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Cytotoxic effects of Acanthamoeba Lysates on t-lymphoblastic cell line (cem-ss) / Iliana Fauzi.

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PERPUSTAKAAN
KOLEJ UNIVERSITI SAINS & TEKNOLOGI MALAYSIA
21030 KUALA TERENGGANU

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CYTOTOXIC EFFECT OF *ACANTHAMOEBA* LYSATES ON T-LYMPHOBLASTIC CELL LINE (CEM-SS)

By :

Iliana Binti Fauzi

Research Report submitted in partial fulfillment of
the requirements for the degree of
Bachelor of Science (Biological Sciences)

Department of Biological Sciences
Faculty of Science and Technology
KOLEJ UNIVERSITI SAINS DAN TEKNOLOGI MALAYSIA
2005

This project should be cited as:

Iliana, F. 2005. Cytotoxic effect of *Acanthamoeba* Lysates on T-Lymphoblastic leukemic cell line (CEM-SS). Undergraduate thesis, Bachelor of Science in Biological Sciences, Faculty of Science and Technology, Kolej Universiti Sains dan Teknologi Malaysia, Terengganu. 88p.

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FAKULTI SAINS DAN TEKNOLOGI
KOLEJ UNIVERSITI SAINS DAN TEKNOLOGI MALAYSIA

PENGAKUAN DAN PENGESAHAN LAPORAN PROJEK PENYELIDIKAN I DAN II

Adalah ini diakui dan disahkan bahawa laporan penyelidikan bertajuk: CYTOTOXIC EFFECT OF *ACANTHAMOEBA* LYSATES ON T-LYMPHOBLASTIC LEUKEMIC CELL LINE (CEM-SS) oleh Iliana binti Fauzi no. matrik: UK 6626 telah diperiksa dan semua pembetulan yang disarankan telah dilakukan. Laporan ini dikemukakan kepada Jabatan Sains Biologi sebagai memenuhi sebahagian daripada keperluan memperolehi Ijazah Sarjana Muda Sains – Sains Biologi, Fakulti Sains dan Teknologi, Kolej Universiti Sains dan Teknologi Malaysia.

Disahkan oleh:

.....

Penyelia Utama
Nama: PROF. MADYA DR. NAKISAH BT. MAT AMIN
Pensyarah
Jabatan Sains Biologi
Fakulti Sains dan Teknologi
Kolej Universiti Sains dan Teknologi Malaysia
21030 Kuala Terengganu.
Cop Rasmi:

Tarikh: 11/4/05

.....

Penyelia Kedua (jika ada)
Nama: DR. MD. MAHFUZUL HOQUE
LECTURER
Department of Biological Science
Faculty of Science & Technology
University College of Science and Technology Malaysia
21030 Kuala Terengganu.
Cop Rasmi

Tarikh: 11/4/05

.....

Ketua Jabatan Sains Biologi
Nama:
Cop Rasmi:

Tarikh: 11/4/05

PROF. MADYA DR. NAKISAH BT. MAT AMIN
Ketua
Jabatan Sains Biologi
Fakulti Sains dan Teknologi
Kolej Universiti Sains dan Teknologi Malaysia
(KUSTEM)
21030 Kuala Terengganu.

ACKNOWLEDGEMENTS

Assalamualaikum w.b.t

I would like to especially thanks my supervisor, Associate Professor Dr. Nakisah Mat Amin for her help, guidance, encouragement, critism and patient, without whom I would not be able to go through my final year project successfully. Above all, thanks you so much for being so generous with ideas. I would also like to thank my co-supervisor, Dr. Mahfuzul Hoque for being helpful and kind in sharing his knowledge for accomplishing my project. My utmost appreciation to Professor Dr. Abdul Manaf Ali, Dr. Chua Tse San, and Dr. Effendy Abdul Wahid for being very helpful throughout my project period.

To all the staffs of Faculty Science and Technology, KUSTEM, thanks you so much for the guidance given. Not forgetting to all the staffs and master students of Laboratory of Biological Molecular and Cell (LBMS), Universiti Putra Malaysia especially to Kak Madihah, Kak Rohaya and Kak Ana, I am truly grateful for the kindness and guidance from which I was able to complete my project work successfully. Special thanks also to Kak Fatimah for sharing her knowledge and giving me guidance while doing this project.

I gratefully acknowledge the following people for their time, advice and generous contributions for making this piece of work possible: staffs of the Biotechnology 3

Laboratory, INOS especially Kak Suhaila and Kak Tie; students master who shared working with me at INOS especially Kak Siti Faezah, Kak Nurul, Kak Afna, Kak Odah, Kak Huda and Abang Jat; Cik Ku Naiza, Cik Azlina and Kak Ezi from Biochemistry Laboratory ; Puan Zarina and Puan Mahidawati from Microbiology Laboratory; and last but not least to the staffs of the Electron Microscopy Preparation Room especially to En. Nasir and Kak Ita. To my fellow group of final year project; Rainee, Harizal and Yew Foo On, thanks a zillion for sharing the work load of keeping the lab tidy with me.

