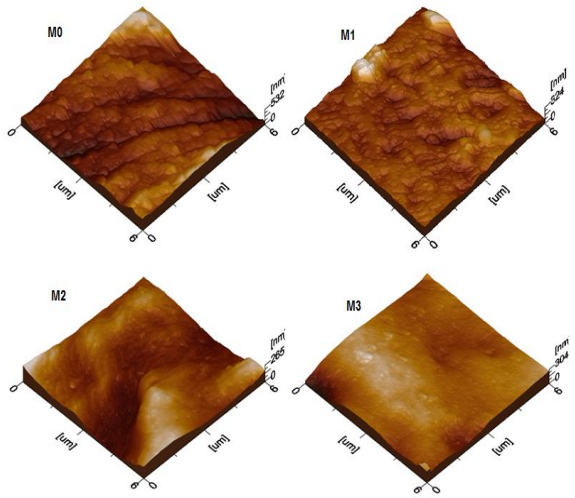


Fig 4. AFM scans of the CA modified with PEG [M0], CA/PEG+5 wt % HA [M1], CA/PEG+10 wt % HA [M2], and CA/PEG+ 15 wt % HA [M3].

Fourier Transform Infra-Red: The part of FTIR spectroscopic analysis is essential in polymeric characterization. The surface characterization of membranes was done using FTIR Spectrum 100 PerkinElmer, MID IR with 1cm^{-1} resolution in transmission mode.IR spectroscopy to examine the molecular structure of the fabricated membranes. It describes the functional group present in the structure and thus dictates the basic characteristics of membranes. The FTIR spectra of the pure CA/PEG modified membranes and CA/PEG blended with HA are collectively presented in Figure 5. The FTIR spectrum of the modified CA membrane was compared with that of the modified CA/HA membranes. In the spectrum of pure CA, the peak at 3417 cm^{-1} was credited to the stretching vibration of the carboxylic acid group (--COOH). The strong peak around 1790 cm^{-1} was allocated due to the stretching mode of the C=O bond, and another peak at 1370 cm^{-1} showed the bending of the C-CH_3 group. The distinctive peak around 1235 cm^{-1} was present due to the asymmetric stretching of the ether C-O-C vibration. The broadening of OH peak in M1, M2 and M3 is because of enhanced OH functional group by hydroxyl apatite addition [10, 11]. This feature express homogenous dispersion of HA in doped solution.

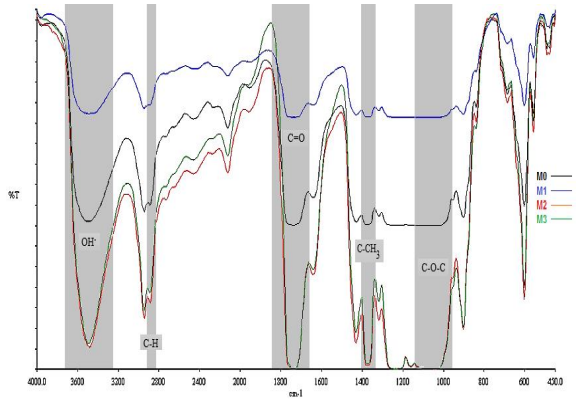


Fig 5. FTIR spectra of the CA/PEG, and CA/PEG +HA blended membranes

Energy Dispersive X-ray spectroscopy: As shown in Fig 6. the EDX spectra shows composition of synthesized membranes [12]. Spectra of M1, M2 and M3 shows presence of calcium (Ca) and Phosphorus (P) along with carbon (C) and oxygen (O) indicating the presence of HA in membranes samples while M0 do not show any Ca or P as HA was not added to M0 casting solution. M0 was used as standard for comparison of membranes having HA and without HA.

