Evaluation of Reverse Osmosis Membranes in Concentration of Beetroot Peel Extract

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Abstract

Recovery of valuable products from organic wastes with conventional extraction method plus modern separation technology is becoming popular in solid waste management. The major attention of this project was to test the efficiencies of two different types of reverse osmosis membranes (RO99 and X20) on juice concentration extracted from peel of beetroot which is "waste". The extractions of beetroot peel were completed using water and ethanol-water (15 v/v%) solvents at 22 °C for 60 minutes. The applied transmembrane pressure, temperature, and recirculation flow rate of membrane separation process were 40 bars, 30 °C, and 400 L/h, respectively. Quantifications of valuable compounds were detected using spectrophotometer. The permeate flux profiles were investigated and lower permeate flux was experienced with RO99 compared to X20 in both methods. Additionally, from the aspect of efficiency, RO99 outstripped X20 membrane on concentration of some compounds such as betalains, and phenolic components. Betaxanthin, betacyanin, antioxidant and TPC contents in final retentates of RO99 membrane concentration were as follow: 292.47±1.93 mg/L, 499.03±1.3 mg/L, 1133.15±25.74 mg/L,1243.96±106.56 mg/L (water solvent) and 337.26±4.31 mg/L, 585.2±5.83 mg/L, 698.55±22.53 mg/L, 1268.87±48.69 mg/L (ethanol-water solvent), respectively. From this experiment, expectation can be made that membrane technology can widen its applications in food and pharmaceutical industries.

Keywords

beetroot peel, reverse osmosis, betalains, phenolic, antioxidant

1 Introduction

Utilizing wastes from food processing in proper way is somehow one of the solutions to environmental pollution problems. In addition, some of the waste parts of fruits and vegetables contain more valuable compounds than the other parts [1, 2].

Beetroot (*Beta vulgaris*) is one of the popular tropical vegetables in Europe because of its high constituent of natural colorant, which is betalain [3], and health-promoting antioxidant rich compounds especially phenolic [4]. Phenolic compounds are well known for their biochemical properties and basically extracted by alcohol-water mixtures [5]. Betalain is also rich in antioxidant capacity [6] and mainly composed of betacyanin (75–95 % of betanin which is responsible for red color) and betaxanthin (95 % of major yellow color giving vulgaxanthin-I) [7]. Being a water soluble compound, extraction of betalain can economically be performed by water. However, the disturbance of protein limits the usage of water as solvent and therefore, preferably, some

kinds of solvents, for example, methanol and ethanol are employed [7]. Ethanol was chosen as the solvent in our project since, unlike methanol, no purification step is required.

The application of membrane technology (mostly ultrafiltration and microfiltration) to separate the various components from the feed stream has been receiving increased attention since the 1980s in different fields. Since that time, some researchers [8, 9] had already done the improvement of the extraction of betalain compounds with the aid of membrane separation. In recent times, this technology is a renaissance especially nanofiltration and reverse osmosis processes (occasionally combined with ultra and microfiltration as pre filtrations), not only for the isolation of different compounds but also for the concentration and/or clarification of the fruit juices [10, 11].

Suitable membranes for different purposes of separation are chosen according to their pore sizes. Reverse osmosis membranes are basically used in water purification

with the purpose of desalination. Even so, their applications in food and beverage industries are continuously expanding mostly in concentration of juices.

Tóth et al. [12] declared that membrane filtration is the only effective technology to separate materials possessing individual molecular weights so that we could extend their usages in various fields. The main purpose of this investigation was to evaluate the efficiency of membranes on concentration of valuable compounds from beetroot peel extracts.

2 Materials and methods
2.1 Crude extract preparation
Beetroots (*Beta vulgaris* L.) variety of Rhonda were purchased from local market. Primarily the beetroots were gently cleaned to remove foreign materials and peeled. The peels were then grounded using GM 200 pulverizer. Aqueous extraction was carried out as control in which the pulp (320 g) was mixed with distilled water (3200 mL) and extraction was achieved by single-batch-type extractor, which was designed with stirrer (LAUDA ECOLINE E100) and thermostat (YELLOWLINE BY IKA OST 20 DIGITAL), at 22 °C for 60 minutes based on our previous experiments. In case of ethanol-water solvent, 15 v/v% ethanol solution was applied instead of distilled water. The measured pH of the crude extracts was in the range of 6 to 7.2. The final extracts were stored under freeze condition until membrane separation experiments.

2.2 Membrane separation
RO membranes of low fouling type Trisep X20 advanced composite membrane (Lenntech) and thin film composite Alfa Laval RO99 membrane with active surface areas of 0.18 m² were applied. Cross-flow filtration process was performed by DDS Filtration Equipment (LAB 20-0.72, Denmark). The average inlet and outlet transmembrane pressure was 40 bars and recirculation flow rate was 400 L/h maintaining the temperature of the stream at 30 °C. During the concentration, the time required to collect each 100 mL of filtrate was recorded and the sample collections were performed at every 500 mL of permeates. After separations, the analytical measurements were carried out.

