Department of **Employment, Economic Development and Innovation**
Biosecurity Queensland

Fish Health Sampling Reports

Gladstone Harbour

As at 8 December 2011

Toward Q2 Tomorrow's Queensland

Queensland Government

Contents

Current situation

Biosecurity Queensland in collaboration with Fisheries Queensland is conducting an investigation into fish health issues reported from the Gladstone area. This report is a summary of the test results to date including preliminary toxicology results.

Biosecurity Queensland has been conducting tests on a range of fish samples, molluscs and crustaceans provided by Fisheries Queensland, individual fishers, and seafood markets from the Gladstone area.

To date, more than 100 fin fish, either whole or tissue samples including 23 sharks, 14 crustacean and two batches of mollusc samples have undergone a range of assessments and testing including gross pathology (external and internal evidence of disease), histopathology (microscopic examination of tissue), microbiology (presence of bacteria, fungi, parasites etc).

Results for three barramundi that have undergone toxicology testing have been received. It must be recognised that this is a very small sample size and the results will be used to inform future toxicology testing programs.

More testing will be required. As such, Fisheries Queensland is continuing to conduct further fish sampling in the Gladstone area and Biosecurity Queensland will continue to test samples submitted.

Samples

Fifty one individual submissions have been received by Biosecurity Queensland laboratories between 30 August 2011 and 28 November 2011.

As of 5 December 2011, these submissions included barramundi (31 whole, 30 tissues); sharks (3 whole, 20 tissues); whiting (1 whole, 12 tissues); spotted cod (1); spangled emperor (1); flathead (2); scat (1); trevally (1); mud crabs (4); prawns (15); Moreton Bay bug (2); 2 batches of scallops; and tissue samples from 10 mullet.

Testing processes

The investigative process involves:

1. Initial assessment of all submissions at the laboratory including description of samples received, their condition and gross pathology (visual signs of disease);
2. Conducting tests to exclude a range of exotic and endemic diseases;
3. Undertaking additional testing where appropriate, including sending samples to other laboratories for specific tests such as toxicology analysis; and
4. Conducting more detailed tests to provide a diagnosis for the identified fish health issue.

Veterinary Diagnostic Assessment: Significant findings

Fin fish

Barramundi

The majority of the barramundi submitted for laboratory tests showed eye lesions ranging in severity from inflammation to ruptured eyes.

Parasitic flatworm infection

Tests results to 2 November 2011, which were previously reported, identified a parasitic flatworm (*Neobenedenia* spp.) as the causative agent of the eye disease in the barramundi. Bacteria were excluded as the cause of the lesions.

This flatworm causes eye injury by way of its feeding and attachment over the surface of the cornea. In eyes that are cloudy, eroded, inflamed, swollen or where there is evidence of haemorrhage, vision is likely to be reduced. In severe cases where the eye has been ruptured, this will cause blindness.

The parasite moves over the surface area of the eye and the skin of the entire fish. A number of barramundi showed hyperaemic (reddened) areas on the skin surface, which is consistent with skin damage caused by the parasite feeding on mucous on the skin surface.

This was the first report of a significant outbreak of *Neobenedenia* spp. in wild barramundi. There is insufficient data to determine the origin of this flatworm and why they are particularly prevalent on the barramundi at this time. Lower winter temperatures are known to predispose fish to immunosuppression and a winter outbreak of *Neobenedenia* spp. High population densities can also result in an increase in the risk of transmission and infection of the parasite. Fish densities would have increased as a result of the spillover of barramundi from Awoonga Dam.

Further results from examination of samples from an additional 16 barramundi were consistent with results reported on 2 November 2011 from barramundi submitted from Gladstone Harbour. An additional new finding was copepods (small crustaceans) found on the gills of some of these barramundi. This is not considered to be an issue of significance at this point but should be followed up and compared with future submissions.

A number of barramundi have shown an increase in the presence of melanomacrophage centres (MMCs) in organs (e.g. liver, spleen and kidney). An increase in MMCs is a common histopathological change associated with a range of possible factors including reduced water quality, starvation, disease or aging. The general function of MMCs is the focalisation, destruction, detoxification and recycling of internal and external materials. The increased number of MMCs is likely a result of the general effects on fish health caused by the flatworm infestation.

Skin lesions and skin abnormalities

The causes of the deep ulcerative lesions in barramundi collected from Gladstone Harbour were not able to be determined. Specific histopathology testing showed that Epizootic Ulcerative Syndrome (EUS), commonly known as red spot disease, was not found in samples collected from the Harbour and was not the cause of these lesions.

Only one case of red spot disease has been found in testing to date. The positive result for red spot disease was found in a barramundi sample taken in September, which was collected from Port Alma outside Gladstone Harbour.

Although red spot disease was not found to be present in the lesions of barramundi from Gladstone Harbour, it does not exclude future cases of red spot in the area. The disease is endemic in Queensland.

Other fin fish species

Skin abnormalities from non-barramundi fish species were generally mild and not due to *Neobenedenia* spp. Fin fish species examined included spangled emperor, spotted cod, whiting, and trevally. Findings included localised skin inflammation, skin erosion, fibrosis (scarring), reddening, dermal haemorrhage (bleeding of the skin), epidermal necrosis (cell death) and oedema (swelling).

No bacterial, parasitic or fungal pathogens were found which could explain the skin conditions on these other fin fish species.

Microscopic examination of gills found no sediment on the gills in any of the samples.

A range of bacteria were isolated from the fish. The lack of distinct invasion or proliferation on the skin and muscle lesions suggests that these bacteria were opportunistic and secondary colonisers following the initial skin damage. These bacteria are not considered to be fish pathogens (i.e. they are not primary infectious agents that cause disease in fish). They are normal microorganisms of the marine environment.

Shark

Shark species examined included lemon shark, bull shark, pig eye shark, whitecheek shark, blacktip shark, whaler shark, graceful shark and narrow sawfish. Sharks presented with scale pocket hyperaemia (pockets of reddening), dermal haemorrhage (bleeding of the skin) and epidermal necrosis (cell death). Histopathological changes associated with the skin lesions included congestion in the connective tissue immediately below the epidermal basement membrane, occasional haemorrhage into the epidermis and /or skin surface and a mild inflammatory cell infiltration into the outer layer of the dermis. No bacterial or fungal pathogens were found which could explain these skin conditions on the sharks.

Skin parasites were found on the surface of 11 of the samples of preserved skin from the sharks. Preliminary identification of these parasites indicates they are small flatworms from the family Microbothriidae, which are different flatworms to the ones found on the barramundi. These have been sent to an aquatic parasitologist for confirmation of identity.

There was no detectable sediment in the gills of the shark samples.

Crustacean

The mud crabs examined indicated an erosive shell disease. These signs are consistent with bacterial infection by *Vibrio* spp. which are organisms found in marine waters. *Photobacterium (Vibrio) damaselae* was isolated from the most recent mud crab submission.

Prawns also showed evidence of shell erosion due to *Vibrio* spp. These bacteria are opportunistic and proliferate on damaged shell to cause erosion due to their chitinolytic enzymes.

A number of prawns including banana, king and tiger showed evidence of endoparasite infection of immature stages of tapeworms. These parasites are not unexpected in wild prawn populations and would not have had a significant impact on prawn health.

Toxicology testing

Toxicology testing was conducted on three barramundi samples taken from two locations – Port Alma (1 barramundi) outside Gladstone Harbour and China Bay (2 barramundi) within Gladstone Harbour. All three fish were considered to be unwell and exhibited clinical signs of ocular pathology due to *Neobenedenia* sp and displayed skin abnormalities.

Testing was conducted on the liver, gill and muscles of each of the three fish, resulting in nine tissue samples being analysed. This is a small sample size and is considered to be a preliminary round of toxicology testing. Results will be used to determine future toxicology testing programs.

These samples were among the first fish with lesions submitted for testing.

The toxicology analysis was conducted by Queensland Health Forensic and Scientific Services at Coopers Plains in Brisbane. Fish samples were tested for organic (e.g. agricultural chemicals) and inorganic compounds (metals).

The results and analysis provided in this report are exposure levels of these chemicals in the animals sampled. Further investigation and analysis is required to determine the toxicological result that these levels could have on aquatic animals in Gladstone Harbour.

For organic analytes the samples were analysed for organophosphorus type pesticides, organochlorine type pesticides, polychlorinated biphenyls (PCBs), synthetic pyrethroid insecticides, polyaromatic hydrocarbons (PAHs), selected herbicides, insecticides and fungicides. For results see Attachment 5.

For inorganic analytes, metal analyses was conducted for: aluminum (Al), arsenic (As), barium (Ba), cadmium (Cd), chromium (Cr), nickel (Ni), copper (Cu), iron (Fe), zinc (Zn), silver (Ag) and selenium (Se). Lead (Pb) and mercury (Hg) were not selected for analysis because the Western Basin Dredging and Disposal Project Environmental Impact Assessment (EIA) for Gladstone Harbour did not identify these metals as of concern as part of the EIA. For results see Attachment 5.

Additional toxicology testing is underway on other fish, crustacean and mollusc samples from Gladstone Harbour. These fish samples include healthy fish collected outside of the Harbour to develop baseline data for comparison with fish from within the Harbour.

Organic chemicals: results and significant findings

The samples submitted were tested for more than 160 chemicals. Samples are reported on a wet weight basis. Of the nine samples tested only one chemical was detected in one of the samples. This preliminary result suggests there is a low probability that organic chemicals are affecting the health of fish in Gladstone Harbour. However, further testing will be required due to the small sample size.

DDE p,p was the only organic chemical found at a detectable level, and this was only in the muscle tissue of one fish from China Bay. DDE p,p is an environmental metabolite (breakdown product) of DDT, a pesticide that was commonly used in the agricultural sector, but has not been permitted in Australia for more than 20 years. As only DDE p,p was detected, it indicates that the fish were not recently exposed to DDT. The chemical level was 0.014mg/kg, which is 0.002mg/kg above the level of detection. This result suggests a low probability that this chemical is at a level that would have caused acute or chronic fish health impacts.

