Article

Performance of Hollow Fiber Ultrafiltration Membranes in the Clarification of Blood Orange Juice

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Abstract: The clarification of blood orange juice by ultrafiltration (UF) was investigated by using three hollow fiber membrane modules characterized by different membrane materials (polysulfone (PS) and polyacrylonitrile (PAN)) and molecular weight cut-off (MWCO) (50 and 100 kDa). The performance of selected membranes was investigated in terms of productivity and selectivity towards total anthocyanin content (TAC), total phenolic content (TPC), and total antioxidant activity (TAA). All selected membranes allowed a good preservation of antioxidant compounds; however, the most suitable membrane for the clarification of the juice was found to be the PS 100 kDa membrane. In optimized operating conditions this membrane exhibited steady-state fluxes of 7.12 L/m^{2}h, higher than those measured for other investigated membranes. Rejections towards TPC and TAA were of the order of 17.5% and 15%, respectively. These values were lower than those determined for PS 50 kDa and PAN 50 kDa membranes. In addition, the PS 100 kDa membrane exhibited a lower rejection (7.3%) towards TAC when compared to the PS 50 kDa membrane (9.2%).

Keywords: blood orange juice; ultrafiltration; clarification; phenolic compounds; antioxidant activity

1. Introduction

Red-colored blood oranges (*Citrus sinensis*) are commonly cultivated in the Mediterranean area. The most common varieties, named *Tarocco*, *Moro*, and *Sanguinello*, are characterized by a red color, of varying intensity and prevalence, due to the presence of anthocyanins found in the flavedo or juice vescicle, mainly consisting of cyanidin-3-glucoside [1,2] and cyanidin-3-(6″-malonyl)-glucoside [3]. These water-soluble pigments contribute, together with other blood orange components (ascorbic acid, flavonoids and hydroxycinnamic acids), to the antioxidant and antiradical activities of the juice [4,5].

These compounds constitute useful markers to allow the recognition and evaluation of nutritional quality in both fresh and processed products [6]. In addition, epidemiological studies highlight a strong relationship between the consumption of fruits and vegetables and health benefits due to the prevention of degenerative processes.

The health-related properties of polyphenols contained in orange juice have been mainly attributed to their antioxidant activity. Indeed, Rapisarda *et al.* [7] demonstrated that the antioxidant efficiency of orange juice may be attributed in a significant part to their content of total phenols while ascorbic acid seems to play a minor role. In addition, for pigmented juices their antioxidant activity appears to be widely influenced by the anthocyanin level.

In recent years, the interest of consumers and producers for bioactive compounds, especially phenolic compounds from natural sources, has remarkably increased. This interest is mainly linked to the increasing demand of natural antioxidants, since they can be used as functional food ingredients for the production of nutraceuticals and cosmetics, or as substitutes of synthetic products in the food industry [8]. Therefore many efforts are involved in the preservation of components which are active in conferring protection against major diseases.

Blood orange juices are naturally cloudy mainly due to the presence of high molecular weight solutes such as pectins which cause problems of haze formation in the extracted juice making the juice highly viscous and difficult to be treated in subsequent processing steps [9].

Conventional clarification processes, based on the use of fining agents, are often discontinuous and require extensive space, due to the intrinsic limitations imposed by the dimensions of vats. They are also characterized by high energy consumption for temperature control and the use of large amounts of coadiuvants and additives (bentonite, gelatines, activated carbon, casein, silica sol, diatomaceous earth, *etc.*) with consequent problems of environmental impact due to their disposal.

Ultrafiltration (UF) membranes have been shown to be of great interest for clarification of fruit juices and have become a commercial success. They are able to retain large species such as microorganisms, lipids, proteins, and colloids while small solutes as for example vitamins, salts, sugars flow together with water. Increased juice yields, continuous operation, reduction of working times, elimination of gelatins, adsorbents, and other filtration aids, easy cleaning and maintenance of the equipment, reduction of waste products are typical advantages of UF over conventional clarification processes [10]. Indeed, UF membranes have been proven to be effective for the clarification of several fruit juices including kiwi fruit [11], mosambi [12], apple [13], orange [14,15], passion fruit [16], pineapple [17], and cactus pear [18].

