



## Animal Care and Use Protocol Aquatic Animals Study Protocol

(กรอกข้อมูลเป็นภาษาไทย หรือ ภาษาอังกฤษ)

Protocol number		This section will be filled by the BUU-IACUC
Date of submission (dd/mm/yyyy)		
Date of approval or request revision (dd/mm/yyyy)		
Date of resubmission (dd/mm/yyyy)		
Date of approval or disapproval (dd/mm/yyyy)		
Date of expiration (dd/mm/yyyy)		

### 1. Protocol title:

(Thai) .....

(English) .....

*If this protocol is a part of the main project, please provide the main project title*

(Thai) .....

(English) .....

### Principal investigator of the main project

Name ..... Affiliation .....

Phone..... E-mail.....

Animal use license no ..... Expired date .....

### 2. Principal investigator of the submitted protocol: *For a student thesis, the principal investigator is the principal adviser and the student is a co-investigator.*

Name ..... Affiliation .....

Phone..... E-mail.....

Animal use license no ..... Expired date .....

### 3. Co-Investigators:

**3.1** Name..... Affiliation .....

Phone..... E-mail.....

Animal use license no ..... Expired date .....

**3.2** Name..... Affiliation .....

Phone..... E-mail.....

Animal use license no ..... Expired date .....

**3.3** Name..... Affiliation .....  
 Phone..... E-mail.....  
 Animal use license no ..... Expired date .....

**4. Contact person in case of emergency**

Name ..... Affiliation .....  
 Phone..... E-mail.....

**5. Type of animal protocol:**

- ☐ Research: in the field of .....
- ☐ Testing/Monitoring, specify .....
- ☐ Teaching: course title/ level .....
- ☐ Biological production, specify .....
- ☐ Animal breeding, specify .....
- ☐ Other, specify .....

**6. Anticipated protocol period:** From ..... to.....  
 (วันเริ่มต้นต้องไม่ก่อนวันที่โครงการได้รับการรับรอง)

**7. Grant proposal:** Please select **ONE** of the following options

- ☐ To be submitted; from.....  
 Amount requested .....  
 Funding period from .....to .....
- ☐ Has been submitted; from.....  
 Amount requested .....  
 Funding period from .....to .....
- ☐ Received from .....  
 Amount requested .....  
 Funding period from .....to .....
- ☐ Other .....

Your signature(s) as Principal investigator (PI) and Co-investigator(s) on this application verify that the information herein is true and correct and that you are familiar with and will comply with the standard of animal care and use established under the ethical guidelines and policies of Burapha University (BUU) and Office of the National Research Council of Thailand (NRCT).

**Principal investigator:** Name .....

\_\_\_\_\_  
(Signature)

\_\_\_\_\_  
(Date)

**Co-investigator:** Name .....

\_\_\_\_\_  
(Signature)

\_\_\_\_\_  
(Date)

**Co-investigator:** Name .....

\_\_\_\_\_  
(Signature)

\_\_\_\_\_  
(Date)

**Co-investigator:** Name .....

\_\_\_\_\_  
(Signature)

\_\_\_\_\_  
(Date)

\*\*\*\*\*

**Approval:**

**BUU-IACUC review:**

☐ Approved

☐ Disapproved

\_\_\_\_\_  
(Chair, BUU-IACUC)

\_\_\_\_\_  
(Date)

**Burapha University**  
**Standardized Research Protocol Format**  
**for Permission of Animal Care and Use**

**1. Protocol title:**

(Thai) .....

(English) .....

**2. Non-technical summary:** *Provide a brief description of the project that is easily understood by non-scientists, expressing its significance and your reasons for undertaking the study.*

.....

.....

**3. Rationale and literature review:** *Include a brief statement of the requirement for the information being sought. Typically, the literature or the experience that led to the proposal will be briefly reviewed, references cited will be provided.*

.....

.....

**4. Objective(s)/hypothesis:** *Provide goal/specific aim of this project.*

.....

.....

**5. Potential benefits of the study:** *Explain how the study is important to human or animal health and the advancement of knowledge*

.....

.....

**6. Experimental design and general procedures:** *Provide a complete description of animal use, including animal species, study groups, and what will be done to the animals. Succinctly outline the formal scientific plan and direction for experimentation. A diagram or chart may be helpful to explain complex design.*

.....

.....

.....

**7. Animal used and justification:****7.1 Description of animals**

Common name	Genus and Species	Strain	Age	Weight	Sex	Number
White Seabass (ปลากะพงขาว)	<i>Lates calcarifer</i>	NA	3 m	500 g	NA	50
		<b>Example</b>				

7.1.1 Special consideration: *List specialized requirements for the research animals.*  
(If any)

.....

