CYTOPATHIC EFFECTS OF *Acanthamoeba* spp IN HUMAN BREAST ADENOCARCINOMA (MCF-7) CELL CULTURE

By

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PENGAKUAN DAN PENGESAHAN LAPORAN PROJEK PENYELIDIKAN I DAN II

Adalah ini diakui dan disahkan bahawa laporan penyelidikan bertajuk: Cytopathic Effect of Acanthamoeba spp In Human Breast Adenocarcinoma (MCF-7) Cell Culture

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LIST OF ABBREVIATIONS

CPE	cytopathic effect
MCF-7	human breast adenocarcinoma cell line designation
PAME	primary amebic meningoencephalitis
GAE	granulomatous amebic encephalitis
CNS	central nerve system
A. castellanii	*Acanthamoeba castellanii*
A. polyphaga	*Acanthamoeba polyphaga*
A. hatchetti	*Acanthamoeba hatchetti*
A. culbertsoni	*Acanthamoeba culbertsoni*
A. rhysodes	*Acanthamoeba rhysodes*
A. griffini	*Acanthamoeba griffini*
A. quina	*Acanthamoeba quina*
A. lugdunensis	*Acanthamoeba lugdunensis*
A. lenticulata	*Acanthamoeba lenticulata*
A. astronyxis	*Acanthamoeba astronyxis*
N. gruberi	*Naegleria gruberi*
N. jadini	*Naegleria jadini*
CCAP	the Culture Collection of Algae & Protozoa
HKL	Hospital Kuala Lumpur
ECACC	European Collection of Cell Cultures
PAS	Page's ameba saline

PBS	phosphate buffered saline
CGM	complete growth media
RPMI 1640	Roswell Park Memorial Institute
FBS	fetal bovine serum
NaCl	sodium chloride
KCl	potassium chloride
KH_2PO_4	potassium dihydrogen orthophosphate
Na_2HPO_4	disodium hydrogen phosphate
$NaHCO_3$	sodium bicarbonate
EDTA	ethylenediaminetetracetic acid disodium salt
dH_2O	distilled water

ABSTRACT

In this investigation, the cytopathic effects (CPE) properties of axenically grown of *Acanthamoeba* spp were conducted *in vitro* on human breast adenocarcinoma, MCF-7 cell cultures. This is the first report describing the CPE of *Acanthamoeba* spp on MCF-7 cell cultures. In this study, *Acanthamoeba* trophozoites were incubated with MCF-7 cell cultures at different amoeba: target cell ratio, which were 1:1, 1:10 and 1:100. The CPE were observed at interval periods of 1h, 12h, 24h, 48h and 72h after the co-cultures incubation. The results showed that the amoebae caused destruction of the monolayer of the cell cultures at varying degrees. The time requested for amoebae to destroy the cultured cells monolayer depended on the amoeba: target-cell ratio. Common morphological features of affected cells alterations such sharpening of cell ends, elongation, packing and picnosis of the nuclei caused by the amoebae were observed.

KESAN SITOFATIK OLEH *Acanthamoeba* spp DALAM KULTUR SEL BARAH PAYU DARA (MCF-7)

ABSTRAK

Dalam kajian ini, kesan sitofatik (CPE) *Acanthamoeba* spp yang dikulturkan secara aksenik ke atas kultur sel barah payu dara dilakukan secara *in vitro*. Laporan ini merupakan pertama kali melaporkan kesan sitofatik yang disebabkan oleh *Acanthamoeba* spp dengan menggunakan kultur sel MCF-7. Dalam kajian ini, trofozoit *Acanthamoeba* dieram dengan 'monolayer' sel barah payu dara pada nisbah ameba: sel barah yang berlainan, iaitu 1:1, 1:10 dan 1: 100. Kesan sitofatik diperhatikan dalam tempoh masa 1 jam, 12 jam, 24 jam, 48 jam dan 72 jam selepas pengeraman ko-kultur. Keputusan yang diperolehi menunjukkan ameba yang digunakan dapat menyebabkan pemusnahan 'monolayer' kultur sel tersebut pada tahap yang berbeza-beza. Tempoh masa yang diperlukan oleh ameba untuk memusnahkan 'monolayer' kultur sel bergantung kepada nisbah dan kepekatan awal ameba. Secara amnya, perubahan rupabentuk kultur sel seperti ketajaman hujung sel, pemanjangan sel dan piknosis nukleus yang disebabkan oleh ameba dapat dilihat.