It is well known that membrane material and molecular weight cut-off (MWCO) may influence the juice quality. In addition, operating parameters, such as transmembrane pressure (TMP), feed velocity

and temperature, have a strong effect on the optimization of membrane performance in terms of maximization of permeate output and minimization of energy consumption. Indeed, one of the main problems of using membranes for clarifying juices is the decay of permeate flux due to membrane fouling which is attributed to the accumulation of macromolecular or colloidal species on the membrane surface and to the possible precipitation of smaller solutes in the membrane pores. Although several studies indicate a positive effect on the permeate flux when TMP is raised, the use of higher TMP values leads to a more accentuated formation of fouling and polarized layers.

This study was aimed at evaluating the performance of different hollow fiber UF membranes in the clarification of depectinized blood orange juice. In particular, the experimental work was addressed to evaluate the influence of membrane material and molecular weight cut-off (MWCO) on the content of antioxidant compounds of the clarified juice. At this purpose, three hollow fiber membrane modules with different molecular weight cut-off (MWCO) (50 and 100 kDa) and membrane material (polysulfone and polyacrylonitrile) were used. The performance of each membrane module in terms of permeate flux was also evaluated in optimized conditions of transmembrane pressure (TMP) and axial feed flow rate (Q_f).

2. Materials and Methods

2.1. Blood Orange Juice

Blood orange juice was obtained by squeezing oranges of *Tarocco* variety harvested in Corigliano Calabro (Cosenza, Italy). Fruits were manually washed with running water in order to remove foreign material from the skin. Hence they were halved and then squeezed by a domestic juicer (Aristarco S.r.l., Treviso, Italy). The pulp was then treated with 1% (*w/w*) of a commercial pectinase from *Aspergillus aculeatus* (Pectinex® Ultra SP-L from *Aspergillus Aculeatus*, Sigma-Aldrich, Milan, Italy). The enzyme is able to hydrolyze both high and low esterified pectins and also partially hydrolyze cellulose and hemicellulose [19]. The puree was incubated for 4 h at room temperature in plastic tanks and then filtered with a nylon cloth. The extracting procedure gave an average juice yield of 43% (*w/w*). The juice was stored at −17 °C and was defrosted to room temperature before use.

2.2. Ultrafiltration Experimental Setup

The clarification of the juice was performed by using a laboratory UF bench plant (DSS LabUnit M10) supplied by Danish Separation System AS, Denmark. The equipment consisted of a 5 L feed tank, a cross-flow pump (ECO type GA4-KDT-TTU), two pressure gauges (0–2.5 bar) located at the inlet (P_{in}) and outlet (P_{out}) of the membrane module, a pressure control valve, and a multitube heat exchanger fed with tap water. The temperature (T) of the feed was controlled by circulating cooling water through the heat exchanger; the axial feed flow-rate (Q_f) and the transmembrane pressure (TMP) were controlled by using a needle concentrate valve and by setting the speed of the pump.

Three different hollow fiber membrane modules, with dimensions of 50 × 330 mm and an effective membrane surface area of 0.16 m², were used to clarify the depectinized juice. They were supplied by China Blue Star Membrane Technology Co. Ltd. (Beijing, China). Their characteristics are reported in Table 1.

Experimental runs were performed according to the total recycle configuration (recycling both permeate and retentate streams in the feed tank) in order to study the effect of transmembrane pressure (TMP) and axial feed flow rate (Q_f) on the permeate flux. TMP was modified in the range 0.3–1.2 bar in fixed conditions of Q_f (100 L/h) and temperature (20 °C). Q_f was modified in the range 25–140 L/h in fixed conditions of TMP (0.6 bar) and temperature (20 °C).

