Lactic acid (2-hydroxypropionic acid) was discovered for the first time in 1780 by a Swedish chemist Scheele and since 1881 it has been produced commercially by fermentation. Lactic acid is frequently used in food industry, esp. for the beverage production and in pharmaceutical industry, chemical industry, or medicine (Víř 1985). The recent growing interest for the manufacture of biodegradable plastics initiates a high demand for lactic acid as the raw material for polylactate production (Det al. 1995; Sova & Sova 2002). Another very promising lactic acid application is the production of environmentally friendly “green” solvents (lactate esters). They can replace traditional solvents made from petrochemical feedstocks (Tsai et al. 1999).

Generally, lactic acid can be manufactured by either biological fermentation or chemical synthesis. While the chemical synthesis produces a racemic mixture of lactic acid (\(\tau^{(+)}\)- and \(\tau^{(-)}\)-forms), fermentation technology is able to selectively synthesise the desired enantiomer due to the high stereospecificity of several lactic acid bacteria. The traditional fermentation process is carried out in the batch mode at a constant pH between 5.5 and 6.0 and at temperatures above 40°C, with molasses or other sugars as the C-source (Kaščák et al. 1996). There are a lot of studies on lactic acid fermentation, on the production of \(\tau^{(+)}\) lactic acid, and on the use of the agricultural cellulosic feedstock and waste for this purpose (Iyer & Lee 1999; Bai et al. 2003a,b; Jin et al. 2003; Wasewar et al. 2003).

Electrodialysis was used for lactic acid recovery from fermentation broth. In the first step, lactate was recovered and concentrated by desalting electrodialysis, and the second step was electroconversion of lactate to lactic acid by water-splitting electrodialysis. The final lactic acid concentration of 151 g/l was obtained. Total energy required in both electrodialysis processes was about 1.5 kWh per 1 kg of lactic acid obtained. The fermentation broth had to be pretreated prior to the electrodialysis experiments. The pretreatment consisted of ultrafiltration, decolourisation, and the removal of multivalent metal ions.

**Keywords**: lactic acid recovery; electrodialysis; pretreatment of fermentation broth
To obtain lactic acid of the required purity, efficient downstream processing is necessary. The conventional fermentation process produces calcium lactate precipitate which has to be reacidified with a strong acid; large amounts of effluents arise, in particular gypsum. It was estimated that downstream processes absorb about 80% of the production costs, indicating an urgent need for more economical downstream technologies, and for bioengineers to design and operate them (Hulse 2004).

Recently, many investigations were focused on the downstream processing of lactic acid from fermentation broth with the aim to reduce costs, to simplify the recovery procedure, to reduce the contents of effluents, and to decrease the negative impact on the surrounding environment (Timmer et al. 1994; Matsumoto et al. 1998; Frieling & Schügerl 1999; Cao et al. 2002).

Membrane techniques, mainly electrodialysis and nanofiltration, have been shown as promising ways for the lactic acid recovery (Jeanet et al. 1996; Bally 2002; Choi et al. 2002). Lee et al. (1998) used a two-stage electrodialysis method for the lactic acid recovery and Kim and Moon (2001) used one-stage electrodialysis with two- and three-compartment water-splitting electrodialysis. Li et al. (2004) proposed an electokinetic membrane bioreactor for the lactic acid production that couples fermentation with in situ lactic acid separation and the product concentration. Börgårdts et al. (1998) studied an integrated process using electrodialysis for the lactic acid recovery. Min-Tian et al. (2003) described the production of L(+) lactic acid by the electrodialysis fermentation.

**MATERIAL AND METHODS**

**Chemicals.** Sodium lactate (p. a. purity) was from Sigma, the other chemicals were chemically pure products of Lachema (Brno, Czech Republic). Demineralised water (resistivity of 18.2 MΩ.cm) was prepared from distilled water in the device Millipore – Q gradient (Molsheim, France). Granulated active charcoal Purolite AC 20G (Purolite International Ltd., UK) was used for decolourisation of the fermentation broth. Chelating resin Purolite S 940 (Purolite International Ltd., UK) and the weak cation exchanger Duolite C-433 (Fluka, Switzerland) were used for the recovery of multivalent metal ions.

