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ORIGINAL ARTICLE

Potential of extraction of Steviol glycosides using cellulose acetate phthalate (CAP) – polyacrylonitrile (PAN) blend hollow fiber membranes

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Abstract Cellulose acetate phthalate (CAP) – Polyacrylonitrile (PAN) blend hollow fibers were spun in the present work and their efficacy in extraction of Steviol glycosides was investigated. Various compositions of blend hollow fibers were characterized in terms of scanning electron microscopy (SEM), permeability, contact angle, pore size distribution and breaking stress. Concentration of CAP was varied up to 5 wt.% in a total polymer concentration of 20 wt.%. It was observed from SEM images that finger like pores in the cross-section became tear drop like with decrease in concentration of CAP, making them denser. Pure PAN membrane showed the highest permeability around 238 l/m^2.h.bar and the lowest molecular weight cut off (MWCO) 12 kDa and the highest pore density 2.8×10^{18} m^{-2}. CAP-PAN (4:16) blend membrane of MWCO 30 kDa was found to be the most suitable for extraction of Steviol glycosides. At 34 kPa transmembrane pressure drop (TMP) and cross flow rate (CFR) 10 l/h, glycoside recovery was 55 % and purity was 30 % under total recycle mode of operation. Steady state permeate flux was 8 l/m^2.h at 102 kPa TMP. A three stage diafiltration was carried out at 34 kPa TMP and 5 l/h flow. Enhanced 94 % recovery and 54 % purity of the Steviol glycosides was obtained under these conditions.

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Keywords CAP-PAN blend membrane · Hollow fiber · Steviol glycoside · Recovery · Purity

Introduction

Advantages of polymer blending are as follows (Robeson 2007): (i) no new synthesis technology of monomer is required; (ii) a wide variety of desired properties can be obtained by blending existing polymers; (iii) development of new monomers is time consuming and expensive; (iv) polymer blends can be scaled up easily.

The above advantages significantly influence the application of polymers and polymeric membranes. Since the first asymmetric membrane was cast by the phase inversion process (Loeb and Sourirajan 1962), preparation of membranes with better anti-fouling properties is an active area of research. The knowledge of polymer blends thus becomes an important tool for membrane scientists. Generally, in ultrafiltration systems, membrane fouling is the biggest challenge that impedes the performance, affecting membrane life. As reported in literature (Xu et al. 1999), anti-fouling characteristics of membranes is proportional to hydrophilicity. Hydrophilic polymers, like polyvinylpyrrolidone (PVP) and polyethylene glycol (PEG) were used as additives to the base polymers like polyetherimide (Xu et al. 1999). There has also been research on the possible improvement in capabilities of blend membranes formed by cellulose acetate and polyurethane (PU) (Sivakumar et al. 1999) and the effect of an additive like PVP (Sivakumar et al. 2001). Performance of blend membranes using polyethersulfone (PES) and polyacrylonitrile (PAN), acrylonitrile butadiene styrene (ABS) and PVP is also explored (Reddy and Patel 2008; Madaeni et al. 2011). Polymeric blend membranes made of polyvinylidenefluoride and polyethersulfone (Wua et al. 2006), polyethersulfone and cellulose acetate phthalate (CAP) are also reported (Rahimpour and Madaeni 2007). It may be noted that all these blend membranes were in flat sheet.

Higher surface area in a compact volume and requirement of lower pressure, make the hollow fiber modules quite popular. In this regard, the research on polymer blend hollow fibers has been picked up recently. Blend hollow fiber membranes from polyacrylonitrile (PAN) and PVP (Qin et al. 2004) poly(vinylidene fluoride) (PVDF) and PVP (Simone et al. 2010), poly(vinylidene fluoride) (PVDF) /Poly methyl methacrylate (PMMA) (Rajabzadeh et al. 2009) and Poly(vinyl butyral) (PVB) and poly(ethylene-*co*-vinyl alcohol) (EVOH) (Fua et al. 2006) have been investigated.

Stevia, being a high intensity sweetener and devoid of calories, is used as substitute for sugar. A lot of research has gone into replacing sugar by Stevioside in carbonated beverages, tea and coffee (Clos et al. 2008; Kroyer 1999; Ye et al. 2013). In this work, the hollow fibers are spun by an ultra low-cost technology using co-axial needle assembly (Thakur and De 2012), rendering them to be highly economical.

Objective of present study was to extend the work of flat sheet membranes (Roy and De 2014a) to a highly scalable platform, viz, hollow fiber membranes. Motivation of the work was to exploit the high selectivity of the PAN rich membranes with hydrophilicity of the CAP rich membranes by blending CAP in PAN. Importantly, the applicability of hollow fiber membranes as a less energy intensive solution for Stevioside extraction was also explored. To the best knowledge of the authors', this is the first report of Stevioside extraction with hollow fiber membranes and hence, a comparative study was also carried out with the previous studies.

Experimental

Materials and methods

Materials Dry stevia leaves powder was procured from M/s, RAS Agro Associates, Maharashtra, India. An in-house evaporator was used to prepare distilled water, which was required for extraction of glycosides, dissolving neutral polymers for measuring rejection, and as an anti-solvent in the gelation bath. High performance liquid chromatograph (HPLC) grade acetonitrile and water was supplied by M/s, Merck (India) Limited, Mumbai, India. 98 % pure Stevioside and Rebaudioside for calibration purpose were obtained from Sigma-Aldrich, USA. Cellulose acetate phthalate (CAP) was procured from M/s, G. M. Chemicals Company, Mumbai, India and polyacrylonitrile (PAN) homopolymer was obtained from M/s, Technorbital Advanced Materials Pvt. Ltd., Kanpur, India. Polymers, like polyethylene glycol (PEG) and polyethylene oxide (PEO) of various molecular weights, (used to obtain the molecular weight cut off (MWCO) of the membranes) were procured from M/s, S R Ltd, Mumbai, India and dextran (average molecular weight: 70 kDa) was obtained from M/s, Sigma Chemicals, USA. The solvent for polymers, i.e., di-methyl formamide (DMF) was purchased from M/s, Merck (India) Ltd., Mumbai, India. The laboratory scale centrifuge (batch size of 200 ml; model number R-24) was supplied by Remi International Ltd., Mumbai, India.

