Research Article

Nanofiltration-based Recovery of Sodium Gluconate from Fermentation Broths of Oil Palm Solid Residue Hydrolysates

Ayla Annisa Liswani[1], Aulya Rahman Arevin[1], Naufal Baariq[1], Ibnu Maulana Hidayatullah[1,2,3,*], Muhamad Sahlan[1,2,3], Masafumi Yohda[4], Heri Hermansyah[1,2,3,*], Apriliana Cahya Khayrani[1,2,3,*]

[1] Department of Chemical Engineering, Faculty of Engineering, Universitas Indonesia, Depok, West Java, 16424, Indonesia
[2] Bioprocess Engineering Study Program, Faculty of Engineering, Universitas Indonesia, Depok, West Java, 16424, Indonesia
[3] Research Center for Biomass Valorizations, Universitas Indonesia, Depok 16424, Indonesia
[4] Department of Biotechnology and Life Science, Tokyo University of Agriculture and Technology, Koganei, Tokyo, 148-4588, Japan

*Corresponding Email: heri.hermansyah@ui.ac.id, apriliana.cahya@ui.ac.id

Abstract

Sodium gluconate (SG), a functional organic salt derived from gluconic acid, is widely utilized in various industrial sectors because of its excellent chelating ability, low toxicity, and biodegradability. SGs can be produced sustainably through the neutralization of gluconic acid, which is obtained from the microbial fermentation of lignocellulosic biomass such as oil palm fronds (OPFs) and empty fruit bunches (OPEFBs). To meet commercial demands, SGs must be recovered in concentrated form, and nanofiltration (NF) offers a promising membrane-based approach for this purpose. This study aimed to optimize the SG concentration from the fermentation broths of oil palm residue hydrolysates via a dead-end NF system, with feed pH as a key parameter. The NF270 membrane demonstrated optimal performance when synthetic fermentation broth was used at pH 8.0 and 9 bar, yielding 5.55 ± 0.16 g L^{-1} SG, a flux of 74.47 ± 0.39 L m^{-2} h^{-1}, $28.04 \pm 2.13\%$ rejection, and $25.21 \pm 1.12\%$ recovery. An increased SG concentration was also achieved in real biomass-derived broths; at pH 8.0, the OPF hydrolysate produced 1.56 ± 0.02 g L^{-1} SG with 35.72 ± 1.36 L m^{-2} h^{-1} flux, whereas the OPEFB hydrolysate yielded 1.52 ± 0.15 g L^{-1} SG with 41.29 ± 1.26 L m^{-2} h^{-1} flux. The results demonstrate that feed pH significantly influences nanofiltration performance, particularly in terms of improving the SG concentration and membrane efficiency across different biomass sources. This study provides the first systematic evaluation of SG recovery from oil palm solid residues via nanofiltration, highlighting its potential as a sustainable and efficient alternative to conventional purification methods.

Introduction

The transformation of lignocellulosic biomass into value-added biochemicals represents a cornerstone of sustainable industrial biotechnology [1]. Among the numerous platform chemicals derived from biomass, gluconic acid stands out for its broad applicability and eco-friendly profile, garnering significant interest from sectors ranging from food and pharmaceuticals [2–3]. This compound is biosynthesized through glucose fermentation facilitated by Gluconobacter oxydans, which play a pivotal role in the selective and efficient oxidation of monosaccharides due to its high selectivity, substrate tolerance, and resistance to lignocellulose-derived fermentation inhibitors [4–5].

The utilization of lignocellulosic biomass from agricultural residues is increasingly recognized as a strategic approach to support the sustainable production of gluconic acid. Among these residues, oil palm byproducts are particularly promising resources [6]. As one of the largest palm oil producers globally, Indonesia generates

substantial volumes of biomass waste, notably oil palm fronds (OPFs) and empty fruit bunches (OPEFBs), which are often discarded or incinerated without under-going any meaningful valorization [7]. These residues are characterized by their high holocellulose content, reaching up to 67% in OPEF and 83% in OPF, making them attractive feedstocks for biochemical conversion [8].

