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**IMPROVEMENT IN PROPIONIC ACID
ACCUMULATION FROM LACTOSE BY
*PROPIONIBACTERIUM ACIDIPROPIONICI***

by

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**DEPARTMENT OF BIOCHEMICAL ENGINEERING AND
BIOTECHNOLOGY**

***SUBMITTED
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OF THE DEGREE OF***

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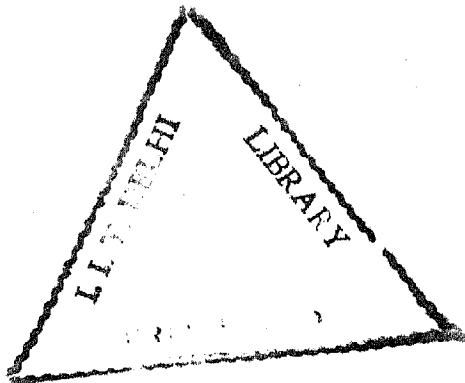
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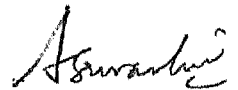
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This is to certify that the thesis entitled, "IMPROVEMENT IN PROPIONIC ACID ACCUMULATION FROM LACTOSE BY *PROPIONIBACTERIUM ACIDIPROPIONICI*", being submitted by Ms. VANDANA GOSWAMI to the Indian Institute Of Technology, Delhi for the award of the degree of **Doctor of Philosophy**, is a record of bonafide research work carried out by her under my supervision. The results contained in this dissertation have not been submitted in part or full to any other University or Institute for the award of any degree or diploma.

Date: 29/4/99



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Dedicated to

MY PARENTS

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Abstract

Batch, fed-batch and continuous fermentation studies were conducted with lactose as the substrate, at a constant temperature of 30⁰C, pH 6.5 and under anaerobic conditions for propionic acid production using *Propionibacterium acidipropionici* (ATCC 4875). In batch fermentations, when the initial concentration of lactose (S_0) was 37, 45, 50, 57 and 73 $g\ l^{-1}$, respectively 16, 19, 22., 25.3, 26.3 $g\ l^{-1}$ propionic acid was produced. Increase in S_0 from 37 $g\ l^{-1}$ to 57 $g\ l^{-1}$ resulted in increase in propionic acid accumulation but further increase in S_0 from 57 $g\ l^{-1}$ to 73 $g\ l^{-1}$ did not increase the propionic acid concentration in the fermentation broth. It, however, led to accumulation of by-products (succinic, acetic and pyruvic acids). Product inhibition studies revealed that specific growth rate of the culture decreased with the increase in propionic/ acetic acid concentration.

The average batch kinetic data ($S_0 \sim 50\ g\ l^{-1}$, pH 6.5), was used to develop an unstructured model for propionic acid fermentation. For optimal estimation of model parameters, a non-linear regression technique assisted by a computer program, was used to minimise the deviations between the model predictions and experimental results. When optimised parameters were used, model simulations were found to be close to experimental results.

Statistical validity of the model was demonstrated with an accuracy of 99%. The model featured typical growth and product formation characteristics of *Propionibacteria*.

Fed-batch fermentations were conducted. Fresh nutrient feeding strategies for fed-batch fermentations were designed and tested to improve propionic acid productivity. Significant improvement in propionic acid concentration was obtained as compared to batch fermentation. Maximum propionic acid concentration and productivity obtained from fed-batch fermentations were 37 g l^{-1} and $0.4 \text{ g l}^{-1} \text{ h}^{-1}$.

In situ cell retention devices were made using stainless steel filter of 10μ and 5μ pore size. Continuous fermentations were conducted at dilution rate, $D = 0.05 \text{ h}^{-1}$ using in situ cell retention devices (spin filter) of 10μ and 5μ . Continuous fermentation using spin filter (pore size 5μ) improved the propionic acid productivity to $0.9 \text{ g l}^{-1} \text{ h}^{-1}$ (approximately four times higher than batch fermentation). The in situ cell retention bioreactor was operated continuously for a period of one week without any clogging/ contamination problems. It was particularly suitable and economical for lab-scale, continuous propionic acid production.

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