2.0 Research Activities in 2008

Research in AKUATROP is progressing very well in 2008. Academic staffs, science and fishery officers were running experiments in their respective field in Aquaculture. Apart from pursuing Research grant, the research is funded by AKUATROP. The research projects conducted by AKUATROP are as follow:

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<th>Researchers</th>
<th>Project Title</th>
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| Prof. Dr. Faizah Shaharom           | 1. Parasitic Crustaceans on Marine Cultured Fish: Taxonomy and Life Cycle *Caligus sp.* from Cultured Seabass (*Lates calcarifer* - FRGS)  
2. Trichodinids (Protozoa: Ciliophora: Peritrichida) from *Cyprinus carpio* (Koi) at Sungai Siput, Perak |
| Assoc. Prof. Dr. Anuar Hasan        | 1. Hybridization between *Pangasius pangasius* and *Pangasius hypophthalmus*. - e-Science  
2. Identification and basic mechanism of useful probiotic bacteria having the key role to boost the growth of slow growing groupers (*Epinephelus spp.*) - FRGS |
| Assoc. Prof. Dr. Abol Munafii       | 1. Sexual Differentiation and Reproductive Aspects of Tomato Clownfish (*Amphiprion frenatus*).  
2. Study on the mechanism of sex determination in protandrous Anemone fish.  
4. Effect of feeding on broodstock maturation and egg quality of *Tor tambroides* in captivity.  
5. Breeding of Marble Goby (*Oxyeleotris marmoratus*)  
| Dr. Zaleha Kassim                  | 1. Mass Production of Live food and Feeding of *Epinephelus spp.* (Grouper) - ABI MOSTI.  
Development of Cyst from Marine Harpacticoid Copepods - NOD MOSTI |
| Dr. Mhd Ikhwanuddin @ Polity Abdullah | 1. Preliminary study of *Macrobrachium rosenbergii* larval stages-development in different temperature range.  
3. Identification and Selection of Probiotic Bacteria from Digestive Tract for the development of Blue Swimming Crab (*Portunus pelagicus*) Seed Production Technology.  
5. Parasite of Blue Swimming Crab (*Portunus pelagicus*), Mud Crab (*Scylla olivacea*) dan Crucifix Crab (*Charybdis feriatus*).  
6. The Fishery of *Portunus pelagicus* and Species Diversity of Portunid Crabs along the South China Sea.  
7. The Occurrence, Distribution and Attachment of Pedunculate Barnacle on the gills of Mud Crab (*Scylla olivacea*) and Blue Swimming Crab (*Portunus pelagicus*) |
| Mr. Liew Hon Jung (Co- Researcher)  | 1. A study on the chemical communication on the reproduction of Tropical Donkey-ear Abalone (*Haliotis asinina*) |
### 2.1 Research Activities Conducted By Non-Academic Staffs

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<td>Parasite of Blue Swimming Crab (<em>Portunus pelagicus</em>), Mud Crab (<em>Scylla olivacea</em>) and Crucifix Crab (<em>Charybdis feriatus</em>)</td>
<td>Mohd Ihwan (Science Officer)</td>
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<td>3</td>
<td>Parasitic Crustaceans on Marine Cultured Fish: In vitro Culture of Parasitic Crustacean for Identification of the Parasites Infection, Lifecycle Studies and Development of Vaccines</td>
<td>Suhairi (Science Officer)</td>
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<tr>
<td>4</td>
<td>The Occurrence, Distribution and Attachment of Pedunculate Barnacle on the gills of Mud Crab (<em>Scylla olivacea</em>) and Blue Swimming Crab (<em>Portunus pelagicus</em>)</td>
<td>Mohd Ihwan (Science Officer)</td>
</tr>
<tr>
<td>5</td>
<td>Identification and Selection of Probiotic Bacteria from Digestive Tract for the development of Blue Swimming Crab (<em>Portunus pelagicus</em>) Seed Production Technology</td>
<td>Mohd Ihwan (Science Officer)</td>
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<td>Genetic diversity in two populations’ blue swimming crab <em>Portunus pelagicus</em> assayed by random amplified polymorphic DNA markers- preliminary assessment</td>
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<td>Optimization of survival in artificial production of juvenile blue swimming crab, <em>Portunus pelagicus</em> (Linnaeus, 1766).</td>
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<td>Wahidah (Asst Science Officer)</td>
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<td>19</td>
<td>Development and maintenance of hatchery system in AKUATROP</td>
<td>Zulkarami (Asst Science Officer)</td>
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2.2 AKUATROP Focus Research

2.2.1 Aquaponic

A community project based on aquaponic system has been set-up at Kg. Telaga Batin, Seberang Takir, Kuala Terengganu. This project is a joint-venture project between Department of Fisheries Terengganu, Department of Agriculture Terengganu, Universiti Malaysia Terengganu and Kumpulan Rakyat Termiskin (KRT) Kg. Telaga Batin. The system was set up by WHK Integrated.

