SCREENING FOR LIPASE ACTIVITY BY MD 3236
BACTERIA ISOLATED FROM SOIL SAMPLES

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SCREENING FOR LIPASE ACTIVITY BY MD 029b BACTERIA ISOLATED FROM CORAL MUCUS

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SCREENING FOR LIPASE ACTIVITY BY MD 029b BACTERIA ISOLATED FROM CORAL MUCUS

By

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CO_2	-	Carbon dioxide
g	-	Gram
g/l	-	Gram per liter
h	-	Hour
HCl	-	Hydrochloride acids
H_2S	-	Hydrogen sulphide
H_2O	-	Water
H_2O_2	-	Hydrogen dioxide
HPLC	-	High-performance liquid chromatography
ID	-	Identity
ml	-	Milliliter
min	-	Minute
M	-	Molar
mg	-	Milligram
MR-VP	-	Methyl Red - Voges-Proskauer test
NaCl	-	Sodium chloride
NaOH	-	Sodium hydroxide
N_2	-	Nitrogen
NO_2	-	Nitrite
NO_3	-	Nitrate
nm	-	Nano meter
O_2	-	Oxigen
pH	-	pH
rpm	-	Rotation per minute
s	-	Second
S	-	Sulphur
SIM	-	Sulfur Reduction Test, Indole Production, Motility
TSI	-	Triple Sugar Iron Test

v/v	-	volume/volume
w/v	-	weight/volume
μm	-	Micrometer
°C	-	Degree Centigrade
μl	-	Micro litter
%	-	Percentage
α	-	Alpha
β	-	Beta
γ	-	Gamma

ABSTRACT

Lipase is a class of enzyme which catalyzes the hydrolysis of long chain triglycerides, which constitutes to the most important group of biotechnological applications. In this study, the ability to produce this enzyme by marine bacteria MD 029b isolated from coral mucus at Pulau Bidong Terengganu, was investigated. The identification of the bacterium was confirmed by several biochemical tests based on their morphological and phenotypic characteristics. The results suggested that the bacteria is *Alteromonas* sp. Prior to the induction of lipase by the bacteria, three different media have been tested to determine the best culture medium: nutrient broth in distilled water, nutrient broth in sea water and Zobell medium; nutrient broth in sea water was found to contain the highest bacterial growth. The preliminary step of lipase induction that is the pre-inoculum preparation has been carried out. The induction of lipase with olive oil as substrate was investigated. Several reaction parameters were studied to obtain the optimum conditions for lipase activity: incubation time, temperature and amount of olive oil as substrate. The result obtained showed that the free fatty acids released after 18 hours of incubation was significantly higher ($P<0.05$) compared to the 6, 12, and 24 hours. There was significant difference ($P<0.05$) between 15°C - 27°C and 27°C - 37°C but no significant difference between 15 - 37°C. Furthermore, no significant difference ($P>0.05$) was observed between 2% to 3% but there was significant difference between 1% to 2% of olive oil. Therefore, the optimum free fatty acids were released after 18 hours of incubation, at 27°C and using 1% to 2% of olive oil as substrate. These results indicate that *Alteromonas* sp is a potential source of lipase producer.

PENYARINGAN AKTIVITI LIPASE DARIPADA BAKTERIA MD 029b YANG DIPENCILKAN DARIPADA MUKUS BATU KARANG

ABSTRAK

Lipase adalah satu kelas bagi enzim-enzim yang memangkinkan hidrolisis rantai trigliseride yang panjang, dan merupakan kumpulan yang paling penting dalam aplikasi-aplikasi bioteknologi. Dalam kajian ini, keupayaan untuk menghasilkan enzim ini daripada bakteria MD 029b yang dipencilkan daripada mukus batu karang di Pulau Bidong Terengganu, telah dikaji. Pencirian untuk mengesahkan identifikasi bakteria, beberapa ujian biokimia telah dijalankan berdasarkan kepada ciri-ciri mofologi dan fenotip. Keputusan ujian mencadangkan bahawa bakteria adalah *Alteromonas* sp. Sebelum induksi lipase oleh bakteria, tiga media berlainan telah diuji untuk menentukan kultur medium yang terbaik: broth nutrien dalam air suling, broth nutrien dalam air laut dan medium Zobell; broth nutrien dalam air laut didapati memberikan pertumbuhan bakteria tertinggi. Langkah persediaan untuk pra-inokulum telah dijalankan sebelum menjalankan induksi lipase. Induksi lipase dengan minyak zaitun sebagai substrat telah dikaji. Beberapa tindak balas parameter telah diuji untuk menentukan aktiviti lipase yang optima: masa pengeraman, suhu, amaun minyak zaitun sebagai substrat. Keputusan menunjukkan asid lemak yang dibebaskan selepas 18 jam pengeraman adalah lebih signifikan ($P<0.05$) berbanding dengan 6, 12 dan 24 jam. Terdapat perbezaan signifikan ($P<0.05$) di antara 15 - 27°C tetapi tiada perbezaan signifikan di antara 15 - 37°C. Tambahan pula, tiada perbezaan signifikan ($P>0.05$) didapati di antara 2% -3% tetapi terdapat perbezaan signifikan ($P<0.05$) diantara 1% - 2% minyak zaitun sebagai substrat. Oleh itu, jumlah asid lemak bebas optima didapati selepas 18 jam pengeraman, pada suhu 27° C dengan menggunakan 1% hingga 2% minyak zaitun. Keputusan-keputusan ini menunjukkan bahawa *Alteromonas* sp adalah sumber pengeluar enzim lipase yang berpotensi.