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Analysis of the desalination process for the biological production of erythritol

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ABSTRACT

The interest and consumption of healthy sugar substitutes is increasing. Among these compounds is erythritol, which is obtained in a biotechnological process. An important step in the biotechnological process is product separation and purification. By fermentation, the conversion of renewable raw materials into erythritol is carried out and a fermentation broth is obtained, which is a solution that requires desalination and purification. The main challenge during the downstream stage is the separation of inorganic salts, mainly sodium chloride, which increases the fermentation yield but becomes a contaminant after the upstream stage. This paper presents ion exclusion as a method of desalting an erythritol solution using ion exchange resin in sodium form. The presented experimental studies and the mathematical model indicate the possibility of increasing efficiency of the process in a multistage system and the possibility of selecting parameters based on process simulations. As a result, it is possible to reduce losses and increase the purity of the product.

Keywords: Erythritol; Bioconversion; Desalination; Preparative liquid chromatography

1. Introduction

Biotechnological processes and the conversion of renewable energy sources (biomass) are increasingly used in industry and are being developed to reduce the share of fossil fuels in the production of chemical compounds [1,2]. An important class of chemicals produced by microorganisms that is being studied includes C4 compounds [3]. Among them, erythritol (butane-1,2,3,4-tetrol) is a four-carbon sugar alcohol that naturally occurs in fruits and fermented food products [4,5]. The low calorific value, low glycemic index, and low chemical reactivity, as well as sweetening properties (70%–80% of sucrose) make it widely used as a sweetener in the food and pharmaceutical industries, especially for diabetics [6,7]. Research on polyol production methods includes chemical synthesis, as well as biotechnological methods. Erythritol is a polyol that is commercially obtained mainly through fermentation processes [8,9]. Both the selection of the appropriate substrate and the selective strain of the microorganism affect the efficiency and cost of biosynthesis. Erythritol can be produced by osmophilic yeast such as *Yarrowia lipolytica*, *Moniliella megachiliensis*, *Candida magnoliae*, *Trichosporonoides oedocephalis* [10–12]. Erythritol bioproduction is most commonly performed with *Y. lipolytica*, which is a widely distributed strain in nature and has the ability to convert substrates such as glucose [13], crude glycerol [14], xylose [15], raw molasses, vegetable oil and waste cooking oil [16–18]. The appropriate selection of parameters, including osmotic pressure, type of substrate, pH, and temperature of the medium, affects erythritol biosynthesis, its concentration, and the final composition of the fermentation broth (including side metabolites) [19,20]. The separation of

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erythritol from the crude fermentation broth is required to obtain a pure product. Reducing the formation of by-products (e.g., citric acid, α-ketoglutaric acid, arabitol, mannitol) facilitates the purification step [21]. Many studies indicate a significant influence of osmotic pressure on the increase in erythritol production. Higher osmotic pressure is achieved by introducing an osmotic stress agent (usually sodium chloride) into the medium [22,23]. For example, the addition of salt (NaCl 10–30 g/L) to the medium not only increases the production of erythritol by *Y. lipolytica*, but also reduces the formation of by-products (mannitol and arabitol) [24]. The sodium chloride used is not converted by microorganisms, and its separation from the bioconversion product is necessary. Both upstream processing and purification of the biosynthetic product affect the quality of the product and its production costs. Downstream processing consists of the separation of biomass, residual substrate and by-products, decolorization, desalination, and crystallization [25]. Separation of impurities can be performed using chromatographic techniques. Liquid chromatography is used both at the analytical and production scale. Preparative chromatography aims to purify and isolate the product with high yield and high purity. Ion exclusion chromatography can be applied for the separation of ionic and non-ionic substances and has been used to efficiently desalinate the erythritol solution. Chromatographic separation of sodium chloride from erythritol was carried out on a cation exchange resin (charge of the ion exchanger, such as charge of ionic substances). Consequently, ionic compounds are excluded from the support pores and elute first, while the non-ionic compounds are eluted later [26].

In preparative chromatography, over-extended mass (or volume) is used to increase the productivity. The overload increases productivity while reducing the purity of the product. Selection of the best separation conditions allows to obtain a pure product in high yield, which may affect the final cost of the process [27]. The possibility of simulating the process with a correct mathematical model reduces the time of separation analysis and selection of the best parameters, as well as the costs associated with experimental tests.

The aim of this work is to analyze the possibility of the separation of erythritol from fermentation broths using preparative chromatography. The issue of bioconversion towards erythritol is very well described in the literature and indicates the possibility of obtaining a product with a high concentration. The problem that limits the implementation of technology in the industry is the efficiency of the separation method. In addition to the aspect of purity of the final product, the costs of the process and the possibility of minimizing waste fractions are also important. Process optimization is possible by selecting the appropriate mathematical model and verifying the results through both experimental and simulation tests.

The paper presents the results of experimental erythritol desalination by ion exclusion chromatography and process simulation using a mathematical model. In the process analysis, sodium chloride was selected as the salt, the amount of which is the highest impurity. Additionally, in conjunction with the separation of sodium chloride, the separation of other inorganic salts present in a small amount occurs.

