Recovery of Lactic Acid from Fermentation Broth by the Two-Stage Process of Nanofiltration and Water-Splitting Electrodialysis

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Abstract A two-stage process of nanofiltration and water-splitting electrodialysis was investigated for lactic acid recovery from fermentation broth. In this process, sodium lactate is isolated from fermentation broth in the first stage of nanofiltration by using an NTR-729HF membrane, and then is converted to lactic acid in the second stage by water-splitting electrodialysis. To determine the optimal operating conditions for nanofiltration, the effects of pressure, lactate concentration, pH, and known added impurities were studied. Lactate rejection was less than 5%, magnesium rejection approximated 45%, and calcium rejection was at 40%. In subsequent water-splitting electrodialysis, both the sodium lactate conversion to lactic acid and sodium hydroxide recovery, were about 95%, with a power requirement of 0.9~1.0 kWh per kg of lactate.

Keywords: lactic acid, nanofiltration, water-splitting electrodialysis

INTRODUCTION

Lactic acid is one of the most widely used food preservatives and a very common substrate for chemical synthesis. Fifty to 60% of the lactic acid produced worldwide is manufactured by fermentation [1-4]. The cost for lactate recovery and the conversion of lactate is an important factor in determining the economic feasibility of a fermentative lactic acid production process. Lactate or lactic acid can be recovered and purified from the fermentation broth by precipitation, extraction, adsorption, distillation, nanofiltration (NF), and electrodialysis (ED) [1,5-9]. Among these processes, ED is considered to be the most environment-friendly.

The transport of lactic acid through NF membranes was studied extensively by Horst *et al.* [10]. The purpose of this study, however, was to develop a mass transfer model for concentrating lactic acid out of a low concentration (1~2%) broth. NF has also been studied as an alternative to evaporation (EV) and to ED in industrial processes for whey demineralization [10]. Whey was concentrated by EV, followed by demineralization of the concentrated whey using electrodialysis [10]. A hybrid process of NF and ED has been developed for treating waste water in the pulp and paper industry [11]. The process was optimized to recover used water from the highly pigmented effluent which also had a high content of organic and organochlorinated compounds and salts. NF was selected for the removal of the organic compounds and partial desalination; ED was used to reduce the sodium chloride concentration.

As for lactic acid recovery methodologies, a 2-stage ED process was proposed [12] where the first step of desalting ED was for recovery of lactate from the fermentation broth and the second step of water-splitting ED (WSED) was to convert lactate to lactic acid (Fig. 1). The desalted broth usually contains a significant amount of hardness ions (Ca^{2+}, Mg^{2+}, *etc.*) due to poor rejection of these ions by the membranes used in desalting ED. These hardness ions can severely foul bipolar membranes in WSED and for this reason an ion-exchange step is required before WSED. NF can be an alternative to desalting ED. The major advantage of using NF for the recovery of lactate from a fermentation broth would be a high hardness rejection, which could obviate the need for an ion-exchange step before WSED, as demonstrated in Fig. 1. A NF membrane has a more open structure than reverse osmosis (RO) membranes and the mean pore size is somewhat larger than that of low molecular weight salts [13]. With NF membranes, ions in an electrolyte solution are retained by the combination of size exclusion and electrical interaction between the ions and the membrane. Impurities, including hardness ions in the fermentation broth, can be retained while lactate is recovered to the permeate in a NF process, especially if a membrane with a proper pore size and electrical properties is used [14].

