

FRI
NEWSLETTER

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AQUATIC INVASIVE SPECIES R&D

MESSAGE FROM THE EDITOR

EDITORIAL BOARD

Dear Readers,

It is a pleasure to welcome you to the latest issue of the FRI Newsletter, Volume 24 (2021), focussing on the theme of aquatic invasive species. The FRI Newsletter is an annual magazine published by the Fisheries Research Institute, Malaysia. This issue includes updates from our FRI researchers on aquatic invasive species R&D, insights to the problems and control measures by the Department of Fisheries Malaysia.

Aquatic (water-dwelling) invasive species are non-native plants, animals, and other organisms that have evolved to live primarily in water (aquatic habitats) rather than on land (terrestrial habitats). Globally, invasive alien species (IAS) are considered to be one of the major threats to native biodiversity, with the International Union for Conservation of Nature (IUCN) citing their impacts as “immense, insidious, and usually irreversible”. They threaten the ecological stability of infested habitats, and native species therein, as those are highly sensitive to the adaptive and pervasive traits of the non-native species. Although some invasive species have little or no detectable effects, many AIS do cause damage to the environment and on human interests. Case histories of their effects are widely reported in primary literature.

It is hard to believe that the FRI Newsletter has reached its 24th year of publication. Of course, this achievement cannot be accomplished single-handedly, so I would like to thank all the contributors for their respective inputs. I look forward to continued contributions of articles, suggestions on themes and other valuable inputs for the upcoming volumes of this publication from all FRI researchers. My team is continuously striving to improve our newsletter, thus, any comments or feedback would be gratefully appreciated and I can be reached via e-mail at norhana@dof.gov.my or wannorhana@yahoo.com.

Let's keep up the good work.

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Aquatic Invasive Alien Species in Malaysia: Status and Challenges in Managing the Conflict and Complexity

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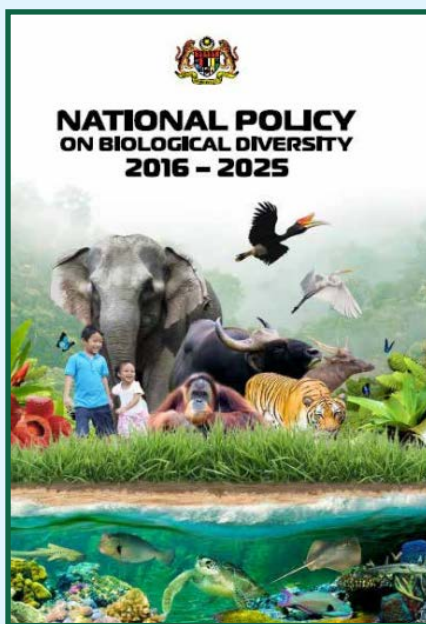
Introduction

The increasing rates of global trade, with corresponding increases in transportation of goods, services and people, largely expand the mixing of biological specimens across the world beyond national boundaries. Even though only a small percentage of transported organisms may become invasive, those which have the capacity to do so will negatively impact the economy of many countries, through losses in agricultural production and enforcement of environmental counteractive measures (Department of Agriculture (DOA), 2021)).

According to the Convention on Biological Diversity (CBD), the term 'Invasive Alien Species' (IAS) refers to "A species, subspecies or lower taxon introduced outside its natural past or present distribution, includes any part, gametes, seed, eggs or propagules of such species that might survive and subsequently reproduce" (CBD, 2002). As a signatory to the CBD, Malaysia is committed to develop and follow the Strategic Plan for Biodiversity (2011-2020) for the conservation and sustainable use of biological diversity resources, in line with the proposed 20 targets to reduce biodiversity loss (CBD, 2010). One of the targets, Aichi Target 9, focuses on identification, prioritisation, and prevention of the introduction and establishment of IAS. This is also aligned with the United Nation's Sustainable Development Goal 15.8 (SDG) - live on land.

In Malaysia, management of IAS is implemented through the National Action Plan on Invasive Alien Species 2021-2025 (NAP IAS 2021-2025) (DOA, 2016) which covers Goal 3 of the National Policy on Biological Diversity (NPBD) 2016-2025 - "We have safeguarded all our key ecosystems, species and genetic diversity", in particular Target No. 11. The NAP - IAS tag along with the three main goals which follow the NPBD Action Plans;

- 11.1 - to address public awareness on IAS;
- 11.2 - risk assessment on IAS; and
- 11.3 - strengthen quarantine inspection and enforcement.



TARGET 11

By 2025, invasive alien species and pathways are identified, priority species controlled and measures are in place to prevent their introduction and establishment.

National Policy on Biological Diversity 2016 - 2025

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Papaya die-back disease (*Erwinia papayae*) first reported in Johor in 2003
Photo by Department of Agriculture

Key indicator 11.1: By 2025, the level of awareness of the public regarding IAS has doubled compared to the 2016 level.

Key indicator 11.2: By 2018, a risk assessment framework for invasive alien species has been established.

Key indicator 11.3: By 2021, the National Action Plan for the Prevention, Eradication, Containment and Control of Invasive Alien Species has been fully implemented.

Target 11 of the National Policy on Biological Diversity, with key indicators

With the alarming numbers of introduced non-native species that may potentially become invasive, concentrated effort must be focussed on minimising or reducing the impacts and negative threats they generate to the natural environment. In Malaysia, the level of awareness on aquatic IAS and the

possible impacts has increased compared to past decades. This article provides an insight on aquatic IAS status and management in Malaysia, including the history of introduction of non-native species. We also present relevant studies by FRI on selected IAS (Red Claw, Red Swamp Crayfish and

Peacock Bass). In addition, the current IAS management practices and proposed way forward to fulfil Target 11 of the NPBD, Aichi Target 9 and SDG 15.8 are also highlighted.

The Pathway of the Introduction of Aquatic Alien Species

Among the first documentation on the alien species in Malaysia was by Ang *et al.*, (1989) which revealed the introduction of Chinese carps by Chinese immigrants in the 1800s. Later, *Tilapia mossambica* was introduced from Java to Malaya by the Japanese during World War II (Chen, 1965) and subsequently spread globally. Following that, many alien fish species were brought into the country through aquaculture activities, fisheries restocking, aquarium trade, sport fishing and for biological control (Khairul-Adha, 2013). The pathways of unintentional introductions of alien fishes in rivers, lakes and wetlands include escape or release from fish farms, use as fish bait, and disposal of unwanted pet aquarium fishes (Mohsin and Ambak, 1983; Salam and Gopinath, 2006; Chong *et al.*, 2010).

The Management of IAS in Malaysia

The National Biodiversity Council (NBC), chaired by the Deputy Prime Minister and attended by Chief Ministers of states, is the highest decision-making body for biodiversity matters and policy (NPBD) (KATS, 2019). The Ministry of Energy and Natural Resources (KeTSA) or formerly known as Ministry of Water, Land and Natural Resources (KATS) serves as the Secretariat and is also the supranational body driving the NPBD mechanism at the federal and state levels (KATS, 2019). In parallel, the National Committee on IAS has been established under the Ministry of Agriculture and Food Industries (MAFI); an inter-ministerial and inter-agency Steering Committee which serves as the main platform to coordinate the management of IAS related issues in Malaysia, including monitoring the implementation of the National Plan of Action on Invasive Alien Species (NAP IAS) 2021-2025. The three main goals of the NAP IAS are:

- i. Improve understanding and public awareness about IAS
- ii. Conduct risk assessment on all introduced exotic species before their release
- iii. Strengthen quarantine inspection and enforcement at entry points and international borders.

Under this main committee, three (3) National Technical committees were set up to address specific commodities

- i. IAS Technical Committee for Aquatic Species,
- ii. IAS Technical Committee for Agriculture and Forestry
- iii. IAS Technical Committee for Terrestrial Animal and Animal Diseases.



National Action Plan on Invasive Alien Species 2021-2025 and the goals (DOA, 2021)

The Department of Fisheries Malaysia (DoF) acts as the Secretariat for the IAS Technical Committee for Aquatic Species. The main roles of DoF are to coordinate strategies and actions for prevention, eradication, containment, and control of aquatic invasive alien species. This is done through risk management approaches along the introduction pathway of aquatic IAS into the country, starting from pre-border, border entry, the introduction of IAS either authorised and intentional, or unauthorised (whether unintentional and intentional) and lastly invasion of wild or public waters. The DoF through the IAS Technical Committee for Aquatic Species also addresses the negative impacts of aquatic IAS. The criteria for the evaluation of invasiveness/risk assessment or IAS in Fisheries and Marine species are divided into three main categories; (i) Fisheries, (ii) Marine and (iii) Diseases. A total of twenty criteria were chosen as common risk factors, with minor modifications for marine and diseases category. Scores were given to the species/pathogen selected to be listed as top IAS of concern for each categories (DOA, 2021).

The following sections highlight information on selected priority aquatic IAS in Malaysia and studies carried out by the Fisheries Research Institute. Three other IAS studies on tilapia, snowflake corals and Charru mussel are featured in the research updates section of this Newsletter.

