

7. APPENDICES

Appendix 1: Reagents and solutions

I. Reagents and solutions

1. Porcine cysticercosis:

Cysticerci examination: 10 % HCl, 10% formalin and 100% alcohol

2. Trichinellosis

Blood serum samples

Microtiter plates coated with *Trichinella* antigen (larval excretory-secretory antigen of *Trichinella spiralis*): 50 µl *Trichinella* E/S antigen per well.

Trichinella-positive control serum (1 ml lyophilized)

Trichinella-negative control serum (1 ml lyophilized)

PBS buffers (Physiological buffer solution)

Sterile distilled waters (Asqua bitus)

Ati-pig IgG-peroxidase conjugates per diluted 1:10 (SIGMA, product N°. A5670)

ABTS buffer, dry matter from Boehringer

Stop solution

Tablets chromogen ABTS

2000 ml of PBS-Tween 20 (pH=7.2-7.4) consist of

KH ₂ PO ₄	0.4 g
---------------------------------	-------

Na ₂ HPO ₄ x 12H ₂ O	5.8 g
-------------------------------------------------------	-------

NaCl	16.0 g
------	--------

KCl	0.4 g
-----	-------

Tween 20	1.0 ml
----------	--------

Distilled water	ad 2000 ml
-----------------	------------

Appendix 2: Survey questionnaires**I. Hospital questionnaire survey of neurocysticercosis in humans in selected hospital**

Date_____/_____/_____;

Name of the Hospital_____ Location_____

Name of the Doctor_____ Title _____

1. Patient's residence areas:_____

2. Number of the patients showing following clinical signs:

a) Seizure_____ b) Violent behavior_____ c) Head ache_____

d) Depression_____

e) Facial palsy_____ Monoparesis or hemiplegia.

3. What is the method of diagnosis used in the hospital?

a) CT scan_____ b) ELISA_____ c) MRI_____ d) Other_____

4. How many epileptic cases are found for the period of:

a) Year 2000_____ b) 2001_____ c) 2002_____ d) 2003_____

5. How many confirmed neurocysticercosis cases:

a) Year 2000_____ b) 2001_____ c) 2002_____ d) 2003_____

6. Age groups:

a) 0-14 year_____ b) 15-35 years_____ c) About_____

7. Sexes:

a) Male_____ b) Female_____

8. Patient's socioeconomic status:

a) Upper_____ b) Middle_____ c) Lower_____

9. Treatment:

a) Surgery_____ b) Medical _____

10. Name of the drugs used:

a)_____

b)_____

c) _____

11. Is the treatment effective?

a) Yes____ b) No____

12. List the patients according to caste:

a) Brahmins____ b) Non-brahmins____

13. Other comments: _____

II. Slaughterhouse questionnaire survey of porcine cysticercosis and trichinellosis

Date____/____/____;

Interviewee name _____ Title _____

Interviewer name _____ Title _____

Name of slaughterhouse _____

Location _____

1. Educational level

a) Low____ b) Medium____ c) High____

2. Slaughter pigs for consumption per day:

a) 1-100____ b) 100-500____ c) 500-2000____ d) more than 2000

3. Average slaughter pigs per day in each slaughterhouse:

a) 50-200____ b) 200-300____ c) 300-600____ d) More than 600____

4. Environment and hygienic aspects in slaughterhouses

a) Good__ b) Poor____

5. Pig origins?

a) Household raising ____ b) Small and medium farm____

c) Intensive farm ____

6. Have you ever heard porcine cysticercosis?

a) Yes____ b) No____

7. Have you ever heard trichinellosis?

a) Yes____ b) No____

8. Inspected pigs for porcine cysticercosis?

a) Yes____ b) No____

9. Which method use for diagnosis?

a) Tongue palpation___ b) Meat inspection___ c) Both___

10. Have you ever found parasitic diseases of slaughter pigs?

a) Yes___ b) No___

11. Have you ever found infected cases of cysticercosis?

a) Yes___ b) No___

12. How many cases per month?

a) 1-3___ b) 3-5___ c) more than 5___

13. Most confirmed cases?

a) Household raising ___ b) Small and medium farms ___

c) Intensive farm ___

14. Inspected pigs for trichinellosis?

a) Yes___ b) No___

15. Ages of slaughter pigs

a) Young___ b) Medium___ c) Old___ d) All___

16. Pig breed for slaughtering?

a) Local___ b) Exotic___ c) Crossed bred___

17. Most ages of infected pigs

a) Young___ b) Medium___ c) Old___ d) All___

18. Most cases infected?

a) Heavy___ b) Light ___

19. Infected carcasses and organs use for:

a) Human consumption___ b) Processing___ c) Culling___

20. Food habits

a) Cooked___ b) Uncooked___ c) Mixed___

21. Disease control and prevention?

a) Yes___ b) No___

22. Other comments: _____

III. Pig farm questionnaire survey of porcine cysticercosis and trichinellosis in 3 provinces

Date ____/____/____;

