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## APPENDIX

Appendix 1 : Turk Island Salt solution + modified BG<sub>11</sub> medium contained the following components:

**1. Preparation of Turk Island Salt solution**

Stock solution A :	KCl	33.3	g
	MgCl <sub>2</sub> .6H <sub>2</sub> O	275.0	g
	CaCl <sub>2</sub> .2H <sub>2</sub> O	73.3	g
and made up to 5 litres with distilled water			
Stock solution B :	MgSO <sub>4</sub> .7H <sub>2</sub> O	374.0	g
and made up to 5 litres with distilled water			

**2. Composition of modified BG<sub>11</sub> medium ( BG<sub>11</sub> medium + NaNO<sub>3</sub> solution)**

NaNO <sub>3</sub>	(75.0g/500ml)	50ml
KH <sub>2</sub> PO <sub>4</sub>	(8.0g/200ml)	5 ml
MgSO <sub>4</sub> .7H <sub>2</sub> O	(15.0g/200ml)	5 ml
CaCl <sub>2</sub> .2H <sub>2</sub> O	(7.2g/200ml)	5 ml
Na <sub>2</sub> CO <sub>3</sub>	(4.0g/200ml)	5 ml
citric acid	(1.2g/200ml)	5 ml
EDTA.Na <sub>2</sub>	(0.2g/200ml)	5 ml

FeSO<sub>4</sub>.7H<sub>2</sub>O (1.2g/200ml) 5 ml

\* Trace element A<sub>5</sub> solution + Co contained the following component in gram per litre ; H<sub>4</sub>BO<sub>4</sub> (2.86), ZnSO<sub>4</sub>.7H<sub>2</sub>O (0.22), CuSO<sub>4</sub>.5H<sub>2</sub>O (0.08), MnCl<sub>4</sub>.4H<sub>2</sub>O (1.81), Na<sub>2</sub>Mo<sub>4</sub>.2H<sub>2</sub>O (0.39) and Co(NO<sub>3</sub>)<sub>2</sub>.6H<sub>2</sub>O (0.049).

Culture medium of *A. halophytica* was prepared by adding all solution of item 2 at indicated volume to 500 ml of Stock solution A and 500 ml of Stock solution B. To this mixture 140.8 g NaCl was added and adjusted pH to 7.6 by slowly adding 2M NaOH then adjusted the final volume to 5 litres with distilled water. The medium was sterilized by autoclaving at 15 lb/in<sup>2</sup> for 15 minutes.

Appendix 2 : Preparation of polyacrylamide gel electrophoresis.

1. Stock reagent

30% Acrylamide, 0.8% bis stock solution

acrylamide	30.3	g
bis-acrylamide	0.8	g
adjust volume to 100 ml		

Solution B

For native-PAGE

Tris	18.2	g
adjust pH to 8.8 with 1M HCl and adjust volume to 100 ml with distilled water		

For SDS-PAGE

Tris	18.2	g
10% (w/v) SDS	4.0	ml
adjust pH to 8.8 with 1M HCl and adjust volume to 100 ml with distilled water		

Solution CFor native-PAGE

Tris 6.0 g

adjust pH to 6.8 with 1M HCl and adjust volume to 100 ml with distilled water

For SDS-PAGE

Tris 6.0 g

10%(w/v) 4.0 ml

adjust pH to 6.8 with 1M HCl and adjust volume to 100 ml with distilled water

Electrophoresis bufferFor native-PAGE

Tris 3.0 g

Glycine 14.4 g

adjust pH 8.8 with 1 M HCl and adjust volume to 1000 ml with distilled water

For SDS-PAGE

Tris	3.0	g
Glycine	14.4	g
SDS	1.0	g

adjust pH to 8.3 with 1M HCl and adjust volume to 1000 ml with distilled water

2. Non-denaturing PAGE7.5 % Separating gel

30% acrylamide solution	2.5	ml
Solution B	2.5	ml
distilled water	5.0	ml
TEMED	10.0	μl
10%(w/v) $(\text{NH}_4)_2\text{S}_2\text{O}_8$	50.0	μl

5% Stacking gel

30% acrylamide solution	1.3	ml
Solution B	2.0	ml
distilled water	4.6	ml
TEMED	10.0	μl
10%(w/v) $(\text{NH}_4)_2\text{S}_2\text{O}_8$	60.0	μl

