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Recovery of Salts from Synthetic Erythritol Culture Broth via Electrodialysis: An Alternative Strategy from the Bin to the Loop

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Abstract: Sustainability and circularity are currently two relevant drivers in the development and optimisation of industrial processes. This study assessed the use of electrodialysis (ED) to purify synthetic erythritol culture broth and for the recovery of the salts in solution, for minimising the generation of waste by representing an efficient alternative to remove ions, ensuring their recovery process contributing to reaching cleaner standards in erythritol production. Removal and recovery of ions was evaluated for synthetic erythritol culture broth at three different levels of complexity using a stepwise voltage in the experimental settings. ED was demonstrated to be a potential technology removing between 91.7–99.0% of ions from the synthetic culture broth, with 49–54% current efficiency. Besides this, further recovery of ions into the concentrated fraction was accomplished. The anions and cations were recovered in a second fraction reaching concentration factors between 1.5 to 2.5 times while observing low level of erythritol losses (<2%), with an energy consumption of 4.10 kWh/m^3.

Keywords: electrodialysis; erythritol downstream; desalination; waste reduction; current efficiency

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1. Introduction

Erythritol is a four-carbon polyol used for foods, pharmaceutical and cosmetic products manufacture, among others [1–5]. Its high relative sweetness [1,2], and low caloric content [3,4] makes erythritol a competitive sugar substitute with promissory increases of demand in the market [6]. Erythritol is produced as an extracellular product in submerged cultures by various osmotolerant microorganisms from yeast, fungi-like yeast and fungi, including *Moniliella tormentosa pollinis*, *Yarrowia lipolytica*, *Moliniella Pollinis*, *Candida magnoliae*, *Candida sorbosivorans*, and *Pseudozyma tsukubaensis* [7–11]. The growth and metabolism of microorganisms are affected by osmotic stress modulated by salts, polyethyleneglycol, and carbon source addition [12–15]. At high osmotic pressure conditions, the growth rate decreases, while osmotolerant erythritol-producing microorganisms favour erythritol production and accumulation as a protective response to hyperosmotic conditions [16–18]. The effect of the hyperosmotic conditions by adding salts has been studied in the production of erythritol using NaCl and KCl as osmotic agents in concentration levels up to 50 g/L increasing the concentration of erythritol up to 50% compared to the control without the addition of salt [17–21].

To accomplish essential cellular tasks (maintenance, growth, and synthesis of products), microorganisms require a source of carbon, macronutrients (N, S, and P), micronutrients (K, Ca, Mg, Na, and Fe), and trace elements (Mn, Zn, Co, Mo, Ni, and Cu) [22–27]. Main components in culture media for erythritol production include salts as KH_2PO_4, $MgSO_4$, NaCl, $CuSO_4$, $FeSO_4$, $MnSO_4$, $CuSO_4$, $(NH_4)_2SO_4$; and other compounds from different sources as surfactants, peptone, and polysorbate 80 [8,28,29]. These compounds are added in concentrations between 0.2 g/L to 26 g/L in the culture stage [7,8,10,28,30–33].

The remaining salts from the culture media have to be separated from the culture broth to obtain pure erythritol, and therefore the non-used salts in the culture broth leave the process as a residual stream that has to be removed in the downstream section [34]. In the last decades, the recovery of nutrients has gained interest, becoming a multidisciplinary challenge for industry, water resources management, soil conservation, among other fields [34–37]. The transition from conventional processes to industrial circular processes represents an opportunity to tackle resource management and climate change concerns. In order to attend the depicted necessities, the European Union has developed projects as *Zero Brine* [36], *INCOVER* [37], *NITROMAN* [38], among other initiatives looking for technological alternatives that are able to reduce the impact caused by the waste and to evaluate the recovery of valuable compounds as phosphorus, sulphate, ammonia, and others. However, there is not enough scientific evidence regarding the consumption and recovery and reutilisation of salts from submerged cultures, especially considering the mandatory demand for salts in these biotechnological processes.

The removal of salts from erythritol culture broth is used as an example of a more general approach for recycling residuals from biotechnological culture and so far has been addressed by adsorption on ion exchange resins in the downstream section [34,39–41]. The principle behind this separation is the removal of salts by exchanging their position with other exchangeable ions (Na^+, H^+, among others) within cationic and anionic exchange resins. However, despite the high performance and efficiency of this conventional approach, downstream regeneration of ion-exchange resins requires chemical substances. It generates large amounts of alkaline and acid residual waste streams mainly composed of the salts removed from the culture broth. This fact represents a drawback to reach sustainable and cleaner production goals.

Alternatives for removing salts include membrane-based technologies, like forward osmosis, membrane distillation and electrodialysis [42]. Electrodialysis (ED) is an environmentally friendly and low energy demanding membrane-based technology suitable for the recovery of ionisable molecules from spent culture broths [42–46]. An electrical potential applied to the ED cell serves as the driving force for separating ionic species from the uncharged matter in an aqueous solution. Studies in ED application include recovery and reutilisation of alkaline fractions [47], separation and purification of L-phenylalanine [48], lactic acid [49], glycerol [50], glutamine [51], and 1,3-propanediol [52].

During the ED batch process, the feed located in the diluted and concentrated reservoir is recirculated in a closed loop through a stack of membranes in the ED batch mode, driven by a directly applied direct current. Cations and anions migrate from the feed solution to a concentrated fraction by going through cation- and anion-selective membranes, respectively. Two fractions are obtained at the end of the ED treatment: a purified product fraction with a low content of ions and a concentrated fraction rich in ions [50,53]. The main factors affecting the performance of ED treatment include the applied current density, the selectivity of the membranes (monovalent or multivalent), the flow of diluted and concentrated fractions, temperature, and pH [50,51,54].

Limiting current density (LCD) is a critical parameter that rules the overall performance of electrodialysis. The LCD value represents a threshold of a maximum current applied to the ED cell, at and above which adverse effects take place in the ED separation process. If the ED is operated in the limiting (at the threshold) or over-limiting (above the threshold) regime, dissociation of water molecules occurs in a thin layer at the membrane/diluted-solution interface. The H^+ and OH^- ions from water dissociation

participate in the transfer of current (through the ED membrane stack), reducing the overall ED process efficiency and increasing the energy consumption. Besides water splitting, fluid leakage, current leakage, and co-ion transfer occur if the threshold current density is exceeded [50].

This paper aims to evaluate the performance of the ED to separate and recover valuable ions from culture-broth process streams. Firstly, remove salts from erythritol culture broth, and then concentrate the same salts in a potentially reusable concentrated fraction. This assessment could become the first stage to evaluate the reutilisation of ions as an approach to treat waste stream from bin to loop.

The experiments were designed in three stages, gradually increasing the complexity of the fed solution. In this sense, the research question was to understand how can the complexity of culture broth solutions affect the performance of ED treatments. For this purpose, three different solutions were tested, and a stepwise voltage approach was used for the experiments. Each stage was judged upon the removal efficiency of ions in the diluted fraction, the concentration factor in the concentrated fraction, and the overall losses of products, by-products, and glucose used as a carbon source. Considering the industrial projection of this ED application, the current efficiency and energy consumption were also determined.

2. Materials and Methods

2.1. Reagents

All the reagents used in this study were purchased from Merck KGaA (Darmstadt, Germany). Anion multi-element standards (Certipur® Anion-Multielement-Standard I (PO_4^{3-}) and II (Cl^-, SO_4^{2-})) and cation multi-element standard (Certipur® Cationic-Multielement-Standard VI) (NH_4^+, K^+, Na^+, Ca^{2+}, Mg^{2+}) were used as standards for Ion-Chromatography. HPLC grade erythritol, glucose, and glycerol were used as the calibration curve standards. Milli-Q-Water was prepared according to DIN ISO 3696 (1991) [55].

2.2. Feed Solutions

The experimental part was developed in three stages, increasing the complexity of the solutions by adding compounds from three different groups present in the erythritol culture media:

2.2.1. Salts from Culture Media

This fraction includes all the ionisable compounds added as macronutrients, micronutrients, and traces elements, namely ammonium sulphate ($(NH_4)_2SO_4$), magnesium sulphate heptahydrate ($MgSO_47H_2O$), potassium dihydrogen phosphate (KH_2PO_4), ferrous sulphate heptahydrate ($FeSO_47H_2O$), manganese sulphate monohydrate ($MnSO_4H_2O$), zinc sulphate heptahydrate ($ZnSO_47H_2O$), calcium chloride dihydrate ($CaCl_2H_2O$), and sodium chloride (NaCl) as the osmotic agent used. These compounds are separated in the concentrated fraction.

2.2.2. Erythritol and By-Products

This fraction included erythritol as the main product, glycerol, and residual glucose as by-products. These compounds are neutral and not dissociable at the operational conditions (erythritol, glycerol, and glucose pKa values 13.9, 14.4, and 12.28, respectively) [56–58]. These compounds should remain in the diluted fraction.

2.2.3. Other Compounds from Culture Media

This fraction includes surfactant (tween 80) and peptone used as surfactant and emulsifier for membrane cell protection. These compounds should remain in the diluted fraction.

