Journal of Environmental Chemical Engineering 12 (2024) 114845

Contents lists available at ScienceDirect

Journal of Environmental Chemical Engineering

journal homepage: www.elsevier.com/locate/jece

Biorefinery approach for the development of an integrated extraction and purification process for the recovery of pectin derived compounds and flavonoids from onion peels

O. Benito-Román *, M.O. Ruiz, M.T. Sanz, S. Beltrán

Department of Biotechnology and Food Science (Chemical Engineering Section), Faculty of Sciences. University of Burgos, Plaza Misael Bañuelos s/n, Burgos 09001, Spain

ARTICLE INFO

Keywords:
Subcritical water extraction
Diafiltration
Membrane
Biorefinery
Oligosaccharides

ABSTRACT

This work aims the valorisation of onion peels by recovering high added value compounds (flavonoids and pectin derived compounds (PDC)) in a three stages water-based cascade process, using subcritical water (SubW) hydrolysis and diafiltration using ceramic tubular membranes. After quercetin and derivatives were recovered in a hydroalcoholic extraction process, the onion peels were subjected to a SubW process that yielded three different hydrolysates (severity factors in the range 2.88–3.29). These hydrolysates resulted to be complex multicomponent mixtures with variable shares of partially hydrolysed PDC, degradation products (HMF, furfural, organic acids) and free monosaccharides. The 50 kDa membrane effectively removed more than 98 % of the impurities present in the hydrolysates after a dilution volume of 3.5. In order to mitigate the observed membrane fouling, a pre-treatment step via centrifugation was used to remove colloidal matter besides colour which allowed the use of the 5 kDa membrane to obtain a retentate that contained over 80 % of the initial PDC with a final purity of 82 % after a dilution volume of only 2.0.

1. Introduction

The food processing industry produces significant amounts of waste, leading to both environmental and economic challenges. A prominent example is onion peel, the dry outer layer of the onion, which is generated not only by the processing industry but also by crop fields or household consumption [1]. In the European Union alone, it is estimated that 0.6 Mt of onion skin waste are generated annually [2], with key producing countries such as the Netherlands (24 % of the total), ahead of Spain (20 %) and France (12 %) [3].

Onion peels are rich in valuable compounds such as flavonoids (mainly quercetin and other derivatives), which are known for their antioxidant, anti-inflammatory, or antimicrobial properties [4] and structural carbohydrates present in the cell walls, such as pectin, hemicellulose and cellulose [5]. Pectin is a complex heteropolysaccharide rich in D-galacturonic acid [6], that consists of at least three distinct structural domains, namely homogalacturonans (linear) and the branched rhamnogalacturonan I and rhamnogalacturonan II [7], with many different potential applications. Despite the presence of valuable compounds, the majority of onion peels are either sent to landfills or used for low-value applications. This not only results in the loss of valuable resources but also exacerbates environmental issues, such as greenhouse gas emissions from landfills and the inefficient use of agricultural by-products. Additionally, it leads to significant economic losses for producers, as the disposal costs of onion waste can be substantial, reaching up to 40 €/t [8]. The sheer volume of onion skins being discarded underscores the pressing need to develop sustainable strategies for their valorisation. The present valorisation strategy aims the recovery of the flavonoids (using different alcohols [9–11] or subcritical water [12,13]) or the simple and complex carbohydrates, such as pectin, pectic oligosaccharides [14,15] or low-lignin raw dietary fibre [5]. This approach, however, is limited to recovering only one type of compound at a time, highlighting the need for a sequential strategy that can efficiently recover all valuable products with high purity.

In this context, waste biorefineries have the potential to serve as a catalyst for transitioning from a linear economy to a circular bioeconomy [16], which seeks to minimize waste and make the most of available resources. These facilities integrate processes that convert waste into raw materials, enabling the production of fuels, energy, and/or chemicals through environmentally friendly methods [17]. Besides the environmental advantages, the development of efficient and

* Corresponding author.
E-mail address: obenito@ubu.es (O. Benito-Román).

https://doi.org/10.1016/j.jece.2024.114845

Available online 20 November 2024

Nomenclature

List of symbols

A	membrane area (m^2)
AcA	acetic acid
ara	arabinose
C	compound concentration ($mg{\cdot}L^{-1}$)
DP	degradation products
DV	diafiltration volume (V_w/V_0)
F	furfural
FA	Formic acid
GalA	galacturonic acid
gal	galactose
GalAO	galacturonic acid oligomeric
glu	glucose
HMF	hydroxymethyl furfural
J	permeate flux ($L\ m^{-2}{\cdot}h^{-1}$)
J*	critical flux parameter defined in Eq. 8 ($L\ m^{-2}{\cdot}h^{-1}$)
J_0	pure water permeate flux ($L\ m^{-2}{\cdot}h^{-1}$)
k	fouling index of Eq. 8
k_c	fouling index of Eq. 9 (s^{-1})
k_{cl}	fouling index of Eq. 11 ($s\ m^{-2}$)
k_s	fouling index of Eq. 10 ($s^{-0.5}m^{-0.5}$)
n	parameter of Eq. 8
P	Pressure (bar)
PDC	pectin derived compounds
POS	pectic oligosaccharides
r_f	fouling resistance (m^{-1})
r_m	membrane hydraulic resistance (m^{-1})
R	retention coefficient estimated by Eq. 3 (%)
R^2	determination coefficient
rha	rhamnose
SubW	subcritical water
SubWH	subcritical water hydrolysate
T	temperature (°C)
TCA	total carboxylic acids
TMS	total monosaccharides
TMP	transmembrane pressure
TPC	total phenolic compounds
Tr	transmission coefficient estimated by Eq. 4
TS	total solids
V	volume (L)
xyl	xylose

Greek symbols

α	selectivity estimated by Eq. 5
ζ	zeta potential
ρ	density (kg/m^3)
μ	viscosity of the permeate (Pa·s)

Subscripts

0	subcritical water hydrolysate used as feed solution
i	each group of hydrolysate compounds
j	each group of hydrolysate compounds, without the oligosaccharide fraction
p	permeate stream
r	retentate stream
w	pure water for the continuous feed diafiltration experiments

economically viable processes for onion peels valorisation could have substantial economic benefits, providing new revenue streams for the food industry and contributing to job creation in rural areas, near the source of raw materials [18]. The high volume of onion production in Europe, which has grown by 25 % in period 2018–2022 [1], guarantees a constant flow of raw material for this facilities. Nonetheless, there is a broader need to optimize the entire supply chain, including factors such as transportation logistics, facility size, and operational strategies [19], in order to ensure the economic viability of the biorefinery.

To optimize the use of onion peels from a biorefinery standpoint, a more comprehensive valorisation approach should target, besides flavonoids, other significant structural components, such as pectin [15]. From the three main waste biorefinery types identified by Sarangi et al. [20] (biological, thermochemical and chemical) the thermochemical type, based on a hydrothermal process using subcritical water (SubW), offers significant potential for obtaining pectin and pectin-derived compounds (PDCs). By carefully controlling the operating conditions, both the extraction and hydrolysis of pectin can be achieved in a single step. SubW is defined as water at temperatures between 100 and 374 °C, maintained in the liquid phase through pressurization. Under these conditions, water demonstrates exceptional solvent and hydrolysis properties due to a notable reduction in its dipole moment, viscosity, and ionic product [21]. Nevertheless, in practical applications, the primary challenge with SubW lies in controlling experimental conditions, as this can lead to the formation of monosaccharides that can further react to form undesired degradation products (organic acids and furfural or hydroxymethylfurfural) [22] and uncontrolled molecular weight loss of pectin [23]. To achieve a high-purity product the elimination of these impurities is essential [22,24].

For this purpose, ultrafiltration (UF) processes have emerged as environmentally friendly methods to efficiently purify, concentrate, or fractionate bioactive compounds from the downstream processing streams obtained after processing agricultural wastes [18]. These membrane-based processes present several advantages, such as the potential scalability, the reduced energy consumption of the elimination of the need of organic solvents or other precipitating agents. As summarized by Sarangi et al. [20] the majority of the research in biorefinery facilities uses the membrane technology to produce whey protein concentrates. This fact opens a window of opportunities for the use of this technology for the production of high purity pectin derived compounds which have to be separated from the low molecular weight impurities formed during the SubW hydrolysis process [25]. The primary challenge in UF processes is fouling, where solutes accumulate on the external surface or within the membrane pores. It has been demonstrated that fouling can be attributed to the presence of pectin [26] or to complex mixtures of suspended matter (pectin and hemicellulose gel, with trapped polyphenols), as demonstrated by Saf et al. [27], when processing samples from biomass. Thus, based on this information, SubW hydrolysates from onion peel waste are expected to have a high fouling potential, making it essential to study their impact on permeate flux [25]. Additionally, strategies to mitigate fouling, such as operating in diafiltration (DF) mode, should be explored. In DF water is added to the feed to dilute it so the transport of the molecules with lower rejection rates (usually those with the lowest molecular weight) are forced to pass through the membrane [28]. The main drawback associated to the DF process is the excessive dilution of the permeate, which would demand a further concentration step [29].

The novelty of this study lies in the development of a comprehensive cascade process to fully valorise the valuable compounds present in onion peels, creating new value chains from this underutilized by-product. Unlike previous approaches, our work not only recovers flavonoids through a conventional hydroalcoholic extraction method

(optimized in prior research [30]), but also introduces an innovative second and third stage. These stages involve subjecting the solid residue, post-flavonoid extraction, to hydrothermal treatment using subcritical water followed by advanced membrane fractionation and separation techniques. The key innovation here is the use of subcritical water as the sole solvent for the recovery of high-purity pectin-derived compounds. Furthermore, the study delves into the effects of feed composition on the ultrafiltration process, operated in diafiltration mode with water as the diluent, and the application of ceramic TiO_2 membranes (50 and 5 kDa molecular weight cut-offs). Additionally, we explore novel fouling mitigation strategies to ensure sustained membrane performance.

2. Materials and methods

2.1. Chemicals and materials

Onion (*Allium cepa*) peel wastes used in this work corresponded to the cultivar Horcal and were kindly provided by "Embutidos Cardeña" (Burgos, Spain). Samples were air-dried in a room at a controlled temperature (25 °C). Subsequently, they were milled using a SM100 cutting mill (Retsch GmbH, Germany), equipped with a 1 mm sieve. The composition of the raw material used in this work was analysed according to the procedure described in Section 2.3.1.

Glucose (glu, 99.5 %), galactose (gal, 99 %), arabinose (ara, 99 %), rhamnose (rha, 99 %), xylose (xyl, 99 %) and furfural (F, 99 %) were supplied by Sigma-Aldrich (USA). 5-hydroxymethyl furfural (HMF, 97 %) was provided by Biosynth Ltd. (United Kingdom); D-galacturonic acid monohydrate (GalA, purity >97 %) was obtained from Alfa Aesar (USA). Formic acid (FA, 98 %) and acetic acid glacial (AcA, 99.7 %) were purchased from Panreac (Spain); orthophosphoric acid (76 %) and sodium hydroxide (99 %) from Cofarcas S.A. (Spain), and ammonium acetate (LiChropurTM, ≥99 %) form Merck (Germany). Phenolic compounds used in this work were provided by Extrasynthese (France), (quercetin 4'-glucoside (QC4'), quercetin 3,4'-diglucoside (QC3,4') and isorhamnetin); Sigma-Aldrich (USA) (quercetin (QC), quercetin 3-glucoside (QC3'), protocatechuic acid, p-cumaric acid, kaempferol and myricetin) and Honeywell Fluka (USA), company that supplied p-hydroxybenzoic acid. Sulphuric acid used for the sample hydrolysis was 96 % pure and was provided by Labbox (Spain), whereas that used for the HPLC mobile phase preparation was 96 % Suprapur provided by Merck (Germany). All chemicals were used without further purification.