Organic food which was produced from aquaponic system is fresh and free from pesticide. Catfish can be harvested after 4 months, lettuce and chinese mustard plant can be harvested after 1 month and can achieved 3 times harvesting for one cycle period (4 months).

This project has become one of the attractions of this place. According to the Chairman of the KRT Telaga Batin, Tuan Haji Che Husin Che Muda, in the year 2008, there were numerous KRT visitors from various places such as KRT Gajah Mati, Kelantan, KRT Sarawak, KRT Rasah, Negeri Sembilan, KRT Cheras and KRT Keramat. Feedback from the visitors show that they are very interested with the system because it is able to produce two products at the same time. Department of Agriculture Sarawak and East Coast Economic Region (ECER) committee have also visited this project. Both of these agencies are interested to set-up this system at their respective places. Aquaponic system also manages to capture the attention of ECER committee.
and has been identified as one of the project in ECER. The discussion to set-up this system at Rantau Manis Kelantan is still in progress.

This project also manages to capture the attention of the Ministry of Agriculture and Agro-Based Industry. On 20th December 2008, the minister has come to visit this place. In his speech, he encourages the community and the participant to work hard to ensure this project achieved its objective. This project has the potential to be one of the ecotourism attractions for the Kg. Telaga Batin.

2.2.2 AKUATROP Crab Projects

Research on *P. pelagicus* has been conducted intensively by a few researchers lead by Dr. Ihkwanuddin @ Polity Abdullah. Mhd. Dr. Ihkwanuddin formerly a staff at Department of Fisheries, Sarawak. He has been working on Portunid crabs; blue swimming crab (*P. pelagicus*), mud-crab (*Scylla olivacea*) and crucifix crab (*Charybdis feriatus*) for over 15 years. His expertise was then utilized by AKUATROP by supervising various aspects of Portunid crabs such as seed production technology, species diversity and parasite occurrence. Among the researches conducted are optimization of survival in artificial production of juvenile crab of blue swimming crab (*P. pelagicus*), identification and selection of probiotic bacteria from digestive tract for the development of blue swimming crab (*P. pelagicus*) seed production technology, parasite of blue swimming crab (*P. pelagicus*), mud crab (*S. olivacea*) and crucifix crab (*C. feriatus*). The Fishery of *P. pelagicus* and species diversity of portunid crabs along the South China Sea (SEA) and the occurrence, distribution and attachment of pedunculate barnacle on the gills of mud crab (*S. Olivacea*) and blue swimming crab (*P. pelagicus*).

Apart from that, research on genetic variation of *P. pelagicus* has been carried out by AKUATROP lecturer, Mrs Norainy Mohd. Husin to determine genetic diversity among different populations. Potential populations for this study are Kedah, Perak, Selangor, Johor, Terengganu, Kelantan, Sarawak and Sabah. Two DNA markers namely Random Amplified Polymorphic DNA (RAPD) and mitochondrial DNA were chosen. The information obtained can be applied to the construction of a genetic-based stock enhancement program and to avoid including inbred founder populations in breeding programs of *P. pelagicus*. 
Tagged males of blue swimming crab (*Portunus pelagicus*).

Juveniles of blue swimming crab (*Portunus pelagicus*).

### 2.2.3 Kelah Breeding for Conservation

*Tor tambroides* is a tropical indigenous species with wide commercial value particularly for human food, recreational, ornamental as well as conservation (Nyugen et al., 2005). These fishes are known to prefer clear, swift-flowing water with stony, pebbly or rocky bottoms, but these habitats have been significantly degraded by dramatic changes associated with agricultural and industrial development, including deforestation, irrigation, dam construction and river management work, as well as over fishing (Ingram et al., 2005).

Due to high market demand of *Tor tambroides* at different stage from juvenile to adult, this fish is facing serious population decline and extinction (Ng, 2004). Again, recent catch statistics indicated that this fish was considered as endangered species (Jalal et al., 2005). Without proper management and knowledge of these high value species, they will face a serious decline
population and extinction. Intensive scientific research on these species should be carried out to conserve and increase their population.