The Red Swamp Crayfish (*Procambarus clarkii*)

The Red Swamp Crayfish originated from the coastal plain of the Gulf from the Florida panhandle to Mexico. It was reported to be the world's most successfully translocated freshwater species (Lodge *et al.*, 2012). This species has a remarkable ability to colonise a wide range of habitats due to its extraordinary ecological plasticity; resistant gene pool to change in population; biological and life cycle adaptation to environmental changes; good tolerance to a wide range of salinity, oxygen and temperature; fast growth rate and flexible feeding strategy (Alcorlo *et al.*, 2004; Gherardi, 2006; Jones *et al.*, 2009). Their burrowing behaviour causes damage to rice fields and river banks (Anastácio & Marques, 1997; Rodríguez *et al.*, 2003). Erosion from river banks increase the water turbidity and lead to cyanobacterial bloom (Yamamoto, 2010). *Procambarus clarkii* is also a carrier for many pathogens and parasites. Consumption of raw crayfish contaminated with *Vibrio mimicus* was associated with gastroenteritis (Mac Eachern, 2010) and paragonimiasis disease caused by the digenean parasite *Paragonimus* sp. (Procop, 2009). Once established, it is difficult to eliminate *P. clarkii* (Holdich, & Lowery, 1988) thus the best mitigation measure is to prevent any new introduction. In addition, a combination method of trapping and bio control using predatory fish species has been demonstrated to be successful (Gherardi *et al.*, 2011).

Procambarus clarkii was reported to be introduced in Malaysia in the year 2018 for culture trial in Federal Land Consolidation and Rehabilitation Authority (FELCRA) paddy fields in Seberang Perak. After evaluation of the risk assessment by the Committee for Incoming Alien Species Application (CIASA), DoF, further importation of this species was banned and later was suggested to be listed in the Regulation of Prohibited Species for the Import Section of the Fisheries Act (1985). Although the culture trial was stopped, elsewhere in Malaysia, this species was traded online as an ornamental species due to its attractive bright red colour, easily bred and handled. *P. clarkii* is among the crayfish species that are popularly sold with many names and varieties such as Papua Tiger (*Cherax peknyii*), Blue Pearl (*Cherax albidus*), White Snow (*P. allenii*), Red Neon (*P. clarkii* var red), and Papua rainbow (*Cherax gherardii*).



Adult Red Swamp Crayfish (*Procambarus Clarkii*)



The imported stock of Red Swamp Crayfish from China in November 2019, showing the individual broodstock with bearing eggs

The Australian Red Claw Crayfish (*Cherax Quadricarinatus*)

The Red Claw Crayfish (*C. quadricarinatus*) is a freshwater crustacean native to northern Australia and Papua New Guinea. It is among the largest decapods with a total length reaching 25 cm and weighing over 600 g (Kozák *et al.*, 2015). *C. quadricarinatus* is physically robust, with a lifespan of up to four to five years (Jones, 1990), and are tolerant to a broad range of environmental conditions. They have simple reproductive cycles and are fast growing, thus making them ideal candidates for aquaculture and the aquarium business. These characteristics also qualify them as an effective invasive species with feral communities developed in 22 countries out of 67 translocated countries/territories (Haubrock *et al.*, 2021).

Among the impacts of *C. quadricarinatus* are predation on native macroinvertebrates benthic communities (Marufu *et al.*, 2018), decline in fish populations (Douthwaite *et al.*, 2018), trophic web alteration (Romero & Jimenez, 2002; Todd, 2017) and reduce stability of river banks due to its burrowing activities (Todd & D' Andrea, 2003; Todd, 2017). Red Claw is also known to carry disease and act as an intermediate host to several pathogens including the digenean lung flukes (*Paragonimus* species), rickettsia-like parasites, *Vibrio cholera*, *V. mimicus*, *enterococci* and *Escherichia coli*. Cultured Red Claw is prone to White Spot Syndrome Virus (WSSV) (Mrugala *et al.*, 2015) and Yellow Head Virus (YHV) and also susceptible to the newly reported crustacean virus, the Decapod Iridescent Virus 1 (DIV1) (Qiu *et al.*, 2019).

The culture of Red Claw in Malaysia began in the late 1990s in Kluang Johor. At present, the species is widely spread in Malaysia with established feral populations leading the public to believe that the species is local (Naquiddin *et al.*, 2016) as in Haubrock (2021). In its home country, Australia, the Red Claw aquaculture slowly developed since the late 1980s, and

despite having all the potential characteristics, the production remained relatively low and not living up to expectations (Lawrence & Jones, 2002; Rigg *et al.*, 2020).

Haubrock *et al.*, (2021) observed that many established populations of Red Claw occur in areas with high native freshwater decapod diversity. Interestingly, this was also observed from a FRI study in Sungai Perai where Red Claw outcompete the native Giant Freshwater Prawn (*Macrobrachium rosenbergii*) and rice field crab 'ketam padi' (*Sayamia sexpunctata*) with nearly half the catch (45.36%) as indicated in Figure 1 (Mohamad-Sufiyani *et al.*, 2018).



Established population of Red Claw in Sungai Perai, Penang

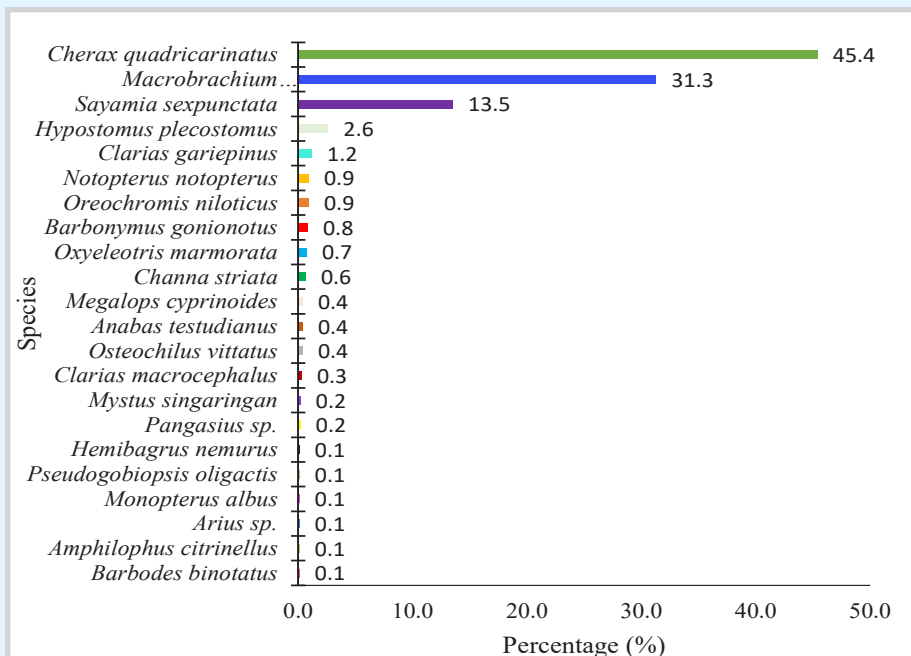


Figure 1: Percentage of aquatic species from Sungai Perai showing dominance of decapods (crustaceans)

Due to its global existence, Red Claw is being considered as an IAS and categorised as a 'major concern' for environmental impact and 'moderate' socio-economic impact. There is an urgent need for more studies on impact assessment and associated risk, and on mitigation of potential threats to biodiversity hotspots, economic and public health in relation to the Red Claw invasion.

Peacock Bass (*Chichla spp.*)

Peacock Bass is the common name for fishes of the genus *Cichla*, a large cichlids, diurnal and predatory freshwater fish from the Amazon, Orinoco basins, and Guiana Rivers in tropical South America (Kullander *et al.*, 2006; Willis *et al.*, 2012; Reis *et al.*, 2012). They were introduced worldwide as aquarium fishes and some are released in the water bodies as game fishes. Peacock Bass are known to be established in the warmer parts of North America and Asia, where they are identified as an invasive species. They may grow to a large size, are piscivorous in nature, and can damage the ecosystem due to their highly predatory behaviour and extensive feeding on smaller native fish (Pelicice *et al.*, 2008).

The introduction of Peacock Bass in Malaysia was highlighted in a popular fishing magazine, Rod & Line (Wong, 1996). An active aquarium fish breeder from Gopeng, Perak purchased 326 peacock bass fry from South America from an importer in Ipoh, a year earlier. Some of the fish died and several of them were accidentally released into a nearby mining pond. Four months later, he found the breeding fry but these were later eaten by otters. Thinking there was no survivor, he raised another aquarium fish

species, the goldfish, in the pond, but only to find them reducing in numbers. While draining the pond to check this out, to his surprise, he realised that he had unintentionally raised the first generation of Peacock Bass in Malaysia. Being an intelligent species, it has survived and is highly adaptable to many unfavourable conditions. The speckled Peacock Bass, *Cichla temensis*, can grow up to 13 kg in weight and 1 m in length and is regarded as the largest species of Cichlid. Taxonomic classification of peacock bass has validated 16 *Cichla* species, generally differing in colour pattern and range, but similar in proportions and most meristic characters. Extensive variations also cause problems in identification in some species, like *C. temensis*. Most *Cichla* species display a pattern of three wide vertical stripes on their bodies, sometimes with smaller intermediate bands with a grey, brown, yellow or green background. They are also recognised with a spot on their tail fins resembling the eyes on a peacock's tail feathers, hence the origin of their common name given as 'peacock'. Many adult species (primarily males), develop a pronounced (nuchal) hump on their foreheads shortly before and during the breeding season. Other physical traits vary greatly, depending on species, at individual level as well as at the developmental stage.



The thriving populations of Peacock Bass in Tasik Raban, Perak



The predatory, piscivorous behaviour of Peacock Bass, feeding on another smaller predator fish, (top) a young Giant Snakehead or Toman, *Channa micropeltes*, and a glassfish (bottom).

Cichla species was already listed under the Fisheries (Prohibition of Import and etc. of Fish) Regulations 1990, Amendment 2011) to be read together with the Fisheries Act 1985 (Act 317) where no import, sale, culture and keeping method in Malaysia, is allowed. Under this regulation, any introduction of a fish species under the list is prohibited and requires a special written approval from the Director-General of Fisheries Malaysia will be evaluated under CIASA. All importation of any species shall comply with rules and regulations stipulated under the Fisheries Act 1985 and MAQIS Act 2011.