Table 1 presents the concentration levels and compounds added to the experiments. The solution fed to the system increased its complexity in each stage (S, SP, and SCB solutions), while the initial concentrated solution (equivalent to the S solution) remained

unchanged during the experiments as the receiving media for the salts. The compounds and concentrations in the feed solutions were defined based on a preliminary characterisation of ions and conductivity of a real erythritol culture broth after the fermentation stage (data not shown). According to these results, the concentrations used in this work corresponded to 50% culture media before fermentation. Considering the lack of studies regarding the consumption of osmotic agents, in this study sodium chloride was added only to adjust the conductivity of feed solutions.

Table 1. Description of the evaluated solutions.

Compound	Molecular Formula	[g/L]	Sample		
			S	SP	SCB
Ammonium sulphate	$(NH_4)_2\ SO_4$	1.4	x	x	x
Magnesium sulphate heptahydrate	$MgSO_4\ 7H_2O$	0.5	x	x	x
Potassium dihydrogen phosphate	KH_2PO_4	4.2	x	x	x
Ferrous sulphate heptahydrate	$FeSO_4\ 7H_2O$	0.0025	x	x	x
Manganese sulphate monohydrate	$MnSO_4\ H_2O$	0.00085	x	x	x
Zinc sulphate heptahydrate	$ZnSO_4\ 7H_2O$	0.0007	x	x	x
Calcium chloride dihydrate	$CaCl_2\ 2H_2O$	0.001	x	x	x
Sodium chloride *	NaCl	0.8	x	x	x
Tween 80	$C_{64}H_{124}O_{26}$	0.25			x
Peptone	n.a.	0.05			x
Erythritol	$C_4H_{10}O_4$	5		x	x
Glycerol	$C_3H_8O_3$	5		x	x
Glucose	$C_6H_{12}O_6$	2		x	x

Note: n.a.: not available. * NaCl was added to adjust conductivity to the measured value in the real culture broth. S: only salts solution, SP: salts + product, SCB: synthetic culture broth.

The compounds added to the three feed solutions are marked with a cross in Table 1. The first stage considers only the salt content in spent culture media (S solution), where both the diluted and the concentrated solution have the same initial composition. The initial concentrated solution is kept the same (S solution) in further stages, whereas the diluted solution complexity gradually increased. The main products and by-products were added to the diluted stream in the second stage (SP solution). Finally, the third stage studied the treatment of the complete erythritol synthetic culture broth (SCB), including tween 80 and peptone, introduced to the diluted stream.

2.3. Experimental Settings

An electrodialysis laboratory unit ED 64004 (PCCell GmbH, Heusweiler, Germany) was used in this work. The ED 64004 consisted of a current generator supplying constant voltage (0–30 V), a control unit, an online measuring system for pH, temperature, conductivity, and voltage (JUMO GmbH & Co. KG, Fulda, Germany), and three independent storage containers for diluted, concentrated and electrode rinsing solution connected in circuits with three magnetically coupled centrifugal pumps NDP 25/4 (ITS-Betzel, Hatterscheim, Germany).

The ED membrane stack was composed of two electrode compartments with 10 identical cells. Each cell unit has an anion exchange membrane (AEM. Ref: PC SA 10x) and a cation exchange membrane (CEM. Ref: PC SK 9x). The effective membrane area was 64 cm^2 per membrane, with 1 mm of spacing between membranes. The anode and cathode materials were Pt/Ir-coated titanium and theV4A steel, respectively. They were placed in the polypropylene electrode housing material.

All the voltages assessed in this approach corresponded to 80% of the limiting current density (LCD) determined using the Cowan and Brown method [59]. A stepwise voltage was the approach used for the application of the direct current in this work. Through this approach, four different voltages (10, 9, 7 and 6 V) were kept constant during a period and changed as soon as the conductivity of the diluted fraction turned from 5.47 mS/cm

(initial value) to 3, 2 and 1.4 mS/cm. The four voltage-conductivity pairs were selected from preliminary assays finding the LCD for different conductivity levels (data not shown).

S, SP, and SCB experiments were performed in triplicates. In situ parameters as pH, conductivity, and temperature were recorded at the beginning and during the treatment. 1.5 L of diluted and concentrated solution were pumped and recirculated to the system at a constant flowrate rate of 15 L/h, whereas the electrode-rinsing solution (0.25 M Na_2SO_4) was recirculated at 150 L/h.

A total of 12 samples per experiment were taken at different interval times, considering the reduction in the concentration of ions. The first nine samples were taken every 5 min, the 10th and 11th were taken every 10 min, and the last sample was taken after the conductivity of the diluted fraction reached a chosen value of 0.28 mS/cm. This value was selected, having as reference tap water conductivities. Due to the short-term characteristics of the experiments, scaling and fouling phenomena were not considered in the analysis.

2.4. Analysis of Anions and Cations with Ion Chromatography (IC)

Inorganic ions (Na^+, NH_4^+, K^+, Mg^{2+}, Cl^-, SO_4^{2-}, PO_4^{3-}) in diluted and concentrated fractions were determined by ion chromatography (IC) on a Dionex ICS 5000+ system (Thermo Scientific, Waltham, MA, USA) equipped with conductivity detectors and an autosampler Dionex AS-DV (Thermo Scientific, Waltham, MA, USA) for simultaneous injection onto both anion and cation columns.

Anions were separated using a Dionex ATV-HC trap pre-column (9 × 75 mm) using gradient elution. Cations were separated on a Dionex CS12A analytical column (4 × 250 mm) with a CG12A guard column (4 × 50 mm) using an isocratic elution. The mobile phases used were 5mM NaOH, 25mM NaOH, and 20 mM methanesulphonic acid. All the flows, gradient specifications, and current suppression programs are described by Linderman et al. (2020) [60].

2.5. Analysis of Erythritol, Glycerol, and Glucose with HPLC

Erythritol, glycerol, and glucose were determined on a High-Performance Liquid Chromatography (HPLC) equipped with a degasser unit DGU-20A3R (Shimadzu, Kyoto, Japan), an autosampler unit SIL-20ACHT (Shimadzu, Kyoto, Japan), a column oven CTO-20A (Shimadzu, Kyoto, Japan), and a refractive index detector RID-20A (Shimadzu, Kyoto, Japan). The mentioned compounds were separated on a Shodex SH1011 column (80 × 300 mm) with a guard column Shodex Sugar SH-G (6.0 × 50 mm). The HPLC was coupled with a refractive index detector RID-20A (Shimadzu, Kyoto, Japan). The separation was performed at an isocratic gradient. The mobile phase used was 5 mM H_2SO_4 at 0.6 mL/min for 20 min. The oven and RID temperature were 50 °C.

2.6. Removal of Ions and Current Efficiency (CE)

The percentage of removal (R_j) of ions was calculated by using Equation (1), where $C_j(t_i)$ means an initial or a given time, and $C_j(t_{i+1})$ means the final time of the period to be evaluated.

$$R_j(\%) = \frac{C_j(t_i) - C_j(t_{i+1})}{C_j(t_i)} \times 100\% \quad (1)$$

Current efficiency (CE) was calculated based on Faraday's Law following the approach suggested by Wu et al. (2011) [47] (Equation (2)). Where n_j^i and n_j^f are the initial and final number of moles of salts in diluted fraction, F is the Faraday constant (96,485.33 sA/mol), I is the current value (A), t is the time (s), N is the number of cell pairs in the stack, and z is the ionic charge (pH-dependent for polyvalent ions).

$$CE(\%) = \frac{F \times \sum_j z_j(n_j^i - n_j^f)}{N \times I \times t} \quad (2)$$

Considering that the energy applied is used to promote the simultaneous migration of cations and anions towards the electrodes, CE was estimated only based on the behaviour of anions in the solution.

2.7. Energy Consumption

The energy required for the desalination (E_d) of the feed solution was calculated following Equation (3), where U is the applied voltage (V), I is the applied current (A), t is the desalination duration (h), and V is the treated volume of the feed solution (m^3).

$$E_d\left(\frac{kWh}{m^3}\right) = \frac{U \times I \times t}{V} \quad (3)$$

Energy consumed by the pumps (E_p) in the diluted, concentrated, and electrode-rinsing solutions was calculated following Equation (4), where Q is the flow of the diluted, concentrated, or electrode-rinsing solution (m^3/s), Δp is the differential pressure (Pa) between the inlet and outlet of the ED stack, η is the pump efficiency (assumed as 75% for all three pumps), and V is the treated volume of the diluted solution (m^3).

$$E_p\left(\frac{kWh}{m^3}\right) = \frac{Q \times \Delta p}{\eta \times V} \quad (4)$$

3. Results

3.1. Chemical Composition of S, SP, and SCB Solutions

Table 2 presents the characterisation of initial monovalent and bivalent ions in the S, SP, and SCB solutions. The preparation process causes minor deviations in the chemical composition of the solutions. Due to the pH levels and the polyvalent nature, sulphur and phosphorus ions can adopt different dissociation values. Considering the initial pH value (Table 3), the main form of these ions was the monovalent dihydrogen phosphate ($H_2PO_4^-$) and sulphate (SO_4^{2-}) with −1 valence.