Metals: results and significant findings

The gill, liver and muscle samples from the three fish samples were tested for the following 11 metals: aluminum (Al), arsenic (As), barium (Ba), cadmium (Cd), chromium (Cr), nickel (Ni), copper (Cu), iron (Fe), zinc (Zn), silver (Ag) and selenium (Se). Samples are reported on a wet weight basis.

Ten metals were detected across the range of samples. Based on an initial analysis of the scientific literature, the majority of detections were considered to be within the normal range for these metals in the types of tissue sampled. It is not unexpected that relatively higher levels of aluminium, iron and zinc were found in some tissues. Based on an initial analysis of scientific literature from Australia and overseas, the levels of metals detected is considered to have a low likelihood as a significant contributor to the reported ill health of fish from Gladstone Harbour.

The levels at which metals would have an impact on fish health differ among fish species and across different environment conditions. Therefore toxicological studies require comparisons across species and environments and across sick and healthy fish. Further testing is being conducted on other fish species, prawns and scallops collected from the harbour including testing of healthy fish and samples taken outside the Gladstone Harbour area. Scientific literature reviews will also continue to be undertaken to further assess levels of metals found in fish.

Further testing

It is important to highlight that these results are from a limited number of samples (three barramundi from two locations). Analysis and results for small sample numbers needs to be reviewed with caution as the samples may not be truly representative of the population. More samples are undergoing toxicology testing to give a broader understanding of potential impacts of chemicals on fish health. It is therefore not possible to reach a definitive conclusion on the impact of organic chemicals and metals based on these three samples. Further testing and analysis is required.

It should be appreciated that even with further testing, it is possible that no definitive diagnosis will be found.

Testing reports to date

Three interim reports have been prepared by Biosecurity Queensland.

Attachment 1

Report 1: Interim Veterinary Diagnostic Assessment (released 30 September 2011)

This first report was released on 30 September 2011 providing results on eight barramundi taken from within Gladstone Harbour and one barramundi taken from Port Alma.

Attachment 2

Report 2: Interim Veterinary Diagnostic Assessment (released 6 October 2011)

Report 2 provided additional results from samples taken from Gladstone Harbour and surrounds including species such as whiting, spotted cod, spangled emperor, prawns and mud crabs.

An explanatory note of findings covering both Report 1 and Report 2 was also released by Fisheries Queensland to provide additional information regarding the results **(Attachment 2.1).**

Attachment 3

Report 3: Interim Veterinary Diagnostic Assessment (released 2 November 2011)

Report 3 provides further results on the testing of barramundi samples along with a range of other fish species that had mild skin abnormalities.

Attachment 4

Report 4: Interim Veterinary Diagnostic Assessment including toxicology summary (released 8 December 2011)

Report 4 provides further results on the testing of a range of other fish species that presented with skin abnormalities, including sharks and crustaceans as well as a summary of toxicology results to date.

Attachment 5

Report 1: Gladstone fish toxicology data (released 8 December 2011)

Report 5 provides results on initial barramundi samples that have undergone toxicology testing.

ATTACHMENT 1

Biosecurity Queensland
Gladstone Fish Health
Interim Veterinary Diagnostic Assessment Report 1
30 September 2011

A. Gross pathology examination of barramundi from submissions P11-74868 (South Tree Inlet) and P11-74922 (Calliope River) showed eye lesions ranging in severity from inflammation to a ruptured eye. The eye lesions were caused by the parasite *Neobenedenia* spp. This parasite is known to cause eye injury by way of their feeding and attachment activities over the surface of the cornea. The role of a primary bacterial cause in the eye lesions is not supported by bacteriological and histopathological evidence. Further examinations are underway to elucidate the potential role of secondary bacterial pathogens. The severity of the eye lesions in some fish will cause blindness in the eye where it is ruptured. In eyes that are cloudy, eroded, inflamed, swollen or haemorrhagic, vision is likely to be reduced. A number of barramundi showed hyperaemic (reddened) areas on the skin surface which is consistent to skin damage caused by the parasite feeding on mucous on the skin surface.

   This is the first report of a significant outbreak of *Neobenedenia* spp. in wild barramundi. There is insufficient data to determine the origin of the *Neobenedenia* spp., why they are particularly involved in the barramundi and at this time period. Lower winter temperatures are known to predispose fish to immunosuppression and consequently a winter outbreak of *Neobenedenia* spp.

B. Gross pathology examination of the barramundi from submission P11-74903 (Port Alma) showed skin lesions. Histopathology examination of the lesions showed they were consistent with Epizootic Ulcerative Syndrome (EUS), caused by *Aphanomyces invaderis*. Specific histopathology testing confirmed the lesion was caused by this fungus. Note that Port Alma is outside of the Gladstone Harbour fishing exclusion zone. The fungus is the primary pathogen and secondary bacteria *Micrococcus* spp., *Moraxella* spp., *Proteus vulgaris, Pseudomonas* spp. isolated from the lesions have followed the skin and muscle damage initiated by the EUS fungus. EUS is endemic to Queensland.

   The causes of the deep chronic necrotic and ulcerative lesions in barramundi in submissions P11-74922 (Calliope River) and P11-74662 (Boyne River) are not able to be determined. EUS was considered to be a differential diagnosis for these lesions. Specific histopathology testing showed this organism was not present and hence not the cause of these lesions. Although light, mixed growths of *Moraxella* and *Pseudomonas* spp. were isolated from skin, these are considered to be bacteria secondary to initial skin damage. Although EUS was not found to be present in the lesions of these fish it does not exclude EUS causing lesions in other fish from Gladstone Harbour.

   Specific bacteriology testing for *Streptococcus agalactiae* and *Streptococcus iniae* have been completed and these organisms were not isolated.

C. Histopathology examination of gills found no sediment on the gills in any of the samples.

D. Most fish examined were noted to have no food in their stomachs and intestines. This suggests a lack of recent feeding which may be explained by visual deficit in fish with eye disease resulting in a reduced capacity to catch prey.

E. The gill myxosporean, consistent with *Henneguya* spp., a spore forming parasite, was identified by histopathology examination. This finding was considered a minor finding with minimal impact on barramundi health at the current level of infestation. The presence of nematodes in the intestine of the barramundi is considered a minor finding with minimal impact on barramundi health at the current level of infestation.

F. Examination of the mud crab showed significant erosion of the carapace body. This type of erosion is consistent with Shell Disease.

G. Examination of the Spotted Cod (P11-75085) and the Spangled Emperor (P11-75194) showed different signs to those of the barramundi. The Spotted Cod had superficial lesions on left side. Testing is ongoing for these submissions.

H. Examination of the prawn submission (P11-75123) is underway but no results to report at this time.

I. This is an interim report and subject to revision and refinement when new information is available pending completion of examinations in progress.

Fish samples were submitted to the Biosecurity Sciences Laboratory (BSL), Aquatic Animal Health Unit to investigate and determine the possible cause or causes of the eye and skin abnormalities. The Tropical and Aquatic Animal Health Laboratory provided valuable diagnostic support for BSL.

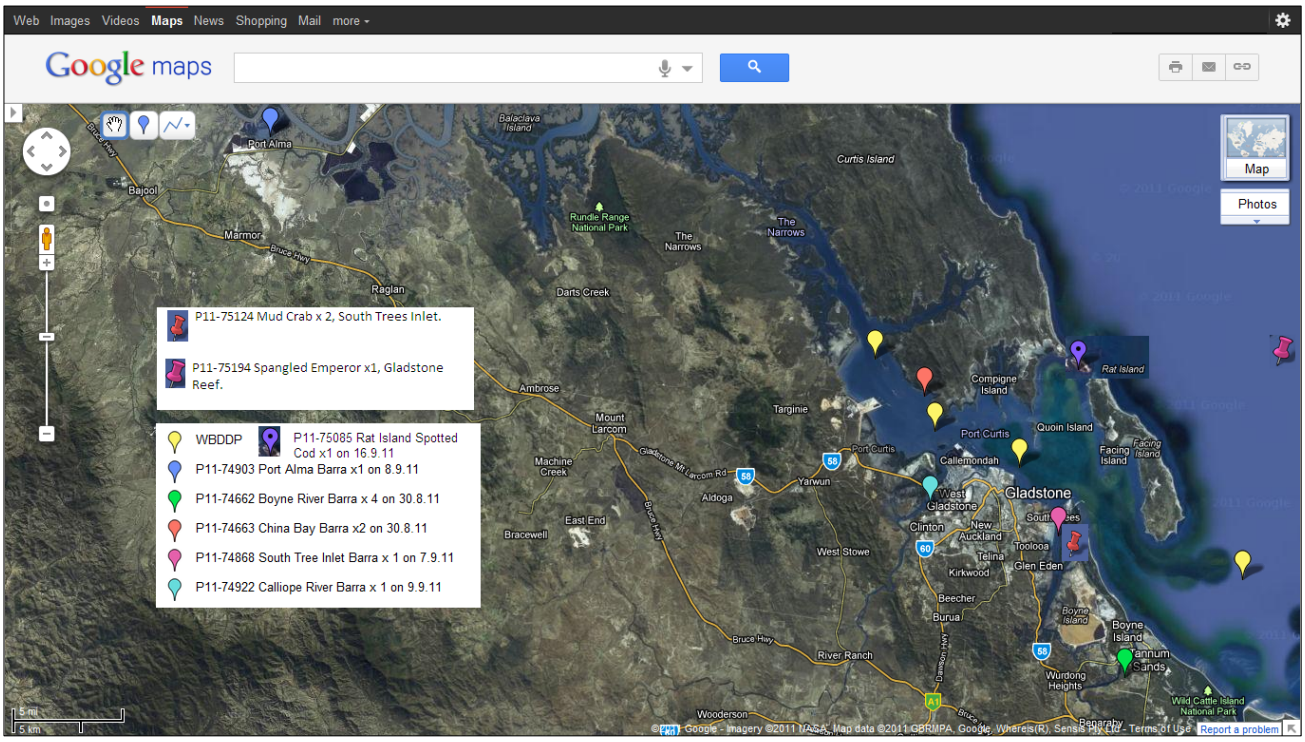
Table 1: summarises the fish and shellfish samples received as of 27.9.11 in connection with Gladstone.