The juice was also clarified according to the batch concentration mode (recycling the retentate in the feed tank and collecting separately the permeate stream) in optimized operating conditions (TMP, 0.5 bar; T, 20 °C; Q_f, 140 L/h) up to a volume reduction factor (VRF) of 3.

The VRF is defined as the ratio between the initial feed volume and the final retentate volume, according to the following equation:

$$VRF = \frac{V_f}{V_r} = 1 + \frac{V_p}{V_r} \tag{1}$$

where V_f, V_p, and V_r are the volume of feed, permeate, and retentate, respectively.

The hydraulic permeability of each membrane module was determined by the slope of the straight lines obtained plotting the water flux values in selected operating conditions *versus* the applied TMP. After the experiments, each membrane module was cleaned in three steps. The first cleaning step was performed recirculating distilled water for 20 min through the membrane module. In the second step the membrane module was submitted to a cleaning procedure using a 0.1% NaOH solution at a temperature of 40 °C for 60 min. Finally the membrane module was cleaned with a 1% *w/w* enzymatic solution (Ultrasil 53, Henkel KGaA, Dusseldorf, Germany) at 40 °C for 60 min and then rinsed with tap water for 20 min. This cleaning protocol allowed to recover about 90%–95% of the initial hydraulic permeability of the UF membranes.

Table 1. Characteristics of hollow fiber ultrafiltration (UF) membrane modules.

Membrane Type	DCQ II-006C-PS50	DCQ II-006C-PS100	DCQ II-006C-PAN50
Membrane material	Polysulfone	Polysulfone	Polyacrylonitrile
Internal diameter of fibers (mm)	2.1	2.1	2.1
Operating pressure (bar)	0–1.5	0–1.5	0–1.2
Operating temperature (°C)	5–45	5–45	5–45
Operating pH	2–13	2–13	2–10
Molecular weight cut-off (kDa)	50	100	50

2.3. Analytical Methods

2.3.1. Physicochemical Analysis

The total soluble solid (TSS) content of the juice samples was measured by an Abbe digital refractometer Bellingham + Stanley 60/DR (Bellingham and Stanley Ltd., Kent, UK) and expressed as °Brix. The suspended solids content was determined in relation to the total juice (%, *w/w*) by centrifuging, at 2000 rpm for 20 min, 45 mL of a pre-weighted sample; the weight of settled solids was determined after removing the supernatant. pH was measured by an Orion Expandable ion analyzer EA 920 pH meter (Allometrics, Inc., Baton Rouge, LA, USA) with automatic temperature

compensation. Viscosity was measured by using a RFS III viscometer (Rheometric Scientific, Piscataway, NJ, USA).

2.3.2. Total Phenolic Content (TPC)

The total phenolic content was estimated colorimetrically by using the Folin-Ciocalteu method [20]. The method is based on the reduction of tungstate and/or molybdate in the Folin-Ciocalteu reagent by phenols in alkaline medium resulting in a blue colored product (λ_{max} 756 nm). The estimation of total phenols was carried out in triplicate and results were expressed as mg/L gallic acid.

2.3.3. Total Antioxidant Activity (TAA)

TAA was determined by an improved version of the ABTS radical cation decolourisation assay in which the radical monocation of the 2,2′-azino-bis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS$^+$) is generated by oxidation of ABTS with potassium persulphate before the addition of the antioxidant [21,22]. This method gives a measure of the antioxidant activity of pure substances and of mixtures by monitoring the reduction of the radical cation as the percentage inhibition of absorbance at 734 nm. Spectrophotometric measurements were performed by using a UV-Visible recording spectrophotometer (UV-160 A, Shimadzu Scientific Instruments, Inc., Japan) at 30 °C. ABTS was dissolved in water at 2 mM concentration: ABTS radical cation was produced by reacting 10 mL of ABTS stock solution with 100 μL of 70 mM potassium persulfate solution (ABTS:$K_2S_2O_8$ = 1:0.35 molar ratio) and allowing the mixture to stand in the dark at room temperature for 12–16 h before use. Work solution was prepared diluting 1 mL of the ABTS radical cation solution to 25 mL with PBS buffer (5 mM Na_2HPO_4, 5 mM NaH_2PO_4, NaCl 9 g/L, pH = 6.8) to a final UV absorbance of 0.70 ± 0.02 at 734 nm. After the addition of 10 μL of sample to 10 mL of ABTS work solution, the absorbance at 734 nm was recorded every min for a total of 6 min. The value at 5 min was used to calculate the results reported as TAA, expressed in terms of mM trolox equivalent. Each determination was performed in triplicate. Results were expressed as mean ± SD of three samples.