2.2. Cascade process for the recovery of bioactive compounds

This study introduced a three-stage cascade process for the valorisation of onion peels, as illustrated in Fig. 1., that is described in the following sections.

2.2.1. First stage: flavonoids extraction process

In order to extract flavonoids present in the raw material, 60 g of onion peels were mixed with 600 mL of an ethanol/water mixture (70/30, v/v) at 37 °C for 1 h, according to the extraction procedure described by Benito-Román et al. [13]. After this treatment ethanol was removed by means a rotary evaporator (model Laborota 4001; Heidolph, Germany) at 40 °C under vacuum from the liquid extract, that was subsequently freeze dried (Labconco Inc., MO, USA) at 0.15 mbar for, at least, 48 h. The composition in terms of flavonoids of the freeze-dried extract was analysed (procedure described in Section 2.3.4), and the flavonoids-free onion peels were dried and served as raw material for the subcritical water hydrolysis process described in Section 2.2.2.

2.2.2. Second stage: subcritical water hydrolysis

The flavonoids-free onion peels, after drying, were subjected to a subcritical water hydrolysis process in the 500 mL batch reactor (maximum specifications: pressure 7 MPa; temperature, 250 °C) presented in Fig. S1 (supplementary material). In each experiment, 350 mL of water plus 15 g of flavonoids-free onion peels were used and the system was pressurized by means of N_2 to achieve the desired working pressure (5 MPa). The extraction conditions used in this work were obtained in a previous research [31], and led to maximize the extraction of pectin derived compounds. Specifically, the hydrolysis conditions selected were 115 °C for 180 min; 125 °C for 150 min and 135 °C for 90 min, that led to three different subcritical water hydrolysates (SubWH), denoted as H-115, H-125 and H-135, respectively. The composition of the liquid hydrolysates was determined following the procedure outlined in 2.3.2.

When using SubW as a reaction media, it is common to use the severity factor for the monitoring of the hydrothermal processing and fractionation of lignocellulosic biomass, in order to optimize the operational conditions, comparison with other biomass (using hydrothermal processes) and as a scale-up parameter [32,33]. In these processes, the rate of structural changes in biomass is influenced by various operational conditions, including temperature, reaction time, pressure, reaction mode, and, in some cases, the presence of auxiliary treatments. The severity factor is crucial because it combines the effects of temperature and time into a single parameter, allowing researchers to make direct comparisons of results across different studies and facilitating the transition from lab-scale experiments to industrial applications. It has been widely used by researchers studying the hydrothermal hydrolysis of several types of biomass [34,35]. It was first introduced by Overend & Chornet [36], and in its simplest way it is calculated according to Eq. (1):

$$\log R_0 = t \cdot \exp\left(\frac{T(t) - 100}{14.75}\right) \quad (1)$$

Where t is the time in min, and T is the temperature of reaction in °C, and 100 °C is the base of temperature (value selected because it is a low temperature with practically no solubilization and depolymerization of the hemicellulose). The exponential term is related to the reaction rate and 14.75 is a typical activation energy for glycosidic bond cleavage of carbohydrates, assuming a hydrothermal process and the overall conversion is first order. The severity factor expressed in Eq. (1) can be

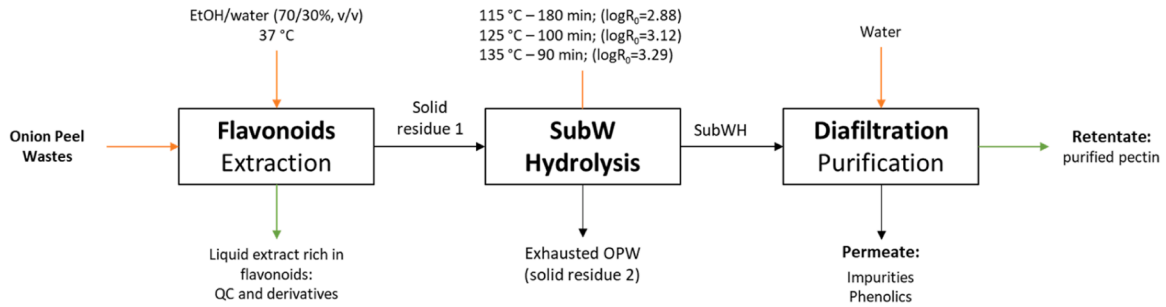

Fig. 1. Cascade process proposed in this work.

modified to include non-isothermal conditions, for instance the heating and cooling periods in which the biomass is exposed to high temperature conditions, resulting in Eq. (2), according to Wang et al. [37]:

$$\log R_0 = \log[R_{0HEATING} + R_{0ISOTHERMAL} + R_{0COOLING}] = \log\left[\int_0^{t_H} \exp\left(\frac{T(t)-100}{14.75}\right)\cdot dt + t\cdot\exp\left(\frac{T(t)-100}{14.75}\right) + \int_0^{t_C} \exp\left(\frac{T(t)-100}{14.75}\right)\cdot dt\right] \quad (2)$$

In Eq. (2), t_H (min) is the time needed to achieve the target temperature, t_C (min) is the time needed for the whole heating–cooling period, t (min) is the retention time, and T represents the treatment temperature (°C). In the present work, the heating period had a constant heating rate of of 5 °C/min and a cooling period (at a rate of 3 °C/min) needed to cool-down the reactor and open it safely. The application of the Eq. (2) lead to severity factors of 2.88±0.02, 3.12±0.01 and 3.29 ±0.01 for the hydrolysates H-115, H-125 and H-135, respectively.

Third stage: diafiltration (DF) of SubWH using different pore size ceramic membranes

The continuous feed diafiltration experiments were carried out using the experimental setup shown in Fig. S2 (supplementary material), operated in diafiltration (DF) mode. In this work, the 7-channel tubular ceramic membranes used (Filtanium™, active layer of TiO_2 supported on titania) were supplied by TAMI industries (France) and had 50 and 5 kDa molecular weight-off (MWCO). These ceramic membranes consisted of a single tubular module (254 mm length, 10 mm outside diameter) with 7 inner channels (2 mm hydraulic diameter) providing an effective filtration area of 0.0132 m^2. According to Trigueros et al. [38], these membranes have a positive charge pH below 4–4.5, being the point of zero charge at pH 4–4.5. The selection of the membranes MWCO was based on a previous study done by the authors [39], and prior results found in the literature where other authors have used the 50 kDa membrane to filtrate pectin from different natural sources, such as citrus [40] or sugar beet [41]. The selection of the 5 kDa membrane was used pursuing a total recovery of the PDC, and as other authors reported in the literature, a membrane with a MWCO in the range 1–5 kDa is needed for that purpose [42].

In each experiment, 100 mL of each subcritical water hydrolysate were placed in a stirred jacketed tank (Mervilab, Spain) and pumped to the membrane module by means of a Masterflex peristaltic pump (HV-7220–57 with a Masterflex L/S EasyLoad II head HV-77201–62). Another Masterflex peristaltic pump (HV-7521–10 with a Masterflex L/S EasyLoad II head HV-77200–50) was used to add deionised water to the feed tank. The pipes were silicone tubing (Materflex L/S 15). Two pressure gauges measured the pressure at the inlet and outlet of the membrane module. Before starting the filtration process, the feed solution was recirculated through the system for 5 min. Then, transmembrane pressure (TMP) and crossflow velocity were adjusted to a constant value of 1.05±0.05 bar and 1.5 m/s, respectively, using the pump speed controller and the valve placed downstream the membrane module, based on the results obtained in a previous work [39]. All experiments were carried out at 25 °C.

The volume in the feed tank was kept constant by fixing diafiltration water volume equal to permeate volume (V_w=V_p), by using the second peristaltic pump, defining the diafiltration volume (DV) as V_w/V_0. Permeate fluxes were measured gravimetrically (±0.001 g) until the end of the experiment. To maintain a constant volume in the feed tank, water was continuously added at the same flow rate as the permeate being withdrawn. This was achieved by adjusting the speed of the water pump to match the permeate flux, ensuring stable system operation throughout the diafiltration process. The permeate and the retentate were periodically analysed and the concentration of different SubWH compounds was represented against the DV ratio in order to evaluate the mass transfer rate, the membrane fouling, pectin derived compounds retention and purification degree. After the DF experiment, the retentate was characterized and ceramic membranes were cleaned to restore the initial water permeability. The membrane was rinsed with deionised water, then cleaned with a phosphoric acid solution (0.1 v/v%, 25 °C and TMP=0.3 bar) for 45 min, and finally with a sodium hydroxide solution (0.2 M, 50 °C and TMP=0.3 bar) for 60 min. After that, the membrane was rinsed again with deionised water and the permeate flux was measured at 25 °C under different TMP values (0.25–1.5 bar), in order to evaluate the membrane cleaning extent.

2.2.2.1. Process parameters. Experimental filtration data have been expressed as percentage variation of permeate and retentate content relative to the initial SubWH content. The retention coefficients of different SubWH compounds, R_i, were estimated as follows [27]:

$$R_i \quad (\%) = \left(1 - \frac{C_{i(p)}}{C_{i(0)}}\right)\cdot 100 \quad (3)$$

Also, an overall transmission coefficient, Tr_i, was calculates as:

$$Tr_i \quad (\%) = \frac{C_{i(p)}\cdot V_{(p)}}{C_{i(r)}\cdot V_{(r)}}\cdot 100 \quad (4)$$

where C_i (mg/L) is the compound concentration, and subscripts *p*, *r* and *0* refer to the permeate, retentate and feed solution (or initial SubWH), respectively. The selectivity, α, of ultrafiltration between pectic fraction and other SubWH compounds (j) was estimated as:

$$\alpha = \frac{Tr_j}{Tr_{PDC}} \quad (5)$$

The recovery factor towards PDC (RF_{PDC}, Eq. 6) at the end of diafiltration and the PDC fraction purity index regarding total soluble solids (PI, Eq. 7) were estimated as follows:

$$RF_{PDC} \quad (wt.\%) = \frac{C_{PDC(r)}\cdot V_r}{C_{PDC(0)}\cdot V_0}\cdot 100 \quad (6)$$

$$PI(wt.\%) = \frac{C_{PDC}}{C_{TS}}\cdot 100 \quad (7)$$

2.2.2.2. Analysis of membrane fouling. The filtration resistances have been estimated by the resistance-in-series model and Darcy's law, that enables the calculation of the total resistance to filtration r_T (m^{-1}), as a combination of the original clean membrane resistance, r_m (m^{-1}) which is the membrane intrinsic resistance to filtration determined using pure water, and the resistances deriving from the different fouling mechanisms r_f (m^{-1}) [43]. The contribution of the fouling to the total resistance, r_f/r_T, has also been calculated.

The membrane fouling mechanisms have been examined using the Hermia's model [44] modified by [45] for crossflow filtration:

$$\frac{dJ}{dt} = -k\cdot(J - J^*)\cdot J^{2-n} \quad (8)$$

where *k* is the fouling index, *J** is the critical flux that should not be exceeded in order to avoid fouling, and *n* indicates the mechanism of fouling: n=2 indicates the complete blocking model, n=1.5 the standard

blocking model, and n=0 the cake layer model. When n is fixed, the following linearized fouling models were obtained by integrating Eq. 7 from t=0 to t and from J_0 to J as the boundary conditions [38]:

$$\ln(J - J^*) = \ln \quad (J_0 - J^*) - k_c \cdot t \quad (\text{for } n = 2) \tag{9}$$

$$\frac{1}{J^{0.5}} = \frac{1}{J_0^{0.5}} + k_s \cdot t \quad (\text{for } n = 1.5) \tag{10}$$

$$\ln\left(\frac{J}{J - J^*}\right) - \frac{J^*}{J} = \left[\quad \frac{J^*}{J_0} - \ln\left(\frac{J_0 - J^*}{J_0}\right) \quad \right] + k_{cl} \cdot (J^*)^2 \cdot t \quad (\text{for } n = 0) \tag{11}$$

where k_c, k_s, k_{cl} are the fouling indexes for complete blocking (s^{-1}), standard pore-blocking ($m^{-0.5} \cdot s^{-0.5}$), and cake layer formation (m^{-2} s) models, respectively. Eqs. 9–11 have been used to fit the experimental permeate flux data. The most predominant fouling mechanism has been identified by comparing their determination coefficients, R^2 [38,46,47].

