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

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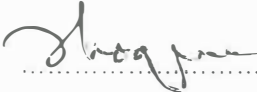
PENGAKUAN DAN PENGESAHAN LAPORAN  
PROJEK PENYELIDIKAN I DAN II

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 memperoleh Ijazah Sarjana Muda Sains – Sains Biologi, Fakulti Sains dan Teknologi, Kolej Universiti Sains dan Teknologi Malaysia.


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## LIST OF ABBREVIATION

rpm	-	round per minute
CD <sub>50</sub>	-	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
NaHCO <sub>3</sub>	-	sodium bicarbonate
PAS	-	Page's amoeba saline
NA	-	nutrient agar
NNA	-	non-nutrient agar



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## 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  $38\mu\text{g/mL}$  and  $75\mu\text{g/mL}$ , respectively. HM and M lysates shared the same  $CD_{50}$  values,  $150\mu\text{g/mL}$ . 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  $CD_{50}$  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 berkadar dengan bilangan sel-sel hidup selepas rawatan. Kepekatan sitotoksik 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  $38\mu\text{g/mL}$  dan  $75\mu\text{g/mL}$  masing-masing. Sel lisat HM dan M mempunyai nilai ( $CD_{50}$ ) yang sama iaitu  $150\mu\text{g/mL}$ . 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.