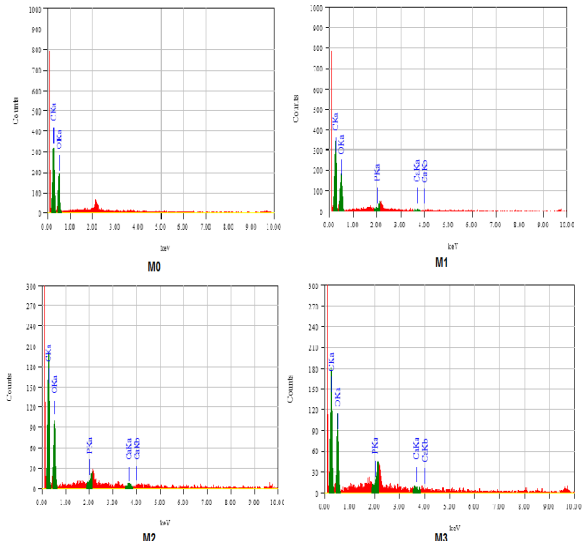


FIG 6. EDX spectra of modified CA and CA/HA membranes revealing C, O, P and Ca as significant elements.

Contact angle measurement and pure water flux: Contact angles, water uptake and degree of swelling are basic characteristic describing the hydrophilicity or hydrophobicity of membrane. Contact angles values of all prepared membranes, including CA/PEG and CA/PEG/HA modified were taken using sisile drop method on using Tantec Contact and are described in Fig 7[13, 14]. For dialysis, reasonable water flux is required which will avoid the excessive loss of water from the patient's body to outside

[15,16]. In this case it was measured using Dead end filtration cell and equation used is given below. and results revealed that case, membrane M2–shows the average value of 169L/hr.m^2. Graphical representation of prepared membranes PWP is also given in Fig 7.

$$Flux\ (J) = \frac{Q}{\Delta t \times A} \quad (1)$$

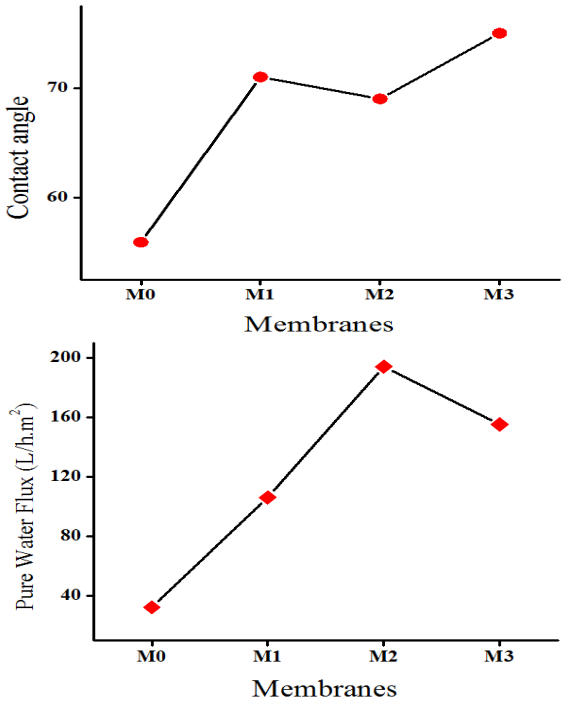


Fig 7. Contact angle measurements and pure water flux of CA/PEG and CA/PEG /HA modified membranes

BSA rejection%: Renal failure patients would experience albumin (≈67 kDa) loss during the dialysis treatment [17]. For an ideal dialysis, it should avoid albumin loss during operation. Fig 8. represents the % rejection of all CA/PEG and CA/PEG/HA blend membranes measured. All the membranes except pure CA/PEG membrane have more than 90% rejection of BSA while membrane M0 has 43.7% rejection which was quite attractive for all dialysis membranes to prevent albumin loss [18]. The membrane M2 with 96.8% rejection gives optimum properties concerning dialysis. All results were measured using using Shidmazu UV mini 1240 at 280nm.