Through pretreatment and enzymatic hydrolysis, the structural carbohydrates within these residues can be depolymerized into fermentable sugars, primarily glucose. This glucose serves as the principal substrate in the microbial production of gluconic acid, which is subsequently neutralized to form sodium gluconate (SG), a water-soluble salt with considerable industrial importance [9]. SG is widely employed as a chelating agent in cleaning formulations, a corrosion inhibitor in metal surface treatments, and a dispersing agent in construction applications [10]. The biodegradability, low toxicity, and functional stability of these compounds make them compelling alternatives to conventional synthetic additives.

Despite its industrial potential, the economic viability of SG production hinges not only on upstream fermentation efficiency but also on the implementation of effective downstream recovery methods [11]. Traditional concentration techniques, such as thermal evaporation and solvent-based extraction, often involve high energy consumption and operational costs. In contrast, membrane-based technologies, such as nanofiltration (NF), offer selective and energy-efficient alternatives for concentrating SG on dilute fermentation broths [12].

Nanofiltration operates on the combined principles of molecular sieving and electrostatic exclusion, with membrane performance being influenced by factors such as solute properties, membrane characteristics, and solution chemistry [13]. The NF270 membrane, composed of polyamide and characterized by a molecular weight cutoff of 200-300 Da and a negatively charged surface, has shown promise for separating low-molecular-weight organics, including sodium-based compounds [14].

Among the various operational parameters, pH plays a decisive role in determining NF performance. Adjustments in pH alter the ionization states of solutes and the surface charge of the membrane, which in turn affect solute rejection and permeate flux [15]. Previous investigations have demonstrated that elevated feed pH improves the retention of gluconic acid in hydrolysates derived from corn cobs and wheat straw [14]. Similar trends were observed in the NF of organic acid-containing domestic waste streams [16]. However, there is a lack of research specifically addressing the NF of sodium gluconate derived from lignocellulosic sources such as OPF and OPEFB. Additionally, optimal conditions are needed to obtain the optimum recovery, flux, and rejection of sodium gluconate.

To address this gap, the present study investigated the use of a dead-end NF system with NF270 membranes for concentrating sodium gluconate produced from oil palm residues. The effect of feed pH on membrane performance was systematically evaluated using both synthetic and biomass-derived fermentation broths, enabling a comparative assessment of sodium gluconate recovery across different substrates.

Materials and methods
1) Materials

Analytical-grade sodium gluconate and sodium hydroxide (Sigma–Aldrich) were used in this study. The fermentation media used were synthetic media, OPF hydrolysate, and OPEFB hydrolysate. The synthetic fermentation medium was composed of glucose, xylose, yeast extract, $MgSO_4 \cdot 7H_2O$, KH_2PO_4, and $(NH_4)_2SO_4$ while the OPF and OPEFB hydrolysates were produced via hydrothermal and enzymatic pretreatment. The NF process was conducted using NF270 flat-sheet membranes (DOW Filmtec), with 50% ethanol used for membrane pretreatment. High-purity nitrogen gas (N_2) was used to apply operational pressure. For HPLC analysis, methanol and phosphoric acid were used as the mobile phases.

2) Method
2.1) Fermentation broth preparation

The fermentation process was conducted via several media: synthetic, OPF hydrolysate, and OPEFB hydrolysate. For the synthetic medium, 50 g of glucose, 25 g of xylose, 20 g of yeast extract, 0.5 g of $MgSO_4 \cdot 7H_2O$, 1.5 g of KH_2PO_4, and 1.5 g of $(NH_4)_2SO_4$ were dissolved in 1 L of demineralized water [17]. OPF and OPEFB biomasses were pretreated hydrothermally (150°C for 40 min) in 0.05 M citrate buffer, followed by enzymatic hydrolysis using cellulase:xylanase at a ratio of 1:3 for 72 hours. Fermentation was then carried out with Gluconobacter oxydans for 72 hours at 220 rpm and 30°C [18]. The fermentation broth obtained was autoclaved (121°C for 15 min) and vacuum filtered.