It is worrying when Malaysia, as one of the economies most dependent on fisheries in the world, does not have enough information on how the aquatic IAS have impacted local biodiversity and altered the structure and functions of the inherent ecosystems. As a cross-cutting issue, the knowledge gap on the true extent of IAS implications on local species and ecosystems have been identified as due to the lack of coordinated and systematic monitoring of IAS.

Conclusion

This article provides brief information on current policy, management and practise of controlling the aquatic IAS in Malaysia. The DoF has taken full responsibility for the action and direction which have been put in place in managing the challenges arising from the conflict of interest in economic development and safeguarding the invaluable biological diversity treasure.

While we are on the right track in terms of policy and strategies, there is still insufficient technical capacity in research, quarantine, control, enforcement at border, risk assessment, effective control and eradication measures. More engagement with the public and industry on IAS issues would be beneficial.

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Intrusion of Invasive Species through Marine Litter: A Threat?

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Introduction

Loads of anthropogenic materials can be seen floating extensively on the surface of the ocean with the most evident phenomenon being the Great Pacific garbage patch in the central North Pacific Ocean. The materials entering the marine environment are usually known as marine debris and 50-80% of these are actually plastic wastes. Negative implications of plastic wastes are well established globally with numerous documentations reporting them as causes of mortality in marine mammals. Mortality among whales and sea turtles are highly associated with plastic wastes whether through ingestion, or due to plastic entanglement. Plastic wastes in the marine environment are also impacting the economy such as the fisheries sector due to ghost nets and abandoned, lost or discarded fishing gear (ALDGF). Alongside the fisheries sector, the tourism sector is also highly affected where heavily contaminated regions will be receiving negative commentary from tourists which in turn will affect tourist flow. Removal of marine debris from the environment requires a huge amount of capital and requires liaison between the government, Non-Governmental Organizations, affected sectors and even the public.

The majority of plastics wastes is not readily bio-degradable and requires hundreds to thousands of years to disintegrate. Defragmentation of plastics is another immense concern among scientists as this lead to the formation of microplastics, which can be defined as plastic with sizes less than 5 mm. Occurrence of microplastics in the world's ocean are ubiquitous with

current studies concerning their ingestion by commercial marine species. Microplastics ingestion rates among marine species are increasing and its availability in the organism's tissues which is later consumed by humans have been worrisome. Alongside the physical occurrence and possible chemical leaching in organisms ingesting microplastics, another recent threat involving plastic debris have come to light where scientists suspected marine litter, particularly plastics and microplastics, may function as vectors for invasive alien species (IAS).

Potential Invasion through Debris

The biological invasions into the marine environment have been associated with various routes such as shipping, navigational canals, aquaculture, and the aquarium trade. However, the transfer of non-native species was also speculated to have been contributed by plastic debris and other floating materials. Due to its buoyancy and persistence, marine litter becomes a potential raft for biota by attaching themselves to glass, metal and paper surfaces, and more frequently towards plastic items. According to National Oceanic and Atmospheric Administration (NOAA), there are four possibilities of invasive species intrusion through marine litter. First, the current range of a species may be extended to a particular coastline accelerated by factors such as the impacts of climate change. Heavily populated coastlines are also cause for concern since they have the ability to mount the debris loads. Second, established invasive species that was introduced from another pathway

such as shipping may use local pathways (marine debris) to expand their range even further. Third, large debris materials have the ability to carry a wide range organisms and species to new locations. Fourth, repeated arrivals of small bio-fouled debris items from a specific location may be critical in leading to multiple inoculations of the same species, overwhelming the local population and inducing genetic constraints.

Records of Intrusion

A study conducted in 2018 found a total of 94 debris items were attached with biota on surveyed beaches at Asturias, Spain with higher prevalence on plastic materials compared to non-plastic items (Photo 1). Genetic analysis was conducted on the 3,300 individuals attached on the materials and found five non-native species, and two were listed in the global invasive species database (GISD) which are *Austrominius modestus* and *Crassostrea gigas*.

Transboundary floating debris may also occur during ecosystem imbalance due to storms or tsunamis. A disastrous tsunami occurred in 2011 at East Japan that generated a massive biological rafting event. A total of 289 living Japanese coastal marine species from 16 phyla were documented at the shores of North America and Hawaii on a floating dock in 2017 (Photo 2). The unexpectedly long survival of these species is reckoned to the multilayer growth, and self-recruitment by other species through reproductive strategies which enable them to reproduce and multiply even during floating.

Identification of invasive species can also be conducted using environmental DNA (eDNA) extraction and metabarcoding with the recovery of biofilm from the litter. The extraction was conducted in the Port of Gijón where marine litter collected at the port consisted of pads, textiles, plastic bags, including expanded polystyrene (EPS) fragments. Only materials with visible biofilm were processed for eDNA extraction (Photo 3). The result discovered a total of 122 species with 3 of these recognized as exotic species. Two species that are considered as IAS in the local ecosystems were also uncovered, namely the brown

alga *Sargassum muticum* and the signal crayfish *Pacifastacus leniusculus*. Few native species were also found on the litter and are considered harmful due to their potential of forming toxic blooms.

Conclusion

The intrusion of invasive species in our waters causes great biodiversity loss and definitely poses threats to biosecurity. Marine debris may act as vectors for the species introduction with the debris being a form of habitat and a medium of transit upon arriving in a new

territory. Due to the rough surface of plastic and their durability at sea, plastic materials serve as an excellent transport mechanism for alien species as they can be transported over long distances. The evidence obtained from studies globally affirmed the negative impacts of marine litter, plastics, and microplastics as dispersal tools for invasive species. Efforts to reduce marine litter from entering the coastal and marine environments should be geared up to avoid further harmful effects towards the marine ecosystem.



Photo 1: Examples of individuals found on litter items. a) Goose barnacles attached on a PET; b) Goose barnacles on a duct tape

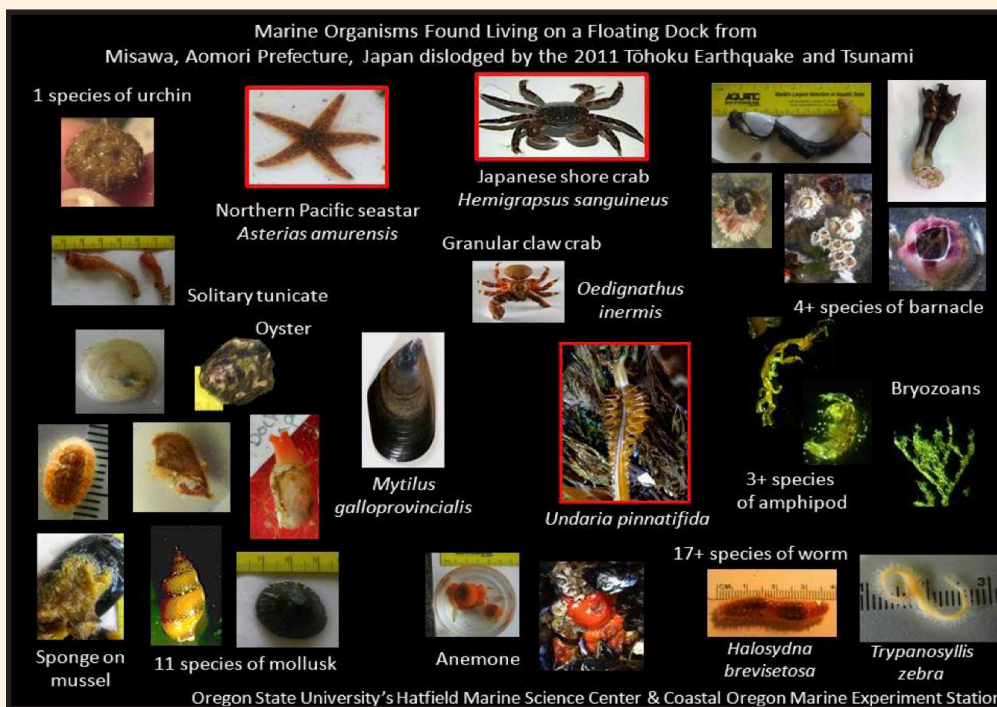


Photo 2: Different organisms found on a floating dock from the 2011 East Japan earthquake

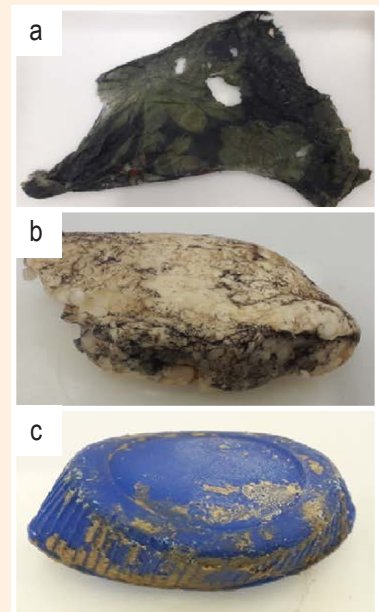


Photo 3: Marine litter samples that were scratched for eDNA extraction. a) Fragmented microplastic; b) Expanded polystyrene; c) Plastic bottle cap

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Tolerance of Flowerhorn Fish (*Cichlasoma hybrid*) to High Salinity, Low Temperature and Low Oxygen Content

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Introduction

The Flowerhorn fish, also known as Hua Luo Han, is a hybrid of hybrids. It was first developed in Malaysia in the 1990s by cross-breeding South American cichlids, mostly *Cichlasoma* spp. (Nico *et al.*, 2007), thus giving its name *Cichlasoma hybrid*. Since then, Flowerhorn became a craze among aquarium enthusiasts due to its attractiveness (Than *et al.*, 2019). Until now, the demands for this artificially created hybrid are still high as they are usually kept for “feng shui” purposes (Ng, 2016). Flowerhorn is very territorial, aggressive and hardy (National Committee on Invasive Alien Species Malaysia, 2018), and can withstand harsh water conditions which most fish species cannot tolerate. Thus, this species is considered as a high-risk species in terms of invasiveness (Saba *et al.*, 2020). According to the National Committee on Invasive Alien Species Malaysia (2018), this species is recognized as an Invasive Alien Species (IAS) in Malaysia. This unusual freshwater fish is listed in the Fisheries Regulations Amendment (2011), Fisheries Act (1985) as a prohibited species to import, sale, culture and keep in Malaysia.