In this study, the surface properties of NTR-729HF membranes were studied by FT-IR and zeta-potential analyses. The recovery of lactate by NF was investigated by using model solutions at various pressures, lactate

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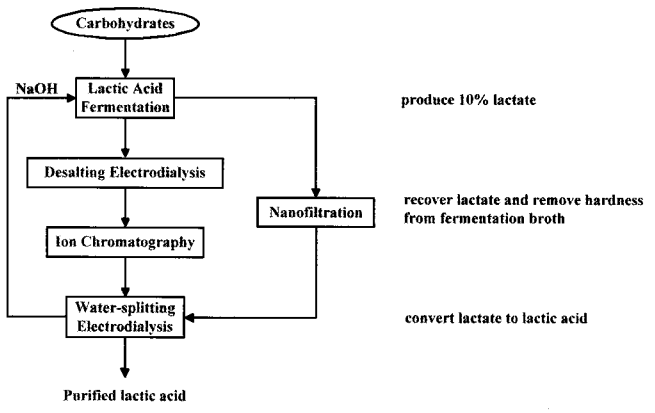

Fig. 1. Lactic acid production by fermentation and membrane separation.

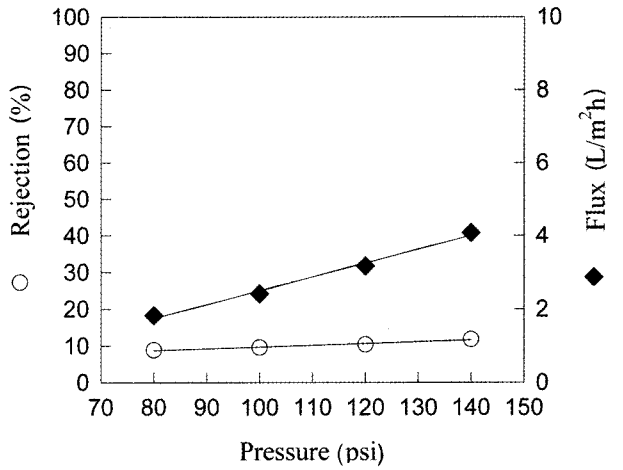

Fig. 2. Rejection and flux of sodium lactate at different pressures.

concentrations, pHs and impurity concentrations, and then some peculiar phenomena caused by high concentration of lactic acid were analyzed. Finally, the recovery of lactate by NF from an actual fermentation broth was performed, and the nanofiltered broth then acidified by use of WSED.

MATERIALS AND METHODS

Chemicals

Lactic acid solutions (Purac Co., Spain) used in this study contained oligomers. To decompose the oligomers into monomers, the solutions were boiled for 5 h at 120°C before use. Feed solutions containing 10% lactic acid were prepared and the pH adjusted to 5.5 by the addition of sodium hydroxide. Magnesium sulfate (Sigma Co., USA) and glucose (Difco Co., USA) were added to the resulting sodium lactate solution to observe the possible effects of known impurities.

Membrane

NTR-729HF (Nitto-Denko Co., Japan) membranes were used for NF. The NTR-729HF membrane is made of poly (vinyl alcohol) and polyamide, respectively.

Equipments and Experimental Conditions

The NF unit used in this study was composed of a high-pressure pump (RUM-2, Japan), a stack (C10-T, Nitto-Denko Co.), a feed reservoir and a cooling water bath for temperature control. The total solution volume in the system was 2 L and the circulation rate was maintained at 0.4 L/min. The retentates and permeates were recycled to the feed reservoir to keep lactate concentrations constant. The operating temperature was 30°C.

Analysis

A HPLC (Hitachi Co., Japan) with an Aminex HPX-87H column (Bio-Rad Laboratories Inc., USA) was used for analyzing lactic acid concentrations. Ion concentration in model solutions and the fermentation broths were measured by ion chromatography (Waters Co., USA) with IonPac AS18 and CS18 columns (Dionex Co., USA) and an inductively coupled plasma method (PQ3, VG elemental LTD., UK), respectively. The concentration of glucose was analyzed by an enzymatic method (Glucose- E Kit, Yeongdong Pharmaceutical Co., Korea). The chemical structures of the membrane surfaces were characterized by Fourier transform infrared spectroscopy (FTIR; model EQUINOX-55, Bruker Co, Germany) using the attenuated total reflectance (ATR) method. A zeta-potential analyzer (ELS-8000, Otsuka Co., Japan), based on electrophoresis, was used for measuring the surface charge change of the membranes. In this method a 10 mM NaCl solution containing polystyrene latex 504 nm particles at various pHs was used as a test solution.