2. Mathematical model of column chromatography

The number of experimental tests can be limited by simulating the concentration profiles of the separated components. In the mathematical description of the chromatographic process, mass transfer phenomena and thermodynamic equilibrium should be taken into account. As a result, a mathematical model of the chromatography column is obtained. The mathematical models of the chromatographic column are classified into heterogeneous, pseudo-homogeneous, and homogeneous, depending on the assumptions and simplifications. General rate model (GR) and lumped pore diffusion model (POR) take into account two mass-balance equations for the solute in the mobile phase and in the stationary phase [28]. In practice, these heterogeneous complex models are used mainly when consideration of high mass transfer resistance is necessary, for example due to the large particle size of the separated compounds [29,30]. In the case of aqueous solutions of alcohols, polyols, and carboxylic acids, the equilibrium dispersive model is often useful in the practical application and simulation of the chromatographic process.

In this work, the equilibrium dispersive model (ED) has been used for simulation of column dynamics:

$$\frac{\partial C_i}{\partial t}+\frac{u}{\varepsilon_t}\frac{\partial C_i}{\partial x}+\frac{\left(1-\varepsilon_t\right)}{\varepsilon_t}\frac{\partial q_i}{\partial t}=D_a\frac{\partial^2 C_i}{\partial x^2} \tag{1}$$

where C_i and q_i are the concentration of solutes in mobile and stationary phases, respectively, ε_t is the total porosity, u is the linear velocity, D_a is the apparent dispersion coefficient, t is the time and x is the distance along the column. The values of C_i and q_i are correlated by the adsorption isotherm equation [31]. The ED model is complemented by appropriate initial and boundary conditions.

Initial conditions: for $t = 0$ ($0 < x < L$, where L is the length of the column), the column is filled with a pure mobile phase and the initial concentrations are as follows:

$$C_i\left(0,x\right)=0 \tag{2}$$

$$q_i\left(0,x\right)=0 \tag{3}$$

At the initial time ($t = 0$ and $0 < x < L$) the concentration substances in the mobile phase and in the stationary phase are equal to zero, while between the mobile and stationary phases is a state of equilibrium.

Boundary conditions at the inlet and outlet of the column are described by Danckwerts-type boundary conditions, according to which the phenomena of dispersion occur only inside chromatographic column:

- at the column inlet ($t > 0$, $x = 0$):

$$u\left(C_{0i}\left(t\right)-C_i\left(t,0\right)\right)=-\varepsilon_t D_a\frac{\partial C_i\left(t,0\right)}{\partial x} \tag{4}$$

- at the column outlet ($t > 0$, $x = L$):

$$\frac{\partial C_i\left(t,L\right)}{\partial x}=0 \tag{5}$$

where C_{0i} is the injection concentration of the sample [32].

The ED model can be used for products with low molecular weights and when the mass transfer kinetics is fast but not infinitely fast. This model is very often used to design and optimize with high accuracy, especially for column efficiency (number of plates) greater than 100 [30].

3. Materials and methods

3.1. Materials

Sodium chloride (99.8%, salt) was purchased from Avantor Corporation (Poland). Erythritol (food product) was purchased from Intenson Corporation (Poland). Sulfuric acid (Chempur, Poland), citric acid (Sigma-Aldrich, Poland), and d-mannitol (Sigma-Aldrich, Poland) were used in quantitative analysis. Chromatographic separation experiments were carried out using monospheric ion exchange resins based on a styrene-divinylbenzene copolymer, strongly acidic cation exchange in sodium form: Lewatit S1567, 0.6 mm and Amberjet 1200Na, 0.6 mm. Fermentation broth obtained by bioconversion of glycerol was used in the research. The fermentation process and the composition of the fermentation broth were presented in the previous work [26]. The main impurity of the fermentation broth was sodium chloride added at the fermentation stage and not metabolized by yeast.

3.2. Analytical methods

Concentrations of erythritol, d-mannitol, citric acid, and inorganic salts were measured by high performance liquid chromatography with a refractive index detector and a Rezex ROA organic acid column (4.6 mm × 250 mm, Phenomenex). The flow rate of the mobile phase (5 mM H_2SO_4) was maintained at 0.4 mL/min with a column temperature of 50°C. During the process, the conductivity of the collected eluate (Cond) was analyzed online using a multifunction meter (Elmetron) measuring the pH, temperature, and conductivity of working solutions. In addition, when the sorption isotherm was determined, the refractive index was measured with a refractometer.

3.3. Experimental studies

3.3.1. Ion exclusion chromatography

The preparative chromatography system included a chromatographic column (volume 0.53 L with bed height of 100 cm), a peristaltic pump, a multifunction meter, and a fraction collector. Experimental studies were carried out for aqueous model solutions and fermentation broths dosed onto the packed column in a amount of 10%–60% of the total bed volume. Distilled water was used as a mobile phase with a space velocity of 1–1.2 h^{-1} (ratio of the flow rate of the mobile phase to the total bed volume). Studies have been carried out on the effect of volume overload on process efficiency at room temperature (20°C) and the separation of fermentation broth was carried out at a temperature of 50°C. The fractions of the column were collected and analyzed using high-performance liquid chromatography (HPLC). Chromatograms of erythritol and salt were determined on quantitative analysis. The results are plotted on a dimensionless scale to compare the tests performed for different separation conditions and for the results obtained from computer simulation. The dimensionless form of the total volume of mobile phase dispensed onto the column (V) with respect to the volume of the column bed (VB) was defined as the ratio of the volume of the eluate to the bed volume (V/VB, where VB = 530 mL). Dimensional-free data were also presented for the concentrations of components determined by HPLC by determining the ratio of the concentration of the component in the tested sample to its initial concentration in the feed (C/C_0, where C_0 is the initial concentration of the component in the feed solution).