At present, studies on the physio-biological aspect of this invasive species are sparse. In Malaysia itself, there is little research work being conducted on this hybrid fish. There are, however, websites and fan pages created by Flowerhorn enthusiasts to share experiences and problems among them. Due to the lack of information on Flowerhorn in the scientific literature, studies about this species is important to understand more about its characteristics, in order to determine how far this fish can tolerate changes of one or more environmental variables. Measuring the limits of alien fish tolerance to environmental parameters is vital in forecasting its eventual geographic distribution (Schofield *et al.*, 2009a). To the author’s knowledge, this is the first report on the physio-biological aspects of Flowerhorn in Malaysia. This study was conducted to determine the survivability and the tolerance limit of Flowerhorn to changes in environmental parameters, i.e. increased water salinity, decreased water

temperature and reduced oxygen content, under controlled condition.

Materials and Methods

The tolerance of Flowerhorn (*Cichlasoma* sp.) to selected lower and upper environmental conditions was investigated in the laboratory at the Fisheries Research Institute (FRI) Batu Maung, Pulau Pinang. For this study, the species chosen was the low graded adult greenish black Flowerhorn (total length of 13-15 cm) bought from a local aquarium shop (Photo 1 (top)). Its nuchal hump was not well developed and the body’s black markings were not as striking as the top graders. No consideration was taken on its gender.

Three experiments, consisting of exposing the fish to continuous increases/decreases of environmental parameters, were conducted: (1) high salinity; (2) low temperature; and (3) low oxygen. For each experiment, four 200 litres glass tanks (two treatments and two controls) were prepared and which $\frac{3}{4}$ were filled with filtered freshwater. A natural photoperiod of 14L:10D was maintained throughout the experiment. The fish were isolated in experiments on high salinity and low temperature. However, for the low oxygen experiment, up to 10 fishes were used in the treatment tank, separated by dividers. The fish were fed with commercial blood worm pellets *ad libitum* twice daily. Good water quality was maintained using biological filters and constant aeration. The water quality was monitored daily for salinity (ppt), temperature (°C) and dissolved oxygen (mg/L). Other water parameters such as ammonia (NH₄) and pH were monitored weekly. With the exception of the low oxygen experiment treatment tank, up to 75% of water was replaced twice a week, taking note that salinity and temperature were adjusted as necessary.

For experiment (1), the water salinity in the treatment tank was gradually increased by 3 ppt from 0 ppt every 3 days, by mixing the water with filtered saltwater. For experiment (2), a chiller was added to the treatment

tank. Then, the temperature in the treatment tank was continuously decreased, every 3 days, by 3°C from normal room temperature (28°C). For experiment (3), the water in the treatment tank was left without aeration and no water changes were made. The top of the treatment tank was covered to prevent natural air from entering the water body.

Flowerhorns were exposed to continuous increases or decreases of these three parameters until end-points are reached. Two end-points were determined i.e. loss of equilibrium (LOE) and death. Dead fishes were then measured and dissected to examine the level of damage to its internal organs (Photo 1 (bottom)). The observation was done under a dissecting microscope.



Photo 1: Live (top) and dead (bottom) Flowerhorn (*Cichlasoma hybrid*)

Results and Discussion

It is important to study the level of resistance of non-native fish and their ability to survive in harsh conditions, so that more can be learned about the species. Through these experiments, we may have a better understanding on how the fish cope with changes in the natural environment. Thus, it will be easier to determine the best resolution in handling or overcoming the subject matter. The fish resistance may collapse when some environment changes surpass the fish’s capacity of adjustment to deal with the changes (Brandao *et al.*, 2018). Among the significant environmental parameters influencing the fish survival are salinity, temperature and oxygen level.

Two end-points were determined i.e. loss of equilibrium (LOE) and death (Table 1). LOE was defined as the fish lying on its side and unable to correct itself (Schofield et al., 2009a). For the salinity tolerance of Flowerhorn, the LOE and death of fish were identified and confirmed at 18 ppt. For tolerance to low temperature, the treated fish started to show the LOE sign at 22°C and later death at the same temperature. For the experiment on tolerance to low oxygen, only one end-point was determined; death at 1.4 mg/l of oxygen.

Table 1: End-points of loss of equilibrium (LOE) and death for all three experiments.

Experiment	LOE	Death
High Salinity (ppt)	18	18
Low Temperature (°C)	22	22
Low Oxygen (mg/l)	-	1.4

Although some cichlids have long been known to be euryhaline or can withstand a wide range of salinity (Oldfield, 2004; Nico et al., 2007), the salinity tolerance of Flowerhorn is undefined. The salinity experiment lasted for 19 days at a salinity of 18 ppt. The fish were able to resist salinity changes from 0 ppt to 15 ppt without mortality. All fish suffered LOE after the water salinity was increased to 18 ppt, before being pronounced dead the next day. Observations showed that the fish suffered massive damage to their internal organs rendering them hard to identify. The result from this study agreed with a few previous studies. Oldfield (2014) reported that the Red Devil fish (*Amphilophus* sp.) can survive the transition of salinity up to 21 ppt, while the Jack Dempseys fish (*Cichlasoma octofasciatum*) preferred the brackish water of 10 ppt. He then concluded that generations of inbreeding, selective breeding, or hybridization may have altered the genetic composition of the fish such that the observed salinity responses were not representative of wild individuals. Another study by Sui et al., (2016) discovered that blood parrotfish, another man-made cross-

bred fish hybridized from male *Cichlasoma citrinellum* and female *C. synspilum*, can easily adapt to salinity increase. Thus, blood parrotfish can move along a salinity gradient. This study suggested that Flowerhorn could be euryhaline.

To date, the effects of temperature on Flowerhorn remain unknown. The low temperature experiment lasted for one week. At 25°C, the treated fish started to become less aggressive than the controlled fish. Exposure to 22°C resulted in the fish becoming paralyzed at the bottom of the tank (LOE). 100% mortality was observed on the next day. Observation through dissection however did not indicate any damage to the internal organs. The lower lethal temperature for this fish was concluded at 22°C. Although Flowerhorns, like most tropical fish, thrived in temperatures between 28-31°C (Than et al., 2019), a study on its sister's cichlid fish (*C. paranaense*) revealed that the fish can still survive a decreased temperature by 6°C, although this did reduce the fish's swimming activity and aggressiveness (Brandao et al., 2018). Another sister's species (*C. urophthalmus*) tolerated lower temperature and managed to survive until it reached the death end-point of 9°C (Schofield et al., 2009a).

Until now, the effects of reduced oxygen and the tolerance of hypoxia on Flowerhorn are unclear. It is imperative to quantify the tolerances of native and non-native fish to hypoxia in light of habitat changes. Incidences of hypoxia happen when dissolved oxygen levels are less than 2.0 mg/l (Schofield et al., 2009b). This experiment was conducted for 18 days. Throughout the experiment only two out of ten treated fish were dead. Dissection of the fish did not show any sign of damage. However, the dead fish showed red streaks on their abdomen indicating the tissues were damaged as a result of high ammonia (Sharpe, 2011). The level of ammonia (NH⁴) in the water was 2 mg/l. The reason for this

species to still survive in very low oxygen is not yet known and requires further studies. However, this Flowerhorn species could be tolerant to hypoxia. The current result is consistent with the findings of Schofield et al., (2009b) which reported that *C. urophthalmus* was very tolerant to hypoxia and able to endure a high level of salinity variation under hypoxic conditions.

The results revealed that this Flowerhorn species have a high tolerance level towards high salinity, low temperature and low oxygen content. Thus, this fish could possibly endure brackish water, sub-tropical waters, or hypoxic water bodies. For this species, broad physiological tolerance to environmental variables could be a key factor facilitating its establishment outside normal ranges. Despite the results revealed here, further studies are necessary on the long-term effects of individual environmental parameters such as salinity, temperature and oxygen, and the combined effects of these parameters, to this fish's behavior. Laboratory experiments such as these may be useful in risk assessment and screening of newly introduced non-native fishes.

Conclusion

The Flowerhorn *Chiclasoma* hybrid is considered to be a tolerant species and exhibits great flexibility regarding environment variables. They may survive in brackish waters, sub-tropical areas, and even water with low oxygen level. This fish must not be released into the wild due to its ability to adapt in various water conditions. As an IAS, Flowerhorn can threaten the existence of native species. These data are critical for the prediction of the invasiveness and spread of this species throughout the various aquatic habitats in Malaysia, and for the development of a national plan of action for IAS.

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Tilapia in Tasik Temenggor: Introduce or Escape?

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The Perak State Fisheries Office, Department of Fisheries (DoF) received a claim in 2020 from anglers on the abundance of large-sized tilapias in Lake or Tasik Temenggor, Banding, Perak. Tasik Temenggor is a man-made lake formed after the construction of Temenggor Dam to generate electricity in 1974. With an area of 152 km², the lake has become a source of income for the community living around it, especially in relation to inland fisheries. It is known as a breeding ground for high value fish like Kelah, Temoleh, Baung, Tengas, Loma, Tengalan, Sebarau and Lampam.