2.2) Neutralization and nanofiltration

The acidic fermentation broth obtained was neutralized via the addition of 1 M NaOH dropwise at a rate of 1 mL min^{-1} under continuous stirring at 220 rpm until the pH of the fermentation broth was adjusted to 7.5–8.0, depending on the experimental conditions. These pH values were selected since slightly alkaline conditions (approximately pH 8) have been reported to increase rejection rates in NF processes involving sodium salts, whereas near-neutral conditions help to maintain stable flux and minimize fouling in ultrafiltration and NF membranes [19–20]. For comparison, the pure SG solution was prepared by dissolving SG salt crystals in water, and the synthetic solution was prepared by mixing gluconic acid with NaOH. Pure SG and synthetic SG

solutions were used to represent a simplified system, enabling direct comparison with fermentation broth and highlighting the influence of complexity on NF performance. The neutralized fermentation broth, pure SG, and synthetic SG were then used for NF, which was conducted via a Sterlitech HP4750 high-pressure stirred cell in a dead-end configuration equipped with an NF270 membrane. The flat-sheet membranes were pretreated by soaking in 50% ethanol for 24 hours, followed by rinsing with 200 mL of deionized water under 9 bar pressure for 30 min. Each filtration process was conducted at 9 bars for 30 min using nitrogen gas as the driving force. The feed volume ranged from 30 to 200 mL depending on the feed type. During this process, the permeate was collected from the system's weep tube, while the retentate remained in the cell. Both fractions were used for subsequent analysis.

3) Analysis and data interpretation

High-performance liquid chromatography (HPLC) analysis was conducted via a Shimadzu system fitted with a GIST C18 column and a UV/VIS detector. The samples for sodium gluconate quantification were filtered through 0.22 μm syringe filters and diluted tenfold to reduce matrix interference. The mobile phase in this analysis consisted of methanol and phosphoric acid in a 1:9 ratio and was operated at a flow rate of 1 mL min^{-1} with a column temperature of 40°C. Sodium gluconate concentrations were determined via a standard calibration curve.

The membrane performance was evaluated by calculating data for three parameters: flux, rejection, and recovery. Flux was calculated as the permeate volume per unit membrane area per unit time. The rejection percentage was determined on the basis of the difference between the feed and permeate SG concentrations. The recovery percentage was obtained by comparing the amount of SG retained in the retentate relative to the total amount present in the feed mixture. These values were used to assess the influence of the feed pH and matrix composition on the NF efficiency. Flux describes the volumetric flow rate of permeate normalized by the membrane area and filtration time and is calculated via Eq.1.

$$J_v = \frac{V_p}{A.t} \qquad \text{(Eq.1)}$$

where J_v is the flux (L m^{-2} h^{-1}), V_p is the volume of permeate collected (L), A is the effective membrane area (m^2), and t is the filtration time (hours). The rejection calculates the percentage of SGs retained by the membrane and is expressed as Eq.2.

$$R_i = \left(1 - \frac{C_{i,P}}{C_{i,F}}\right) \times 100\% \qquad \text{(Eq.2)}$$

where $C_{i,P}$ is the concentration of SG in the permeate (g L^{-1}) and where $C_{i,F}$ is the concentration in the feed solution (g L^{-1}). A higher rejection indicates greater retention of solute by the membrane. Recovery represents the fraction of solute retained in the retentate relative to its initial amount in the feed and is calculated as Eq.3.

$$R(\%) = \frac{V_R.C_R}{V_F.C_F} \qquad \text{(Eq.3)}$$

where V_R and C_R are the concentration and volume of SG in the retentate, respectively, and where V_F and C_F refer to those in the feed solution. Recovery is a measure of how much of a target compound is captured and concentrated during NF.