Pure water flux measurements were performed before and after concentration step in order to estimate membrane resistance and fouling resistance. After the concentration process, distilled water was used for rinsing and removing the polarization layer completely. According to Darcy's Law, permeate flux J (m³/(m² × s)) for pure water can simply be demonstrated as:

$$J = \frac{TMP}{\mu \times R_m},$$

(1)

where TMP means the applied transmembrane pressure difference (Pa) as driving force; μ is the dynamic viscosity of permeate (Pa × s); R_m represents membrane resistance (1/m) to mass transfer [13].

Subsequently, membrane resistance (R_m) and fouling resistance (R_f) can be derived from Darcy's Law:

$$R_m = \frac{1}{\mu \times a_1}$$

(2)

$$R_f = \frac{1}{\mu \times a_2} - R_m,$$

(3)

where a_1 and a_2 means slope of pure water flux curves before and after measurement versus transmembrane pressure difference.

The permeate flux (L/(m² × h)) of the sample was simply calculated from volume of filtrate ΔV (L) divided by Δt (h), the time required to collect the filtrate, and active surface area of membrane A_m (m²) [14].

$$J_x = \frac{\Delta V}{A_m \times \Delta t}$$

(4)

According to the research of Bánvölgyi et al. [15], Volume Reduction Ratio (VRR) was approved by feed volume V_0 (m³) and volume of retentate V_R (m³) or volume of permeate V_P (m³) following the Eq. (5);

$$VRR = \frac{V_0}{V_R} = \frac{V_0}{V_0 - V_P}.$$

(5)

Based on the concentrations of permeate C_p (mg/L) and retentate C_R (mg/L), retention (%) can be estimated as follows (Eq. (6)) [15]:

$$R = \left(1 - \frac{C_P}{C_R}\right) \times 100.$$

(6)

2.3 Spectrophotometric analysis
Spectronic Genesys 5 Spectrophotometer (MILTON ROY, U.S.A.) was applied for the quantification of betalain compounds. The colors were detected as absorption at individual wavelengths of 480 nm for betaxanthin [16] and 535 nm for betacyanin [17]. The appropriate dilution was done for each sample and the concentrations of the respective betalain compounds were calculated using Eq. (7):

$$BC = \frac{A \times MW \times DF \times 1000}{\varepsilon \times L} \ [\text{mg/L}],$$

(7)

where A is the absorbance; MW is the molecular weight (g/mol); DF is the dilution factor; ε is the molar extinction coefficient (L/(mol × cm)) and L is the path length (cm).

For quantification of betaxanthin and betacyanin, molar extinction coefficients and molecular weights for respective compounds i.e. betaxanthin (ε = 48,000 L/(mol × cm) and MW = 308 g/mol), and for betacyanin (ε = 60,000 L/(mol × cm) and MW = 550 g/mol) were applied [18].

Additionally, spectrophotometric method was applied for Total Phenolic Compound (TPC, 760 nm) and antioxidant (593 nm) assays. TPC contents of each sample were analyzed according to Folin-Ciocalteu method [19] following employed procedure. The sample solution (20 µL) was mixed with 1250 µL of Folin reagent and 230 µL of methanol-distilled water solution. After exactly 1 minute, 1000 µL of sodium carbonate solution (0.7 M) was added to the sample solution [20]. The mixture was then put in thermal bath which maintained the temperature at 50 °C and absorbance measurement was carried out at 760 nm after 5 minutes of incubation. The calibration was done by using (0.3 mM) gallic acid as standard and gave R^2 value of 0.99. TPC was calculated according to the Eq. (8):

$$\text{TPC} = \frac{A \times 2500 \times DF}{S \times a_3} \left[\frac{\text{mg GAE}}{\text{L}} \right], \tag{8}$$

whereby A is the measured absorbance; S is the amount of sample (µL); a_3 is the slope of calibration curve; DF is the dilution factor.

Ferric Reducing Antioxidant Power (FRAP) method was applied to investigate the antioxidant capacity in reference to Benzie and Devaki [21]. FRAP reagent was prepared with 250 µL of acetate buffer solution (pH = 3.6), 25 µL of ferric chloride solution (30 mM), and 25 µL of TPTZ solution (10 mM dissolved in 40 mM HCl). FRAP reagent (1500 µL) was mixed with (30 µL) distilled water followed

by sample solution (20 µL) and incubated for 5 minutes at room temperature in the dark. After exactly 5 minutes, the absorbance measurement was performed at 593 nm on Genesys 5 UV-Visible spectrophotometer. The calibration was realized with ascorbic acid (10 mM) instead of sample solution and expressed as ascorbic acid equivalent and R^2 value of 0.99 was depicted from the calibration curve. The calculation was done using the Eq. (9);

$$AC = \frac{A \times 1550 \times DF}{S \times a_4} \left[\frac{\text{mg ASE}}{\text{L}} \right], \tag{9}$$

where A is the absorbance; S is the amount of sample (µL); a_4 is the slope of calibration curve; DF is the dilution factor. All measurements were triplicated under the same conditions. Data was evaluated by performing significant test based on variance analysis model through Microsoft Excel 2010 software.

3 Results and discussions
3.1 Statistical analysis
Results of statistical evaluation proved that significant level (α) for each sample is quite satisfactory (99.99 % significant level) p-value less than 0.001.