RESULTS AND DISCUSSION

Effects of Pressure on Flux and Rejection

The rejection of lactic acid and the solution flux were measured at four different pressures (80, 100, 120, and 140 psig). The lactate concentration and pH were 100 g/L and 5.5, respectively. As shown in Fig. 2, the pressure increase from 80 to 140 psig resulted in a slight increase in the rejection of lactate, but a 2-fold increase in the solution flux.

Model equations which can describe the flux in NF have been reported by various research groups [10,15,17]. Generally, the solvent and solute fluxes can be described by a function of pressure and osmotic pressure differences and the extended Nernst-Plank equation, respectively:

$$J_{water} = A(\Delta P - \Delta \pi) \quad (1)$$

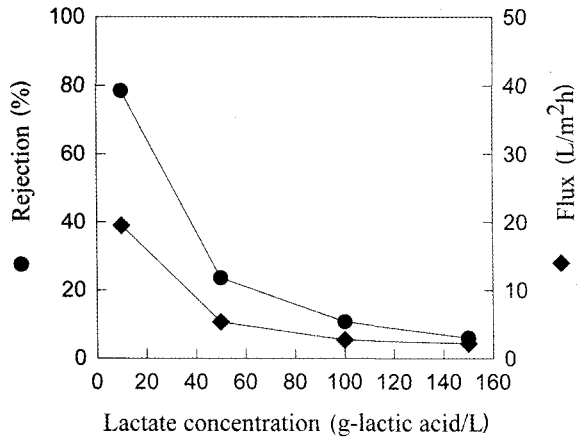

Fig. 3. Rejection and flux of sodium lactate at different lactate concentrations.

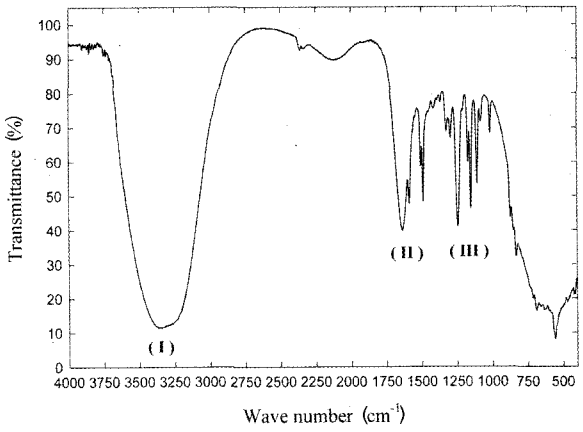

Fig. 4. FT-IR photograph of NTR-729HF membrane surface by attenuated total reflectance method.

$$J_{\text{lactate}} = -D^{\text{m}}\left(\frac{dc^{\text{m}}}{dx} + C^{\text{m}}\frac{zF}{RT}\frac{d\Psi^{\text{m}}}{dx}\right) + K_{\text{c}}J_{\text{water}}C^{\text{m}} \quad (2)$$

where J_{water} is water flux, A permeability, ΔP pressure difference, $\Delta\pi$ osmotic pressure difference, J_{lactate} lactate flux, D^{m} diffusivity, C^{m} lactate concentration at the membrane surface, x coordinate in the flow direction, z valency, F the Faraday constant, ψ^{m} electrical potential, R the gas constant, T temperature, and K_{c} the convective coupling coefficient [16]. According to Eq. (1), the solvent flux is proportional to the pressure difference, such linearity is evident as shown in Fig. 2.

According to Eq. (2), the transport of lactate ions is determined by the combination of diffusion and convection. At a low pressure the effect of diffusion is more dominant than that of convection. But at a high pressure it is reversed [10]. In this study, the increased effect of convection with pressure resulted in an increased transport of water, sodium and lactate, but the degree of increase was higher for water than for sodium and lactate. Therefore, the permeate was diluted and consequently the rejection of solutes was seen to increase with pressure.