Membrane synthesis

Solution preparation The total polymer weight percent was fixed at 20 wt.%, since dissolving more than 20 wt.% of PAN and CAP in DMF made the solution highly viscous and spinning was difficult. Four different solutions were made, with wt% of CAP being 5, 4, 2 and 0 and PAN wt% being 15, 16, 18 and 20, respectively. PAN and CAP are two immiscible polymers, as reported by others (Guyot and Benevise 1962; Siahkolah and Walsh 1974; Barani and Bahrami 2007), and both of them can be blended by dissolving in dipolar aprotic solvents, like dimethyl formamide (DMF) (Barani and Bahrami 2007). The polymers, with their respective combination of wt %, were dissolved in DMF under constant stirring at around 200 rpm with temperature being maintained at 60 °C. It took about 6 h for the complete dissolution of the polymers in the solvent. Higher CAP concentrations yielded much porous hollow fiber membranes of poor mechanical strength. These were found not suitable to carry out the experiments and since, the objective of present work is to explore the scope for industrial applications, higher CAP concentration membranes were excluded from this work.

Membrane spinning As mentioned earlier, the membranes were spun using an indigenous technology of co-axial syringe assembly. Details of the set up have been discussed by Thakur and De (2012). The syringe assembly consisted of an inner core (through which the anti-solvent, i.e., water, flows) and the outer annulus. Annular space had the polymer solution flowing through it, which in contact with water in the inner core (flow of water driven by pump), had phase inversion and solidified. The fibers (inner core filled with water), then came in contact with water in the gelation bath, placed under the set up, completing the phase inversion process. Conditions of spinning are reported by Mukherjee and De (2014). Fibers were then wound up using a variable voltage variable frequency drive and spool assembly and were immersed in distilled water for 24 h to ensure the completion of phase inversion.

Hollow fiber cartridge preparation Sixty such fibers were potted inside high density polyethylene (HDPE) pipes, sealed on both sides by adhesive. The inner diameter, outer diameter and length of each fiber in the cartridge are 400 μm, 700 μm and 15 cm. The effective area for filtration was 0.011 m^2 for such cartridges.

Experimental procedure

Experiments were performed for a) characterization of the membranes; b) choosing the optimum membrane and c) detailed study using the optimum membrane under continuous and batch mode to observe the effects of operating conditions on extraction of Stevioside. The ensuing discussion is structured accordingly.

Experimental set up

The set up is illustrated in Fig. 1. Feed was pumped into the cartridge using a booster pump. Pressure gauges (marked 8 and 2) were used to maintain a pressure differential across the membrane. The feed flowed in and flowed out axially. Permeate flowed radially out. Retentate from the cartridge was recycled back to feed tank through the flowmeter, (where the flow is controlled by the valve marked 3), completing the circuit.

Membrane characterization

Surface morphology, molecular weight cut off (MWCO) permeability and pore density Scanning electron microscope (SEM) images (model: ESM-5800, JEOL, Japan) were used to study the surface morphology of the membranes. Membranes were dried in the dessicator for 1 h before fracturing in liquid nitrogen. Then, they were placed on stubs and gold coated and viewed at the desired magnification.

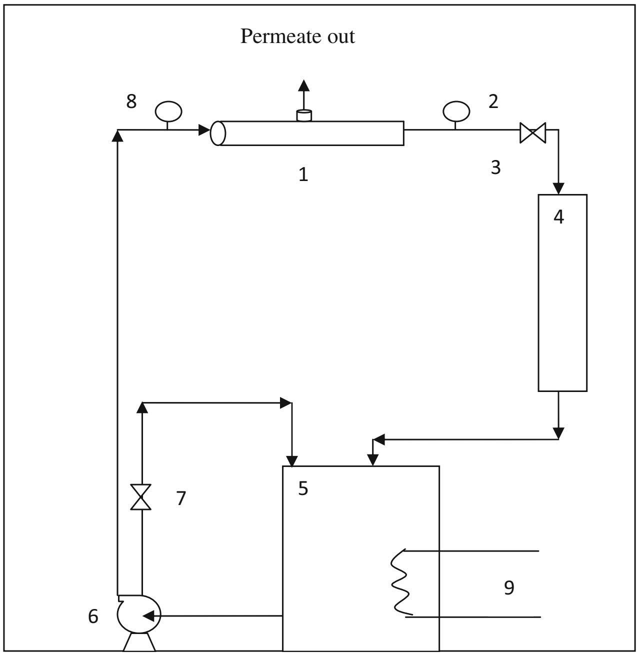

Fig. 1 Details of the experimental set up

Different molecular weight of neutral polymers of PEG ((4 kDa, 6 kDa, 10 kDa, 20 kDa, 35 kDa, 70 kDa, 100 kDa) was used to measure the MWCO of the membranes, by measuring the rejection of respective polymers. A 10 kg/m^3 concentration solution of each of the polymers was fed to the feed tank. Rejection was measured at low pressure (14 kPa) and high flow rate (10 l/h) conditions to minimize concentration polarization. Permeate was collected at 5 min intervals and concentration was measured using an Abbe type refractometer supplied by M/s, Excel International, Kolkata, India. The observed rejection (% R_{obs}) was calculated as:

$$R_{obs} = \left(1 - \frac{C_P}{C_0}\right) \times 100\% \tag{1}$$

where, C_P and C_0 are the concentration of the permeate and feed, respectively. Rejection values were plotted against molecular weight of the polymer and molecular weight corresponding to 90 % rejection was estimated as MWCO of the membrane.