Interviewee name _____ Title _____

Interviewer name _____ Title _____

Name of farm or village _____

Name of owner _____

Location _____

Date of establishment ____/____/____

1. Educational level

a) Low ____ b) Medium ____ c) High ____

2. Sizes of pig production

a) House hold raising ____ b) Small & medium ____ c) Intensive ____

3. Pig production system?

a) Free ranging ____ b) Mixed ____ c) Indoor ____

4. Target of pig production?

a) Piglet ____ b) Fattening ____ c) Mixed ____

5. Buying piglets from outside for raising?

a) Yes ____ b) No ____

6. Age of pigs for selling:

a) 2-4 month ____ b) 6 month ____ c) 7-12 month ____

d) More than 12 month ____

7. Pig breed for raising:

a) Local ____ b) Exotic ____ c) Crossed bred ____

8. Do they have an identification number?

a) Yes ____ b) No ____

9. Types of feed:

a) Ratio feed ____ b) Leftovers ____ c) Mixing ____ d) Traditional ____

10. Getting feeds from outside?

11. Drinking water for pigs and human uses:

- a) Tap water___ b) Underground water___ c) Pond water___ d) River___
12. Condition of water:
- a) Direct use___ b) Treatment___
13. Condition of the holdings: Floor type
- a) Concreted___ b) un-concreted___ c) Mixed
14. Environment of pig raising areas:
- a) Clean___ b) Dirty___
15. Waste management:
- a) Good___ b) Poor
16. Having problems with parasitic diseases
- a) Yes___ b) No___
17. Have you ever heard porcine cysticercosis?
- a) Yes___ b) No___
18. Have you ever heard of *Taenia solium*?
- a) Yes___ b) No___
19. Have you ever heard of trichinellosis?
- a) Yes___ b) No___
20. Investigate pigs for cysticercosis?
- a) Yes___ b) No___
21. Which method use for diagnosis?
- a) Tongue palpation___ b) Meat inspection___ c) No inspected___
22. Investigate pigs for trichinellosis?
- a) Yes___ b) No___
23. Have ever confirmed porcine cysticercosis?
- a) Yes___ b) No___
24. How many infected cases of cysticercosis per month?
- a) 1-3 case___ b) 3-5___ c) More than 5___
25. Age of infected pig confirmed porcine cysticercosis?
- a) 2-4 month___ b) 6 month___ c) 7-12 month___
- d) More than 12 month___
26. Infected pigs sale for:
- a) Human consumption___ b) Processing ___ c) Culling___

27. Food habits

a) Cooked___ b) Uncooked___ c) Mixed___

28. Are there sufficient toilets?

a) Yes___ b) No___

29. In the farms or villages, are there human cases of *Taenia solium* or neurocysticercosis?

a) Yes___ b) No___

30. Disease control and prevention?

a) Yes___ b) No___

31. Is the measure effective?

a) Yes___ b) No___

32. Treatment:

a) Conventional___ b) Traditional___ c) Mixed___

33. Is the treatment effective?

a) Yes___ b) No___

34. Other comments:

Appendix 3. Degree of infection by cysticerci in organs and muscles (n=47)