The research presented in this work was focused on the downstream stage, which, when analyzing the composition of the broth, requires, first of all, the separation of inorganic salts (mainly sodium chloride) and carboxylic acids such as citric acid (whose elution profile is the same as for sodium chloride – according to curves shown in the paper in Fig. 13).

3.3.2. ED model parameters

The total porosity is calculated according to equation [30]:

$$\varepsilon_t = \frac{t_{r0} \times u}{L} \tag{6}$$

where t_{r0} is the column dead time $\varepsilon_t V_B / \dot{V}$ and L is length of the column. In practice, the total porosity is determined by means of not retained, pore-penetrating tracer substance. A sodium chloride solution was used as an inert substance. The bed porosity was measured in a small column (7 mm ID) with different length (10–20 cm).

The number of theoretical plates N (column efficiency) was experimentally determined on the basis of the retention time t_r of erythritol (10 g/L aqueous solution) and the half height peak width $w_{1/2}$ according to the equation:

$$N = 5.54 \frac{t_r}{w_{1/2}} \tag{7}$$

The apparent dispersion coefficient D_a is related to the column efficiency:

$$D_a = \frac{\text{HETP} \times u}{2\varepsilon_t} \tag{8}$$

where HETP is the height equivalent to a theoretical plate (L/N) [33,34]. Experimentally determined parameters are summarized in Table 1.

Table 1
Model parameters Lewatit S1567 Na^+

Experimental value	Lewatit S1567 Na^+, 0.6 mm
ε_t (–)	0.41 ± 0.01
HETP (cm)	0.92 ± 0.01
K (–) for $q^* = KC^*$	0.327

Notes: Total porosity ε_t, the height equivalent to a theoretical plate HETP and linear isotherm constant K are presented in Table 1.

3.3.3. Adsorption equilibrium

The adsorption isotherm is an equation related to the amount of a substance adsorbed on a sorbent and the equilibrium concentration of a solute in a solution at a given temperature. Two different methods of isotherm determination were tested: the batch static method and the adsorption–desorption method [30]. Both methods are based on mass balance and in both cases the tests were carried out at room temperature. In the static method, beakers containing a known amount of sorbent and a solution were shaken until they were equilibrated. Preliminary studies indicated that better results are obtained with the adsorption–desorption method compared to the static method. Phase equilibrium data were obtained using the adsorption–desorption method. In this method, a column with bed volume V_B (2 mL) is equilibrated with a solution of known concentration of the component by continuously dosing the feed (C_i of erythritol in the range of 10–100 g/L). In the next step, desorption with deionized water has been carried out until the eluate is pure water, and the collected eluate is analyzed to obtain the mass of desorbed component *m* [30]:

$$m = \varepsilon_t \cdot V_B \cdot C_F^m + \left(1 - \varepsilon_t\right) \cdot V_B \cdot q^*\left(C_F^m\right) \tag{9}$$

where C_F^m is the feed concentration and in this case is also the equilibrium concentration in the mobile phase.

3.4. Computational studies

The simulation elution profiles were determined on the basis of the ED model supplemented with experimentally determined parameters. Simulation studies were performed using Chromatographic Column Software (created by Krzysztof Kaczmarski, Rzeszow University of Technology, Department of Chemical Engineering).

3.5. Parameters of mathematical modeling of preparative chromatography

The model parameters and the adsorption isotherm for the Lewatit S1567 resin were determined. Adsorption equilibrium for erythritol was measured using the adsorption–desorption method. The sodium chloride is defined as an inert compound and the isotherm parameters were determined only for erythritol. Experimental studies were carried out on three columns with a volume of 2 mL, obtaining repeatable results for all. In each case, equilibrium was achieved very quickly and linear curves for erythritol were obtained. On the basis of the experimental curve, the constant for the linear isotherm model was determined (Fig. 1).

4. Results

On the basis of the experimental tests performed, the concentration profiles for model solutions containing erythritol and sodium chloride were determined. Studies were carried out on the column with a volume of 530 mL (column diameter 2.6 cm, bed height 100 cm) at room temperature. In the next step, simulation tests were carried out for the parameters used in the experimental studies. The ED model was used supplemented with experimentally determined model constants.

Studies on the effect of volume overload on process efficiency have been described in the previous article [35]. The feed solution was composed of 150 g/L erythritol and 30 g/L sodium chloride (4% of the salt content in the product – 96% purity after the desalination of the aqueous solution was assumed). Chromatographic separation experiments were carried out using a strongly acidic cation exchange resin in sodium form (based on a styrene-divinylbenzene copolymer, Lewatit S1567, 0.6 mm). For low overload (10%–20%) a very high dilution of the erythritol solution was obtained. At 50%–60% column loading, the smallest degree of product dilution (approximately 2 times relative to the feed) and the lowest separation efficiency (32%–38%) were obtained. It was found that the separation efficiency decreases with volume overloading and the process carried out for a 30%–40% column load allows the highest possible separation efficiency. The data obtained were compared with the concentration profiles determined in the chromatographic column program. An exemplary comparison (for 60% column loading) of the computer simulations with experimental profiles for erythritol (Ery) and sodium chloride (Cond) is presented in Fig. 2.