Date received /Case no.	Sample Type, Number, Condition	History	Findings
30.8.11 P11-74662	Whole barramundi x 4 Dead >24h on ice	Caught from Boyne River, Gladstone. Eye and skin lesions reported	Ocular pathology. One fish with deep necrotic lesion. EUS not found
30.8.11 P11-74663	Whole barramundi x 2 Dead >24h on ice	Caught from China Bay, Gladstone. Eye lesions reported	Ocular pathology
5.9.11 P11-74796	Whole Moreton Bay bug x 1 Dead >2 weeks, decomposed as not on ice in transit to lab	Coordinates 23.28.70-151.30.50 23.22.80-151.26.20	Samples unsuitable for testing
7.9.11 P11-74868	Whole barramundi x 1 Dead >24h on ice	Caught from South Tree Inlet, Gladstone. Eye lesions reported	Ocular pathology caused by flatworm *Neobenedenia* spp.
8.9.11 P11-74903	Whole barramundi x 1 Dead >24h on ice	Caught from Port Alma, Gladstone. Skin lesions reported	Severe deep chronic necrotic lesions. Lesions caused by EUS
9.9.11 P11-74922	Whole barramundi x 1 Dead >24h on ice	Caught from Calliope River, Gladstone. Eye and skin lesions reported	Ocular pathology caused by flatworm. *Neobenedenia* spp. EUS not found
16.9.11	Whole spotted cod x 1	Caught from Rat	Superficial lesions on

Date received /Case no.	Sample Type, Number, Condition	History	Findings
P11- 75085	Dead <24h on ice	Island, Gladstone. Skin lesions reported	left side. Testing ongoing
20.9.11 P11-75123	Whole prawn x 2 Dead >24h on ice	Location being clarified	To be reported
20.9.11 P11-75124	Whole mud crab x2 Live	Caught from mouth of South Tree Inlet, Gladstone	Erosion of carapace consistent with Shell Disease
22.9.11 P11-75194	Spangled Emperor x1 Dead >24h on ice	Caught from Gladstone Reef	To be reported
27.9.11 P11-75286	Whiting	Caught outside Gladstone Harbour closure area	To be reported

Note: As all fish were received by the laboratory as dead fish on ice, there is a risk that Neobenedenia *spp. have detached from the fish, therefore reducing the probability of detection.*

The barramundi samples have been sent by Gladstone fish markets representing fish caught by a number of fishers in Gladstone. The map following shows the approximate locations of the barramundi sampled in relation to the Western Basin Dredging and Disposal Project (WBDDP) sites. The WBDDP sites for dredging are activated in stages with the dumping site outside of Gladstone Harbour. Map of fish samples submitted for diagnostic examination to Biosecurity Queensland is as follows:



ATTACHMENT 2

Biosecurity Queensland
Gladstone Fish Health
Interim Veterinary Diagnostic Assessment Report 2
6 October 2011

A. The cause of the severe ulcerative lesions on the barramundi samples from Gladstone Harbour could not be determined. EUS (red spot) and bacterial infection have been ruled out as causative agents. The skin damage is serious and increases the likelihood of mortality in affected fish.

   A range of bacteria were isolated from the affected barramundi. However the lack of distinct invasion or micro-colony establishments in the skin and muscle lesions suggests that they are not fish pathogens. Bacteria isolated include: *Micrococcus* spp.; *Moraxella* spp.; *Proteus vulgaris* and *Pseudomonas* spp.

B. The Spotted Cod, Spangled Emperor and Whiting did not show eye disease nor have these fish been observed to carry *Neobenedenia* spp., even at subclinical levels.

   EUS was not detected in the Spotted Cod, Spangled Emperor or Whiting skin lesions. A protozoan *Uronema*-like parasite was found in the Spangled Emperor. This was not considered to be significant. Histopathology of the Whiting showed skin fibrosis, but there were no other significant findings. Histopathology showed the skin lesions of the Spotted Cod were due to superficial abrasions. This could be caused by mild trauma.

C. Histopathology examination of the gills of all fish submitted from Gladstone Harbour found no sediment on the gills of these samples.

D. Prawn and mud crab samples examined indicated an erosive shell disease which is most likely the result of bacterial infection by *Vibrio* spp. which are organisms found in marine waters. These bacterial are opportunistic and cause shell fouling with erosion due to chitinolytic activity. The mud crab had a low grade viral infection in the hepatopancreas, but is not considered significant.

This is an interim report and subject to revision and refinement when new information is available pending completion of examinations in progress.

Table 1: summarises the fish and shellfish samples received as of 6.10.11 in connection with Gladstone Harbour Fish Health investigation.

Date received /Case no.	Sample Type, Number, Condition	History	Findings
30.8.11 P11-74662	Whole barramundi x 4 Dead >24h on ice	Caught from Boyne River, Gladstone. Eye and skin lesions reported	Ocular pathology. One fish with deep necrotic lesion. EUS not found
30.8.11 P11-74663	Whole barramundi x 2 Dead >24h on ice	Caught from China Bay, Gladstone. Eye lesions reported	Ocular pathology
5.9.11	Whole Moreton Bay	Coordinates	Samples unsuitable for

Date received /Case no.	Sample Type, Number, Condition	History	Findings
P11-74796	bug x 1 Dead >2 weeks, decomposed as not on ice in transit to lab	23.28.70-151.30.50 23.22.80-151.26.20	testing
7.9.11 P11-74868	Whole barramundi x 1 Dead >24h on ice	Caught from South Tree Inlet, Gladstone. Eye lesions reported	Ocular pathology caused by flatworm *Neobenedenia* spp.
8.9.11 P11-74903	Whole barramundi x 1 Dead >24h on ice	Caught from Port Alma, Gladstone. Skin lesions reported	Severe deep chronic necrotic lesions. Lesions caused by EUS
9.9.11 P11-74922	Whole barramundi x 1 Dead >24h on ice	Caught from Calliope River, Gladstone. Eye and skin lesions reported	Ocular pathology caused by flatworm *Neobenedenia* spp. EUS not found
19.9.11 P11-75082	Mud crab X1	Found dead Shelley's Beach	Erosion of carapace consistent with shell disease. Cause of death not able to be determined due to post mortem degeneration.
16.9.11 P11- 75085	Whole spotted cod x 1 Dead <24h on ice	Caught from Rat Island, Gladstone. Skin lesions reported	Superficial lesions on left side due to superficial abrasions
20.9.11 P11-75123	Whole prawn x 2 Dead on ice >24h	Location being clarified	Erosive shell disease
20.9.11 P11-75124	Whole mud crab x2 Live	Caught from mouth of South Tree Inlet, Gladstone	Erosion of carapace consistent with Shell Disease. Low grade viral infection of hepatopancreas
22.9.11 P11-75194	Spangled Emperor x1 Dead >24h on ice	Caught from Gladstone Reef	Intestinal trematode cysts. Protozoan *Uronema*-like parasite
27.9.11 P11-75286	Whiting X1 Dead >24h on ice	Caught outside Gladstone Harbour closure area	Skin fibrosis
4.10.11 P11-75412	Barramundi X1	Water outlet Calliope	To be reported

Note: As all fish were received by the laboratory as dead fish on ice, there is a risk that Neobenedenia *spp. have detached from the fish therefore reducing the probability of detection.*

A preserved collection of the *Neobenedenia* spp., a capsalid monogenean flatworm, has been forwarded for confirmatory identification. Results expected in 2-3 weeks.

Samples of barramundi from submissions P11-74663 (China Bay) and P11-74903 (Port Alma) have been forwarded for toxicological study including: heavy metals; agrichemical residues and polyaromatic hydrocarbons (PAH) residues. Results expected 6-8 weeks.

ATTACHMENT 2.1

Explanatory note to Biosecurity Queensland's Interim Veterinary Diagnostic Assessment Report 2 (released on 6 October 2011)

This explanatory note was to provide further explanation of findings in Biosecurity Queensland's Interim Veterinary Diagnostic Assessment Report 2 which was released on 6 October 2011. Information provided in this explanatory note was prepared by Fisheries Queensland.

Red-spot disease has not been ruled out as the causative agent of the lesions seen on fish around the Gladstone area.

We believe the parasitic flatworm may be the major cause of redness and illness in the barramundi collected from the Harbour and may have led to some of the lesions we have seen.

The Biosecurity Queensland report from 6 October 2011 includes results from 8 fish tested from Gladstone Harbour.

Fish samples tested from Port Alma, which is just outside Gladstone Harbour, did test positive for red-spot disease.

The Harbour fish that showed lesions did not test positive for red-spot disease, however the symptoms of lesions were consistent with the disease. All these fish did test positive for the parasitic flatworm (fluke). The interim veterinary diagnostic assessments are focused on reporting on the samples that have been received for testing. It would be an error to draw conclusions from these results in isolation.

Experts strongly advise readers to refer to the earlier report dated 30 September 2011 which states *'although EUS* (red-spot disease) *was not found to be present in the lesions of these fish it does not exclude EUS causing lesions in other fish from Gladstone harbour'*. We believe that the estimated 30,000 barramundi washed over the spillway at Awoonga Dam between December 2010 and March 2011 due to overtopping has contributed significantly to the number of fish seen with symptoms.

The presence of these fish in the Harbour is reflected in the dramatically increased catches of barramundi by commercial fishers during this year.

Our hypothesis is fish suffered physical stress, which combined with the stress of the relocation, would have also made them susceptible to disease.