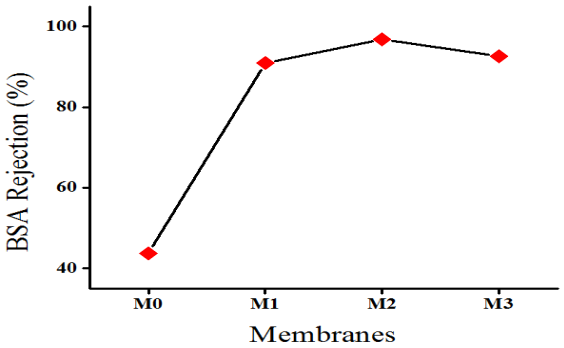


Fig 8. BSA rejection % of CA/PEG and CA/PEG /HA modified membranes

Urea clearance%: Urea is the main component of uremic toxins and is generally adopted to estimate the excellence of hemodialysis membranes [19, 20]. For a good hemodialysis membrane urea reduction should be at least 60% [21]. Here, urea concentration was measure using setup shown in fig 9 and concentrations were recorded using Total Organic Carbon Analyzer (TOC-500, Shimadzu) using equation 2 given below. Fig.9 illustrates the urea reduction of different membranes. Membrane M2 shows the highest urea reduction of 55.2% in contrast to the CA/PEG membrane with 49.4% urea reduction, which is higher than commercial parameters of good dialysis membrane as described by Eknoyan [22].

$$\text{Urea clearance } \% = \frac{C_i - C_f}{C_i} \times 100 \quad (2)$$

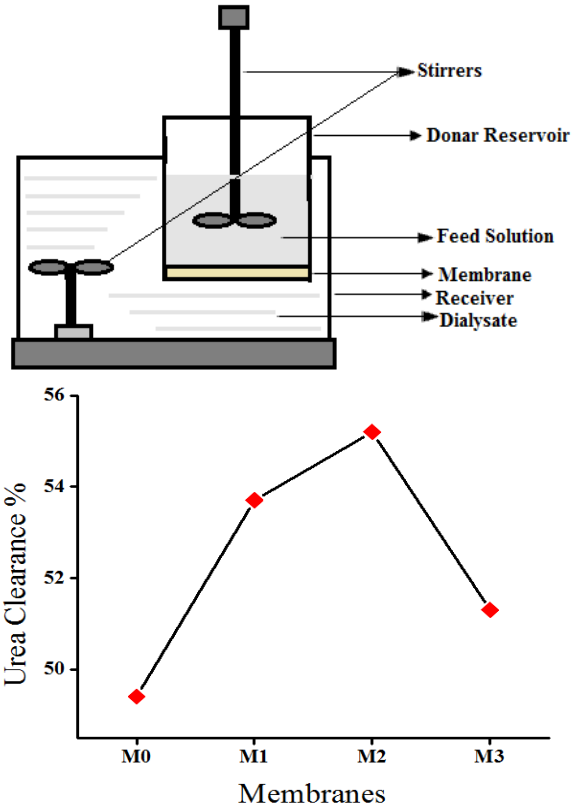


Fig 9. Schematic presentation of Urea testing cell and Urea Clearance % of CA/PEG and CA/PEG /HA modified membranes

Summary:

The thought of assimilation of HA to CA/PEG resulted in the formation of membranes with better surface modification and performance. Synthesized membranes were tested for contact angle, water flux, BSA rejection and urea clearance %. SEM, EDX, AFM and FTIR characterization was carried out to inspect the morphology of membranes. The CA/PEG/HA blend membranes have high polarity and strong hydrogen bonding as proven by the FTIR spectra. The presence of HA in CA/PEG membranes

altered the organization of void structures the tear and sponge-like bodies converted into finger-like regular channels and their pore radius also decreased as shown by SEM images. The blended membranes have developed hydrophilicity and protein resistance confirmed by contact angle and protein rejection experiments respectively. The BSA rejection was increased from 43.7% in case of unmodified CA/PEG membranes to 96.8% for modified membranes. These modified membranes showed appreciable uremic waste clearance relative to the unmodified CA membranes. The percentage urea clearance was increased to 55.2% from already reported value of 52%. Categorically, the presented study has an extensive impact on utilization in dialysis.

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