2.3. Samples characterization: analytical methods

2.3.1. OSW characterization: structural components and elemental analysis

The composition of the structural components present in onion peel wastes was determined following the protocol NREL/TP-510–42618. In brief, solid samples were subjected to acid hydrolysis and then the resulting sugar monomers were determined by HPLC, using the chromatographic system described in Section 2.3.2. Moisture, ash content and lignin (both soluble and insoluble) were also determined according to the above mentioned NREL protocol. The extractives content of the raw material was determined following the protocol described in the NREL technical report NREL/TP-510–42619.

2.3.2. Determination of structural components and degradation products

Identification and quantification of sugars and their degradation products in SubW extracts was carried out following protocol NREL/TP-510–42623. For that purpose, the HPLC system (Agilent Technologies) equipped with a Biorad Aminex HPX-87 H column (300×7.8 mm, Bio-Rad) coupled with the guard column Micro-Guards Cation H^+ (Bio-Rad), and a variable wavelength detector (VWD) and a refractive index detector (RID). 0.005 M sulfuric acid as mobile phase (0.6 mL/min) was used. The column and detectors were maintained at 40 °C and the total running time was 67 min per injection. The monomeric sugars and degradation products in the SubW extracts were first determined after centrifugation (3170 g, 15 min, 4 °C) and filtration through a 0.22 μm pore size syringe filter (Scharlab, Spain). Total sugars were determined after the acid hydrolysis of the sample according to the NREL/TP-510–42618 Analytical Procedure. The oligomeric galacturonic acid (GalAO) was determined as the difference between the total galacturonic acid (determined after the acid hydrolysis) and the free galacturonic acid, and the PDC fraction was calculated as the difference between total GalA and sugar content after acid hydrolysis and the free GalA and sugar monomers content. The following compounds, glucose (glu), galactose (gal), xylose (gyl), rhamnose (rha), arabinose (ara), other compounds such galacturonic acid (GalA), formic (FA) and acetic (AcA) acids, and sugar dehydration products, 5-hydroxymethyl furfural (HMF) and furfural (F) formed during the SubW hydrolysis process [48], were identified by HPLC, comparing the retention time with pure standards. The formation of organic acids are produced as degradation products from the monomers (which are saccharides) of biomass structural components when exposed to high temperatures, such as those in subcritical water conditions, as extensively describe Kabyemela et al. [48] in a seminal study on this topic, proposing reaction pathways and the associated kinetics. Their proposed mechanism indicates that organic acids, primarily acetic and formic, are the final degradation products (HMF or furfural), along with other reaction intermediates. More specifically, in studies involving onion peels [31] and galacturonic acid, the pectin monomer [49] under subcritical water conditions, it has been demonstrated that formic acid and furfural are the primary degradation products detected. For the specific identification of galactose and xylose, the K-ARGA and K-XYLOSE assay kits (Megazyme Ltd., Ireland) were used.

2.3.3. Pectin derived compounds molecular weight distribution

The molecular weight distribution of the samples was determined employing high-pressure size exclusion chromatography coupled to refraction index detector (HPSEC-RID). The chromatography equipment consisted of an Agilent 1260 Infinity II LC system (Agilent Technologies, CA, USA) with a guard pre-column (PL Aquagel-OH, 7.5×50 mm, 8 μm) and two columns (PL Aquagel-OH 30 and 40, 7.5×50 mm, 8 μm) from Agilent Technologies linked in series. Characterization of SubWH was performed at 35 °C. 10 μL of each sample were eluted in isocratic mode with 0.01 M ammonium acetate, at a flow rate of 0.7 mL/min. In addition, a pullulan standard set (0.342 – 400 kDa) provided by PSS Polymer Standards Service GmbH (Mainz, Germany) was used for calibration and data were analyzed with Agilent OpenLab Data Analysis 2.5 software. Using that software, the weight-average molecular weight (Mw) and polydispersity (PDI, which is calculated as ratio between weight average molecular weight and the number average molecular weight) of all the samples. All the results presented in terms of molecular weight are relative to the calibration curve obtained using the pullulan standards. Standards and samples were filtered through 0.22 μm syringe filters.

2.3.4. Phenolics identification

Each sample was characterized by High Performance Liquid Chromatography using a Diode Array Detector (HPLC-DAD, Agilent 1100, CA, USA) with a Kinetex® Biphenyl column (250×4.6 mm, particle size 5 μm, pore size 100 Å) supplied by Phenomenex (CA, USA). The mobile phase consisted of (A) ammonium acetate 5 mM with acetic acid (1 %, v/v) in water and (B) ammonium acetate 5 mM with acetic acid (1 %, v/v) in acetonitrile. A detailed description of the method is presented in Benito-Román et al. [13]. The total phenolic compounds (TPC) were calculated as the sum of the individual phenolics detected at 280 nm.

2.3.5. Particle size distribution

The droplet size distribution of the SubWH was measured using the Mastersizer 2000 (Malvern Panalytical, Malvern, United Kingdom) device. Around 5 mL of the hydrolysate was suspended in distilled water, at stirring rate around 1000 rpm. Each emulsion droplet size was measured three times and the DeBroukere mean, D[3,4] was reported.

2.3.6. ζ-potential measurement

The ζ-potential measurement of the SubWH was conducted using the Zetasizer Nano ZS apparatus (Malvern Panalytical, Malvern, United Kingdom), using the Laser Doppler Velocimetry technique, using the DTS1061 disposable folded capillary cell. Five replicates of 12 measurements were performed for each sample at 20 °C.

2.3.7. Browning degree (BD)

BD was determined by UV–Vis spectrophotometry at 420 nm [50] using a Hitachi U-2000 UV/Vis spectrophotometer.

2.3.8. pH

The pH of the samples was measured by means of a pH-meter (Crison GLP 22).

2.3.9. Density measurements

The density of the samples was measured by means of the densimeter DMA 5000 (Anton Paar, Austria). The results presented in this work show average values of 3 measurements with the calculated standard deviation.

2.3.10. Statistical analysis

All the statistical calculations were done using Statgraphics 19-X64. The significance of the differences was determined based on an analysis of the variance with the Fisher's least significant difference (LSD) procedure at p-value ≤0.05. To estimate the kinetic parameters, a simple regression model was used in Statgraphics 19-X64. In addition, experimental results were then compared to those of the model prediction through the values of the Root Mean Square Deviation (RMSD), calculated according to Eq. 12:

$$\mathrm{RMSD} = \sqrt{\frac{\sum_{i=1}^{n}\left(\mathrm{Response}_{exp} - \mathrm{Response}_{calc}\right)^2}{N}} \quad (12)$$

where N is the number of experimental data.

3. Results and discussion

3.1. Raw material composition

The composition of the raw material utilized in this study is outlined in Table 1. Onion peel wastes are a rich source of extractives (15.4 ±0.7 % of the total weight), from which 75 % were water soluble and the remaining ethanol-soluble extractives. Among the water-soluble extractives, QC4 and protocatechuic acid arose as the most abundant (75 % and 6 %, respectively), whereas QC was the ethanol extractive most abundant (70 %) of the total. The composition of the extractives suggests that an appropriate recovery strategy would involve using an ethanol/water mixture to efficiently recover the majority of extractives present in the raw material. Regarding the structural components of the raw material, cellulosic derived compounds together with pectin are present in a remarkable extent, 33.3±0.2 g/100 g of raw material and 18.9±0.3 g/100 g of raw material, respectively.

3.2. Flavonoids extraction from onion peel wastes: first stage of cascade process

Extraction with an ethanol/water mixture (70 %, v/v) yielded an extract that, after ethanol removal by evaporation and subsequent freeze-drying, accounted for 10 % of the initial raw material. This dry product was enriched in QC4' (88±2 mg/g of solid product) and QC, which accounted for the 56±2 mg/g of the solid product, followed by QC3,4' with nearly 10±1 mg/g of solid product presence in the solid product. The composition of the freeze-dried product in terms of individual phenolics is summarized in Table S1 (supplementary material). After the ethanol/water extraction process, solid residue (see Fig. 1) were subjected to the analysis for the determination of the composition, as reported in Table 1. It is shown that the ethanol/water extraction process did not affect the structural components of the onion peels, so the resulting solid was subjected to the subcritical water hydrolysis process in order to release pectin derived compounds.

Table 1
Composition of onion peel wastes (OPW) used in this work (expressed in dry basis) as received and after the extractives recovery (raw material for the SubW hydrolysis in the cascade process).

	g/100 g OPW	
Component	**OPW (as received)**	**OPW (residue 1)**
Extractives	15.4±0.7	5.7±0.7
Glucan	33.3±0.2	39.3±2.1
Galacturonan	18.9±0.3	26.6±0.9
Soluble lignin	12.5±0.4	11.5±0.2
Insoluble lignin	4.8±0.6	3.3±0.1
Protein	4.2±0.1	2.6±0.1
Galactan+Arabinan+Xylan	4.7±0.4	4.9±0.4
Ashes	7.5±0.4	7.9±0.6

3.3. Subcritical water hydrolysates (SubWH) composition: second stage of cascade process

The physical properties of the subcritical water hydrolysates obtained under three different experimental conditions is detailed in Table 2. It is important to note that the subcritical water hydrolysates were obtained under different combinations of temperature and time, consequently, the severity factor ($\log R_0$) was calculated for each extraction condition using Eq. 2. The calculated severity factors were 2.88±0.02, 3.12±0.01, and 3.29±0.01 for SubWH H-115, H-125, and H-135, respectively. These severity factors provide a basis for comparing the composition results of each hydrolysate.

Overall, no substantial differences were observed in the main physical properties of the subcritical water hydrolysates, such as pH, absorbance at 420 nm, or zeta potential. However, the particle size of the dissolved biomass noticeably decreased with increasing extraction severity, highlighting SubW's ability to promote hydrolysis reactions. Increases in the extraction severity increase the biomass hydrolysis yield (expressed as the total solids content in the subcritical water hydrolysates), but the change from 3.12 to 3.29 led to similar results. The concentration of the main species present in the subcritical water hydrolysates (pectin derived compounds, degradation products and free monosaccharides) as a function of the severity factor, is presented in Fig. 2. More specifically, in Fig. 2a the concentration of pectin derived compounds and free monosaccharides is presented. Notably, H-125 ($\log R_0$=3.12) exhibits the highest concentration of pectin derived compounds, indicating that this temperature-time relationship achieved the best extraction performance, recovering approximately 32 % of the structural pectin present in the onion peels by-product. It can be observed that their concentration of free sugar monomers (monosaccharides) increases with a rise in the severity factor. This is attributed to the heightened degree of hydrolysis of the structural pectin present in the hydrolysate. These findings indicate that elevating the severity factor from 2.88 to 3.29 in the subcritical water treatment results in an increased quantity of residual compounds or impurities in the hydrolysates.

Concerning the degradation compounds formed during the SubW hydrolysis process, compounds from hydrolysis (FH and AcH), Fig. 2(b) illustrates that their concentration increases the higher the severity factor. This observation is attributed to the extended period of hydrolysis reactions occurring at elevated severity factors. Additionally, sugar dehydration products (F and HMF) also exhibit an increasing trend with an increase in the severity factor. This findings align with the results presented by Pińkowska et al. [51], who conducted SubW extractions of sugar beet pulp pectins, also observing an increase in impurity content (formic acid, lactic acid, levulinic acid, oxalic acid, HMF, and furfural) with the increase in temperature and extraction time. These findings suggest the hydrolysis of dissolved pectin derived compounds and its conversion into degradation products, as demonstrated by Benito-Román et al. [49], who reported the degradation of GalA to form formic acid and furfural in SubW. After the analysis of the SubWH composition, it appears that H-125 is the one that provides the best results in terms of PDC concentration and presence of impurities.