As one of the 21 centers of excellence in the world, AKUATROP has taken a step forward in carrying out a few research to manage and conserved species from extinct in our eco-system and to have better management for aquaculture. AKUATROP in conjunction with kelah World and Department of Fisheries Malaysia has agreed to run a number of researches on this species. Currently, one international student; Endryeni Mulyadi from Padang, Sumatera is carrying out her research on induced breeding of Kelah, *Tor tambroides* (Bleeker, 1854) and larval development. Up coming research for kelah are broodstock management interm of particularly on cultured system, water quality and nutrition. Research on larval management covers a few topics like nutrition, water quality management, and larval handling. These projects are carried out by postgraduate and undergraduate students under Assoc. Prof. Dr. Abol Munafi Ambok Bolong.

AKUATROP Kelah Hatchery is located near to the Netloft complete with cement broodstock tanks, backup generator system, two high capacity chiller system, sand filters, UV light, 24 hours aeration supply, recirculating water system and raceways for larvae.

![Kelah (Tor tambroides) broodstock.](image-url)
Kelah juveniles which are successfully bred using induce breeding methods.

2.2.4 Horseshoe crab project

The horseshoe crab has descended from mud dwelling primitive arthropods called trilobites which lived in the Precambrian seas, nearly 600 million years ago. After 150 million years or so, the horseshoe crab evolved into its present shape and surprisingly archeal animal body has not shown any phenotypic change even after a span of 350 million years. The horseshoe crab belongs to the benthic community and specifically migrates on the shore from the deeper waters for the purpose of breeding.

The most diversified Paleozoic group is represented only by four species of the horseshoe crab with discernible morphology and three genera confirmed, so far, all over the world. The abundance of three species of the horseshoe crab like *Carcinoscorpius rotundicauda*, *Tachypleus gigas* and *Tachypleus tridentatus* along the coast of Malaysia is an interesting phenomenon. Largest specimen of a *T. tridentatus* (7.5 kg) was collected for the first time along the coast of Papar Sabah which is a new record.
The largest specimen of a horseshoe crab ever reported *Tachypleus tridentatus* (7.5 kg) was collected for the first time along the coast of Papar, Sabah which is a new record.

The blood of the horseshoe crab is generally known as haemolymph which is white in colour but it turns into blue when exposed to air. The blue colour of the blood is attributed to haemocyanin, a copper based molecule that carries oxygen throughout the circulatory system of the crab. The white blood cells of horseshoe crab are known as amoebocytes which are elliptical in shape with a prominent nucleus. The blood cells aggregate quickly when exposed to minute amount of endotoxins or harmful bacteria. The presence of the endotoxins in human beings is generally assessed with the help of rabbit vaccine test. Apart from the method being not very sensitive, it results in the death of a large number of rabbits. In recent times the biomedical properties of the horseshoe crab have assumed great importance due to unanticipated marketing of Limulus Amoebocyte Lysate as an endotoxin tester in food, drug and pharmaceutical industries. The lysate test has been proven to be a better substitute for the rabbit vaccine test because of its efficiency in terms of quick result and economy. The pyrogen test with the help of lysate is simple, sensitive, accurate and easy to perform.
Very little studies have been done on the biotechnological potential to date in Malaysia. There is an urgent need to carry out more comprehensive biotechnological studies on three species of horseshoe crabs found along the coast of Malaysia. As the horseshoe crab migrate regularly towards the shore for breeding purpose, it is rather prudent on our part to protect the breeding beaches from destruction and environmental degradation. The adequate measures should be adopted in protecting the valuable breeding beaches along the coast of Malaysia. Regular plantation of trees along the coast to control beach erosion, protection of sand dune vegetation on the beaches, ban on removal of sand gravel from the breeding beaches for construction purpose, deforestation of mangrove, enforcement of the technique of removal of blood by entrepreneurs without sacrificing and disturbing the normal life cycle of the horseshoe crab are the other important measures to be strictly followed by the environmentalists to protect this valuable creature from the extinction along the coast of Malaysia.