The addition of so many large barramundi in the Boyne River below the dam would have significantly increased competition for food and increased physical stress levels of all fish.

The likely significant increase in stress levels has lowered the ability of fish to tolerate these parasites on their skin. As a result, the fish attempt to remove them by rubbing against hard objects, causing the redness.

In severe cases the redness may become an open wound (lesion), which can attract secondary infections.

Further targeted sampling and testing seeks to confirm the relationship between the parasite and the redness.

The Department of Employment, Economic Development and Innovation remains open and transparent with our testing of fish in the Gladstone area showing signs of lesions and cloudy eyes.

That’s why we make test results conducted by Biosecurity Queensland publicly available.

We will continue collecting more samples of barramundi and other fish for testing and will make the results of those tests publicly available.

The closure was lifted as we had identified the likely causes of the symptoms, and Queensland Health could not find any association with fish disease and human health concerns, and symptoms identified in the sick fishers.

The identification of likely diseases producing the symptoms led Queensland Health to determine that there was no risk to human health or unsafe food, providing safe food procedures are followed.

ATTACHMENT 3

Biosecurity Queensland
Gladstone Fish Health
Interim Veterinary Diagnostic Assessment Report 3
2 November 2011

A. As of 24 October 2011, laboratory analysis of fish from Gladstone included 26 submissions of 32 whole barramundi, tissues from 15 barramundi, 1 whole spotted cod, 1 spangled emperor, 1 whiting, 1 scat, 1 trevally, 3 sharks, 3 mud crabs, 2 prawns and 1 Moreton Bay bug. Tests conducted include gross pathology, histopathology, and bacteriology. (For further details of submissions and interim results refer to Table 1). Toxicology testing to examine fish tissue residues of heavy metals, agrichemicals and polyaromatic hydrocarbons (PAHs) is in progress with results expected by the end of November 2011.

B. Key results to date indicate that Gladstone barramundi have eye and/or skin disease in up to 80% of all fish submitted for testing at the Biosecurity Queensland Biosecurity Sciences Laboratory and is due in significant part to *Neobenedenia* flatworm. The identity of this capsalid monogenean (flatworm) has been independently confirmed by an expert fish parasitologist in South Australia as *Neobenedenia* spp. The infection rate of *Neobenedenia* flatworms on all fish submitted for testing is 86%, with the eye lesion rate at 79% and skin abnormality rate at 100%. The eye disease manifests as cloudiness, redness and eye rupture. The skin disease manifests mostly as redness and in some fish as ulceration. There is no evidence for Epizootic Ulcerative Syndrome (EUS) commonly known as red spot disease.

C. The barramundi kidney, spleen and particularly in the liver have significantly increased numbers of melanomacrophage centres (MMCs). This is potentially a bio-indicator of catabolic processes in the fish. The catabolic changes related to an increased number of MMCs, probably reflect the processing of an increased rate of red blood cell death and other deteriorated cells due to chronic skin and eye damage caused by *Neobenedenia* spp. or by the affected osmoregulation.

D. Skin abnormalities from non-barramundi fish species submitted for testing are generally mild and are not due to *Neobenedenia* spp. Sharks presented with scale pocket hyperaemia, dermal haemorrhage and epidermal necrosis. Whiting presented with localised skin erosion, inflammation and fibrosis. Trevally presented with localised epidermal hyperplasia and degeneration. Spangled emperor showed localised inflammation of skin and presence of abnormal cells. Spotted cod displayed localised skin inflammation and oedema. No bacterial, parasitic or fungal pathogen were found which could explain these skin diseases.

E. A range of bacteria were isolated from the submitted fish, being a mixed culture of bacterial species. The lack of distinct invasion or proliferation in the skin and muscle lesions suggests that the isolated bacteria are not fish pathogens. These include *Micrococcus* spp., *Moraxella* spp., *Proteus vulgaris, Pseudomonas* spp., and *Pseudomonas fluorescens.*

F. The prawn and mud crab samples examined indicated an erosive shell disease. The signs are consistent with bacterial infection by *Vibrio* spp. which are organisms found in marine waters. The bacteria are opportunistic and proliferate on damaged shell to cause erosion due to their chitinolytic enzymes.

Table 1: Interim summary of samples received as of 24 October 2011 in connection with Gladstone Harbour Fish Health investigation.

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
30.8.11 P11-74662	Whole barramundi fish x 4 Dead >24h on ice	Caught from Boyne River, Gladstone. Eye and skin lesions reported	1. Ocular pathology with corneal hyperaemia, haemorrhage, lymphocytic inflammation, oedema and erosion. Sclera, conjunctiva, choroid and iris tissues with hyperaemia/congestion and inflammation. 2. One fish with chronic, deep necrotic muscle. No evidence of EUS (red spot disease). 3. Absence of observable sediment on the gills. 4. Stomach empty. 5. Gills with myxosporean *Henneguya*-like spp. spore forming parasite, low to moderate level infestation. 6. Intestine with low infestation of larval nematodes in granulomas of the submucosa and muscle layers. 7. Heart with chronic low level infection due to *Cruoricola*-like spp. blood fluke eggs. 8. Liver – incidental coccidian-like parasite. 9. Kidney and spleen in some sections with heavy melanomacrophage centres (MMCs).
30.8.11 P11-74663	Whole barramundi fish x 2 Dead >24h on ice	Caught from China Bay, Gladstone. Eye lesions reported	1. Ocular pathology with corneal oedema and erosion, inflammation in choroid with eosinophilic granulocytes and congestion. 2. No evidence of EUS (red spot disease). 3. Stomach empty. 4. Absence of observable sediment on the gills. 5. Gills with myxosporean *Henneguya*-like spp. spore forming parasite, low level infestation. 6. Intestine with moderate infestation of nematodes in granulomas of the submucosa and muscle layers. 7. Heart with chronic low level infection due to *Cruoricola*-like spp. blood fluke eggs.
5.9.11 P11-74796	Whole moreton bay bug x 1 Dead >2 weeks, decomposed as not on ice in	Coordinates 23.28.70-151.30.50 23.22.80-151.26.20	1. Possible injury and secondary microbial invasion of the lesion with host response producing melanisation of the affected area. Gills fouling.

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
	transit to lab.		
7.9.11 P11-74868	Whole barramundi fish x 1 Dead >24h on ice	Caught from South Tree Inlet, Gladstone. Eye lesions reported	1. Ocular pathology (keratitis, necrosis) and blindness (ruptured eye) caused by infestation with monogenean capsalid fluke *Neobenedenia* spp. 2. No evidence of EUS (red spot disease). 3. Absence of observable sediment on gills. 4. Stomach empty. 5. No evidence of bacterial disease. 6. Gill myxosporean *Henneguya*-like spp. at low level causing localised gill infection. 7. Intestine – low level larval nematode granulomas. 8. Heart and kidney with chronic low level infection due to *Cruoricola*-like spp. blood fluke eggs.
8.9.11 P11-74903	Whole barramundi fish x 1 Dead >24h on ice	Caught from Port Alma, Gladstone. Skin lesions reported	1. Skin-muscle – severe deep, ulcerative, necrotic and haemorrhagic lesions. Fungal pathogen (*Aphanomyces invadens/invaderis*) consistent with that which causes Epizootic Ulcerative Syndrome (EUS) or red spot disease observed in lesions. 2. Mixed growth of bacteria *Micrococcus* spp., *Moraxella* spp., *Proteus vulgaris, Pseudomonas* spp. from skin lesion. Spleen – no aerobic growth. These bacteria are not primary fish pathogens. 3. Absence of observable sediment on gills.
9.9.11 P11-74922	Whole barramundi fish x 1 Dead >24h on ice	Caught from Calliope River, Gladstone. Eye and skin lesions reported	1. Ocular pathology with corneal erosion, generalised, severe, keratitis with focal haemorrhages and significant eosinophilic granulocyte infiltration. Oedema and separation of corneal stroma. This is caused by *Neobenedenia* spp. 2. Skin-muscle – severe deep, ulcerative, necrotic and haemorrhagic lesions. Light, mixed growth of *Moraxella* and *Pseudomonas* spp. isolated from skin. No aerobic growth from eye (aqueous humour, spleen). These bacterial are secondary contaminants of the skin lesion, not causative. 3. Gill, heart and kidney with chronic moderate to high level infection due to *Cruoricola*-like spp. blood fluke

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
			eggs. 4. No evidence of EUS (red spot disease). 5. Stomach empty. 6. Absence of observable sediment on the gills.
16.9.11 P11-75082	Mud crab female x1	Shelley Beach, Rockhampton Submitted by Biosecurity Queensland	1. Advanced post mortem degeneration. 2. Extensive erosion of the lateral carapace. 3. Three caudal sets of gill filaments were absent. 4. No obvious abnormalities were apparent.
16.9.11 P11- 75085	Whole spotted cod fish x 1 Dead <24h on ice	Caught from Rat Island, Gladstone. Skin lesions reported	1. Cataract in one eye, normal other eye. 2. Superficial skin abrasions and erythema one side of body. 3. No evidence of EUS (red spot disease). 4. No significant bacterial pathogen isolated. 5. Absence of observable sediment on the gills. 6. No evidence of systemic bacterial infection.
20.9.11 P11-75123	Whole prawn x 2 Dead on ice >24h	Location being clarified	1. Erosive shell lesions and melanisation, subcuticular fibrosis and inflammation. 2. A mixed bacterial population isolated, including *Moraxella* spp., *Pseudomonas* spp., *Vibrio splendidus*. The shell lesions can be caused by *Vibrio*.spp (chitinolytic activity). 3. Absence of observable sediment on the gills.
20.9.11 P11-75124	Whole mudcrab x2 Live	Caught from mouth of South Tree Inlet, Gladstone	1. Erosive shell lesions and melanisation (biofouling and bacteria), subcuticular inflammation. 2. A mixed bacterial population isolated, including *Shewanella putrefasciens, Vibrio* spp. *Vibrio splendidus I, Alcaligenes faecalis, Moraxella* spp., *Vibrio alginolyticus.* 3. Gill – sediment fouling and gregarine-like organisms. 4. Hepatopancreatic viral infection.
22.9.11 P11-75194	Spangled Emperor x1 Dead >24h on ice	Caught from Gladstone Reef	1. Protozoan-like infestation of skin. 2. Intestinal trematode cysts. 3. Absence of observable sediment on the gills. 4. No evidence of EUS (red spot disease). 5. No significant bacterial pathogen