7.1.2 Source/ Vendor

☐ Nature: *Perform without contravention to law and careful execution.*  
*Recognizing the health of animals, endangered species and*  
*Ecosystems.*

☐ Laboratory animals: *Specify the source with genetic quality and health*  
*certificates.*

.....

☐ Commercial source, specify .....

☐ Other, specify .....

7.1.3 Transportation .....

.....

.....

7.1.4 Prevention of injury and/or infection during transportation

.....

.....

7.1.5 Quarantine

☐ No

☐ Yes, specify the method, location and duration

.....

.....

**7.2 Scientific justification for animal species; number requested; and data analysis.**

7.2.1 Animal model and species justification: *Provide a scientific justification for the*  
*choice of animal model(s). What physiological and morphological characteristics*  
*does this animal possess that make it the best possible model?*

.....

.....

7.2.2 Number of animals required: *Provide an explanation of how the numbers of*  
*animals to be used in each group or total were appropriate. Number of animals*  
*used in the experiment should be based on scientific and statistical requirements*  
*to achieve objectives.*

.....

.....

- 7.2.3 Data analysis/statistical method: *List the statistical test(s) planned or describe the strategy intended to evaluate the data.*
- .....
- .....

## 8. Animal care

### 8.1 Study location: *Study area where the animals will be housed.*

.....

.....

### 8.2 Aquatic housing system

- ☐ Open system
- ☐ Closed system
- ☐ Semi-open system
- ☐ Other e.g. sheltered, outdoor or naturalistic system, specify.....

### 8.3 Macroenvironment (Animal holding room/space)

#### 8.3.1 Temperature

- ☐ Ambient
- ☐ Other, specify (°C).....

#### 8.3.2 Humidity, specify (%).....

#### 8.3.3 Ventilation system, specify.....

#### 8.3.4 Illumination

##### 8.3.4.1 Light source

- ☐ Natural
- ☐ Fluorescent/ LED ..... Lux
- ☐ Other, specify ..... Intensity ..... Lux

##### 8.3.4.2 Photoperiod .....

#### 8.3.5 Noise and vibration control

- ☐ No
- ☐ Yes, describe .....

### 8.4 Microenvironment: *i.e., water that directly contacts with the animals*

#### 8.4.1 Water system

- ☐ Recirculation system
- ☐ Flow-through/single-pass system
- ☐ Static system
- ☐ Other, specify .....

#### 8.4.2 Type of water

- ☐ Freshwater
- ☐ Seawater
- ☐ Brackish water

#### 8.4.3 Source of water .....

## 8.4.4 Water quality treatment and control

☐ Water pre-treatment and chemical removal

☐ No

☐ Yes, please specify (chemicals/ozone/UV/etc.) .....

☐ Water quality control

☐ Parameters, specify.....

☐ Salinity (ppt), specify .....

☐ Frequency of water quality testing, specify .....

☐ Water changing schedule: ..... days; changing .....%

## 8.4.5 Life support system

☐ No

☐ Yes, specify

☐ using life support system or filter system

☐ Other, specify .....

## 8.4.6 Water temperature control

☐ Ambient temperature

☐ Will be set at the range ..... to ..... °C

## 8.4.7 Behavioral management

☐ No

☐ Yes, environmental enrichment will be provided to elicit

Appropriate behaviors

## 8.4.8 Social management

☐ Single housing because .....

☐ Social housing, number of animals per tank .....

8.4.9 Sanitation: *describe the materials and methods used at the animal housing facility.*

.....

.....

**8.5 Food**

## 8.5.1 Type of food

☐ Commercial feed

☐ Other, specify .....

## 8.5.2 Feeding schedule, specify .....

**8.6 Aquatic animal tank/pool**

8.6.1 Size ..... Volume.....

8.6.2 Material .....

8.6.3 Stocking density (Number of animals per liter/ton).....

**8.7 Substrate**

☐ No

☐ Yes, specify .....

**9. Health monitoring:** *Describe the criteria used for health evaluation while the animals are on study.*

.....

.....

**10. Animal welfare:**

**10.1 Literature search for duplication:** *This search must be performed to prevent unnecessary duplication of previous experiments.*

10.1.1 Database(s) searched .....

10.1.2 Date of search: *must be within six months prior to IACUC meeting*  
dd/mm/yyyy .....

10.1.3 Period of information searched: *range of years searched*  
.....

10.1.4 Keywords used in search .....

.....

10.1.5 Results of search: *provide a narrative description of the results of the literature search.*

☐ No .....

☐ Yes, explain why it is scientifically necessary to duplicate any previous work?  
.....