Artificial insemination of a female horseshoe crab  Artificial insemination of a male horseshoe crab

This marine creature can play a vital role in improving the economy of the people of Malaysia. Besides meeting the local demand of the horseshoe crab for research and other purposes, a considerable amount of foreign money can also be earned by exporting the final or semi final products to the companies actively involved in producing pharmaceutical products from horseshoe crab on commercial scale. This needs a dedicated team of persons of competence and dedication to chalk out a detailed proposal along with the policy makers for implementing this project for nobel cause. The team should comprise of the persons like economists, urban specialists, architects, marketing survey specialists, representation of medical specialists as well as research administrators. Not the least but the last, the active and sincere supports from the local authorities will certainly play a vital role in implementing such an important programme for the benefit of society.
2.3 Research Abstract and Summary

2.3.1 Aquatic Organism Health

**Research Title**: Parasitic Crustaceans on Marine Cultured Fish: *In vitro* Culture of Parasitic Crustacean for Identification of the Parasites Infection, Lifecycle Studies and Development of Vaccines.

**Project Leader**: Prof. Dr. Faizah Shaharoum
**Researcher**: Suhairi Mazelan

Caligidae are the most commonly reported species infecting on the body of marine or brackish cultured fishes. In line with the 9th Malaysian Plan (RMK9) which highlights aquaculture industry as the first thrust core development under agriculture sector, this study focus on the development of vaccine to prevent infestation of the parasitic crustacean for a long term mission especially to help the aquaculturists who are facing this problem. To ensure the success of this research, Institute of Tropical Aquaculture (AKUATROP); Universiti Malaysia Terengganu is collaborating with Marine Fish Production and Research Centre (PPPIL), Tanjung Demong, Terengganu and National Aquaculture Fisheries Research Institute (NAFISH), Batu Maung, Penang in undertaking this research.

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Specimen of *Caligus* sp.

**Research Title**: Parasite of Marine Ornamental Fish
**Project Leader**: Prof. Dr. Faizah Mohd Shaharoum
**Researcher**: Wahidah Wahab

This study was done by Institute of Tropical Aquaculture (AKUATROP) Aquatic Organisms Health Unit staff to determine the cause of abnormality or fatality to the cultured fish in the Universiti Malaysia Terengganu (UMT) marine hatchery. Most of the marine ornamental fish that were examined were used as broodstock for several breeding studies in AKUATROP. The ectoparasites were Monogenea, Turbellaria and *Cryptocaryon Irritans*, which were mainly found on the gills. The endoparasites which were digenea, nematode, unidentified cysts and unidentified flagellates were found in the intestine, stomach and liver. The morphological characteristics of these parasites were described and captured using 80i Eclipse Advanced Research Microscope.
Figure 1: Digital image of parasites found on external organ of Vermiculated Anglefish (*Chaetodontoplus mesoleucus*), False Clown fish (*Amphiprion ocellaris*) and Pink Skunk Clownfish (*Amphiprion perideraion*) (a) *Cryptocaryon irritans* which are found on the skin (b) Turbellaria in skin and (c) *Ichthyobodo sp* in gill

Figure 3.0: (d) Digital image of *digenea* which are found from the intestine of False Clown fish (*Amphiprion ocellaris*) (e) Unidentified cyst (f) Close up on the head part of the Nematode

**Research Title**: Identification, Prevalence, Mean Intensity and Abundance of Trichodinids (Protozoa: Ciliophora: Peritrichida from *Cyprinus Carpio* (Koi) At Sungai Siput, Perak

**Project Leader**: Prof. Dr. Faizah Shaharoum

**Researcher**: Syaidatunorhani Abdul Manan

In this study, even though *Trichodina acuta* and *Trichodina nobilis* were the highest species in station 1 and station 2, the dominant species infected *Cyprinus carpio* in this study was *Trichodina heterodentata* with the prevalence of about 95%. Therefore, it can be concluded that overall, within the diversity of Trichodinids, *Trichodina heterodentata* widely infects Koi fish in Perak.

Result shows that the asexual reproduction of family Trichodinidae was by binary fission that starts with the division of nucleus, continued by the division process of adhesive disc and the development of radial pins, which is finally followed by the process of developing new denticles starting from the central part, blade and thorns (rays) of the denticle. Old denticles will be reabsorbed to the center part of the disc and obscured.
Research Title: The Occurrence, Distribution and Attachment of Pedunculate Barnacle on the gills and body of Blue Swimming Crab (Portunus pelagicus)

Project Leader: Dr. Mhd Ikhwanuddin Abdullah
Researcher: Mohd Ihwan Zakariah

The focused of this study are to investigate the occurrence, distribution and attachment barnacle that attached on the Crab’s gill, to identify the species of barnacle and to find out the effect of barnacle attachment on the Crab’s gill.