**Fermentation broth.** Real fermentation broth was obtained through lactic acid fermentation using *Lactobacillus plantarum* L10 as the producent strain (the strain was obtained from the Collection of Department of Dairy and Fat Technologies, ICT Prague, Czech Republic). The lactate concentration ranged from 17 to 88 g/l. The initial medium consisted, respectively, of (g/l): glucose, 40–100; yeast extract, 5.3; KH₂PO₄, 2.6; CH₃COONa.3H₂O, 6.6; ammonium citrate 2.6; MgSO₄.7H₂O, 0.24; MnSO₄.4H₂O. The fermentation temperature was 37°C and pH was kept at 5.8 by means of NaOH (20 g/l).

**Electrodialysis equipment.** The electrodialysis laboratory unit BEL-500 (Berghof, Germany) consisted of a control unit (adjustable outputs of voltage from 0 to 50 V and of current from 0 to 3.9 A), a measuring device (conductivity and voltage), and 3 independent circuits with pumps and storage containers (for the diluate, the concentrate, and the electrode solution). The membrane stack ED 0 (Mega, Czech Republic) with 20 pairs of ion exchange membranes Ralex CMH and Ralex AMH was used for the desalting in the electrodialysis experiments. The effective membrane area was 180 cm², the distance between the membranes was 1 mm. Stack Type 500 with 4 bipolar membranes Neosepta BP-1 and 5 cation exchange membranes Neosepta CMB (Tokuyama Corp., Japan) was used for water-splitting electrodialysis, the distance between membranes being 0.5 mm. The effective membrane area was 57.6 cm².

**Operating conditions.** Electrodialysis experiments were carried out in the batch mode. Desalting electrodialysis: The electrode solution (Na₂SO₄ – 25 g/l), the concentrate (sodium lactate – initial concentration 3–104 g/l), and the diluate (the pretreated fermentation broth – the initial concentration from 15 to 88 g/l) were circulated through the corresponding compartments of the desalting stack at the flow rate of 2.4 l/min. For the constant current period, voltage of 1.5 V per one pair and current density of 7.8 mA/cm² were used. For the constant voltage period, voltage of 18 V was used. The experiments were terminated when the lactate concentration in the diluate dropped to 1–2 g/l.

In the case of the two-level electrodialysis, only the depleted diluate was replaced by another fresh fermentation broth after the first level. The conditions of the second level were the same as mentioned above.

For water-splitting electrodialysis, the following solutions were used: NaOH – 20 g/l (electrode...
solution), NaOH – 1 g/l (concentrate), and the concentrate obtained from desalting electrodialysis (diluate). Current density of 67.7 mA/cm² and voltage of 12 V were applied. The circulation flow rate was 2 l/min. When conductivity in the diluate reached the value of 5 mS/cm, the experiments were terminated.

Ultrafiltration. UF cartridge TFF 300 kD (Millipore, USA) was used for the clarification of the fermented broth.

Decolourisation, the removal of multivalent metal ions. These operations were carried out in glass columns filled with the above mentioned materials. The flow rate of the cell-free fermentation broth through the column was 1 bed volume per hour (decolourisation), resp. 2 bed volumes per hour (removing of multivalent metal ions).

Analytical methods. Lactate was determined by HPLC (Laboratorní přístroje Praha, Czech Republic); column – Ostion LG KS in H-cycle; refractometer detector RIDK 101; mobile phase – H₂SO₄ (c = 0.005 mol/l), flow rate of the mobile phase 0.5 ml/min; column temperature 85°C. The concentration of lactic acid and the concentration of NaOH were determined by titration with standard solutions of NaOH (c = 0.025 mol/l) or HCl (c = 1 mol/l), respectively, using phenolphthalein as indicator. The colour intensity of the fermentation broth was measured by spectrophotometer at the wavelength of 400 nm relative to water. Multivalent metal ions were determined by AAS method, and the biomass as a dry weight at 105°C.

Calculations. The equations were taken from Lee et al. (1998).