The claim from the anglers raised a concern in the DOF and the Fisheries Research Institute (FRI) since there are significant tilapias farming activities in Tasik Temenggor. Since 2007, a large scale farming operation was started in an approved Aquaculture Zone at Tasik Temenggor using the Norwegian Genomar Supreme Tilapia, an improved strain derived from the GIFT (Genetically Improved Farm Tilapia) strain. The GIFT is a product of systematic selective breeding and genetic improvement programs by the WorldFish and partners in 1988 using *Oreochromis niloticus*. The farm operator uses a polar circle HDPE cage, which is said to control and minimize the number and severity of escapee events. After all these years it is a big question mark whether the cultured tilapia have escaped from the cages and breed in the wild. The fact that tilapia consume a wide range of natural food organisms make them the preeminent predators especially towards fish larvae.

To verify the claim, the Molecular Biology Laboratory, Impact Assessment Division, FRI Batu Maung, Penang organized a tilapia sampling trip at Pusat Perikanan Darat Banding, Perak on 5th March 2020. Tilapia fin clip samples were collected from two groups; i) from Tasik Temenggor or wild (code: TB), and ii) from cages (code: TR). Genomic DNA was extracted from the fin clips using DNeasy Blood and Tissue Kit (Qiagen, Germany). Polymerase Chain Reaction for mitochondrial mtDNA Control Region (CR) was carried out by using the primer set ORMT-F: 5'-CTAACTCCCAAAGCTAGGAATTCT-3',

ORMT-R: 5'-CTTATGCAAGCGTCGATGAAA-3'. The amplified fragments with expected size of 450 bp were sequenced. All sequences were compared with the reported tilapia mtDNA CR sequences in the GenBank (NCBI) and aligned by a clustalW in MEGA X. All those sequences were analysed with three recognized species of tilapia including the *O. aureus* (blue tilapia), *O. mossambicus* (Mozambique tilapia) and *O. niloticus* (Nile tilapia) (GenBank accession number MN384756.1, KY587515.1 and AY833491.1). A neighbor-joining (NJ) tree was constructed with the Kimura Two-parameter distance model by MEGA X with *Amphilophus labiatus* (JX402377.1) as the outgroup to root the tree. It successfully separated TB (wild) and TR (cage) groups in different branches where TB individuals are grouping together with Nile tilapia and TR with blue tilapia. Based on the tree diagram (Figure 1), the Control Region gene successfully separated the tilapias species.



A: Cage's samples (TB); B: Lake's samples (TR)



Fin Clipping



TB



TR

Genetic distance for individuals from TB, TR and reference sequence was calculated as supporting data to the phylogenetic tree results. Based on Table 1, the nearest genetic distance is at a reading of 0.0030 which is between TB (01 and 03) with *O. niloticus*, followed by 0.0211 between TR (10 and 13) with *O. aureus*. Genetic distance is a measure of the genetic divergence between species or between populations within a species, whether the distance measures time from a common ancestor or degree of differentiation. The genetic distance between TR and TB gives a value of 0.1595 close to the distance shown between *O. niloticus* and *O. aureus* which are 0.1615. This indicates that TR and TB are *O. niloticus* and *O. aureus*, respectively. It supports the results produced through phylogenetic trees.

The event that we encountered in Tasik Temenggor is not an isolated case or only

happened in Malaysia. Other countries have also faced similar situations. The Mississippi river in the USA has a problem with Asian carps dominating the river system and causing serious damage to the native fish populations. In Australia, invasive trout's pose the greatest threat towards little native fish. In Lake Lanao, Philippine, accidentally introduced Thai catfish (*Clarias batrachus*) was a mistake. And the lists continue.

So, are those tilapias caught by the anglers' escapees from the cages? What do you think? Based on the results alone, we may say that the origins of the Nile tilapia caught by the anglers are not from the cages. It may have been introduced accidentally or intentionally into the lake. At present, we don't have sufficient data on distribution of Tilapias in Malaysia, neither the domesticated nor the wild type. It is high time for us to look into it.

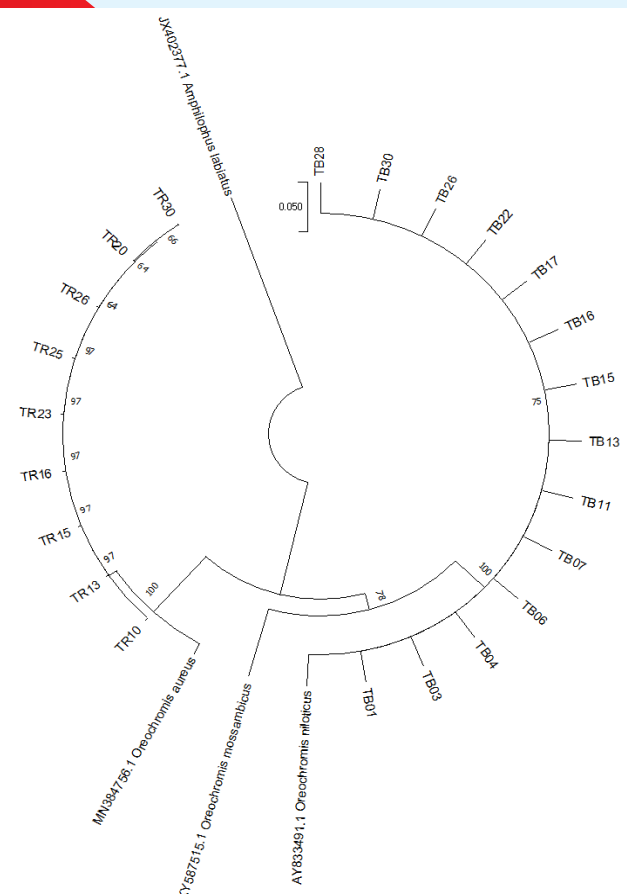


Table 1: The genetic distance between individual based on mtDNA CR sequence.

	1	2	3	4	5	6	7	8
1. TR10								
2. TR13	0.0000							
3. TB01	0.1595	0.1595						
4. TB03	0.1595	0.1595	0.0000					
5. KY587515.1_Oreochromis _mossambicus	0.1749	0.1749	0.1012	0.1012				
6. MN384756.1_ Oreochromis_aureus	0.0211	0.0211	0.1657	0.1657	0.1772			
7. AY833491.1_Oreochromis _niloticus	0.1554	0.1554	0.0030	0.0030	0.1049	0.1615		
8. JX402377.1_Amphilophus _labiatus	0.6938	0.6938	0.6748	0.6748	0.7091	0.7202	0.6816	

Figure 1: Phylogenetic tree constructed with tilapia mtDNA CR sequences using Kimura 2-parameter distance model. Bootstraps values greater than 50 are shown. *O. niloticus* (AY833491.0), *O. mossambicus* (KY587515.1), *O. aureus* (MN384756.1) and *Amphilophus labiatus* (JX402377.1) are reference tilapia species and the accession numbers were obtained from the NCBI database

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Mytella charruana: From Eastern Pacific Ocean to Tebrau Strait

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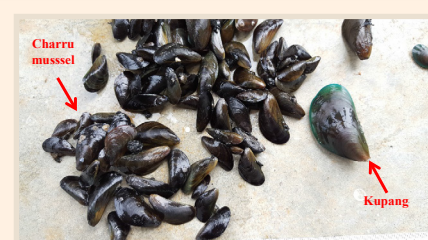
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Mytella charruana (d'Orbigny, 1846), commonly known as charru mussel, is a tropical mussel native to the eastern Pacific Ocean from Guaymas Sonora, Mexico, south to Ecuador and west to the Galapagos Islands. This mussel can reach up to 65 mm in shell length and is distinguished by its exterior dark-brown/black shell with radial patterns. The interior shell colour is iridescent purple. It is considered a euryhaline species, as it has been found from marine environments (salinity greater than 25 ppt) to estuarine (salinity of 5 ppt), and not recorded in freshwater environments (0-5 ppt). Individuals are attached to the substrate, generally as epibionts, by a fibrous tissue; *M. charruana* is a filter feeder and competes for space and other resources with other benthic organisms.

The Tebrau Strait, (also known as the Johore Strait, Straits of Johor, Selat Johor, Selat Tebrau, and Tebrau Reach), is a strait separating the state of Johor on the southern

tip of peninsular Malaysia in the north from Singapore and its islands in the south. It connects to the Straits of Malacca in the west, and the Singapore Strait in the southeast. This strait, which is a deep waterway, is also used as a shipping lane. Johor has three ports, namely the Johor Port, the Port of Tanjung Pelepas and the Tanjung Langsat Port. Apart from that, the Tebrau Strait is also used for mariculture and fisheries. With all of these factors at play, the risk of biological invasions is high.