Pig N°	Degree of infection by cysticerci in muscles and organs of carcasses													
	To	Es	He	Di	Li	Lu	S	Ki	Br	Ext	Int	Tri	Fl	HI
1	L	L	H	L	N	N	N	N	N	H	H	L	H	H
2	L	N	L	L	N	N	N	N	N	H	H	H	H	H
3	N	L	L	L	N	N	N	N	N	L	H	H	H	H
4	H	H	H	H	N	N	N	N	N	L	L	L	L	L
5	L	N	L	L	N	N	N	N	N	L	H	H	H	H
6	N	L	L	L	N	N	N	N	N	H	H	H	H	H
7	L	L	L	L	N	N	N	N	N	H	H	L	H	H
8	H	H	H	H	N	N	N	N	N	L	L	L	L	H
9	L	L	H	L	N	N	N	N	N	H	H	H	H	H
10	N	L	L	L	N	N	N	N	N	L	N	L	L	H
11	N	L	L	L	N	N	N	N	N	H	H	H	H	H
12	L	L	H	H	N	N	N	N	N	L	L	L	H	H
13	H	H	H	H	N	N	N	N	N	H	H	H	H	H
14	N	N	L	L	N	N	N	N	N	H	H	H	H	H
15	N	L	L	L	N	N	N	N	N	L	L	H	H	H
16	L	N	L	L	N	N	N	N	N	H	H	L	H	H
17	H	H	H	L	N	N	N	N	N	N	N	L	L	L
18	N	L	L	L	N	N	N	N	N	N	L	L	N	N
19	N	L	L	L	N	N	N	N	N	H	H	H	H	H
20	N	L	L	L	N	N	N	N	L	H	H	H	H	H
21	L	L	H	L	N	N	N	N	N	H	H	H	H	H
22	N	N	L	L	N	N	N	N	N	H	L	H	H	H
23	H	H	L	H	N	N	N	N	N	L	L	L	H	H
24	L	L	L	H	N	N	N	N	N	H	H	H	H	H
25	H	L	H	H	N	N	N	N	L	H	H	H	H	H
26	L	L	H	H	N	N	N	N	L	H	L	L	H	H

Appendix 3(continued). Degree of infection by cysticerci in organs and muscles (n=47)

Pig N ^o	Degree of infection by cysticerci in muscles and organs of carcasses													
	To	Es	He	Di	Li	Lu	S	Ki	Br	Ext	Int	Tri	Fl	Hi
27	H	N	L	L	N	N	N	N	N	H	H	H	H	H
28	N	L	L	L	N	N	N	N	N	H	H	H	H	H
29	L	L	H	H	N	N	N	N	N	N	N	L	N	L
30	L	N	H	H	N	N	N	N	N	H	H	H	H	H
31	L	L	L	L	N	N	N	N	N	H	L	L	H	H
32	N	L	H	H	N	N	N	N	N	H	H	H	H	H
33	L	N	L	H	N	N	N	N	N	L	L	H	H	H
34	H	L	H	L	N	N	N	N	N	H	H	H	H	H
35	L	L	H	H	N	N	N	N	N	H	H	H	H	H
36	L	L	L	L	N	N	N	N	N	L	N	L	L	L
37	H	H	L	H	N	N	N	N	N	H	H	H	H	H
38	N	N	L	L	N	N	N	N	N	H	L	L	L	H
39	H	H	L	H	N	N	N	N	N	L	N	L	N	N
40	L	L	H	L	N	N	N	N	N	H	H	L	H	H
41	L	L	L	L	N	N	N	N	N	H	H	H	H	H
42	H	H	H	H	N	N	N	N	N	L	L	L	H	H
43	N	L	H	H	N	N	N	N	N	H	H	H	H	H
44	N	N	L	L	N	N	N	N	N	H	H	L	H	H
45	L	L	H	L	N	N	N	N	N	H	H	H	H	H
46	H	H	H	H	N	N	N	N	N	L	H	H	H	H
47	N	L	L	L	N	N	N	N	N	H	H	H	H	H
T.L	19	28	27	29	0	0	0	0	3	13	12	19	6	4
T.H	12	9	20	18	0	0	0	0	0	31	30	28	38	41

Note: N, negative = 0 or no cysts; L, light infection (Low) \leq 1-100 cysts per organ; H, heavy infection (High) $>$ 100 cysts per organ; TL, total light infection; TH, total heavy infection.

Key: To, tongue; Es, esophagus; He, heart; Di, Diaphragm; Li, liver; Lu, lungs; Ki, kidney; S, spleen; Br, brain; Int, internal masseter; Ext, external masseter; Tri, triceps brachii; Fl, forelimbs and Hi, hind limbs.