Compaction was carried out with distilled water at 104 kPa for 2 h. After compaction, feed tank (as in Fig. 1), was charged with distilled water and steady state permeate flux was recorded at five transmembrane pressure drops (Banerjee and De 2012) The flux was calculated as:

$$v_w = \frac{Q}{A\Delta T} \tag{2}$$

where, vw, Q, A and ΔT are pure water flux, volumetric flow rate of permeating water, effective filtration area and sampling time, respectively. Pure water flux was plotted with transmembrane pressure drop. It resulted in a straight line through origin and membrane permeability was obtained from the slope of this straight line. Pore density and average pore radius were calculated as per the method used by the authors in the previous work (Roy and De 2014a).

Extraction of steviol glycosides Dry Stevia leaf powder was mixed with distilled water at a ratio of 1:14 (g/ml), and was kept in a water bath at around 78±1 °C for 56 min (Chhaya et al. 2013). The aqueous extract was allowed to cool to room temperature and then cloth filtered (3 μm pore size), before analyzing the colour, clarity, total solids content and concentration of Stevioside and Rebaudioside A. Colour and clarity were measured using UV–VIS Spectrophotometer (supplied by M/s, Perkin Elmer, model 420A, Connecticut, USA) (Roy and De 2014a). Total solids were measured gravimetrically. Steviol glycosides, Steviosides and Rebaudioside A were measured using HPLC method (Roy and De 2014a).

Stevioside and Rebaudioside A were determined by using amino (NH_2) reverse-phase HPLC column ((4.6 mm ID, 250 mm length and 5 μm particle size, supplied by M/s Perkin Elmer, USA). Acetonitrile and water at a ratio of

80:20 (volume basis) was the mobile phase, and the flow rate was maintained at 1 ml/min. Stevioside and Rebaudioside A present in sample were detected using a UV detector at 210 nm wavelength. Pure component of each was used for generating calibration curve. The following equations were used for recovery and purity (Roy and De 2014a, b; Fuh and Chiang 1990; Zhang et al. 2000; Reis et al. 2009; Vanneste et al. 2011; Mondal et al. 2012; Chhaya et al. 2012; Rao et al. 2012a, b):

$$\text{Glycoside recovery} = \frac{\text{Concentration}(\text{Stevioside}+\text{Rebaudioside})\text{in permeate}}{\text{Concentration}(\text{Stevioside}+\text{Rebaudioside})\text{in feed}} \times 100\% \tag{3}$$

$$\text{Purity} = \frac{\text{Concentration of}(\text{Stevioside}+\text{Rebaudioside})\text{in permeate}}{\text{Concentration of total solids in permeate}} \times 100\% \tag{4}$$

Primary clarification The cloth filtered Stevia feed was centrifuged at a speed of 8500 rpm for a centrifugation time of 25 min (Chhaya et al. 2013).

UF experiments

Selection of the optimum membrane Centrifuged Stevia extract was kept in feed tank of the filtration unit. The operating conditions for the selection of optimum membrane were transmembrane pressure (TMP) of 34 kPa and cross flow rate (CFR) 10 l/h. Feed was pumped to the hollow fiber cartridge and was recycled back to the feed tank. Pressure and flow rate were maintained using the valves in the bypass and retentate line. Permeate was collected at regular intervals and recycled back to the tank under total recycle mode. From the cumulative volume versus time plot, the flux decline profile was generated.

UF experiments with the optimum membrane As described in the preceding section (Selection of optimum membrane), UF runs were carried out in similar manner, with the TMP chosen as 14, 34, 68 and 102 kPa and CFR at 5, 10 and 15 l/h. Thus, 12 experiments were performed, with 3 sets of flow rates for each of the TMP values. TMP across the module was measured by taking arithmetic average of inlet and outlet pressure gauge readings of the cartridge.

Experiments under batch concentration mode Batch runs were carried out at three sets of TMP and cross flow rates, of 102 kPa and 15 l/h, 34 kPa, 5 l/h and 34 kpa and 10 l/h. The experimental procedure remained the same, except that the permeate was not recycled back into the tank. Thus, volume in the tank continuously decreased and experiments were carried out till the volume in the tank decreased to half of its initial volume. Flux decline was recorded throughout the duration and samples were collected at regular intervals for analysis.

Multi stage diafiltration Diafiltration mode was carried out at TMP of 34 kPa and flow rate of 5 l/h. The feed volume was 1.5 L and ultrafiltration was carried out till 500 ml of permeate was collected. At this point, 500 ml of distilled water was added as make up volume to the feed tank to maintain it at 1.5 L. This mode of filtration is known as constant volume diafiltration and three stages of diafiltration were carried out for the present study. Samples for analysis were taken after completion of each stage for analysis. Permeate flux was noted for the entire duration of the study.

Membrane cleaning Permeability of the membrane was measured at the beginning of each run as described previously (section on Membrane characterization). Membranes were cleaned using an acid–base treatment. The membrane was first charged with 0.1 N HCl, in a total recycle mode for 30 mins, then with 0.2 M NaOH for 30 mins in the same mode. After this, membranes were cleaned with water, to purge NaOH completely and pH was brought to normal. In this way, the complete recovery of membrane permeability was achieved within ±2 %.

