

APPENDIX

Appendix A

Reagents and dye

Gram staining

Acetone alcohol

Ethyl alcohol (ethanol, 95 %) 700 ml

Acetone 500 ml

Crystal violet dye

Crystal violet 10 g

Distilled water 1000 ml

Gram's solution iodine

Iodine 1 g

Potassium iodide 2 g

Distilled water 300 ml

Safranin (staining solution)

Safranin (90 % dye content) 0.25 g

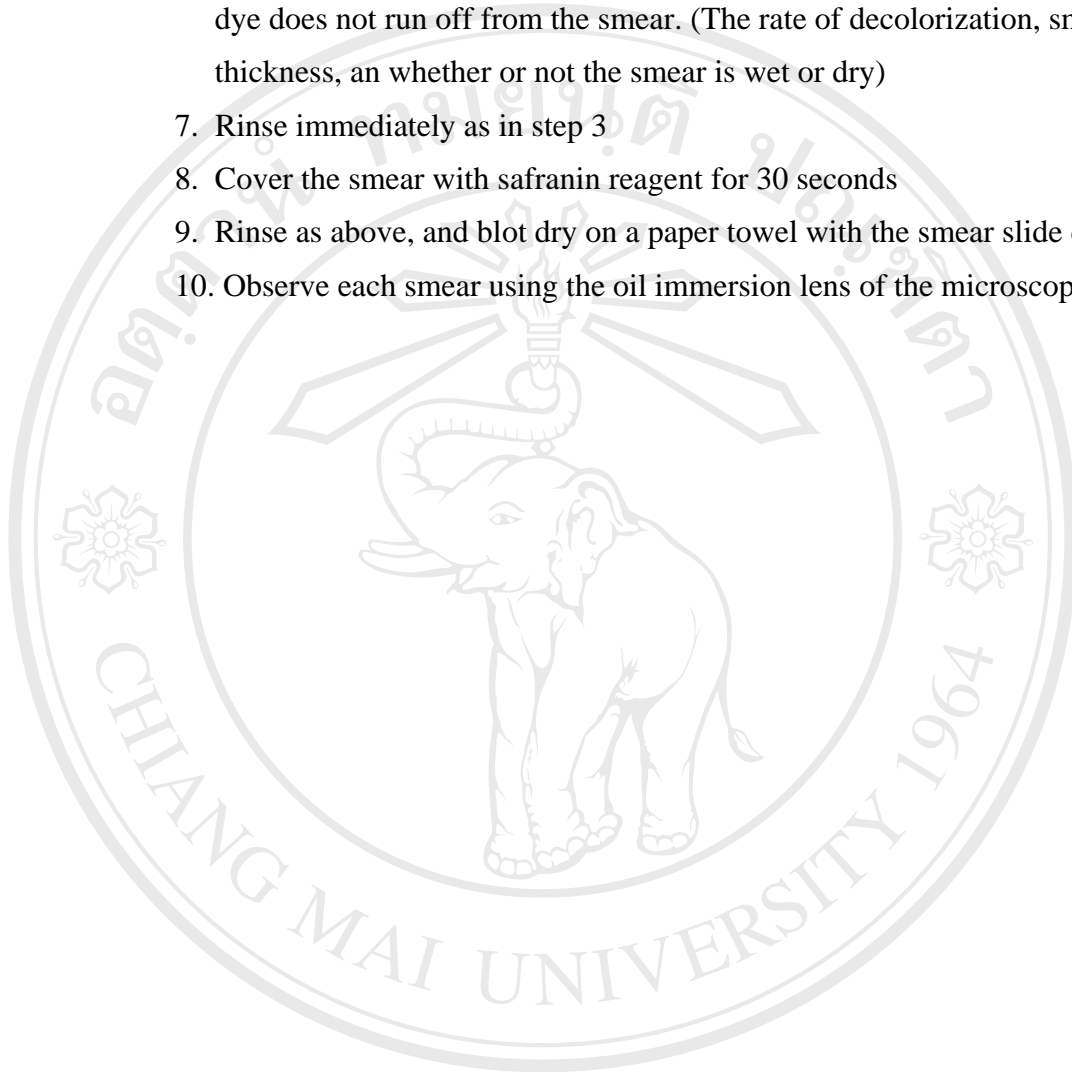
Alcohol (95 %) 10 ml

Distilled water 100 ml

Procedure

1. Prepare separate, thin smears of bacteria. Air dry and heat fix them
2. Cover the smears with crystal violet for 1 min
3. Rinse slides in slowly running tap water for five seconds
4. Next, rinse smears with the iodine reagent and then apply the same reagent for 1 min

5. Rinse as in step 3
6. Apply the alcohol reagent slowly. Continue to add the decolorizer until dye does not run off from the smear. (The rate of decolorization, smear thickness, and whether or not the smear is wet or dry)
7. Rinse immediately as in step 3
8. Cover the smear with safranin reagent for 30 seconds
9. Rinse as above, and blot dry on a paper towel with the smear side down.
10. Observe each smear using the oil immersion lens of the microscope.



ลิขสิทธิ์มหาวิทยาลัยเชียงใหม่
Copyright © by Chiang Mai University
All rights reserved

Appendix B

Reagents and method for determination of enzyme activity and protein properties

Protein determination by Lowry method (1951)

Reagents

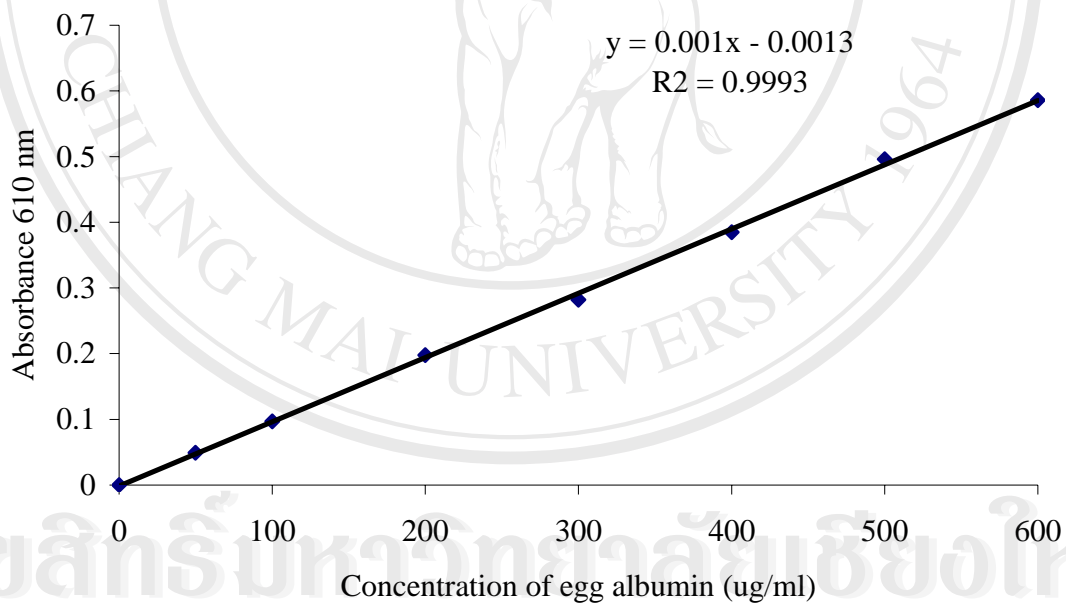
1. Distilled water
2. Solution A : 20 g Na_2CO_3 in 1 L of 0.1 M NaOH
3. Solution B : 0.5 g $\text{CuSO}_4 \cdot \text{H}_2\text{O}$, 1 g tartaric acid in 100 ml of H_2O
4. Solution C : Solution A : Solution B = 50 :1 (Freshly prepared)
5. Solution D : Diluted the commercial: Phenol reagent (Folin-Ciocalteu reagent) 1: 2

Procedure

1. Add 1 ml of solution C (to 0.1 ml of protein solution)
2. Incubate 10 min at 30 °C
3. Add 0.1 ml of Solution D
4. Incubate for 20 min at 30 °C
5. Measure the sample at 610 nm
6. To prepare calibration curve, add a different concentration of egg albumin Solution to test tub and perform the protein assay as described above (1-5) Plot the absorbance of each standard protein concentration as a function of the protein concentration
7. The amount of protein in sample is calculated by the correlation between absorbance and standard protein concentration

Table B1 Absorbance at 610 nm of various concentration of BSA by Lowry method

Egg albumin ($\mu\text{g/ml}$)	A_{610}
0	0.052
50	0.097
100	0.198
200	0.282
400	0.385
500	0.496
600	0.586

**Figure B1** Calibration curve of standard D=BSA concentration versus absorbance at 610 nm

Protein determination by spectrophotometer at 280 nm

Procedure

1. Set absorbance at 280 nm to zero with distilled water in quartz cuvette.
2. Remove the distilled water and add sample solution to the cuvette then measure the absorbance of the protein solution.
3. To calculate the protein concentration using the following formula

$$\text{Concentration (mg/ml)} = \text{absorbance of protein solution at 280 nm}$$

Protease assay and calculation

Reagents

1. 1 % Casein hamersten
0.5 g of casein hamersten in 50 ml of 0.1 M potassium phosphate buffer (KPB)
2. 1 M potassium phosphate buffer (pH 6.8)
Solution A 1 M KH_2PO_4 136.09 g/l
Solution B 1 M 174.18 g/l
3. 12 % Trichloroacetic acid (TCA)
12 g TCA in 100 ml of distilled water and then diluted to 7 % TCA
4. Solution A: 20 g Na_2CO_3 in 1 l of 0.1 M NaOH
Solution B: 0.5 g $\text{CuSO}_4 \cdot \text{H}_2\text{O}$ in 100 ml of distilled water
Solution C: 1 g of tartaric acid in 100 ml of distilled water