2.3.4. Anthocyanins

Anthocyanins were assessed by using an HPLC system (Hitachi D-7000 System) equipped with a pump, a UV-Vis detector, and a data acquisition system. Chromatographic separation was performed by using a Luna C 18(2) column (250 × 4.6 mm, 5 μm Phenomenex, Torrance, CA, USA); the following conditions were used: V = 1 mL min^{-1}; T = 25 °C; λ = 518 nm. The mobile phase was a mixture of H_2O/HCOOH (9:1) as solvent A and H_2O/HCOOH/CH_3CN (4/1/5) as solvent B.

Anthocyanins separation was achieved by using the following linear gradient: starting condition, 88% A, 12% B; 26 min, 70% A, 30% B; 35 min, 100% B; 43 min, 88% A, 12% B; 46 min 88% A, 12% B. Anthocyanins were identified by matching the retention time and their spectral characteristics against those of standards (cyanidin 3,5-diglucoside, delphinidin 3-glucoside, cyanidin 3-glucoside, peonidin 3-glucoside).

Quantification was made according to the linear calibration curves of standard compounds [1].

The rejection (R) of UF membranes towards specific compounds was calculated as follows:

$$R = (1 - \frac{C_p}{C_f}) \cdot 100$$

(2)

where C_f and C_p are the concentration of a specific component in the permeate and feed, respectively.

2.4. Statistical Analysis

Analyses of physicochemical parameters were performed in triplicate. Results were given as mean ± standard deviation. One-way analysis of variance (ANOVA) was used to compare the means. Differences were considered to be significant at $p < 0.05$. Statistical analyses were performed with use of Microsoft Excel software (version 2010; Microsoft Corporation; Redmond, WA, USA).

3. Results and Discussion

3.1. Physicochemical Properties of Blood Orange Juice

The physicochemical characteristics of the depectinized juice used in the UF experiments are reported in Table 2. The results related to the TSS, pH, TAA, and TPC evaluation are in agreement with data reported in literature for blood orange juice [6,23,24]. The total anthocyanin content of the juice was estimated to be 7.56 ± 0.87 mg/L. This value, calculated as the sum of individual anthocyanins determined by HPCL analyses, is lower than the amount of anthocyanins determined in orange juices of Tarocco varieties (between 41 and 133 mg/L) according to the data reported by Mondello *et al.* [1]. According to Maccarone *et al.* [3] cyanidin-3-glucoside resulted as the main component of the fraction [3].

3.2. Effect of Operating Conditions on Permeate Flux

Figure 1 shows the effect of TMP on the steady-state permeate flux in selected operating conditions of axial feed flow rate (100 L/h) and temperature (20 °C) for all the investigated membranes. At low pressures the permeate flux resulted proportional to the applied pressure; a further increase in pressure did not improve the permeate flux and a limiting flux value was reached. According to the gel polarization model, the existence of a limiting flux is related to the concentration polarization phenomenon that arises as the feed solution is convected towards the membrane where the separation of suspended and soluble solids from the bulk solution takes place. The formation of a viscous and gelatinous-type layer is responsible for an additional resistance to the permeate flux in addition to that of the membrane [25].