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
			isolated.
27.9.11 P11-75286	Whiting x1 Dead >24h on ice	Caught outside exclusion zone. No specified location	1. Skin fibrosis. 2. No evidence of EUS (red spot disease). 3. No significant bacterial pathogen isolated.
4.10.11 P11-75412	Barramundi x 1 Dead > 72h	Caught from Calliope River near the hot water outlet on 30.9.11. Eye lesions reported. Submitted by Fisheries Queensland	1. Eye lesions – *Neobenedenia* flatworm isolated from body. 2. Absence of observable sediment on gills. 3. Kidney and spleen with severe melanomacrophage centres (MMCs). 4. Heart – moderate numbers of *Cruoricola*-like spp. Fluke eggs. 5. Spleen and muscle low level microsporidian parasite. 6. No evidence of EUS (red spot disease). 7. No significant bacterial pathogen isolated.
6.10.11 P11-75466	Barramundi x 1 Dead 61h	Witt Island. Submitted by Fisheries Queensland	1. Redness of pelvic fins. 2. Kidney – severe levels of MMCs. 3. Intestine – moderately level nematode granulomas. 4. No evidence of EUS (red spot disease). 5. No significant bacterial pathogen isolated.
6.10.11 P11-75467	Barramundi x 2 Dead 61h	Hamilton Point. Submitted by Fisheries Queensland	1. Eye – cornea with ulceration, inflammation, necrosis, hyperaemia, oedema. 2. Skin – mild redness ventral body, tail and pectoral fin bases. 3. Kidney – severe levels of black MMCs. 4. Liver – central area of hepatitis and fibrosis. 5. No evidence of EUS (red spot disease). 6. No significant bacterial pathogen isolated.
6.10.11 P11-75468	Barramundi x 1 Dead 61h	Tide Island. Submitted by Fisheries Queensland	1. Skin – a few superficial red marks on left side more than right side. 2. Kidney – severe levels of black MMCs. 3. Liver – quite large and moderate level of light brown MMCs. 4. Intestine with low level nematodes. 5. Low level *Cruoricola*-like spp. fluke. 6. No evidence of EUS (red spot disease). 7. No significant bacterial pathogen isolated.

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
7.10.11 P11-75528	Barramundi x 1 Dead > 48h	Hamilton Pt. Submitted by Fisheries Queensland	1. Skin- There was moderate hyperaemia of the ventral midline, but no skin ulcers. 2. There was slight hyperaemia of the right eye. 3. Liver – severe level brown MMCs. Spleen – moderate dark brown MMCs. Kidney – severe level black MMCs. 4. Spleen – *Pseudomonas fluorescens* bacteria isolated, but no evidence of systemic bacterial disease. 5. Low level intestinal nematode granulomas, *Henneguya.* 6. No evidence of EUS (red spot disease).
7.10.11 P11-75529	Barramundi x 1 Dead > 48h	GL001. Submitted by Fisheries Queensland	1. Left eye ruptured and collapsed. Right eye – corneal opacity and peripheral hyperaemia. 2. Gills – moderate level *Henneguya* spp. and low level *Cruoricola*-like spp. fluke. 3. Kidney – severe MMCs, liver and spleen – moderate MMCs. 4. No evidence of EUS (red spot disease). 5. No significant bacterial pathogen isolated.
7.10.11 P11-75530	Barramundi x 1 Dead > 48h	SAM004-FXLW 24.035 115.325 Submitted by Fisheries Queensland	1. Skin – no skin ulcers were evident, but there was mild hyperaemia of the ventral midline. Skin – *Moraxella* spp. Spleen – no aerobic growth. Unidentified Gram positive motile bacillus from skin sample. 2. Eye – there was hyphaema of the left eye, as well as flocculent yellow material in the anterior chamber. 3. Spleen – moderate level dark-brown MMCs. Kidney – moderate level black MMCs. 4. Gills – low level *Henneguya* spp. plasmodia. 5. No evidence of EUS (red spot disease).
7.10.11 P11-75531	Barramundi x 1 Dead > 48h	SAM002-FXLW Submitted by Fisheries Queensland	1. Eye – cornea with keratitis, sloughing of cornea. 2. Spleen – severe black MMCs, Kidney – severe black MMCs. 3. Intestine – moderate level nematode granulomas. 4. No evidence of EUS (red spot disease). 5. No significant bacterial pathogen

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
			isolated.
7.10.11 P11-75566	Barramundi x 14 Dead >48h	Tide island x1 South Trees Inlet, Northside x1 FXLW 151.325/24.035 x1 Turkey beach x1 Turkey beach FYGT x1 GBL002 x1 Witt Island x1 FUNX fish5 x 1 SAM005-FXLW x1 Barra x 2 FUNX site 1 x1 GBL003 x1 Hamilton harbour x1 Submitted by Fisheries Queensland	1. 9 fish have eye lesions of varying severity from cloudiness, hyperaemia to rupture. 2. 7 fish have skin changes mainly with redness either localised or more regional in distribution, but no ulceration. 3. Fish were unsuitable for internal examination.
7.10.11 P11-75569	Shark (Lemon) x 1 Dead >48h	FXLW SAM001. Submitted by Fisheries Queensland	1. Skin – some areas of redness along flank and ventrally. Skin – separation of epidermis and scales from dermis and individual cell necrosis of epidermis.
7.10.11 P11-75571	Scat marine species x1 Dead >48h	Location to be advised. Submitted by Fisheries Queensland	1. Skin – small ulcer, middle top of head. Small red lesion left side. Ventral redness. 2. Dermatitis with focal lymphocytes and some necrotic inflammatory cells. Sloughing of mucous cells and epidermis. 3. No EUS granuloma formation.
7.10.11 P11-75573	Trevally x1Dead >48h	Location to be advised. Submitted by Fisheries Queensland	1. Tail – redness. Pectoral fin – redness. Skin – localised epidermal hyperplasia and degeneration. No EUS granuloma formation. 2. Tail – redness. Pectoral fin – redness.
13.10.11 P11-75654	Bull shark x 2 Dead >24h	Location to be advised	1. Shark 1 – body with red marks at mid to caudal left side worse than right side. Anus bleeding. Skin with 2-3+ mucous mixed with blood. Shark 2 – Body with ventral rash onto left side of abdomen and one area under mandible. 2. Skin – dermal haemorrhage and epidermal erosion and necrosis, scale pocket hyperaemia with inflammatory cells – eosinophilic granulocytes. Skin – scale pocket hyperaemia, epithelial erosion. Dermis with haemorrhage (mild) and

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
			in a linear fashion.
24.10.11 P11-75875	Formalin fixed tissues (gills, skin, internal organs), ectoparasites from 15 barramundi. Bacterial swabs for culture from 5 barramundi. Frozen tissues from 1 barramundi for toxicology (gills, liver, muscle).	Location to be advised. Submitted by Fisheries Queensland	Testing in progress.

ATTACHMENT 4

Biosecurity Queensland
Gladstone Fish Health
Interim Veterinary Diagnostic Assessment and Toxicology Report 4
8 December 2011

A. As of 8 December 2011, laboratory analysis of fish from Gladstone included 51 submissions of: barramundi (31 whole, 30 tissues); sharks (3 whole, 20 tissues); whiting (1 whole, 12 tissues); 1 whole spotted cod; 1 spangled emperor; 2 flathead; 1 scat; 1 trevally; 4 mud crabs, 15 prawns, 2 Moreton Bay bug, 2 batches of scallops and tissue samples from 10 mullet of mixed species. Tests conducted include gross pathology, histopathology, and bacteriology. (For further details of submissions and interim results to date refer to Table 1). Toxicology testing to examine fish tissue residues of heavy metals, agrichemicals and polyaromatic hydrocarbons (PAHs) was undertaken.

B. Further results of the examination of samples from an additional 16 barramundi were consistent with results reported on 2 November from barramundi submitted from Gladstone Harbour. The key finding to date indicate that Gladstone barramundi have eye and/or skin disease that is, in significant part, due to Neobenedenia flatworm. The eye disease manifests as cloudiness, redness and eye rupture. The skin disease manifests mostly as redness and in some fish as ulceration. An additional new finding was copepods found on the gills of some of these barramundi. This is not considered to be an issue of significance at this point but should be followed up and compared with future submissions. There is no evidence to date that Epizootic Ulcerative Syndrome (EUS), commonly known as red spot disease, is causing fish disease in Gladstone Harbour.

C. The barramundi kidney, spleen and particularly in the liver have significantly increased numbers of melanomacrophage centres (MMCs). This is potentially a bio-indicator of catabolic processes in the fish. The catabolic changes related to an increased number of MMCs, probably reflect the processing of an increased rate of red blood cell death and other deteriorated cells due to chronic skin and eye damage caused by Neobenedenia sp. or by the affected osmoregulation. An increase in the size and frequency of MMCs is a common histological change associated with either disease, starvation, reduced water quality or old age

D. Tissue samples from two submissions P11-74663 (X2 barramundi from China Bay) and (P11-74903 X1 barramundi from Port Alma) were sent to QHFSS for toxicology for organic and inorganic compounds. Tissue samples submitted for toxicology testing consisted of gill, liver and muscle from each animal. Samples are reported on a wet weight basis. For organic analytes (Attachment 5: Laboratory Reference SSP29151) the samples were analysed for organophosphorus type pesticides, organochlorine type pesticides, polychlorinated biphenyls (PCBs), synthetic pyrethroid insecticides, polyaromatic hydrocarbons (PAHs), selected herbicides, insecticides and fungicides. DDE p,p was the only analyte detected at a concentration of 0.014 mg/kg in sample P11-74663-10 muscle (China Bay). For inorganic analytes (Attachment 5: Reference 11PE14-156:Uta), metal analyses was conducted for: aluminium (Al), arsenic (As), barium (Ba), cadmium (Cd), chromium (Cr), nickel (Ni), copper (Cu), iron (Fe), zinc (Zn), silver (Ag) and selenium (Se). Lead (Pb) and mercury (Hg) were not selected for analysis because these metals were reported as below the limit of detection in the Western Basin Dredging and Disposal Project Environmental Impact Assessment (EIA) for Gladstone Harbour.