Special grateful also to my beloved parent, Hj. Fauzi bin Hj. Yaa'cob and Hjh. Wan Norizan binti Wan Mahmood; and my siblings; Iqram, Izazi Fakhira and Izzul Faruq who always giving me supports and advices without feeling bored. All the love and support given each time have strengthened up my soul and gave me confident to accomplish this study.

Last but not least, to all my course mates and person who did not mention here, thanks for helping and being supportive to me. May Allah bless all of you.

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LIST OF ABBREVIATION

rpm	-	round per minute
CD_{50}	-	cytotoxic dose of 50%population
RPMI	-	Roswell Park Memorial Institute Medium Culture
FBS	-	Fetal Bovine Serum
PBS	-	Phosphate Buffer Saline
DMSO	-	Dimetil Sulfoxide
MTT	-	3-[4,5-dimethylthioazol-2-yl]-2-5-diphenyltetrazolium Bromide
ELISA	-	Enzyme Linked Immunosorbant Assay
NaHCO3	-	sodium bicarbonate
PAS	-	Page's amoeba saline
NA	-	nutrient agar
NNA	-	non-nutrient agar

LIST OF APPENDIXES

ABSTRACT

Cytotoxic effects of four amoeba lysates were studied on T-lymphoblastic leukemic cell line. Three types of amoeba lysates were isolated from marine sources and identified as M, HM, and HU. A lysate derived from a pathogenic species was identified as AK lysate. This amoeba were isolated from corneal scrapping of a patient who had Keratitis. Cytotoxic activities of amoeba lysates were tested against CEM-SS (T-lymphoblastic leukemia) using MTT, a colorimetric tetrazolium-based assay. The level of MTT cleavage in this assay i.e. transformation of tetrazolium salt to form formazan by viable cells was found to be directly proportional to the number of cells. The concentration of lysates that killed cells by 50% (CD_{50}) with respect to untreated cell population, varied among the cell lysates tested. AK lysate was found to be the most potent lysate followed by HU lysates with the CD_{50} of 38μg/mL and 75 μg/mL, respectively. HM and M lysates shared the same CD_{50} values, 150 μg/mL. Cytotoxic effect of amoeba lysates were also studied on time-course cytotoxic effect which viability of CEM-SS cells were calculated within 24 hours to 72 hours after treated with each amoeba lysates at their CD50 values. Amoeba lysates showed reduction of cell viability denoted by rapid fall of cell viability within 24 hours with AK lysate showed the most potent to cytotoxic effect towards CEM-SS cell line followed by HU, HM and M. The results of this study suggest that these amoeba lysates have potential to be used as anti-leukemic agents.

KESAN SITOTOKSIK OLEH SEL-SEL LISAT *ACANTHAMOEBA* PADA SEL-SEL T-LIMFOBLASTIK LEUKEMIA (CEM-SS).

ABSTRAK.

Kesan sitotoksik oleh 4 sel lisat amoeba telah dikaji ke atas sel-sel T-limfoblastik leukemia. Tiga jenis sel lisat amoeba diperolehi daripada sumber laut dikenal pasti sebagai M, HM, dan HU. Satu spesies patogenik dikenal pasti sebagai sel lisat AK, diperolehi daripada kornea pesakit yang menghidap Keratitis. Aktiviti sitotoksik 4 jenis sel lisat ini telah diuji ke atas sel-sel CEM-SS (T-Limfoblastik leukemia) menggunakan kaedah MTT. Tahap penukaran MTT penting di mana penukaran garam tetrazolium, MTT kepada bentuk formazan oleh sel-sel hidup adalah berkadaran dengan bilangan sel-sel hidup selepas rawatan. Kepekatan sitotosik sel lisat amoeba yang membunuh 50% (CD_{50}) sel-sel CEM-SS adalah berbeza dengan merujuk kepada populasi sel–sel kawalan. Sel lisat AK dikenal pasti sebagai sel lisat yang paling kuat memberi kesan sitotoksik pada sel-sel CEM-SS diikuti oleh sel lisat HU dengan nilai CD_{50} iaitu 38μg/mL dan 75μg/mL masing-masing. Sel lisat HM dan M mempunyai nilai (CD_{50}) yang sama iaitu 150 μg/mL. Ujian ketoksikan terhadap masa turut dijalankan di mana kebolehidupan sel-sel CEM-SS dikira selepas dirawat dengan sel-sel lisat amoeba setiap 24 jam selama 72 jam. Penurunan kadar kebolehidupan sel-sel CEM-SS yang cepat berlaku dalam tempoh 24 jam. Sel lisat AK menunjukkan nilai sitotoksik yang paling kuat diikuti oleh HU, HM dan M. Hasil kajian ini mencadangkan bahawa sel-sel lisat amoeba ini berpotensi untuk digunakan sebagai agen anti leukemia.