Mix solution A: B: C, 50 : 1 : 1 (Freshly prepared)

Folin-Ciocalteu reagent 1 : 2

Protease assay

1. Add 0.25 ml of diluted enzyme to 0.25 ml of 1 % casein hamersten
2. Incubate at 40 °C for 10 min
3. Incubate at ambient temperature for 30 min
4. An then centrifuge at 8,000 rpm for 10 min
5. Added mix solution in No. 4 0.625 ml to 0.25 ml of supernatant
6. Incubate at ambient temperature for 10 min
7. Add 0.125 ml of Folin-Ciocalteu reagent
8. Incubate at 40 °C for 30 min at 40 °C
9. Measure protease activity at 660 nm

Enzyme activity can be calculated from net absrobance by calibration to the standard curve of tyrosine

One unit of protease activity was defined as the amount of enzyme catalyzes the formation of 1 μ tyrosine per min under the given condition

$$\text{Protease activity (U/ml)} = \frac{(E-E_0 \times b \times c)}{ES \times a \times T}$$

E = Absorbance of the reaction mixture of enzyme and substrate

E₀ = Absorbance of control as the samples that the substrate incubated with the denature enzyme by addition of TCA

a = Volume of the enzyme (0.25 ml)

b = The volume of enzyme and sbustrate (0.5 ml)

c = Dilution factor

ES = Slope value from standard calibration curve (tyrosine) (3.2781)

T = Time

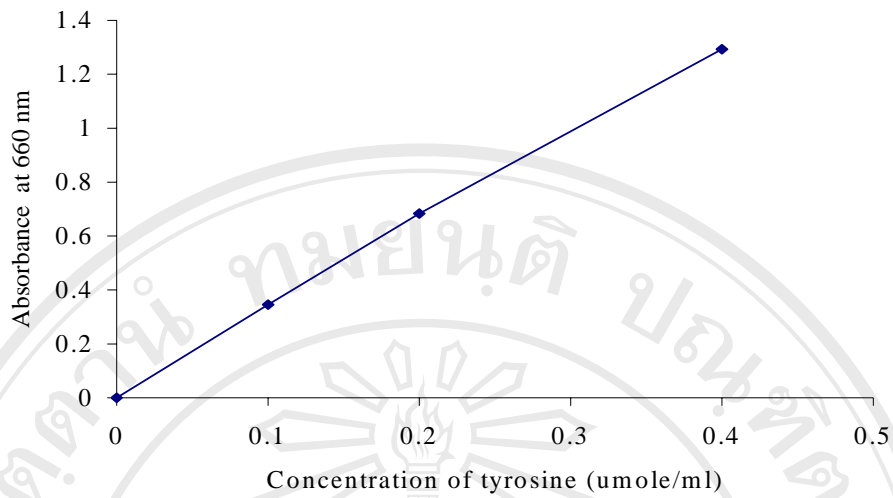


Figure B2 Standard tyrosine

SDS-PAGE reagents

Acrylamide stock solution

Tris-HCl (pH 8.8)

Tris-HCl 9 (pH 6.8)

10% sodium dodecyl sulfate

TEMED (N'N'N',N' - tetramethylenediamine)

10 % ammonium persulfate (APS)

distilled water

Electrophoresis buffer

Tris 1.8 g

Glycine 8.4 g

SDS 0.3 g

Distilled water 300 ml

ลิขสิทธิ์มหาวิทยาลัยเชียงใหม่

Copyright © by Chiang Mai University

All rights reserved

Coomassie gel staining

0.25 % Coomassie brilliant blue (w/v) in 50 % methanol, 10 % acetic acid

Coomassie gel destaining

25 % methanol, 7 % acetic acid

Procedure**Preparation of gel**

1. Clean the glass, spacer, combs and upper buffer reservoir of the gel apparatus with detergent and completely dry.
2. Assemble the glass plate sandwich and the spacer
3. Prepare the monomer solution for separate gel by combining all of the reagents in Table B2.
4. Gently mix the 33 μ l of APS and into the solution in step 3.
5. Carefully introduce the solution into glass plate sandwich using an autopipette to about 0.5 cm below the short plate.
6. Gently layer about 1-5 mm of distilled water on top of the gel solution and allow gel to polymerize for 30-60 min.
7. Prepare concentrate gel monomer solution by combining all of the reagents in Table B3.
8. Deaerated the monomer solution for at least 15 min.
9. Pour off the water covering the concentrate gel and dry the area above it with filter paper.
10. Insert comb into glass plate sandwich and tilt the comb at a slight angle to provide a way for bubbles to escape.
11. Add 10 % APS and 10 μ l of TEMED to the deaerated monomer solution and carefully pour the concentrate gel solution on the top of separate gel.
12. Align the comb in its proper position and allow the gel to polymerize for 30-45 min.

Table B2 Formulation of separate gel

Component	Volume
Distilled water	2.24 ml
Tris-HCl, pH 8.8	1.25 ml
10 % SDS	50 μ l
Acrylamide/bis	1.41 ml

Table B3 Formulation of concentrate gel

Component	Volume
Distilled water	1.03 ml
Tris-HCl, pH 6.8	0.415 ml
10 % SDS	16.5 μ l
Acrylamide/bis	0.185 ml

Sample preparation

1. Combine protein sample and treatment buffer in an Eppendroff tube.
2. Boil the sample for 3 min and then centrifuge for 1 min.

Electrophoresis

1. Place the gel into electrophoresis chamber and attach both gels to electrode assembly.
2. Add electrophoresis buffer to inner and outer reservoir and remove the comb from the concentrate gel carefully.
3. Load the prepared sample into the well in the stacking by layering them under electrode buffer.
4. Attach the electrode plugs to power supply.
5. Turn on power supply to 100 V. until the Bromophenol blue tracking dye front reaction the bottom of the gel.

Dye staining with coomassie brilliant blue

1. Pick up the gel and transfer to a small container containing Coomassie gel stain.
2. Agitate for 30 min on a slow shaker
3. Pour out stain.
4. Add Coomassie destain and continue slow shaking. Change the destain solution several times, until the background has been satisfactorily removed.

Native Molecular weight determination

Molecular weight of Native enzyme was determined by gel filtration chromatography on a Superdex 200 PC High resolution column and 10 mM KPB containing 100 mM NaCl as running buffer. Protein standards consisted of cytochrome C (12,400 Da), carbonic anhydrase (29,000 Da), β -amylase (66,000 Da), alcohol dehydrogenase (150,000 Da) and albumin (200,000 Da).

Molecular weight determination

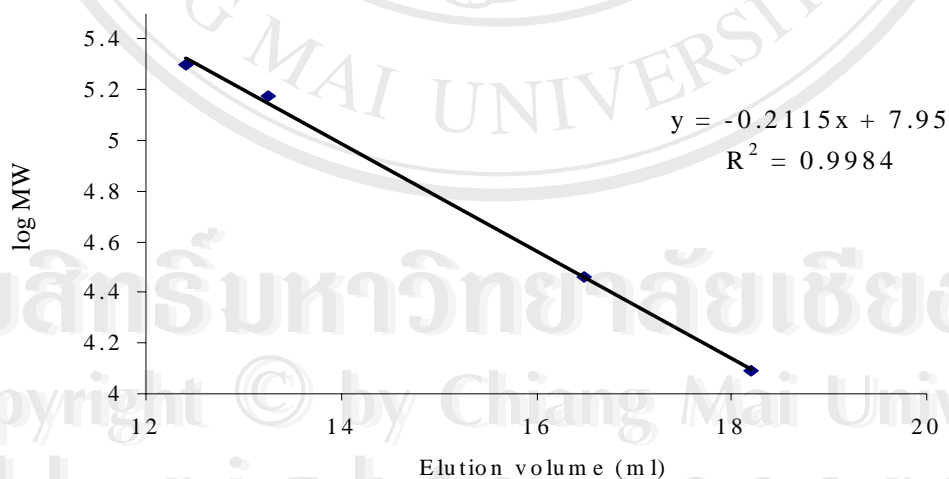


Figure B3 Standard curve of log molecular weight and elution volume

SDS-PAGE is used to determine the molecular weight of protein by comparison of protein mobility with those of several marker proteins of known molecular weight.