For all the investigated membranes a limiting flux was observed at an applied pressure of 0.6 bar. As expected, in the selected operating conditions, PS membranes with MWCO of 100 kDa exhibited higher fluxes when compared to 50 kDa membranes.

Figure 2 shows the effect of axial feed flow rate (Q_f) on the steady-state permeate flux in fixed conditions of TMP (0.6 bar) and temperature (20 °C). For all the investigated membranes an increase in the flow rate led to higher permeate fluxes. According to the film model an increase in the recirculation velocity reduces concentration polarization, enhances the mass transfer coefficient, and increases the permeation flux [26].
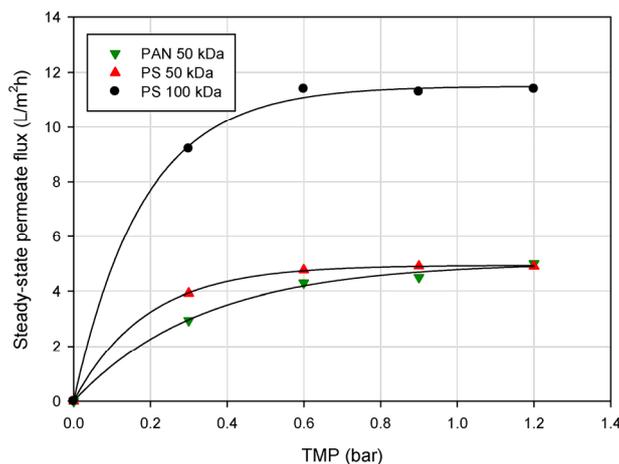
Figure 1. Ultrafiltration (UF) of blood orange juice. Effect of TMP on permeate flux (total recycle configuration; Q_f, 100 L/h; T, 20 °C).

Table 2. Physicochemical characterization of depectinized blood orange juice.

| pH	3.5 ± 0.1
TSS (°Brix)	10.2 ± 0.2
Suspended solids (%, *w/w*)	12.0 ± 0.2
Viscosity (mPa s)	1.90 ± 0.04
TPC (mg GAE/L)	907.22 ± 66.34
TAA (mM Trolox)	6.50 ± 0.46
TAC (mg/L)	7.56 ± 0.87
Cyanidin-3-glucoside (mg/L)	5.98 ± 0.75
Cyanidin-3,5-diglucoside (mg/L)	0.20 ± 0.04
Delfinidin-3-glucoside (mg/L)	0.84 ± 0.15
Peonidin-3-glucoside (mg/L)	0.54 ± 0.01

TSS, total soluble solids; TPC, total phenolic content; GAE, gallic acid equivalent; TAA, total antioxidant activity; TAC, total anthocyanin content.
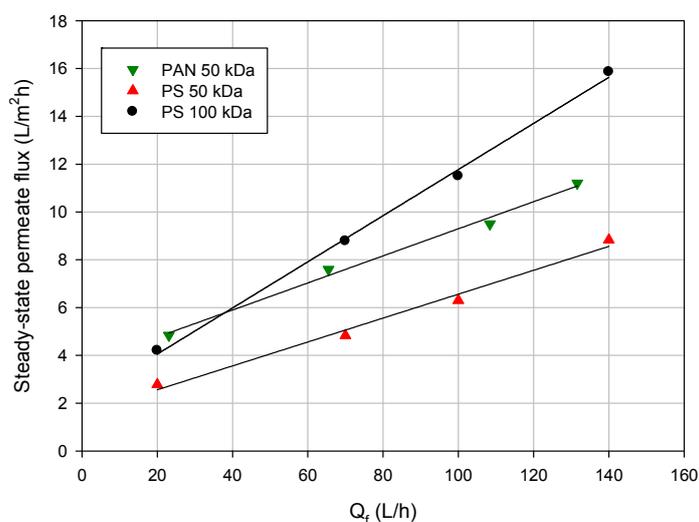
Figure 2. UF of blood orange juice. Effect of Q_f on permeate flux (total recycle configuration; TMP, 0.6 bar; T, 20 °C).

