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BIOFILM DEVELOPMENT IN A FLUIDIZED BED BIOREACTOR FOR AEROBIC PHENOL DEGRADATION

A thesis presented in partial fulfilment of the requirements for the degree of Master of Technology

in Biotechnology and Bioprocess Engineering at Massey University, Palmerston North, New Zealand

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Ki te wao nui a Tane, ki nga awa a Tangaroa ki uta, ki nga moana a Tangaroa ki tai. Ki te ika a Maui, ki te waka a Maui hoki.

To the land of the long white cloud.

ABSTRACT

The main objective of this thesis was to follow the biofilm development during start-up of a fluidized bed bioreactor with the help of digital image processing. A mixed microbial culture immobilized on activated carbon particles was grown on phenol as sole carbon source in an aerobic liquid-solid fluidized bed bioreactor. The effect of different reactor temperatures and of different inlet phenol concentrations on the system behaviour during start-up was investigated.

The phenol inhibition kinetics of the culture was studied in batch culture experiments. Three substrate inhibition models (Teissier-Edwards, Haldane and Aiba-Edwards models) were fitted to the experimental data. There was no statistically significant difference in the goodness of fit between the equations. The phenol concentrations at which the fitted functions go through their maximum value were between 57 and 88 mg/l, corresponding to specific growth rates of between 0.64 and 0.65 h⁻¹.

A fluidized bed system was developed and tested. The test runs showed that the most critical part of the apparatus was the liquid distributor at the bottom of the fluidized bed reactor. Other critical factors that were decided on during the test runs were initial bed expansion, flow rate, support particle size, and amount of support particles used, these parameters all being interdependent.

The fluidized bed experiments proved that the use of image analysis techniques is a very effective means of measuring the mean biofilm thickness on fluidized support particles. Micrographs of the bioparticles were analyzed with the help of a software-controlled system. The software identified the circumference of the particle core and the bioparticle. The mean biofilm thickness was calculated

from the projected areas and the perimeters of the bioparticle and the particle core applying a simple trapezoid formula.

In all fluidized bed experiments, the bed stratified into layers (in most cases two or three) containing bioparticles with different biofilm thickness and different biofilm structure. The main focus was on the development of the biofilm in the top layer. The phenol reduction was only small due to a very short hydraulic retention time. Conversely, the dissolved oxygen concentration in the outlet reached very low values. Thus, the system was oxygen-limited.

Different reactor temperatures led to distinct differences in the morphology of the biofilm in the top layer. Without temperature control, i.e. at ~17°C, and at 30°C, a loose, fluffy, unevenly shaped, thick biofilm developed, whereas at 25°C the biofilm was firm and relatively even in shape, the final thickness remaining far below the values reached by the fluffy biofilm. Since the biofilm that developed at 25°C showed the most favourable characteristics, this temperature was used for the experiments examining the effect of different inlet phenol concentrations.

The biofilm thickness in the top layer increased the fastest at an inlet phenol concentration of 100 mg/l, followed by 35 mg/l, then 330 mg/l and finally 520 mg/l. In the batch culture experiments, the same order had been found for the specific growth rates at phenol concentrations of the above values. In the case of the few observations obtained at non-inhibitory phenol concentrations, the biofilm density increased with increasing phenol concentration. At inhibitory phenol concentrations the flow patterns in the reactor were very different, thus these patterns were the dominating factor influencing the biofilm density.

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NOMENCLATURE

A projected area of a particle

b biofilm thickness

CoA Coenzyme A

d diameter

d_L long diameter of an ellipsoid

d_s short diameter of an ellipsoid

d_{equiv.} equivalent diameter of a sphere

F_{i1} F-ratio between Model i and Model 1

HRT hydraulic retention time

i inhibition constant

K, k₁, k₂ kinetic constants

K_i inhibition constant

K_s saturation constant

m, n constants

NAD Nicotinamide adenine dinucleotide

NADPH Nicotinamide adenine dinucleotide phosphate (reduced)

P perimeter

R² coefficient of determination

r_s volumetric substrate uptake rate

S substrate concentration

S₀ initial substrate concentration

S' substrate concentration at the onset of the exponential growth

phase

S* threshold substrate concentration (below which organisms grow

apparently without inhibition)

S_m total inhibition concentration

Std. standard deviation

x biomass concentration

Y_{x/s} growth yield coefficient

Greek letters

μ specific growth rate

μ_m maximum specific growth rate

μ* maximum observable specific growth rate

 σ^2 variance

Subscripts

b bioparticle

c carrier particle

i inlet

o outlet