![Figure 1: Picture shows the area of barnacle attachment in the P. pelagicus. The samples were taken from Tanjung Dawai, Kedah.](image)

Research Title: Isolation, identification and characterization of probiotic microbes to enhance the growth of brown marbled grouper, Epinephelus fuscoguttatus (Forsskal 1775)

Project Leader: Assoc. Prof. Dr. Anuar Hassan
Researcher: Mithun Sukumaran and Nurhidayah Mohd Razif

The project successfully completed 85% of the main objectives. The project was guided and run by the deputy director of the institute, Asoc. Prof. Dr. Anuar Hassan, with a Ph.D. scholar Mr. Mithun Sukumaran (India) and M.Sc. student Miss. Nurhidayah, M.R. (Malaysia). Two international conferences are being presented and one communicated (Asia-Pacific aquaculture 2009) with the findings of the project and 4 papers including 2 international papers are in progress. Two products from the project, for the ‘Malaysian innovation’ are also in progress.
2.3.2 Breeding Technology

**Research Title**: Hybridization between *Pagasianodon hypophthalmus* (Sauvage, 1878) and *Pangasius pangasius* (Hamilton, 1822)

**Project Leader**: Assoc. Prof. Dr. Anuar bin Hassan

**Researcher**: Dr. Shahreza bin Md. Sheriff and Mithun Sukumaran

The main objective of the project was focused on the cross breeding of the two important species of the fish. The growth performance studied have been completed whereas the, molecular characterization of the offspring of the parental and crossbred fishes are in progress. In the proposed project the experiments on cross breeding of the two cat fishes were successfully completed where parental and hybrid strains were produced. The molecular characterization of the project headed by Dr. Shahreza is ongoing with the studies focused on the comparison of expression of traits by the cross bred and parental strain offspring. The output of the project so far could be considered by successful completion of a master degree project done by Mr. Agus Putra Abdul Samad (Indonesia). Mr. Agus has also communicated one paper in the national journal and presented two papers in international conferences.

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**Research Title**: Sexual Dimorphism In The Morphometric Characteristics Of False Clownfish, *Amphiprion Ocellaris*

**Project Leader**: Assoc. Prof. Dr. Abol Munafi Ambok Bolong

**Researcher**: Lokman Nor Hakim Norazmi

The purpose of this study is to observe and determine morphologically the characteristics that differentiate sex in *A. ocellaris*. Since the reasons for this sexual dimorphism are still unknown, further studies are recommended on the sexual determination mechanism and differentiation process of *A. ocellaris*. The combination of both traditional morphometric parameters and Truss network measurements has helped to strongly proven the sexual dimorphism in *A. ocellaris*. The whole body morphology of *A. ocellaris* has been able to be analyzed since both methods have covered its whole body. The result of this study provides information to help breeders solve the problems in sex identification of *A. ocellaris* broodstocks thus increasing its production in captivity.

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**Research Title**: Study on the gonad structure of Pink Skunk Clownfish, *Amphiprion perideraion*

**Project Leader**: Assoc. Prof. Dr. Abol Munafi Ambok Bolong

**Researcher**: Sarmiza Saperi

Histological study was carried to describe and differentiate the gonad structure of Pink Skunk Clownfish, *Amphiprion perideraion* according to sex. 13 families of *A. perideraion* were collected from Pulau Bidong, Terengganu. Each family consists of a female, a male and two to four non-breeders. The ovotestis contains of previtellogenic oocytes and spermatocytes with the oocytes dominating the ovotestis. This result indicates that the non-breeders of *A. perideraion* might have the ability to switch their sex directly to female subjected on the surrounding factors. The ovary filled with developing ovarian tissues in all stages with vitellogenic oocytes dominating the ovary. Absence of the testicular tissues reveals that the sex inversion from male to female is an irreversible process. It can be concluded that the gonad structure of *A. perideraion* is similar and can be typically observed in other protandrous hermaphroditic fishes.
Research Title: Effects Of 17 Α-Methyltestosterone on the Juveniles of False Clownfish (Amphiprion Ocellaris)

Project Leader: Assoc. Prof. Dr. Abol Munafi Ambok Bolong
Researcher: Muhammad Abduh Yazed

False clownfish (Amphiprion ocellaris) is the most common marine ornamental species that can be found in aquaria. This bright orange coloured fish with three white stripes is famous among hobbyist because it can easily adapt and breed in captivity. However, the production of this species in captivity is still low due to limited broodstock mass. This species has the ability to change sex from male to female to meet the requirement of its social structure. Hormone treatment has been used widely to produce monosex population in fish species. In this experiment, the juveniles of A. ocellaris are treated with 17 α-methyltestosterone (MT) to investigate the effects of this androgen on the gonads of A. ocellaris juveniles at different concentration.