Model concentration profiles using the ED model and determined the experimental parameters are similar to the experimental profiles (Fig. 2). The obtained results are similar with regard to both the elution time of the components and the shape and width of the chromatographic peaks.

The single stage batch ion exclusion was carried out according to the scheme presented in Fig. 3. The feed (erythritol solution for purification) was loaded into the chromatographic column in the amount of 10%–60% of bed volume and then the water (deionized water) was continuously dosed. The eluate was collected as three fractions: saline solution (waste), sodium chloride, and erythritol solution (recycle fraction), and purified erythritol solution (product). The collection of the above fractions was determined on the elution profiles.

In the next step, a multistage batch preparative chromatography system was applied. The feed composition was

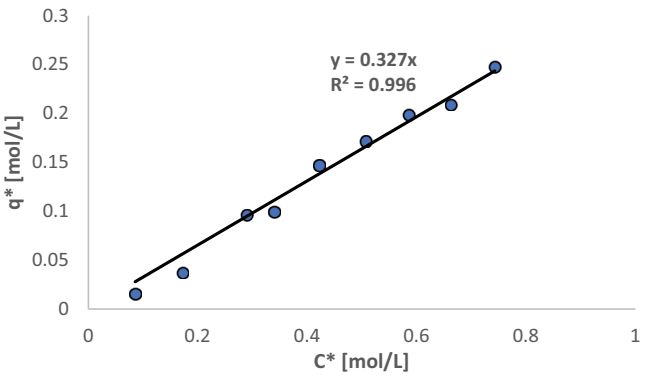

Fig. 1. Erythritol isotherm on Lewatit S1567 at room temperature (21°C). *C** equilibrium concentration of erythritol in the mobile phase, *q** equilibrium concentration of erythritol in the stationary phase (symbols-experimental data, solid line-fitting curve).

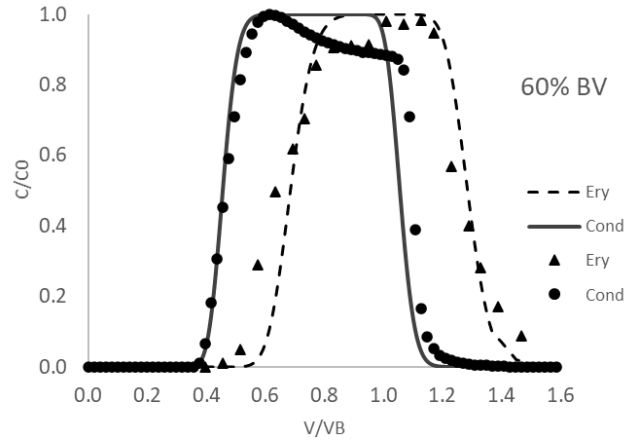

Fig. 2. Comparison of the experimental (symbols) and simulated (lines) elution profiles of 1.23 mol/L erythritol (Ery) and 0.51 mol/L sodium chloride (Cond) for 0.53 L column packed with Lewatit 0.6 mm (space velocity 1.2 h^{-1}), feed solution 318 mL (60% bed volume). *V*/VB – eluent volume ratio, where *V* is the total volume of the mobile phase dispensed onto the column, and VB is the volume of the column with bed; C/C_0 – component concentration ratio, where *C* is the component concentration in the tested sample and C_0 is the initial concentration of the component in the feed solution).

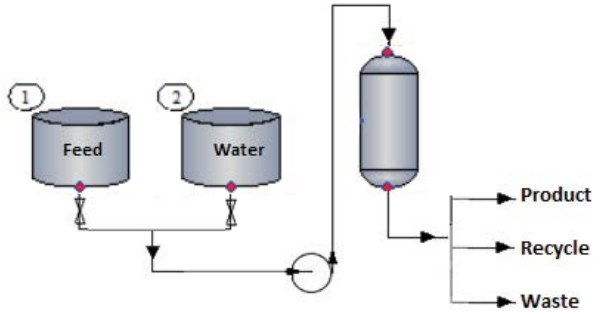

Fig. 3. Diagram of single-stage batch preparative chromatography.

the same as in a single separation system (150 g/L erythritol and 30 g/L sodium chloride). The feed was loaded into the first column in the amount of 34% bed volume (180 mL) and then water was continuously dosed. The recycle (93 mL) of the first column was mixed with feed (87 mL) and purified in the second column. The process was repeated for two consecutive columns as shown in Fig. 4.

The results obtained for the single-column system presented in Fig. 5 indicate that the simulation profiles are similar to the experimental profiles. The arrows in the diagram are the collection points of the subsequent eluate fractions (waste, recycle, and product).