Appendix 4. Summary results of questionnaire surveys in pig slaughterhouses around Phnom Penh / Cambodia (n=70)

N o.	Questions	0 (%)	1 (%)	2 (%)	3 (%)	4 (%)	Total No.
1	Educational level (1=Low, 2=Medium, 3=High)		60	31	9		70
2	Slaughter pigs for consumption per day (1= 1-100, 2=100-500, 3=500-2000, 4=more 2000)		0	3	89	8	70
3	Average slaughter pigs per day in each slaughterhouse: (1=50-200,2=200-300, 3=300-600 4=More than 600)		11	67	22	0	70
4	Environment and hygienic aspects in slaughterhouses (1=Good, 2=Poor)		0	100			70
5	Pig origins? (1=Household raising, 2= Small and medium farm, 3=Intensive farm)		40	33	27		70
6	Have ever heard of porcine cysticercosis? (1=Yes, 0=No)	36	64				70
7	Have ever heard of trichinellosis? (1=Yes, 0=No)	61	39				70
8	Inspected pigs for cysticercosis? (1=Yes, 0=No)	43	57				70
9	Which method? (1=Tongue palpation, 2=Meat inspection, 3=Both)		13	31	56		70
10	Find parasitic diseases of slaughter pigs? (1= Yes, 0=No)	37	63				70
11	Confirmed infected cases of cysticercosis? (1=Yes, 0=No)	37	63				70
12	How many cases per month? (1=1-3, 2=3-5, 3=more than 5)		63	31	6		70
13	Most confirmed cases? (1= Household raising, 2= Small and medium farms, 3= Intensive farm)		74	26	0		70

Appendix 4(continued). Summary results of questionnaire surveys in pig slaughterhouses around Phnom Penh/ Cambodia (n=70)

N o.	Questions	0 (%)	1 (%)	2 (%)	3 (%)	4 (%)	Total No.
14	Inspected pigs for trichinellosis? (1=Yes, 0=No)	100	0				70
15	Ages of slaughter pigs (1=Young, 2=Medium, 3=Old, 4=All)		9	64	13	14	70
16	Pig breed? (1=Local, 2=Exotic, 3=Crossed bred)		50	24	26		70
17	Most ages of infected pigs (1=Young, 2=Medium, 3=Old and 4=All)		10	36	24	30	70
18	Most cases infected? (1= Heavy, 2=Light)		57	43			70
19	Infected carcasses and organs use for: (1=Human consumption, 2=Processing, 3=Culling)		14	64	22		70
20	Food habits (1=Cooked, 2=Uncooked, 3=Mixed)		57	16	27		70
21	Disease control and prevention? (1=Yes, 0=No)	76	24				70

Appendix 5. Summary results of questionnaire surveys of pig farms in 3 provinces / Cambodia, (n=132)

N o.	Questions	0 (%)	1 (%)	2 (%)	3 (%)	4 (%)	Total No.
1	Educational level (1=Low, 2=Medium, 3=High)		79	16	5		132
2	Sizes of pig production (1= House hold raising, 2=Small &medium and 3=Investment)	=	85	15	0		132
3	Pig production system? (1= Free ranging, 2= Mixed, 3= Indoor)		86	11	3		132
4	Target of pig production? (1=Piglet, 2= Fattening, 3=Mixed)		31	45	24		132
5	Buying piglets from outside for raising? (1=Yes, 0=No)	35	65				132
6	Age of pigs for selling (1= 2-4 month, 2=6 month, 3=7-12 month, 4=More than 12 month)		32	22	36	10	132
7	Pig breed for raising (1=local, 2=Exotic, 3=Crossed bred)		89	3	8		132
8	Identification number (1=Yes, 0=No)	99	1				132
9	Types of feeds (1=Ratio feed, 2=Leftovers, 3=Mixing, 4=Traditional)		5	17	24	54	132
10	Getting feeds from outside? (1=Yes, 0=No)	55	45				132
11	Drinking water for pigs and human uses: (1=Tap water, 2=Underground, 3=Pond, 4=River)		0	17	66	17	132
12	Condition of water: (1=Direct use, 2=Treated)		93	7			132
13	Condition of holdings: floor type (1=Concreted, 2=Un-concreted 3=Mixed)		4	78	18		132
14	Environment of pig raising areas (1=Clean, 2=Dirty)		17	83			132
15	Waste management (1=Good, 2=Poor)		11	89			132
16	Having problems with parasitic diseases (1=Yes, 2=No)	17	83				132

Appendix 5 (continued). Summary results of questionnaire surveys of pig farms in 3 provinces / Cambodia, (n=132)

