APPENDICES

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APPENDIX A

INSTRUMENTS

List of instruments used in this study	7
Instruments	
Centrifuge	
ELISA reader	
Incubator	
Laminar Flow	
Microcentrifuge	
pH meter	
Refrigerator (-20°)	
Sepectrophotometer UV-1201	
Water bath	

X A NTS Source Kendo Laboratory, Germany Tecan, Austria Thermo electron corporation, USA NUAIRE, USA NUAIRE, USA Kendro, Germany Precisa, Switzerland Whirlpool, Thailand Shimadzu Co., Japan Memmert, Germany

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APPENDIX B

REAGENTS AND BUFFERS PREPARATION

1. Regent for Cell culture

1.1. Dulbecco's Modified Eagle Medium (DMEM)

DMEM powde	er	13.54 g
NaHCO ₃		0.200 g
HEPES		1.150 g
ddH ₂ O		800 ml

Stirred until dissolved and adjust pH with acetic acid. Dissolved in ddH₂O and adjust volume to 1,000 ml. Filtered through 0.2 μ m Millipore membrane filter Mixed and stored at 4 °C.

1.2. Complete DMEM culture medium

DMEM medium89 mlFetal bovine serum (FBS)10 mlPennicillin (10,000 units/ml)/1 ml

Streptomycin (10,000 units/ml)

.3. Phosphate buffer saline (PBS pH 7.2)		
NaCl	8.000 g	
KCl	0.200 g	
Na ₂ HPO ₄	1.150 g	

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KH₂PO₄

0.200 g

Distilled water

800 ml

Adjust pH to 7.2 by adding 1N HCl or 1N NaOH

Adjust volume to 1000 ml.

Filter with 0.2 μ m millipore filter, Store at room temperature

1.4. 0.25% Trypsin EDTA

Disodium EDTA	0.025 g
Trypsin	0.25 g
PBS	100 ml

1.5. Hank's Balanced Salt Solution (HBSS)

KCl	0.400 g
KH ₂ PO ₄	60 mg
NaCl	8 g
NaHCO ₃	0.350 g
NaH ₂ PO ₄ •7H ₂ O	90 mg
Glucose	1000 g

addH20 100 ml Copyright[©] by Chiang Mai University All rights reserved

2. Reagent for ELISA

2.1. Phosphate buffer saline (PBS pH 7.2)

NaCl	8.000 g
KCl	0.200 g
Na ₂ HPO ₄	1.150 g
KH ₂ PO ₄	0.200 g
Distilled water	800 ml
Adjust pH to 7.2 by adding 1N	HCl or 1N NaOH

Adjust volume to 1000 ml.

Filter with 0.2 µm millipore filter, Store at room temperature

2.2. Coating buffer (0.1 M Carbonate/Bicarbonate pH 9.6)

Na ₂ CO ₃	1.06 g	
NaHCO ₃	1.26 g	
Distilled water	200 ml	
Mix well, Adjust pH to 9.6 with concentrated HCl		
Adjust volume to 250 ml, Storage at 4° C		

2.3. Blocking buffer (2% Skimmed milk)

Skimmed milk PBS 2 g 100 ml

Mix well, prepare before use

2.4. Stopping solution (1 N HCl)

HCl	82.8 ml
Sterile distilled water	917.2 ml

Slowly dropwise HCl to distilled water, Store at room temperature

2.5. 0.05% Tween-PBS

PBS pH 7.2	500 ml
Tween	250 ml

Mix well, Store at room temperature

Reagent of XTT assay 3.

3.1. XTT reagent

XTT powder 5 mg 5 ml

Incomplete DMEM

3.2. N-methyl dibenzopyrazine methyl sulfate (PMS)

PMS powder 1.53 mg

 $1 \, \mathrm{ml}$ PBS

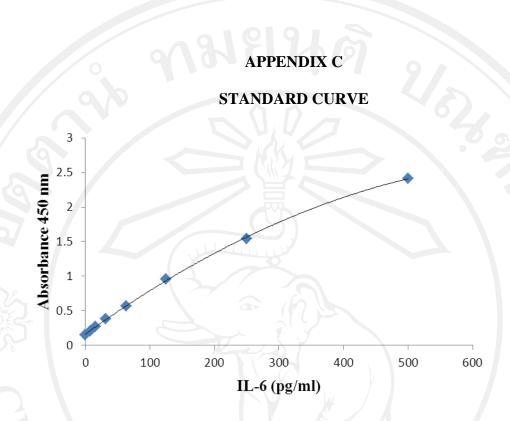


Figure I. The standard curve of IL-6 concentration determined by ELISA

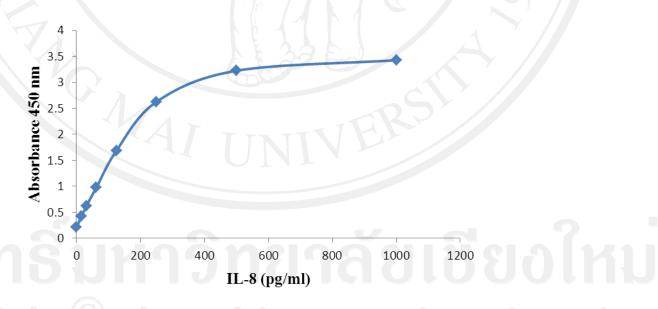


Figure II. The standard curve of IL-8 concentration determined by ELISA

CURRICULUM VITAE

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2005	Certificated of high school	Pasang School
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Presentations and publications

- Udomsom S, Khunatorn Y, Taneyhill K. Toxicity Testing of Particulate Matter Collected from Chiang Mai using the Developed Direct Alveolar Epithelial Cells-Air Exposure Prototype. Proceedings: 1st ASEAN Plus Three Graduate Research Congress. 2012. (Oral presentation and proceeding)
- 2. Udomsom S, Khunatorn Y, Taneyhill K. Toxicity Testing of Particulate Matter Collected from Chiang Mai using the Developed Direct Alveolar Epithelial Cells-Air Exposure Prototype. The conference on Recent Advances in Diagnosis and Monitoring of Liver Diseases at the Annual Meeting of the Faculty of Associated Medical Sciences, Chiang Mai University. 2012. (Poster presentation)

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