Results and discussions

Membrane characterization

Surface morphology, molecular weight cut off (MWCO) and permeability SEM images of the cross section of hollow fibers of various compositions are presented in Fig. 2. As reported by the authors (Roy and De 2014a), PAN polymer produces a dense, homophasic structured membrane which gradually loses its compactness with increasing CAP weight percent. This can be attributed to the inter polymer interaction parameter, quantified by Barani and Bahrami (2007), defined by Δb. This parameter assumes more negative values (−0.05 to −0.15) if the polymers have a decreasing interaction between them. Thus, as CAP weight percent is increased, Δb becomes more negative, and produces a more porous structure. Structure property is confirmed by Fig. 2. It is evident

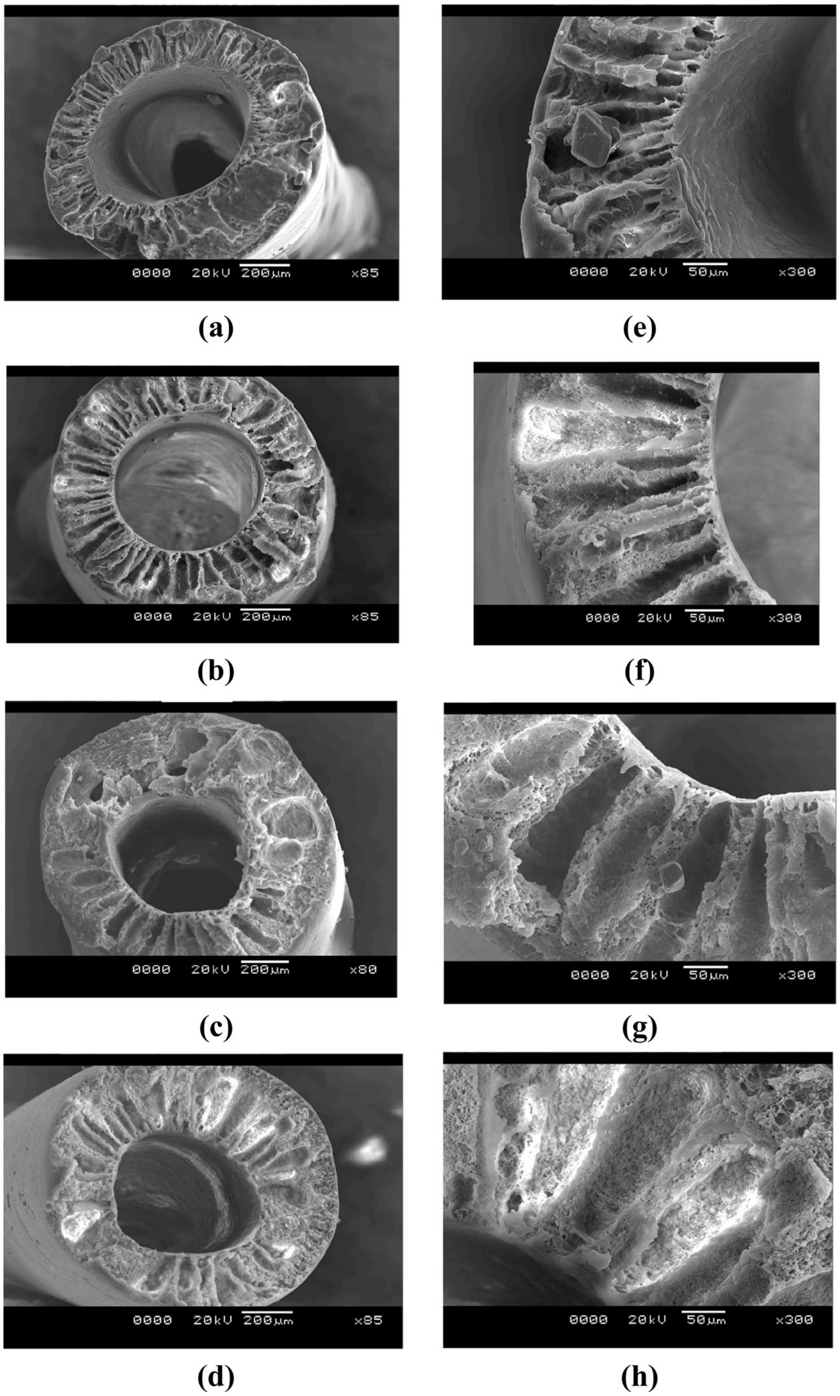

Fig. 2 SEM images of membrane (**a**) cross section of PAN: CAP 15:5; (**b**) cross section of PAN: CAP 16:4; (**c**) cross section of PAN: CAP 18:2; (**d**) cross section of PAN: CAP 19:0; (**e**) thickness of PAN: CAP 15:5; (**f**) thickness of PAN: CAP 16:4; (**g**) thickness of PAN: CAP 18:2; (**h**) thickness of PAN: CAP 20:0

that as CAP weight percent gets reduced, the pore density increases (increasing homophasic compact structure). The finger like pores decrease and structure becomes denser as one moves from the CAP rich composition to the CAP depleted composition. Another phenomenon to be noted is that the CAP rich composition produces bigger sized pores than CAP depleted one. MWCO, permeability and pore densities of various membranes are presented in Table 1. It is clear that MWCO values increase with CAP concentration. These are 12, 20, 30 and 44 kDa corresponding to CAP concentration 0, 2, 4 and 5 wt.%, respectively. This trend is in direct corroboration with the SEM images that membranes become more porous with CAP concentration in the blend.