Furthermore, phenolic compound analysis of subW hydrolysate still revealed concentrations ranging from 355±3 to 178±13 mg/L, with 20–50 % being flavonoids, as it is presented in Table 3. This is indicating that a remarkable concentration of phenolic compounds was still present in the biomass after the first stage in which the raw material was treated with an ethanol/water mixture to recover the flavonoids. Despite prior extraction, residual phenolic compounds persist within this by-product, tightly bound to onion peel structural components and were released only during the SubW hydrolysis process, which make them a common by-product in the pectin production process [52], which is worth separating and fractionating. Ferulic and p-coumaric acids were the primary phenolic acids found in cell walls, frequently esterified with pectins and arabinoxylans or cross-linked with other polysaccharides of the cell wall

Table 2
Physical properties of the subcritical water hydrolysates obtained under different conditions. CF indicates that the SubWH has been centrifuged.

	H-115 ($logR_0$=2.88)	H-125 ($logR_0$=3.12)	H-135 ($logR_0$=3.29)	H-125CF ($logR_0$=3.12)
Total Solids (TS, g/L)	9.0±0.8^a	11±1^b	10.6±0.1^b	9.5±0.1^a
Zeta Potential (ξ, mV)	−12.6±0.4^b	−12.8±0.6^b	−12.3±0.8^b	−11.2±0.4^a
Conductivity (mS/cm)	1.08±0.03^a	1.29±0.03^b	1.39±0.06^b	1.37±0.03^b
pH	3.47±0.05^b	3.44±0.06^b	3.57±0.04^c	3.34±0.03^a
Absorbance (420 nm)	2.18±0.01^b	2.22±0.02^b	2.23±0.03^b	1.33±0.05^a
D[3,4] (μm)	149±5^d	87±3^c	73±3^b	0.178±0.012^a

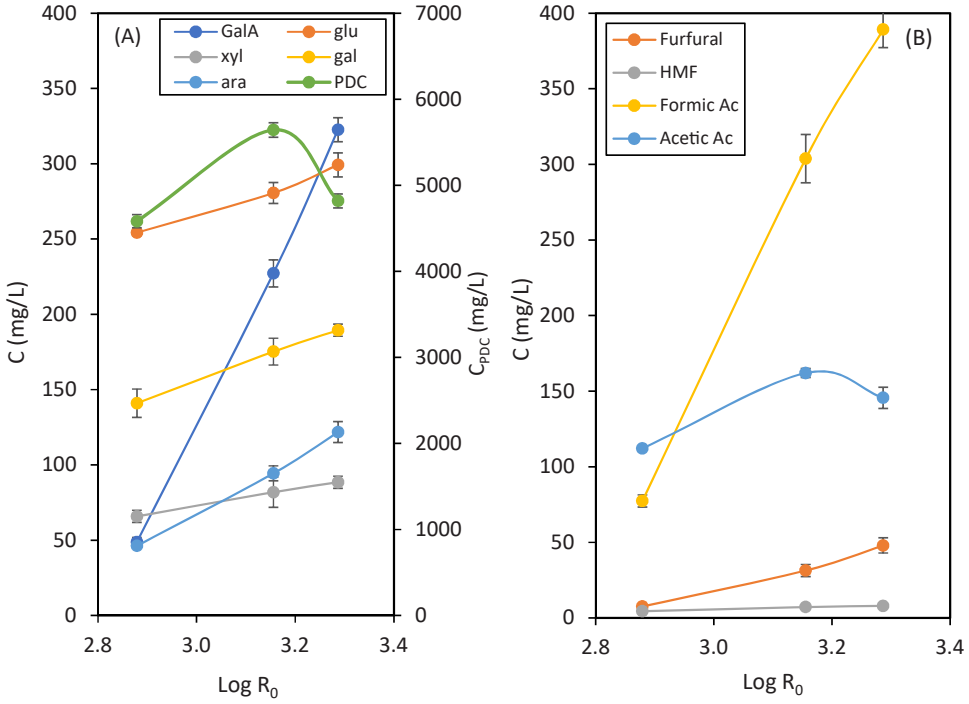

Fig. 2. Concentration of pectin derived compounds and free monosaccharides (a) and degradation products (organic acids, furfural and HMF) (b) as a function of the severity factor ($logR_0$).

Table 3
Phenolics content in the subcritical water hydrolysates obtained under different conditions. CF indicates that the SubWH has been centrifuged.

	H-115 ($logR_0$=2.88)	H-125 ($logR_0$=3.12)	H-135 ($logR_0$=3.29)	H-125CF ($logR_0$=3.12)
Total Phenolics (mg/L)	177±4^a	279±5^b	354±3^c	269±4^b
Protocatechuic A.	44.8±3.1^c	32.9±2.6^a	40.4±0.1^b	31.0±2.2^a
p-cumaric A.	nd	2.7±0.1^a	8.9±0.2^b	2.3±0.3^a
p-hydroxybenzoic A.	40.5±0.2^a	161±1^b	240±2^c	159±1^b
QC	38.8±0.2^d	30.4±0.3^c	20.5±0.4^a	29.6±0.2^b
QC4'	48.1±0.3^c	39.5±0.2^b	31.9±0.2^a	37.9±0.3^b
QC3,4'	3.8±0.1^a	9.3±0.5^c	8.3±0.1^b	9.1±0.3^c
QC3	nd	0.8±0.1^a	2.1±0.1^b	0.8±0.1^a
Kaempferol	nd	0.2±0.0^a	0.2±0.0^a	0.2±0.0^a
Myricetin	0.9±0.1^a	1.6±0.1^b	1.7±0.2^b	1.7±0.1^b
Isorhamnetin	nd	nd	0.1±0.0	nd

[53], so the hydrolysis of the cell wall may help to release them. The release of increasing amounts of flavonoids with the increase of the severity extraction conditions demonstrates the ability of SubW to induce the hydrolysis of the cell walls and release valuable bioactive compounds. In this sense the presence of cumaric acid increased by five times when working at 135 °C compared to 125 °C, and similar results were obtained for p-hydroxybenzoic acid. Regarding QC and QC4', the increase in the severity conditions decreased the extraction yield, but a clear peak was observed for the most water soluble QC3,4'. The subcritical water hydrolysis led to a flavonoids profile similar to that obtained in the conventional hydroalcoholic extraction process, in which QC4' and QC were the most abundant.

The analysis of the molecular weight distribution for each of the three subcritical water hydrolysates (SubWH) is depicted in Fig. 3. This figure demonstrates that the SubWH are complex multimodal distributions. However, it is generally observed that higher-intensity extraction conditions lead to a reduction in the molecular weight of pectin derived compounds and an increased presence of different families [54],

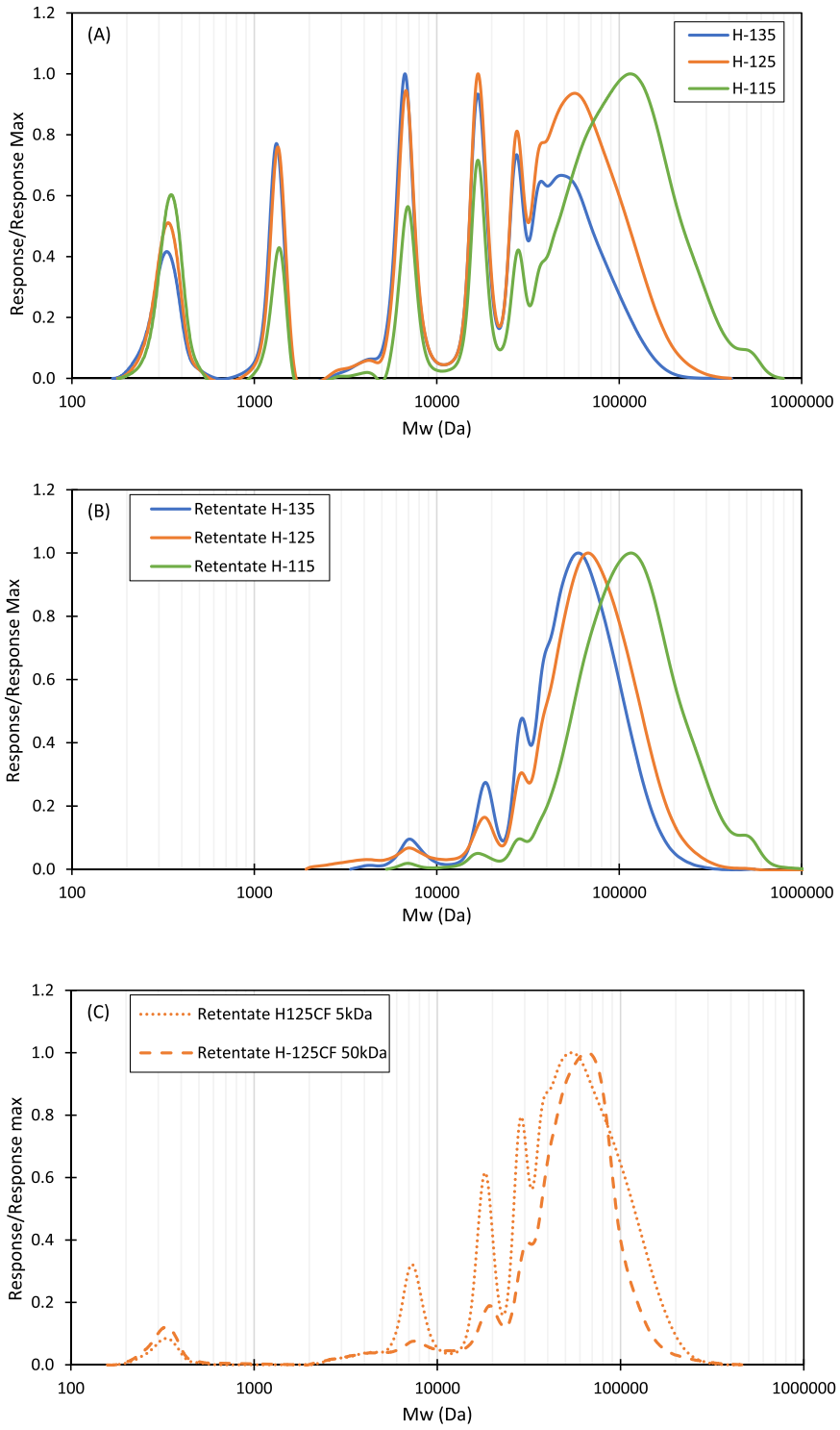

Fig. 3. GPC profiles: (A) Different SubW hydrolysates used in this work; (B) Molecular weight distribution of the retentates obtained using the 50 kDa membrane for the three hydrolysates used in this work; and (C) the retentates obtained using the 50 and 5 kDa membranes when performing the DF of the hydrolysate H-125 centrifuged (H-125CF).

including an increase in the low molecular weight families that would correspond to monomers or degradation products, as can be observed in Fig. 3. The analysis of the molecular weight distribution reveals that 28.5 % of the molecules present in the H-135 hydrolysate have a molecular weight higher than 50 kDa, whereas this share increases to 41 % for H-125 and 68 % in the H-115 hydrolysate. The multimodal distributions phenomenon is widely observed in SubW extraction processes for pectin: the presence of multimodal distributions and an augmented proportion of low molecular weight populations with increasing extraction severity [55,56]. In the hydrolysate H-115, the most abundant fraction has a weight-average molecular weight (Mw) of 126 kDa with a polydispersity of 1.41; for 125 °C, a value of 78 kDa with a polydispersity of 1.25; and finally, for 135°C, a value of 70 kDa with a polydispersity of 1.14. It is concluded that the extraction temperature significantly affects the molecular weight of pectin derived compounds, as the structure undergoes more extensive fragmentation, leading to the

production of different families of compounds. Results obtained by Klinchongkon et al. [23] in the SubW extraction of pectin from passion fruit indicate that an increase in extraction temperature leads to a decrease in molecular weight and polydispersity. Furthermore, those authors reported molecular weights in the range from 18 to 259 kDa using temperature conditions between 120 and 140 °C.