**10.2 Briefly describe how you have considered each of the following alternatives (the 3Rs) or why they are not applicable.**

10.2.1 Replacement of animals: *e.g., with in vitro models, computer models or less sentient animals.*  
.....  
.....

10.2.2 Reduction in the number of animals: *e.g., using appropriate statistical methods in the design and analysis of the study; reduction in experimental variability by using animals of defined genetic or microbiological status; sharing tissue among investigators.*  
.....  
.....

10.2.3 Refinement of experimental procedures to minimize pain or Distress: *e.g. early endpoints; use of analgesics, anesthetics or techniques that reduce stress in the animal.*  
.....  
.....

**10.3 Anesthesia:**

☐ No

☐ Yes, select and describe or specify the followings

Type of anesthesia

☐ Non-chemical method, *describe* .....

☐ Chemical method, *specify the followings*

a) Name of anesthesia used .....

b) Dosage .....

c) Route of administration .....

d) Stage of anesthesia .....



**11. Surgery : Does this Protocol involve survival surgery ?**

☐ No                      ☐ Yes

If yes, please answer the following

**11.1 Type of surgical procedures, check all that apply**

Procedure: ☐ Underwater,                      ☐ Out of water

Technique: ☐ Non-recirculating, ☐ Re-circulating

**11.2 Location:** Give room number for conducting the proposed procedures

.....  
 .....

**11.3 Surgeon/Qualification:** Indicate who will perform the surgery, and his/her qualifications, training, or experience in the proposed procedure.

.....  
 .....

**11.4 Surgical procedure:** Describe in detail any surgical procedures planned and/ or add a Reference

.....  
 .....

**11.5 Pre- and Post-operative provisions:** Detail the provisions for both pre-and post-operative care, including frequency of monitoring, supportive care, analgesia, and wound care.

.....  
 .....

**11.6 Describe long-term care of any chronic survival procedures:** .....

.....  
 .....

**12. Blood, body fluid, tissue and organ collection.**

Are the animal survived during blood/ body fluid collection?

☐ No                      ☐ Yes, please provide information in the table below

Procedures	Anatomic location	Needle size/ catheter size and length	Volume collected (ml)	Frequency (per day or week)
Blood withdrawal				
Body fluid withdrawal				
Tissue/ organ				
Other please describe e.g. mucous				

**13. Animal Restraint:**

☐ No    ☐ Yes

If yes, describe device, duration of restraint, frequency of observation, conditioning procedures and steps to assure comfort and well-being.

.....  
 .....

If prolonged restraint is used, must provide justification

.....  
 .....

#### 14. Food and water deprivation, or dietary manipulation:

##### 14.1 Does this protocol involves food and water deprivation, or dietary manipulation?

☐ No ☐ Yes

If **yes**, describe methods for assessing conditions, pain, discomfort, stress, and distress during the course of study. Include clinical signs and symptoms expected.

.....  
 .....

##### 14.2 Provide detail of these procedures in Table below

	Amount restricted/ added	Duration	Compound supplemented	Compound deleted	Frequency
Food restriction					
Nutrient alterations					
Other					

#### 15. Tumor and disease models, toxicity testing:

☐ No ☐ Yes

If **yes**, describe methods for assessing physical conditions, pain, discomfort, stress, and distress during the course of study. Including clinical signs or manifestations expected from the procedure.

.....  
 .....

What criteria will be used to determine a humane endpoint before severe morbidity and death ?

.....  
 .....

#### 16. Behavioral studies:

☐ No ☐ Yes

If **yes**, describe

##### 16.1 Types of behavioral manipulation

.....  
 .....

##### 16.2 The protocol involving the use of testing apparatus or aversive stimulus and detail of duration and frequency of the testing periods.

.....  
 .....

**17. Endpoints:** *Specific humane endpoints must be clearly defined in all animal protocols.*

**17.1 Study/ experimental endpoint:** *Describe the endpoint for the animals in this protocol when the scientific aims and objectives have been reached.*

.....

.....

**17.2 Humane (Early) endpoint is used:** *The animals are humanely euthanized prior to the expected date of study termination.*

☐ No                      ☐ Yes

If **yes**, please specify early endpoint **criteria** used are .....

.....

**17.3 Death or moribund as an endpoint is used:**

☐ No

☐ Yes, answer the following

17.3.1 Criteria that establish when the endpoint has been reached, and describe how animals will be monitored and care for

.....

.....

17.3.2 Identification of personnel responsible for evaluating animal condition, record keeping, and notification of the investigator and/or veterinarian to perform euthanasia.

.....

.....

