

Efficiency of RO/NF Membranes at Removal of Veterinary Antibiotics

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Abstract The production of pharmaceuticals has increased rapidly during the last several decades while they have been used for the health of human and animals. Routes of environmental exposure include the release of treated wastewater, land disposal of livestock manures and municipal biosolids (i.e., sewage sludge), and the use of medicated aquaculture feed. This study represents the membrane treatment technology (RO/NF) for the individual removal of antibiotic residues (sulfamethoxazole, trimethoprim, ciprofloxacin, dexamethasone and febantel) and their mixture. According to the results obtained in this work the use of RO (LFC-1 and XLE) and tight NF90 membrane are recommended to achieve an excellent level of removal (> 95%) of all selected veterinary antibiotics (VAs). Nanofiltration NF270, NF and HL membranes showed lower rejection of VAs in binary solutions, but a much higher one in a mixture solution, due to synergistic effect.

Keywords: removal efficiency; RO/NF membranes; veterinary antibiotics

INTRODUCTION

It is now well established that pharmaceuticals are ubiquitous contaminants of wastewater effluents and among all the emerging pollutants, pharmaceuticals are those which have become of increasing concern, due to the possible appearance of resistant bacterial cultures as a consequence of their extensive use. Sources of pharmaceuticals in the environment are mainly due to pharmaceutical industries, hospitals, households, and the disposal of expired or unused medicine, aquaculture and animal feeding (Shao et al., 2009). Veterinary antibiotics (VAs) belong to the biological class of pharmaceuticals and they are used to treat disease and to protect the health of animals. According to a report by the Union of Concerned Scientists (UCS), 16 million kg of antibiotics are used annually in the US, and approximately 70% of them are used for non-therapeutic purposes. Antibiotic use in animal feeding has increased from nearly 91,000 kg in 1950 to 9.3 million kg in 1999. In the EU in the 1990s, a total use of 5 million kg of antibiotics was reported (Zhao et al., 2010). Therefore the presence of antibiotics in wastewater has increased in recent years and their abatement is a challenge.

Recently, a growing number of studies worldwide have shown the presence of a number of different VAs in animal wastes, surface and ground waters, rivers sediments and in soils at concentrations that could have potential impacts on the ecosystems (Zhao et al., 2010). More often, these compounds occur at ng L⁻¹ or µg L⁻¹ concentrations (Batt et al., 2007; Christensen et al., 2009; Sim et al., 2010) and they have potential risks to the aquatic ecosystem such as endocrine disruption and

severe effects. Since they originally possess specific biological features, they are hardly biodegradable and thus give rise to pharmaceutical pollution which is an emerging environmental issue worldwide. They are commonly derived from municipal, agricultural and industrial wastewater sources and pathways due to the often inadequate treatment in conventional wastewater treatment plants (WWTPs) (Brown et al., 2006; Karthikeyan and Meyer, 2006; Batt et al., 2007; Gebhardt and Schröder, 2007; Avisar et al., 2010). As a result, some of those micropollutants will be discharged into the receiving body of water or be used for irrigation, and may result in ground water pollution (Avisar et al., 2009). The new treatments like advanced oxidation processes (AOP), are not suitable because degradation products can be more toxic than the parent compound (De Witte et al., 2010). Membrane filtration processes using RO and NF showed the excellent removal (> 90%) of different pharmaceuticals (Kim et al., 2007; Watkinson et al., 2007; Radjenović et al., 2008). RO and NF are recognized as the leading water treatment technologies for the 21st century, due to their high separation effectiveness and environmental acceptability ("green technology"). The intensive research and development of membrane preparation in the last decade has resulted in their reduced market prices and application in various fluid treatments. In this way RO and NF membranes became competitive in the clean technology of water treatment.

In this paper two reverse osmosis (RO) and four nanofiltration (NF) membranes were used for the laboratory investigation of their efficiency in the removal of five different veterinary antibiotics. Also membranes were characterized using modified fine – pore model.

METHODS

Materials and standards

Chemical standards for trimethoprim (TMP), dexamethasone (DEXA) and febantel (FEBA) were purchased from Veterina (Kalinovica, Croatia), ciprofloxacin (CIPRO) from Pliva (Zagreb, Croatia) and sulfamethoxazole (SMETOX) from Sigma (Steinheim, Germany). All antibiotic standards (Table 1) used were of high purity grade (> 93.6%).

Binary solutions of individual standards (10 mg L⁻¹) were prepared in Milli-Q water, with the exception of FEBA, which was dissolved in ethanol due to its poor solubility in water. Antibiotics were also tested in mixtures. One mixture was SMETOX and FEBA, and the second one consisted of all five antibiotics.