In general, it is possible to conclude that increases in the severity of the extraction conditions increases the dissolution of pectin, yet concurrently expedite their degradation, leading to a higher quantity of impurities that must be separated from the pectin derived compounds in the subsequent membrane stage. Moreover, PDC also suffer a decrease in the molecular weight.

3.4. Diafiltration process using the 50 kDa membrane: third stage of cascade process

The DF of the subcritical water hydrolysates obtained in the second stage of the cascade process, was primary investigated using a 50 kDa membrane in order to separate the most abundant PDC families present in the hydrolysates (according to the MW distribution presented in Fig. 3a) and the performance of the process analysed in terms of permeate flux, membrane fouling and composition of the retentate.

3.4.1. Permeate flux and fouling

In Fig. 4a, the results of the permeate flux profile (J) as a function of the diafiltration volume (V_w/V_o) are shown for the DF process of the various hydrolysates used as feedstock. The permeate flux of pure water through the 50 kDa membrane was 83.1±1.1 L/(h·m^2). In contrast, the initial permeate flux for the SubWH was 8.1, 8.6 and 9.4 L/(h·m^2) for H-135, H-125 and H-115, respectively, which is indicating a dramatic reduction in the permeate flux. Moritz et al. [57] noted that the extent and reversibility of fouling depend on the zeta potential of both, the membrane surface and the solution being treated. If both have opposite charge, fouling layers form rapidly, leading to a steep decline in permeate flux, as it is possible to observe in Fig. 4a, in relation to the zeta potential values reported in Table 2. Related to that, is has been observed that the decrease in the particle size of the hydrolysates increases permeate flux depletion and fouling, which may be related with the ability of the small molecules formed to enter the membrane pores and hinder permeate flux.

The fouling characterization was carried out using the resistance-in-series model. As seen in Table 4, the total filtration resistance (r_T) was a

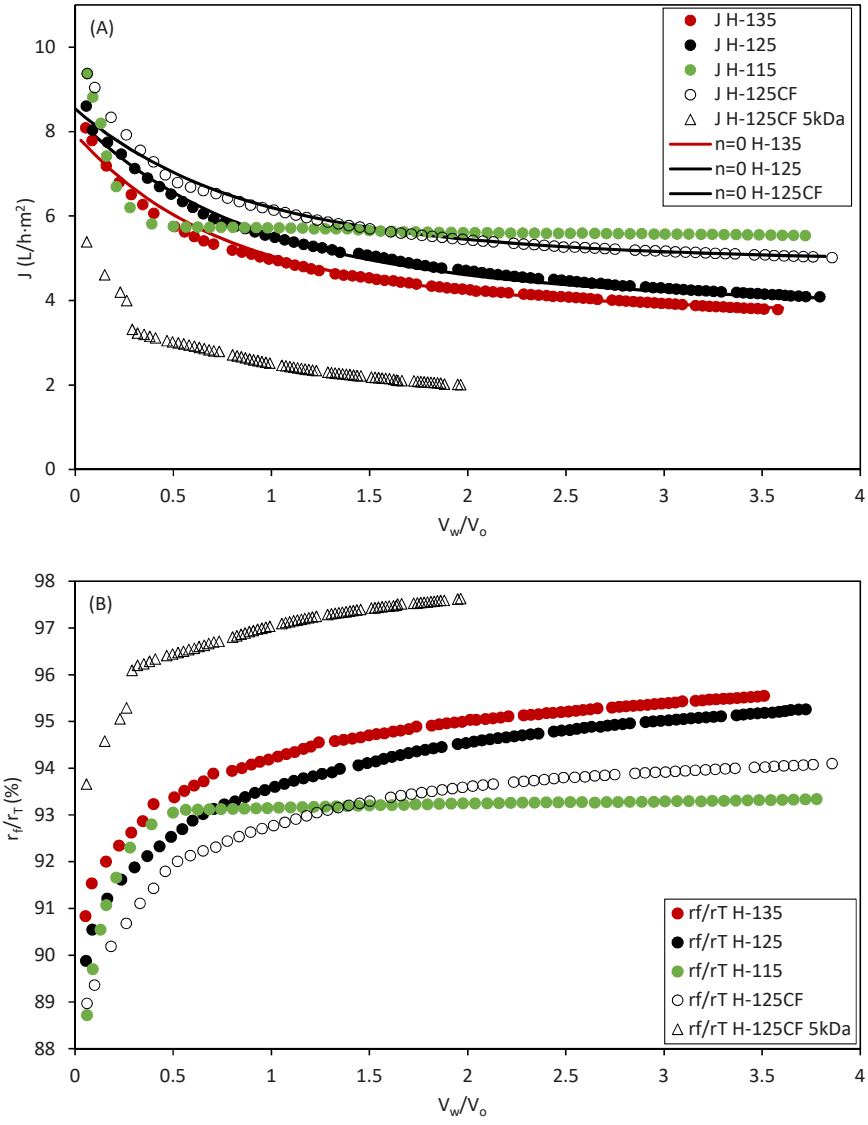

Fig. 4. Results of UF in terms of permeate flux (a) and the contribution of fouling resistance to the total filtration resistance (b) as a function of the diafiltration volume for the hydrolysates used in this study. In figure (a), the solid line represents the permeate flux calculated using the Hermia model (Eq. 11, for=0, cake layer formation). CF indicates that the SubWH has been centrifuged.

Table 4
Fouling indexes (k_c, k_s and k_{cl}) of three fouling models for the filtration of the subcritical water hydrolysates processed in this work. CF indicates that the SubWH has been centrifuged.

		H-115 ($logR_0$=2.88)	H-125 ($logR_0$=3.12)	H-135 ($logR_0$=3.29)	H-125CF ($logR_0$=2.88)	H-125CF ($logR_0$=2.88)
Membrane MWCO	kDa	50	50	50	50	5
Fouling Contribution	r_T (10^{-13}, m^{-1})	4.4–7.5	4.8–10	5.2–11	4.4–8.3	7.5–20.9
	r_m (10^{-12}, m^{-1})	4.90	4.90	4.90	4.90	4.90
	r_F (10^{-13}, m^{-1})	3.9–7.0	4.4–9.9	4.7–11	4.0–7.8	7.0–20.5
Cake Layer Formation	k_{cl} (m^{-2}·s)	$7.4{\cdot}10^{7}$	$5.8{\cdot}10^{7}$	$7.2{\cdot}10^{7}$	$6.00{\cdot}10^{7}$	$10.8{\cdot}10^{7}$
(n=0, Eq. 11)	R^2	0.927	0.995	0.992	0.997	0.989
	RMSD	0.418	0.086	0.111	0.075	0.097
Standard Blocking	k_s ($s^{-0.5}$ $m^{-0.5}$)	$4.7{\cdot}10^{-3}$	$8.5{\cdot}10^{-3}$	$8.5{\cdot}10^{-3}$	$7.3{\cdot}10^{-3}$	$1.66{\cdot}10^{-2}$
(n=1.5, Eq. 10)	R^2	0.562	0.884	0.846	0.799	0.864
	RMSD	0.820	0.439	0.457	0.504	0.396
Complete Blocking	k_c (s^{-1})	$1.97{\cdot}10^{-4}$	$1{\cdot}10^{-4}$	$1{\cdot}10^{-4}$	$1.46{\cdot}10^{-4}$	$6.1{\cdot}10^{-5}$
(n=2, Eq. 9)	R^2	0.897	0.987	0.979	0.982	0.897
	RMSD	0.371	0.313	0.326	0.353	0.374

function of the hydrolysate composition. Whereas the highest resistance was observed for the H-135 and H-125 hydrolysates and kept increasing during the DF process (Fig. 4b) the H-115 hydrolysate provided a different fouling pattern, since the ratio r_f/r_T remained constant after a fast increase after a diafiltration volume of only 0.5. The explanation for this observed phenomenon can be found in the differences in composition for the different hydrolysates considered. H-125 and H-135 had similar solid content; however, the hydrolysate obtained at the highest temperature underwent the greatest degree of hydrolysis, as it is demonstrated by the presence of more sugar monomers, degradation products and phenolics together with the decrease in the molecular weight of the PCD previously demonstrated, which could interact with the membrane hindering the permeate flux. In all cases, it is observed that the membrane resistance ($r_m = 4.90{\cdot}10^{12}$ m^{-1}) is much lower than the fouling resistance (r_f), with r_f being the resistance that contributes the most to the final value of r_T and the decrease in J during UF for these three studied hydrolysates. In Fig. 4b, it can be observed that the contribution of r_f has been very high (89–96 %), justifying the significant fouling potential of these hydrolysates. This high membrane fouling may be attributed to the combined effect of the presence of colloidal matter and the oligosaccharide fraction (mainly constituted by glucose, galactose and free GalA), both identified as compounds with high fouling capacity in UF processes [38,46]. Additionally, depending on the hydrolysates, the contribution of r_f has varied significantly, reaching a value close to 93 % in UF for H-115 but exceeding 95 % for H-125 and H-135. This indicates that differences in the composition of the hydrolysates affect the degree of membrane fouling and the permeate flux profile.

The predominant fouling mechanism of the membrane was evaluated using the Hermia model (Eq. 8). In Table 4, it is observed that for all three hydrolysates, the R^2 value is higher for the fit with the cake formation model, indicating that the formation of a cake layer on the membrane surface is the fouling mechanism that most contributes to the increase in r_F during UF. The permeate flux obtained using the Hermia model (with cake layer formation) is presented in Fig. 4a. On one hand, hydrolysates H-125 and H-135 exhibited a continuous permeate flux decline after the initial rapid permeate decline, indicating a further fouling phenomenon. Muñoz-Almagro et al. [58] pointed out to the formation a gel layer on the surface of the membrane when ultrafiltering sunflower pectin derived solutions, whereas Jin et al. [24] also reported a dramatic permeate flux decline, which was related to the complexity of the pectin extracts, that cause membrane concentration polarization and fouling. Moreover, the presence of small organic molecules may be interacting with the membrane, increasing the fouling effect; in this sense, Saf et al. [27] indicated that phenolic compounds adsorption on the membrane, together with a gel layer formation, might be responsible for membrane fouling and permeate flux decline, when ultrafiltering olive mill wastewater in a tubular ceramic multichannel membrane (MWCO 150 kDa) operated in batch mode at 48 °C. On the other hand, a slightly different pattern was observed for the H-115 hydrolysate: the J value exhibits a rapid initial drop, followed by a relatively constant trend thereafter. This behaviour accounts for the inferior R^2 value associated with fitting the experimental data to the cake layer fouling mechanism. As previously noted, SubW hydrolysates have a very complex composition with a mixture of pectin derived compounds (that can be either linear or branched) and small polar molecules, so, a combined effect of macromolecule accumulation on the membrane surface and pore blocking is possible. Specifically, it has been observed that in processes involving oligosaccharides, fouling is more severe due to these molecules' tendency to aggregate with others via hydrogen bonding [59], increasing the likelihood of cake formation on the membrane surface. Gimenes et al. [60] indicated that the shape and degree of branching of the pectin derived compounds affects its filtrability through the membrane, since the application of an external force such as transmembrane pressure can induce changes in the spatial conformation of the molecule, leading to what is known as *"coil-stretch"* deformation [61], which promotes its insertion into the membrane pores, and a reduction in the permeate flux as a consequence.