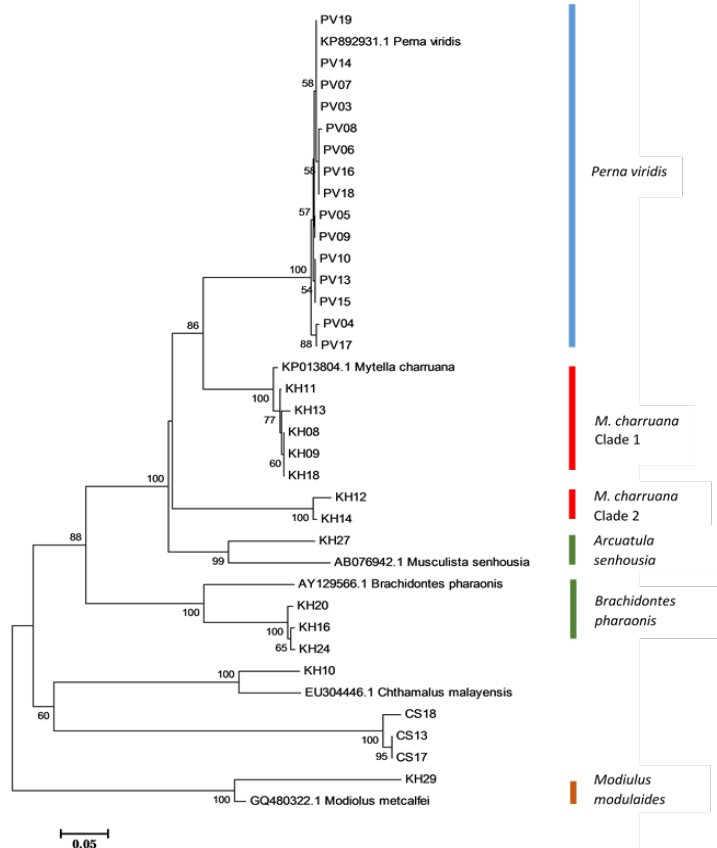
The Tebrau Strait is well known for its mussel farm (*Perna viridis* aka kupang or siput sudu). In April 2016, mussels previously identified as kupang hitam or black mussel was reported in in the eastern and western parts of the Tebrau Strait. In August 2016 it has begun to spread to the mussel's smart line farm in Teluk Jawa, Masai. It was estimated that the percentage of black mussel in the smart line system at that time was up to 50% of the total system available.



Kupang or green mussel compared to its rival, charru mussel

To confirm the species identity, we did DNA barcoding of the newly detected species aiming at the cytochrome oxidase 1 (mtCO1) gene using universal LCO and HCO primers to ascertain its identity and using Basic Local Alignment Search Tool (BLAST). There was a 100% identity match over ~510 bases to Haplotype H46 of *Mytella charruana*, with a native range on the Pacific coast of the Americas from Guaymas, Mexico to Ecuador. Introduction of this species was very likely via ballast water or through fouled ship hulls.

Phylogenetic trees (Figure 1) were



constructed using the Neighbor-Joining method from the cytochrome oxidase subunit 1 (CO1) sequences. PV is mussel (kupang) samples, while KH are samples other than mussel. Samples from GenBank (accession number) were used for comparison.

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Figure 1: Phylogenetic trees constructed using the Neighbor-Joining method.

Freshwater Stingray Species in Sungai Perak: New Record for Malaysia

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Stingrays are synonymous with marine life. Not many Malaysians are aware that some stingrays can live and breed in a freshwater environment for a lifetime without entering marine waters. In Peninsular Malaysia, freshwater stingrays have been recorded in Sungai Pahang (2004), Sungai Perak (2016), and Sungai Kelantan (2019). Apart from these main rivers, stingrays are also found in several tributaries such as Sungai Jelai (Sungai Pahang branch), Sungai Sembrong in Johor (Sungai Endau branch), and Sungai Pergau (Sungai Kelantan branch). Stingray species found in Sungai Jelai and Sungai Kelantan was confirmed as *Fluviotrygon signifer* (Compagno & Roberts, 1982). In 2019, another species, namely *Urogyrnus polylepis* (Bleeker, 1852) or locally known as 'ikan pari', was recorded in Sungai Sembrong, Johor. It is believed that many other stingray species are living in Malaysia's main rivers and tributaries especially in rural areas. Thus, there is a need to take into account the existence of these species in Malaysia. In order to achieve that, Institut

Sumber Marin Asia Tenggara (ISMAT) and Fisheries Research Institute, Kampung Acheh (FRIKA) have been working closely together with the local fishers in Sungai Perak since 2016 to study the existence of freshwater stingrays, especially on species that have not been recorded.

During this study, ISMAT collected five freshwater stingray specimens (two males and three females) from Tanjung Belanja and Kampung Teluk Kepayang in Perak Tengah District, adjacent to Sungai Perak. According to local fishers, these stingrays were caught once in a while using traps or fishing rods. An attempt was made to identify the species using morphometric (measuring and meristic (counting specimens' specific organs) methods. In addition, the x-ray images of all samples were taken to obtain evidence on cartilage layout to distinguish it from the *F. signifer* previously recorded in Sungai Kelantan, Sungai Perak and Sungai Pahang. Species identification was confirmed molecularly at the Conservation

Genetic Laboratory, ISMAT.

The results indicated that all five specimens from Sungai Perak were confirmed as *Fluviotrygon kittipongi* (Vidthayanon & Roberts, 2006) or roughback whipray and locally known as 'pari sungai'. This species is from the family Dasyatidae and was first recorded in Thailand in 2006. According to the book 'Ray of the World' by Last *et al.*, published in 2016, this species was only found in Thailand and Indonesia. Globally, this species is listed as endangered in the International Union for Conservation of Nature (IUCN). Hence, the discovery of this species in Sungai Perak is a new record for Malaysia.

The photos of *F. kittipongi* are shown below (Photo 1a and Photo 1b). This species is categorized as small-sized freshwater stingrays with a long tail and wide spread of denticles (a small, tooth-like structure; placoid scales of cartilaginous fish) in the middle part of the dorsal. The denticles are

absent in juveniles. The shape and colour pattern of *F. kittipongi* are very similar to *F. signifer* found in Sungai Pahang and Sungai Kelantan but can be distinguished by the lateral part of both pectoral fins which are greyish brown. However, for *F. signifer*, the anterior and posterior sides of the pectoral fins curve outwards like a semicircle (Photo 2 a and Photo 2b). In addition, sharper and wider denticles are visible at the top of the tail compared to *F. signifer*. The shape of the disk is oval and thin. The top of the disc's colour is bright, or orange mixed with brown, after all the mucus covering it is cleared. The length of the disk exceeds the width of the disk. The snout of *F. kittipongi* is sharp, and the bulge is very clearly visible compared to the wider snout of *F. signifer*. Disc widths for all specimens from Sungai Perak were 17 cm, 22.3 cm, 23.5 cm, 27.3 cm, and 27.9 cm, respectively. Presently, these findings show that Sungai Perak recorded at least two species of freshwater stingrays namely *F. signifer* and *F. kittipongi*.



Photo 2a: The dorsal view of *Fluvitrygon signifer*



Photo 1b: The ventral view of *Fluvitrygon kittipongi*

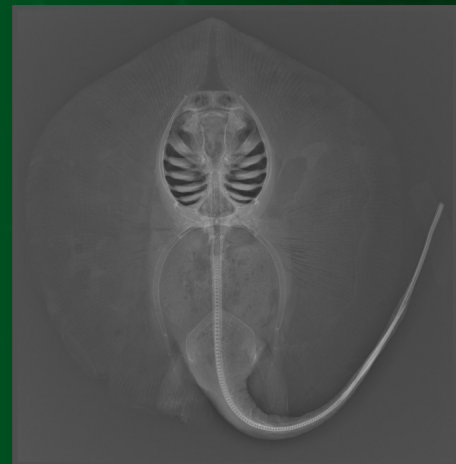


Photo 3: X-ray image of *Fluvitrygon kittipongi* specimen



Photo 4a: Upper row of teeth



Photo 4b: Lower row of teeth



Photo 1a: The dorsal view of *Fluvitrygon kittipongi*



Photo 2b: The ventral view of *Fluvitrygon signifer*

Although at present only three freshwater species have been recorded in Peninsular Malaysia, it is likely that there are other species that have been caught but not identified yet, especially in rural areas. ISMAT will continue to conduct this study in Malaysia to obtain more information on freshwater stingray species especially in remote areas. This study is vital to update the information on the biodiversity of freshwater stingrays in the country and further improve on the sustainable management of the resources.

The Invasive Snowflake Coral (*Carijoa riisei*): Threats to Malaysia's Coral Reef Community

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Snowflake coral is an octocoral (Octocorallia: Cnidaria), a soft coral that originated from tropical Western Atlantic (Bassari, 2015). This octocoral is a fast-growing species that can pose a serious threat to native coral reefs (National Committee on Invasive Alien Species Malaysia, 2018). Snowflake coral was first discovered in Pulau Payar Marine Park, Kedah in 2005 (National Committee on Invasive Alien Species Malaysia, 2018). It occupied a certain area of the reefs, usually the shaded areas at more than 10 meters

depth. The competition for space between the snowflake coral and the existing species such as *Dendronephthya* spp. (carnation corals) and *Tubastrea micrantha* (black tree coral) has led to the depletion of these native corals in those areas (Abdullah, 2015). The reason for the snowflake coral inhabiting Pulau Payar is still unknown and further studies need to be carried out to investigate its current status.

The latest coral survey in Pulau Payar was

conducted in 2020 by the National Marine Park Research Centre (PPTLN), FRI Batu Maung, Penang. Among the objectives was to determine the current status and density of snowflake coral at the Coral Garden dive site (6°05'20"N; 100°02'09"E). It was carried out using the 0.5 m x 0.5 m quadrat and transects line method. Coral garden depth profile is a mixture of slope and flat reef with a maximum depth of 20 meters during high tide. During this survey, a 50 m line transect was laid at 15 to 17 m depth, perpendicular

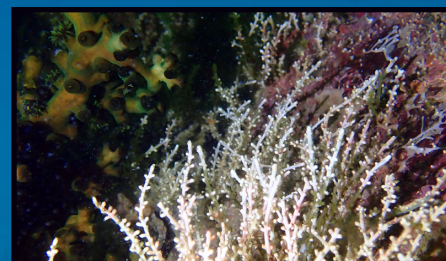
with the reef slope. The quadrat was photographed at 1 m interval along the transect line and analysed using the Coral Point Count with Excel extension (CPCe) software.