Reverse osmosis and nanofiltration waterworks

Removal of VAs was tested in the laboratory set-up as described previously (Košutić et al., 2000), with commercially available reverse osmosis and nanofiltration membranes at a working pressure of 15 bar. Reverse osmosis membranes were XLE from Dow/FilmTec, Midland MI and LFC–1 from Hydranautics, Oceanside, CA and nanofiltration membranes NF90, NF270 and NF from Dow/FilmTec and HL from Desal, Osmonics, GE Infrastructure Water Process Techn., Vista, CA. The surface area of the membranes was 10.75 cm². First, the preserved membranes were washed with demineralized water without pressure and then pressurized at 20 bar for 5h. After pressure pretreatment, the pure water flux was measured.

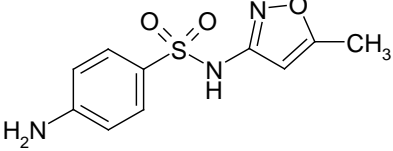
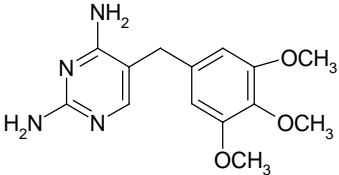
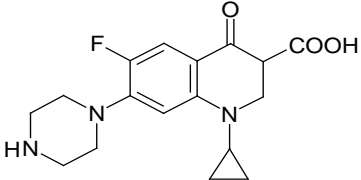
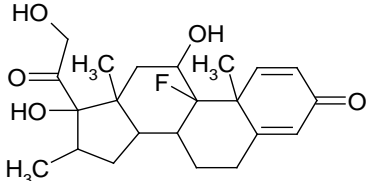
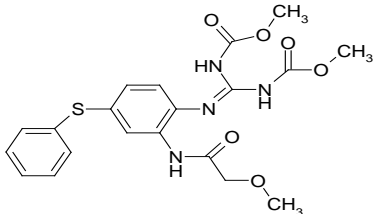
Conditions for determining pore size and pore size distributions in the selective membranes' layer, have been described previously (Košutić et al., 2006).

Chemical analysis

The concentrations of inorganic salts (feed concentrations were 300 mg L⁻¹) were determined by the conductometer SCHOTT Instruments Lab 960, and those for markers' (total carbon) were

determined by the carbon analyzer Shimadzu TOC-V_{WS}. Concentrations of sulfates were determined with Dionex ICS-3000.

Table 1. Selected veterinary antibiotics, their structure and physico-chemical properties.

Antibiotic	Structure	Mol. weight g mol ⁻¹	pK _a ^a	logK _{OW} ^b	d _c nm
Sulfamethoxazole		253.28	pK ₁ 1.83 pK ₂ 5.57	0.89	0.734
Trimethoprim		290.32	pK ₁ 3.23 pK ₂ 6.76	0.91	0.779
Ciprofloxacin xHClxH ₂ O		331.35	pK ₁ 6.68 pK ₂ 8.63	0.28	0.826
Dexamethasone		392.46	n.a.	1.94	0.889
Febantel		446.48	n.a.	1.53	0.941

^a pK_a values were retrieved from Babić et al. (2007).

^b log K_{OW} values are for neutral molecule form; based upon REPHAD Database. <http://sunasce.com/fkit/rephad.php> (accessed 25 July 2010)

The analyses of VAs were performed using a Varian ProStar 500 (Walnut Creek, California, USA) HPLC system consisting of a ProStar autosampler, ProStar 230 tertiary pump system, ProStar 330 diode array detector, and thermostatted column compartment. The column temperature was set to 30 °C and the injection volume was 30 µL. C18 Synergy Fusion 150 mm×4.6 mm, particle size 4 µm column (Phenomenex) was used to separate all investigated compounds in the mixture. The mobile phase used in the chromatographic separation consisted of a binary mixture of solvents A (0.01% formic acid in water) and B (0.01% formic acid in acetonitrile). A simultaneous mobile phase gradient program was used: the elution started with a 2.5 min linear gradient from 100% A to 8% B, followed by a 3.5 min linear gradient to 10% B, a 5 min linear gradient to 30% B, a 4 min linear gradient to 60% B and finally a 3 min linear gradient to 95% B which was maintained for 10 min and then a 0.1 min linear gradient back to 100% of A. The flow rate was 0.5 mL min⁻¹. The separation was monitored at an absorbance wavelength of 254 nm.