Table 2. Ion concentration in feed S, SP and SCB solutions.

Ion	S * [mg/L]	SP [mg/L]	SCB [mg/L]
Cl^-	426.9	399.6 ± 17.6	431.5 ± 8.2
SO_4^{2-}	1162.2	1088.6 ± 29.8	1165.7 ± 11.0
$H_2PO_4^-$	1281.7	1216.8 ± 47.7	1236.5 ± 7.8
Na^+	272.1	279.4 ± 31.2	289.6 ± 1.5
NH_4^+	230.4	221.8 ± 15.0	248.9 ± 34.5
K^+	515.4	516.9 ± 75.6	588.1 ± 13.6
Mg^{2+}	67.2	71.6 ± 15.3	69.1 ± 2.6

Note: * Only one sample was characterised.

Table 3. Physicochemical parameters of the diluted and concentrated fraction at the initial and final stages of the ED separation process.

Parameter	Units	Diluted Fraction					
		S		SP		SCB	
		Initial	Final	Initial	Final	Initial	Final
pH		4.59–4.60	3.92–4.07	4.59–4.63	4.29–4.30	4.53–4.55	4.23–4.42
Temperature	°C	21.7 ± 2.9	20.2 ± 0.5	21.2 ± 1.1	20.8 ± 0.5	20.8 ± 3.8	20.2 ± 0.2
Conductivity	mS/cm	5.57 ±0.03	0.28 ± 0.00	5.41 ± 0.05	0.28 ± 0.00	5.43 ± 0.03	0.28 ± 0.00

Table 3. *Cont.*

Parameter	Units	Concentrated Fraction					
		S		SP		SCB	
		Initial	Final	Initial	Final	Initial	Final
pH		4.59–4.60	4.40–4.59	4.56–4.64	4.39–4.52	4.58–4.59	4.48–4.72
Temperature	°C	21.7 ± 2.9	20.3 ± 1.1	22.2 ± 1.1	20.8 ± 0.5	24.2 ± 1.4	19.8 ± 0.5
Conductivity	mS/cm	5.57 ± 0.03	11.79 ± 0.51	5.57 ± 0.02	10.02 ± 0.02	5.54 ± 0.01	11.2 ± 0.06

According to the measured concentrations in Table 2, an average erythritol culture media would need more anions (72% wt.) than cations (28% wt.); and more total monovalent (69% wt.) than bivalent ions (31% wt.). Differences in the amounts of ions added per species are expressed in the concentration gradients. These differences are considered in other sections for the analysis of removal and current efficiency.

3.2. Physicochemical Parameters

Table 3 presents the physicochemical parameters measured at the beginning and the end of each experiment. A slight drop between 0.13–0.67 units in pH value was observed in the diluted fraction and between 0.01 and 0.19 in S and SP feed solutions in the concentrated fraction. However, this level of pH variations did not affect the overall removal of cations and anions from the diluted fraction (Table 4). Temperature control plays an important role in avoiding effects caused by temperature depending processes as diffusion and changes in the affinity and selectivity through the cation-exchange and anion-exchange membranes [61]. Finally, the reduction and increase of the conductivity values in the diluted and concentrated fraction, respectively, represent in situ proof of the effectiveness of the ED treatment for the removal of ions from erythritol synthetic culture broth.

Table 4. Average removal efficiency per ion.

Sample	Solution		
	S [%]	SP [%]	SCB [%]
Cl^-	99.3 ± 0.1	99.0 ± 0.3	99.0 ± 0.3
SO_4^{2-}	98.6 ± 0.3	95.4 ± 0.6	97.3 ± 0.0
$H_2PO_4^-$	86.6 ± 0.8	91.0 ± 0.6	93.9 ± 0.7
Na^+	86.3 ± 1.5	88.4 ± 3.2	91.9 ± 1.0
NH_4^+	92.6 ± 1.5	91.8 ± 0.8	94.5 ± 1.6
K^+	94.6 ± 0.1	94.5 ± 0.6	94.9 ± 0.6
Mg^{2+}	92.9 ± 0.6	93.7± 0.5	91.7 ± 0.7
Total av.	93%	93%	96%

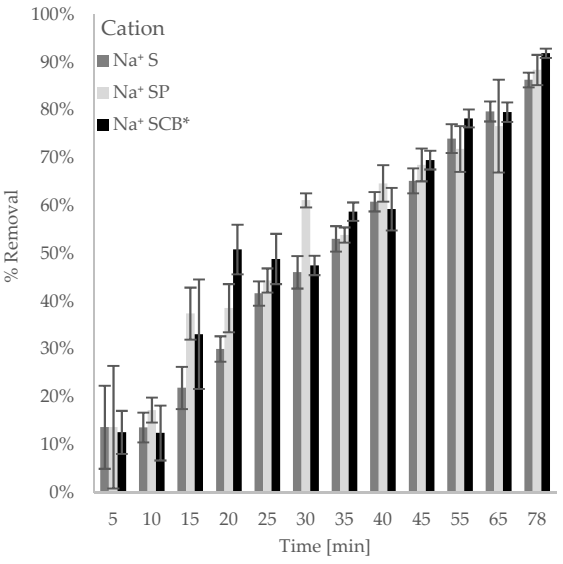
3.3. Desalination of Product in the Diluted Fraction

A final conductivity value of 0.28 mS/cm was reached after 78 min for S and SP solutions and min 84 for SCB solution, representing an increase of 8% in the treatment time compared to S and SP solution. This 8% increase is related to the differences in the composition of the solutions involved. SCB solution included two substances of different nature: the non-ionic surfactant, Tween 80, and the protein hydrolysate, peptone, widely used in biotechnological cultures.

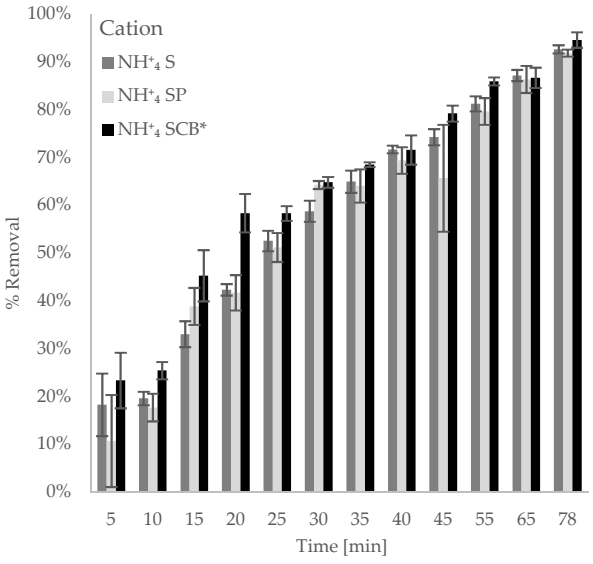
Table 4 shows the removal efficiency of ions in the diluted fraction, the total removal efficiency values were 93%, 93%, and 96% for S, SP, and SCB solutions. In all cases, the removal of individual anions and cations was higher than 86%. Among anions, chloride exhibited the highest and dihydrogen phosphate the lowest removal efficiency. For the cations, the highest removal efficiency was reached for potassium and the lowest for sodium.

The ion removal efficiency over time is presented in Figure 1. Similar removal efficiency was observed within the three solutions, meaning that complex solutions (SCB) presented

the same general behaviour as the simplest solution (S). The addition of non-charged compounds to the SP solution did not affect the treatment time and removal of ions kept at the same level as S solutions. Slightly large error bars were observed for some ions in solutions, this is due to the difference in concentration levels and the accuracy of the analytical tools. Besides that, the concentration values exposed in the error bars can be explained by differences during sampling, or the response of the electrodes to the application of current. Notwithstanding, the samples followed a clear trend for ion removal. This consideration is also valid for the concentration of products going to the concentrate fraction (Figure 2).