3.3. Effect of Volume Reduction Factor on Permeate Flux

Figure 3 shows the effect of VRF on the permeate flux in optimized operating conditions of TMP and Q_f for all the investigated membranes. Results showed that the permeate flux decreased gradually by increasing the VRF due to concentration polarization and gel formation. In particular, the permeate flux *vs.* VRF curve is characterized by an initial period in which a rapid decrease of permeate flux occurs; a second period, up to VRF 1.5, corresponding to a smaller decrease of permeate flux; and a third period characterized by a steady-state value. In the selected operating conditions, PS membranes with a MWCO of 100 kDa exhibited steady-state permeate fluxes of 7.12 L/m^2h; lower steady-state fluxes (of the order of 3.5–4 L/m^2h) were detected for 50 kDa membranes.
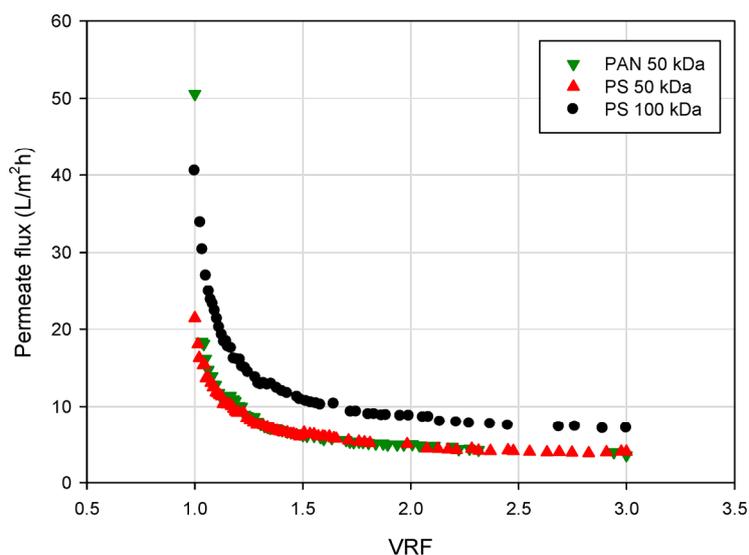
Figure 3. UF of blood orange juice. Effect of VRF on permeate flux (batch concentration configuration; TMP, 0.5 bar; Q_f, 130 L/h; T, 20 °C).

3.4. Effect of UF on Chemical Parameters of Blood Orange Juice

Suspended solids in the depectinized juice were completely removed independently by the used membranes: the resulting permeate was a clear juice with a brilliant red color.

Table 3 shows the effect of the different UF membranes on some characteristic properties of blood orange juice. The TSS content of permeates decreased slightly with UF. In addition, TSS levels appeared to be higher in the retentate than in the permeate fraction: this phenomenon can be attributed to the presence of a high suspended solids content in the pulpy products that can interfere with the measurement of the refractive index. These observations corroborate the results obtained by Toker *et al.* [27] in the clarification of blood orange juice with different UF membranes.

Table 3. Effect of different hollow fiber UF membranes on TSS, TPC, TAC, and TAA of blood orange juice.

UF Membrane	Sample	TSS (° Brix)	TPC (mg GAE/L)	TAC (mg/L)	TAA (mM Trolox)
PS 100 kDa	Feed	10.0 ± 0.2 [a]	835.42 ± 0.43 [a]	7.33 ± 0.14 [a]	6.0 ± 0.8 [a,b]
	Permeate	9.8 ± 0.2 [a]	687.08 ± 0.22 [b]	6.79 ± 0.13 [b]	5.1 ± 0.2 [a]
	Retentate	-	950.83 ± 0.21 [c]	7.71 ± 0.15 [a]	7.1 ± 1.2 [b]
PS 50 kDa	Feed	10.2 ± 0.2 [a]	920.00 ± 0.47 [a]	6.83 ± 0.13 [a]	6.6 ± 0.5 [a]
	Permeate	10.0 ± 0.2 [a]	705.00 ± 0.34 [b]	6.20 ± 0.12 [b]	5.1 ± 0.3 [b]
	Retentate	-	1217.08 ± 0.82 [c]	7.02 ± 0.14 [a]	8.9 ± 1.0 [c]
PAN 50 kDa	Feed	10.4 ± 0.2 [a]	966.25 ± 0.27 [a]	8.52 ± 0.17 [a]	6.9 ± 0.9 [a]
	Permeate	10.2 ± 0.2 [a]	738.33 ± 0.19 [b]	8.03 ± 0.16 [b]	5.3 ± 0.4 [b]
	Retentate	-	1095.80 ± 0.70 [c]	8.80 ± 0.17 [a]	7.8 ± 0.8 [a]

