Engineering is to design a process using known knowledge.			
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## **Research Interests:**

## **Research Area: Biochemical engineering**

## **Selected Achievements in the Last Five Years:**

• Periplasmic release of a recombinant creatinase from *Escherichia coli* using osmotic shock [Biochem. Eng. J., 19, 211 (2004).]

A modified osmotic shock involving pretreatment of the cells with divalent cation (Ca2+ or Mg2+) for periplasmic release of a recombinant creatinase from Escherichia coli was proposed. The pretreatment enhanced the release of lipopolysaccharide (LPS) when the cells were subsequently treated with ethylenediaminetetraacetate (EDTA), thus increased the permeability of the outer membrane. A protocol capable of dealing with suspensions of increased cell concentration was also suggested.

• Abatement of concentration polarization in ultrafiltration [J. Membr. Sci., 238, 1 (2004).]

A novel method using *n*-hexadecane/water two-phase flow to abate concentration polarization in ultrafiltration was proposed. Compared with a corresponding air-sparged ultrafiltration, the *n*-hexadecane/water ultrafiltration was shown to have a higher permeate flux when concentrating a crude lipase solution. The benefits from the *n*-hexadecane/water two-phase flow were found to be common in various ultrafiltration practices.

• Fed-batch production of lipase by Acinetobacter radioresistens using Tween 80 as the carbon source [Biochem. Eng. J., 19, 25 (2004).]

Lipase production by Acinetobacter radioresistens was examined in fed-batch cultures using Tween 80 as the carbon source. Data obtained from fed-batch cultures with DO- and pH-stat feedings showed that specific growth rate was the intrinsic factor that determined the efficiency of lipase synthesis. Based on a growth-associated pattern for lipase formation, the production of lipase with fed-batch culture could be simulated satisfactorily.

• Recovery of lipase by adsorption at the *n*-hexadecane-water interface [J. Chem. Technol. *Biotechnol.*, **78**, 1128 (2003).]

A novel process based on the hydrophobic adsorption at the n-hexadecane-water interface was developed for recovery of Acinetobacter radioresistens lipase from a pre-treated fermentation broth. Advantages of the proposed process include simple operation, low

operational cost, environmentally friendly, no requirement of pre-concentration for the enzyme solution, and negligible enzyme denaturation.

• Recovery of *Acinetobacter radioresistens* lipase by hydrophobic adsorption to *n*-hexadecane coated on a nonwoven fabric [*Biotechnol. Prog.*, **19**, 464 (2003).]

A simple and clean adsorption/desorption process was proposed for recovering Acinetobacter radioresistens lipase from fermentation broth. The adsorbent used was n-hexadecane coated on a hydrophobic nonwoven fabric (NWF). n-Hexadecane has a melting point of  $1\tilde{6}18^{\circ}$ C, and its affinity for lipase decreases markedly from liquid to solid state. Accordingly, performing the adsorption and desorption above and below, respectively, the melting point would need no extraneous materials for separation. The benefits of this process include easy preparation of adsorbent, low operational cost, no extraneous materials needed, negligible enzyme denaturation, high efficiency, and simple process simulation.

• Effect of specific growth rate on the production of a recombinant nuclease by *Escherichia* coli [Biochem. Eng. J., 14, 101 (2003).]

The effect of specific growth rate on the production of Vibrio vulnificus nuclease was investigated in fed-batch cultures of a high-yield recombinant Escherichia coli. The synthesis of nuclease was dependent on , irrespective of the feeding methods. When concerning the compromise between the nuclease yield and its production rate, the linear-gradient feeding method, being simple and adaptable, was shown to be adequate for the nuclease production.

• Production of nuclease in fed-batch culture of recombinant *Escherichia coli* [J. Chin. Inst. Chem. Engrs., **34**, 507 (2003).]

Strategies for enhancing the production of Vibrio vulnificus nuclease using a recombinant Escherichia coli were explored. Using a batch culture, a maximum nuclease yield of 3500 U/mL was obtained. Using a pH-stat fed-batch culture with the DO level maintained above 15% air saturation and with intermittent addition of nutrients, a nuclease yield of 12000 U/mL was achieved.

• A pseudo-exponential feeding method for control of specific growth rate in fed-batch cultures [*Biochem. Eng. J.*, **10**, 227 (2002).]

A simple feeding method for controlling specific growth rate in fed-batch culture was developed. This method applies a constant feed rate using a concentrate reservoir and two mixing chambers in series to simulate the exponential feeding. Fed-batch cultures with Escherichia coli showed that the present feeding method could sustain the cells growing at predetermined specific growth rates.

• Production of *Acinetobacter radioresistens* lipase with repeated batch culture in presence of nonwoven fabric [*Biotechnol. Bioeng.*, **76**, 214 (2001).]

Cultivation of Acinetobacter radioresistens on n-hexadecane for lipase production was investigated with repeated batch culture in the presence of a hydrophobic nonwoven fabric. The fabric was shown to be able to disperse n-hexadecane, to enhance cell growth and, in turn, lipase production.

## **Researches in Progress:**

• Hyaluronic acid fermentation—development of the medium, examination of fermentation conditions, and design of an efficient recovery process