Research Title: Early growth performance of hybrid red tilapia under marine water Environment

Project Leader: Mr. Liew Hon Jung
Researcher: Anuar Deraman

Tilapia are economically important food fish not only for tropical region but also popular in United State and European countries. They are primarily cultured in diverse farming open water system like freshwater ponds, tanks, concrete tanks, rice paddies, cages in lakes, rivers and reservoir following either intensive or super-intensive practices. In present study our results also support that hybrid tilapia can be introduced to real marine water environment like abundance of shrimp ponds, estuaries or even sea cages.

Research Title: Mass Fry Production of Epinephelus sp. (grouper)

Researcher: Dr. Zaleha Kassim

This project involves collaboration with a few researchers from several IPTAs and Fisheries Department. UMT’s research team led by Dr. Zaleha is now in the process to scale up production of planktonic and benthic microalgal collected from Bidong Island and several coastal areas in Terengganu such as lagoon in Merchang and Setiu. There are about seven species of microalgae are now maintained in 20L carboys and many more in the isolation plates. Rotifers and copepods are also collected from wild during field sampling. The culture is maintained and prepared for mass production before feeding test experiment. To date, the team manage to isolate and up-scale several local species of microalgae from Bidong island. At the same time they consistently maintain 12millions cells/L common microalgae Chlorella sp., Isochrysis sp. and Nannochloropsis sp. in our lab.

Project Leader: Dr. Mhd. Ikhwanuddin @ Polity Bin Abdullah

Researcher: Azmie Ghazali

The crab fishery and culture operations are expected to continue to grow in the future but the crab culture operations have to depend solely on juvenile collected from the wild, which will vary in size, age and with the seasons. So it is likely that management controls on the indiscriminate collection of natural juvenile will be needed to prevent over exploitation of the wild caught juvenile. Therefore, this research aim is to improve further the hatchery protocol of the seed production technology of blue swimming crab juveniles (*P. pelagicus*) for the commercial production. Below is the flow chart of the blue swimming crab, *P. pelagicus* project:–

![Flowchart](image)

**Figure 1: Workflow for optimization of survival in artificial production of juvenile blue swimming crab, *Portunus pelagicus* (Linnaeus, 1976).**

Research Title: Preliminary study of *Macrobrachium rosenbergii* larval stages development in different temperature range.

Project Leader: Dr. Mhd Ikhwanuddin @ Polity Abdullah

Researcher: Abd Halim Safi

In Malaysia, giant freshwater prawn farming has been practiced by adopting traditional methods, where the juvenile/postlarvae were collected from the wild and reared in semi intensive farms before the first artificial seed production technique was attempted. Freshwater prawn culture has attracted more attention in the recent years due to its export potential and increasing demand as luxury protein. Prawn is a poikilothermic animal which the water temperature is probably the most important environmental factor which directly affects the survival, growth and metabolism of prawn. The main objective of this project is to determine the stages development in different temperature.
Mate the mature male and female *M. rosenbergii* in 60 L HDPE tank.

Acclimatize the berried female *M. rosenbergii* in brackishwater (5 ppt) condition for 1-2 days until the eggs hatch.

Newly hatched *M. rosenbergii* larvae.
Research Title: A Study on Chemical Communication on The Reproduction of Tropical Donkey-Ear Abalone Haliotis asinina

Project Leader: Dr. Hii Yii Siang
Researcher: Mr. Liew Hon Jung and Hua Thai Nhan

In this study we found that the reproductive H. asinina produced a chemical stimulant that attracts other individuals for spawning. This study provides the information for further success of induced spawning of the species; especially in estimation of reproduction timing and control reproduction performance.

Project Title: Ecology and Biology Of Mussel In Sea Farming Area
Project Leader: Dr. Zaleha Kassim
Researcher: Ahmad Lutfi Omar

This study investigates the recruitment and early growth of Perna viridis on rope collector hanged from mussel culture platform in Muar coastal area in The Straits of Malacca. Analysis of shell length by FiSAT II software shows that the species in the study area is over exploited.