Experimental data for the second, third and fourth columns were also compared with the profiles obtained from

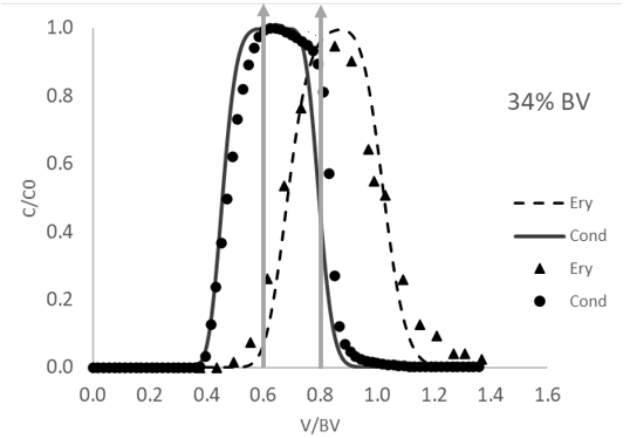

Fig. 5. Comparison of the experimental (symbols) and simulated (lines) elution profiles of 1.23 mol/L erythritol (Ery) and 0.51 mol/L sodium chloride (Cond) for a single-stage system (space velocity 1.2 h^{-1}), feed solution 180 mL (34% bed volume). *V*/BV – eluent volume ratio, where *V* is the total volume of the mobile phase dispensed onto the column, and BV is the volume of the column with bed; C/C_0 – component concentration ratio, where *C* is the component concentration in the tested sample and C_0 is the initial concentration of the component in the feed solution).

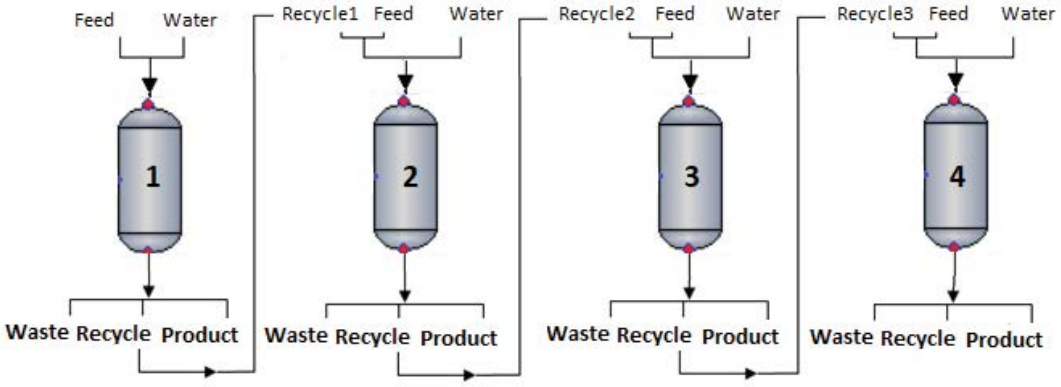

Fig. 4. Preparative chromatography cyclic process (recirculation with the feed reduces the impact of reducing the amount of feed to be processed in the second and third columns, while reducing the amount of polyol losses present in the recycling); optimal collection time for recycle: from 0.6 to 0.8 *V*/BV (106 mL with optimal rate 1.2 h^{-1}) and for product: from 0.8 to 1.1 *V*/BV (159 mL with optimal rate 1.2 h^{-1}).

mathematical modeling (Figs. 6–8). The erythritol content in the recycle was lower than in the feed, with the result that the solution passed to the next column was variable, with a lower concentration of erythritol.

Analyzing the obtained results, the loss of erythritol in the waste fraction was determined at a level of 4% relative to the amount dosed to the column. The amount of erythritol in the recycled fraction was 30%. Therefore, the single-column process is associated with a large loss of product in both fractions (waste and recycle). Desalination of the recycling fraction makes it possible to reduce product losses and consequently increase the efficiency of the process. However, this value is limited due to the decrease in erythritol concentration during recycling to the next steps. Process simulation based on mathematical modeling is a useful tool for the analysis and selection of optimal separation conditions.

As can be seen in Fig. 9, compared to a single system, the highest increase in efficiency is observed for the two-stage desalination process. Yield is defined as the ratio of the amount of erythritol in the product fraction to the total amount of erythritol dosed to the column. The recycle fraction can be purified in a four-column system. The successive steps of separation do not significantly increase the recovery of erythritol. Additionally, the proposed system of mixing recycle with fermentation broth allows to obtain a high

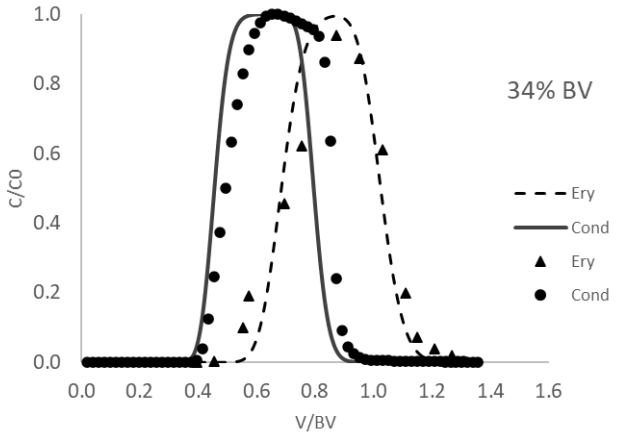

Fig. 6. Comparison of the experimental (symbols) and simulated (lines) elution profiles of 0.97 mol/L erythritol (Ery) and 0.55 mol/L sodium chloride (Cond) for a two-stage system (space velocity 1.2 h^{-1}), feed solution 180 mL (34% bed volume). *V*/BV – eluent volume ratio, where *V* is the total volume of the mobile phase dispensed onto the column, and BV is the volume of the column with bed; C/C_0 – component concentration ratio, where *C* is the component concentration in the tested sample and C_0 is the initial concentration of the component in the feed solution).