3.1.1 Color compounds concentration
Quantification of betalain compounds in initial feeds, retentates and permeates (500 mL, 1000 mL, 1500 mL, 2000 mL, and final volume) collected during concentration processes with two types of reverse osmosis membranes (RO99 and X20) were conducted by spectrophotometric analysis. The resulting betaxanthin and betacyanin contents in each sample were shown in Tables 1 and 2. As we can

Table 1 Betalains, total phenolic compounds, and antioxidant contents in crudes and retentates of RO99 type reverse osmosis membrane separation

Solvent	Sample	Betaxanthin (mg/L)	Betacyanin (mg/L)	TPC (mg/L)	Antioxidant (mg/L)
Water	Initial	75.46±0.45	117.33±0.32	187.95±18.54	107.7±13.05
	R (500mL)	89.06±1.3	142.27±0.65	299.67±22.91	138.8±41.83
	R (1000mL)	130.13±1.93	216.33±1.21	358.54±17.3	170.65±3.22
	R (1500mL)	182.75±0.81	309.47±0.7	551.03±11.4	195.68±12.04
	R (2000mL)	249.22±1.34	419.65±0.65	868.06±26.83	359.51±25.74
	Final	292.47±1.93	499.03±1.3	1243.96±106.56	1133.15±25.74
Ethanol-water	Initial	87.27±0.93	159.87±0.7	332.13±24.21	173.08±2.67
	R (500mL)	108.06±3.63	196.17±6.16	370.62±10.21	121.35±6.95
	R (1000mL)	127.05±1.45	232.1±0.65	488.38±2.61	133.49±22.45
	R (1500mL)	180.18±2.95	329.63±2.92	754.08±31.7	289.73±14.63
	R (2000mL)	304.41±2.95	551.47±4.54	1005.44±78.57	648.49±74.01
	Final	337.26±4.31	585.2±5.83	1268.87±48.69	698.55±22.53

Table 2 Betalains, total phenolic compounds, and antioxidant contents in crudes and retentates of X20 type reverse osmosis membrane separation

Solvent	Sample	Betaxanthin (mg/L)	Betacyanin (mg/L)	TPC (mg/L)	Antioxidant (mg/L)
Water	Initial	84.44±0.45	167.93±1.3	188.43±2.81	126.99±13.9
	R (500mL)	52.36±2.22	101.93±0.32	133.20±17.23	344.44±17.81
	R (1000mL)	67.5±1.13	129.07±0.97	539.31±0.00	469.26±61.57
	R (1500mL)	112.93±1.13	215.6±1.3	726.12±0.00	1175.56±30.79
	R (2000mL)	190.96±1.13	357.87±3.31	969.79±96.49	1274.73±5.13
	Final	253.33±1.52	450.27±2.07	1003.9±13.78	2428.53±35.92
Ethanol-water	Initial	94.71±0.19	192.13±0.00	214.43±34.46	424.02±17.84
	R (500mL)	66.99±1.13	125.77±0.92	217.67±17.23	420.88±37.71
	R (1000mL)	84.44±1.77	154.73±2.8	471.09±10.34	440.23±10.26
	R (1500mL)	120.38±1.59	219.27±0.97	492.20±48.24	761.94±87.23
	R (2000mL)	136.55±0.00	261.8±0.00	718.81±106.82	1148.95±20.52
	Final	366.78±0.00	586.3±4.54	869.88±120.61	1925.41±118.02

see in Table 1, betaxanthin content of initial feeds for RO99 membrane concentrations were 75.46±0.45 mg/L (water solvent) and 87.27±0.93 mg/L (ethanol-water solvent).

Concentration of betaxanthin compound in both extracts increased continuously in each sample and final retentates contained up to 292.47±1.93 mg/L and 337.26±4.31 mg/L in water and ethanol-water extracts, individually. Betalain compounds content in each sample during RO filtrations were depicted in Figs. 1 and 2 in the function of VRR. Concentration ratios (C_R/C_0) of betaxanthin in final retentates (RO99) were almost the same for both water and ethanol-water extracts as displayed in Fig. 1.

According to Fig. 2, the trend of betacyanin concentration ratio for water extract exceeded ethanol-water data. This means that RO99 membrane had greater effect on concentration of water extract. The amount of betacyanin measured in water and ethanol-water extracts were 117.33±0.32 mg/L (initial), 499.03±1.3 mg/L (final), and 159.87±0.7 mg/L (initial), 585.2±5.83 mg/L (final), respectively.
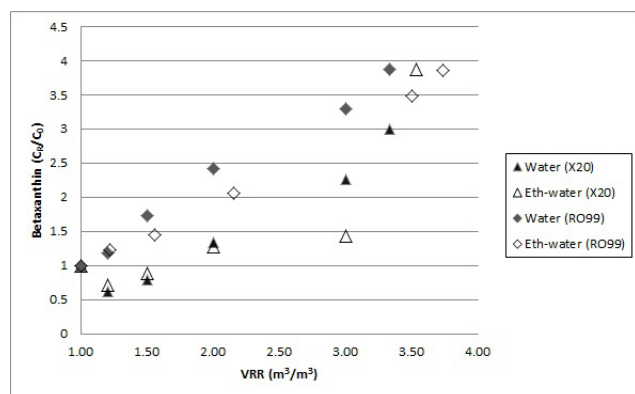