### Sample buffer

1.25 M Tris-HCl pH 6.8 : glycerol : distilled water (1:2:2 v/v)  
were added with trace amount of bromo-phenol blue.

### SDS-PAGE

#### 10% Separating gel

30% acrylamide solution	3.3	ml
Solution B	2.5	ml
distilled water	4.2	ml
TEMED	5.0	μl
10%(w/v) $(\text{NH}_4)_2\text{S}_2\text{O}_8$	50.0	μl

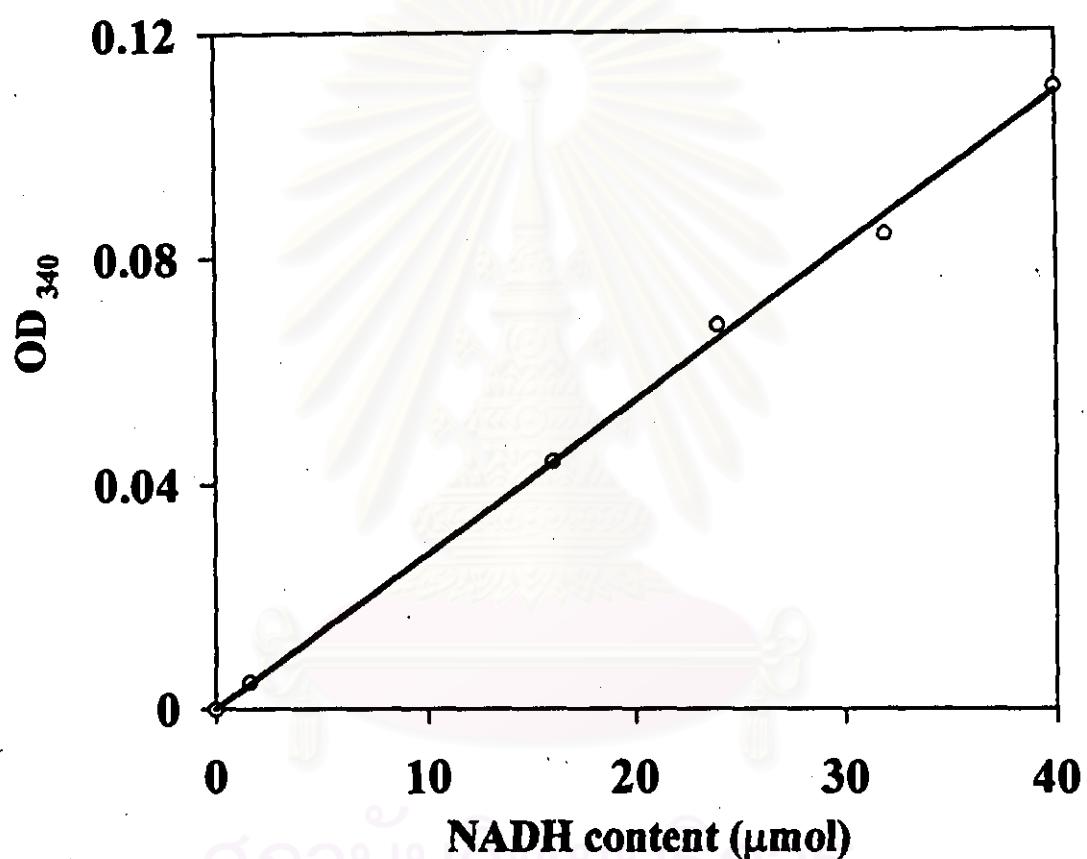