(**a**)



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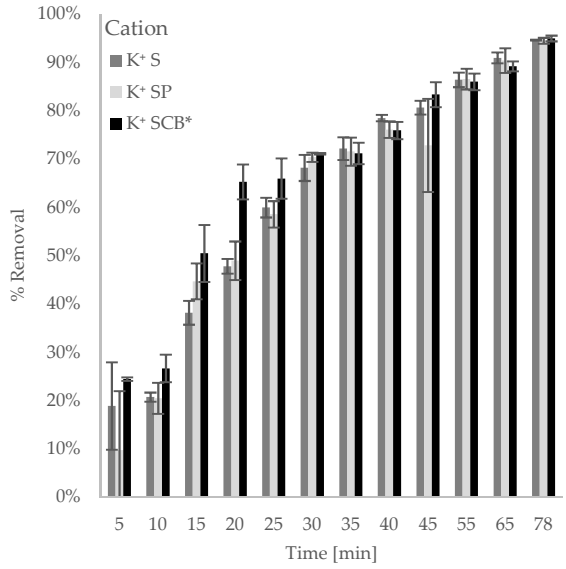
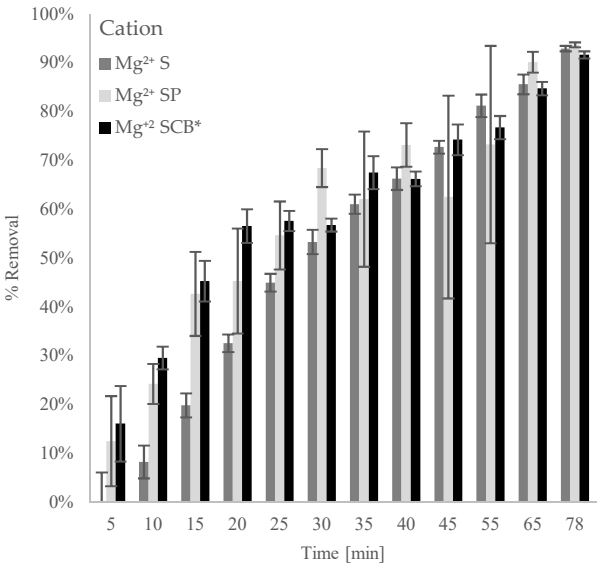
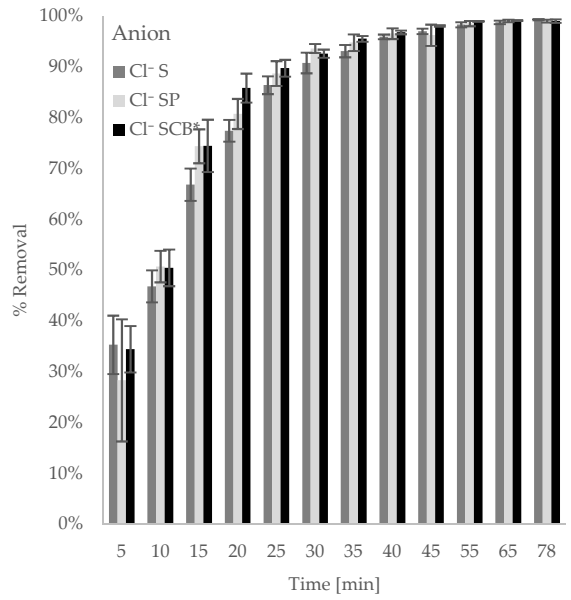


(**c**)



(**d**)

Figure 1. *Cont.*



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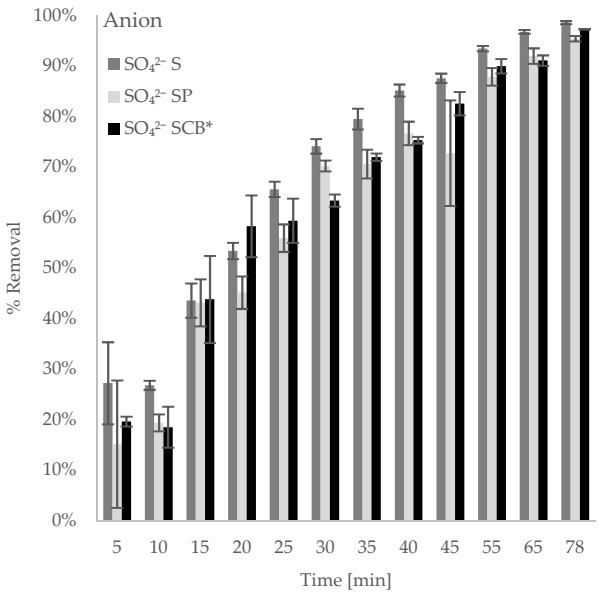
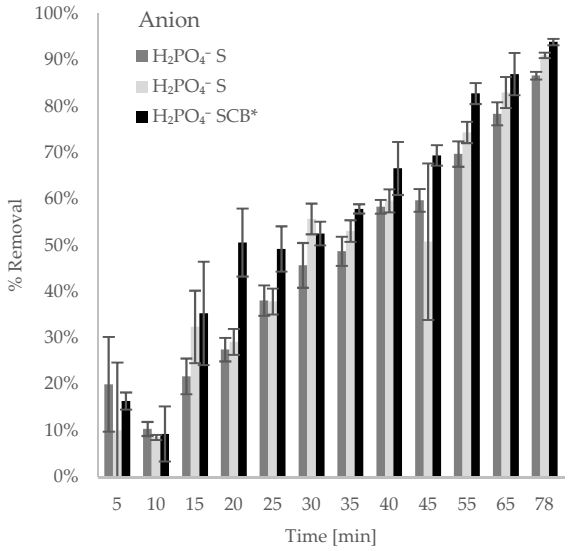


(**f**)



(**g**)

Figure 1. Removal of ions in diluted fraction. (**a**) sodium; (**b**) ammonia; (**c**) potassium; (**d**) magnesium; (**e**) chloride; (**f**) sulphate; (**g**) dihydrogen phosphate for salts solution (S), salts + product solution (SP), and synthetic culture broth solution (SCB). * Final time for SCB samples was 84 min.

Rakicka et al. (2016) [62] developed a study for the desalination of an erythritol culture broth using ion exclusion resins (Lewatit S3428 and S2568H). Three different fractions were obtained: a saline waste, recycle, and product fraction with a 95% degree of desalination (based on NaCl removal).

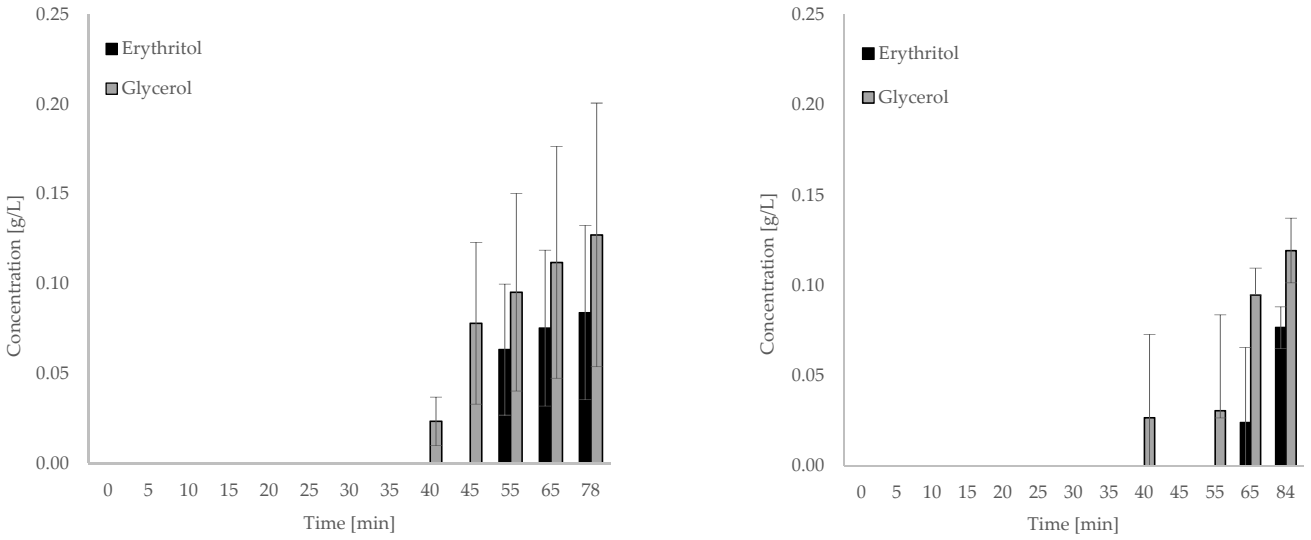


Figure 2. Concentration of products in the concentrated fraction (**a**) salts + product solution (SP); (**b**) synthetic culture broth solution (SCB).

The results obtained in this study exhibited a remarkable performance of ions removal (96%), comparable to the reported by Rakicka et al. (2016) [62]. However, an advantage of ED is the absence of a waste stream, compared to large volumes of saline waste, operation recycles and regeneration-wastewater generated for ion-exchange treatments.

3.4. Performance of Ion-Removal Rate in S, SP, and SCB Samples

The ion-removal rate is defined as the average percentage of ions removed divided per time (%removed/min); this rate is strongly influenced by the concentration of ions in the diluted fraction and the voltage applied. Hence, reductions in the ion-removal rate within the time depend on the driving force given by the electrical potential, the mobility of the ions, the molecule charges, and concentration gradient (Table 2). The breakpoint time (BKP) indicates that sudden reductions in the removal rate occur, caused by the downturn in the driving force.

By identifying the BKP it was possible to characterise two periods of ions removal: a fast removal period and a slow removal period. Therefore, the determination of BKP time allows one to understand the processes and determine possible alternatives to improve CE, avoid back diffusion, product losses, and reduce energy consumption. Table 5 presents the breakpoint time (BKP), the reduction in the ion-removal rate after the BKP, the amounts of ions removed (%) until BKP and the average values of the rate of ions removal before and after BKP (% removed/min).