**18. Euthanasia/ Disposition of animals:**

☐ **Euthanasia, please select method**

☐ Chemical

- Substance and dose used for euthanasia .....
- Route of administration .....

☐ Mechanical, specify .....

☐ Other, specify .....

☐ **Transfer**

☐ Transfer to another research project

- Protocol number .....
- PI .....

☐ Transfer to a slaughter house, specify .....

☐ Transfer to a teaching course .....

☐ **Other**, specify .....

**19. Necropsy:** If animals are to be necropsy:

☐ No                      ☐ Yes

• Location

.....

• Who will do it, and what is their experience in the technique to be used?

.....

- Personnel protective equipment (PPE)
- .....

**20. Animal tissue and carcasses disposal:** *Describe method used to dispose animal tissue and carcasses.*

.....

.....

**21. Biohazard/Safety:** Is the protocol involved any biohazardous agents?

- ☐ No                      ☐ Yes

If yes, specify .....

- ☐ Infectious agent(s) is (are) used : .....

Please provide the certificate of biosafety approval

- ☐ Biohazardous chemical(s), carcinogen(s) or radioactive(s) material is (are) used, specify .....

**21.1 Provide a list of any potential biohazards associated with this proposal:**

*Specify Biosafety Level (1 or 2). Please see biosafety guidelines (page 107 - 174) to detail. (<http://research.buu.ac.th/web2015/file/Guideline.pdf>)*

- ☐ Biosafety level 1 .....

- ☐ Biosafety level 2 .....

**21.2 Explain any safety precautions or programs designed to protect personnel from biohazards and any surveillance procedures in place to monitor potential exposures.**

.....

.....

**21.3 Explain how the waste is decontaminated and disposed.**

.....

.....

**21.4 List primary safety equipment and personnel protective equipment requirements.**

.....

.....

**21.5 List procedures if accident, injury or illness occurs.**

.....

.....

**21.6 List specific treatment provision for accidental exposure.**

.....

.....

**21.7 List relevant occupational medical health provision.**

.....

.....

**22. Qualifications of personnel:** *List all individuals who will be involved in this protocol. If personnel do not have experience, state how they will be trained.*

Name/Status	Qualification/Recent trainings in the use and care of laboratory animals	Responsibilities in the project

**23. List of References used in writing protocol.**

.....  
 .....

**24. Assurances:** As the PI on this protocol, I verify that the information herein is true and correct and that I am familiar with and will comply with standard of animal care and use established under the ethical guidelines and policies of Burapha University; Office of the National Research Council of Thailand (NRCT) and Animal for Scientific Purposes Act B.E.2558 (2015). Additionally, I acknowledge my responsibilities and provide assurances for the followings:

**24.1 Animal use:** The animals authorized for use in this protocol will be used only in the activities and in the manner described herein, unless a modification is specifically approved by the BUU-IACUC prior to its implementation.

**24.2 Duplication of effort:** I have made every effort to ensure that this protocol is not an unnecessary duplication of previous experiments.

**24.3 Statistical assurance:** I assure that I have consulted with a qualified individual who evaluated the experimental design with respect to the statistical analysis, and that the minimum number of animals needed for scientific validity will be used.

**24.4 Biohazard/safety:** I have taken into consideration and made the proper coordination regarding all applicable rules and regulations concerning radiation protection, biosafety, recombinant issues, and so forth, in the preparation of this protocol.

**24.5 Training:** I verify that the personnel performing the animal procedures/ manipulations described in this protocol are technically competent and have been properly trained to ensure that no unnecessary pain or distress will be caused to the animals as a result of the procedures/manipulations.

**24.6 Responsibility:** I acknowledge the inherent moral, ethical and administrative obligations associated with the performance of this animal use protocol, and I assure that all individuals associated with this project will demonstrate a concern for the health, comfort, welfare, and well-being of the research animals. Additionally, I pledge to conduct this study in the responsibility for implementing animal use alternatives where feasible, and conducting humane and lawful research.

**24.7 Scientific review:** This proposed animal use protocol has received appropriate peer scientific review and is consistent with good scientific research practice.

**24.8 Painful procedures:** A signature for this assurance is required by the Principal Investigator if the research being conducted has the potential to cause more than momentary or slight pain or distress even if an anesthetic or analgesic is used to relieve the pain and/or distress.

I am not conducting biomedical experiments, which may potentially cause more than momentary or slight pain or distress to animals.

**24.9 Research studies:** The BUU-IACUC will be notified of any changes in the proposed project, or personnel, relative to this application. I will not proceed with animal experiment until approval by the BUU-IACUC is granted.

Signature .....

(Principal investigator)

Date .....