#### 5% Stacking gel

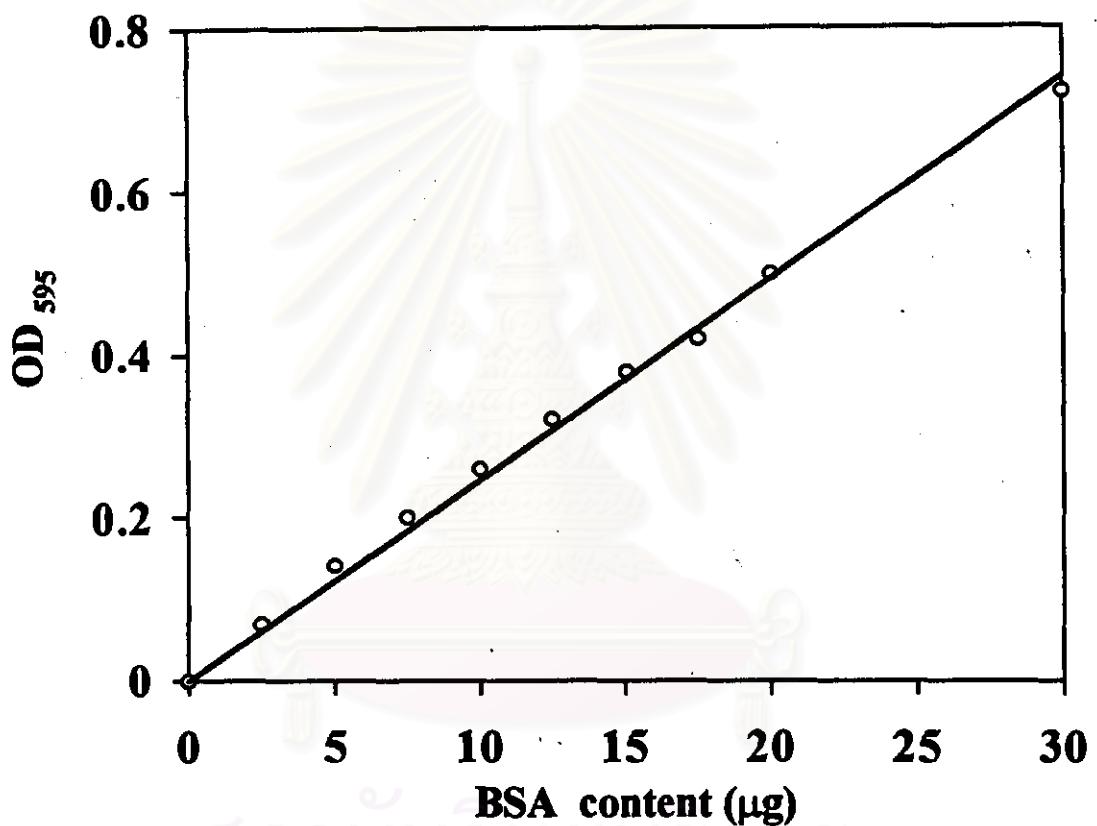
30% acrylamide solution	1.3	ml
Solution B	2.0	ml
distilled water	4.6	ml
TEMED	10.0	μl
10%(w/v) $(\text{NH}_4)_2\text{S}_2\text{O}_8$	60.0	μl

Sample buffer

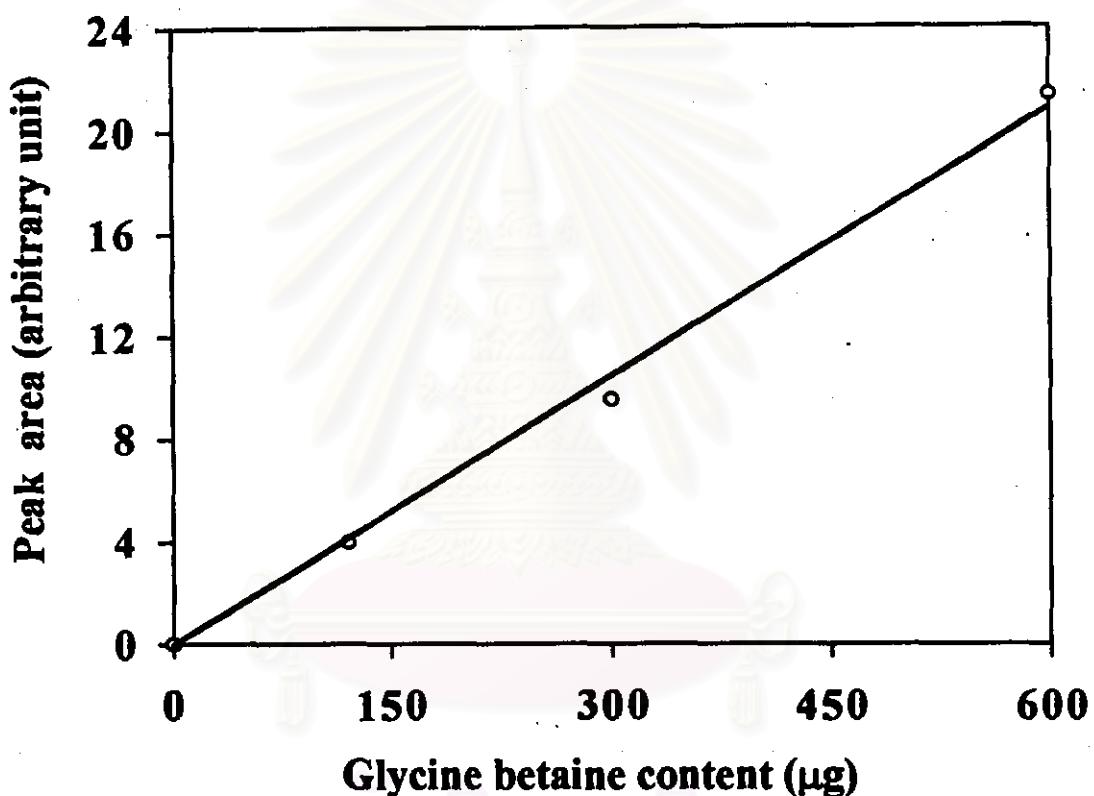
1 M Tris-HCl	0.6	ml
50%(v/v) glycerol	5.0	ml
10%(w/v) SDS	2.0	ml
2- mercaptoethanol	0.5	ml
1%(w/v) bromophenol blue	1.0	ml
distilled water	0.9	ml