Procedure

1. After gel electrophoresis and staining. Measure the distance of protein migration.
2. Plot the logarithms of the molecular weight of the protein standard
3. Read molecular weight of the unknown protein from the graph

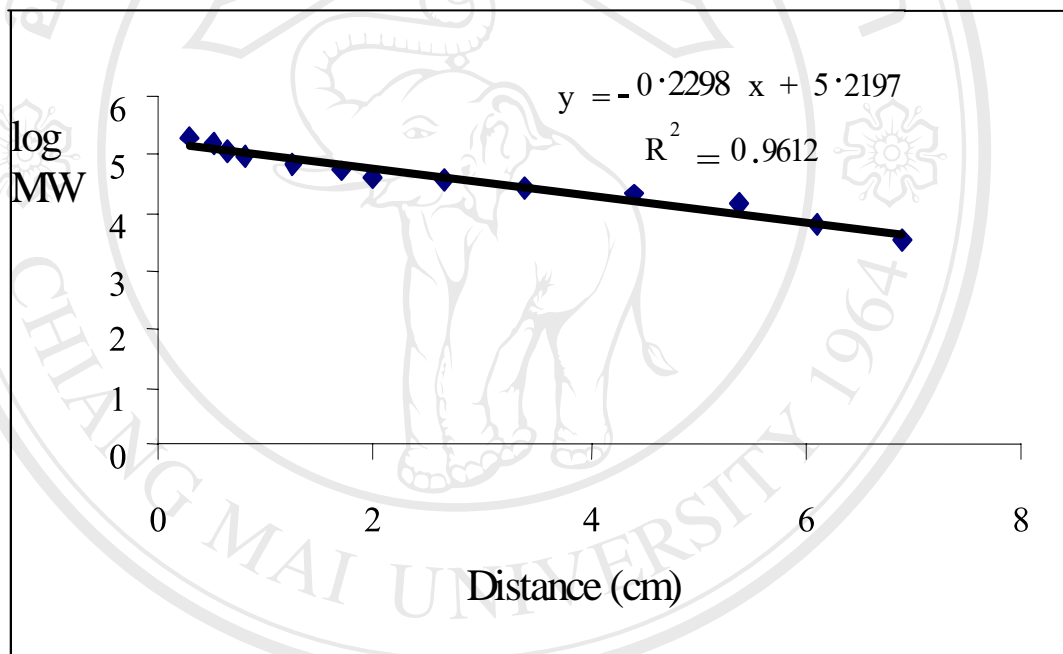


Figure B4 Semilogarithmic graph of molecular weight

0.5 M MES

Dissolve 1.066 g in distilled water and adjust to pH 5.5-7.0 with HCl and add water to a final volume of 10 ml.

1 M MOPS

Dissolve 2.0926 g MOPS base in distilled water and adjust to pH 7.5-8.5 with HCl and add water to final volume of 10 ml.

1 M HEPES

Dissolve 2.3831 g HEPES base in distilled water and adjust to pH 7.5-9.0 with NaOH and add water to final volume of 10 ml.

1 M KPB

Dissolve of solution A: 1 M KH_2PO_4 1.361 g on distilled water 10 ml HEPES base in distilled water and solution B: 1 M K_2HPO_4 and mixed solution A : solution B (39.0 ml : 61.0 ml for pH 7.0 and mixed solution A : solution B (5.3 ml : 94.7 ml for pH 8.0

1 M Tris-HCl

Dissolve 1.2114 g Tris base in distilled water and adjust to pH 9.0-11.0 with NaOH and add water to final volume of 10 ml.

0.5 M CAP

Dissolve 1.1066 g base in distilled water and adjust to pH 9.0-11.0 with NaOH and add water to final volume of 10 ml.

0.5 M CHES

Dissolve 1.0365 g base in distilled water and adjust to pH 9.0-11.0 with NaOH and add water to final volume of 10 ml.

1M EDTA

Dissolve 0.0744 g Ethylene diamine tetraacetic acid (EDTA) base in distilled water and add water to final volume of 200 μl and neutralize by 1M Na_2HCO_3

1M PMSF

Dissolve 0.0350 g Phenylmethylsulphonyl fluoride (PMSF) base in absolute ethanol and add ethanol to final volume of 200 μl .

1M DTT

Dissolve 0.0310 g Dithioerythreitol base in distilled water and add water to final volume of 200 μl .

1M PCMB

Dissolve 0.0714 g p-Chloromercuri benzoic acid (PCMB) base in 1 M NaOH and add water to final volume of 200 μl and then centrifuge before use.

1M TPCK

Dissolve 0.0704 g Tosyl-L-phenylalanine chloromethyl ketone base in distilled water and add water to final volume of 200 μ l.

1M Mercaptoethanol

Dissolve 0.0156 g Ethylene mercaptoethanol base in distilled water and add water to final volume of 200 μ l.

100 mM o-Phenanthroline

Dissolve 0.0198 g o-Phenanthroline base in distilled water and add water to final volume of 100 μ l.

100 mM α',α' -Dipyridyl

Dissolve 0.0156 g α',α' -Dipyridyl base in distilled water and add water to final volume of 100 μ l.

100 mM NaCl

Dissolve 0.0292 g NaCl base in distilled water and add water to final volume of 500 μ l .

100 mM KCl

Dissolve 0.0373 g KCl base in distilled water and add water to final volume of 500 μ l .

100 mM $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$

Dissolve 0.0735 g CaCl_2 base in distilled water and add water to final volume of 500 μ l .

100 mM $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$

Dissolve 0.0989 g $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$ base in distilled water and add water to final volume of 500 μ l.

100 mM $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$

Dissolve 0.1017 g $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$ base in distilled water and add water to final volume of 500 μ l.

100 mM ZnCl₂

Dissolve 0.0682 g ZnCl₂ base in distilled water and add water to final volume of 500 µl.

100 mM CuCl₂·2H₂O

Dissolve 0.0852 g CuCl₂ base in distilled water and add water to final volume of 500 µl.

100 mM CoCl₂·6H₂O

Dissolve 0.1190 g CoCl₂·6H₂O base in distilled water and add water to final volume of 500 µl.

100 mM FeSO₄·7H₂O

Dissolve 0.1390 g FeSO₄·7H₂O base in distilled water and add water to final volume of 500 µl.

100 mM NiCl₂·6H₂O

Dissolve 0.1188 g NiCl₂·6H₂O base in distilled water and add water to final volume of 500 µl.

2.5 mM Leu-pNA

0.0036 g dissolve in distilled water and add water to final volume of 5 ml

2.5 mM Lys-pNA 0.0054 g

dissolve in distilled water and add water to final volume of 5 ml

2.5 mM Arg-pNA

0.0045 g dissolve in distilled water and add water to final volume of 5 ml

2.5 mM Gly-pNA

0.0024 g dissolve in distilled water and add water to final volume of 5 ml

2.5 mM Met-pNA

0.0034 g dissolve in distilled water and add water to final volume of 5 ml

2.5 mM Phe-pNA

0.0036 g dissolve in 5 ml of DMSO and add water to final volume of 5 ml

2.5 mM Pro-pNA

0.0044 g dissolve in distilled water and add water to final volume of 5 ml

2.5 mM Gly-Phe-pNA

0.0043 g dissolve in distilled water and add water to final volume of 5 ml

2.5 mM Gly-Pro-pNA

0.0041 g dissolve in distilled water and add water to final volume of 5 ml

2.5 mM Val-Ala-pNA

0.0039 g dissolve in distilled water and add water to final volume of 5 ml

2.5 mM H-Gly-Arg-pNA

0.0044 g dissolve in distilled water and add water to final volume of 5 ml

2.5 mM CBZ-GGL-pNA

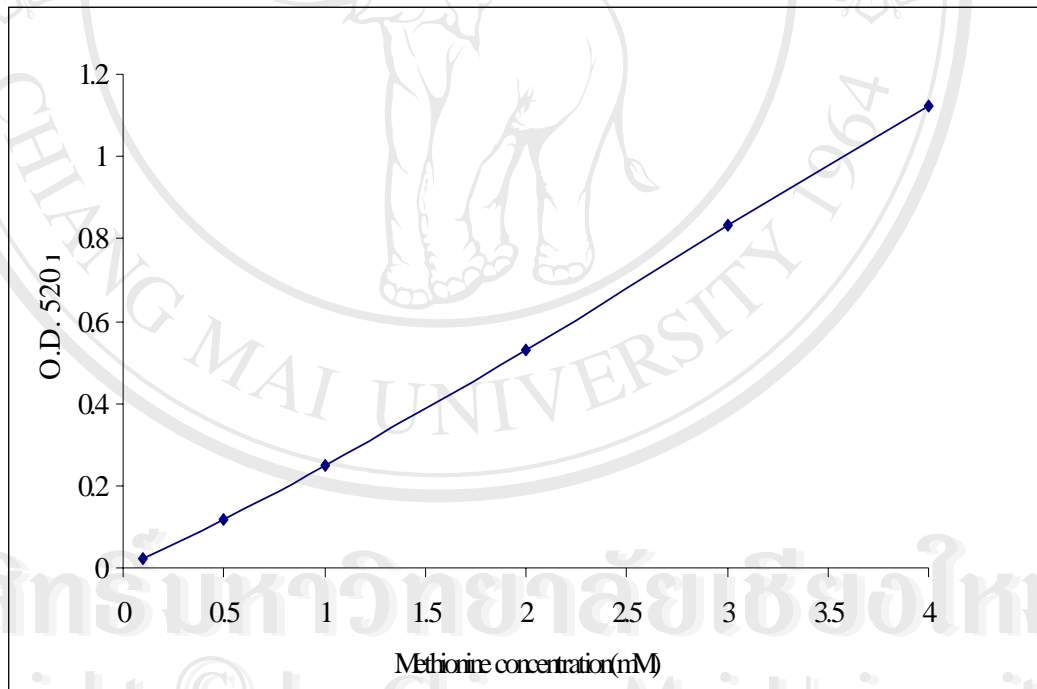
0.0062 g dissolve in distilled water and add water to final volume of 5 ml

Standard methionine (TNBS method)

Prepare D-methionine 0.1, 0.5, 1, 2, 3 and 4 mM. Each concentration were take 200 μ l and add 100 μ l of 0.25 M NaOH. Incubate at 30 °C for 10 min and after that add 0.125 M $\text{Na}_2\text{B}_4\text{O}_7$ 100 μ l. And add 0.2 M TNBS 40 μ l incubate at 30 °C for 10 min. After that added $\text{Na}_2\text{SO}_3 \cdot \text{NaH}_2\text{PO}_4$ solution for 800 μ l and then measure at absorbance 520 nm.

