

## *E.coli* Vaccine Cell Concentration

*E. coli* were successfully concentrated 17x from an initial concentration of  $1.8 \times 10^6$  cells/ml.

### Introduction

Fatal infant bovine diarrhea (calf scours) is prevented by an *E. coli* vaccine grown in batch fermentation. Once the culture reaches the desired density, cells are concentrated as the first step toward recovering cell wall-associated antigens. Selection of the proper filtration module and process conditions is essential for the cost-effective concentration of cells. A fast processing time with minimal membrane fouling is the key consideration. This bulletin describes the successful concentration of *E. coli* using hollow fiber cross-flow filtration.

### Process Conditions

Given that a short processing time was of prime importance, a 0.2 mm rated KrosFlo® module (part number K22M-230-01W) containing 2.1 m<sup>2</sup> of membrane surface was used to concentrate a 53-liter batch. A positive displacement lobe-type pump provided a recirculation rate of 45 L/min. The filtrate flow was shut off until recirculation was well established. Inlet pressure was maintained at 11 psig throughout the run. Retentate and filtrate flows were unrestricted at 0 psig, so the average trans-membrane pressure was 5.5 psig. The process fluid temperature was 10°C at the beginning of the run and rose to 15°C by the end.



KrosFlo Modules with process volume from 3 to 1,000 liters

### Results

The 53 liter batch was concentrated to just over 3 liters in only 24 minutes. This represented a 17-fold volume reduction at a steady state flux of 58 L/m<sup>2</sup>hr.

### Discussion

This concentration process is often done by continuous centrifugation. Centrifuges have certain disadvantages such as lengthy cleaning cycles, mechanical complexity (i.e. rotating seals) and significant product loss. And unless special containment features are incorporated hazardous aerosols can be created, especially when processing pathogens. Alternatively, hollow fiber cross-flow filtration offers short processing times, mechanical simplicity, safer process conditions and higher product yields and recovery. Single use modules provide consistent lot-to-lot performance without the risk, time loss and expense of cleaning procedures, thus making validation easy.



KrosFlo® Pharma Production System for processing volumes from 100 to 10,000 liters