Effects of Lactate Concentrations

The rejection of lactate and the solution flux were measured at four different lactate concentrations (10, 50, 100, and 200 g/L). As shown in Fig. 3, the increase in lactate concentration resulted in decreased flux and rejection.

The flux is dependent on the osmotic pressure difference when the operating pressure is fixed. The osmotic pressure difference is represented by the van't Hoff equation.

$$\Delta\pi = RT(C^{\text{m}} - C^{\text{p}}) \quad (3)$$

where C^{p} is lactate concentration in the permeate. As the bulk lactate concentration increases, the lactate concentration on the membrane surface (C^{m}) and thus ($C^{\text{m}} - C^{\text{p}}$) increases, resulting in an osmotic pressure difference (increase). It is in this way that the reduced flux at high lactate concentrations can be described (Fig. 3). The decrease in rejection with lactate concentration can be explained in two ways. Firstly, the acceleration of diffusion by the increased concentration gradient which is proportional to ($C^{m} - C^{p}$), where the accelerated lactate diffusion together with the reduced flux will result in a decreased rejection at high lactate concentrations and, secondly a screening effect due to a confined space surface charge of the membrane. As the lactate concentration increases, the confined space surface charge is screened by counter ions (sodium ions) in the feed. This screening effect can facilitate the transport of lactate ions through the membrane.

The recovery rate (≡ solution flux × permeate concentration) is an important factor for evaluating the performance of the NF process. It will be profitable to have a low rejection (high permeate concentration) and a high flux. But the two factors change with lactate concentration in the opposite manner of each other, implying that there may exist an optimum feed lactate concentration. However, the recovery rate monotonically increased with lactate concentration in the range tested in this study (10~150 g/L) (Table 1).

Effects of pH

The pH of a lactate solution is very important in determining the transport of lactate ions since the extent of dissociation of sodium lactate and the membrane surface charge change with the pH. The surface layer of NTR-729HF membrane and its electrostatic properties were investigated at different pHs. As shown in Fig. 4, the membrane surface was observed to have hydroxyl (I), acyl (II), and amide (III) groups, and the surface charge changed from slightly positive to large negative values as pH changed from 2.5 to 6.5 (Fig. 5).

The solution flux and the lactate rejection were measured at different lactate concentrations (10, 50, 100, and 150 g/L) and pH values (2.5, 3.5, 4.5, 5.5, and 6.5) (Fig. 6); the operating pressure was maintained at 120 psig. In the cases of 10 and 50 g/L of lactate concentration, the

Table 1. Recovery rate of lactate at different lactate concentrations

Feed concentration (C^f)[a] (g/L)	Permeate concentration (C^p)[a] (g/L)	Flux (L/m²h)	Recovery rate (g/m²h)
10	2.0	19.5	38.7
50	37.0	5.33	197.4
100	88.9	2.67	237.1
150	140.4	2.17	304.1

[a]Lactic acid equivalent.

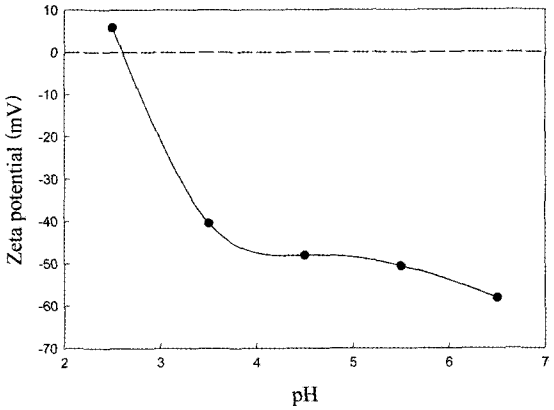

Fig. 5. Zeta potential of NTR-729HF membrane surface at different pHs.

rejection increased and the flux decreased with pH as expected. The fraction of dissociated lactate ions increased as the pH increased, and thus more lactate ions were rejected by the negatively charged membrane. The increase in rejection resulted in an osmotic pressure difference which made, in turn, the solution flux decrease.