The effect over the mobilities of ions is depicted in the reduction of the ion-removal rate presented at different BKP time as shown in Table 5. In the first place, the ion mobility is decelerated due to the decreasing ionic strength of the diluted fraction among all three solutions (S, SP, and SCB), especially after the BKP time. In the second place, differences in ion mobilities between stages with increasing solution complexity can be observed, as discussed in the following paragraphs.

In S solutions, two BKP times were observed. The trend reflected by the average value of ions-removal rate before and after BKP time allows determining that chlorine was the ion with the highest mobility and overall removal. Notwithstanding the different levels of concentration described in Table 2, phosphate and sodium have a lower average value of ions-removal rate before and after BKP time, demonstrating lower mobilities than the other ions despite the initial concentration. Chloride was the first ion presenting a reduction in the removal rate of 90.3% after 25 min of treatment (BKP time). By that time, approximately

86.4% of the total chloride ions were already removed from the diluted fraction. The second BKP time was observed at 40 min for the other ions in solution with reduction levels in the removal rate between 52.3–81.8% when approximately 58.3–85.1% of the total ions were already removed (SO_4^{2-} > K^+ > NH_4^+ > Mg^{2+} > Na^+ > $H_2PO_4^-$).

Table 5. Breakpoint time for ions removal in S, SP, and SCB samples.

Ion	S			SP			SCB		
	BKP Time *	Reduction in Removal Rate **	Removed at BKP Time ***	BKP Time *	Reduction in Removal Rate **	Removed at BKP Time ***	BKP Time *	Reduction in Removal Rate **	Removed at BKP Time ***
	[min]	%	%	[min]	%	%	[min]	%	%
K^+	40	77.8	78.5	55	78.1	86.5	45	84.0	83.3
NH_4^+	40	69.0	71.7	55	62.4	79.6	45	78.0	79.2
Na^+	40	53.5	60.8	40	61.6	64.7	45	64.4	69.5
Mg^{2+}	40	61.8	66.3	65	80.3	90.2	45	71.5	74.3
Cl^-	25	90.3	86.4	25	92.8	88.7	25	92.6	85.8
SO_4^{2-}	40	81.8	85.1	55	79.0	87.9	45	78.5	82.5
$H_2PO_4^-$	40	52.3	58.3	55	40.9	74.4	55	74.3	82.8

	Average ions-removal rate before and after BKP [%removal/min]													
	K^+		NH_4^+		Na^+		Mg^{2+}		Cl^-		SO_4^{2-}		$H_2PO_4^-$	
	t < BKP	t > BKP	t < BKP	t > BKP	t < BKP	t > BKP	t < BKP	t > BKP	t < BKP	t > BKP	t < BKP	t > BKP	t < BKP	t > BKP
S	2.0	0.4	1.8	0.5	1.5	0.7	1.6	0.6	3.5	0.3	2.1	0.4	1.5	0.7
SP	1.6	0.3	1.5	0.4	1.6	0.6	1.4	0.3	3.5	0.3	1.6	0.3	1.3	0.7
SCB	1.9	0.3	1.5	0.4	1.5	0.5	1.7	0.5	4.3	0.3	1.8	0.4	1.5	0.4

Note: * BKP time: breakpoint time. ** reduction in the average ion-removal rate after the BKP. *** % of ions removed until reaching the BKP. t < BKP: time before BKP. t > BKP: time after BKP.

SP solutions presented slower removal rates compared to S solutions. In addition, four BKP times at 25, 40, 55, and 65 min were observed indicating a late onset of reduction in the removal rate, while slightly higher overall removal values than S samples were reached.

Despite requiring a longer time to reach the final conductivity value, the SCB solution presented similar levels of the total reduction in the removal rate (74.3–92.6%) compared to SP. In this case, three different BKP times 25, 45, and 55 min were obtained at the moment when approximately 69.5–95.8% of ions were removed. The average value of ions-removal before the BKP point showed an increase compared to SP data. SCB results allow us to make conclusions about three possible phenomena presented. The first regards the effects of non-charged compounds on possible polarisation concentration phenomena presented due to concentration gradients generated between the bulk liquid and the interfacial layer, which increases within the concentration of non-charged compounds. The second regards increases in the membrane resistance due to electrostatic and hydrophobic interactions presented between peptone and the membrane surfaces, specifically on cationic membranes [63,64]. The third phenomena relate to possible effects of polysorbate 80 as a non-charged surfactant on the increase of the average ions-removal rate and the mobility of the ions due to minimisations of the polarisation concentration phenomena, resulting in consequence in slightly higher overall removal values (Figure 1) while demanding longer treatment times. These results are in line with the study developed by Gohil et al. (2005) [65], who described the reduction in the polarisation concentration phenomena caused by the addition of surfactants. The authors proved the formation of a layer of surfactants due to their amphiphilic nature (hydrophobic head and hydrophilic tail). The ions can accumulate temporarily over this layer, reducing the concentration gradients in the membrane-solution interfacial layer, and therefore reducing possible polarisation concentration effects [65].

3.5. Current Efficiency (CE)

Current efficiency (CE) is a measure of the current utilisation to transmit ions instead for other uses, such as water splitting. An efficient process can separate the ions while ensuring a continuous movement and transfer of ions between the cathode and anode.

Table 6 presents a comparison of the results obtained in this work and other solutions with similar and even higher complexity levels like lignocellulosic effluent, molasses, and brine solutions from downstream processes and waste treatment. This table includes the energy application approach, CE, and removal values. Studies that have reported high CE

values (90–220%) indicating the dependence of the energy approach used and the initial levels of ions in diluted fraction by the order of 2–414 g/L [66–69]. By understanding this dependence, appropriated current densities or current application approaches can be selected, avoiding back diffusion of ions and water splitting, and therefore reach high CE [70]. These conditions can be obtained by applying the appropriate arrangement of the operating parameters such as voltage and feed flow rate with the initial concentration of ions. In this work, CE values between 49% and 54% were obtained for S, SP, and SCB solutions (Table 6), meaning that around 54% of the energy applied is used to transport the ions, rather lower than most of the studies presented in Table 6. Even though it was not rigorously followed, there were no significant changes in the volume observed. However, water splitting and electro-osmosis transport could be responsible for the remaining 46% of the energy provided.

Table 6. Current efficiency values.

Treated Sample	Initial Concentration [g/L]	Energy Application Approach	Removal/ Concentration	Current Efficiency	Ref.
Saline solution	10	Constant voltage 6 V	3.92%	47.54%	[68]
Formic acid solution	276–414	Constant current density	140% *	100–220%	[70]
Saline solution	29.8	Non-uniform current 5.75 A (total)	92.7%	94%	[71]
Brine from pickled prunes	100	Constant current density <10 mA/cm^2	87.6%	90–95%	[66]
Lignocellulosic effluent [1,2]	14.05	Constant current density 39.1 mA/m^2	~96%	~95%	[67]
Sugarcane juice [2]	2.03	Constant current density 17.2 mA/m^2	>90%	~75%	[67]
Molasses [2]	81.2	Constant current density 101.6 mA/m^2	~60%	~104%	[67]
S	3.90	Stepwise voltage 10, 9, 7 and 6 V	93%	54 ± 3%	This work
SP	3.80	Stepwise voltage 10, 9, 7 and 6 V	93%	49 ± 4%	This work
SCB	4.00	Stepwise voltage 10, 9, 7 and 6 V	96%	53 ± 2%	This work

Note: * Concentration factor. [1] hydrolysate from acid hydrolysis. [2] Only cations were considered.

3.6. Recovery of Salts in the Concentrated Fraction

At the end of the treatment, the concentration of ions in the concentrated fraction was 1.5 to 2.5 times higher than the initial concentrations (Table 7). These concentration levels are in agreement with the increases in conductivity presented in Table 3. The results showed different behaviours analogously with the composition of the feed solutions. It can be observed that cation selectivity decreases as the complexity of the solution increases. This fact is analogous to the trend of ions removal presented in the last section and connected to the possible effects of peptone over CEM. With the concentration levels obtained, the number of ions in the concentrated solution is similar to the initial ions used for the culture stage, this represents an attractive alternative to reuse the ions either into erythritol culture media or other processes to reduce waste, close loops, and go deep into circular economy standards.

Table 7. Concentration factor in the concentrated fraction.

Stage	Compound						
	Na^+	NH_4^+	K^+	Mg^{2+}	Cl^-	SO_4^{2-}	$H_2PO_4^-$
S	2.4 ± 0.1	2.1 ± 0.1	2.3 ± 0.1	2.7 ± 0.0	1.9 ± 0.2	1.9 ± 0.1	1.5 ± 0.1
SP	2.5 ± 0.3	1.8 ± 0.2	2.0 ± 0.3	1.4 ± 0.1	2.6 ± 0.6	2.3 ± 0.3	2.3 ± 0.4
SCB	2.1 ± 0.2	1.8 ± 0.3	2.0 ± 0.5	1.5 ± 0.0	2.4 ± 0.7	2.4 ± 0.6	2.1 ± 0.3

3.7. Losses of Products in the Concentrated Fraction

The concentration of erythritol, glycerol, and glucose in the concentrated fraction (Figure 2) was measured to determine the losses of product passing the membrane. Concentrations of 0.13 g/L and 0.12 g/L of glycerol and 0.08 g/L of erythritol were determined in the concentrated fractions of SP and SCB solutions after 40 min of treatment. The presence of products in the concentrated fraction was determined after the BKP time when the rate of removal of ions decreased significantly for all the ions. These concentrations represented losses of about 2.31% and 1.96% of glycerol and 1.7% and 1.39% of erythritol in SP and SCB solutions.