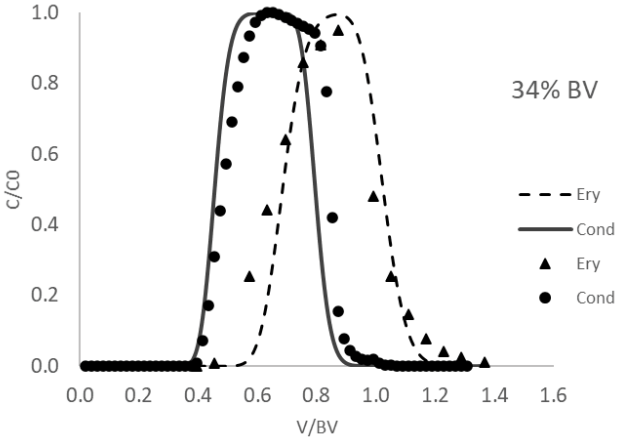

Fig. 7. Comparison of the experimental (symbols) and simulated (lines) elution profiles of 0.89 mol/L erythritol (Ery) and 0.55 mol/L sodium chloride (Cond) for a three-stage system (space velocity 1.2 h^{-1}), feed solution 180 mL (34% bed volume). *V*/BV – eluent volume ratio, where *V* is the total volume of the mobile phase dispensed onto the column, and BV is the volume of the column with bed; C/C_0 – component concentration ratio, where *C* is the component concentration in the tested sample and C_0 is the initial concentration of the component in the feed solution).

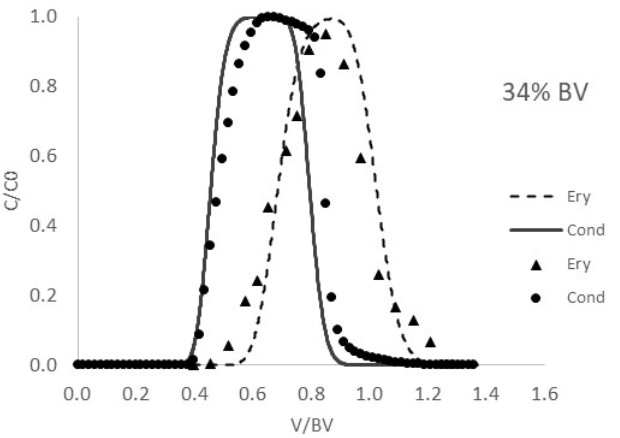

Fig. 8. Comparison of the experimental (symbols) and simulated (lines) elution profiles of 0.86 mol/L erythritol (Ery) and 0.55 mol/L sodium chloride (Cond) for a four-stage system (space velocity 1.2 h^{-1}), feed solution 180 mL (34% bed volume). *V*/BV – eluent volume ratio, where *V* is the total volume of the mobile phase dispensed onto the column, and BV is the volume of the column with bed; C/C_0 – component concentration ratio, where *C* is the component concentration in the tested sample and C_0 is the initial concentration of the component in the feed solution).

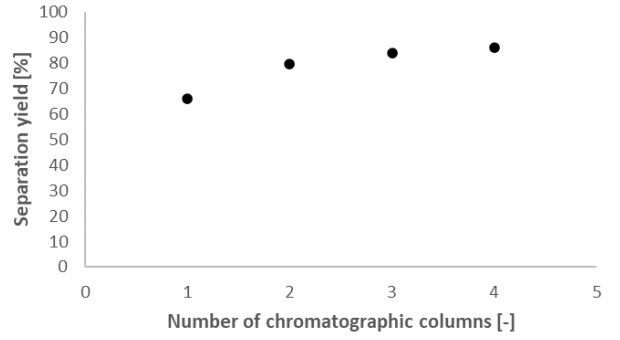

Fig. 9. Effect of the number of columns on erythritol separation yield (34% column loading, volume 0.53 L).

degree of purification without further dilution of the product fraction. The separation process is carried out under steady-state conditions (constant volume and composition of the feed). The mathematical model allows us to choose the conditions of separation, which have been verified experimentally. In Rakicka-Pustułka et al. [36] presents a process based on a 5-step system, in which only recycling is subject to separation. Based on the results presented in this paper, it can be concluded that a better solution is to use a smaller number of separation columns with partial recycling in the feed.

The results for erythritol (Fig. 9) correspond to the curve obtained for another polyol. For example, Fig. 10 shows the simulation data for the ion exclusion of propane-1,3-diol carried out on a 10.5 L column. The higher efficiency of propane-1,3-diol is related, among others, to the higher affinity of propane-1,3-diol for the resin, resulting in a longer retention time.

The use of the mathematical simulation of the process allows to limit the number of experimental tests. On the basis of experimental and simulation tests, it is possible to select the optimal solution limiting product loss while obtaining the highest possible efficiency and product solution concentration.

Chromatographic separation is related to the dilution of the product. Column overload is used to limit the degree of dilution. Fig. 11 shows a simulation data determined on the basis of a mathematical model, in which the effect of column overload on the concentration of product fractions was analyzed. As can be seen, 30%–40% overload makes it possible to obtain the most concentrated product fraction. A further increase in the column overload, which would lead to a reduction in the separation efficiency, does not increase the concentration of the product. It can be concluded that from the point of product concentration an overcharge of approximately 35% can be used. The result is a high degree of desalination (90%) while minimizing product losses. The final stage is ion exchange in the cation–anion exchanger system. A high degree of desalination in the chromatography process reduces the frequency of regeneration of the bed in the ion exchange stage (final polishing stage). In addition to the economic advantage, a lower amount of waste fractions is obtained.