In the cases of 100 and 150 g/L of lactate concentration, the flux decreased with pH, however, the lactate rejection also decreased with pH on the contrary in cases of low lactate concentration. This unexpected phenomenon can be explained by the effect of total salt concentration. In the cases of high lactate concentrations, a very large amount of NaOH needs to be added to raise the solution pH (Fig. 7). This implies that the total salt concentration is very high in a high-pH solution. The decrease in rejection due to the high total salt concentration (as evidenced by Fig. 3) at a high pH was considered to be more than enough to counter balance the increase in rejection due to the intrinsic effect of a high pH on rejection.

Overall, especially in the cases of 100 and 150 g/L lactate concentration, the effect of the pH on the rejection and flux was insignificant over pH 5.5. Lactate fermentation in this study was carried out at pH 5.5, and for this reason, the subsequent experiments were carried out at pH 5.5.

Effects of Residual Glucose

It is important to know how impurities in fermentation broth influence the recovery of lactate. The rejection and

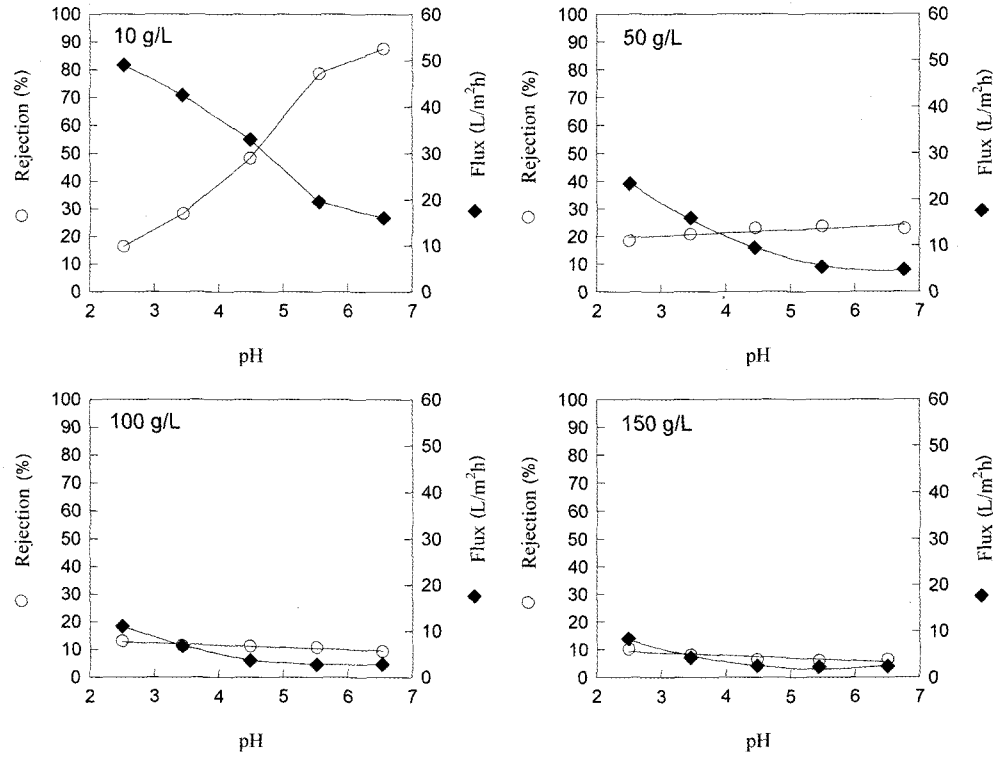

Fig. 6. Lactate rejection and solution flux at different pHs and concentrations.