2.3.3 Aquaculture Engineering

Research Title: Preliminary study of the wild caught grouper juvenile Epinephelus coioides from Bukit Keluang Besut, Terengganu in recirculating aquaculture system (RAS).
Project Leader: Assoc. Prof. Dr. Abol Munafi Ambok Bolong
Researcher: Abd Halim Safi

Institute of Tropical Aquaculture, AKUATROP has developed their own recirculating aquaculture system (RAS) to study the feeding and growth performance of wild caught young grouper. The preliminary result of this study is focused on growth and feeding wild-caught young grouper in RAS. The experiment was done at Saltwater Hatchery Unit, AKUATROP, Universiti Malaysia Terengganu (UMT) in Terengganu, Malaysia. The growth and feeding performance of wild-caught young grouper in RAS were conducted in 10 fiber glass tanks (1000L) filled with 700L of UV-disinfection seawater gentle aeration were provided. Epinephelus coioides which have been acclimated were randomly selected, recorded the wet weight and total length, and then placed in culture tanks. Mortality was recorded daily and dead fish were removed from the culture tanks. Wet weight and total length data were collected weekly to determine the growth of E. coioides. Grading process was conducted weekly to prevent cannibalisms. Dissolved oxygen, temperature, pH and salinity in culture tank were monitored daily. The following water quality parameters were analyzed every 10 days such ammonia, nitrite and nitrate.
Figure 1: Fiberglass tank for RAS at AKUATROP

Figure 2: Orange-spotted grouper E. coioides as a preliminary culture species

Figure 3: RAS main components system at AKUATROP

Figure 4: Cannibalism occurred while the study was conducted

Figure 5: Orange-spotted grouper length at 60 days of culture

Figure 6: Orange-spotted grouper length at 120 days of culture

Figure 7: Orange-spotted grouper length at 150 days of culture

Figure 8: Orange-spotted grouper length at 180 days of culture
2.3.4 Aquaculture Biotechnology

**Research Title**: Genetic Variability among Hatchery Reared *Epinephelus fuscoguttatus* Broodstocks

**Project Leader**: Dr. Shahreza Md. Sheriff

**Researcher**: Dr. Nur Asma Ariffin, Rosmawati Maludin, Sufian Mustafa

A study was conducted to determine the genetic variability among four populations of *Epinephelus fuscoguttatus* reared in hatchery of PPPIL, Tanjong Demong by using Random Amplified Polymorphism DNA (RAPD) analysis. A total of 15 individuals belonging to Bali, Sabah, Langkawi and Kedah populations were screened using 20 RAPD primers. Three primers (OPA1, OPA11 and OPA20) produced reproducible DNA fragments which separated all the four populations. High genetic similarity index (SI) was found among individuals in all populations indicating low intra-population genetic variability. The average SI values for Langkawi, Kedah, Sabah and Bali populations were 0.989 ± 0.01, 0.985 ± 0.01, 0.983 ± 0.02 and 0.907 ± 0.07 respectively. Phylogenetic analysis demonstrated four well-differentiated clusters according to population. UPGMA dendogram showed that Sabah population was closely related to Kedah population than Langkawi and Bali. The present study confirmed that RAPD method proved convenient to study the genetic variability of the hatchery population broodstocks. The valuable finding from this study is important for further breeding program studies in our continuing effort to increase the quality and production of this species.

**Research Title**: Role of Green Mussel in Modern Biotechnology

**Researcher**: Dr. Anil Chatterji

Marine green mussels are bivalve shellfishes possessing two shells in molluscan group. Mussels contribute substantially to the total marine fish catch of the world. Mussels are nutritious with high protein and thus they are consumed generally as raw or cooked, canned, smoked and even small quantities are frozen. Recent studies conducted at various places have shown that bivalves (green mussels) are not only an inexpensive source of proteins for human consumption but also possess some important activities having many applications in the modern biological science. Studies in countries like Japan, Netherlands, Italy and several countries showed that the extract prepared from the mussels could effectively be used to cure many viral diseases and also to prevent the spread of viral diseases to others. Recent studies conducted at various places have strongly shown that mussel’s extracts have both prophylactic and therapeutic properties and can be useful in various disease managements.