Table B4 Standard methionine

Methionine mM	A ₅₂₀
0.1	0.022
0.5	0.120
1	0.247
2	0.532
3	0.834
4	1.123

**Figure B5** Standard methionine

Appendix C

The characteristic of isolated bacteria

Table C1 Gram's staining of bacteria isolated from various sources

Sources	Code	Gram's stain
Nam-pla1 (Chonburi) (1 month)	PC253 PC185 PC255	+ve, rods +ve, cocci +ve, rods
Nam-pla1 (Chonburi) (2 months)	PC185 PC255	+ve, cocci +ve, rods
Nam-pla1 (Chonburi) (3 months)	PC188 PC189	+ve, cocci +ve, rods
Nam-pla1 (Chonburi) (4 months)	PC214 PC258	+ve, rods +ve, cocci
Nam-pla1 (Chonburi) (5 months)	PC191 PC216	+ve, rods +ve, rods
Nam-pla1 (Chonburi) (6 months)	PC191 PC216	+ve, rods +ve, rods
Nam-pla 2 (Chonburi) (12 months)	TP165 TP259 TP262	+ve, cocci +ve, rods +ve, rods

Table C1 (continued)

Source	Code	Gram 's stain
Nam-pla 3 (Sukhothai)	NN156	-ve,rods
	NN157	-ve,rods
	NN240	+ve,rods
	FT176	+ve,rods
Nam-pla 4 (Ratchaburi)	FR26	+ve,cocci
Nam-pla 5 (Rayong)	TR89	+ve,cocci
	TR90	+ve,cocci
	TR87	+ve,cocci
Nam-pla 6 (Samutprakarn)	SS61	+ve,rods
	SS63	+ve,cocci
Nam-pla 7 (Samutprakarn)	K1	+ve,rods
	K2	+ve,rods
	K3	+ve,rods
	K4	+ve,rods
	K5	+ve,rods
	K6	-ve,rods
	K7	+ve,rods
	K8	+ve,rods
	K9	+ve,cocci
	K10	+ve,cocci
	K11	+ve,rods
	K12	+ve,rods
	K13	-ve,rods
	K14	+ve,rods
	K15	+ve,cocci

Table C1 (continued)

Source	Code	Gram's staining
	K16	+ve,cocci
	K17	+ve,rods
	K18	+ve,rods
	K19	+ve,rods
	K20	+ve,rods
	K21	+ve,rods
	K22	+ve,rods
Ka-pi 1 (Maejo market Chiang Mai)	KJ16	+ve,rods
	KJ81	-ve,rods
	KJ119	+ve,rods
Ka-pi 2 (Sukhothai)	KM1	-ve,rods
	KM2	+ve,rods
	KM151	-ve,rods
	KM232	+ve,rods
Ka-pi 3 (Kardluang market Chiang-Mai)	KK11	-ve,rods
	KK12	+ve,rods
	KK19	+ve,rods
	KK22	+ve,rods
	KK51	+ve,cocci
	KK92	+ve,cocci
	KK112	+ve,rods
	KK119	+ve,rods
	KK126	-ve,rods
	KK127	+ve,rods
	KK128	+ve,rods
KK137	+ve,rods	
KK138	+ve,rods	

Table C1 (continued)

Source	Code	Gram's staining	
Ka-pi 4 (Sukhothai)	K23	+ve,rods	
	K24	+ve,rods	
	K25	+ve,rods	
	K26	+ve,rods	
	K27	-ve,rods	
	K28	+ve,rods	
	K29	+ve,rods	
	K30	+ve,rods	
	K31	-ve,rods	
	K32	+ve,rods	
	K33	+ve,rods	
	K34	+ve,rods	
	K35	+ve,rods	
	Ka-pi 5 (Sukhothai)	K36	+ve,rods
		K37	+ve,rods
K38		+ve,rods	
K39		-ve,rods	
K40		+ve,rods	
K41		+ve,cocci	
K42		+ve,cocci	
K51		+ve,rods	
K52		+ve,rods	
K53	+ve,rods		

Table C1 (continued)

Source	Code	Gram's staining
Pla-ra 1 (Maejo market Chiang Mai)	RJ26	+ve,rods
	RJ29	+ve,rods
	RJ31	+ve,cocci
	RJ58	+ve,rods
	RJ79	-ve,rods
	RJ85	+ve,rods
	RJ89	+ve,rods
	RJ90	+ve,rods
	RJ93	+ve,rods
	RJ102	+ve,rods
	RJ103	+ve,rods
	RJ109	+ve,rods
	RJ110	-ve,rods
RJ129	+ve,rods	
Pla-ra 2 (Sukhothai)	RC179	+ve,rods
	RC192	+ve,rods
	RC194	+ve,rods
	RC195	+ve,rods
	RC210	+ve,rods
	RM143	-ve,rods
	RM146	-ve,rods
	RM235	+ve,rods
RM249	+ve,rods	

Table C1 (continued)

Sources	Code	Gram's stain
Pla-ra 3 Kard-luang market (Chiang Mai)	KK22	+ve, cocci
	KK23	+ve, rods
	KK38	-ve, rods
	KK61	-ve, rods
Pla-ra 4 (Chiang Mai)	K54	+ve, rods
	K55	+ve, rods
	K56	+ve, rods
	K57	+ve, cocci
	K58	+ve, rods
	K59	-ve, rods
	K60	-ve, rods
	K61	+ve, rods
Pla-chom1 (Sukhothai)	PJ52	+ve, rods
	PJ139	+ve, rods
Pla-chom 2 (Khonkan)	PJ78	+ve, rods
Naw-mai-dong (Maejo market Chiang Mai)	NJ7	+ve, rods
	NJ33	+ve, rods
	NJ73	-ve, rods
	NM154	+ve, rods
	NM181	+ve, rods

Table C1 (continued)

Source	Code	Gram's staining
Tai-pla1 (Trang)	TP6	+ve,rods
	TP57	-ve,rods
Kaey (Trang)	KT7	-ve,rods
	KT9	-ve,rods
Pu-chem Maejo market Chiang Mai)	PD62	+ve,rods
	PD67	+ve,rods
Pung-pla (Kardluang Chiang Mai)	PP59	-ve,rods
	PP134	+ve,rods
	PP147	+ve,rods
Gra-tiem-dong (Sukhothai)	KT56	+ve,rods
Tao-chieo 1 (Kardluang Chiang Mai)	TK21	+ve,rods
	TK53	+ve,rods
	TK54	-ve,rods
	TK145	+ve,rods
	TK160	+ve,rods
	TK161	+ve,rods
	TK167	+ve,rods
	TK170	+ve,rods
Tao-chieo 2 (Sukhothai)	TN103	+ve,rods
Tao-chieo 3 (Sukhothai)	TS101	+ve,rods

Table C1 (continued)

Source	Code	Gram's staining
Sea water (Rayong)	SW1	-ve,rods
Salt (Sukhothai)	SM25	+ve,cocci
Pla-too-chem (Samutsakorn)	PT12	+ve,rods
Sea shell solid-waste (Rayong)	SW13	+ve,rods
Sea sand (Rayong)	SN75	+ve,cocci
Phak-kard-dong (Sukhothai)	PD69	-ve,rods
Hua-phak-Kkrd-chem (Sukhothai)	HK5	+ve,rods
	HK20	+ve,rods
Soil 1 (Samutsongkram)	SS36	+ve,cocci
	SS48	+ve,rods
	SS59	+ve,rods
Soil 2 (Samutprakarn)	SS50	+ve,rods
	SS77	+ve,rods

Table C2 The ability of growth on Nutrient agar (NA) at various concentrations of sodium chloride

Sources	Code	Sodium chloride concentration (%)					
		0	5	10	15	20	25
Nam-pla 1	PC253	+	+	+	+	-	-
	PC185	+	+	+	+	-	-
	PC255	+	+	+	+	-	-
	PC188	+	+	+	+	+	-
	PC189	+	+	+	+	-	-
	PC208	+	+	+	+	-	-
	PC212	+	+	+	+	-	-
	PC213	+	+	+	+	-	-
	PC214	+	+	+	+	-	-
	PC258	+	+	+	+	-	-
	PC191	+	+	+	+	-	-
	PC216	+	+	+	+	-	-
	PC164	+	+	+	+	-	-
PC219	+	+	+	+	-	-	
Nam-pla 2	TP165	+	+	+	+	+	+
	TP259	+	+	+	+	+	-
	TP262	+	+	+	+	-	-
Nam-pla 3	NN156	+	+	+	+	-	-
	NN157	+	+	+	+	-	-
	NN240	+	+	+	+	-	-
	FT176	+	+	+	+	-	-
Nam-pla 4	FR26	+	+	+	+	+	+

Table C2 (continued)

Source	Code	Sodium chloride concentration (%)					
		0	5	10	15	20	25
Nam-pla 5	TR89	+	+	+	+	+	+
	TR90	+	+	+	+	+	+
	TR87	+	+	+	+	+	+
Nam-pla 6	SS61	+	+	+	+	+	+
	SS63	+	+	+	+	+	+
Nam-pla 7	K1	+	+	+	+	-	-
	K2	+	+	+	+	-	-
	K3	+	+	+	+	-	-
	K4	+	+	+	+	-	-
	K5	+	+	+	+	-	-
	K6	+	+	+	+	-	-
	K7	+	+	+	+	-	-
	K8	+	+	+	+	-	-
	K9	+	+	+	+	-	-
	K10	+	+	+	+	-	-
	K11	+	+	+	+	-	-
	K12	+	+	+	+	-	-
	K13	+	+	+	+	-	-
	K14	+	+	+	+	-	-
	K15	+	+	+	+	-	-
	K16	+	+	+	+	-	-

Table C2 (continued)