RESULTS AND DISCUSSION

Membrane characterization

In Table 2, rejection factors of inorganic salts, sodium and calcium chloride, and magnesium sulfate, are displayed. Rejection factors for sodium and calcium chloride are low for nanofiltration membranes. Membranes were tested with magnesium sulfate because the results for sodium and calcium chloride couldn't be compared to the manufacturers' data. They test membranes with magnesium sulfate and the results obtained in this work are in agreement with their results. The appreciably higher rejection factor for sodium and calcium chloride by the RO rather than the NF membranes are evident. In the case of RO membranes there are little differences of rejection of sodium and calcium chloride in contrast to wide differences of their values for NF membranes. Both facts point to differences in the active layer porosities of the examined membranes, which have been shown in PSD curves (Fig. 1). The PSDs of all the NF membranes are located at evidently wider pores, which are responsible for the lower sodium chloride retention. Thus, the size exclusion mechanism is prevailing. The higher rejection of calcium chloride observed with NF membranes are the consequence of the additional charge exclusion which resulted from the repelling action of the NF membranes' electric charge on the divalent ions. Membrane XLE had little less rejection of sodium chloride than expected but it can be explained with the higher pore size as shown by the PSD curve.

Table 2. Rejection factors of inorganic salts with flux and effective number of pores.

	NaCl	CaCl ₂	MgSO ₄ ·7H ₂ O	J_w	N_{eff}
	%	%	%	L m ⁻² h ⁻¹	10 ¹⁶ m ⁻²
LFC-1	96.5	98.1	99.7	38.38 ± 2.18	0.874
NF90	89.9	98.0	99.4	77.92 ± 6.05	1.413
NF270	13.6	28.1	90.7	109.16 ± 18.82	1.188
NF	24.6	37.3	98.2	98.53 ± 14.57	1.004
XLE	89.5	96.4	99.2	69.38 ± 11.00	1.199
HL	27.1	82.9	94.1	88.48 ± 7.05	0.758

According to Matsuura (1993) there are few transport models for RO/NF, one of them is the fine-pore model, which is the most realistic and was used in this work. The pore size and pore size distributions (PSDs) were determined for all membranes by the modified examination method based on the specific solutes (markers) transport. The results are presented in Fig. 1 and there are noticeable differences in PSD curves between RO and NF membranes. PSDs for RO membranes are unimodal, with the most pores of the size 0.78 nm (LFC-1) and 0.88 nm (XLE). Pores are smaller than 1 nm, and this is confirmation that these are RO membranes. The PSDs of two tight NF membranes have the main peak at a similar pore size, 0.79 nm for the NF90 and 0.70 nm for the NF270 type membrane, respectively. In the case of the NF270 type membrane there is also an additional peak at 1.56 nm, making the distribution similar to those of the loose NF membranes. Hence, loose NF membranes show bimodal PSD curves with two clearly separated peaks. The pores with highest incidence in this case are those of 0.72 nm (NF) and 0.73 nm (HL) followed by large pores, sized between 1.32 and 2.03 nm.

Removal of veterinary antibiotics

Rejection (R) of each investigated compound was calculated as:

$$R(\%) = \frac{c_f - c_p}{c_f} \times 100(\%),$$

where c_f and c_p are concentrations (mg L^{-1}) of a compound in feed and permeate stream, respectively. The rejections of the selected veterinary antibiotics in laboratory scale RO and NF treatment are depicted in Fig. 2.