It is a fact that MWCO and permeability are two factors which are not always dependent on each other, i.e., higher MWCO membranes need not necessarily have higher permeability and vice versa (Roy and De 2014a; Vanneste et al. 2011). It is also evident that PAN rich membranes produce a

Table 1 MWCO, hydraulic permeability and pore densities of the spun PAN:CAP hollow fibers

PAN : CAP	MWCO (kDa)	Permeability (l/m^2.h.bar)	Mean pore radius (nm)	Pore densities (no of pores/m^2)
15:5	44	150	6.5	2.5×10^{17}
16:4	30	110	5.0	2.5×10^{17}
18:2	20	150	4.2	5×10^{17}
20:0	12	235	3.1	2.6×10^{18}

denser structure than CAP rich membranes. From the calculated pore densities, it is clear that the PAN rich membranes have a higher density of pores resulting to higher permeability than the CAP rich ones. The pore density decreases from around 2.6×10^{18} to around 5.0×10^{17} m^{-2} (around 80 % decrease) with a small increment of CAP wt% (0 to 2 wt.%). The drop in pore density is not so sharp from CAP 2 to 4 wt.%, as it decreases from 5.0×10^{17} to around 2.5×10^{17} m^{-2} (decrease of about 50 %) and becomes almost invariant for CAP 5 wt.% as well. This is reflected in variation of permeability as well. Permeability decreases from 235 l/m^2.h.bar to 150 l/m^2.h.bar to 110 l/m^2.h.bar for CAP wt% 0, 2 and 4, respectively. The pore density of 30 kDa and 44 kDa (CAP wt% 4 and 5, respectively) are almost equal, but the permeability of 44 kDa membranes is more and almost equal to 20 kDa membrane, since the pore sizes are much bigger but lesser in number than the 20 kDa membrane (as discussed in the previous section).

Selection of optimum membrane The produced hollow fiber membranes are tested with Stevia extract and their performance in terms of permeate flux, recovery and purity of Steviol glycosides are evaluated. The permeate flux profiles for all the experiments are found to be similar. Stevia extract causes immediate pore blocking (Roy and De 2014b) and therefore steady state was reached quite early. Thus, the steady state values are reported. One representative flux profile is included in supplementary figure (Fig. S1). The performance of various membranes under steady state is shown in Fig. 3. As observed from this figure, the permeate flux increases from 4.5 to about 8.0 l/m^{2}h with increase in CAP concentration upto 2 wt.%. Further increase in CAP concentration to 2 wt.%, the permeate flux decreases to 4 l/m^{2}h and it again increases to about 8.5 l/m^{2}h when CAP is increased to 5 wt.%. As discussed in previous section, MWCO of pure PAN membrane is 12 kDa. A plant extract like Stevia contains cell debris, high molecular weight cellulosic materials etc., that foul the membrane quite fast. Higher molecular weight solutes form a gel-type of layer over the membrane surface causes severe fouling leading to low value of steady state flux 4.5 l/m^2.h at 34 kPa TMP. Membrane with 2 wt.% CAP has larger sized pores with MWCO 20 kDa. Thus, although its permeability is less compared to pure PAN membrane, its permeate flux is quite high, about 7.6 l/m^{2}h. For 4 wt.% CAP membrane, MWCO is 30 kDa but it has the lowest permeability about 110 l/m^{2}h bar. Thus, this membrane shows the lowest permeate flux of 4 l/m^{2}h. For 5 wt.% CAP membrane, MWCO is the highest, 44 kDa with slightly higher permeability compared to CAP 4 wt.% membrane. Since, the size of pores of this membrane is the highest (as is evident from SEM image, Fig. 2b), the permeate flux is also the highest (8.5 l/m^{2}h) among the four membranes.

Pure PAN and 2 wt.% CAP membranes show almost similar values of glycoside recovery (both 55 %) and purity (25 % and 30 %). Since, these membranes are in lower range MWCO of UF, a dense gel layer consisting of higher molecular weight rejected solutes is formed on the membrane surface. The glycosides are again rejected by this layer (equivalent to a dynamic membrane) to some extent leading to lower values of recovery and purity. The nature of compactness of gel layer in both of the membranes is similar. This observation is inferred from the close values of recovery and purity of these two membranes. For 4 wt.% CAP (30 kDa MWCO) membrane, pore sizes are bigger. Thus, the gel layer may not be compact compared to earlier two membranes (Mondal et al. 2012; Rai et al. 2006). Thus, more Steviol glycosides permeate through the membrane, resulting to higher recovery and purity (45 %). Therefore, it is fair to assume that most of the solids present in the extract have molecular weight between 12 and 30 kDa and they can permeate through pure PAN, 2 wt.% and 4 wt.% CAP membranes almost to the same extent. Since, more glycosides permeate through 30 kDa membrane (4 wt.% CAP), its purity and recovery both are the highest. On the other hand, for 5 wt.% CAP membrane, MWCO is the highest 44 kDa. Its pore size is slightly larger than 30 kDa membrane. However, in view of the fouling effects, recovery of the glycoside is almost the same as that of 30 kDa membrane (about 60 %). Having large pore size, this membrane also allows other solids to permeate and thus purity of glycosides is reduced to only 32 %. Therefore, in view of maximum recovery and purity of Steviol glycosides, 4 wt.% (30 kDa) CAP blend membrane is selected as the optimum one to study effects of operating conditions. It may be noted that permeate flux can be enhanced by adopting other well known techniques (Gomaa et al. 2011). The glycoside flux can be obtained from the recovery and steady state flux values (by multiplying