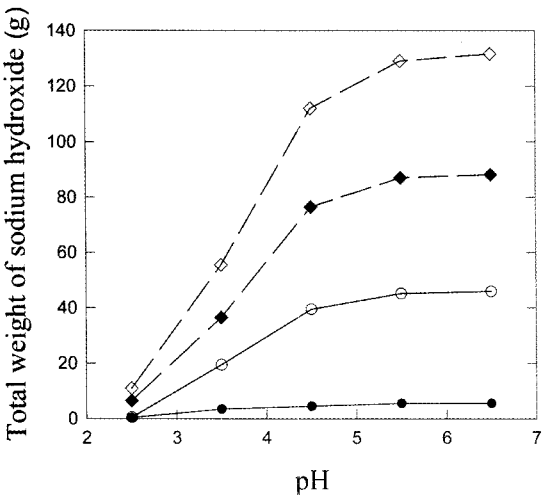

Fig. 7. Quantity of sodium hydroxide added for pH adjustment.

Table 2. Recovery of lactate from fermentation broth

Components	Feed concentration	Rejection (%)	NF45 Rejection (%)
Lactate (g/L)	90.0	4.9	20.0
Glucose (g/L)	0.42	45.9	60.9
Magnesium (ppm)	99.8	45.3	83.5
Calcium (ppm)	45.2	40.0	65.0

the solution flux were measured in the presence of glucose. The rejections of lactate and glucose were measured under four different glucose concentrations (0.5, 1, 2, and 4 g/L) when the lactate concentration was maintained at 100 g/L, as shown in Fig. 8. The rejection of glucose decreased slightly from 38 to 34% with its concentration, while the rejection of lactic acid and the solution flux were not influenced by glucose concentration.

Hardness Rejection

The lactate concentration was varied from 10 to 90 g/L and, the operating pressure and pH were 120 psig and 5.5, respectively. As shown in Fig. 9, the rejection of magnesium ions (0.01 M) showed high values, although they decreased from 97.2 to 78.4% with lactate concentration, implying that an ion-exchange step can be omitted or at least minimized by using nanofiltration for the recovery of lactate.

Recovery of Lactate from Fermentation Broth

The recovery of lactate from actual fermentation broth with a sodium lactate concentration of 90 g/L was then performed. As noted in Table 2, the rejection of lactate was less than 5% and the solution flux was at 2.8 L/m^{2}h. This result was comparable to that of model solutions (data not shown). The rejection of glucose, magnesium and calcium were 45.9, 45.3, and 40.0%, respectively.

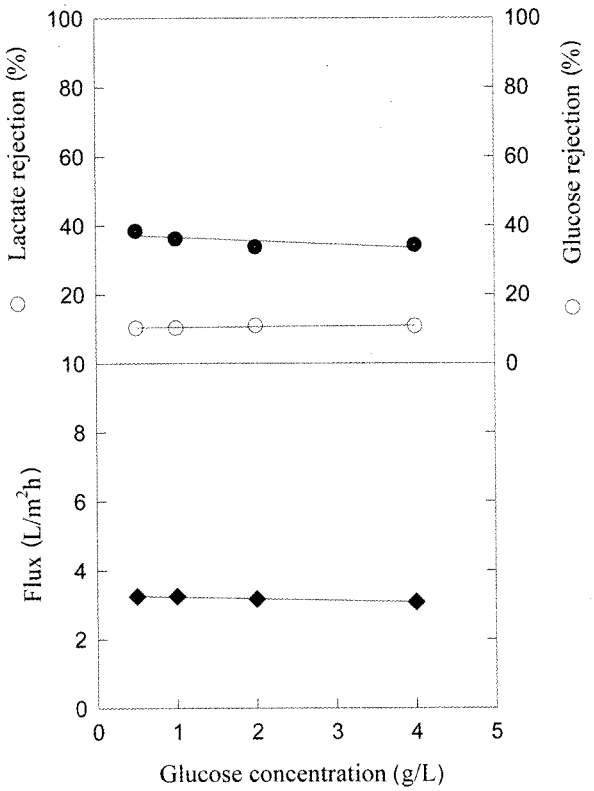

Fig. 8. Rejection of sodium lactate and glucose and solution flux at different concentrations of added glucose.