All in all, the filtration of onion peels subcritical water hydrolysates in diafiltration mode significantly reduced the fouling in comparison to the filtration in total recirculation mode [39]. The dilution effect of the feed concentration (due to the continuous addition of water in the feed tank and solute removal) during diafiltration seems to play a favourable role in limiting fouling phenomena.

3.4.2. *Impurity transport profile across the membrane*

To assess differences between compounds and among types of hydrolysates, the kinetic results of membrane transport have been expressed as the ratio of the concentration of each compound in the permeate (C_P) throughout UF to its initial concentration in the feed (C_F). Fig. 5 depicts the C_P/C_F profile as a function of the diafiltration volume (V_w/V_0) obtained during UF of the various hydrolysates used as feedstock. Fig. 5a-d demonstrate that the transport rate across the membrane for all lower molecular weight compounds, such as free monosaccharides (FMS), free galacturonic acid (GalA), total carboxylic acids (TCA), and sugar dehydration products (SDP, as the sum of HMF and furfural), has been consistently similar and independent of the chemical composition of the hydrolysate used as feedstock. In all cases, more than 90–95 % of these impurities are removed in the permeate with diafiltration volumes between 3 and 3.5, establishing this as the minimum dilution factor for the purification of this type of hydrolysate.

Table S3 compiles the results of the total transmission (Tr, Eq. 4) achieved for these three hydrolysates at the end of UF using a diafiltration volume of only 3.5. It is important to note that the final transmission of free monosaccharides, free galacturonic acid, sugar dehydration products and carboxylic acids with a 50 kDa membrane has

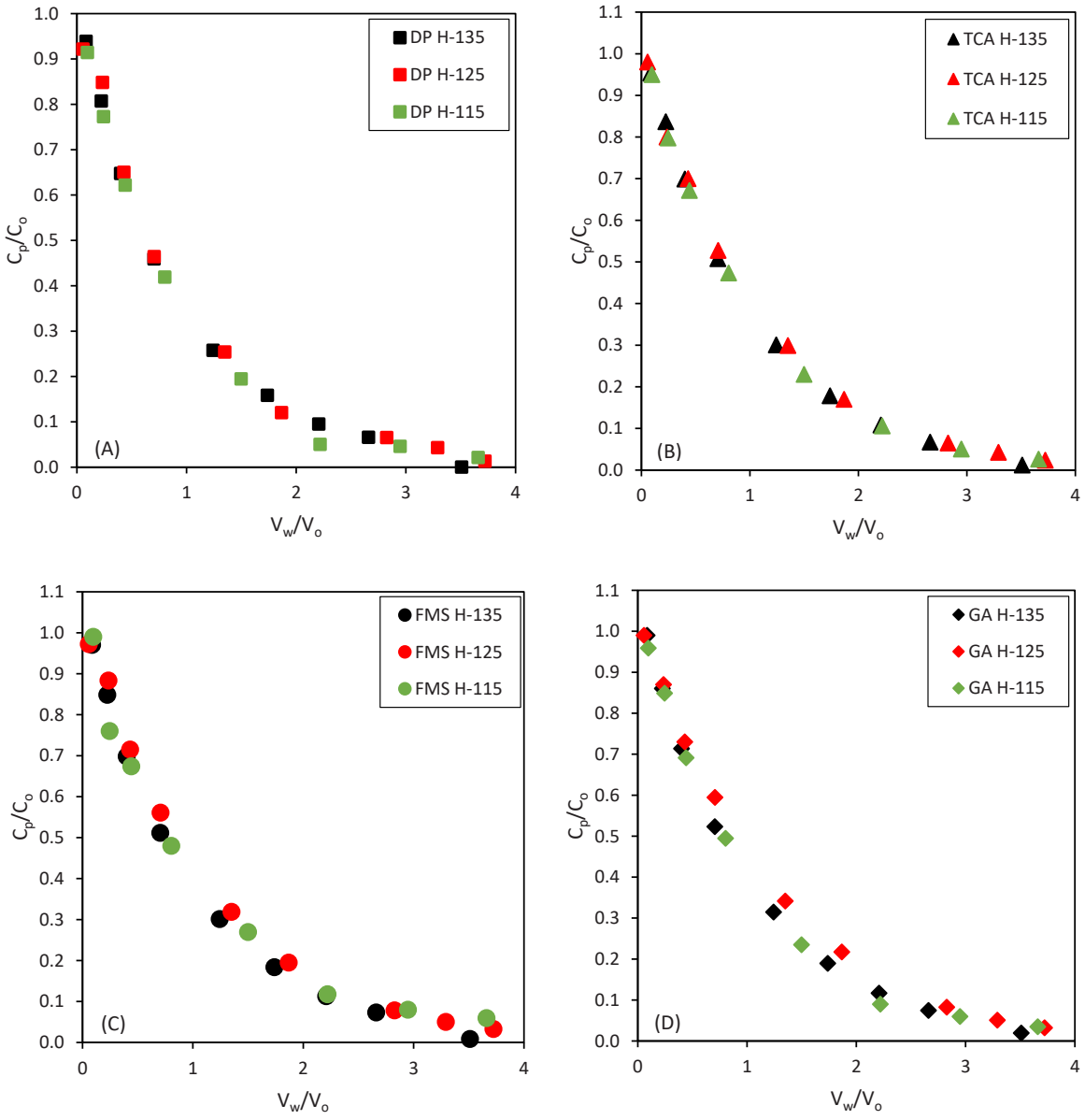

Fig. 5. Results of the variation in permeate concentration (CP/C_0) as a function of dilution factor:(a), sugar dehydration products (DP, sum of HMF and F); (b), total carboxylic acids (TCA); (c), Free monosaccharides (FMS) and (d), free galacturonic acid (GA). UF conditions: 50 kDa membrane, 25 °C, TMP= 1 bar, 17.5 L/min feed flow.

been significantly above 97 %, and the values are nearly identical for all three hydrolysates studied. Moreover, most of these low molecular weight compounds were not detected in the analyses conducted on the final retentates obtained after UF, which also indicates a high selectivity of the membrane versus pectin derived compounds, as indicates the values of α presented in Table S3.

With these results, it is possible to conclude that UF in diafiltration mode at 25 °C with a 50 kDa membrane, TMP = 1 bar, and diafiltration volumes of around 3.5 provide retentates with minimal presence of low molecular weight impurities typically found in SubWH obtained from onion peels waste. No phenolic compounds were detected in the retentate, as they have been fully transferred to the permeate fraction. Other authors have reported the transference of phenolics (considered as impurities in pectin products) but in lower extents. For instance Cho et al. [52] reported that polyphenols in pectin solution decreased about 34 % at two volumes of diafiltration, and finally decreased about 57 % at six volumes of diafiltration, using a regenerated cellulose membrane, whereas Vladisavljević et al. [62] reported that the content of anthocyanin was reduced by 65 % when the red raspberry juice was UF through ceramic membranes with a MWCO of 30 and 50 kDa. These authors indicated that anthocyanins rejection mechanism is not only dependent on the molecular size exclusion but also on the interaction of these molecules with the membrane surface, and it is likely that those molecules have binding affinity to the inorganic oxide particles of the ceramic membranes, being preferably retained as a consequence, and decreasing the permeate flux.

All in all, the permeate comprises a mixture of dehydration products, monomeric sugars, organic acids, phenolics, and low molecular weight PDCs, necessitating a second separation stage to isolate the remaining high-value components present in that stream, such as the phenolic compounds. However, colloidal matter and most coloured compounds have not been removed in the permeate. This is evident as the absorbance at 420 nm measured in these process streams was 1.43±0.02, 1.57±0.03, and 1.56±0.02 for H-115, H-125, and H-135, respectively. Although there is a significant decrease compared to the value in the subcritical water hydrolysate used as a feed in the membrane process

(see Table S1, supplementary material), it indicates a necessity for including a pre-treatment stage before diafiltration to remove them beforehand. This pre-treatment is crucial to enhance the purification degree of the pectin-derived compounds, a topic that will be further discussed in Section 3.5.

3.4.3. Evaluation of the PDC composition in the final retentate

The results of the retention factor (Eq. 6) of the PDC in the final retentate obtained after UF of the different hydrolysates with a dilution factor of 3.5 resulted to be 82±1 % for the hydrolysate H-115, but it was significantly lower 58±1 % and 56±1 % for the hydrolysates H-125 and H-135, respectively. This outcome indicates that a 50 kDa membrane has an excessively large pore diameter for the characteristics of the pectin derived compounds fraction present in the hydrolysate H-115, which in the feed around 70 % of the total PDC had a molecular weight higher than 50 kDa (based on the pullulan calibration curve used in this work). The retentate after the DF of the H-115 hydrolysate had a mayor fraction (92 % of the total) that had a weight average molecular weight (Mw) of 135 kDa, with two minor fractions. With regards the H-135 and H-125, the final retentates had a similar molecular weight distribution, with a major fraction (83 % and 89 %, respectively) with Mw of 74 and 87 kDa, respectively. The degree of hydrolysis observed was higher for the H-135 hydrolysate, as it can be observed in Fig. 3b, in which minor fractions with Mw of 28, 18 and 7 kDa remained in the retentate. A closer analysis of the molecular weight distribution revealed that the fraction of PDC with higher MW than 50 kDa was 61 % in H-135 and 69 % in H-125, indication that some of the PDC in the retentate still have molecular weight lower than the membrane MWCO. On the other hand, in Fig. 3b, it can be observed that, for all hydrolysates, compounds with a molecular weight lower than 4 kDa have not been detected. Considering these results, it is necessary to decrease the membrane pore diameter to a value lower than 7 kDa and higher than 4 kDa to achieve an increase in the retention efficiency of the PDC fraction from the hydrolysate H-125, established as optimal in the extraction stage.

The purity of pectin-derived compounds isolated from the permeate increases as the intensity of the hydrolysis process decreases. For instance, the highest purity achieved (calculated using Eq. 7) was 74.6 ±0.9 % for H-115, which decreased to 68.3±1.3 % for H-125 and even lower to 64.1±1.1 % for H-135. This trend can be attributed to the fact that higher intensity conditions during hydrolysis promote the breakdown of other structural components with molecular weights similar to those of pectin-derived compounds. These components cannot be separated during the diafiltration process. Indeed, the purity of the extracts prior to the membrane process was 29.0 %, 34.7 %, and 38.0 % for H-135, H-125, and H-115, respectively. This suggests that the DF process enhances the purity of the products derived from pectin, making it an effective method for removing impurities, allowing the fractionation of the PDC. As can be observed by comparing Fig. 3a and Fig. 3b the fraction below 20 kDa was almost completely removed from the hydrolysates being moved to the permeate. A further processing of the permeate would lead to the recovery of this PDC lower molecular weight.

By analyzing the quantity of pectin-derived compounds present in the retentate, it becomes possible to calculate the overall mass balance for these compounds, considering the raw material used in the cascade process. This mass balance enables the determination of the overall recovery efficiency for the process in terms of pectin-derived compound recovery. The results indicate that subcritical water hydrolysis at 125 °C provided the highest extraction yield, achieving 24 % recovery. According to these results, in Section 3.5 the purification using a 5 kDa membrane is studied, in order to retain the low molecular weight families lost using the 50 kDa so the overall recovery yield of PDC is increased.

3.5. Improvements in the DF process of hydrolysate H-125

The purification study conducted so far has demonstrated excellent results in terms of impurities removal. Fouling limits the permeate flux, so in order to enhance the permeate flux, a pre-treatment phase was incorporated to eliminate fouling substances. Subsequently, the pre-treated hydrolysate was subjected to diafiltration using a 5 kDa MWCO membrane, aiming to retain the low molecular weight fractions in the permeate.