Product losses in ED can be related to different factors: the charge of molecules in solution susceptible to pH value and pH changes (e.g., amphoterism), affinity by membrane material, and diffusional effects [48,69,72]. Considering the non-amphoteric nature of the substances and the pH stability of the process (Table 2), a movement of products due to protonation and electric charge is unfeasible. Consequently, losses of product going through the membranes can be mainly associated with diffusional phenomena like diffusion-driven for the difference in concentrations or electro-osmosis caused by water cotransport [69,72]. Besides losses of valuable material, this diffusional phenomenon can cause biofouling, reducing the life of the membranes and the performance of separation.

To illustrate the level of losses in ED, Shen et al. (2005) [44] reported overall losses of about 16% of glutamine (initial concentration glutamine 32 g/L) while removing 95% of ammonium sulphate using ED, which are notably higher than the levels obtained in this work. This fact confirms the suitability of the use of ED in the purification of erythritol [64]. A possible alternative for reducing diffusional effects can be assessed through the decrease of treatment times by adopting a different strategy for the application of voltage.

3.8. Energy Consumption

Total energy consumption includes the energy required for the desalination and the energy consumed to pump the water fractions and electrode-rinsing solution through the ED stack. The total average energy consumption for the triplicates of S, SP, and SCB solutions is 3.96 kWh/m^3, 3.87 kWh/m^3, and 4.10 kWh/m^3 of the treated solution, respectively. The energy consumed for the treatment of the SCB solution is increased compared to the S and SP solutions due to the increased desalination time required for the same diluted fraction quality. The energy consumed for desalination makes roughly 40%, for pumping the diluted and concentrated fractions 10%, while the pumping of the electrode-rinsing solution used around 50% of the total energy in all tested solutions (S, SP, and SCB).

The energy consumption for culture broth desalination presented in this work is in a similar range to the values reported by Doornbusch et al. (2020) [73]. They attained 4.88–1.72kWh/m^3 for desalting NaCl solution (approx. conductivity 49.3 mS/cm) to drinking water quality, however without accounting for the energy consumed for the pumping of the electrode-rinsing solution.

4. Discussion

The selectivity for removing ions presented in this study did not follow any pattern based on molar mass, charge, or initial concentration. For the cations, a higher selectivity towards potassium and magnesium than sodium was observed, which is in correspondence

to results reported by Luo et al. (2020) [73]. Whereas the anions removal showed significantly higher selectivity for chloride compared to dihydrogen phosphate. Several authors have discussed ion selectivity remarking on some important issues such as the interaction between the ions and the AEM/CEM. These interactions include the groups attached to the membrane skeleton, membrane moisture, mobility in the membranes, rheological conditions [73–77].

The main effect caused by the addition of Tween 80 and peptone to SCB solution was the increase in the treatment time, without reductions in the ions-removal efficiency. (Figure 1). The increase in the treatment time is related to interactions between peptone and the ion exchange membranes. In long-term ED processes, this fact might represent membrane fouling, increasing the energy consumption, and affecting the overall performance of the separation. Ruiz et al. (2007) [62], Mikhaylin and Bazinet (2016) [78], and Nichka et al. (2020) [63], reported fouling on ion-exchange membranes due to the electrostatic and hydrophobic interactions between protein fractions and membranes. These interactions cause material deposition over the membrane surface, producing alterations that increase the membrane electrical resistance, reducing the rate of ion migration through the membranes and the overall performance of the separation. Some alternatives are used to mitigate fouling effects, namely the cleaning-in-place (CIP) routines and the implementation of pulsed electric field routines during treatment [64].

The ED application was capable of removing up to 96% of ions in the complex SCB sample. Chloride showed a separation behaviour significantly better in terms of mobility and removal efficiency in all the feed solutions evaluated, followed by sulphate, potassium, and ammonium. In general terms, the experimental results demonstrated a higher affinity and selectivity for anions than cations, explained by the faster movement of anions than cations at the same conditions. Similar results were reported by Kooistra (1967), who described the polarisation curves for the characterisation of ion-exchange membranes [79].

Considering that ED was demonstrated to be a suitable alternative for removing ions from erythritol culture broth, additional research work is required to improve CE and to increase the knowledge about membrane performance. These efforts could be directed towards the assessment of samples with a higher concentration of ions (concentrated culture broths, increasing the driving force), determining the maintenance requirements for the membrane and increase in the effectivity of the current applied (reduction of the resistance). To address current effectiveness, it is recommendable to explore different current application approaches, including constant current density [66] or new approaches as pulsed electric field routines [64].

5. Conclusions

Electrodialysis proved to be a suitable alternative to remove and recover salts from synthetic culture broth (SCB) solution, obtaining a remarkable desalination of 96%, comparable with values reported in the literature. The ion-removal rates followed the same trends in the three solutions (S, SP, and SCB), suggesting that ion selectivity and mobility are influenced by interactions between the species in solution and the groups attached to the membrane.

The salts removed from the synthetic culture broth and other solutions were concentrated 1.5 to 2.5 times in a second fraction. Producing a solution with similar characteristics to the initial culture media. Besides, less than 2% of products were found in the concentrate fraction after 40 min of treatment, with average energy consumption up to 4.10 kWh/m^3.

Current efficiency values of 49–54% were observed during the experiments. These values are still below the average reported in the literature. To improve the CE performance and address product losses, it is necessary to implement strategies to reduce the resistance and improve energy utilisation. The strategies include the concentration of the fed solution, reduction in the total treatment time, and the assessment of different current applications (e.g., constant current treatment or pulsed electric treatment).

ED for culture broth desalination represents an attractive option to be implemented in biotechnological downstreaming as an alternative to remove ions, returning them either into culture media or other processes, reducing waste generation, and closing loops moving towards circular economy processes.

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References

1. Martău, G.A.; Coman, V.; Vodnar, D.C. Recent advances in the biotechnological production of erythritol and mannitol. *Crit. Rev. Biotechnol.* **2020**, *40*, 608–622. [CrossRef]
2. Boesten, D.M.P.H.J.; den Hartog, G.J.M.; de Cock, P.; Bosscher, D.; Bonnema, A.; Bast, A. Health effects of erythritol. *Nutrafoods* **2015**, *14*, 3–9. [CrossRef]
3. Kasumi, T. Fermentative production of polyols and utilization for food and other products in Japan. *Japan Agric. Res. Q.* **1995**, *29*, 49–55.
4. BeMiller, J.N. Carbohydrate and Noncarbohydrate Sweeteners. In *Carbohydrate Chemistry for Food Scientists*; Ball, M., Ed.; Elsevier: Amsterdam, The Netherlands, 2018; pp. 371–399. ISBN 9780128120699.
5. Hao, H.X.; Hou, B.H.; Wang, J.K.; Zhang, M.J. Solubility of erythritol in different solvents. *J. Chem. Eng. Data* **2005**, *50*, 1454–1456. [CrossRef]
6. Ahuja, K.; Rawat, A. *Erythritol Market Size by Form (Powder, Granular), by Application (Beverage, Bakery, Confectionery & Dairy Products, Personal Care, Pharmaceutical), Regional Outlook, Application Potential, Price Trends, Competitive Market Share & Forecast, 2020–2026*; Global Market Insights: Selbyville, DE, USA, 2020.
7. Guo, J.; Li, J.; Chen, Y.; Guo, X.; Xiao, D. Improving Erythritol Production of Aureobasidium pullulans from Xylose by Mutagenesis and Medium Optimization. *Appl. Biochem. Biotechnol.* **2016**, *180*, 717–727. [CrossRef]
8. Kang, P.; Li, L.; Yan, L.; Ju, X.; Hu, C.; Yao, X. Enhancement of erythritol production in Trichosporonoides oedocephalis by regulating cellular morphology with betaine. *Chem. Pap.* **2019**, *73*, 2065–2072. [CrossRef]
9. Ghezelbash, G.R.; Nahvi, I.; Malekpour, A. Erythritol production with minimum By-product using Candida magnoliae mutant. *Appl. Biochem. Microbiol.* **2014**, *50*, 292–296. [CrossRef]
10. Kobayashi, Y.; Iwata, H.; Mizushima, D.; Ogihara, J.; Kasumi, T. Erythritol production by Moniliella megachiliensis using nonrefined glycerol waste as carbon source. *Lett. Appl. Microbiol.* **2015**, *60*, 475–480. [CrossRef] [PubMed]
11. Regnat, K.; Mach, R.L.; Mach-Aigner, A.R. Erythritol as sweetener—wherefrom and whereto? *Appl. Microbiol. Biotechnol.* **2018**, *102*, 587–595. [CrossRef]
12. Zhu, D.; Luo, F.; Zou, R.; Liu, J.; Yan, Y. Integrated physiological and chloroplast proteome analysis of wheat seedling leaves under salt and osmotic stresses. *J. Proteom.* **2021**, *234*, 104097. [CrossRef]
13. Yang, C.; Ji, X.; Pan, W.; Liu, Y.; Zhou, L.; Chen, Q.; Tang, X. Paliperidone ascending controlled-release pellets with osmotic core and driven by delayed osmotic pressure. *J. Drug Deliv. Sci. Technol.* **2018**, *48*, 193–199. [CrossRef]
14. Tian, X.; Wang, Y.; Chu, J.; Mohsin, A.; Zhuang, Y. Exploring cellular fatty acid composition and intracellular metabolites of osmotic-tolerant mutant Lactobacillus paracasei NCBIO-M2 for highly efficient lactic acid production with high initial glucose concentration. *J. Biotechnol.* **2018**, *286*, 27–35. [CrossRef] [PubMed]