E. Skin abnormalities from non-barramundi fish species submitted for testing are generally mild and are not due to Neobenedenia sp. The whiting presented with localised skin erosion, inflammation and fibrosis. Trevally presented with localised epidermal hyperplasia and degeneration. Spangled emperor showed localised inflammation of skin and presence of abnormal cells. Spotted cod displayed localised skin inflammation and oedema. No bacterial, parasitic or fungal pathogens were found which could explain these skin diseases.

F. Sharks presented with scale pocket reddening, dermal haemorrhage and epidermal necrosis. The common histopathological changes present included congestion in the connective tissue immediately below the epidermal basement membrane, occasional haemorrhage into the epidermis and /or skin surface and a mild inflammatory cell infiltration into the outer layer of the dermis. Skin flukes were found on the surface of 11 of the samples of preserved skin from the sharks. Preliminary identification of these parasites as monogenean from the family of Microbothriidae.

G. A range of bacterial species were isolated from submitted fish species. The lack of distinct invasion or proliferation in the skin and muscle lesions suggests that in these cases the isolated bacteria are not pathogenic to these fish. Bacteria species isolated included Micrococcus spp., Moraxella spp., Proteus vulgaris, Pseudomonas spp., and Pseudomonas fluorescens.

H. The mud crab samples examined indicated an erosive shell disease. These signs are consistent with bacterial infection by Vibrio spp. which are organisms found in marine waters. Photobacterium (Vibrio) damaselae was isolated from the most recent mud crab submission. Prawns also showed evidence of shell erosion due to Vibrio spp. These bacteria species are opportunistic and proliferate on damaged shell to cause erosion due to their chitinolytic enzymes. Vibrio and Photobacterium are commonly isolated from healthy shellfish. A number of banana, king and tiger prawns showed evidence of endoparasite infection of immature stages of tapeworms. These parasites are not unexpected in wild prawn populations and would not have had a significant effect on prawn health.

Table 1: Interim summary of samples received as of 28 November 2011 in connection with Gladstone Harbour Fish Health investigation.

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
30.8.11 P11-74662	Whole barramundi fish x 4 Dead >24h on ice	Caught from Boyne River, Gladstone. Eye and skin lesions reported	1. Ocular pathology with corneal hyperaemia, haemorrhage, lymphocytic inflammation, oedema and erosion. Sclera, conjunctiva, choroid and iris tissues with hyperaemia/congestion and inflammation. 2. One fish with chronic, deep necrotic muscle. No evidence of EUS (red spot disease). 3. Absence of observable sediment on the gills. 4. Stomach empty. 5. Gills with myxosporean *Henneguya*-like spp. spore forming parasite, low to moderate level infestation. 6. Intestine with low infestation of larval nematodes in granulomas of the

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
			submucosa and muscle layers. 7. Heart with chronic low level infection due to *Cruoricola*-like spp. blood fluke eggs. 8. Liver – incidental coccidian-like parasite. 9. Kidney and spleen in some sections with heavy melanomacrophage centres (MMCs).
30.8.11 P11-74663	Whole barramundi fish x 2 Dead >24h on ice	Caught from China Bay, Gladstone. Eye lesions reported	1. Ocular pathology with corneal oedema and erosion, inflammation in choroid with eosinophilic granulocytes and congestion. 2. No evidence of EUS (red spot disease). 3. Stomach empty. 4. Absence of observable sediment on the gills. 5. Gills with myxosporean *Henneguya*-like spp. spore forming parasite, low level infestation. 6. Intestine with moderate infestation of nematodes in granulomas of the submucosa and muscle layers. 7. Heart with chronic low level infection due to *Cruoricola*-like spp. blood fluke eggs. 8. Toxicology testing results (Attachment 5: Report Reference SSP29151 and 11PE14-156:Uta)
5.9.11 P11-74796	Whole Moreton bay bug x 1 Dead >2 weeks, decomposed as not on ice in transit to lab.	Coordinates 23.28.70-151.30.50 23.22.80-151.26.20	1. Possible injury and secondary microbial invasion of the lesion with host response producing melanisation of the affected area. Gills fouling.
7.9.11 P11-74868	Whole barramundi fish x 1 Dead >24h on ice	Caught from South Tree Inlet, Gladstone. Eye lesions reported	1. Ocular pathology (keratitis, necrosis) and blindness (ruptured eye) caused by infestation with monogenean capsilid fluke *Neobenedenia* spp. 2. No evidence of EUS (red spot disease). 3. Absence of observable sediment on gills. 4. Stomach empty. 5. No evidence of bacterial disease. 6. Gill myxosporean *Henneguya*-like spp. at low level causing localised gill infection. 7. Intestine – low level larval nematode granulomas. 8. Heart and kidney with chronic low level infection due to *Cruoricola*-like spp. blood fluke eggs.

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
8.9.11 P11-74903	Whole barramundi fish x 1 Dead >24h on ice	Caught from Port Alma, Gladstone. Skin lesions reported	1. Skin-muscle – severe deep, ulcerative, necrotic and haemorrhagic lesions. Fungal pathogen (*Aphanomyces invadens/invaderis*) consistent with that which causes Epizootic Ulcerative Syndrome (EUS) or red spot disease observed in lesions. 2. Mixed growth of bacteria *Micrococcus* spp., *Moraxella* spp., *Proteus vulgaris, Pseudomonas* spp. from skin lesion. Spleen – no aerobic growth. These bacteria are not primary fish pathogens. 3. Absence of observable sediment on gills. 4. Toxicology testing results (Attachment 5: Report Reference SSP29151 and 11PE14-156:Uta).
9.9.11 P11-74922	Whole barramundi fish x 1 Dead >24h on ice	Caught from Calliope River, Gladstone. Eye and skin lesions reported	1. Ocular pathology with corneal erosion, generalised, severe, keratitis with focal haemorrhages and significant eosinophilic granulocyte infiltration. Oedema and separation of corneal stroma. This is caused by *Neobenedenia* spp. 2. Skin-muscle – severe deep, ulcerative, necrotic and haemorrhagic lesions. Light, mixed growth of *Moraxella* and *Pseudomonas* spp. isolated from skin. No aerobic growth from eye (aqueous humour, spleen). These bacterial are secondary contaminants of the skin lesion, not causative. 3. Gill, heart and kidney with chronic moderate to high level infection due to *Cruoricola*-like spp. blood fluke eggs. 4. No evidence of EUS (red spot disease). 5. Stomach empty. 6. Absence of observable sediment on the gills.
16.9.11 P11-75082	Mud crab female x1	Shelley Beach, Rockhampton Submitted by Biosecurity Queensland	1. Advanced post mortem degeneration. 2. Extensive erosion of the lateral carapace. 3. Three caudal sets of gill filaments were absent. 4. No obvious abnormalities were apparent.
16.9.11 P11- 75085	Whole spotted cod fish x 1 Dead <24h on ice	Caught from Rat Island, Gladstone. Skin lesions reported	1. Cataract in one eye, normal other eye. 2. Superficial skin abrasions and erythema one side of body. 3. No evidence of EUS (red spot disease). 4. No significant bacterial pathogen

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
			isolated. 5. Absence of observable sediment on the gills. 6. No evidence of systemic bacterial infection.
20.9.11 P11-75123	Whole prawn x 2 Dead on ice >24h	Gladstone Harbour	1. Erosive shell lesions and melanisation, subcuticular fibrosis and inflammation. 2. A mixed bacterial population isolated, including *Moraxella* spp., *Pseudomonas* spp., *Vibrio splendidus*. The shell lesions can be caused by *Vibrio*.spp (chitinolytic activity). 3. Absence of observable sediment on the gills.
20.9.11 P11-75124	Whole mud crab x2 Live	Caught from mouth of South Tree Inlet, Gladstone	1. Erosive shell lesions and melanisation (biofouling and bacteria), subcuticular inflammation. 2. A mixed bacterial population isolated, including *Shewanella putrefasciens, Vibrio* spp. *Vibrio splendidus I, Alcaligenes faecalis, Moraxella* spp., *Vibrio alginolyticus.* 3. Gill – sediment fouling and gregarine-like organisms. 4. Hepatopancreatic viral infection.
22.9.11 P11-75194	Spangled Emperor x1 Dead >24h on ice	Caught from Gladstone Reef	1. Protozoan-like infestation of skin. 2. Intestinal trematode cysts. 3. Absence of observable sediment on the gills. 4. No evidence of EUS (red spot disease). 5. No significant bacterial pathogen isolated.
27.9.11 P11-75286	Whiting x1 Dead >24h on ice	Caught outside exclusion zone. No specified location	1. Skin fibrosis. 2. No evidence of EUS (red spot disease). 3. No significant bacterial pathogen isolated.
4.10.11 P11-75412	Barramundi x 1 Dead > 72h	Caught from Calliope River near the hot water outlet on 30.9.11. Eye lesions reported. Submitted by Fisheries Queensland	1. Eye lesions – *Neobenedenia Sp.* flatworm isolated from body. 2. Absence of observable sediment on gills. 3. Kidney and spleen with severe melanomacrophage centres (MMCs). 4. Heart – moderate numbers of *Cruoricola*-like spp. Fluke eggs. 5. Spleen and muscle low level microsporidian parasite. 6. No evidence of EUS (red spot disease). 7. No significant bacterial pathogen isolated.
6.10.11 P11-75466	Barramundi x 1 Dead 61h	Witt Island. Submitted by	1. Redness of pelvic fins. 2. Kidney – severe levels of MMCs.