RESULTS AND DISCUSSION

A sequence of five separative steps for the lactic acid recovery from the fermentation broth was proposed and tested. The separation consisted of three pre-treating steps (ultrafiltration, decolourisation, the removal of multivalent metal ions), and of two electrodialysis procedures (desalting electrodialysis and water-splitting electrodialysis) (Figure 1).

Pretreatment of fermentation broth

The fermentation broth had to be pretreated prior to the electrodialysis experiment due to the high demands of the electrodialysis membranes, especially bipolar ones, on the quality of the feed solution used. The cells can be deposited on the membrane surface and create clusters in the space between the membranes, the dyes from the fermentation broth can be adsorbed on the membranes. Both phenomena significantly decrease the electrodialysis efficiency (Czytko et al. 1987; Heriban et al. 1993). Multivalent metal ions (Ca, Mg, Fe, etc.) cause irreversible damage of the bipolar
membranes, they require, consequently, less than 1 mg multivalent metal ions per litre.

**Ultrafiltration**

The broth after lactic acid fermentation contained 3.35 g/l of biomass, the cells having been removed from it by a spiral-wound module. The cell free permeate and concentrate containing 51.1 g/l of biomass were obtained. Relatively considerable fouling of membranes was found. To use a different type of the membrane cartridge would seem advisable.

**Decolourisation**

The renewable granulated active charcoal Purolite AC 20 was chosen and used for decolourisation. The results were reported earlier (Hábová et al. 2001).

The flow of the fermentation broth through the column influenced significantly the decolourisation degree. If the flow rate was as slow as 1 bed volume per hour only, the amount of the fermentation broth corresponding to the 10-fold bed volume was decolourised by 90%. At a higher flow rate, the decolourisation capacity decreased. When the decolourisation was carried out in a batch mode, the decolourisation degree was equal to that mentioned above; more decolourising agent, however, was used up.

Regeneration was done successively by water flow, by boiling with 4% HCl, by water flow, by boiling with 4% HCl and by water flow. Regenerated charcoal showed practically the same decolourisation capacity as the initial batch and its value after 15 decolourising cycles was lower by 5%.

**Removal of multivalent metal ions**

Two ion exchangers were tested. The chelating resin Purolite AC 20 G showed better results than the weak cation exchanger Duolite C-433 (Table 1). Purolite AC 20 G was used for the removal of multivalent metal ions from the fermentation broth designated for the electrodialysis experiments.

The compositions of the initial fermentation broth and of that treated with resin are given in Table 2. Depleted broth contained less than 1 mg/l of multivalent metals and the bipolar membrane could be used without risk of irreversible membrane damage.

**Electrodialysis**

Two-stage electrodialysis method was used for lactic acid recovery. Sodium lactate was removed from the pretreated fermentation broth and was concentrated by desalting electrodialysis. Further it was converted to lactic acid by water-splitting electrodialysis using bipolar membranes. Lactic acid was recovered from the model solutions and from the pretreated fermentation broth. The experiments with model solutions were focused on the determination of suitable conditions for the electrodialysis experiments and on the investigation of the time course under different conditions. The results obtained with model solutions were published earlier (Hábová et al. 2001, 2004).

**Desalting electrodialysis**

For desalting electrodialysis the fermentation broth was used with three different levels of lactate concentration: 18, 36, and 87 g/l. The results of the

---

**Table 1. Comparison of resins used for removal of multivalent metal ions (flow rate was 2 bed volumes/h)**

<table>
<thead>
<tr>
<th>Ion exchanger</th>
<th>Purolite S 940</th>
<th>Duolite C-433</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial bivalent ions concentration (mmol/l)</td>
<td>1.845</td>
<td>1.845</td>
</tr>
<tr>
<td>Final bivalent ions concentration (mmol/l)</td>
<td>0.0025</td>
<td>0.05</td>
</tr>
<tr>
<td>Treated bed volumes</td>
<td>42</td>
<td>24</td>
</tr>
<tr>
<td>Retained ion efficiency (%)</td>
<td>99.8</td>
<td>97.2</td>
</tr>
<tr>
<td>Convenient for process</td>
<td>yes</td>
<td>no</td>
</tr>
</tbody>
</table>