3.5.1. Pre-treatment of the hydrolysate H-125 before diafiltration

The hydrolysate H-125 exhibit dark-colored colloidal matter, which, as presented in Section 3.4.2, was not possible to remove after diafiltration using the 50 kDa membrane, and was primarily responsible for the permeate flux depletion and fouling. To improve the long-term operability of the membranes and reduce fouling, several strategies can be employed [63]: besides a careful selection of the membrane MWCO and selection of the optimal operating conditions (that have already been done in this work), it is possible to pre-treat the feed solution remove large particles or impurities that contribute to fouling. Therefore, in order to remove it, the hydrolysate was centrifuged (10 min, 3170 g). This pretreatment has been successfully used in the past by Turano et al. [64], who achieved an organic matter reduction of about 90 %, as well as an increase of the permeate flux by 100 %, when using olive mill wastewater as raw material. Table 2 and Table S2 shows the characterization of the hydrolysate after removing the colloidal matter (named H-125CF), and its comparison with the initial hydrolysate (H-125), and Fig. 6 shows the particle size distribution of both hydrolysates. It is noteworthy that the value for the absorbance at 420 nm decreased notably after the centrifugation pre-treatment (down to 1.33), which meant a reduction of 60 % in the color of the hydrolysate. Regarding the solids content of the hydrolysate, it was reduced after centrifugation by 14 %, and also a small decrease in the concentration of PDC was detected, around 7 %. In summary the centrifugation step reduced the solids dissolved with a small reduction in the PDC content.

The removal of the colloidal matter increased the permeate flux (from 4 L/(h·m^2) to 5 L/(h·m^2), achieved after a DV equal to 3.5, and reduced fouling, with a contribution to the total resistance from 89 % to 94 % in comparison to 90–95 % achieved with the non-centrifuged H-125 hydrolysate (Figs. 4a and 4b). These results are in agreement with the results presented by Singh et al. [65], who remarked the significant effect of colloidal matter and lignin content on membrane fouling. According to the results presented in Table S3, it can be concluded that UF in diafiltration mode at 25 °C with a 50 kDa membrane and DV of 3.5 shows slight changes when removing colloidal matter from the feed in terms of transmission and selectivity for the main impurities considered in this work, but increased the transmission of PDC from 41.8 % to

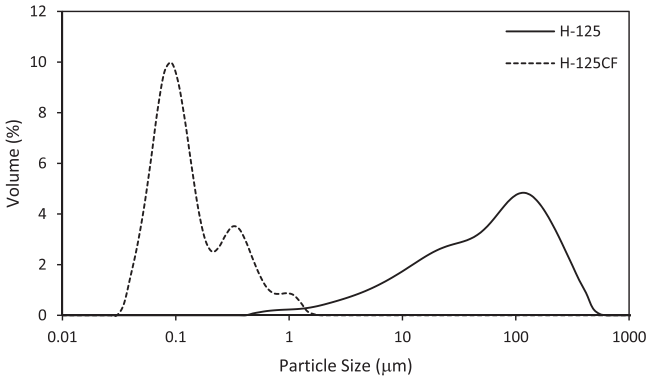

Fig. 6. Particle size distribution of the subcritical water hydrolysate obtained at 125 °C before (H-125) and after centrifugation (H-125CF).

46.3 %. The combined effect of the diafiltration and colloidal removal lead to a higher transmission of the compounds through the membrane as the concentration polarization effect is mitigated [28]. Subsequently, since the centrifugation stage did not affect the composition of the hydrolysate, it was processed using a lower MWCO membrane (5 kDa), in order to retentate all the PDC families present in the hydrolysate. When using this membrane, an important reduction in the permeate flux was observed (Fig. 4a) together with an increase of the filtration resistance (Fig. 4b) in comparison with the results obtained with the same hydrolysate and the 50 kDa membrane, after a DV of 2. The fouling analysis reported in Table 4, allowed to conclude the high fouling rate observed when processing the hydrolysate H-125CF observed when using the 5 kDa membrane.

3.5.2. Transport profile of impurities through 50 kDa and 5 kDa membranes

To assess the differences between compounds and the use of membranes with different pore sizes (50 and 5 kDa) for the hydrolysate H-125CF, the kinetic results of membrane transport have been expressed as the ratio of the concentration in the permeate of each compound (C_P) during UF to its initial concentration in the feed (C_F). Fig. S3 depicts the C_P/C_F profile as a function of the DV obtained during UF using different membrane pore sizes. As can be observed, the transport rate through the membrane is similar for all compounds with lower molecular weight, such as free monosaccharides (FMS), free galacturonic acid (GalA), total carboxylic acids (TCA) and sugar dehydration products (furfural and HMF). For degradation compounds and carboxylic acids, the transport rate has proven to be independent of the chemical composition of the hydrolysate with colloidal matter, regardless of the use of 50 kDa or 5 kDa membranes. For the 50 kDa membrane, more than 90–95 % of these impurities are removed in the permeate, reaching DV between 3 and 3.5, establishing itself as the minimum dilution factor for purification. In contrast, for the 5 kDa membrane, almost 90 % of impurities have been removed in the permeate at a DV of 2, therefore, a higher dilution factor will be needed to achieve a higher impurities removal.

Table S3 summarizes the results of the total transmission (Tr, Eq. 4) achieved for centrifuged H-125 using 50 kDa and 5 kDa membranes. The final transmission of impurities with a 50 kDa membrane has been high, exceeding 97 % using a DV of 3.5. In the case of using a 5 kDa membrane, values higher than 90 % are only achieved for the TCA and the sugar dehydration products, and a DV of 2. The transmission of the PDC was more than 2 times lower when using the 5 kDa membrane in comparison with the 50 kDa one. Related to these results, it was observed an increase in the selectivity (calculated using Eq. 5) when the 5 kDa membrane is used.

3.5.3. Evaluation of the PDC composition in the final retentate

The results of the retention factor (Eq. 6) of the PDC in the final retentate obtained after UF with 50 kDa and 5 kDa membranes with dilution factors of 3.5 and 2, respectively, are presented from resulted to be 54±1 % for UF with a 50 kDa membrane and 85±1 % for UF with a 5 kDa membrane. This outcome indicates that with a 50 kDa membrane, the PDC fraction with a molecular size less than 50 kDa passes through the membrane. In contrast, with the 5 kDa membrane, the entire PDC fraction with molecular sizes greater than 4 kDa is retained. The difference between the retained percentage of centrifuged and non-centrifuged feed is due to the removal of colloidal oligosaccharides through centrifugation, confirmed by the molecular weight distribution analysis shown in Fig. 3c. With these results, it can be concluded that UF in diafiltration mode at 25 °C requires a 5 kDa membrane to increase the retention efficiency of the PDC fraction to a value exceeding 80 % for the H-125CF hydrolysate established as optimal in the extraction stage, to obtain PDC in a concentration of approximately 4.6 g/L. It is possible to use a lower membrane size for the purification of the hydrolysate, increasing the retention of the PDC as the transmission of the impurities remains at high rates. This fact allows, despite the higher fouling observed for the 5 kDa membrane, to have a retentate with high purity after a diafiltration volume of only 2, in which all the PDC present in the initial hydrolysate are present. Then, this purified retentate can be further fractionated using different MWCO membranes not operated in DF mode, which avoids the dilution effect concomitant of the DF processes.

The purity of pectin-derived compounds isolated from the permeate increases after the centrifugation process. The pectin isolated from the retentate obtained using the 50 kDa membrane had a purity of 80.5 ±0.8 % whereas the obtained from the 5 kDa DF process was 77.6 ±1.2 %, both significantly higher than the obtained from the H-125 hydrolysate without centrifugation (which was 68.3±1.3 %), which indicates the convenience of centrifuging the extract prior to the DF process, since the purity is increased and the filtration process improved.

4. Conclusions

The cascade process presented in this work allowed to obtain valuable fractions of phenolic compounds (mainly quercetin and derivatives) and pectin derived compounds with high purity from agricultural wastes by coupling water-based technologies, such as subcritical water hydrolysis (that yields complex mixtures) and diafiltration to obtain high purity streams. Fouling is the main limitation of the filtration process, that can be mitigated by the inclusion of a pre-treatment stage that removes colour and colloidal matter. By doing this, a high purity permeate can be obtained in which most of the PDC fractions are retained using a 5 kDa membrane. This purified retentate can be further fractionated using a different MWCO membranes.

Funding

This work was supported by the Agencia Estatal de Investigación (AEI) though projects PID2020–116716RJ-I00/ AEI / 10.13039/501100011033, PID2019–104950RB-I00 / AEI / 10.13039/501100011033 and TED2021–129311B-I00.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.jece.2024.114845.

Data availability

Data will be made available on request.

References

[1] N.A. Sagar, Y. Kumar, R. Singh, C. Nickhil, D. Kumar, P. Sharma, H. Om Pandey, S. Bhoj, A. Tarafdar, Bioresour. Technol. 362 (August) (2022) 127870, https://doi.org/10.1016/j.biortech.2022.127870.

[2] A. Bains, K. Sridhar, B.N. Singh, R.C. Kuhad, P. Chawla, M. Sharma, Chemosphere 343 (August) (2023) 140178, https://doi.org/10.1016/j.chemosphere.2023.140178.

[3] EUROSTAT, Fruit and vegetable production in 2022. Fruit and Vegetable Production in 2022. Available at: https://ec.europa.eu/eurostat/web/products-eurostat-news/w/ddn-20240301-1#:~:text=For example%2C Italy accounted for,%25) and Portugal (9%25).&text=The Netherlands was the leading,%25) and France (12%25.

[4] M. Kumar, M.D. Barbhai, M. Hasan, S. Punia, S. Dhumal, Radha, N. Rais, D. Chandran, R. Pandiselvam, A. Kothakota, M. Tomar, V. Satankar, M. Senapathy, T. Anitha, A. Dey, A.A.S. Sayed, F.M. Gadallah, R. Amarowicz, M. Mekhemar, Biomed. Pharmacother. 146 (2022), https://doi.org/10.1016/j.biopha.2021.112498.