Source	Code	Sodium chloride concentration (%)					
		0	5	10	15	20	25
Nam-pla 7	K17	+	+	+	+	-	-
	K18	+	+	+	+	-	-
	K19	+	+	+	+	-	-
	K20	+	+	+	+	-	-
	K21	+	+	+	+	-	-
	K22	+	+	+	+	-	-
Ka-pi 1	KJ16	+	+	+	+	-	-
	KJ81	+	+	+	+	-	-
	KJ119	+	+	+	+	-	-
Ka-pi 2	KM1	+	+	+	+	-	-
	KM2	+	+	+	+	-	-
	KM151	+	+	+	+	-	-
	KM232	+	+	+	+	-	-
Ka-pi 3	KK11	+	+	+	+	-	-
	KK12	+	+	+	+	-	-
	KK19	+	+	+	+	-	-
	KK22	+	+	+	+	-	-
	KK51	+	+	+	+	-	-
	KK92	+	+	+	+	+	+
	KK112	+	+	+	+	-	-
	KK119	+	+	+	+	-	-
	KK126	+	+	+	+	-	-
	KK127	+	+	+	+	-	-

Table C2 (continued)

Source	Code	Sodium chloride concentration (%)					
		0	5	10	15	20	25
Ka-pi 3	KK128	+	+	+	+	-	-
	KK137	+	+	+	+	-	-
	KK138	+	+	+	+	-	-
Ka-pi 4	K23	+	+	+	+	-	-
	K24	+	+	+	+	-	-
	K25	+	+	+	+	-	-
	K26	+	+	+	+	-	-
	K27	+	+	+	+	-	-
	K28	+	+	+	+	-	-
	K29	+	+	+	+	-	-
	K30	+	+	+	+	-	-
Ka-pi 4	K31	+	+	+	+	-	-
	K32	+	+	+	+	-	-
	K33	+	+	+	+	-	-
	K34	+	+	+	+	-	-
	K35	+	+	+	+	-	-
Ka-pi 5	K36	+	+	+	+	-	-
	K37	+	+	+	+	-	-
	K38	+	+	+	+	-	-
	K39	+	+	+	+	-	-
	K40	+	+	+	+	-	-

Table C2 (continued)

Source	Code	Sodium chloride concentration (%)					
		0	5	10	15	20	25
Ka-pi 5	K41	+	+	+	+	-	-
	K42	+	+	+	+	-	-
	K43	+	+	+	+	-	-
	K44	+	+	+	+	-	-
	K45	+	+	+	+	-	-
	K46	+	+	+	+	-	-
	K47	+	+	+	+	-	-
	K48	+	+	+	+	-	-
	K49	+	+	+	+	-	-
	K50	+	+	+	+	-	-
	K51	+	+	+	+	-	-
	K52	+	+	+	+	-	-
	K53	+	+	+	+	-	-
Pla-ra 1	RJ26	+	+	+	+	-	-
	RJ29	+	+	+	+	-	-
	RJ31	+	+	+	+	-	-
	RJ58	+	+	+	+	-	-
	RJ79	+	+	+	+	-	-
	RJ85	+	+	+	+	-	-
	RJ89	+	+	+	+	-	-
	RJ90	+	+	+	+	-	-
	RJ93	+	+	+	+	-	-
	RJ102	+	+	+	+	+	+
	RJ103	+	+	+	+	-	-

Table C2 (continued)

Source	Code	Sodium chloride concentration (%)					
		0	5	10	15	20	25
	RJ109	+	+	+	+	+	+
	RJ110	+	+	+	+	-	-
	RJ129	+	+	+	+	-	-
Pla-ra 2	RC179	+	+	+	+	-	-
	RC192	+	+	+	+	-	-
	RC194	+	+	+	+	+	+
	RC195	+	+	+	+	-	-
	RC210	+	+	+	+	-	-
	RM143	+	+	+	+	-	-
	RM146	+	+	+	+	+	+
	RM235	+	+	+	+	+	+
	RM249	+	+	+	+	-	-
Pla-ra 3	KK22	+	+	+	+	-	-
	KK23	+	+	+	+	-	-
	KK38	+	+	+	+	-	-
	KK61	+	+	+	+	-	-
Pla-ra 4	K54	+	+	+	+	-	-
	K55	+	+	+	+	-	-
	K56	+	+	+	+	-	-
	K57	+	+	+	+	-	-
	K58	+	+	+	+	-	-
	K59	+	+	+	+	-	-
	K60	+	+	+	+	+	+
	K61	+	+	+	+	-	-
	K62	+	+	+	+	-	-

Table C2 (continued)

Source	Code	Sodium chloride concentration (%)					
		0	5	10	15	20	25
Pla-chom1	PJ52	+	+	+	+	-	-
	PJ139	+	+	+	+	-	-
Pla-chom 2	PJ78	+	+	+	+	+	+
Naw-mai-dong	NJ7	+	+	+	+	+	+
	NJ33	+	+	+	+	-	-
	NJ73	+	+	+	+	-	-
	NM154	+	+	+	+	-	-
	NM181	+	+	+	-	-	-
Bu-du	BD82	+	+	+	+	+	+
Tai-pla1	TP6	+	+	+	+	+	-
	TP57	+	+	+	+	+	-
Kaey	KT7	+	+	+	+	+	+
	KT9	+	+	+	+	-	-
Pu-chem	PD62	+	+	+	+	+	-
	PD67	+	+	+	+	-	-
Sai-pla	PP59	+	+	+	+	-	-
	PP134	+	+	+	+	+	+
	PP147	+	+	+	+	-	-
Gra-tiem-dong	KT56	+	+	+	+	-	-
Tao-chieo1	TK21	+	+	+	+	-	-
	TK53	+	+	+	+	-	-
	TK54	+	+	+	+	-	-
	TK145	+	+	+	+	-	-
	TK160	+	+	+	+	-	-

Table C2 (continued)

Source	Code	Sodium chloride concentration (%)					
		0	5	10	15	20	25
Tao-chieo1	TK161	+	+	+	+	-	-
	TK167	+	+	+	+	+	+
	Tk170	+	+	+	+	-	-
Tao-chieo 2	TN103	+	+	+	+	-	-
Tao-chieo 3	TS101	+	+	+	+	-	-
Sea water	SW1	+	+	+	+	-	-
Salt	SM25	+	+	+	+	+	+
Fermented fish	PT12	+	+	+	+	+	+
Sea solid waste	SW13	+	+	+	+	-	-
Sea sand	SN75	+	+	+	+	+	+
Phak-kard-dong (Pickled mustard)	PD69	+	+	+	+	-	-
Hua-phak-kard- chem	HK5	+	+	+	+	+	+
	HK20	+	+	+	+	-	-
Soil 1	SS36	+	+	+	+	+	+
	SS48	+	+	+	+	-	-
	SS59	+	+	+	+	+	+
Soil 2	SS50	+	+	+	+	+	+
	SS77	+	+	+	+	-	-
Soil 3	SS31	+	+	+	+	+	-
	SS54	+	+	+	+	+	-
	SS55	+	+	+	-	-	-
	SS56	+	+	+	-	-	-
	SS57	+	+	+	+	+	+

Table C3 The ability of growth of bacteria at various temperatures

Sources	Code	Temperature (°C)					
		35	40	45	50	55	60
Nam-pla1	PC253	+	+	+	-	-	-
	PC185	+	+	+	-	-	-
	PC255	+	+	+	-	-	-
	PC188	+	+	+	-	-	-
	PC189	+	+	+	-	-	-
	PC208	+	+	+	+	+	-
	PC212	+	+	+	-	-	-
	PC213	+	+	+	+	+	-
	PC214	+	+	+	-	-	-
	PC258	+	+	+	-	-	-
	PC191	+	+	+	-	-	-
	PC216	+	+	+	+	+	-
	PC164	+	+	+	-	-	-
	PC219	+	+	+	-	-	-
Nam-pla2	TP165	+	+	+	-	-	-
	TP259	+	+	+	-	-	-
	TP262	+	+	+	-	-	-
Nam-pla3	NN156	+	+	+	-	-	-
	NN157	+	+	+	-	-	-
	NN240	+	+	+	-	-	-
	FT176	+	+	+	-	-	-

Table C3 (continued)

Source	Code	Temperature (°C)					
		35	40	45	50	55	60
Nam-pla 4	FR26	+	+	+	-	-	-
Nam-pla 5	TR89	+	+	+	-	-	-
	TR90	+	+	+	-	-	-
	TR87	+	+	+	-	-	-
Nam-pla 6	SS61	+	+	+	-	-	-
	SS63	+	+	+	-	-	-
Nam-pla 7	K1	+	+	+	-	-	-
	K2	+	+	+	-	-	-
	K3	+	+	+	-	-	-
	K4	+	+	+	-	-	-
	K5	+	+	+	-	-	-
	K6	+	+	+	-	-	-
	K7	+	+	+	-	-	-
	K8	+	+	+	-	-	-
	K9	+	+	+	-	-	-
	K10	+	+	+	-	-	-
	K11	+	+	+	-	-	-
	K12	+	+	+	-	-	-
	K13	+	+	+	-	-	-
	K14	+	+	+	-	-	-
	K15	+	+	+	-	-	-
	K16	+	+	+	-	-	-

Table C3 (continued)