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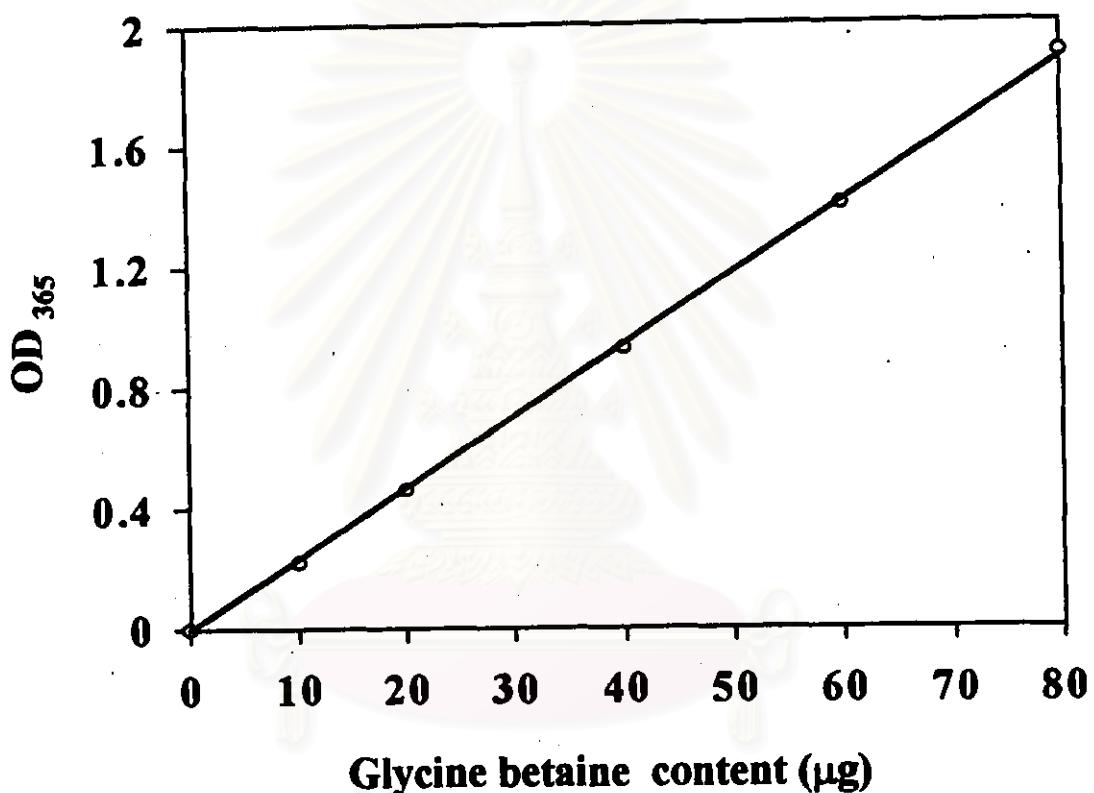
Appendix 3 : Standard curve of NADH

Appendix 4 : Standard curve of BSA

Appendix 5 : Standard curve of glycine betaine determined by  
<sup>1</sup>H-NMR



Appendix 6 : Standard curve of glycine betaine determined by  
tri-iodide assay



## **BIOGRAPHY**

Miss Uthaiwon Kumarb was born on November 7 ,1972 in Petchburi, Thailand. She graduated with a Bachelor Degree of Science in Biology from Faculty of Science, Silapakorn University, Nakhonpathom, Thailand in 1993 and studied for a Master Degree in Biochemistry program since 1994.



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