N o.	Questions	0 (%)	1 (%)	2 (%)	3 (%)	4 (%)	Total No.
17	Have ever heard of porcine cysticercosis? (1=Yes, 0=No)	40	60				132
18	Have ever heard of <i>T.solium</i> ? (1=Yes, 0=No)	54	46				132
19	Have ever heard of trichinellosis? (1=Yes, 0=No)	83	17				132
20	Investigate pigs for cysticercosis? (1=Yes, 0=No)	34	66				132
21	Which method? (1=tongue palpation, 2=Meat inspection, 3=No inspected)		66	0	34		132
22	Investigate pigs for trichinellosis? (1=Yes, 0=No)	100	0				132
23	Have ever confirmed porcine cysticercosis? (1=Yes, 0=No)	55	45				132
24	How many infected cases of cysticercosis per month (0= No case, 1=1-3, 2=3-5, 3=more than 5)	54	28	18			132
25	Age of infected pig confirmed porcine cysticercosis? (1=4 month, 2=6 month, 3=7-12 month, 4= More than 12 month.		24	26	40	10	132
26	Infected pigs sale for: (1=Human consumption, 2=Processing, 3=Culling)		14	61	25		132
27	Food habit (1=Cooked, 2=Uncooked, 3=Mixed)		59	6	35		132
28	Sufficient toilets? (1=Yes, 0=No)	84	16				132
29	Human cases of <i>Taenia solium</i> ? (1=Yes, 0=No)	70	30				132
30	Disease control and prevention? (1=Yes, 0=No)	79	21				132
31	Is the measure effective? (1=Yes, 0=No)	89	11				132
32	Treatment: (1=Conventional, 2=Traditional, 3=Mixed)		41	33	26		132
33	Is the treatment effective? (1=Yes, 0=No)	33	67				132

CURRICULUM VITAE

1. General background:

Family name: THAN
 First name: Sovyra
 Date of birth: 04 February 1968
 Place of birth: Siem Reap, Cambodia
 Nationality: Khmer (Cambodian)
 Marital status: Married
 Language skill: English, Russian and Khmer- very good
 Home Address: #47, St.464, Toulthom Pong II, Chamkarmon,
 Phnom Penh, Cambodia
 Phone: (855) 12 727 196, 12 592 090
 E-mail: t_sovyra@hotmail.com or
sovyra@yahoo.com
 Office Address: #74, Monivong Blvd, Daun Penh,
 Phnom Penh, Cambodia
 Department of Animal Health and
 Production
 E-mail: t_sovyra@hotmail.com or
sovyra@yahoo.com

2. Education

<u>Date from- to</u>	<u>Institution</u>	<u>Degree or Diploma</u>
2000-2002	Pannasatra University of Cambodia, Phnom Penh, Cambodia	Diploma of English for Academic Purpose
1986-1993	Kharkov Zoo-Veterinary	DVM, MSc.vet.sc.

	Institute	
	Kharkov, Ukraine	
1983-1986	Toul Kok High School	Diploma of High School
	Phnom Penh, Cambodia	
1980-1986	Siem Reap Secondary School	Diploma of Secondary School
	Siem Reap Province, Cambodia	

3. Professional Experience

<u>Date from- to</u>	<u>Description</u>
Oct. 2003-Sept.2005	Attended MSc. Course Program in Veterinary Public Health, Joint International Post-Graduate Studies in VPH between Faculties of Veterinary Medicine of Freie Universitat Berlin, Germany and Chiang Mai University, Thailand.
Mar.2003-Apr.2003	National Consultant of EC-project on Support to Veterinary and Livestock Services in Cambodia.
Aug.2002-Apr.2003	Member of National Consultation Committee (NCC) of FAO on Domesticated Animal genetic Resources.
May2002-June2003	Member of National Reformed Consultation Committee on Veterinary Sub-Degree for hygiene and inspection of animal meats and animal products in slaughterhouses, slaughtering sites and Markets.
Jan.2002-Feb. 2002	National Consultant of PRASAC- project on Assessment of nutrition and forage

	development in Cambodia.
-Jan.2001-Nov.2001	Supervisor of Bachelor Thesis's students from Royal University Agriculture of Cambodia
Jun.2001-to date	Chief of Livestock Division of Animal Production Office, Department of Animal Health and Production.
Jan.1997-Apr.1997	Supervisor of Thesis's students from Prek Leap Agriculture College
Oct.1996-Feb.1997	Chief of inspecting sector of Animal Quarantine Station.
1995-2001	Manager of own pig farm
1994-2001	Technical staff of Animal Production.
1993-2003	Manager of Small Animal and Pet Clinic in Phnom Penh.