Results and discussion
1) Characterization of the neutralized feed solutions

The neutralization process plays a crucial role in the preparation of fermentation broths for NF because sufficient conversion of gluconic acid into its salt form, SG, is achieved. Adjustment of the pH to 7.5 and 8.0 with NaOH directly influenced this conversion, as higher pH promoted more ionization of gluconic acid. The concentrations of SG obtained at pH 7.5 and 8.0 from various feed solutions are shown in Table 1. The highest initial concentration of SG was obtained in the synthetic fermentation broth because of its controlled composition. In contrast, the OPF and OPEFB hydrolysate broths presented lower SG concentrations, which was attributed to the complexity of the biomass-derived substrates and the presence of microbial metabolites. Thus, the volume of NaOH required to reach the desired pH was greater for these lignocellulosic broths, indicating a higher initial acid content.

This feed solution concentration is used as initial concentration data to further compare SG recovery and quantify the performance of the NF membrane, whereas in the NF membrane process, the pH value of the feed solution affects the membrane surface charge [14]. The surface charge of the membrane determines the mechanism by which the membrane rejects the feed. A feed pH that exceeds the membrane's isoelectric point will result in a negatively charged membrane surface, and the membrane's performance will be influenced by charge repulsion or the Donnan exclusion effect [23]. Thus, the influence of charge repulsion effects on membrane performance enhances the ability of a membrane to reject ionic compounds [24].

Table 1 SG concentrations (g L^{-1}) at pH 7.5 and 8.0 from various feed solutions

Feed solution	pH 7.5	pH 8.0
Pure sodium gluconate	3.500	3.500
Synthetic sodium gluconate	3.500	3.500
Synthetic broth	7.600	7.700
OPF hydrolysate broth	2.202	2.205
OPEFB hydrolysate broth	4.360	4.370

2) Characterization of the NF membrane

The NF270 membrane used for the NF process was a polyamide thin-film composite membrane with a molecular weight cutoff (MWCO) of approximately 200-400 Da, which encompasses the molecular weight of sodium gluconate (218.14 Da), indicating that electrostatic repulsion dominates the rejection mechanism [22]. The membrane's water permeability was determined to be approximately 20.55 L m^{-2} h^{-1} bar^{-1} when it was operated at 9 bar, which is consistent with the literature values from Sahlan et al. [4], where the water permeability was 20.34 L m^{-2} h^{-1} bar^{-1} at a pressure of 9 bar. A study by Tomczak [22] reported a membrane permeability of 9.8 L m^{-2} h^{-1} bar^{-1} for the NF270 at 5 bar. This confirmed that the membrane functioned properly and within its expected performance range and that NF270 permeability increased with increasing membrane pressure.

3) Effect of feed pH on NF flux

The NF flux was calculated via the flux equation and is presented in Figure 1. The feed pH influences the NF flux, as increasing the pH affects the feed concentration, membrane permeability, and membrane resistance. As shown in Figure 1, the NF flux increased with increasing pH. A study by Nilsson et al. [25] reported enhanced membrane permeability following alkaline pretreatment at pH 10.4 and 9.8 for 40 and 60 minutes, respectively. The study also revealed an increase in NF membrane flux after conditioning at pH 11.5. The greater presence of Na$^+$ ions at pH 8.0 than at pH 7.5 results in more cations carrying a charge opposite to that of the negatively charged NF270 membrane. This facilitates easier permeation of components through the membrane, contributing to increased flux.

The flux of the synthetic SG solution obtained showed minor deviations, confirming the suitability for positive control and approximation for flux discussion. In contrast, both the synthetic fermentation broth and the OPEFB hydrolysate broth exhibited substantially lower fluxes, with reductions of 129.361±1.652 L m^{-2} h^{-1} and 162.405 ±1.457 L m^{-2} h^{-1}, respectively, compared with the pure synthetic SG solution. This reduction can be attributed to the higher solute concentrations and the presence of additional components in the fermentation-derived feeds, which increase the osmotic pressure and concentration polarization, thereby hindering water transport through the membrane.