15. Kim, S.I.; Choi, H.K.; Kim, J.H.; Lee, H.S.; Hong, S.S. Effect of osmotic pressure on paclitaxel production in suspension cell cultures of Taxus chinensis. *Enzyme Microb. Technol.* **2001**, *28*, 202–209. [CrossRef]
16. Gutierrez, C.; Abee, T.; Booth, I.R. Physiology of the osmotic stress response in microorganisms. *Int. J. Food Microbiol.* **1995**, *28*, 233–244. [CrossRef]
17. Kim, S.; Lee, K.; Kim, J.; Oh, D. Erythritol production by controlling osmotic pressure in Trigonopsis variabilis. *Biotechnol. Lett.* **1997**, *19*, 727–729. [CrossRef]
18. Li, L.; Gu, L.; Ju, X.; Hu, C.; Fu, J.; Cheng, H.; Kang, P. Osmotic pressure regulation using KCl for enhanced erythritol production using trichosporonoides oedocephalis ATCC 16958. *Food Sci. Technol. Res.* **2017**, *23*, 793–800. [CrossRef]
19. Liu, X.; Yu, X.; Xia, J.; Lv, J.; Xu, J.; Dai, B.; Xu, X.; Xu, J. Erythritol production by Yarrowia lipolytica from okara pretreated with the in-house enzyme pools of fungi. *Bioresour. Technol.* **2017**, *244*, 1089–1095. [CrossRef] [PubMed]
20. Frigaard, N. Sugar and Sugar Alcohol Production. In *Genetically Engineered Foods*; Grumezescu, A., Butu, A., Eds.; Elsevier: Helsingor, Denmark, 2018; pp. 31–47. ISBN 9780128115190.
21. Hernández-Pérez, A.F.; Machado, F.; De Souza Queiroz, S.; Vaz De Arruda, P.; Chandel, A.K.; Das Graças, M.D.A. Biotechnological production of sweeteners. In *Biotechnological Production of Bioactive Compounds*; Elsevier: Amsterdam, The Netherlands, 2019; pp. 261–292. ISBN 9780444643230.
22. Chan, E.C.S. Microbial nutrition and basic metabolism. In *Handbook of Water and Wastewater Microbiology*; Mara, D., Horan, N., Eds.; Elsevier: London, UK, 2003; pp. 3–33. ISBN 9780080478197.
23. Devanthi, P.V.P.; Gkatzionis, K. Soy sauce fermentation: Microorganisms, aroma formation, and process modification. *Food Res. Int.* **2019**, *120*, 364–374. [CrossRef]
24. Reihani, S.F.S.; Khosravi-Darani, K. Influencing factors on single-cell protein production by submerged fermentation: A review. *Electron. J. Biotechnol.* **2019**, *37*, 34–40. [CrossRef]
25. Ren, H.Y.; Kong, F.; Ma, J.; Zhao, L.; Xie, G.J.; Xing, D.; Guo, W.Q.; Liu, B.F.; Ren, N.Q. Continuous energy recovery and nutrients removal from molasses wastewater by synergistic system of dark fermentation and algal culture under various fermentation types. *Bioresour. Technol.* **2018**, *252*, 110–117. [CrossRef] [PubMed]
26. He, L.; Lv, H.; Xing, Y.; Wang, C.; You, X.; Chen, X.; Zhang, Q. The nutrients in Moringa oleifera leaf contribute to the improvement of stylo and alfalfa silage: Fermentation, nutrition and bacterial community. *Bioresour. Technol.* **2020**, *301*, 122733. [CrossRef]
27. Zhang, Y.; Meng, L.; Ai, M.; Qiao, Y.; Liu, G.; Fan, X.; Lv, X.; Feng, Z. Nutrient requirements of Lactobacillus casei Shirota and their application in fermented milk. *LWT* **2020**, *118*, 108735. [CrossRef]
28. Koh, E.S.; Lee, T.H.; Lee, D.Y.; Kim, H.J.; Ryu, Y.W.; Seo, J.H. Scale-up of erythritol production by an osmophilic mutant of Candida magnoliae. *Biotechnol. Lett.* **2003**, *25*, 2103–2105. [CrossRef] [PubMed]
29. Jeya, M.; Lee, K.M.; Tiwari, M.K.; Kim, J.S.; Gunasekaran, P.; Kim, S.Y.; Kim, I.W.; Lee, J.K. Isolation of a novel high erythritol-producing Pseudozyma tsukubaensis and scale-up of erythritol fermentation to industrial level. *Appl. Microbiol. Biotechnol.* **2009**, *83*, 225–231. [CrossRef] [PubMed]
30. Saran, S.; Mukherjee, S.; Dalal, J.; Saxena, R.K. High production of erythritol from Candida sorbosivorans SSE-24 and its inhibitory effect on biofilm formation of Streptococcus mutans. *Bioresour. Technol.* **2015**, *198*, 31–38. [CrossRef] [PubMed]
31. Mirończuk, A.M.; Rakicka, M.; Biegalska, A.; Rymowicz, W.; Dobrowolski, A. A two-stage fermentation process of erythritol production by yeast Y. lipolytica from molasses and glycerol. *Bioresour. Technol.* **2015**, *198*, 445–455. [CrossRef] [PubMed]
32. Rakicka, M.; Rywińska, A.; Lazar, Z.; Rymowicz, W. Two-stage continuous culture—Technology boosting erythritol production. *J. Clean. Prod.* **2017**, *168*, 420–427. [CrossRef]
33. Mirończuk, A.M.; Furgała, J.; Rakicka, M.; Rymowicz, W. Enhanced production of erythritol by Yarrowia lipolytica on glycerol in repeated batch cultures. *J. Ind. Microbiol. Biotechnol.* **2014**, *41*, 57–64. [CrossRef] [PubMed]
34. Daza-Serna, L.; Serna-Loaiza, S.; Masi, A.; Mach, R.L.; Mach-Aigner, A.R.; Friedl, A. From the culture broth to the erythritol crystals: An opportunity for circular economy. *Appl. Microbiol. Biotechnol.* **2021**, *105*, 4467–4486. [CrossRef] [PubMed]
35. Ward, A.J.; Arola, K.; Thompson Brewster, E.; Mehta, C.M.; Batstone, D.J. Nutrient recovery from wastewater through pilot scale electrodialysis. *Water Res.* **2018**, *135*, 57–65. [CrossRef]
36. European Commission. *Recovering Resources from Industrial Wastewater to Minimise Environmental Impact*; Zero Brine Project; Cordis EU Research Results; European Commission: Brussel, Belgium, 2020.
37. European Commission. *Wastewater Treatment Progresses towards a Circular Economy*; Incover Project; European Commission: Brussel, Belgium, 2019.
38. European Commission. *Unlocking New Value from Urban bioWASTE*; Value Waste Project; European Commission: Brussel, Belgium, 2018.
39. VCM. Nitroman Project. Interreg EU Project. 2019. Available online: https://www.vcm-mestverwerking.be/nl/kenniscentrum/20262/nitroman (accessed on 16 May 2021).
40. Ding, Y.; Sartaj, M. Optimization of ammonia removal by ion-exchange resin using response surface methodology. *Int. J. Environ. Sci. Technol.* **2016**, *13*, 985–994. [CrossRef]
41. Richardson, J.; Harker, J.; Backhurst, J. Ion Exchange. In *Coulson and Richarson's Chemical Engineering by Coulson & Richardson*; Butterworth Heinemann: Oxford, UK, 2002; pp. 1053–1075.
42. Xie, M.; Shon, H.K.; Gray, S.R.; Elimelech, M. Membrane-based processes for wastewater nutrient recovery: Technology, challenges, and future direction. *Water Res.* **2016**, *89*, 210–221. [CrossRef] [PubMed]