As has been shown in experimental studies, collecting fractions for waste, recycling, and product with the simultaneous re-treatment of the recycling fraction allows the efficiency of the process.

Division of fractions affects their composition and quantity. Reducing waste fraction increases the amount of salt in the recycle, but, as shown in Fig. 12, it reduces product losses. As can be seen, the use of a mathematical model allows one to determine many relationships that facilitate the selection of process parameters while limiting the number of experimental tests.

The fermentation broth experiments were carried out according to the procedure described above. Fig. 13a and b show exemplary results obtained for broths obtained as a result of glycerol fermentation by strain of *Yarrowia lipolytica*. The fermentation broth purified on the ion exclusion consisted of the main product: erythritol in the amount of 112 g/L and by-products and unreacted components of the medium: inorganic salts 39 g/L, citric acid 9 g/L, mannitol 5 g/L. As can be seen, elution curves for ionic compounds – inorganic salts and carboxylic acids – have the same course. The similar relation occurs for polyols – erythritol and mannitol have a very similar affinity to the resin. This is an important aspect that should be considered in the crystallization step. However, both erythritol and mannitol are used as sweeteners with similar properties, so the aspect is not as

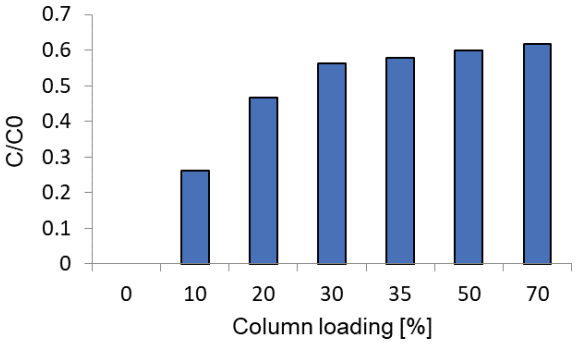

Fig. 11. Effect of column overload on the concentration of the product fraction determined on the basis of the mathematical model data (assumption of 90% desalination of the product, C_0 – the polyol concentration in the feed, C – the polyol concentration in the product fraction).

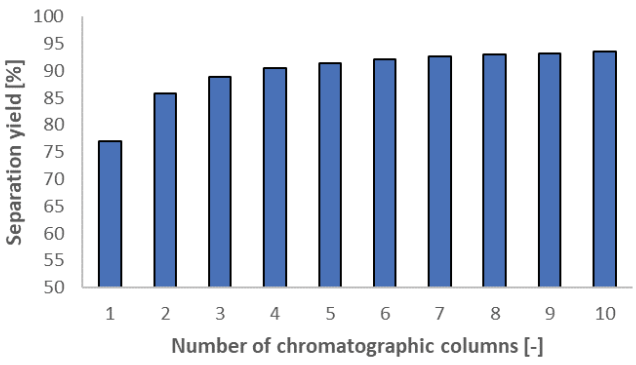

Fig. 10. Effect of the number of columns on the yield of propane-1,3-diol separation (30% column loading, column volume 10.5 L).

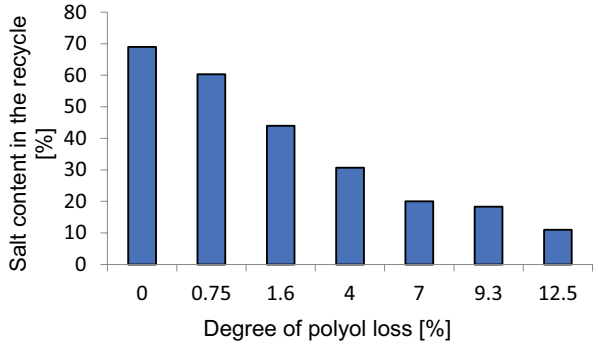

Fig. 12. Effect of salt content in the recycle on polyol loss determined on the basis of the mathematical model data (assumption of 90% desalination of the product).