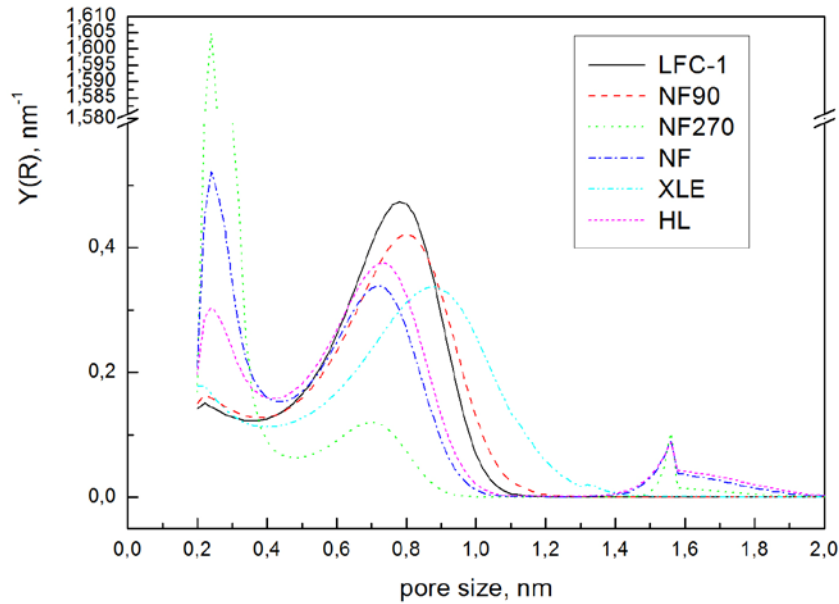


Fig. 1. Pore size distributions of examined RO and NF membranes at 15 bar.

As systematized by Bellona et al. (2004), rejection of solutes on RO/NF membranes will be affected by solute and membrane properties, feed composition and operating conditions. Solute can be rejected on RO and NF membranes by one or a combination of three basic mechanisms: size exclusion (sieve steric effect), charge exclusion (electrical, Donnan) and physico-chemical interactions between solute, solvent and membrane.

Considering the MWCO (100 Da) of the investigated RO membranes (Xu et al., 2006), all the compounds should be well rejected by the mechanism of steric hindrance. Rejections of all VAs were higher than 95%, which is presented in Fig. 2 (VAs are presented with increasing molecular weight). Also rejection higher than 97% for the tight nanofiltration NF90 membrane was to be expected because its MWCO is 100 – 200 Da (Boussu et al., 2006; Radjenović et al., 2008) and this membrane is at the narrow pore end of the NF separation range. The MWCO value 150 – 300 Da of other nanofiltration membranes (Zhang et al., 2004; Boussu et al., 2006; López-Muñoz et al., 2009) may explain its various rejections (higher than 15%). All VAs with a molecular weight higher than 300 g mol^{-1} are rejected almost completely ($> 97\%$).

In the last column of Table 1 the values for the effective diameter of and organic component in water (d_c) are presented. The relationship between the molecular weight of an organic component and its effective diameter in water (Geens et al., 2006) is:

$$d_c = 0.065 \cdot (M_w)^{0.438}.$$

Although the molecular weight is not a direct measure of the dimensions of a molecule, it still reflects the molecular size, and it is a readily accessible parameter. This correlation is valid for the molecular weight range where nanofiltration typically operates (up to ± 600 Da) (Van der Bruggen and Vandecasteele, 2002). It was an expected rejection increase, with the increase of effective diameter for all investigated membranes. For RO membranes rejection is very high ($> 95\%$). There is not a big difference in their rejection because the effective diameter of all investigated VAs (> 0.734 nm) is similar to the pore sizes of LFC-1 (0.78 nm) and XLE (0.88 nm). A slightly lower rejection of SMETOX and TMP with XLE membrane can be explained by its having bigger pores

than LFC-1. All other VAs are completely removed with RO membranes because d_c is bigger than the pore size of the investigated membranes. Pore size of the tight NF90 membrane is very close to RO membranes so the rejection is higher than 97%.