Fig. 1 Variation of betaxanthin color compound content with VRR during concentration by RO99 and X20 reverse osmosis membranes
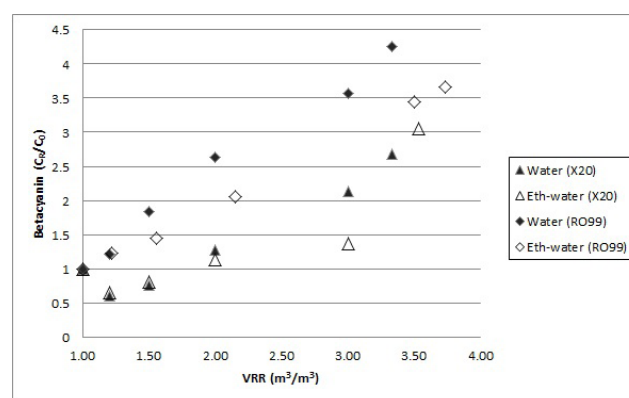

Fig. 2 Variation of betacyanin color compound content with VRR during concentration by RO99 and X20 reverse osmosis membranes

In the case of X20 type RO filtrations, the initial amounts of betalain compounds in water extract were 84.44±0.45 mg/L (betaxanthin) and 167.93±1.3 mg/L (betacyanin) while aqueous ethanol extract contains 94.71±0.19 mg/L of betaxanthin and 192.13±0.00 mg/L of betacyanin. It was noted that these feed concentrations outweighed 500 mL and 1000 mL samples as shown in Table 2.

The possible explanation to this might have been that some amount of water left in the dead space of the pipes of the equipment and caused the dilution of the feed when separation process was started. The amount of betalains went up later on against crude extracts and final concentrates reached of betaxanthin 253.33±1.52 mg/L and betacyanin 450.27±2.07 mg/L in water extract whilst the final concentrates of ethanol-water extract exhibited 366.78±0.00 mg/L and 586.3±4.54 mg/L of betaxanthin and betacyanin, respectively.

In overall point of view, the greater amount of both color compounds was maintained in aqueous ethanol extracts. It proved that 15 v/v% aqueous ethanol solvent

affected more on the extraction of color compounds than water however these compounds are water soluble [22]. Besides, the amount of betacyanin extracted was exhibited to be double of betaxanthin concentration in all cases. The possible explanation is due to the less sensitive property of betacyanin to process conditions than betaxanthin. The concentration ratio of betacyanin was the superiority among the final extracts meaning that the membranes concentrated more amount of betacyanin than betaxanthin. It is rational since betacyanin has larger molecular weight (550 g/mol) than betaxanthin (308 g/mol) [23].

3.2 Total Phenolic Compounds (TPC)

As regard the first, the superior amount of phenolic compounds were beheld in ethanol-water crude extracts (332.13±24.21 mg/L and 214.43±34.46 mg/L) as against water extract (187.95±18.54 mg/L and 188.43±2.81 mg/L) in RO99 and X20 feeds (Tables 1 and 2). Being the batch type extraction and filtration processes, the rationale could be that the profiles of the beetroot peel extracts depended upon the variety and maturation stage of the raw beetroots as reported by Bucur et al. [24]. Nevertheless the efficiency of membranes on concentration of this compound was seen to be higher in water extracts according to Fig. 3.

Furthermore, less amount of TPC was found in final extract of ethanol-water (869.88±120.61 mg/L) with regard to water extract (1003.9±13.78 mg/L) in X20 membrane concentrations. In contrast, large amounts of TPC were retained in both final extracts of RO99 filtrations with quite similar amounts as 1243.96±106.56 mg/L (water) and 1268.87±48.69 mg/L (ethanol-water), separately.

The trends acted almost the same way in all concentration processes. Concentration ratio of TPC became higher in each filtrate along with expanding VRR than
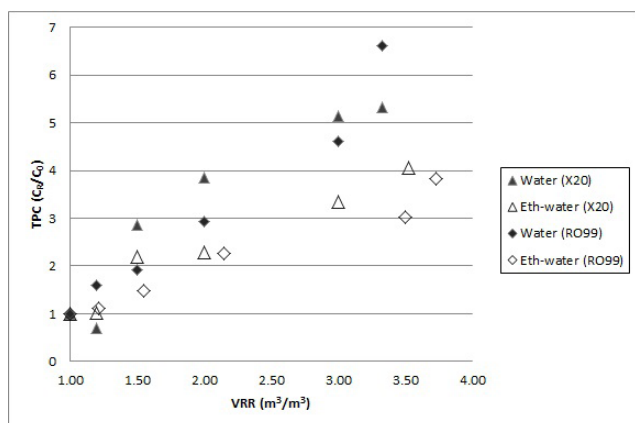
initial ones except the case of water extract (X20) where TPC content was a bit nether in 500 mL sample than in the feed. It might have been the same possible explanation with betalain compounds separation as discussed earlier.