Baruah et al. [26] reported that higher feed concentrations lead to decreased NF flux due to increased osmotic pressure and concentration polarization. However, for the OPF and OPEFB hydrolysates, despite having a higher initial SG concentration, the synthetic fermentation broth still exhibited greater flux than both hydrolysate broths did. The presence of lignin, an impurity in OPF and OPEFB feeds with a molecular weight of approximately 509.4 Da, may decrease permeate flow by causing fouling on the membrane surface, as it exceeds the MWCO of NF270 (300 Da). The metabolites produced by Gluconobacter oxydans during fermentation include 2-ketogluconic acid (2-KGA) and 2,5-diketogluconic acid (2,5-DKGA), which may also interfere with NF performance and reduce flux. A statistical analysis was conducted to determine the significance of the pH increase on the NF flux across all feed types, resulting in p values less than 0.05 ($p < 0.05$), which indicated that the pH change altered the membrane surface charge and influenced the NF process, thus impacting the flux performance.
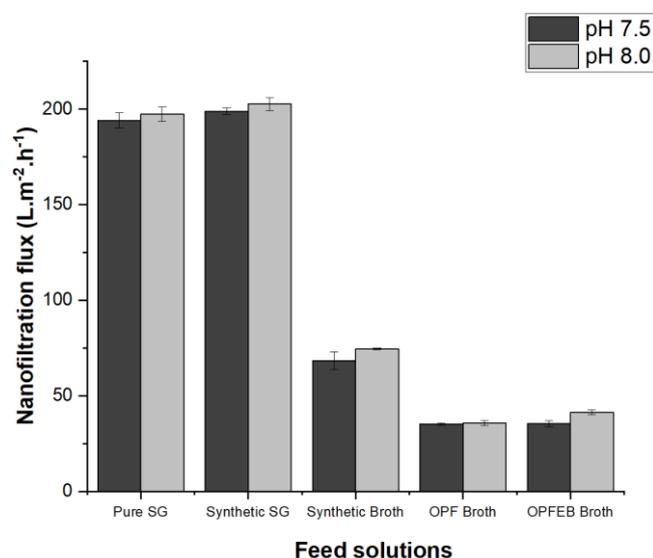

Figure 1 Effect of pH on the NF flux of various feed solutions.

4) Effect of feed pH on SG rejection

The rejection of SG was calculated via the rejection equations and the recovery equation to provide a more comprehensive understanding of membrane performance. As stated in the previous discussion, the membrane separation mechanism in this study was influenced by charge repulsion due to the negatively charged nature of the NF270 membrane. According to Donnan's theory, salt rejection depends on the properties of coions that share the same charge as the membrane [27]. In the case of SG, the ions have equivalent valence charges, but the Na$^+$ ion is more stable because of its closed-shell electron configuration, whereas the C$_6$H$_{11}$O$_7^-$ anion behaves more

like a neutral molecule in aqueous environments [28]. The SG rejection results are shown in Figure 2.

As illustrated in Figure 2, SG rejection generally decreased with increasing pH for all feed types, except for the pure SG solution. This is due to differences in ionic behavior between the two types of solutions. The pure SG solution was prepared by dissolving SG salt crystals in water and contained a lower degree of ion dissociation than did the synthetic SG solution. The synthetic solution is prepared by mixing gluconic acid with NaOH, resulting in more fully dissociated and mobile ions, which lead to greater ion flux and lower observed rejection. The decreased SG rejection with increasing pH can be attributed to the greater addition of NaOH during neutralization, which increases the concentration of Na^+ ions in the feed. These additional Na^+ ions, being coions with respect to the membrane, reduce the membrane's ability to reject the gluconate salt, resulting in decreased SG rejection at higher pH. The secondary metabolites produced by Gluconobacter oxydans most likely contribute to reduced feed per-meability. This explained why the highest rejection values of the OPF and OPEFB feed solutions were 46.549± 4.431% and 83.876±0.442% at pH 7.5 and 29.042± 0.803% and 65.080±3.454% at pH 8.0, respectively. Decreased feed permeation is strongly correlated with lower NF flux, which leads to increased SG rejection. The increase in feed pH had a statistically significant effect on SG rejection ($p < 0.05$), where the increased presence of freely mobile Na^+ ions at higher pH levels influenced the ion balance near the membrane interface and reduced the membrane rejection efficiency for SG.