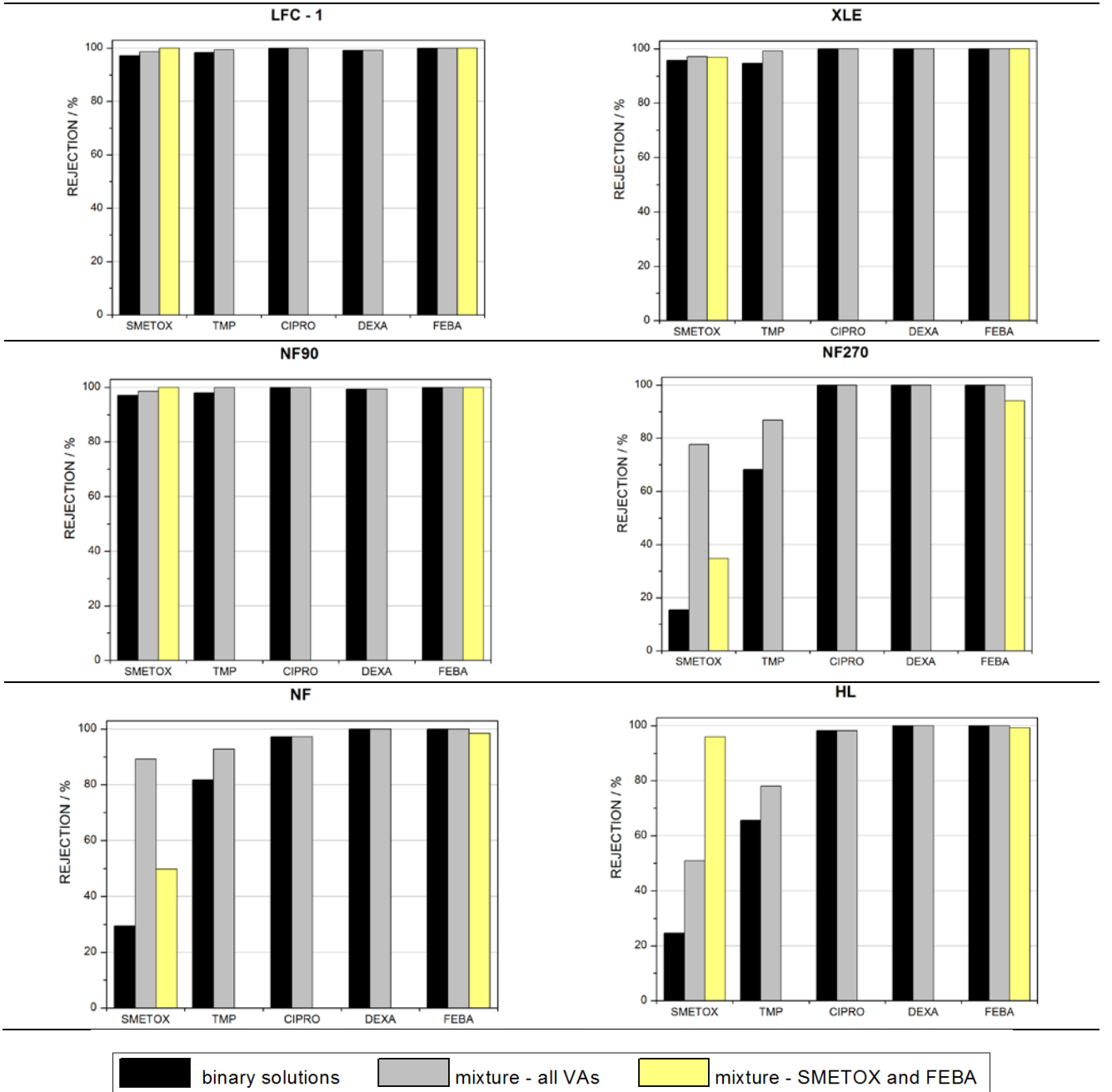


Fig. 2. Rejection factors of all investigated VAs in binary solutions and their mixture, and rejection factors of SMETOX and FEBA in their mixture.

For NF membranes there is a greater effect of effective diameter on rejection. These membranes have larger pores, higher than 1.32 nm, and the rejection of smaller VAs (SMETOX and TMP) is lower (from 15 to 82%) and it increases with d_c .

The increase of rejection with the increase of molecular weight, i.e. effective diameter in water, show the effect of the steric hindrance mechanism, i.e. larger molecule rejection is higher.

The mixture of SMETOX and FEBA shows a higher level of rejection than in binary solutions. This proves that the physico-chemical interactions between solute, solvent and membrane had an

influence on the rejection of these components. This was also proven in the mixture of all five selected antibiotics (Fig. 2).

Results, from this work, showed retention efficiency of selected VAs by different RO and NF membranes examined in a laboratory set-up. The next step of this work will be the investigation of the removal of VAs in different water matrices and also the finding of an optimal pretreatment method to reduce the fouling phenomena in real wastewater treatment. The membrane cleaning procedure, cleaning chemicals and the frequency of cleaning will be determined and optimized in a pilot plant.

CONCLUSIONS

1. The pore size and PSDs of selected RO/NF membranes were determined using the fine – pore model; the pore sizes of RO and tight NF membranes are less than 1 nm and their PSDs are unimodal, while loose NF membranes (HL and NF) have bigger pores, between 1.32 and 2.03 nm, with bimodal PSDs.
2. A great removal of all VAs was achieved with RO (LFC, XLE) and tight nanofiltration (NF90) membranes and a good removal of bigger VAs molecules was achieved with other NF membranes. Obtained results confirm the usability of the examined RO and NF membranes for the removal of veterinary antibiotics from water, especially the XLE and NF90 membranes of high productivity.
3. According to rejection factors of veterinary antibiotics and membrane characteristics it can be concluded that the main separation mechanism was size exclusion in the case of RO and tight NF membranes while for loose NF membranes there was an impact of two other mechanisms (charge exclusion and physico-chemical interactions).

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