**Table 2. Composition of initial and treated fermentation broth (mg/l)**

<table>
<thead>
<tr>
<th></th>
<th>Ca</th>
<th>Mg</th>
<th>Fe</th>
<th>Mn</th>
<th>Zn</th>
<th>Na</th>
<th>K</th>
</tr>
</thead>
<tbody>
<tr>
<td>Initial fermentation broth</td>
<td>28.3</td>
<td>38.8</td>
<td>0.64</td>
<td>4.36</td>
<td>0.44</td>
<td>16500</td>
<td>935</td>
</tr>
<tr>
<td>Treated fermentation broth</td>
<td>0.39</td>
<td>0.47</td>
<td>0.04</td>
<td>0.02</td>
<td>0.02</td>
<td>16800</td>
<td>868</td>
</tr>
</tbody>
</table>
Desalting electrodialysis experiments are shown in Table 3. The highest lactate concentration reached was 185 g/l. The comparison of the lactate recovery from the model solutions and from the fermentation broth showed that, in the case of fermentation broth with lower initial lactate concentrations, the transport rate decreased, the energy consumption increased, and the current efficiency decreased. Nevertheless, at higher lactate concentrations in the fermentation broth a very good agreement with the model solutions was found.

Figure 2 shows the time course of two-level desalting electrodialysis of the pretreated fermentation broth.

![Time course of desalting electrodialysis](image.png)

Table 3. Desalting electrodialysis experiments

<table>
<thead>
<tr>
<th>Experiment</th>
<th>Operating time (min)</th>
<th>$V_{D_0}/V_{K_0}$</th>
<th>Concentration (g/l)</th>
<th>Current efficiency (%)</th>
<th>Energy consumption (kWh/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>initial diluate</td>
<td>final diluate</td>
<td>initial concentrate</td>
</tr>
<tr>
<td>DM 1</td>
<td>120</td>
<td>1.87</td>
<td>18.1</td>
<td>1.3</td>
<td>3.9</td>
</tr>
<tr>
<td>DM 2</td>
<td>165</td>
<td>1.87</td>
<td>17.3</td>
<td>1.3</td>
<td>28.7</td>
</tr>
<tr>
<td>DM 3</td>
<td>195</td>
<td>1.87</td>
<td>22.2</td>
<td>1.2</td>
<td>43.0</td>
</tr>
<tr>
<td>DM 4</td>
<td>195</td>
<td>7.50</td>
<td>36.3</td>
<td>2.0</td>
<td>2.8</td>
</tr>
<tr>
<td>DM 5</td>
<td>180</td>
<td>7.50</td>
<td>36.9</td>
<td>2.8</td>
<td>104</td>
</tr>
<tr>
<td>DM 6</td>
<td>215</td>
<td>7.50</td>
<td>36.6</td>
<td>0.9</td>
<td>4.8</td>
</tr>
<tr>
<td>DM 7</td>
<td>195</td>
<td>7.50</td>
<td>36.2</td>
<td>1.2</td>
<td>107</td>
</tr>
<tr>
<td>DM 8</td>
<td>215</td>
<td>4.38</td>
<td>88.3</td>
<td>3.7</td>
<td>96.4</td>
</tr>
<tr>
<td>DM 9</td>
<td>220</td>
<td>4.38</td>
<td>85.8</td>
<td>1.2</td>
<td>93.6</td>
</tr>
</tbody>
</table>

$V_{D_0}$ – initial volume of diluate, $V_{K_0}$ – initial volume of concentrate

Table 4. Water-splitting electrodialysis

<table>
<thead>
<tr>
<th>Experiment</th>
<th>Operating time (min)</th>
<th>Concentration (g/l)</th>
<th>Current efficiency (%)</th>
<th>Energy consumption (kWh/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>initial lactate</td>
<td>final lactic acid</td>
<td>final NaOH</td>
</tr>
<tr>
<td>WM 1</td>
<td>390</td>
<td>145</td>
<td>121</td>
<td>43</td>
</tr>
<tr>
<td>WM 2</td>
<td>430</td>
<td>171</td>
<td>151</td>
<td>53</td>
</tr>
</tbody>
</table>
at the end of each level. The concentration of lactate obtained during the two-level electrodialysis was about 4-times higher than that in the fermentation broth. The current efficiency was 64% and the energy consumption was 0.34 kWh/kg.