3.3 Antioxidant assay

The antioxidant contents in ethanol-water crude extracts, the feeds for RO99 and X20 membrane filtrations, were recorded as 173.08±2.67 mg/L and 424.02±17.84 mg/L howbeit the water could extract only 107.7±13.05 mg/L and 126.99±13.9 mg/L, respectively (Tables 1 and 2). As we mentioned earlier, specific properties of raw materials could be responsible for these differences in value albeit size, maturity, and ripeness were basic parameters for the beetroot selection. As we can see in Tables 1 and 2, ethanol-water extracts contain less amount of antioxidant in final retentates than water extract, with 698.55±22.53 mg/L and 1925.41±118.02 mg/L as opposed to 1133.15±25.74 mg/L and 2428.53±35.92 mg/L for RO99 and X20 membranes, individually. Fig. 4 represents antioxidant components in each sample varied with VRR.

From the Fig. 4, it was noticed that more antioxidant was concentrated with increasing Volume Reduction Ratio (VRR) in all processes. The trend of concentration ratio of water extract (X20) was astray from others and even overtook drastically the concentration trends of RO99. The huge amount of antioxidant in the final concentrates of water extracts implies that the membranes were quite effective on concentration of different types of water soluble compounds which exhibit antioxidant property. This is reasonable as betalain and phenolic compounds are highly correlated with antioxidant capacity as reported by Bucur et al. [24] and Kavalcová et al. [4].
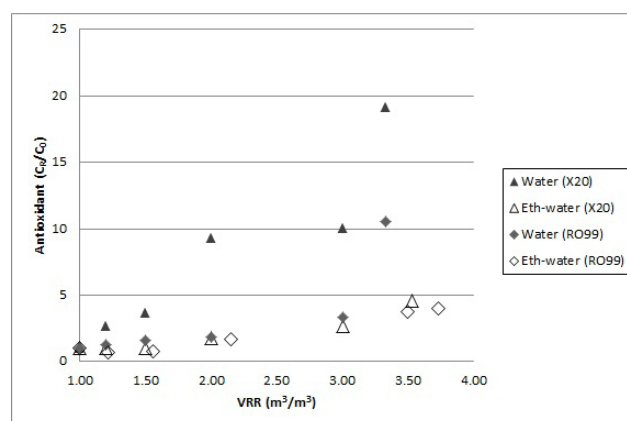

Fig. 3 Total phenolic compound content with VRR during concentration by RO99 and X20 reverse osmosis membranes

Fig. 4 Variation of antioxidant capacity in each sample with VRR during concentration by RO99 and X20 reverse osmosis membranes

3.4 Permeate Flux Measurement

Flux behavior for reverse osmosis filtration of beetroot peel extracts with membrane type of RO99 in the function of volume reduction ratio is shown in Fig. 5. Both water and ethanol-water extracts were fed to the system at 40 bars of transmembrane pressure adjusting the stream temperature at 30 °C. As depicted in Fig. 5, the permeate flux of water extract behaved obviously different from of ethanol-water. Initially, the fluxes of both extracts were tapered off until around VRR 2. That fluxes behavior shows the initial accumulation of the compounds on and/or in the membranes which causes membrane fouling and concentration polarization. The same observation was reported by Couto et al. [25]. Moreover, the flux of ethanol-water reached 16.9 L/(m^2 × h) at VRR = 3.73 after about 35 minutes of concentration time whilst the permeate flux of water extract could reach VRR = 3.33 with 41.27 L/(m^2 × h) permeate flux in very short time (12 minutes). Due to their difference in polarity, ethanol-water solvent could extract some more compounds than only water leading to formation of polarized layer. Nevertheless, physical property of water such as dielectric constant and molecular weight, which exceeded alcoholic solvent, could attribute to elevated flux in water extract as well [26].

Fig. 6 reveals the permeate flux profile of beetroot peel extracts with X20 membrane type filtration. As we can see from Fig. 6, reduction of permeate flux was manifested with elevated process time at fixed transmembrane pressure (40 bars). Like in the case of RO99, the concentration process of water extract was quite faster than ethanol-water extract. It took only 15 minutes for water extract to reach VRR = 3.33 whereas, for ethanol-water, one hour time was necessary to reach the same level of VRR. Besides, the
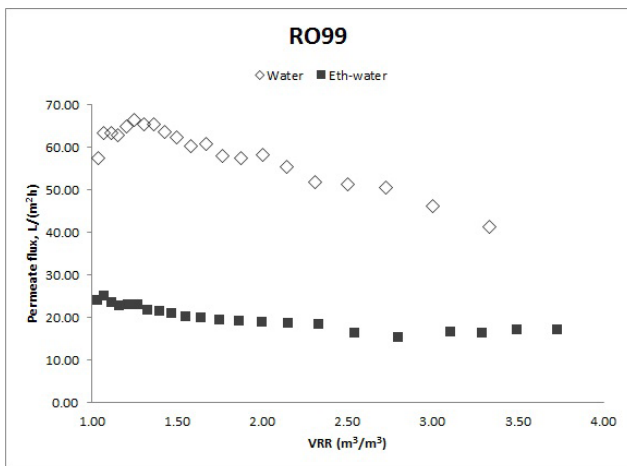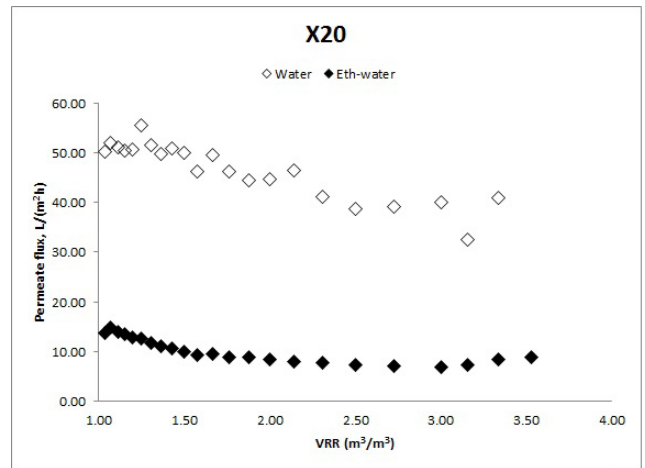