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
		Fisheries Queensland	3. Intestine – moderately level nematode granulomas. 4. No evidence of EUS (red spot disease). 5. No significant bacterial pathogen isolated.
6.10.11 P11-75467	Barramundi x 1 Dead 61h	Hamilton Point. Submitted by Fisheries Queensland	1. Eye – cornea with ulceration, inflammation, necrosis, hyperaemia, oedema. 2. Skin – mild redness ventral body, tail and pectoral fin bases. 3. Kidney – severe levels of black MMCs. 4. Liver – central area of hepatitis and fibrosis. 5. No evidence of EUS (red spot disease). 6. No significant bacterial pathogen isolated.
6.10.11 P11-75468	Barramundi x 1 Dead 61h	Tide Island. Submitted by Fisheries Queensland	1. Skin – a few superficial red marks on left side more than right side. 2. Kidney – severe levels of black MMCs. 3. Liver – quite large and moderate level of light brown MMCs. 4. Intestine with low level nematodes. 5. Low level *Cruoricola*-like spp. fluke. 6. No evidence of EUS (red spot disease). 7. No significant bacterial pathogen isolated.
7.10.11 P11-75528	Barramundi x 1 Dead > 48h	Hamilton Pt. Submitted by Fisheries Queensland	1. Skin- There was moderate hyperaemia of the ventral midline, but no skin ulcers. 2. There was slight hyperaemia of the right eye. 3. Liver – severe level brown MMCs. Spleen – moderate dark brown MMCs. Kidney – severe level black MMCs. 4. Spleen – *Pseudomonas fluorescens* bacteria isolated, but no evidence of systemic bacterial disease. 5. Low level intestinal nematode granulomas, *Henneguya.* 6. No evidence of EUS (red spot disease).
7.10.11 P11-75529	Barramundi x 1 Dead > 48h	Calliope River Submitted by Fisheries Queensland	1. Left eye ruptured and collapsed. Right eye – corneal opacity and peripheral hyperaemia. 2. Gills – moderate level *Henneguya* spp. and low level *Cruoricola*-like spp. fluke. 3. Kidney – severe MMCs, liver and spleen – moderate MMCs. 4. No evidence of EUS (red spot

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
			disease). 5. No significant bacterial pathogen isolated.
7.10.11 P11-75530	Barramundi x 1 Dead > 48h	Upper tidal limits of Boyne River Submitted by Fisheries Queensland	1. Skin – no skin ulcers were evident, but there was mild hyperaemia of the ventral midline. Skin – *Moraxella* spp. Spleen – no aerobic growth. Unidentified Gram positive motile bacillus from skin sample. 2. Eye – there was *hyperaema* of the left eye, as well as flocculent yellow material in the anterior chamber. 3. Spleen – moderate level dark-brown MMCs. Kidney – moderate level black MMCs. 4. Gills – low level *Henneguya* spp. plasmodia. 5. No evidence of sediment in the gills 6. No evidence of EUS (red spot disease).
7.10.11 P11-75531	Barramundi x 1 Dead > 48h	Upper tidal limit of Boyne River Submitted by Fisheries Queensland	1. Eye – cornea with keratitis, sloughing of cornea. 2. Spleen – severe black MMCs, Kidney – severe black MMCs. 3. Intestine – moderate level nematode granulomas. 4. No evidence of sediment in the gills 5. No evidence of EUS (red spot disease). 6. No significant bacterial pathogen isolated.
7.10.11 P11-75566	Barramundi x 14 Dead >48h	Tide island X1 South Trees Inlet Northside X1 Boyne River x1 Turkey Beach X1 Turkey Beach X1 Calliope River X1 Witt Island x1 Boyne River X1 Boyne River X1 Boyne River X 2 Boyne River X1 Calliope River X1 Hamilton Point x1 Submitted by Fisheries Queensland	1. 9 fish have eye lesions of varying severity from cloudiness, hyperaemia to rupture of the eye. These changes have been caused by *Neobenedenia* sp. capsilid flukes. 2. 7 fish have skin changes mainly with redness either localised or more regional in distribution, but no ulceration. 3. Fish were unsuitable for internal examination.
7.10.11 P11-75569	Shark (Lemon) x 1 Dead >48h	Wild Cattle Island Submitted by Fisheries Queensland	1. Skin – some areas of redness along flank and ventrally. Skin – separation of epidermis and scales from dermis and individual cell necrosis of epidermis.

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
			2. Minimal sediment detected on the gills.
7.10.11 P11-75571	Scat marine species x1 Dead >48h	Gladstone Harbour Submitted by Fisheries Queensland	1. Skin – small ulcer, middle top of head. Small red lesion left side. Ventral redness. 2. Dermatitis with focal lymphocytes and some necrotic inflammatory cells. Sloughing of mucous cells and epidermis. 3. No EUS granuloma formation. 4. This scat had a skin ulceration with a cellular inflammatory response. There was no evidence of a fungal infection, thus no Epizootic Ulcerative Syndrome.
7.10.11 P11-75573	Trevally x1 Dead >48h	Boyne River Bridge Submitted by Fisheries Queensland	1. Tail – redness. Pectoral fin – redness. Skin – localised epidermal hyperplasia and degeneration. No EUS granuloma formation. 2. Tail – redness. Pectoral fin – redness. 3. Two encysted metazoan parasite stages were detected in the dermal layer of the skin. 4. No evidence of sediment in the gills 5. No EUS granuloma formation.
13.10.11 P11-75654	Bull shark x 2 Dead >24h	Gladstone Harbour	1. Shark 1 – body with red marks at mid to caudal left side worse than right side. Anal bleeding. Skin with 2-3+ mucous mixed with blood. 2. Shark 2 – Body with ventral rash onto left side of abdomen and one area under mandible. 3. Skin – dermal haemorrhage and epidermal erosion and necrosis, scale pocket hyperaemia with inflammatory cells – eosinophilic granulocytes. Skin scale pocket hyperaemia, epithelial erosion. Dermis with haemorrhage (mild) and in a linear fashion. 4. No evidence of sediment in the gills. 5. No bacteria were isolated from the spleen. 6. Few encysted larval parasites in the wall of stomach, but not expected to have significant effect on shark health.
24.10.11 P11-75875	Formalin fixed tissues (gills, skin, internal organs), ectoparasites from 15 barramundi. Bacterial swabs for culture from 5	Boyne River Submitted by Fisheries Queensland	1. The range of fish samples demonstrate that an ophthalmitis and dermatitis was present 2. Histological examination of skin showed well established inflammation in the dermis and loss of the epidermis that rarely extended more deeply. Underlying hypodermal connective tissue and skeletal muscle appear normal.

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
	barramundi.		3. No bacteria were isolated from the spleen. 4. New finding on gills of barramundi from Gladstone harbour of one or two copepods attached to gill filaments 5. No detectable sediment on the gills. 6. Fresh samples forwarded to QHFSS for toxicology testing.
28.10.11 P11 - 76001	Mud crab, adult (Live)	Location to be advised by submitter	1. The significant change was the erosion of the shell creating a hole into the gill chamber. Although the opportunist bacterial pathogen *Photobacterium (Vibrio) damselae* was isolated from the haemolymph there were no significant internal lesions to indicate a septicaemia in this mud crab or that the haemocele had been opened to the external environment. 2. There was no detectable sediment on the gills. 3. No other infectious agents, or pathology indicating active infections, were present in this mud crab.
31.10.11 P11-76075	Flathead X2 (Fresh)	Spoil Ground Submitted by Queensland Fisheries	Held frozen.
1.11.11 P11-76076	Moreton Bay Bug (Fresh)	6 miles East of Facing Island Submitted by Queensland Fisheries	Held frozen.
02.11.11 P11-76085	Tiger Prawn [Fixed]	Spoil Ground by Fisheries Queensland	1. Other than the immature tapeworm parasite stages in the digestive organ, no significant changes were seen in this prawn. The cestode pleurocercoids are not unexpected. Previously seen a similar infestation in wild *Penaeus esculentus* caught off Bowen and it is likely they have no significant affect on the health of the prawn. 2. There was no detectable sediment on the gills.
31.10.11 P11-76086	Banana Prawn (Fixed)	Eastern leads off Tannum (Spoil Grounds) Submitted by Fisheries Queensland	1. The only significant change in this banana prawn was the one area in ventral thorax of chronic inflammation. This lesion suggests this prawn has a healthy inflammatory response. 2. A couple of endoparasites were detected in this prawn; a few immature tapeworm parasite stages in the digestive organ and some gregarines

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
			in the hindgut. These parasites are not unexpected in a wild prawn and would not have had a significant effect on its health. 3. There was no detectable sediment on the gills.
02.11.11 P11-76087	Eastern King Prawn [Fixed]	Spoil Grounds Submitted by Fisheries Queensland	1. There was an unusual proliferation of connective tissue and granular cells around the nerve cord in the thorax of this king prawn. This proliferation has been described from wild *Penaeus japonicus* in Queensland previously and does not seem to be associated with poor health although the actual significance is unknown. 2. A couple of endoparasites were detected in this prawn; some immature tapeworm parasite stages in the digestive organ and some gregarines in the hindgut. These parasites are not unexpected in a wild prawn and would not have had a significant effect on its health. 3. There was no detectable sediment on the gills.
02.11.11 P11-76088	Tiger Prawn [Fixed]	Tannum Submitted by Fisheries Queensland	1. Other than immature tapeworm parasite stages and a few unidentified parasites encysted in internal tissues, no significant changes were seen in this tiger prawn. These parasites are not unexpected in a wild prawn and would not have had a significant effect on its health. 2. There was no detectable sediment on the gills.
31.10.11 P11-76089	Barramundi X1 tissue [Fixed]	Calliope River Submitted by Fisheries Queensland	1. The proliferation of gill epithelial cells on some gill filaments of this barramundi appeared to be a response to lodgement of moderate numbers of *Cruoricola-like sp.* Blood fluke eggs were present in the gill tissue. There was no detectable sediment on the gills.
31.10.11 P11-76090	Fixed and Fresh Scallops	6-10 miles outside of Gladstone Harbour Submitted by Fisheries Queensland	1. Fresh samples forwarded to QHFSS for toxicology testing 15.11.11.
31.10.11 P11-76091	Sharks x 7 Fixed Swabs Parasites	Calliope River Submitted by Fisheries	1. Skin flukes on the surface of 5 of the samples of preserved skin. Preliminary identification of these as monogenean