The preliminary result showed that the density of the snowflake coral at the Coral Garden was 6.11%. The distribution of this species is sparse where it's mostly concentrated at sheltered areas with low light penetration. A previous study by Normah (2016) found that the density of this coral at Coral Garden itself was 37.70%. The difference between these two studies may be due to the difference in the methods used. The previous study used the random quadrat method instead of transect line. In the random method, the quadrat was placed at selected spots compared to the transect method where it was placed along the laid transect. This could make a difference in term of density measurement. The other factor was the continuous eradication programs conducted by the Marine Park authority that have effectively hindered the growth of this invasive coral. In general, the data obtained from these studies will be useful for managing and monitoring the invasion of snowflake coral in the Pulau Payar Marine Park.

Even though this finding shows the population and distribution of snowflake coral at Pulau Payar Marine Park were less compared to the previous study, regular monitoring of this alien species is still required. Monitoring activities can alert marine parks managers to take action to reduce or eliminate threats of snowflake coral to native species. Permanent monitoring transects will give comprehensive data on population and distribution of snowflake coral. Furthermore, further research such as on DNA, reproduction, ecology and ecological impact are also important to understand the behaviour of this invasive coral species.



Snowflake coral (*Carijoa riisei*) with eight tentacles on each polyp



Carijoa riisei competing for space with the native species of *Tubastrea micrantha*

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Biological Control of the Red Claw Crayfish (*Cherax quadricarinatus*)

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The freshwater red claw crayfish (*Cherax quadricarinatus*) was globally introduced for aquaculture although it is considered as an alien species and reported to have invaded natural environments and disrupt the ecosystems. Since the species is already cultured in Malaysia, strict biosecurity measures must be implemented since red claw has been declared as an invasive species. Biological control is a known method for controlling unwanted pest population by using another organism of predatory behaviour. In this study, interspecific interactions were observed between red claw juveniles to native fish species namely the climbing perch (*Anabas testudineus*), common snakehead (*Channa striata*) and walking catfish (*Clarias batrachus*). Prey-predator interactions were recorded in three sets of tanks measuring 41 x 30 x 25 cm, using two sizes of crayfish juveniles (1-3 cm TL) and two ratios of prey-predator. Smaller crayfish juveniles (1 cm TL) were released to interact with predators in a ratio of 3:1, while for larger crayfish the ratio was 2:1.

Predatory were 100 percent for both red claw sizes with smaller juveniles killed within one hour of interaction while larger (2-3 cm TL) crayfish managed to survive for three days before they were completely killed. Further assessment will be repeated with different sizes, ratios and other species of fish.



Interspecific interactions assessment between the climbing perch, *Anabas testudineus* and the freshwater crayfish, *Cherax quadricarinatus*

Farmed Type and Non-Native Fish Species for Aquaculture: Benefits or Threats?

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Aquaculture is expanding around the world as an approach to support fish supply to the consumers. The stocks being used for aquaculture are described as 'farm type' aquatic organisms by the Food and Agriculture Organisation (FAO) (FAN, 2020). The 'Farm type' refers to a farmed aquatic organism that can be a strain, variety, hybrid, triploid, monosex group or other genetically altered form of species. The 'farm type' is categorized further into primary and secondary farmed types. The primary farmed types refer to the domesticated and improved stocks which are mass produced by aquaculture activity. They have been domesticated for decades, bred in captivity and their economic important traits have been improved through selective breeding. Most of the domesticated stocks are largely indistinguishable from their wild relatives.

However, they are not to be released into public waters to ensure the authenticity of the original stock is preserved. It has been reported that similar genetic makeup of Atlantic salmon populations across 13 Swedish rivers was retrieved as a consequence of fish farming of the species (The Guardian, 21st April 2021). This is due to the cultured salmon that escaped from their designated culture areas possibly mixing up with wild salmon and producing offsprings with inferior genetic makeup. This has diminished their ability to adapt to environmental changes and may decrease the population.

With the advancement of technology, secondary farmed types are produced from primary farmed types through genetic manipulation, hybridization, polyploidy and genetic engineering. These secondary farmed types must also be prevented from escaping into the environment as their genetic components are totally different from their wild relatives.

Currently, the increasing use of non-native species for aquaculture is alarming as the escapees pose a serious threat to our native species. For instance, African catfish (*Clarias gariepinus*), tilapia, Pacific oyster (*Crassostrea gigas*), red claw (*Cherax quadricarinatus*), Asian red tail catfish (*Hemibagrus wyckioides*), priacucu

(*Araipama gigas*) and peacock bass are among the invasive species known to be present in Malaysia. They have been illegally introduced by farmers for aquaculture based on their fast growth rates relative to the native stocks. The existence of these species may cause detrimental effects on environment, economic loss as well as the extinction of native species. This is due, to some extent, that some of them are predatory and fast breeding species which may dominate the habitats. A number of cases regarding the presence of non-native fishes in public

water were reported for the last ten years in Malaysia (Khairul-Adha, 2013, Naquiddin *et al.*, 2013, BERNAMA 17th April 2017, NST 17th April 2017, Ahmad *et al.*, 2020, Saba *et al.*, 2020, NST, 22nd February 2019, 3rd April 2021). In addition, this problem became worse with some religious belief, such as Buddhism belief that releasing the African catfish into the river can avoid disease and bad luck. This has contributed to the increased number of the non-native species in public waters.



Cherax quadricarinatus (photo by Fisheries Research Institute, Glami Lemi)



Peacock bass (*Cichla* spp.) found in Tasek Raban (photo by Fisheries Research Institute, Glami Lemi)



Asian red tail catfish (*Hemibagrus wyckioides*) (photo by Fisheries Research Institute, Glami Lemi)

The current aquaculture practice has also posed significant risks of unintentional introductions from culture systems. This is usually associated with broken fish net cages. For instance, the recent case of illegally cultured red tail catfish that escaped into Sungai Perak. This mass escape poses a serious threat to the river's ecosystem which is a favourable habitat for large stocks of giant freshwater prawn (*Macrobrachium rosenbergii*) and other aquatic species. It will affect the existing food webs and increase the risk of disease transfer in this habitat.

The abundance of this predator will also create competition with the native species in occupying the water bodies. It is challenging to reduce or eliminate the non-native species that are present in our environment. One of the crucial steps to be taken is promoting public awareness to educate and stop people from introducing or releasing non-native species into our rivers. In this regard, not only the Department of Fisheries has to enforce strict rules to curb this, the public are also responsible to end this threat.

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Managing Incoming Alien Species through Committee for Incoming Alien Species Application (CIASA), Department of Fisheries Malaysia

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The demand for non-native species for aquaculture or aquarium fish has increased in recent years. The rate of introduction is expected to increase due to the consistent growth in the aquaculture industry and aquarium trade. Both industries have been considered as the most important routes for alien fish species introduction into local freshwater ecosystems. The database on alien species in Malaysian fisheries is incomplete, especially in the study of alien species impacts, where they become invasive species in aquatic environments. Given the importance of these issues and the high demand from the aquaculture industry, the Department of Fisheries through the Biosecurity Fisheries Division has established the Committee for Incoming Alien Species Application (CIASA). The objective of CIASA is to minimize the import risk through Import Risk Analysis (IRA) for all applications of alien species for aquaculture purposes. Alien species herewith refer to any exotic or non-native species originating from outside Malaysia. The method used for IRA was based on the Manual on Risk Analysis for The Safe Movement of Aquatic Animals (FWG/1/2002). Under this manual, the import risk analysis focuses on two elements; (i). The ecological and genetic risk assessment process; and (ii). Pathogen, parasite, or fellow traveller risk potential. On each element, probability of establishment (e.g. in the intended and beyond the area of introduction) and consequence of the

establishment of an aquatic organism (e.g. ecological impact on native ecosystems and genetic impacts on the local population, if established) need to be determined for each application. The overall import risk analysis will be classified as high, medium or low based on the probability of establishment and the consequences of the establishment. High IRA will be rejected while medium and low may be approved subject to possible mitigation. All IRA findings will be presented at fisheries' top management committee meetings and the approval will be based on the final decision of the top management in the Department of Fisheries. There are two types of members involved during the IRA process; (i) The permanent member consists of the appointed chairman of CIASA and secretariat from the Fisheries Biosecurity Division, and (ii) Temporary resources and experts based on the alien species on each application. Temporary members for IRA are normally from the Department of Fisheries and institutions of higher learning.

Since 2010, CIASA received 52 applications with only 37 of them fulfilling the criteria to do the IRA, and only 23 or 62% were approved (Fig. 1). Most of the applications were for fish (73%), bivalve (15%), and followed by 4% each from seaweed, shrimp, and sea cucumber. The majority of incoming alien species come from China and Australia (Fig. 2). The popular alien species under the fish category were for sturgeon (*Acipenser*

spp.) and bass (*Macquaria* spp.) and pacific oyster (*Crassostrea gigas*) under the bivalve category. Since its establishment in 2010, CIASA has experienced some issues and challenges such as loopholes in the e-permit system, inconsistent mitigation action, limited regulations on live aquatic animal movement and existing alien species in Malaysian waters. Several recommendations on the strengthening of the DoF structure on CIASA were identified. Among them are i) Providing clear terms of reference (TOR) for the committee; ii) Assigning members and alternates; iii) Providing further training on import risk analysis and other risk sectors (e.g. genetics, ecology, etc.); iv) Providing adequate funding (including funding for external reviewers); v) Initiating the preparation of a national statement of ALOP/ALOR; and vi) Developing a strategy to communicate the risks and benefits of import risk analysis. Finally, to minimize the risk of incoming alien species, CIASA must be part of a comprehensive management strategy on efforts to regulate new introductions of alien species, implement a surveillance data programme, and involve stakeholders particularly when high risk alien species become invasive alien species.