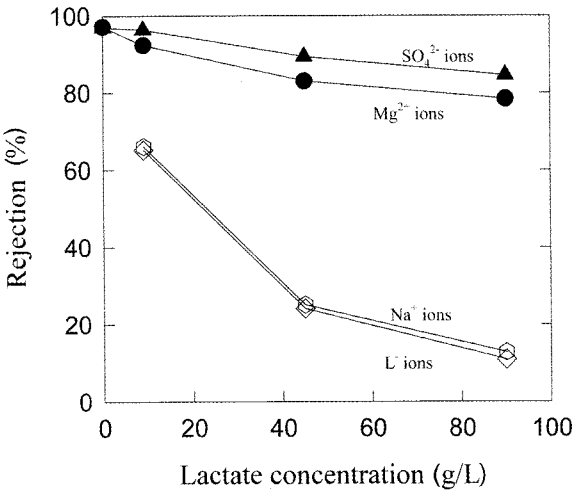

Fig. 9. Rejection of ions and the flux with 0.01 M $MgSO_4$ added at different sodium lactate concentrations.

The lower magnesium rejection in actual fermentation broth in comparison with that in a model solution (Fig. 9) is considered to be due to the influences by the various impurities in the fermentation broth. The level of hardness rejection is significantly higher than that of electrodialysis. However, careful optimization of culture medium to minimize hardness ions concentration is needed in the stage of lactate fermentation when the NTR-

Table 3. Lactate conversion and sodium hydroxide recovery by WSED

Experiments	Feed lactate concentration[a] (g/L)	Time[b] (min)	Sodium hydroxide recovery (%)	Current efficiency (%)	Energy consumption (kWh/kg)
1	72	65	97.4	77.3	1.00
2	72	60	94.1	83.7	0.91

[a]Lactic acid equivalent.
[b]Stopping conditions: pH<1.5 and conductivity<5 mS/cm.

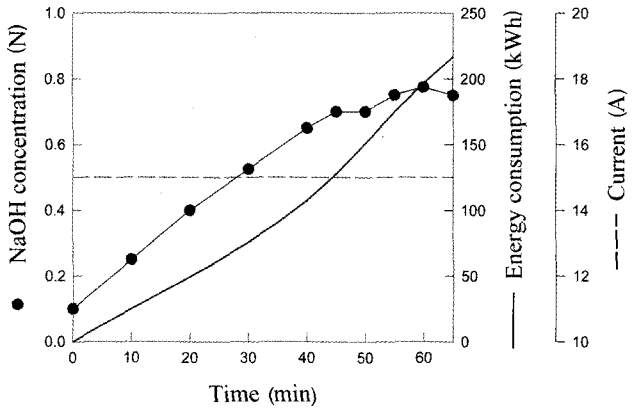

Fig. 10. Water splitting electrodialysis of nano-filtered fermentation broth.

729HF membrane is used for nanofilltration.

A second membrane (NF45) was tested as an alternative to NTR-729HF which was known to have a high hardness rejection. As can be seen in Table 2, the rejections of magnesium and calcium ions (83.5 and 65.0%, respectively) were much higher than those with NTR-729HF membrane, demonstrating the possibility of omitting the ion-exchange step through the use of the NF45 membrane. But, the major drawback of using the NF45 membrane was found to be the much higher rejection of lactate (20%) than the NTR-729HF membrane (5%).

Acidification of the Nano-Filtered Broth by WSED

Nano-filtered fermentation broth was treated by WSED to convert the lactate to lactic acid and simultaneously to recover the sodium hydroxide which is to be recycled to the fermentation process for the control of culture pH (Table 3 and Fig. 10). A constant-current mode operation was performed at a current of 15 A. During the course of collecting a large amount of nano-filtered fermentation broth for the lactate solution, a rather diluted lactate solution of 72 g/L was obtained. With the stopping conditions of pH<1.5 and conductivity<5 mS/cm, the necessary operation time was in the range of 60~65 min. The sodium hydroxide recovery was 94~97%, and the power requirement was 0.9~1.0 kWh per kg of lactate.