5) Effect of feed pH on SG recovery

The purified SG primarily accumulates in the permeate because of Donnan's exclusion within the membrane. However, as discussed in the previous sections, some SGs also accumulate in the retentate. To assess the membrane performance, the concen-tration of SG in the retentate was quantified via HPLC to calculate the recovery percentage of SG. The recovery percentage reflects the proportion of SG retained in the retentate relative to the total SG initially present in the feed solution. The results of this quantification are presented in Figure 3.

The SG recovery percentage shown in Figure 3 decreased with increasing pH for all feed types, except for the pure SG solution. The trend in SG recovery closely mirrored the trend observed in SG rejection, as presented in the previous section. This correlation shows that the rejection percentage is a suitable indicator of the membrane's ability to retain solutes and its capacity to recover unpurified SGs in the retentate. The changes in feed pH did not significantly affect SG recovery ($p > 0.05$). This finding is reasonable, considering that recovery

analysis was based solely on the concentration of SG remaining in the retentate and does not reflect the full solute profile of the system. The recovery percentage only indicates the amount of SG that was not trans-mitted through the membrane and not the complete amount of purified SG.
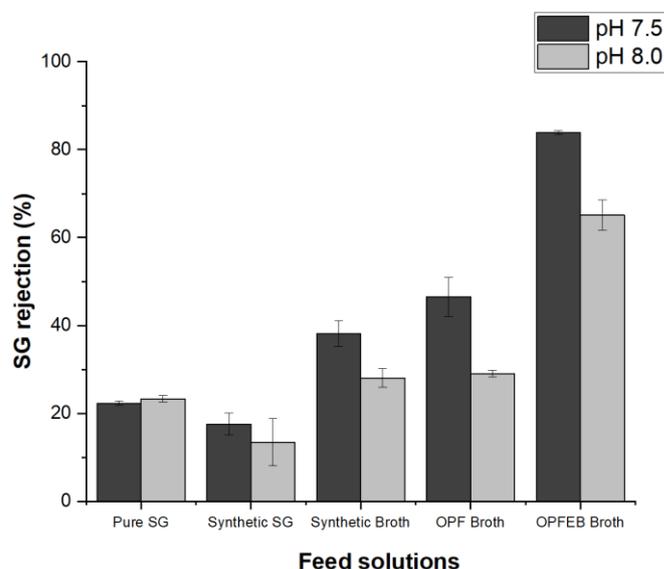

Figure 2 Effect of pH on SG rejection in various feed solutions.
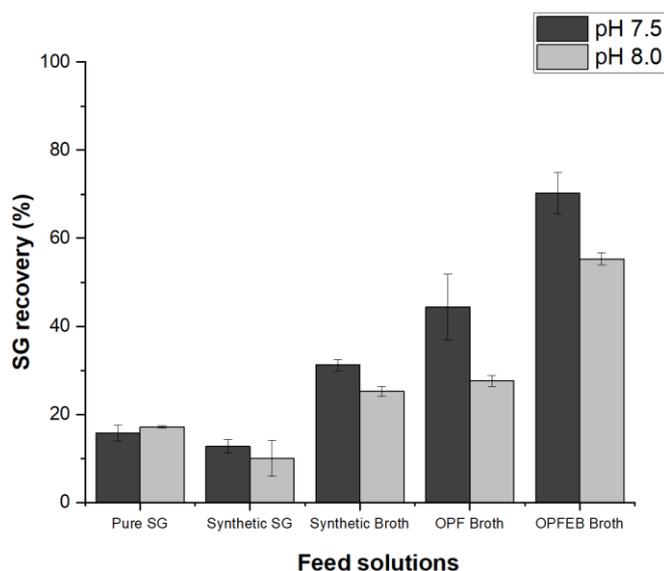

Figure 3 Effect of pH on SG recovery from various feed solutions.