43. Prochaska, K.; Staszak, K.; Woźniak-Budych, M.J.; Regel-Rosocka, M.; Adamczak, M.; Wiśniewski, M.; Staniewski, J. Nanofiltration, bipolar electrodialysis and reactive extraction hybrid system for separation of fumaric acid from fermentation broth. *Bioresour. Technol.* **2014**, *167*, 219–225. [CrossRef] [PubMed]
44. Shen, J.; Duan, J.; Liu, Y.; Yu, L.; Xing, X. Demineralization of glutamine fermentation broth by electrodialysis. *Desalination* **2005**, *172*, 129–135. [CrossRef]
45. Phanthumchinda, N.; Thitiprasert, S.; Tanasupawat, S.; Assabumrungrat, S.; Thongchul, N. Process and cost modeling of lactic acid recovery from fermentation broths by membrane-based process. *Process Biochem.* **2018**, *68*, 205–213. [CrossRef]
46. Cheryan, M.; Parekh, S.R. Separation of glycerol and organic acids in model ethanol stillage by electrodialysis and precipitation. *Process Biochem.* **1995**, *30*, 17–23. [CrossRef]
47. Wu, R.C.; Xu, Y.Z.; Song, Y.Q.; Luo, J.A.; Liu, D. A novel strategy for salts recovery from 1,3-propanediol fermentation broth by bipolar membrane electrodialysis. *Sep. Purif. Technol.* **2011**, *83*, 9–14. [CrossRef]
48. Sun, Z.; Gao, X.; Zhang, Y.; Gao, C. Separation and purification of L-phenylalanine from the fermentation broth by electrodialysis. *Desalin. Water Treat.* **2016**, *57*, 22304–22310. [CrossRef]
49. Hboová, V.; Melzoch, K.; Rychtera, M.; Sekavová, B. Electrodialysis as a useful technique for lactic acid separation from a model solution and a fermentation broth. *Desalination* **2004**, *162*, 361–372. [CrossRef]
50. Vadthya, P.; Kumari, A.; Sumana, C.; Sridhar, S. Electrodialysis aided desalination of crude glycerol in the production of biodiesel from oil feed stock. *Desalination* **2015**, *362*, 133–140. [CrossRef]
51. Shen, J.Y.; Duan, J.R.; Yu, L.X.; Xing, X.H.; Xu, P. Desalination of glutamine fermentation broth by electrodialysis. *Process Biochem.* **2006**, *41*, 716–720. [CrossRef]
52. Wu, R.C.; Ren, H.J.; Xu, Y.Z.; Liu, D. The final recover of salt from 1,3-propanadiol fermentation broth. *Sep. Purif. Technol.* **2010**, *73*, 122–125. [CrossRef]
53. Kresnowati, M.T.A.P.; Regina, D.; Bella, C.; Wardani, A.K.; Wenten, I.G. Combined ultrafiltration and electrodeionization techniques for microbial xylitol purification. *Food Bioprod. Process.* **2019**, *114*, 245–252. [CrossRef]
54. Scarazzato, T.; Panossian, Z.; Tenório, J.A.S.; Pérez-Herranz, V.; Espinosa, D.C.R. A review of cleaner production in electroplating industries using electrodialysis. *J. Clean. Prod.* **2017**, *168*, 1590–1602. [CrossRef]
55. *ISO 3696:1987*; ISO Water for Analytical Laboratory Use; Specification and Test Methods; International Organization for Standardization: Geneva, Switzerland, 1991.
56. National Center for Biotechnology Information. *PubChem Compound Summary for CID 222285, Erythritol*; National Center for Biotechnology Information: Bethesda, MD, USA, 2021.
57. National Center for Biotechnology Information. *PubChem Compound Summary for CID 753, Glycerol*; National Center for Biotechnology Information: Bethesda, MD, USA, 2021.
58. Wang, T.; Meng, Y.; Qin, Y.; Feng, W.; Wang, C. Removal of furfural and HMF from monosaccharides by nanofiltration and reverse osmosis membranes. *J. Energy Inst.* **2018**, *91*, 473–480. [CrossRef]
59. Cowan, D.A.; Brown, J.H. Effect of Turbulence on Limiting Current in Electrodialysis Cells. *Ind. Eng. Chem.* **1959**, *51*, 1445–1448. [CrossRef]
60. Lindemann, M.; Widhalm, B.; Kuncinger, T.; Srebotnik, E. An integrated process for combined microbial VOC reduction and effluent valorization in the wood processing industry. *Bioresour. Technol. Rep.* **2020**, *11*, 100471. [CrossRef]
61. Chaabouni, A.; Guesmi, F.; Louati, I.; Hannachi, C.; Hamrouni, B. Temperature effect on ion exchange equilibrium between CMX membrane and electrolyte solutions. *Water Reuse Desalin.* **2015**, *4*, 535–541. [CrossRef]
62. Rakicka, M.; Rukowicz, B.; Rywińska, A.; Lazar, Z.; Rymowicz, W. Technology of efficient continuous erythritol production from glycerol. *J. Clean. Prod.* **2016**, *139*, 905–913. [CrossRef]
63. Nichka, V.S.; Geoffroy, T.R.; Nikonenko, V.; Bazinet, L. Impacts of flow rate and pulsed electric field current mode on protein fouling formation during bipolar membrane electroacidification of skim milk. *Membranes* **2020**, *10*, 200. [CrossRef]
64. Ruiz, B.; Sistat, P.; Huguet, P.; Pourcelly, G.; Araya-Farias, M.; Bazinet, L. Application of relaxation periods during electrodialysis of a casein solution: Impact on anion-exchange membrane fouling. *J. Memb. Sci.* **2007**, *287*, 41–50. [CrossRef]
65. Gohil, G.S.; Nagarale, R.K.; Shahi, V.K.; Rangarajan, R. Micellar-enhanced electrodialysis: Influence of surfactants on the transport properties of ion-exchange membranes. *Sep. Purif. Technol.* **2005**, *47*, 1–9. [CrossRef]
66. Der Pan, W.; Chiang, B.; Chiang, P. Desalination of the Spent Brine from Pickled Prunes Processing by Electrodialysis. *J. Food Sci.* **1988**, *53*, 134–137. [CrossRef]
67. Luiz, A.; McClure, D.D.; Lim, K.; Leslie, G.; Coster, H.G.L.; Barton, G.W.; Kavanagh, J.M. Potential upgrading of bio-refinery streams by electrodialysis. *Desalination* **2017**, *415*, 20–28. [CrossRef]
68. Sadrzadeh, M.; Mohammadi, T. Treatment of sea water using electrodialysis: Current efficiency evaluation. *Desalination* **2009**, *249*, 279–285. [CrossRef]
69. Sun, B.; Zhang, M.; Huang, S.; Wang, J.; Zhang, X. Limiting concentration during batch electrodialysis process for concentrating high salinity solutions: A theoretical and experimental study. *Desalination* **2021**, *498*, 114793. [CrossRef]
70. Luo, G.S.; Pan, S.; Liu, J.G. Use of the electrodialysis process to concentrate a formic acid solution. *Desalination* **2002**, *150*, 227–234. [CrossRef]
71. Doornbusch, G.; Swart, H.; Tedesco, M.; Post, J.; Borneman, Z.; Nijmeijer, K. Current utilization in electrodialysis: Electrode segmentation as alternative for multistaging. *Desalination* **2020**, *480*, 114243. [CrossRef]

72. Han, L.; Galier, S.; Roux-de Balmann, H. Ion hydration number and electro-osmosis during electrodialysis of mixed salt solution. *Desalination* **2015**, *373*, 38–46. [CrossRef]
73. Luo, T.; Roghmans, F.; Wessling, M. Ion mobility and partition determine the counter-ion selectivity of ion exchange membranes. *J. Memb. Sci.* **2020**, *597*, 117645. [CrossRef]
74. Luo, T.; Abdu, S.; Wessling, M. Selectivity of ion exchange membranes: A review. *J. Memb. Sci.* **2018**, *555*, 429–454. [CrossRef]
75. Sata, T.; Sata, T.; Yang, W. Studies on cation-exchange membranes having permselectivity between cations in electrodialysis. *J. Memb. Sci.* **2002**, *206*, 31–60. [CrossRef]
76. Sata, T. Studies on anion exchange membranes having permselectivity for specific anions in electrodialysis—Effect of hydrophilicity of anion exchange membranes on permselectivity of anions. *J. Memb. Sci.* **2000**, *167*, 1–31. [CrossRef]
77. Stenina, I.; Golubenko, D.; Nikonenko, V.; Yaroslavtsev, A. Selectivity of transport processes in ion-exchange membranes: Relationship with the structure and methods for its improvement. *Int. J. Mol. Sci.* **2020**, *21*, 5517. [CrossRef]
78. Mikhaylin, S.; Bazinet, L. Fouling on ion-exchange membranes: Classification, characterization and strategies of prevention and control. *Adv. Colloid Interface Sci.* **2016**, *229*, 34–56. [CrossRef]
79. Kooistra, W. Characterization of ion exchange membranes by polarization curves. *Desalination* **1967**, *2*, 139–147. [CrossRef]