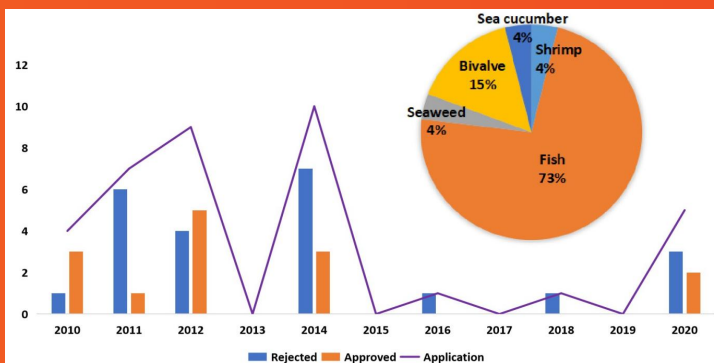


Figure 1: Total incoming application by stakeholders and categories of alien species in Malaysia from 2010 until 2020



Figure 2: Source of incoming alien species application

An Integrated Fish-Rice Culture at Jelebu Negeri Sembilan

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A trial on the integrated fish-rice farming system was conducted in Kg Sungai Buloh, Simpang Durian, Jelebu to add value to the rice production system. The objectives are to collect information on the integrated fish-rice culture system, on the species selection and the growth efficacy during the paddy planting period. The African catfish (*Clarias gariepinus*) was chosen because of its fast growth characteristics, almost complementing the paddy culture duration and having high demand in Negeri Sembilan either as fresh or processed products (smoked fish). In this trial, a batch of 3.5 inches catfish fry with the average weight of 6.0 ± 1.3 g was stocked into the trial plots

with a 1.5 pieces/m² stocking density, after 30-days of rice planting. Pellet feeding (32% protein) was used to increase the rate of fish growth. After 48 days of culture, the fish were harvested gradually until the paddy was fully harvested. The final results were 225.9 kg fish with an average weight of 86.6 ± 28.1 g were gathered. The Food Conversion Ratio (FCR) of every kilogram of fish was acceptable, i.e. 1.26. We have received positive responses from the rice farmers, citing additional income and, if this study is to be repeated, proper control measures must be put in place to reduce the impact of this alien species to the local environment.



Catfish caught during fishing competition held at the paddy field

MOMENTS @ FRI

Proficiency Testing Program for Bacteriology

On 1st until 14th June 2021, the Bacteriology Laboratory, Impact Assessment Research Division, FRI Batu Maung participated in an inter-laboratory Proficiency Testing organized by the Centre for Environment, Fisheries and Aquaculture Science (CEFAS), United Kingdom for analysis of fecal pollution indicator (*Escherichia coli*) in shellfish tissue and seawater. The Proficiency Testing was also participated by the Kuala Lumpur Fisheries Biosecurity Lab and the Kuantan Fisheries Biosecurity Lab. The objective of this program is to assess the competency of the laboratories in conducting the test.



Activities during the inter-laboratory proficiency testing

Launch of Wastetronic (IOT of Marine Fish Farming and Nursing)

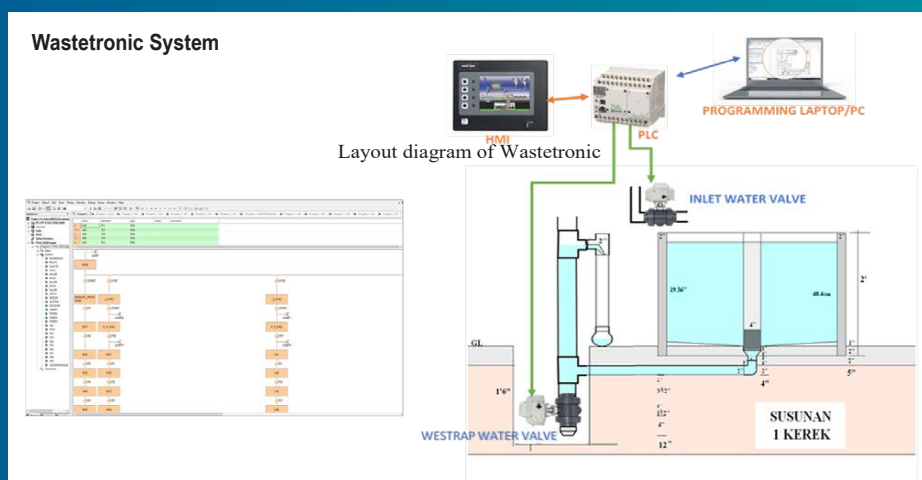
Wastetronic: It combines manual sewage disposal activities into an automated system which is controlled according to the operating mode of the inlet and outlet electric valves by using PLC (Computer Logic Program). The system is equipped with water quality control sensors and observation of low oxygen parameters and high ammonium content. The supposed value will direct the oxygen support equipment to function and the disposal of livestock/rearing water, as well as new water infiltration, is made whenever required. This activity is connected to an internet system to be controlled remotely by using devices such as computers and smart phones/mobile phones.

This innovation has improved the quality of livestock/rearing water according to the mode of operation that has been programmed depending on the suitability of fish size and rearing time. Water quality data will always be updated according to the selected time, while the automatic feeder control can be established according to the appropriate time period and operating frequency. Security monitoring can be done by placing warning lights and sirens when there is no electricity and CCTV is also installed to assist in real-time operation. Electrical outage warnings can also be monitored online whenever a notice is sent to the livestock system operator. For the purpose of application in a livestock system, two versions are already installed, CENT-RAS (nursed) and GROW-OUT RAS (Grow out).

The IOT (Internet of things) systems was successfully launched by the Deputy Minister of the Ministry of Agriculture and Food Industry (MAFI), YB Dato' Hj. Che Abdullah bin Mat Nawi on the 1st of April 2021. This innovation called WASTETRONIC was a collaborative project between FRI Tg Demong, Terengganu and Polytechnic Muadzam Shah, Pahang.



YB Dato' Hj. Che Abdullah bin Mat Nawi, Deputy Minister of the Ministry of Agriculture and Food Industry during launching ceremony at the FRI Tg. Demong, Terengganu



The lay-out of Wastetronic system



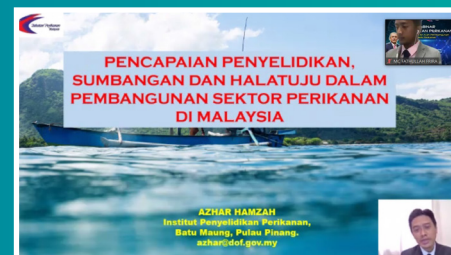
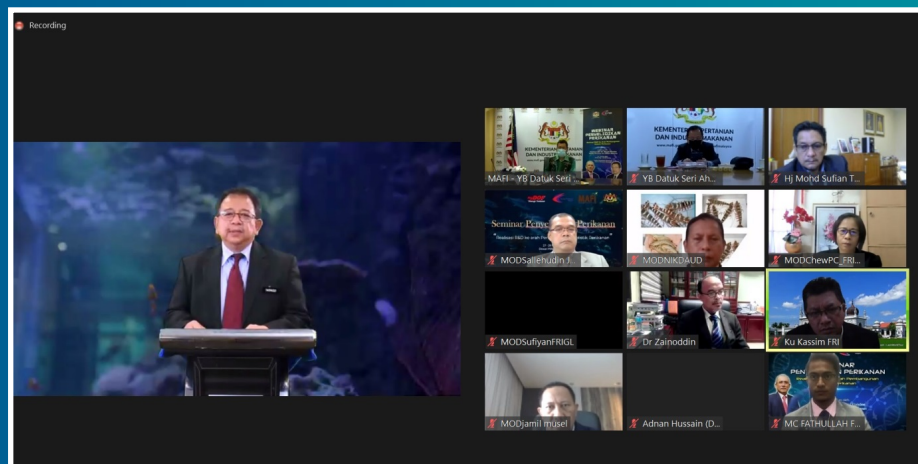
YB Dato' Hj. Che Abdullah bin Mat Nawi, received books from Dr Zainuddin bin Jamari, Senior Director, FRI Batu Maung, Penang

The First FRI Research Webinar

The FRI Research Webinar 2021 was successfully conducted on 27th and 28th May 2021 with the FRI Glami Lemi, Innovation, Promotion and Commercialization Unit, FRI Batu Maung and Bahagian Penyebaran Maklumat Department of Fisheries, Putrajaya as the organizer. The webinar was launched by the Deputy Minister YB Datuk Seri Haji Ahmad bin Hamzah from the Ministry of Agriculture and Food Industry. A total of 200 participants from the Department of Fisheries Malaysia, researchers, lecturers and students from local universities and also farmers took part in the webinar. YBrs Tuan Ahmad Tarmidzi bin Ramly, the Director-General of DoF and Dr Zainuddin Jamari, Senior Director of FRI also joined the webinar. Researches in various fields conducted by FRI were successfully translated into 19 oral presentations papers and 41 posters. There were also 10 selected innovations exhibited during the webinar. Since its establishment in 1957, FRI has been executing progressive research related to fisheries especially on aquaculture, capture fisheries and fish health and management.



The opening speech by the Deputy Minister of Agriculture and Food, YB Datuk Seri Haji Ahmad bin Hamzah



First presentation by the Deputy Senior Director of the FRI, Dr Azhar bin Hamzah

Welcoming speech by YBrs Tuan Ahmad Tarmidzi, the Director-General of DoF