Source	Code	Temperature (°C)					
		35	40	45	50	55	60
	K17	+	+	+	-	-	-
	K18	+	+	+	-	-	-
	K19	+	+	+	-	-	-
	K20	+	+	+	-	-	-
	K21	+	+	+	-	-	-
	K22	+	+	+	-	-	-
Ka-pi 1	KJ16	+	+	+	-	-	-
	KJ81	+	+	+	-	-	-
	KJ119	+	+	+	-	-	-
Ka-pi 2	KM1	+	+	+	+	-	-
	KM2	+	+	+	-	-	-
	KM151	+	+	+	-	-	-
	KM232	+	+	+	+	+	-
Ka-pi 3	KK11	+	+	+	-	-	-
	KK12	+	+	+	-	-	-
	KK19	+	+	+	+	-	-
	KK22	+	+	+	-	-	-
	KK51	+	+	-	-	-	-

Table C3 (continued)

Sources	Code	Temperature (°C)					
		35	40	45	50	55	60
Ka-pi 3	KK92	+	+	-	-	-	-
	KK112	+	+	+	-	-	-
	KK119	+	+	+	+	-	-
	KK126	+	+	+	-	-	-
	KK127	+	+	+	+	-	-
	KK128	+	+	+	+	-	-
	KK137	+	+	+	-	-	-
	KK138	+	+	+	+	-	-
Ka-pi 4	K23	+	+	+	-	-	-
	K24	+	+	+	-	-	-
	K25	+	+	+	-	-	-
	K26	+	+	+	-	-	-
	K27	+	+	+	-	-	-
	K28	+	+	+	-	-	-
	K29	+	+	+	-	-	-
	K30	+	+	+	-	-	-
	K31	+	+	+	-	-	-
	K32	+	+	+	-	-	-
	K33	+	+	+	-	-	-
	K34	+	+	+	-	-	-
	K35	+	+	+	-	-	-

Table C3 (continued)

Source	Code	Temperature (°C)					
		35	40	45	50	55	60
Ka-pi 5	K36	+	+	+	-	-	-
	K37	+	+	+	-	-	-
	K38	+	+	+	-	-	-
	K39	+	+	+	-	-	-
	K40	+	+	+	-	-	-
	K41	+	+	+	-	-	-
	K42	+	+	+	-	-	-
	K43	+	+	+	-	-	-
	K44	+	+	+	-	-	-
	K45	+	+	+	-	-	-
Ka-pi 5	K46	+	+	+	-	-	-
	K47	+	+	+	-	-	-
	K48	+	+	+	-	-	-
	K49	+	+	+	-	-	-
	K50	+	+	+	-	-	-
	K51	+	+	+	-	-	-
	K52	+	+	+	-	-	-
	K53	+	+	+	-	-	-
Pla-ra1	RJ26	+	+	+	-	-	-
	RJ29	+	+	+	-	-	-
	RJ31	+	+	+	-	-	-
	RJ58	+	+	+	-	-	-
	RJ79	+	+	+	-	-	-
	RJ85	+	+	+	+	-	-
	RJ89	+	+	+	+	-	-

Table C3 (continued)

Source	Code	Temperature (°C)					
		35	40	45	50	55	60
Pla-ra1	RJ90	+	+	+	+	-	-
	RJ93	+	+	+	+	+	-
	RJ102	+	+	+	-	-	-
	RJ103	+	+	+	-	-	-
	RJ109	+	+	+	-	-	-
	RJ110	+	+	+	-	-	-
	RJ129	+	+	+	-	-	-
Pla-ra 2	RC179	+	+	+	-	-	-
	RC192	+	+	+	-	-	-
	RC194	+	+	+	-	-	-
	RC195	+	+	+	-	-	-
	RC210	+	+	+	+	-	-
	RM143	+	+	+	-	-	-
	RM146	+	+	+	+	+	-
	RM235	+	+	+	-	+	-
	RM249	+	+	+	-	-	-
Pla-ra 3	KK22	+	+	+	-	-	-
	KK23	+	+	+	-	-	-
	KK24	+	+	+	-	-	-
	KK38	+	+	+	-	-	-
	KK61	+	+	+	-	-	-

Table C3 (continued)

Sources	Code	Temperature (°C)					
		35	40	45	50	55	60
Pla-ra4	K54	+	+	+	-	-	-
	K55	+	+	+	-	-	-
	K56	+	+	+	-	-	-
	K57	+	+	+	-	-	-
	K58	+	+	+	-	-	-
	K59	+	+	+	-	-	-
	K60	+	+	+	-	-	-
	K61	+	+	+	-	-	-
	K62	+	+	+	-	-	-
Pla-chom 1	PJ52	+	+	+	-	-	-
	PJ139	+	+	+	-	-	-
Pla-chom 2	PJ78	+	+	+	-	-	-
Naw-mai-dong	NJ7	+	+	+	+	-	-
	NJ33	+	+	+	-	-	-
	NJ73	+	+	+	-	-	-
	NM154	+	+	+	-	-	-
	NM181	+	+	+	-	-	-
Bu-du	BD82	+	+	+	-	-	-

Table C3 (continued)

Sources	Code	Temperature (°C)					
		35	40	45	50	55	60
Tai-pla	TP6	+	+	+	-	-	-
	TP57	+	+	+	-	-	-
Kaey	KT7	+	+	+	-	-	-
	KT9	+	+	+	-	-	-
Pu-chem	PD62	+	+	+	+	-	-
	PD67	+	+	+	+	-	-
Pung-pla	PP59	+	+	+	+	+	-
	PP134	+	+	+	+	+	-
	PP147	+	+	+	+	+	-
Gra-tiem dong	KT56	+	+	+	-	-	-
Tao-chieo1	TK21	+	+	+	+	-	-
	TK53	+	+	+	+	+	-
	TK54	+	+	+	+	+	-
	TK145	+	+	+	+	+	-
	TK160	+	+	+	-	-	-
	TK161	+	+	+	+	-	-
	TK167	+	+	+	-	-	-
	TK170	+	+	+	-	-	-
Tao-chieo2	TN103	+	+	+	+	-	-
Tao-chieo3	TS101	+	+	+	-	-	-
Sea water	SW1	+	+	+	-	-	-
Salt 1	SM25	+	+	+	-	-	-
Pla-too-chem	P15	+	+	+	-	-	-

Table C3 (continued)

Sources	Code	Temperature (°C)					
		35	40	45	50	55	60
Sea shell solid waste	SW13	+	+	+	-	-	-
Sea sand	SN75	+	+	+	-	-	-
Phak-kard-dong	PD69	+	+	+	-	-	-
Hua-pak-kard-chem	HK5	+	+	+	-	-	-
	HK20	+	+	+	+	+	-
Soil 1	SS36	+	+	+	-	-	-
	SS48	+	+	+	+	-	-
	SS59	+	+	+	+	+	-
Soil 2	SS50	+	+	+	-	-	-
	SS77	+	+	+	-	-	-
Soil 3	SS31	+	+	+	-	-	-
	SS54	+	+	+	-	-	-
	SS55	+	+	+	-	-	-
	SS56	+	+	+	+	+	-
	SS57	+	+	+	+	-	-

Remark + = growth

- = no growth

Table C4 Protease production of isolated bacteria on skim milk agar containing various sodium chloride concentrations

Source	Code	Diameter of clear zone (cm)					
		Sodium chloride concentration (%)					
		0	5	10	15	20	25
Nam-pla 1	PC253	1.0	1.0	0	0	0	0
	PC185	0.7	0.8	0	0	0	0
	PC255	1.0	0.8	0	0	0	0
	PC188	0	0	0	0	0	0
	PC189	0	0	0	0	0	0
	PC208	0.7	0.7	0	0	0	0
	PC212	0.8	0.8	0	0	0	0
	PC 213	0	0.7	0	0	0	0
	PC 214	0.5	0	0	0	0	0
	PC 258	0	0	0	0	0	0
	PC 191	0.3	0	0	0	0	0
	PC 216	0.9	1.2	0	0	0	0
	PC 164	0.3	0	0	0	0	0
	PC 219	1.0	1.5	0	0	0	0
Nam-pla 2	TP165	0	0	0	0	0	0
	TP259	1.2	1.6	0	0	0	0
	TP262	0.5	1.0	0	0	0	0
Nam-pla 3	NN156	0	0	0	0	0	0
	NN157	0.4	0	0	0	0	0
	NN240	1.2	1.6	0	0	0	0

Table C4 (continued)

Sources	Code	Diameter of clear zone (cm)					
		Sodium chloride concentration (%)					
		0	5	10	15	20	25
Nam-pla 4	FT176	0.3	0	0	0	0	0
	FR26	0	0	0	0	0	0
Nam-pla 5	TR89	0.7	0	0	0	0	0
	TR90	0	0.9	0	0	0	0
	TR87	1.0	0	0	0	0	0
Nam-pla 6	SS61	0.8	0.4	0	0	0	0
	SS63	0	0	0	0	0	0
Nam-pla 7	K1	1.1	1.0	0	0	0	0
	K2	1.5	1.3	0	0	0	0
	K3	0.9	0.7	0	0	0	0
	K4	1.2	1.1	0	0	0	0
	K5	1.0	1.0	0	0	0	0
	K6	0	0	0	0	0	0
	K7	1.3	0.9	0	0	0	0
	K8	0	0	0	0	0	0
Nam-pla 7	K9	0	0	0	0	0	0
	K10	0.2	0.2	0	0	0	0
	K11	1.1	1.0	0	0	0	0
	K12	1.4	1.1	0	0	0	0
	K13	0.2	0.1	0	0	0	0

Table C4 (continued)