**Research Title**: Genetic diversity in two populations’ blue swimming crab *Portunus pelagicus* assayed by random amplified polymorphic DNA markers

**Project Leader**: Mrs. Norainy Mohd Husin

**Researcher**: Fara Nor Asmira Jastan

The objective of this study is to determine the genetic diversity of two populations of *P. pelagicus* in Malaysian waters using PCR-RAPD. The basic information obtained can be applied to the construction of a genetic-based stock enhancement program and to avoid including inbred founder populations in breeding programs of *P. pelagicus*. 
2.3.5 Live Feed Culture

**Research Title**: Feeding Behaviour of Tomato Anemonefish, *Amphiprion fennatus* in Captivity  
**Project Leader**: Assoc. Prof Dr. Abol Munafi Ambok Bolong  
**Researcher**: Lokman Nor Hakim Norazmi

This study focused on the gathering of some basic information on the clownfish feeding behavior in captivity. Samples were collected from Bidong Island. Feeding behavior of both sexes were observed where the fish taste the food items before they totally consume the food. All the information was gathered for the journal publications.

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**Research Title**: Feeding *Artemia franciscana* (Kellogg) larvae with bacterial heat shock protein, protects from *Vibrio campbellii* infection  
**Researcher**: Dr. Yeong Yik Sung

Among their numerous physiological effects, heat shock proteins (Hsps) are potent immunomodulators, a characteristic reflecting their potential as therapeutic agents and which led to their application in combating infection. As an example, the up-regulation of endogenous Hsp70 in the branchiopod crustacean *Artemia franciscana* (Kellogg) is concurrent with shielding against bacterial infection. To better understand this protective mechanism, gnotobiotic Artemia were fed with *Escherichia coli* treated to over-produce different prokaryotic Hsps. This was shown to increase larval resistance to experimental *Vibrio campbellii* exposure. Immunoprobing of Western blots showed that the enhanced resistance to *V. campbellii* correlated with DnaK production in *E. coli*. A definitive role for DnaK was then demonstrated by feeding Artemia larvae with transformed bacteria over-producing only this protein, although other Hsps such as DnaJ and grpE also provided tolerance against *Vibrio* infection. Feeding of bacteria synthesizing selected Hsps is therefore suggested as an alternative to antibiotic use as a means of enhancing resistance of Artemia larvae to bacterial infection, which may have potential applications in aquaculture.

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**Research Title**: Ingestion of bacteria overproducing DnaK attenuates *Vibrio* infection of *Artemia franciscana* larvae  
**Project Leader**: Dr. Yeong Yik Sung

Feeding of bacterially encapsulated heat shockproteins (Hsps) to invertebrates is a novel way to limit *Vibrio* infection. As an example, ingestion of Escherichia coli overproducing prokaryotic Hsps significantly improves survival of gnotobiologically cultured Artemia larvae upon challenge with pathogenic *Vibrio campbellii*. The relationship between Hsp accumulation and enhanced resistance to infection may involve DnaK, the prokaryotic equivalent to Hsp70, a major molecular chaperone in eukaryotic cells. In support of this proposal, heat-stressed bacterial strains LVS 2 (*Bacillus* sp.), LVS 3 (*Aeromonas hydrophila*), LVS 8 (*Vibrio* sp.), GR 8 (*Cytophaga* sp.), and GR 10 (*Roseobacter* sp.) were shown in this work to be more effective than nonheated bacteria in protecting gnotobiotic Artemia larvae against *V. campbellii* challenge. Immunoprobing of Western blots and quantification by enzymelinked immunosorbent assay revealed that the amount of DnaK in bacteria and their ability to enhance larval resistance to infection by *V. campbellii* are correlated. Although the function of DnaK is uncertain, it may improve tolerance to *V. campbellii* via immune stimulation, a possibility of significance from a fundamental perspective and also because it could be applied in aquaculture, a major method of food production.
Research Title : Taxonomy and Ultrastructure of Phytal Harpacticoid Copepods from Malay Peninsula
Project Leader : Dr. Zaleha Kassim
Researcher : Nurul Huda Ahmad Ishak

The objectives of this research were to do a taxonomic identification on the dominant species of harpacticoid copepods associated with seaweed and seagrass in Malay Peninsula; and to determine the ultrastructure of phytal harpacticoids using Scanning Electron Microscope (SEM) method.

Research Title : Development of Cyst from Marine Harpaticoid Copepods
Project Leader : Dr. Zaleha kassim

The study embark several objectives that are to determine the spatial and seasonal pattern of cyst in natural environment of Malaysian coast, to determine the environmental condition to induce cyst development in marine harpaticoid copepods and to predict the period of encystment of harpaticoid copepods under manipulated environmental condition in laboratory. We manage to determine the potential environmental cues that trigger diapauses stage in coastal harpaticoids and identification of species with positive indication of entering diapause stage is in progress.