REFERENCES

[1] Timmer, J. M. K., H. C. van der Horst, and T. Robbertsen (1993) Transport of lactic acid through reverse osmosis and nanofiltration membranes. *J. Memb. Sci.* 85: 205-216.
[2] Wee, Y.-J., J.-N. Kim, J.-S. Yun, and H.-W. Ryu (2005) Optimum conditions for the biological production of lactic acid by a newly isolated lactic acid bacterium, *Lactobacillus* sp. RKY 2. *Biotechnol. Bioprocess Eng.* 10: 23-28.
[3] Liu, T., S. Miura, T. Arimura, M.-Y. Tei, E. Y. Park, and M. Okabe (2005) Evaluation of L-lactic acid production in batch, fed-batch, and continuous cultures of *Rhizopus* sp. MK-96-1196 using an airlift bioreactor. *Biotechnol. Bioprocess Eng.* 10: 522-527.
[4] Huang, L. P., B. Jin, P. Lant, X. Qiao, J. Chen, and W. Sun (2004) Direct fermentation of potato starch in wastewater to lactic acid by *Rhizopus oryzae*. *Biotechnol. Bioprocess Eng.* 9: 245-251.
[5] Prescott, S. C. and C. G. Dunn (1959) *Industrial Microbiology.* 3rd ed., McGraw-Hill, New York, NY, USA.
[6] Cockrem, C. M. and P. D. Johnson (1993) Recovery of lactate esters and lactic acid from fermentation broth. *US Patent* 5,210,296.
[7] Evangelista, R. L., A. J. Mangold, and Z. L. Nikolov (1994) Recovery of lactic acid by sorption. *Appl. Biochem. Biotechnol.* 45: 131-144.
[8] Vick Roy, T. B., D. K. Mandel, D. K. Dea, H. W. Blanch, and C. R. Wilke (1983) The application of cell recycle to continuous fermentative lactic acid production. *Biotechnol. Lett.* 5: 665-670.
[9] Glassner, D. A and R. Datta (1990) Process for production and purification of lactic acid. *EP* 0,393,818.
[10] Van der Horst, H. C., J. M. K. Timmer, T. Robbertsen, and J. Leenders (1995) Use of nanofiltration for concentration and demineralization in the dairy industry: Model for mass transport. *J. Memb. Sci.* 104: 205-218.
[11] Geraldes, V. and M. N. de Pinho (1995) Process water recovery from pulp bleaching effluents by an NF/ED hybrid process. *J. Memb. Sci.* 102: 209-221.
[12] Lee, E. G., S.-H. Moon, Y. K. Chang, I.-K. Yoo, and H. N. Chang (1998) Lactic acid recovery using two-stage electrodialysis and its modelling. *J. Memb. Sci.* 145: 53-66.
[13] Eriksson, P. (1988) Nanofiltration extends the range of membrane filtration. *Environ. Prog.* 7: 58-62.
[14] Kang, S. H., Y. K. Chang, and H. N. Chang (2004) Recovery of ammonium lactate and removal of hardness from fermentation broth by nanofiltration. *Biotechnol. Prog.* 20: 764-770.
[15] Han, I. S. and M. Cheryan (1995) Nanofiltration of model acetate solutions. *J. Memb. Sci.* 107: 107-113.
[16] Bowen, W. R. and H. Mukhtar (1996) Characterisation and prediction of separation performance of nanofiltration membranes. *J. Memb. Sci.* 112: 263-274.
[17] Garba, Y., S. Taha, N. Gondrexon, and G. Dorange (1999) Ion transport modelling through nanofiltration membranes. *J. Memb. Sci.* 160: 187-200.
[18] Kang, S. H. and Y. K. Chang (2005) Removal of organic acid salts from simulated fermentation broth containing succinate by nanofiltration. *J. Memb. Sci.* 246: 49-57.

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