6) Effect of feed pH on final SG concentration

The final SG concentration obtained through HPLC analysis serves as one of the key indicators of NF success as a method for determining the salt concen-ration. In this study, SG was expected to be purified primarily into the permeate phase as a result of the Donnan exclusion effect of the membrane. The final concentrations of SG, as determined via HPLC equipped with a C18 column, are shown in Figure 4.
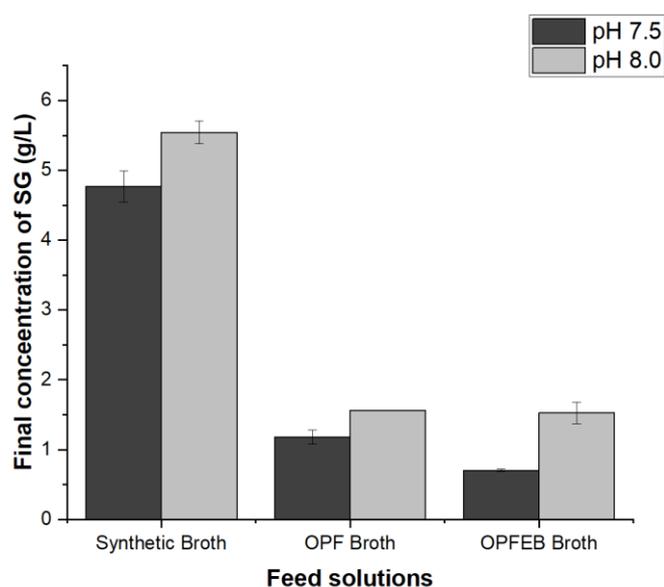
Figure 4 Final concentrations of SG from various feed solutions.

In general, the final SG concentration after NF is closely associated with the initial SG concentration present in the feed solution. Each feed solution had a different initial SG concentration, depending on the total volume of NaOH added during the pH adjustment process. The higher the pH is, the greater the volume of NaOH needed, which in turn results in a higher concentration of SG formed through the neutralization of gluconic acid with NaOH. As shown in Figure 4, the final SG concentrations increased proportionally with increasing pH across all feed types. The lowest final SG concentrations were observed in the feed solutions derived from OPF and OPEFB hydrolysates: 1.177 ± 0.098 g L^{-1} and 1.563 ± 0.018 g L^{-1} for OPF and 0.703 ± 0.018 g L^{-1} and 1.523 ± 0.151 g L^{-1} for OPEFB, respectively. This is due to incomplete conversion of gluconic acid during fermentation by *Gluconobacter oxydans*, resulting in the presence of residual impurities that were not removed prior to fermentation, as well as incomplete transformation of glucose into gluconic acid, with some glucose instead being converted into side products such as 2-ketogluconic acid (2-KGA) and/or 2,5-diketogluconic acid (2,5-DKGA). Statistical analysis revealed that increasing the pH had a statistically significant effect on the final SG concentration across all feed types ($p < 0.05$). However, the SG from the NF process in this study was not yet purified to its maximum potential, and a potential solution would be to extend the NF duration to increase the final product concentration of SG.

7) Findings in relation to previous studies

A comparative study was conducted and revealed that the findings of this study are in line with those of previous studies. The SG yield, represented by the final SG concentration, along with key NF membrane performance parameters such as the nanofiltration flux, rejection percentage, and recovery percentage, were compared with the results reported in similar studies. A summary of the SG yield and nanofiltration membrane performance from this study and related literature is presented in Table 2.