[5] A. Vojvodić, D. Komes, I. Vovk, A. Belščak-Cvitanović, A. Bušić, Food Res. Int. 89 (2016) 565–573, https://doi.org/10.1016/j.foodres.2016.07.023.
[6] W.J. Li, Z.G. Fan, Y.Y. Wu, Z.G. Jiang, R.C. Shi, J. Sci. Food Agric. 99 (12) (2019) 5283–5292, https://doi.org/10.1002/jsfa.9729.
[7] F. Dranca, M. Oroian, Food Res. Int. 113 (February) (2018) 327–350, https://doi.org/10.1016/j.foodres.2018.06.065.
[8] I.G. Osojnik Črnivec, M. Skrt, D. Šeremet, M. Sterniša, D. Farčnik, E. Štrumbelj, A. Poljanšek, N. Cebin, L. Pogačnik, S. Smole Možina, M. Humar, D. Komes, N. Poklar Ulrih, Waste Manag 126 (2021) 476–486, https://doi.org/10.1016/j.wasman.2021.03.033.
[9] K.A. Lee, K.T. Kim, H.J. Kim, M.S. Chung, P.S. Chang, H. Park, H.D. Pai, Food Sci. Biotechnol. 23 (2) (2014) 615–621, https://doi.org/10.1007/s10068-014-0084-6.
[10] E.Y. Jin, S. Lim, S. oh Kim, Y.S. Park, J.K. Jang, M.S. Chung, H. Park, K.S. Shim, Y. J. Choi, Food Sci. Biotechnol. 20 (6) (2011) 1727–1733, https://doi.org/10.1007/s10068-011-0238-8.
[11] T. Piechowiak, K. Grzelak-Błaszczyk, R. Bonikowski, M. Balawejder, LWT - Food Sci. Technol. 117 (2020) 108614.
[12] F. Salak, S. Daneshvar, J. Abedi, K. Furukawa, Fuel Process. Technol. 112 (2013) 86–92, https://doi.org/10.1016/j.fuproc.2013.03.001.
[13] Ó. Benito-Román, B. Blanco, M.T. Sanz, S. Beltrán, Antioxidants 9 (12) (2020) 1–20, https://doi.org/10.3390/antiox9121233.
[14] N. Babbar, S. Baldassarre, M. Maesen, B. Prandi, W. Dejonghe, S. Sforza, K. Elst, Carbohydr. Polym. 146 (2016) 245–252, https://doi.org/10.1016/j.carbpol.2016.03.011.
[15] Ó. Benito-Román, P. Alonso-Riaño, E. Díaz De Cerio, M.T. Sanz, S. Beltrán, J. Environ. Chem. Eng. 10 (3) (2022) 107439, https://doi.org/10.1016/j.jece.2022.107439.
[16] S.F. Ahmed, M. Kabir, A. Mehjabin, F.T.Z. Oishi, S. Ahmed, S. Mannan, M. Mofijur, F. Almomani, I.A. Badruddin, S. Kamangar, Energy Rep. 10 (March) (2023) 3073–3091, https://doi.org/10.1016/j.egyr.2023.09.137.
[17] R. Melgosa, M.T. Sanz, S. Beltrán, J. Supercrit. Fluids 169 (2021), https://doi.org/10.1016/j.supflu.2020.105121.
[18] V. Venugopal, A. Sasidharan, T. Rustad, J. Agric. Food Chem. 71 (46) (2023) 17494–17509, https://doi.org/10.1021/acs.jafc.3c03126.
[19] I.M. Bowling, M. Ponce-ortega, M.M. El-halwagi, (2011) 6276–6286.
[20] P.K. Sarangi, A.K. Singh, S. Sonkar, K.P. Shadangi, R.K. Srivastava, V.K. Gupta, J. Parikh, U.K. Sahoo, M. Govarthanan, Ind. Crops Prod. 205 (July) (2023) 117488, https://doi.org/10.1016/j.indcrop.2023.117488.
[21] J.M. Prado, D. Lachos-Perez, T. Forster-Carneiro, M.A. Rostagno, Food Bioprod. Process. 98 (2016) 95–123, https://doi.org/10.1016/j.fbp.2015.11.004.
[22] B. Gómez, B. Gullón, C. Remoroza, H.A. Schols, J.C. Parajó, J.L. Alonso, J. Agric. Food Chem. 62 (40) (2014) 9769–9782, https://doi.org/10.1021/jf503475b.
[23] K. Klinchongkon, P. Khuwijitjaru, S. Adachi, Biosci. Biotechnol. Biochem. 81 (4) (2017) 712–717, https://doi.org/10.1080/09168451.2016.1277941.
[24] L. Jin, F. Zuo, Y. Gao, S. Sui, D. Zhang, J. Food Eng. 335 (March) (2022) 111158, https://doi.org/10.1016/j.jfoodeng.2022.111158.
[25] S. Yammine, R. Rabagliato, X. Vitrac, M.M. Peuchot, R. Ghidossi, Oeno One 53 (3) (2019) 487–497, https://doi.org/10.20870/oeno-one.2019.53.3.2343.
[26] A. Echavarria, J. Pagán, A. Ibarz, J. Food Eng. 102 (4) (2011) 334–339, https://doi.org/10.1016/j.jfoodeng.2010.09.009.
[27] C. Saf, M. Villain-Gambier, M. Belaqziz, I. Ziegler-Devin, D. Trebouet, N. Ouazzani, Process Saf. Environ. Prot. 164 (June) (2022) 119–128, https://doi.org/10.1016/j.psep.2022.06.010.
[28] S. Saidi, A. Deratani, M.P. Belleville, R.Ben Amar, Food Res. Int. 65 (PC) (2014) 453–461, https://doi.org/10.1016/j.foodres.2014.04.026.
[29] M. Yazdanshenas, A.R. Tabatabaeenezhad, R. Roostaazad, A.B. Khoshfetrat, Sep. Purif. Technol. 47 (1–2) (2005) 52–57, https://doi.org/10.1016/j.seppur.2005.06.004.
[30] Ó. Benito-Román, B. Blanco, M.T. Sanz, S. Beltrán, Food Bioprod. Process. 130 (2021) 92–105, https://doi.org/10.1016/j.fbp.2021.09.005.
[31] Ó. Benito-Román, R. Melgosa, A.E. Illera, M.T. Sanz, S. Beltr, Food Hydrocoll. 153 (Augurst) (2024) 109957, https://doi.org/10.1016/j.foodhyd.2024.109957.
[32] H.A. Ruiz, M. Galbe, G. Garrote, D.M. Ramirez-Gutierrez, E. Ximenes, S.N. Sun, D. Lachos-Perez, R.M. Rodríguez-Jasso, R.C. Sun, B. Yang, M.R. Ladisch, Bioresour. Technol. 342 (August 2021) (2021), https://doi.org/10.1016/j.biortech.2021.125961.
[33] A. Singh, M. Tsai, C. Chen, R. Rani, A. Kumar, V. Tambat, C. Dong, Bioresour. Technol. 367 (September 2022) (2023) 128271, https://doi.org/10.1016/j.biortech.2022.128271.
[34] M. Nikkhah, D. Bishnu, Biomass-.-. Convers. Biorefinery (2024) 21229–21243, https://doi.org/10.1007/s13399-023-04383-7.
[35] Ó. Benito-Román, E. Alonso, M.J. Cocero, J. Supercrit. Fluids 73 (2012) 120–125, https://doi.org/10.1016/j.supflu.2012.09.014.
[36] R.P. Overend, E. Chornet, Philos. Trans. R. Soc. Lond. Ser. A, Math. Phys. Sci. 321 (1561) (1987) 523–536, https://doi.org/10.1098/rsta.1987.0029.
[37] Z.W. Wang, M.Q. Zhu, M.F. Li, J.Q. Wang, Q. Wei, R.C. Sun, Biotechnol. Biofuels 9 (1) (2016) 1–16, https://doi.org/10.1186/s13068-016-0552-8.
[38] E. Trigueros, M.T. Sanz, S. Beltrán, M.O. Ruiz, J. Memb. Sci. 660 (July) (2022), https://doi.org/10.1016/j.memsci.2022.120822.
[39] M.O. Ruiz, Benito-Román, S. Beltrán, M.T. Sanz, J. Memb. Sci. 708 (June) (2024), https://doi.org/10.1016/j.memsci.2024.123054.
[40] Y. Neggaz, M.L. Vargas, A.O. Dris, F. Riera, R. Alvarez, Sep. Purif. Technol. 54 (1) (2007) 18–27, https://doi.org/10.1016/j.seppur.2006.08.017.
[41] M. Fernández-Delgado, E. del Amo-Mateos, M. Coca, J.C. López-Linares, M. T. García-Cubero, S. Lucas, Ind. Crops Prod. 194 (January) (2023), https://doi.org/10.1016/j.indcrop.2023.116304.
[42] M. Ramos-Andrés, B. Aguilera-Torre, J. García-Serna, J. Clean. Prod. 321 (2021), https://doi.org/10.1016/j.jclepro.2021.128923.
[43] G. Di Bella, D. Di Trapani, Membr. (Basel) 9 (2) (2019), https://doi.org/10.3390/membranes9020024.
[44] J. Hermia, Inst. Chem. Eng. Trans. 60 (3) (1982) 183–187.
[45] R.W. Field, D. Wu, J.A. Howell, B.B. Gupta, J. Memb. Sci. 100 (3) (1995) 259–272, https://doi.org/10.1016/0376-7388(94)00265-Z.
[46] T. Qi, X. Chen, W. Shi, T. Wang, M. Qiu, X. Da, J. Wen, Y. Fan, Sep. Purif. Technol. 278 (August 2021) (2022) 119589, https://doi.org/10.1016/j.seppur.2021.119589.
[47] M.C. Vincent Vela, S. Álvarez Blanco, J. Lora García, E. Bergantiños Rodríguez, Chem. Eng. J. 149 (1–3) (2009) 232–241, https://doi.org/10.1016/j.cej.2008.10.027.
[48] B.M. Kabyemela, T. Adschiri, R.M. Malaluan, K. Arai, Ind. Eng. Chem. Res. 38 (8) (1999) 2888–2895, https://doi.org/10.1021/ie9806390.
[49] Ó. Benito-Román, M.T. Sanz, S. Beltrán, J. Supercrit. Fluids 206 (April) (2024) 106155, https://doi.org/10.1016/j.supflu.2023.106155.
[50] S.I.F.S. Martins, W.M.F. Jongen, M.A.J.S. Van Boekel, Opera N. 73 (6) (2008) 36, https://doi.org/10.2307/3717028.
[51] H. Pińkowska, P. Wolak, M. Krzywonos, A. Złocińska, Waste Biomass-.-. Valoriz. 12 (8) (2021) 4587–4598, https://doi.org/10.1007/s12649-020-01317-6.
[52] C.W. Cho, D.Y. Lee, C.W. Kim, Carbohydr. Polym. 54 (1) (2003) 21–26, https://doi.org/10.1016/S0144-8617(03)00133-4.
[53] M. Naczk, F. Shahidi, J. Pharm. Biomed. Anal. 41 (5) (2006) 1523–1542, https://doi.org/10.1016/j.jpba.2006.04.002.
[54] E. Rincon, E. Espinosa, M.T. García-domínguez, A.M. Balu, F. Vilaplana, L. Serrano, A. Jim, Carbohydr. P 272 (July) (2021) 118477, https://doi.org/10.1016/j.carbpol.2021.118477.
[55] I. Pereira Dias, S. Fernanda, D. Estuardo, L. Fetzer, M. Lúcio, J. Léa, M. Silveira, Int. J. Biol. Macromol. 146 (2020) 431–443, https://doi.org/10.1016/j.ijbiomac.2019.12.261.
[56] S. Pedraza-Guevara, R.F. do Nascimento, M.H.G. Canteri, N. Muñoz-almagro, M. Villamiel, M.T. Fernández-Ponce, L. Casas, C. Mantell, E.J. Martinez de la Ossa, E. Ibañez, J. Supercrit. Fluids 171 (ember 2020) (2021) 105133, https://doi.org/10.1016/j.supflu.2020.105133.
[57] T. Moritz, S. Benfer, P. Árki, G. Tomandl, Sep. Purif. Technol. 25 (1–3) (2001) 501–508, https://doi.org/10.1016/S1383-5866(01)00080-6.
[58] N. Muñoz-Almagro, M. Prodanov, P.J. Wilde, M. Villamiel, A. Montilla, Food Chem. 318 (January) (2020) 126476, https://doi.org/10.1016/j.foodchem.2020.126476.
[59] R. Jiraratananon, D. Uttapap, C. Tangamornsuksun, J. Memb. Sci. 129 (1) (1997) 135–143, https://doi.org/10.1016/S0376-7388(97)00046-X.
[60] M.L. Gimenes, V.R. Silva, F. Hamerski, A.P. Scheer, Int. J. Chem. Eng. Appl. 5 (3) (2014) 281–286, https://doi.org/10.7763/ijcea.2014.v5.394.
[61] K.J. Hwang, P.Y. Sz, J. Memb. Sci. 347 (1–2) (2010) 75–82, https://doi.org/10.1016/j.memsci.2009.10.008.
[62] G.T. Vladisavljević, P. Vukosavljević, M.S. Veljović, Food Bioprod. Process. 91 (4) (2013) 473–480, https://doi.org/10.1016/j.fbp.2013.05.004.
[63] C. Cai, W. Sun, S. He, Y. Zhang, X. Wang, Front. Environ. Sci. Eng. 17 (10) (2023), https://doi.org/10.1007/s11783-023-1726-9.
[64] E. Turano, S. Curcio, M.G. De Paola, V. Calabrò, G. Iorio 209 (2002) 519–531.
[65] P. Singh, N.A. Manikandan, M. Purnima, K. Pakshirajan, G. Pugazhenthi, J. Water Process Eng. 38 (August) (2020) 101615, https://doi.org/10.1016/j.jwpe.2020.101615.