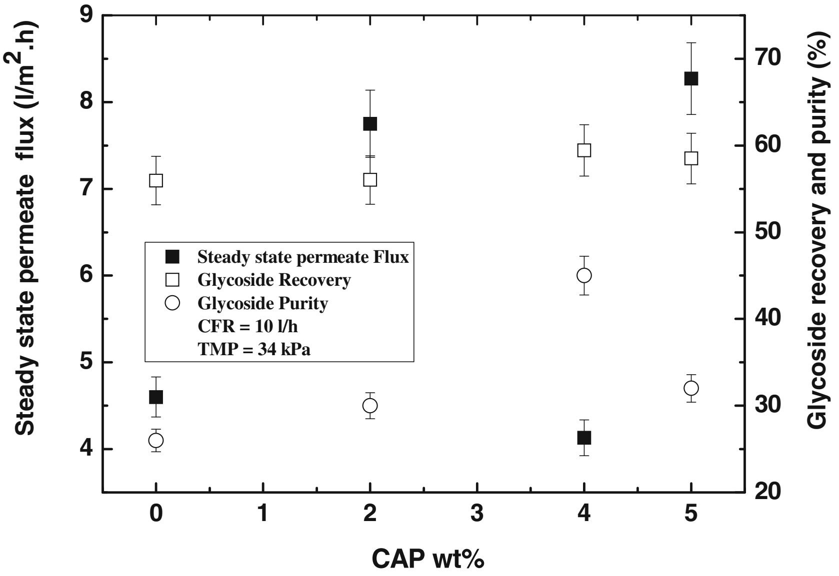

Fig. 3 Steady state flux, recovery and purity against CAP wt%

volumetric flux with glycoside concentration) and they are in the order of 4.35×10^{-2}, 2.19×10^{-2}, 3.91×10^{-2} and 2.25×10^{-2} kg/m^{2}h for the 44, 30, 20 and 12 kDa membranes, respectively.

The present work is a significant improvement over the previous reported work on flat sheet membranes of same composition (Roy and De 2014a). It was seen that in the previous study, the optimum membrane was found to be 90 kDa, since it yielded highest flux, and recovery (68 %) but was poor on purity (34 %), in a stirred dead end system. However, in the present study, the recovery was competitive enough (60 %) with a 30 kDa membrane in a cross flow geometry. Moreover the system yielded much higher purity (45 %) with a higher specific permeate flux (0.07–0.023 compared to 0.032–0.054 l/m^2.h.kPa).

Effects of operating conditions

Total recycle mode Variation of recovery and purity of glycosides with TMP and CFR is shown in Table 2. Two distinct trends are observed. First, glycoside recovery decreases with TMP and CFR has marginal effect on it. For example, recovery decreases from 63 % to 48 % as TMP increases from 17 to 102 kPa. As TMP increases, gel layer over the membrane surface becomes more compact and it acts as a dynamic membrane, thereby, rejecting more glycosides and reducing its recovery. Increased CFR imparts more shearing on gel layer thereby reducing its thickness. But, it has insignificant effects on gel layer characteristics. Therefore, recovery of Steviol glycoside remains unaffected by CFR. Second, purity of the glycoside decreases with TMP but remains invariant with CFR. At higher TMP, as discussed earlier, gel layer becomes compact and it rejects other solutes as well, increasing its purity. However, as expected, CFR does not have any effect on purity. All the experiments were conducted in triplicate and standard deviations (σ) (Kapur and Saxena 2001) associated with individual operating conditions are reported in Table 2. It is evident that for TMP variation (at constant CFR), σ varies between 8 and 10 % for recovery and 11–13 % for purity of the mean value (μ). For CFR variation (at constant TMP), the corresponding values are 2 % for recovery and 4 % for purity. Therefore, majority of the data are beyond μ±σ for constant CFR and all are within μ±σ for constant TMP. This statistical observation is in line with the earlier discussion that the effect of CFR is marginal and that of TMP is more significant.

Batch concentration mode Experiments were conducted in batch concentration mode for various TMP and CFR. Variation of permeate flux profiles is presented in Fig. 4a. The experiments were conducted till attainment of pseudo steady state flux. Therefore, duration of each experiment is different. It is clear that at CFR=5 l/h, permeate flux is much higher for 102 kpa compared to 34 kPa. Pseudo steady state (PSS) flux was 5.7 l/m^{2}h at 102 kPa and it is 3.2 l/m^{2}h at 34 kPa. At higher TMP, membrane fouling is more severe, thereby attainment of PSS occurs after 120 min. On the other hand, at 34 kPa, PSS occurs at 60 min. Effect of CFR is also shown in this figure. At 34 kPa, increase in CFR from 5 to 15 l/h leads to increase in PSS from 3.2 to 4.7 l/m^{2}h. Onset of PSS at CFR=15 l/h is after 30 min. This indicates less fouling at higher CFR.

Volume reduction factor values for various operating conditions varied between 0.4 and 0.5. Variation of glycoside recovery and purity with TMP and CFR are shown in Fig. 4b. As discussed earlier, recovery decreases and purity increases with TMP. At CFR=5 l/h, PSS recovery decreases from 55 to 38 % as TMP increases from 34 to 102 kPa. Effect of CFR on recovery is marginal. At TMP=102 kPa, fouling

Table 2 Recovery and Purity of Steviol glycosides

	TMP=17 kPa			TMP=34 kPa			TMP=68 kPa			TMP=103 kPa		
	CFR=5 l/h	CFR=10 l/h	CFR=15 l/h	CFR=5 l/h	CFR=10 l/h	CFR=15 l/h	CFR=5 l/h	CFR=10 l/h	CFR=15 l/h	CFR=5 l/h	CFR=10 l/h	CFR=15 l/h
Recovery (%)	62.5±3.125	62±3.1	64±3.2	60±3	61±3.05	63±3.15	55±2.75	57±2.85	58±2.9	49±2.45	51±2.55	52±2.6
Purity (%)	46±2.3	43±2.15	42±2.1	48±2.4	45±2.25	43±2.15	54±2.7	53±2.65	50±2.5	59±2.95	58±2.9	56±2.8
TMP variation for a given CFR		CFR=5 l/h		CFR=10 l/h		CFR=15 l/h						
Recovery (σ/μ)		0.10		0.08		0.09						
Purity (σ/μ)		0.11		0.14		0.13						
CFR variation for a given TMP		17 kPa		34 kPa		68 kPa			103 kPa			
Recovery (σ/μ)		0.01		0.02		0.02			0.03			
Purity (σ/μ)		0.04		0.05		0.04			0.02			

