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ศูนย์วิทยทรัพยากร  
จุฬาลงกรณ์มหาวิทยาลัย



APPENDICES

ศูนย์วิทยทรัพยากร  
จุฬาลงกรณ์มหาวิทยาลัย

## APPENDIX I

## REAGENTS AND EQUIPMENT

## 1) Media and reagent

Absolute ethanol	Merck, Germany
Agarose (ultrapure)	GIBCO BRL, USA
Arabinose	Sigma, USA
EDTA	Amresco, USA
Ethidium bromide	Amresco, USA
Glacial acetic	Merck, Germany
Glucose	Difco, USA
Horse serum	GIBCO BRL, USA
Hydrochloric acid	Merck, Germany
Inositol	Sigma, USA
Lactose	Oxoid, USA
Manitol	Difco, USA
Mannose	Sigma, USA
Methanol	Merck, Germany
Sodium acetate	Merck, Germany
Sodium chloride	Merck, Germany
Sucrose	Hermann, Germany
Tris base	Sigma, USA
Triton X	Merck, Germany
Trypticase soy broth	Oxoid, USA

## 2) Equipments

ABI prism 310 automated sequencer	Perkin Elmer, USA
Chemi doc	BIORAD, USA
Gelmate 2000 electrophoresis	Toyobo, Japan
PCR machine 2400 model	Perkin Elmer, USA
Spectrophotometer	BIORAD, SmartSpec <sup>™</sup> 3000, USA



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## APPENDIX II

## REAGENTS PREPARATION

## 1) 50x Tris-acetate buffer (TAE)

Tris base	242 g
Glacial acetic acid	57.1 ml
0.5 M EDTA, pH 8.0	100 ml

Adjust volume to 1,000 ml with distilled water and sterilize by autoclaving

## 2) 1.5% Agarose gel

Agarose powder	0.3 g
1x TAE buffer	20 ml
10 mg/ml Ethidium bromide	2.0 $\mu$ l

## 3) 0.85% NaCl

NaCl	0.85 g
Distilled water	100 ml

Dissolve 0.85 g of NaCl in 80 ml of distilled water. Adjust volume to 100 ml and sterilize by autoclaving

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## 4) 0.5 M EDTA

Add 186.1 g of disodium ethylene diamine tetraacetate.  $2\text{H}_2\text{O}$  to 800 ml of distilled water. Stir vigorously on a magnetic stirrer. Adjust the pH to 8.0 with NaOH (20 g of NaOH pellets). Dispense into aliquots and sterilize by autoclaving. The disodium salt of EDTA will not go into solution until the pH of the solution is adjusted to approximately 8.0 by the addition of NaOH

## 5) TE buffer (pH 8.0)

50 mM Tris-HCl, pH 8.0

10 mM EDTA, pH 8.0

Preparation (10 ml)

1 M Tris-HCl, pH 8.0	0.5 ml
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0.5 M EDTA, pH 8.0	0.2 ml
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Distilled water	9.3 ml
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## 6) Lysis buffer

0.85% NaCl

TE buffer

1% Triton X

## 7) 10 mg/ml Ethidium bromide

Ethidium bromide	1 g
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Distilled water	100 ml
-----------------	--------

Stir on a magnetic stirrer for several hours to ensure that the dye has dissolved.

Wrap the container in aluminium foil or transfer to a dark bottle and store at  $4^\circ\text{C}$

## 8) 3 M Sodium acetate, pH 8.0

Sodium acetate, 3 H<sub>2</sub>O                      408.1 g

Distilled water                                      800 ml

Adjust pH to 8.0 with glacial acetic acid

Adjust volume to 1,000 ml



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Miss Phattaraporn Orataiwun was born on February 22, 1977 in Nakornratchasima, Thailand. She graduated with a Bachelor degree of Science in Biology from Faculty of Science, Mahasarakham University in 1998.



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