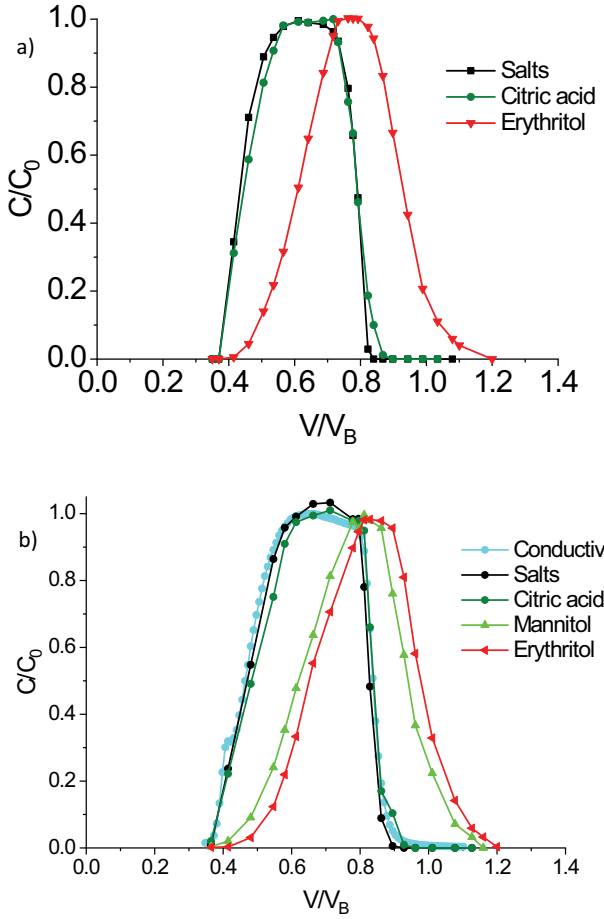

Fig. 13. Experimental elution profiles for ion exclusion (35% bed volume) of fermentation broth for: (a) 0.53 L column packed with Lewatit S1567, 0.6 mm, (b) 10.5 L column packed with Lewatit S1567, 0.6 mm. *V*/VB – eluent volume ratio, where *V* is the total volume of mobile phase dispensed onto the column, and VB is the volume of the column with bed; C/C_0 – component concentration ratio, where *C* is the component concentration in the tested sample and C_0 is the initial concentration of the component in the feed solution).

significant as the need to separate salts and acids from the final product.

In the last step, a process analysis was carried out for the broth solution obtained by fermentation and the data was compared with the simulation results. Fermentation broth (with the following composition: 101 g/L of erythritol, 3 g/L of mannitol, 45 g/L of inorganic salts, and 3 g/L of organic acids) was filtered for biomass separation. The resulting clear solution was subjected to ion exclusion on a 530 mL column packed with Amberjet 1200Na. The desalination process was carried out at a temperature of 50°C, the pH value of the broth was 4. The experimental results were compared with the computer simulation and presented in Fig. 14. The proposed mathematical model is applicable both for model solutions and for real solutions with a more complex composition. Furthermore, studies have shown the possibility of using a different ion exchange resin with the same parameters in exclusion chromatography (monodisperse, strongly

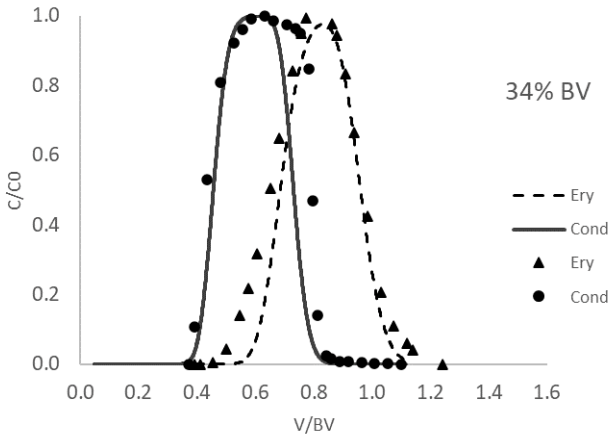

Fig. 14. Comparison of the experimental (symbols) and simulated (lines) elution profiles of 0.83 mol/L erythritol (Ery) and 0.77 mol/L sodium chloride (Cond) for 0.53 L column packed with Amberjet 0.6 mm (space velocity 1 h^{-1}), feed solution 180 mL (34% bed volume). *V*/BV – eluent volume ratio, where *V* is the total volume of the mobile phase dispensed onto the column, and BV is the volume of the column with bed; C/C_0 – component concentration ratio, where *C* is the component concentration in the tested sample and C_0 is the initial concentration of the component in the feed solution).

acidic cation exchange resin in sodium form, gel-type beads, 0.6 mm).

Process optimization using appropriately selected mathematical models and kinetic data is a very important tool that increases the potential possibility of implementing biotechnological techniques in industry. In the literature, this issue is increasingly applied to bioconversion products, including carboxylic acids [37,38]. Therefore, an important aspect is the possibility of simulating preparative chromatography for polyols. Daza-Serna et al. [39] presented the methods to obtain erythritol by fermentation and the analysis of the separation methods. Well-developed fermentation methods require the use of efficient purification techniques. The main impurities that require separation are inorganic salts, which are necessary in the bioconversion stage (sodium chloride). Among the separation methods, methods based on sorption and chromatography have great potential. The research results presented in this paper are an important tool that supports the optimization of the desalination process and the selection of the most appropriate parameters.

5. Conclusions

Ion exclusion leads to the separation of inorganic salts from the erythritol solution with a high degree of desalination. A multistage process increases the efficiency of the process, and the introduction of recycling reduces product losses in a multicolumn system. Mathematical modeling can be successfully used as a tool that supports the selection of process parameters. Simulation curves similar to the experimental profiles can be obtained using the equilibrium dispersive model (ED). Based on the dynamic adsorption–desorption method, a linear adsorption isotherm was selected and the

isotherm constant was determined (K = 0.327). Separation efficiency decreases with volume overload, and the process carried out for a 30%–40% column load allows the highest possible separation efficiency. Separation efficiency decreases with volume overload. The process carried out at 30%–40% column load allows for the highest possible separation efficiency and the lowest dilution of the product fraction. Separation of inorganic salts (sodium chloride) and carboxylic acids (citric acid) was obtained at over 90% for a loss of 4% of the product in the waste fraction.

Declaration of competing interest

The authors declare that they have no competing interests.

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