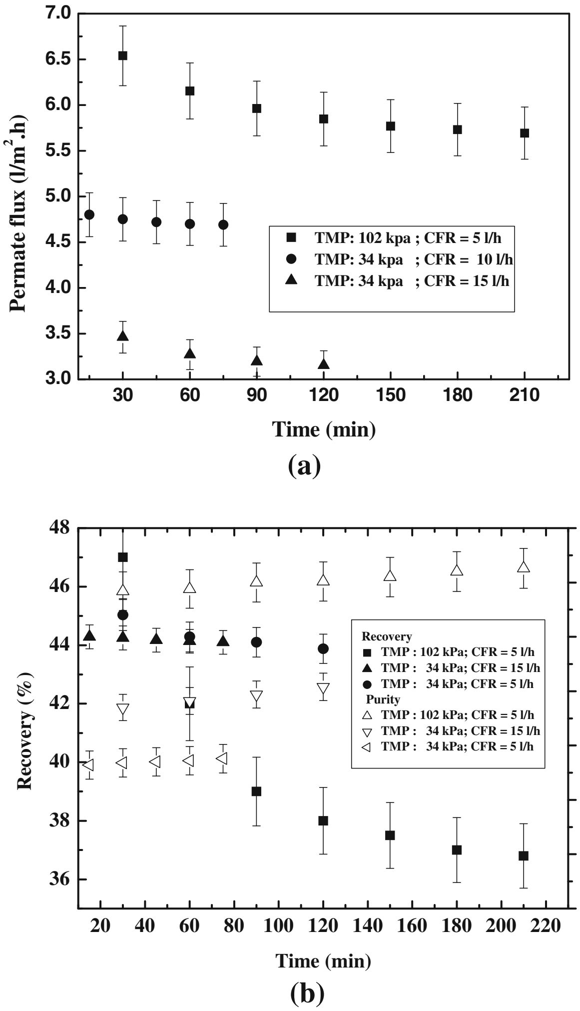

Fig. 4 Experimental results for the batch runs. **a** Flux profiles; **b** Recovery and Purity

becomes progressively severe with filtration duration as the feed is concentrated and glycosides are trapped in the gel layer and recovery decreases. This effect is not significant at lower TMP.

In line with total recycle experiments, purity is higher at higher TMP. For example, at CFR=5 l/h, PSS purity is about 61 % at TMP=102 kPa and it is about 50 % at TMP=34 kPa. However, with time, purity increases marginally (from 59 to 61 %) due to rejection of other solutes. Again, effect of CFR is not very prominent on purity. At higher TMP, more solutes are rejected by the gel layer and purity of glycosides increase.

Improvement of recovery and purity using multistage dialfiltration The results are presented in Fig. 5. A close look at Fig. 5a reveals that during first and second stages of diafiltration, the flux enhancement is minimal. While in stage 1, 5 h was required for 500 ml of permeate collection, in second stage, this was about 4 h and 30 min. However, during 3rd and 4th stages, there was marked increase in permeate

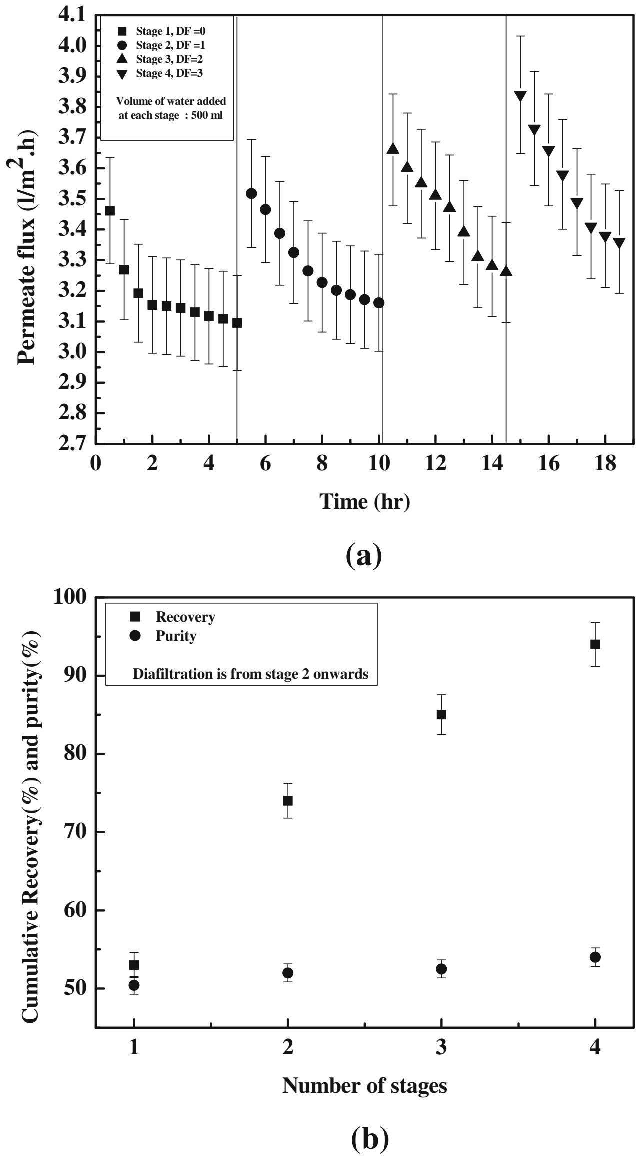

Fig. 5 Experimental results in dialfiltration mode: (**a**) Permeate flux profiles; (**b**) Cumulative recovery and stage wise purity profile at end of each stage

flux. 210 and 180 min were required for collection of 500 ml permeate in these two stages. This can be explained by less membrane fouling by the diluted feed. Initial flux of the membrane increases from 3.45 l/m^{2}h in the first stage to 3.8 l/m^{2}h in the 3rd stage.