Sources	Code	Diameter of clear zone (cm)					
		Sodium chloride concentration (%)					
		0	5	10	15	20	25
	K14	0.9	0.8	0	0	0	0
	K15	0	0	0	0	0	0
	K16	0.1	0.1	0	0	0	0
	K17	0.8	0.7	0	0	0	0
	K18	0.8	0.8	0	0	0	0
	K19	1.2	0.9	0	0	0	0
	K20	1.1	0.9	0	0	0	0
	K21	1.5	1.3	0	0	0	0
	K22	0.7	0.6	0	0	0	0
Ka-pi 1	KJ16	1.5	0.65	0	0	0	0
	KJ81	1.2	1.4	0	0	0	0
	KJ119	1.0	1.8	0	0	0	0
Ka-pi 2	KM1	0.9	1.0	0	0	0	0
	KM2	1.2	1.5	0	0	0	0
	KM151	1.2	1.9	0	0	0	0
	KM232	1.5	1.8	0	0	0	0
Ka-pi 3	KK11	0	1.0	0	0	0	0
	KK12	1.3	1.5	0	0	0	0
	KK19	1.6	2.0	0	0	0	0
	KK22	1.1	1.5	0	0	0	0
	KK51	1.2	1.7	0	0	0	0
	KK92	1.2	1.4	0	0	0	0

Table C4 (continued)

Sources	Code	Diameter of clear zone (cm)					
		Sodium chloride concentration (%)					
		0	5	10	15	20	25
Ka-pi 3	KK112	1.6	1.5	0	0	0	0
	KK119	1.0	1.6	0	0	0	0
	KK126	1.0	1.6	0	0	0	0
	KK127	1.1	1.6	0	0	0	0
	KK128	1.1	1.5	0	0	0	0
	KK137	0.8	1.2	0	0	0	0
	KK138	1.5	1.9	0	0	0	0
Ka-pi 4	K23	1.2	1.3	0	0	0	0
	K24	1.1	1.1	0	0	0	0
	K25	0.9	0.7	0	0	0	0
	K26	1.0	0.9	0	0	0	0
	K27	0.7	0.6	0	0	0	0
	K28	0.3	0.4	0	0	0	0
	K29	0.5	0.5	0	0	0	0
	K30	0.7	0.7	0	0	0	0
	K31	0.9	0.7	0	0	0	0
	K32	1.3	1.2	0	0	0	0
	K33	1.1	1.0	0	0	0	0
	K34	0.8	0.7	0	0	0	0
	K35	1.1	1.1	0	0	0	0

Table C4 (continued)

Sources	Code	Diameter of clear zone (cm)					
		Sodium chloride concentration (%)					
		0	5	10	15	20	25
Ka-pi 5	K36	1.0	1.1	0	0	0	0
	K37	0.8	0.6	0	0	0	0
	K38	0	0	0	0	0	0
	K39	0.6	0.6	0	0	0	0
	K40	0.5	0.5	0	0	0	0
	K41	0.8	0.7	0	0	0	0
	K42	0.3	0.3	0	0	0	0
	K43	1.1	1.0	0	0	0	0
	K44	1.2	0.9	0	0	0	0
	K45	1.1	0.8	0	0	0	0
	K46	1.0	1.0	0	0	0	0
	K47	0	0	0	0	0	0
	K48	0.6	0.6	0	0	0	0
	K49	0.8	0.7	0	0	0	0
K50	0.4	0.4	0	0	0	0	
Ka-pi 5	K51	0.5	0.4	0	0	0	0
	K52	1.8	1.6	0	0	0	0
	K53	1.6	1.5	0	0	0	0

Table C4 (continued)

Sources	Code	Diameter of clear zone (cm)					
		Sodium chloride concentration (%)					
		0	5	10	15	20	25
Pla-ra 1	RJ26	1.5	1.6	0	0	0	0
	RJ29	0.8	0	0	0	0	0
	RJ31	0	0	0	0	0	0
	RJ58	0.9	0.8	0	0	0	0
	RJ79	0	1.2	0	0	0	0
	RJ85	1.3	1.7	0	0	0	0
	RJ89	1.6	1.7	0	0	0	0
Pla-ra 1	RJ93	0.9	1.2	0	0	0	0
	RJ102	1.5	1.8	0	0	0	0
	RJ103	1.0	1.2	0	0	0	0
	RJ109	1.2	1.7	0	0	0	0
	RJ110	1.5	1.0	0	0	0	0
	RJ129	1.8	1.7	0	0	0	0
Pla-ra 2	RC179	0.4	0	0	0	0	0
	RC192	1.8	1.8	0	0	0	0
	RC194	2.0	1.4	0	0	0	0
	RC195	0	0	0	0	0	0
	RC210	1.4	1.7	0	0	0	0
	RM143	1.7	1.2	0	0	0	0
	RM146	1.3	1.1	0	0	0	0
	RM235	1.2	1.0	0	0	0	0
	RM249	1.7	1.7	0	0	0	0

Table C4 (continued)

Sources	Code	Diameter of clear zone (cm)					
		Sodium chloride concentration (%)					
		0	5	10	15	20	25
Pla-ra 3	PR22	1.1	1.5	0	0	0	0
	PR23	1.6	1.3	0	0	0	0
	PR24	1.3	1.8	0	0	0	0
	PR38	0	0	0	0	0	0
	PR61	1.5	2.0	0	0	0	0
Pla-ra 4	K54	1.1	0.9	0	0	0	0
	K55	1.2	1.1	0	0	0	0
	K56	1.0	1.0	0	0	0	0
	K57	0.7	0.7	0	0	0	0
	K58	0.8	0.8	0	0	0	0
	K59	0.2	0.2	0	0	0	0
	K60	0.4	0.4	0	0	0	0
	K61	1.9	1.9	0	0	0	0
	K62	1.2	1.0	0	0	0	0
Pla-chom 1	PJ52	1.2	1.7	0	0	0	0
	PJ139	1.0	0.4	0	0	0	0
Pla-chom 2	PJ78	0	0	0	0	0	0
Naw-mai-dong	NJ7	1.0	1.1	0	0	0	0
	NJ33	1.4	1.6	0	0	0	0
	NJ73	1.3	1.3	0	0	0	0
	NM154	0	0	0	0	0	0
	NM181	0.5	1.0	0	0	0	0
Bu-du	BD82	0	0	0	0	0	0

Table C4 (continued)

Sources	Code	Diameter of clear zone (cm)					
		Sodium chloride concentration (%)					
		0	5	10	15	20	25
Tai-pla 1	TP6	1.2	1.4	0	0	0	0
	TP57	1.2	1.2	0	0	0	0
Koey	KT7	0	0	0	0	0	0
	KT9	0	0	0	0	0	0
Pu-chem	PD62	1.1	0.5	0	0	0	0
	PD67	1.1	1.2	0	0	0	0
Pung-pla	PP59	1.4	1.7	0	0	0	0
	PP134	1.4	1.4	0	0	0	0
	PP147	1.2	1.0	0	0	0	0
Gra-tiem-dong	KT56	0	0	0	0	0	0
Tao-chieo1	TK21	1.5	1.2	0	0	0	0
	TK53	1.0	1.0	0	0	0	0
	TK54	0.9	0	0	0	0	0
	TK145	1.4	0	0	0	0	0
	TK160	0.4	0	0	0	0	0
	TK161	1.2	0	0	0	0	0
	TK167	0.7	0	0	0	0	0
	TK170	0	0	0	0	0	0
Tao-chieo2	TN103	2.1	1.5	0	0	0	0

Table C4 (continued)

Sources	Code	Diameter of clear zone (cm)					
		Sodium chloride concentration (%)					
		0	5	10	15	20	25
Tao-chieo3	TS101	2.0	0.9	0	0	0	0
Sea Water	SW1	0	0	0	0	0	0
Salt 1	SM25	0	0	0	0	0	0
Pla-too-chem	P15	1.5	1.0	0	0	0	0
Sea shell solid waste	SW13	1.5	0.5	0	0	0	0
Sea sand	SN75	0.8	0	0	0	0	0
Phak-kard-dong	PD69	2.0	1.5	0	0	0	0
Hua-pak-kard-chem	HK5	0	0	0	0	0	0
	HK20	1.8	1.5	0	0	0	0
Soil 1	SS36	0.7	1.2	0	0	0	0
	SS48	2.0	1.2	0	0	0	0
	SS59	1.8	1.0	0	0	0	0
Soil 2	SS50	0.7	0	0	0	0	0
	SS77	2.0	1.0	0	0	0	0
Soil 3	SS31	1.8	1.2	0	0	0	0
	SS54	1.8	1.2	0	0	0	0
	SS55	1.3	0.5	0	0	0	0
	SS56	1.9	1.3	0	0	0	0
	SS57	1.8	1.2	0	0	0	0

Table C5 Protease production of isolated bacteria in PY broth in test tube shaking 150 rpm at room temperature for 48 h

Sources	Code	Specific activity (U/mg)
Nam-pla 1	PC253	0.030
	PC185	0.026
	PC255	0.030
	PC188	0.020
	PC189	0.011
	PC208	0.038
Nam-pla 1	PC212	0.029
	PC 213	0.018
	PC 214	0.014
	PC 258	0.013
	PC 191	0.014
	PC 216	0.043
	PC 164	0.014
	PC 219	0.047
Nam-pla 2	TP165	0.017
	TP259	0.051
	TP262	0.026
Nam-pla 3	NN156	0.054
	NN157	0.064
	NN240	0.049
Nam-pla 4	FT176	0.025
	FR26	0.012
Nam-pla 5	TR89	0.017
	TR90	0.017
Nam-pla 6	TR87	0.017
	SS61	0.052
	SS63	0.012

Table C5 (continued)