For each membrane, mean values within a column with different letters are significantly different at $p < 0.05$.

As shown in Table 3, the TPC of clarified juice was affected by the MWCO of selected membranes. The reduction of the phenolic content in the UF permeate was more pronounced with decreasing the MWCO from 100 to 50 kDa. Indeed, rejections of 50 kDa membranes towards phenolic compounds were of about 23%, while a rejection of 17.75% was detected for the PS 100 kDa membrane (Table 4). The positive relationship between MWCO and TPC is in agreement with data reported by other authors in the clarification of blood orange [27], pineapple [28], and apple juice [29] with UF membranes. In addition, a strong relationship was observed between the rejection of UF membranes towards phenolic compounds and the TAA rejection. 50 kDa membranes exhibited TAA rejections of about 23%, while the PS 100 kDa membrane showed a TAA rejection of about 15%. These results can be attributed to the strong contribution of polyphenols to the TAA of the blood orange juice. As shown by Roginsky and Lissi [30] the total phenols determined by Folin-Ciocalteu method can be correlated to the antioxidant activity determined by different methods including ABTS and 2,2-diphenyl-1-picrylhydrazyl (DPPH) assays.

Table 4. Rejection of different UF membranes towards TPC, TAC, TAA, and TSS of blood orange juice.

UF Membrane	Rejection (%)			
	TSS	TPC	TAC	TAA
PS 100 kDa	2.0 ± 0.2 [a]	17.75 ± 0.43 [a]	7.4 ± 0.14 [a]	15.0 ± 0.8 [a]
PS 50 kDa	1.9 ± 0.2 [a]	23.37 ± 0.47 [b]	9.2 ± 0.13 [b]	22.7 ± 0.5 [b]
PAN 50 kDa	1.9 ± 0.2 [a]	23.60 ± 0.30 [b]	5.7 ± 0.17 [c]	23.2 ± 0.9 [b]

For each membrane, mean values within a column with different letters are significantly different at $p < 0.05$.

The content of total anthocyanins was not particularly affected by the UF process. For all the selected membranes the TAC of permeate fractions was slightly lower than that measured in the depectinized juice (Table 3). Consequently the bright red color of the original juice was perfectly preserved in the clarified juice. The rejection of the investigated membranes towards TAC was in the range of 5.7%–9.2% with the PAN 50 kDa membrane exhibiting the lowest anthocyanins retention. Previous studies reported a loss of TAC in clear blood orange juice of 9.4% when the juice is clarified with a tubular 15 kDa made in polyvinylidenefluoride (PVDF) [15].

The content of individual anthocyanins determined in the permeate and retentate fractions of the investigated membranes is reported in Table 5. For the cyanidin-3-glucoside, the main component of the anthocyanin fraction, the PAN 50 kDa membrane showed the lowest rejection (of about 5.7%). For the PS membranes the rejection towards this compound was of 7.2% for the 100 kDa membrane and 8.8% for the 50 kDa membrane and therefore correlated to the MWCO of the membrane.

Table 5. Effect of different hollow fiber UF membranes on individual anthocyanins of blood orange juice.

