MATHEMATICAL MODEL ON THE CELLULASE ENZYMES PRODUCTION AS A FUNCTION OF VISCOSITY

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This thesis is dedicated to my parents

Mostafa** and **Narjes

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Abstract

As cellulosic material is the most abundant and renewable source of organic matter, ethanol can be produced cheaply from cellulosic waste. The cellulose must be hydrolyzed to glucose before it can be fermented to ethanol. The enzymatic hydrolysis of cellulose using cellulase enzymes is considered an environmental- friendly and less energy intensive alternative.

White-rot fungi belong to the wood-destroying *basidiomycetes* and are well known as the only micro-organisms responsible for the enzyme hydrolysis of all major wood polymers, including lignin, cellulose and hemicelluloses.

On the other hand, the production cost of cellulase enzymes is a limitation in ethanol fermentation. Therefore, the goal is to produce the enzymes economically by means of high-yield cellulase fermentation.

Different type of bacteria and fungi can be used to achieve to this goal. In the present study, the ability of *Pycnoporus sanguineus*, a white rot fungus to produce cellulase was investigated since there are no reports of its cellulase production available. Different types of basal media were tested and enzyme activity was measured in the presence of cellulose powder, carboxymethylcellulose (CMC), Tween 80 and yeast extract. The cost of each medium is an important consideration. The highest amount of reducing sugar production was observed when CMC and Tween 80 were

used as the components of the medium. This formulation is the most cost effective among the medium formulation.

The study also presents the development of a mathematical model for cellulase production in submerged fermentation as a function of medium's viscosity. To achieve this, the fungus was grown in two different media on agar plates. Then, the inoculation was carried out from these plates to a medium formulation in the shake flasks using variable concentration of CMC for determination of CMCase activity. Enzyme activity and viscosities of media were measured simultaneously. Mathematical function was explored for each media and then a unique model was reported for each media.

Abstrak

Memandangkan sebatian selulos merupakan bahan organik yang banyak dan sumber yang boleh diperbaharui secara semula jadi, etanol dapat dihasilkan dengan kos yang rendah daripada sisa-sisa selulos. Selulos harus di hydrolisiskan kepada glukos sebelum proses penapaian etanol dapat dilakukan. Pencernaan (hidrolisis) selulose dengan menggunakan enzim selulase merupakan satu kaedah yang tidak mencemarkan alam sekitar dan tidak membebankan.

Sebaliknya, kulat putih reput kepunyaan basidiomycetes memusnahkan kayu dan dikenali sebagai mikroorganisma sahaja bertanggungiawab untuk pemineralan semnua polimer kayu yang utama, termasuk lignin, selulosa dan hemicelluloses.

Penggunaan enzim selulose dalam proses penapaian etanol memerlukan kos penghasilan yang tinggi. Oleh itu, tujuan utama kajian ini adalah untuk menghasilkan enzim ini dalam kuantiti yang banyak tanpa mengeluarkan kos yang tinggi.

Berbagai jenis bacteria dan kulat boleh digunakan untuk mencapai matlamat ini. Dalam kajian ini, kemampuan *Pycnoporus sanguineus* iaitu cendawan pelapuk putih yang dapat menghasilkan selulase telah dikaji dan diselidik kerana tiada laporan kajian mengenai pengahasilan enzim ini daripada spesies tersebut. Berbagai jenis media dasar diuji dan aktiviti enzim diukur dengan adanya serbuk selulosa carboxymethyl cellulose (CMC), Tween 80 and ekstrak yis.

Kemudian, kos setiap media akan menjadi pelaburan. Jumlah tertinggi mengurangkan pengeluaran gula apabila CMC dan Tween 80 telah digunakan sebagai media. Mujurlah media ini adalah media termurah antara yang lain yang telah disiasat.

Disertasi ini juga menerangkan pembangunan model matematik untuk penghasilan selulase dalam fermentasi terendam sebagai fungsi dari viskositi medium. Untuk mencapai matlamat ini, kulat ditumbuhkan dalam dua media yang berbeza pada plat agar . kemudian, inokulasi dilakukan daripada plat agar ke dalam flask yang mengandungi medium yang telah diformulasikan dimana kepekatan CMC telah dimanipulasikan untuk penentuan aktiviti CMCase. Aktiviti enzim dan viskositi media diukur serentak. Fungsi matematik yang menyeluruh telah dijalankan dan model yang unik telah dipersembahkan untuk setiap media.

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Publication

The following paper has been published from this thesis:

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Abbreviations

AR	Average Residual
Avicelase	Avicel hydrolysis activity
BG	β-glycosidase
CBH	Cellobiohydrolase
CMC	Carboxymethylcellulose
CMCase	Carboxymcthyl cellulase
Ea	Enzyme activity
EG	Endoglucanase
Fp	Filter paper
FPase	Filter paper hydrolyse
mol. wt.	molecular weight
Glu	Glucose
R^2	Coefficient of Multiple Determination
R_a^2	Adjusted Coefficient of Multiple Determinations
RSS	Residual Sum of Squares
SEE	Standard Error of the Estimate
SR	Sum of Residuals
T	Time