Sources	Code	Specific activity (U/mg)
Ka-pi 1	K21	0.088
	K22	0.037
	KJ16	0.020
	KJ81	0.051
	KJ119	0.078
Ka-pi 2	KM1	0.030
	KM2	0.050
	KM151	0.060
	KM232	0.070
Ka-pi 3	KK11	0.012
	KK12	0.096
	KK19	0.095
	KK22	0.077
	KK51	0.078
	KK92	0.025
	KK112	0.050
	KK119	0.070
	KK126	0.085
	KK127	0.057
	KK128	0.102
	KK137	0.090
	KK138	0.057

Table C5 (continued)

Sources	Code	Specific activity (U/mg)
Ka-pi 4	K23	0.060
	K24	0.088
	K25	0.050
	K26	0.056
	K27	0.013
	K28	0.056
	K29	0.012
	K30	0.056
	K31	0.032
	K32	0.030
	K33	0.037
	K34	0.018
	K35	0.038
	Ka-pi 5	K36
K37		0.056
K38		0.026
K39		0.043
K40		0.037
K41		0.060
K42		0.060
K43		0.032
K44		0.050
K45		0.056
K46		0.008
K47		0.043
K48		0.051
K49		0.052
K50		0.026
K51		0.064
K52		0.115
K53		0.109

Table C5 (continued)

Sources	Code	Specific activity (U/mg)
Pla-ra 1	RJ26	0.063
	RJ29	0.013
	RJ31	0.008
	RJ58	0.032
	RJ79	0.064
	RJ85	0.079
	RJ89	0.057
	RJ93	0.038
	RJ102	0.022
	RJ103	0.045
	RJ109	0.045
	RJ110	0.029
	RJ129	0.025
Pla-ra 2	RC179	0.015
	RC192	0.075
	RC194	0.090
	RC195	0.012
	RC210	0.084
	RM143	0.029
	RM146	0.036
	RM235	0.052
RM249	0.060	

Table C5 (continued)

Sources	Code	Specific activity (U/mg)
Pla-ra 3	KK22	0.067
	KK23	0.055
	KK24	0.049
Pla-ra 3	KK38	0.018
	KK61	0.052
Pla-ra 4	K54	0.032
	K55	0.052
	K56	0.040
	K57	0.060
	K58	0.047
	K59	0.026
	K60	0.060
	K61	0.123
	K62	0.073
Pla-chom 1	PJ52	0.071
	PJ139	0.075
Pla-chom 2	PJ78	0.014
Naw-mai-dong	NJ7	0.04
	NJ33	0.018
	NJ73	0.018
	NM154	0.040
	NM181	0.030

Table C5 (continued)

Sources	Code	Specific activity (U/mg)
Bu-du	BD82	0.023
Tai-pla 1	TP6	0.029
	TP57	0.043
Kaey	KT7	0.032
	KT9	0.033
Pu-chem	PD62	0.016
	PD67	0.017
Pung-pla	PP59	0.023
	PP134	0.030
	PP147	0.040
Gra-tiem-dong	KT56	0.013
Tao-chieo1	TK21	0.050
	TK53	0.014
	TK54	0.013
	TK145	0.030
	TK160	0.030
	TK161	0.030
	TK167	0.033
	TK170	0.032
Tao-chieo2	TN103	0.091

Table C5 (continued)

Sources	Code	Specific activity (U/mg)
Tao-chieo3	TS101	0.030
Sea water	SW1	0.021
Salt 1	SM25	0.028
Pla-too-chem	P15	0.026
Sea shell solid waste	SW13	0.011
Sea sand	SN75	0.031
Phak-kard-dong	PD69	0.039
Hua-pak-kard-chem	HK5	0.018
	HK20	0.040
Soil 1	SS36	0.039
	SS48	0.018
	SS59	0.018
Soil 2	SS50	0.022
	SS77	0.071
Soil 3	SS31	0.026
	SS54	0.070
	SS55	0.047
	SS56	0.061
	SS57	0.062

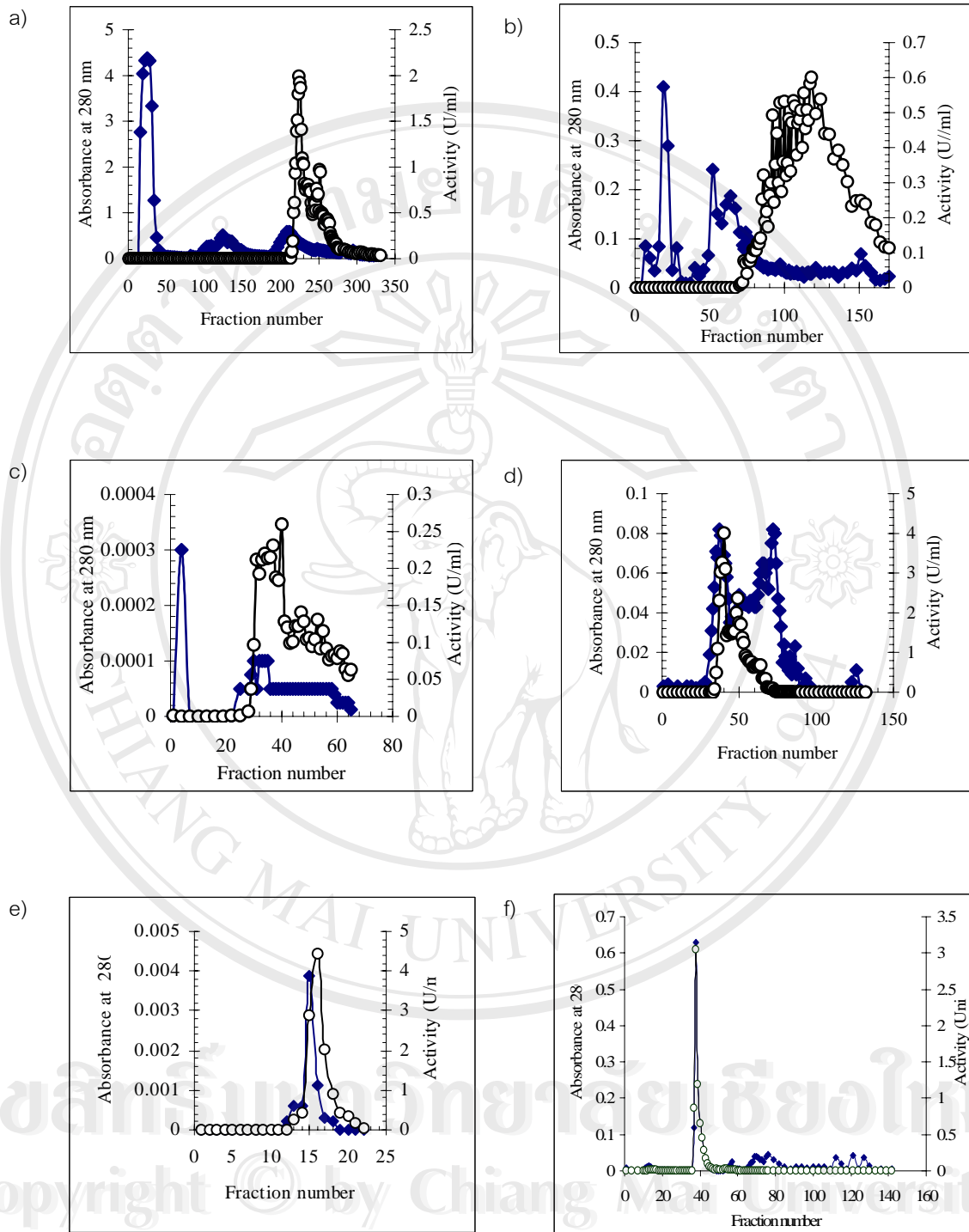


Figure C1 Elution protein profile (protein absorbance at 280 nm) a) DEAE-Toyopearl b) Hydroxyapatite c) Mono Q d) Gel filtration e) MonoQ f) Hydroxyapatite (Absorbance 280 nm, ◆), (Activity (U/ml), ○)

CURRICULUM VITAE

- Name:** Ms. Somkid Deejing
- Date of birth:** August 6, 1971
- Place of birth:** Sukhothai
- Academic background:** B.S. (Biology) in the major of Biology,
Faculty of Science, Narasuan University, 1991.
- M.S. (Microbiology) in the major of
Microbiology, Faculty of Science, Kasetsart
University in 1995.
- Working experience:** Lecturer in the Department of Biology,
Faculty of Science, Maejo University in
1995-present.
- Scholarship:** Received a UDC scholarship of Ministry of
Education in 2000.
- Received a scholarship from Graduate School
Foundation of Chiang Mai University in 2001.
- Received a scholarship from the Institute for the
Promotion of Teaching Science and Technology
in 2001.

Received a Japanese Government (Monbusho) Scholarship for research student in 2002-2004.

Academic award: Award from oral presentation in 34th Annual Meeting of Kasetsart University in the topic "Hybridization of *Saccharomyces cerevisiae* and *Zygosaccharomyces rouxii* by protoplast fusion technique for construction of halotolerant ethanol producing yeast" in 1997.

Academic conference and publication:

Oral presentation in the topic "Purification and characterization of aminopeptidase from *Bacillus* sp." in the Kushu Branch Annual Meeting of Japan Society for Bioscience, Biotechnology and Agrochemistry. September 19, 2003 at Kagoshima University, Kagoshima, Japan.

Deejing, S., Moriguchi, M. and Lumyong, S. 2005. Characterization of various protease from salt-tolerant bacteria isolated from Thai fermented foods. Chiang Mai Journal of Science. (*in press*).

Deejing, S., Yoshimune, K., Lumyong, S. and Moriguchi, M. Purification and characterization of hyperthermotolerant leucine aminopeptidase from *Geobacillus thermoleovorans* 47b. Journal of Industrial Microbiology and Biotechnology. (*in press*).