Fig. 6 Changes in permeate flux values with VRR during the concentration process by reverse osmosis type X20

final VRR 3.53 was achieved with ethanol-water extract after 1 hour and 12 minutes of separation time. The accumulation of foulants, cake formation, pore plugging on/in the membrane layers might be the responsible ones for the resistance to permeation [13].

From pure water flux measurements after concentration process, fouling resistances of the membranes were calculated as depicted in Fig. 7. It can be assumed that concentration of ethanol-water extract by RO99 membrane was more effective than of water extract without interfering membrane feature, therefore significant diminished in fouling resistance was demonstrated compared to water extract. Being regarded as non-porous, reverse osmosis membranes are somehow high in fouling propensity on account of cake layer formation [27]. Since we used low fouling resistance type, fouling of X20 type RO membrane was negligible.
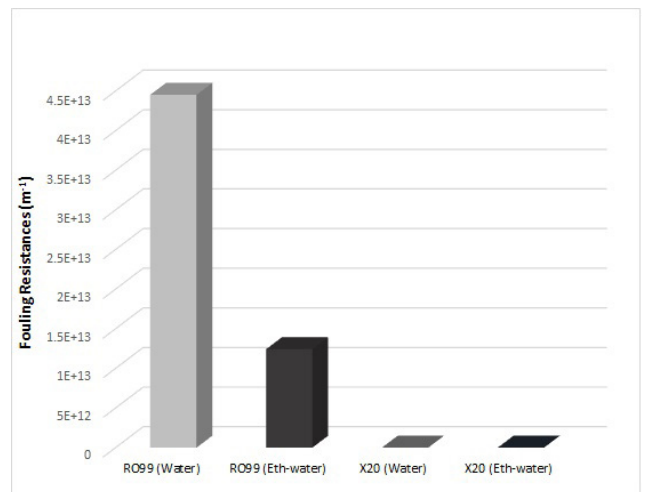

Fig. 5 Permeate flux values changing with VRR during the concentration process by reverse osmosis type RO99

Fig. 7 Fouling resistances of RO99 and X20 membranes

Retention percentages were calculated from the concentration data of final permeates and retentates (Fig. 8) In RO99 membrane concentration processes, betaxanthin and betacyanin retentions in both water and ethanol-water extracts were amounting from 96 % to 99 %. In case of X20 membrane, retentions of betaxanthin and betacyanin were deducted about 6 % and 9 % in water extract, and 17 % and 23 % in ethanol-water extract as opposed to RO99. Since permeates of each membrane did not show the presence of TPC and antioxidant contents, retention was supposed to be 99.99 % for those compounds not only in water extract but also in ethanol-water.

4 Conclusion

Based on all results, concentration level accomplished by RO99 was superior to X20. Retentions of antioxidant and total phenolic compounds reached 99.99 % in both types of extract solutions and betalain retentions were over 95 % as well. Therefore, it is estimated that the filtrations of those valuable compounds with RO99 membrane were fully performed. In our work, extraction process was carried out with only 1:10 ratio of peel-to-solvent. In order to improve the yield of valuable compounds, further investigations with improvements in extraction step are necessary. In accordance with our experimental results, the conclusion comes up with that membrane technology can be applied effectively in concentration or separation of valuable compounds from vegetable wastes.
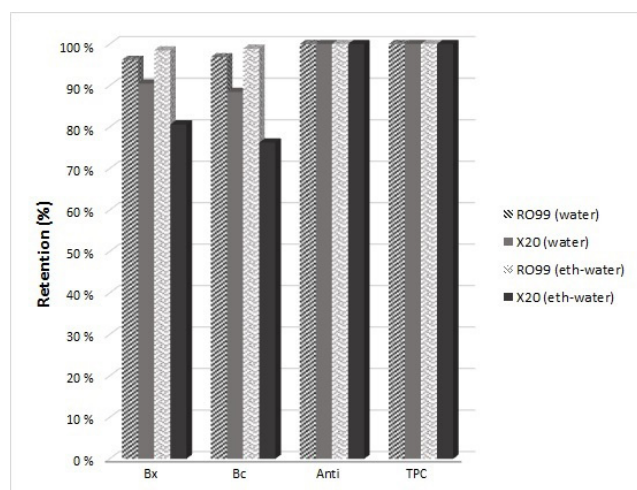