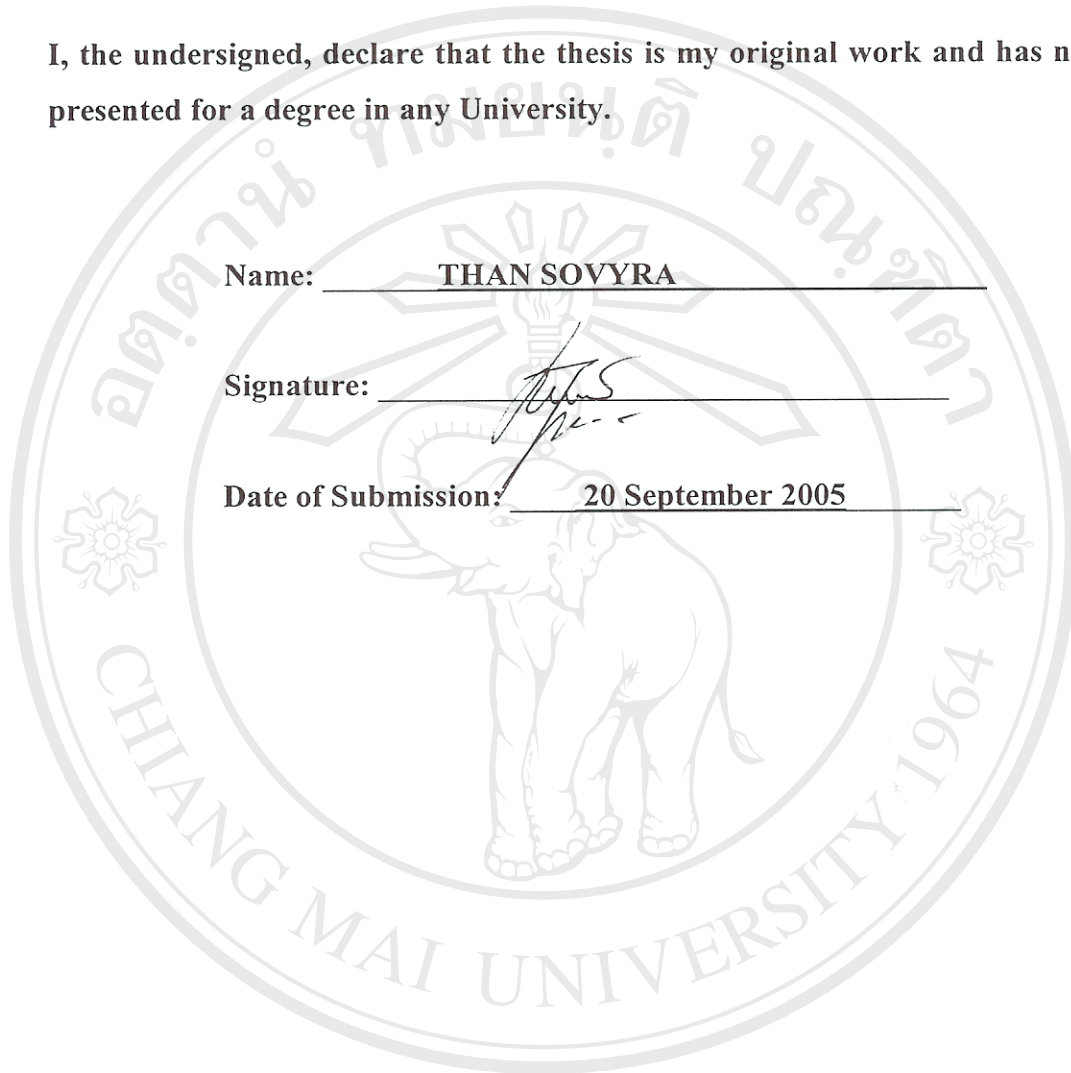
DECLARATION

I, the undersigned, declare that the thesis is my original work and has not been presented for a degree in any University.

Name: THAN SOVYRA

Signature: 

Date of Submission: 20 September 2005



ลิขสิทธิ์มหาวิทยาลัยเชียงใหม่
Copyright © by Chiang Mai University
All rights reserved