Feed dilution affects steviol glycoside recovery. The growth of gel layer is arrested by diluted feed resulting in more permeation of glycosides, yielding to better recovery. Cumulative steviol glycoside recovery is presented in Fig. 5b. It is observed that during 1st stage the recovery is around 54 % and it rises to 94 % at end of 3rd stage of diafiltration. However, the rise in purity is marginal from 50 to 54 %.

Summary of the results of this study with comparison to other reports is presented in Table 3. It can clearly be observed that the present study has some distinct advantages. Specific energy consumption and specific permeate flux were calculated as:

$$\textit{Specific}\ \text{flux} = \frac{J}{\Delta P} \tag{5}$$

$$\textit{Specific}\ \text{energy} = \frac{Q \times \Delta P}{a \times J \times \eta} \tag{6}$$

where, Q is flow rate, η is pump efficiency (70 %), a is membrane area, ΔP TMP and J is permeate flux.

As per specific energy consumption is considered, the hollow fiber system in this work has minimum energy consumption, i.e., 93 kWh per m^3 of extract processed per m^2 filtration area. Specific permeate flux is quite high 0.023 to 0.07 l/m^{2}h.kPa compared to other studies. Only Vanneste et al. (2011) reported a higher specific flux (0.07–0.44 l/m^{2}h.kPa) for one of their membranes. Recovery and purity after first stage of UF are also at higher ends compared to other studies. About 30 to 36 % flux decline indicates that the membrane fouling is not very high compared to other reports due to hydrophilicity imparted by addition of CAP to the blend. Thus, overall, it can be concluded that the performance of the first reported hollow fiber module using CAP-PAN blend for extraction of Steviol glycosides is the best among the cited references in Table 3. Further, improvement of recovery and purity of glycosides is possible by using multistage diafiltration as presented subsequently.

Conclusions

Hollow fibers in CAP-PAN blend were spun for the first time and their application in extraction of Steviol glycosides was investigated. The major conclusions from this work were itemized below:

(i) Denser membrane morphologies were attained as the concentration of CAP in the blend was reduced.
(ii) MWCO of the membranes increased from 12 to 44 kDa as the concentration of CAP in the blend increased from 0 to 5 wt.%.
(iii) Permeability of 4:16 CAP-PAN blend membrane was the lowest 110 l/m^2.h.bar, even though its MWCO was 30 kDa that was found to be suitable for extraction of Steviol glycosides.
(iv) Recovery of glycosides was about 70 % at 14 kPa TMP and purity was about 60 % at 102 kPa TMP, under total recycle mode.
(v) In the batch concentration mode, membrane fouling was more and the recovery and purity of glycosides varied in the range 38–55 % and 59–61 % in the TMP range 34 to 102 kPa.

Table 3 Comparison between the present study and reported literature on Steviol glycoside extraction by ultrafiltration membranes

MWCO	Membrane module	Membrane material/Make	Operating conditions	Flux decline (%)	Steviol glycoside recovery in 1st stage (%)	Steviol glycoside purity in 1st stage (%)	Specific permeate flux ($l/m^2.h.kPa$)	Specific energy consumption ($kWh/m^2.m^3$) Based on 1 m^2 filtration area	Reference
UF 25 kDa	Tubular	PCI, BX-6, FP-100	ΔP: 1200 kPa	80	33 (25 kDa)	45	0.0167	127,551	Fuh and Chiang 1990
UF 100 kDa			ΔP: 800 kPa CFR:25 l/m		53 (100 kDa)		0.075		
UF 2.5 kDa	Rectangular cross flow	Liumar Technologies, Ottawa, Canada	ΔP: 440 kPa CFR:1.5 l/h	8	20	NA	0.079	NA	Zhang et al. 2000
UF:(Pore size: 0.05 µm)	Tubular ceramic	NETZSCH unit with α-Al2O3/TiO_2 membrane material	ΔP: 200–600 kPa CFR:1.86×10^{-4} m^3/s	33–50	45	50	0.058–0.125	4571	Reis et al. 2009
UF 5, 20, 30 kDa	Rectangular cross flow	GE GK, GE GM, UH004, GE PT, UP005, UC005 and lab cast membranes, PES	ΔP: 300 kPa CFR: 400 l/h	20–60	39	30	0.072–0.444	7936	Vanneste et al. 2011
UF 30 kDa	Stirred cell	Permionics Membranes Pvt. Ltd., India, PES	ΔP: 276–690 kPa 600–1800 rpm	20–46	32–47	61–65	0.024–0.061	–	Mondal et al. 2012
UF 30 kDa	Rectangular cross flow	Permionics Membranes Pvt. Ltd., India, PES	ΔP: 276–552 kPa CFR: 60–120 l/h	13–28	40–71	58–70	0.014–0.054	3174	Chhaya et al. 2012
UF 30 kDa	Spiral wound	Permionics Membranes Pvt. Ltd., India, PES	ΔP: 200–500 kPa CFR: 1020 l/h	75	–	65	0.006–0.028	52,040	Rao et al. 2012a, b
UF 7–104 kDa	Stirred cell	CAP-PAN blend lab cast membranes	ΔP: 276 kPa, 500 rpm	43–71	13–52	20–36	0.032–0.054	–	Roy and De 2014a
UF 104 kDa	Stirred cell	CAP-PAN blend lab cast membranes	ΔP: 138–551 kPa 500–1250 rpm	59–65	53–73	29–38	0.021–0.034	–	Roy and De 2014b
UF 12–44 kDa	Hollow fiber	CAP-PAN blend lab spun hollow fiber membranes	ΔP: 14–103 kPa CFR: 5–15 l/h	30–36	49–63	46–59	0.07–0.23	93	Present study.

(vi) In four stage dialfiltration mode, at 34 kPa transmembrane pressure and 5 l/h flow rate, the cumulative recovery of glycosides was 94 %, with purity of 54 %.

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