Fig. 8 Retention percentages of RO99 and X20 membranes

Nomenclature

Δt time required to collect the filtrate (h)
ΔV volume of filtrate (L)
A sample absorbance
A_m active surface area of membrane (m^2)
a_1 slope of pure water flux curve before the concentration
a_2 slope of pure water flux curve after the concentration
a_3 slope of TPC calibration curve
a_4 slope of AC calibration curve
AC antioxidant capacity (mg/L)
Bc   betacyanin compounds concentration (mg/L)
BC betalain compounds concentration (mg/L)
Bx   betaxanthin compounds concentration (mg/L)
C_0 feed concentration (mg/L)
C_p concentration of permeate (mg/L)
C_R concentration of retentate (mg/L)
DF dilution factor
J permeate flux of pure water (m^3/(m$^2 \times$ s))
J_x permeate flux of the sample (L/(m$^2 \times$ h))
L path length (cm)
MW molecular weight (g/mol)
R_m membrane resistance (1/m)
R_f fouling resistance (1/m)
S amount of sample (µL)
TMP applied transmembrane pressure difference (Pa)
TPC   concentration of total phenolic compounds (mg/L)
V_0 feed volume (m^3)
V_P volume of permeate (m^3)
V_R volume of retentate (m^3)
μ dynamic viscosity of permeate (Pa $\times$ s)
ε molar extinction coefficient (L/(mol $\times$ cm))

References

[1] Osorio-Esquivel, O., Ortiz-Moreno, A., Álvarez, V. B., Dorantes-Álvarez, L., Giusti, M. M. "Phenolics, betacyanins and antioxidant activity in *Opuntia joconostle* fruits", Food Research International, 44(7), pp. 2160–2168, 2011.
https://doi.org/10.1016/j.foodres.2011.02.011

[2] Sawicki, T., Bączek, N., Wiczkowski, W. "Betalain profile, content and antioxidant capacity of red beetroot dependent on the genotype and root part", Journal of Functional Foods, 27, pp. 249–261, 2016.
https://doi.org/10.1016/j.jff.2016.09.004

[3] Choo, W. S. "Betalains: Application in Functional Foods", In: Mérillon, J. M., Ramawat, K. G. (eds.) Bioactive Molecules in Food, Springer, Cham, Switzerland, 2019, pp. 1471–1498.
https://doi.org/10.1007/978-3-319-78030-6_38

[4] Kavalcová, P., Bystrická, J., Tomáš, J., Karovičová, J., Kovarovič, J., Lenková, M. "The content of total polyphenols and antioxidant activity in red beetroot", Potravinarstvo: Slovak Journal of Food Sciences, 9(1), pp. 77–83, 2015.
https://doi.org/10.5219/441

[5] Murkovic, M. "Phenolic Compounds", In: Trugo, L., Finglas, P. M. (eds.) Encyclopedia of Food Sciences and Nutrition, Academic Press, Oxford, UK, 2003, pp. 4507–4514.
https://doi.org/10.1016/B0-12-227055-X/00914-7

[6] Sawicki, T., Wiczkowski, W. "The effects of boiling and fermentation on betalain profiles and antioxidant capacities of red beetroot products", Food Chemistry, 259, pp. 292–303, 2018.
https://doi.org/10.1016/j.foodchem.2018.03.143

[7] Cai, Y. Z., Sun, M., Corke, H. "Characterization and application of betalain pigments from plants of the Amaranthaceae", Trends in Food Science & Technology, 16(9), pp. 370–376, 2005.
https://doi.org/10.1016/j.tifs.2005.03.020

[8] Bayindirli, A., Yildiz, F., Özilgen, M. "Modelling of Sequential Batch Ultrafiltration of Red Beet Extract", Journal of Food Science, 53(5), pp. 1418–1421, 1988.
https://doi.org/10.1111/j.1365-2621.1988.tb09290.x

[9] Lee, Y. N., Wiley, R. C., Sheu, M. J., Schlimme, D. V. "Purification and Concentration of Betalaines by Ultrafiltration and Reverse Osmosis", Journal of Food Science, 47(2), pp. 465–471, 1982.
https://doi.org/10.1111/j.1365-2621.1982.tb10105.x

[10] Ghosh, P., Pradhan, R. C., Mishra, S., Rout, P. K. "Quantification and Concentration of Anthocyanidin from Indian Blackberry (Jamun) by Combination of Ultra- and Nano-filtrations", Food and Bioprocess Technology, 11(12), pp. 2194–2203, 2018.
https://doi.org/10.1007/s11947-018-2176-4

[11] Mereddy, R., Chan, A., Fanning, K., Nirmal, N., Sultanbawa, Y. "Betalain rich functional extract with reduced salts and nitrate content from red beetroot (*Beta vulgaris* L.) using membrane separation technology", Food Chemistry, 215, pp. 311–317, 2017.
https://doi.org/10.1016/j.foodchem.2016.07.132

[12] Tóth, A. J., Gergely, F., Mizsey, P. "Physicochemical treatment of pharmaceutical process wastewater: distillation and membrane processes", Periodica Polytechnica Chemical Engineering, 55(2), pp. 59–67, 2011.
https://doi.org/10.3311/pp.ch.2011-2.03