A study conducted by Sahlan et al. [4] was the most comparable study in scope and investigated the preliminary recovery of SG from fermentation broths prepared with synthetic media and OPF hydrolysate via the NF process with the NF270 membrane. Table 2 shows that differences in feed pH have a substantial effect on SG yield and NF performance. These differences are closely related to the SG formation mechanism, where a lower pH results in fewer available Na$^+$ ions to bind with C$_6$H$_{11}$O$_7^-$ anions, thereby limiting the formation of sodium gluconate. This effect was also reflected in the flux values, as higher pH improves membrane permeability, likely due to enhanced ionization and reduced resistance at the membrane interface.

In the case of the OPF hydrolysate fermentation broth, the percentage of SG rejection at pH 5.2 ($66.34\pm2.88\%$) was significantly greater than that at pH 7.5 ($46.549\pm4.431\%$) and pH 8.0 ($29.042\pm0.803\%$). Feed solutions with low or no charge are more difficult to permeate, as electrostatic repulsion between the uncharged feed and the negatively charged NF270 membrane is minimal, resulting in greater solute retention. As the feed pH increases, the degree of dissociation of SG increases, leading to a greater proportion of negatively charged gluconate ions in solution. Simultaneously, the NF270 membrane surface becomes more negatively charged because of the deprotonation of functional groups. This results in stronger electrostatic repulsion between the solute ions and the membrane surface, which supports solute transport across the membrane. Consequently, solute rejection increases, whereas permeate flux may decrease, depending on the balance between charge repulsion and osmotic resistance. Consequently, a larger portion of SG accumulates in the retentate, leading to a higher recovery percentage. This comparative study reinforces the conclusion that increasing the feed pH significantly influences both the SG yield and the NF membrane performance. The use of NF under alkaline conditions, particularly at pH 8.0, enhances the flux and supports the Donnan exclusion effect, while also illustrating that a lower pH may increase rejection and recovery at the expense of permeate productivity.

Table 2 Result comparisons of SG yield and NF270 membrane performance

Feed solutions	pH	SG concentration (g L^{-1})	Flux (L m^{-2} h^{-1})	Rejection (%)	Recovery (%)	Reference
Synthetic fermentation broth	4.0	0.05±0.01	6.24 ± 0.59	20.68±0.01	92.52±0.53	[4]
	7.5	4.77±0.23	68.36 ± 4.57	38.09±2.92	31.19±1.32	This study
	8.0	5.55±0.16	74.47 ± 0.39	28.04±2.13	25.21±1.12	This study
OPF hydrolysate broth	5.2	0.13±0.01	18.15 ± 2.23	66.34±2.88	97.35±0.22	[4]
	7.5	1.18±0.10	35.24 ± 0.68	46.55±4.43	44.38±7.50	This study
	8.0	1.56±0.02	35.72 ± 1.36	29.04±0.80	27.56±1.29	This study
OPEFB hydrolysate broth	7.5	0.70±0.02	35.45 ± 1.55	83.88±0.42	70.19±4.70	This study

Conclusions

This study successfully optimized sodium gluconate (SG) recovery from oil palm residue-derived fermentation broths via nanofiltration with NF270 membranes under a dead-end configuration. The experimental findings in this study showed that an increase in feed pH from 7.5 to 8.0 resulted in higher NF flux and lower SG rejection for all feed types. The highest flux was observed in the synthetic fermentation broth, followed by the OPEFB and OPF hydrolysates, while the highest rejection was obtained for the lignocellulosic broths. This study highlights the inverse effect of pH on flux and rejection, driven by changes in ion mobility and membrane charge interactions. Additionally, increasing the pH led to an increase in the final SG concentration in the retentate. Among the feed types, synthetic fermentation broth resulted in the highest yield of SG recovery, whereas hydrolysate-based media resulted in a lower value. This is attributed to more complete neutralization of gluconic acid at higher pH values. The effectiveness of the SG concentration also depended on the fermentation conditions, where the synthetic medium allowed for more selective conversion of glucose to gluconic acid by *Gluconobacter oxydans*, whereas biomass-derived feeds were affected by impurities and competing metabolic pathways, leading to reduced SG yields.

Acknowledgements

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