Due to a high lactate concentration and the lowest concentration of multivalent metal ions, concentrates DM 7 and DM 9 were used for the water-splitting electrodialysis experiments. The other concentrates had too low lactate concentrations or contained more than 1 mg of multivalent metal ions per litre and could not be used, consequently, for the bipolar membrane treatment.

Water-splitting electrodialysis

Water-splitting electrodialysis using bipolar membrane was used for electroconversion of sodium lactate to lactic acid. Time profile of water-splitting electrodialysis is obvious from Figure 3. Table 4 shows the results of two experiments with sodium lactate which was recovered from fermentation broth and concentrated by desalting electrodialysis. The values obtained are very similar to those achieved with model lactate solutions. The final lactic acid concentrations of 121 and 151 g/l, corresponding, respectively, to 92 and 95% conversion were obtained; the energy consumption was about 1 kWh/kg. The final base concentration was 43 and 53 g/l and the current efficiency was 70–80%.

Conclusions

Lactic acid separation from fermentation broth was studied comprehensively and the results obtained showed a very good agreement with the literature sources (Lee et al. 1998; Bailly 2002).

The results confirm that two stage electrodialysis is a suitable and efficient technique for the recovery of lactate ions from the pretreated fermentation broth and the subsequent conversion into lactic acid with respect to environmental aspects.

In the first electrodialysis step, the final lactate concentration of up to 175 g/l was obtained while the final lactic acid concentration of 151 g/l was reached in the second step. The total energy required in both electrodialysis processes representing the energy consumption for the lactate transfer and for its electroconversion to lactic acid was about 1.5 kWh/kg of lactic acid obtained.

References


Bailly M. (2002); Production of organic acids by bipolar electrodialysis: realizations and perspectives. Desalination, 144: 157–162.


Received for publication February 2, 2004
Accepted after corrections April 7, 2004

Souhrn


Pro separaci kyseliny mléčně z fermentačního média byla použita dvoustopová elektrodialýza. V prvním stupni byl laktát sodný separován a nakoncentrován pomocí odsolovací elektrodialýzy a následně ve druhém stupni byl pomocí elektrodialýzy s bipolárními membránami konvertován na kyselinu mléčnou. V prvním stupni bylo v 1 litru získáno až 175 g laktátu a z něj ve druhém stupni 151 g kyseliny mléčné. Celkově bylo třeba na získ 1 kg

Souhrn


Pro separaci kyseliny mléčně z fermentačního média byla použita dvoustopová elektrodialýza. V prvním stupni byl laktát sodný separován a nakoncentrován pomocí odsolovací elektrodialýzy a následně ve druhém stupni byl pomocí elektrodialýzy s bipolárními membránami konvertován na kyselinu mléčnou. V prvním stupni bylo v 1 litru získáno až 175 g laktátu a z něj ve druhém stupni 151 g kyseliny mléčné. Celkově bylo třeba na získ 1 kg

Received for publication February 2, 2004
Accepted after corrections April 7, 2004
kyseliny mléčné vynaložit 1,5 kWh elektrické energie. Před elektrodialýzou muselo být fermentační médium upravováno. Úprava spočívala v ultrafiltraci, odbarvování a odstranění vícemocných iontů kovů.

**Klíčová slova:** izolace kyseliny mléčné; elektrodialýza; předúprava fermentačního média

---

**Corresponding author:**

Ing. Věra Hábová, Vysoká škola chemicko-technologická v Praze, Fakulta potravinářské a biochemické technologie, Ústav kvasné chemie a bioinženýrství, Technická 5, 166 28 Praha 6, Česká republika
tel.: + 420 224 354 127, fax: + 420 224 355 051, e-mail: vera.habova@vscht.cz