[13] Miller, D. J., Kasemset, S., Paul, D. R., Freeman, B. D. "Comparison of membrane fouling at constant flux and constant transmembrane pressure conditions", Journal of Membrane Science, 454, pp. 505–515, 2014.
https://doi.org/10.1016/j.memsci.2013.12.027

[14] Liu, M., Lü, Z., Chen, Z., Yu, S., Gao, C. "Comparison of reverse osmosis and nanofiltration membranes in the treatment of biologically treated textile effluent for water reuse", Desalination, 281, pp. 372–378, 2011.
https://doi.org/10.1016/j.desal.2011.08.023

[15] Bánvölgyi, S., Horváth, S., Stefanovits-Bányai, É., Békássy-Molnár, E., Vatai, G. "Integrated membrane process for blackcurrant (*Ribes nigrum* L.) juice concentration", Desalination, 241(1–3), pp. 281–287, 2009.
https://doi.org/10.1016/j.desal.2007.11.088

[16] Kushwaha, R., Kumar, V., Vyas, G., Kaur, J. "Optimization of Different Variable for Eco-friendly Extraction of Betalains and Phytochemicals from Beetroot Pomace", Waste and Biomass Valorization, 9(9), pp. 1485–1494, 2018.
https://doi.org/10.1007/s12649-017-9953-6

[17] Singh, A., Ganesapillai, M., Gnanasundaram, N. "Optimizaton of extraction of betalain pigments from beta vulgaris peels by microwave pretreatment", In: 14th International Conference on Science, Engineering and Technology (14th ICSET-2017), Vellore, India, IOP Conference Series: Materials Science and Engineering, 263(3), Article number: 032004, 2017.
https://doi.org/10.1088/1757-899X/263/3/032004

[18] Ravichandran, K., Saw, N. M. M. T., Mohdaly, A. A. A., Gabr, A. M. M., Kastell, A., Riedel, H., Cai, Z., Knorr, D., Smetanska, I. "Impact of processing of red beet on betalain content and antioxidant activity", Food Research International, 50(2), pp. 670–675, 2013.
https://doi.org/10.1016/j.foodres.2011.07.002

[19] Bueno, F. G., Machareth, M. A. D., Panizzon, G. P., Lopes, G. C., Mello, J. C. P., Leite-Mello, E. V. S. "Development of a UV/Vis spectrophotometric method for analysis of total polyphenols from *Caesalpinia peltophoroides* Benth", Química Nova, 35(4), pp. 822–826, 2012.
https://doi.org/10.1590/S0100-40422012000400031

[20] Koczka, N., Stefanovits-Bányai, É., Ombódi, A. "Total Polyphenol Content and Antioxidant Capacity of Rosehips of Some *Rosa* species", Medicines, 5(3), Article number: 84, 2018.
https://doi.org/10.3390/medicines5030084

[21] Benzie, I. F. F., Devaki, M. "The ferric reducing/antioxidant power (FRAP) assay for non-enzymatic antioxidant capacity: concepts, procedures, limitations and applications", In: Apak, R., Capanoglu, E., Shahidi, F. (eds.) Measurement of Antioxidant Activity & Capacity: Recent Trends and Applications, John Wiley & Sons Ltd., Hoboken, NJ, USA, 2018, pp. 77–106.
https://doi.org/10.1002/9781119135388.ch5

[22] Delgado-Vargas, F., Jimenez, A. R., Paredes-López, O. "Natural Pigments: Carotenoids, Anthocyanins, and Betalains – Characteristics, Biosynthesis, Processing, and Stability", Critical Reviews in Food Science and Nutrition, 40(3), pp. 173–289, 2010.
https://doi.org/10.1080/10408690091189257

[23] Tamba, A., Servent, A., Mertz, C., Cissé, M., Dornier, M. "Coupling of pressure-driven membrane technologies for concentrating, purifying and fractionizing betacyanins in cactus pear (*Opuntia dillenii* Haw.) juice", Innovative Food Science & Emerging Technologies, 52, pp. 244–255, 2019.
https://doi.org/10.1016/j.ifset.2018.12.008

[24] Bucur, L., Țarălungă, G., Schroder, V. "The betalains content and antioxidant capacity of red beet (*Beta vulgaris* L. subsp. *vulgaris*) root", Farmacia, 64(2), pp. 198–201, 2016.

[25] Couto, D. S., Corrêa Cabral, L. M., da Matta, V. M., Deliza, R., de Grandi Castro Freitas, D. "Concentration of pineapple juice by reverse osmosis: physicochemical characteristics and consumer acceptance", Food Science and Technology, 31(4), pp. 905–910, 2011.
https://doi.org/10.1590/S0101-20612011000400012

[26] Kim, I. C., Jegal, J., Lee, K. H. "Effects of aqueous and organic solutions on the performance of polyamide thin-film composite nanofiltration membranes", Journal of Polymer Science Part B: Polymer Physics, 40(19), pp. 2151–2163, 2002.
https://doi.org/10.1002/polb.10265

[27] Zhu, X., Elimelech, M. "Colloidal Fouling of Reverse Osmosis Membranes: Measurements and Fouling Mechanisms", Environmental Science & Technology, 31(12), pp. 3654–3662, 1997.
https://doi.org/10.1021/es970400v