Membrane type	Anthocyanins	Feed (mg/L)	Permeate (mg/L)	Retentate (mg/L)
PS 100kDa	Cyanidin-3,5-diglucoside	0.24 ± 0.01 [a]	0.20 ± 0.01 [b]	0.29 ± 0.01 [c]
	Cyanidin-3-glucoside	5.64 ± 0.11 [a]	5.23 ± 0.10 [b]	6.00 ± 0.12 [c]
	Delfinidin-3-glucoside	0.91 ± 0.01 [a]	0.90 ± 0.01 [ab]	0.88 ± 0.01 [b]
	Peonidin-3-glucoside	0.54 ± 0.01 [a]	0.46 ± 0.01 [b]	0.54 ± 0.01 [a]
PS 50kDa	Cyanidin-3,5-diglucoside	0.17 ± 0.01 [a]	0.16 ± 0.01 [a]	0.18 ± 0.01 [a]
	Cyanidin-3-glucoside	5.46 ± 0.10 [a]	4.98 ± 0.10 [b]	5.56 ± 0.11 [a]
	Delfinidin-3-glucoside	0.67 ± 0.01 [a]	0.60 ± 0.01 [b]	0.70 ± 0.01 [a]
	Peonidin-3-glucoside	0.53 ± 0.01 [a]	0.46 ± 0.01 [b]	0.58 ± 0.01 [c]
PAN 50kDa	Cyanidin-3,5-diglucoside	0.19 ± 0.01 [a]	0.18 ± 0.01 [a]	0.24 ± 0.01 [b]
	Cyanidin-3-glucoside	6.84 ± 0.13 [a]	6.45 ± 0.13 [b]	6.94 ± 0.13 [a]
	Delfinidin-3-glucoside	0.95 ± 0.02 [a]	0.90 ± 0.02 [b]	0.98 ± 0.02 [a]
	Peonidin-3-glucoside	0.54 ± 0.01 [a]	0.50 ± 0.01 [b]	0.64 ± 0.01 [c]

For each specific compound, mean values within a row with different letters are significantly different at $p < 0.05$.

According to the results, all selected membranes allowed very good preservation of anthocyanins in the clarified orange juice. This is a very important aspect in relation to the juice processing, including also an eventual concentration step, and its marketing value. Indeed, because of its high cost the red dye of red orange juice based drinks is often achieved by the addition of natural and synthetic dyes (*i.e.*, red berries, allura red, *etc.*) reducing the amount of blood orange juice in the final product [8]. Therefore, in the interest of consumers, the preservation of the initial anthocyanin content of the fresh juice should be guaranteed for the entire processing cycle and commercial duration of the juice.

4. Conclusions

The clarification of blood orange juice using hollow fiber UF membranes with different membrane material and MWCO has been studied. The performance of selected membranes has been investigated in terms of productivity and selectivity towards compounds contributing to the antioxidant activity of the juice.

In optimized operating conditions of pressure and feed flow rate, PS membranes with a MWCO of 100 kDa exhibited steady-state permeate fluxes of 7.12 L/m^{2}h; lower steady-state fluxes (of the order of 3.5–4 L/m^{2}h) were detected for 50 kDa membranes.

According to the results, all selected membranes allowed a good preservation of the original composition of the blood orange juice in terms of antioxidant compounds and anthocyanin content. Therefore, the clarification process of the juice based on the exclusive use of membrane filtration, was found to be an effective method for clear blood orange juice production with high quality attributes.

On the basis of permeate flux data and chemical composition of the clarified juice, the most suitable membrane for the clarification of the juice was found to be the PS 100 kDa membrane.

Author Contributions

Alfredo Cassano conceived and designed the experiments. Experimental activities on laboratory scale with ultrafiltration membranes were carried out by Carmela Conidi and Fitim Destani. Carmela Conidi was also involved in the analytical characterization of permeate and retentate samples. All Authors contributed to the interpretation and discussion of experimental results.

Conflicts of Interest

The authors declare no conflict of interest.

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