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
		Queensland	from the family of Microbothriidae. 2. Due to possible autolysis of the skin prior to preservation it is not possible to know if the absence of epidermis in some samples was due to necrosis and sloughing or just a post mortem degeneration artefact. The bacteriology was not significant with a mixed over growth of contaminating bacteria. 3. No significant isolates from bacteriology. 4. Common changes present included congestion in the connective tissue immediately below the epidermal basement membrane, occasional haemorrhage into the epidermis and /or skin surface and a mild inflammatory cell infiltration into the outer layer of the dermis.
08.11.11 P11-76191	Scallops (Fresh)	Bundaberg Submitted by Fisheries Queensland	1. Fresh samples forwarded to QHFSS for toxicology testing 15.11.11.
08.11.11 P11-76192	Shark samples – Fixed parasites	Rhodds Harbour Submitted by Fisheries Queensland	1. The common change see in these shark skin samples was a mild to moderate inflammatory cell infiltrate into the dermis. 2. No detectable sediment on the set of gills examined. 3. Due to possible autolysis of the skin prior to preservation it is not possible to know if the absence of epidermis in some samples was due to necrosis and sloughing or just a post mortem degeneration artefact. 4. The mixed growth of bacteria was not considered significant.
09.11.11 P11-76200	Whiting –	Rhodds Harbour Submitted by Fisheries Queensland	Testing in progress.
09.11.11 P11-76202	Barramundi –	Gladstone Harbour Submitted by Fisheries Queensland	Testing in progress.
09.11.11 P11- 76203	Mullet –	Rhodds Harbour Submitted by Fisheries Queensland	Testing in progress.
09.11.11 P11-76205	Diamond Scale Mullet.	Turkey Beach Submitted by	Testing in progress.

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
		Fisheries Queensland	
14.11.11 P11-76276	Shark x 3	Fitzroy River Submitted by Fisheries Queensland	1. The skin fluke previously seen on the preserved skin samples from bull sharks (P11-76091) were again present on the skin from the bull shark and the pigeye shark: identification to be confirmed. 2. There were no significant bacteria identified from the submitted swabs. 3. There may have been some post mortem degeneration of these samples, it is not clear if the lifting and sloughing of parts of the epidermis of these sharks was an artefact or not. All three sharks had an inflammatory cell infiltration of the dermis. This was mild in the pigeye shark; moderate in the bull shark where there was also congestion in the connective tissue immediately below the epidermal basement membrane; and moderate to severe in the saw shark. In the saw shark the dermal inflammation extended into the subcutaneous skeletal muscle.
14.11.11 P11-76277	Shark x 1	Kolan River Bundaberg Submitted by Fisheries Queensland	1. The skin fluke previously seen on the preserved skin samples from bull sharks from the Calliope River (P11-76091) were present on this bull shark specimen; identification to be confirmed. 2. There were no significant bacteria identified from the submitted swab. 3. There was a mild superficial congestion and a mild inflammatory cell infiltration of the dermis of this bull shark.
14.11.11 P11-76278	Barramundi x 2 Fresh tissue, gill, liver – held frozen	Fitzroy River Submitted by Fisheries Queensland	Testing in progress.
14.11.11 P11-76279	Sand Whiting x 3 (Fixed) Gill muscle liver (fresh)	Kolan River Bundaberg Submitted by Fisheries Queensland	Testing in progress. Fresh samples forwarded to QHFSS for toxicology testing.
14.11.11 P11-76280	Sand Whiting x 2 muscle gill, liver (fresh)	7 mile Turkey Beach Submitted by Fisheries Queensland	Testing in progress. Fresh samples forwarded to QHFSS for toxicology testing.
14.11.11	Mullet x 2	Kolan River	Testing in progress.

Date received/ Case no.	Sample Type, Number, Condition	History	Interim Findings
P11-76281	Gill muscle liver (fresh)	Bundaberg Submitted by Fisheries Queensland	Fresh samples forwarded to QHFSS for toxicology testing.
14.11.11 P11-76282	Mullet x 2 Muscle gill liver (fresh)	7 mile (Turkey Beach) Submitted by Fisheries Queensland	Testing in progress. Fresh samples forwarded to QHFSS for toxicology testing.
21.11.11 P11-76412	Banana Prawns (fresh)	Gladstone Harbour Submitted by Fisheries Queensland	Testing in progress Fresh samples. forwarded to QHFSS for toxicology testing.
28.11.11 P11-76561	Barramundi X2 Muscle, liver gill (fresh)	Awoonga Dam Fitzroy River Submitted by Fisheries Queensland	Testing in progress. Fresh samples forwarded to QHFSS for toxicology testing.

ATTACHMENT 5

Gladstone Fish Toxicology Data Report 1

Toxicology analysis of chemicals and metals provided by Queensland Health and Forensic Scientific Services.

Queensland Government

CaSS | Forensic and Scientific Services
A CLINICAL AND STATEWIDE SERVICE

CERTIFICATE OF ANALYSIS

CLIENT: Biosecurity Sciences Laboratory
Building 12
39 Kessels Rd
Coopers Plains QLD 4108

ATTN: Roger Chong

Laboratory Reference	: SSP29151
Client Order Number	: -
Quote Number	: -
Client Project	: -
Client Batch Reference	: -
Date Received	: 14/10/2011
Date Commenced	: 18/10/2011
Laboratory Number/s	: 11KE8976-8984

CC: Stephen Were

Submitting Authority : DEEDI

Number of Samples : 9

Reason for Analysis : Environmental Investigation

Method/s of Analysis : 22276 – Analysis of Seafood for Organophosphorus, Organochlorine and Synthetic Pyrethroid Pesticides

Remarks : This method was also used for the determination of PAH's and PCB's.

: The stated Limits of Reporting vary due to the differing weights of sample available for analysis.

……
Scott Turner
Senior Chemist, Organics Laboratory
24/11/2011

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Organics Laboratory Number		11KE8976	11KE8977	11KE8978	11KE8979	11KE8980	11KE8981
Client Reference		P11-7466-8	P11-7466-9	P11-7466-10	P11-7466-11	P11-7466-12	P11-7466-13
Collected Date		-	-	-	-	-	-
Sample Description	Units	Gill	Liver	Muscle	Gill	Liver	Muscle
Organochlorine Pesticides by GCMS							
CHLORDANE cis	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CHLORDANE trans	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CHLORDENE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CHLORDENE EPOXIDE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CHLORDENE, 1-HYDROXY	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CHLORDENE, 1-OH-2,3-EPOXY	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DDD o,p	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DDD p,p	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DDE o,p	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DDE p,p	mg/kg	< 0.031	< 0.16	**0.014**	< 0.028	< 0.13	< 0.012
DDT o,p	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DDT p,p	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DICOFOL p,p bd	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DIELDRIN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
ENDOSULFAN alpha	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
ENDOSULFAN beta	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
ENDOSULFAN ETHER	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
ENDOSULFAN SULPHATE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
ENDRIN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
ENDRIN ALDEHYDE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
HCH-a	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
HCH-b	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
HCH-d	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
HEPTACHLOR	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
HEPTACHLOR EPOXIDE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
LINDANE (HCH-g)	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
METHOXYCHLOR	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
NONACHLOR cis	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
NONACHLOR trans	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
OXYCHLORDANE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012

Organics Laboratory Number	Units	11KE8982	11KE8983	11KE8984
Client Reference		P11-74903-2	P11-74903-1	P11-74903-3
Collected Date		-	-	-
Sample Description	Units	Gill	Liver	Muscle
Organochlorine Pesticides by GCMS				
CHLORDANE cis	mg/kg	< 0.74	< 0.059	< 0.025
CHLORDANE trans	mg/kg	< 0.74	< 0.059	< 0.025
CHLORDENE	mg/kg	< 0.74	< 0.059	< 0.025
CHLORDENE EPOXIDE	mg/kg	< 0.74	< 0.059	< 0.025
CHLORDENE, 1-HYDROXY	mg/kg	< 0.74	< 0.059	< 0.025
CHLORDENE, 1-OH-2,3-EPOXY	mg/kg	< 0.74	< 0.059	< 0.025
DDD o,p	mg/kg	< 0.74	< 0.059	< 0.025
DDD p,p	mg/kg	< 0.74	< 0.059	< 0.025
DDE o,p	mg/kg	< 0.74	< 0.059	< 0.025
DDE p,p	mg/kg	< 0.74	< 0.059	< 0.025
DDT o,p	mg/kg	< 0.74	< 0.059	< 0.025
DDT p,p	mg/kg	< 0.74	< 0.059	< 0.025
DICOFOL p,p bd	mg/kg	< 0.74	< 0.059	< 0.025
DIELDRIN	mg/kg	< 0.74	< 0.059	< 0.025
ENDOSULFAN alpha	mg/kg	< 0.74	< 0.059	< 0.025
ENDOSULFAN beta	mg/kg	< 0.74	< 0.059	< 0.025
ENDOSULFAN ETHER	mg/kg	< 0.74	< 0.059	< 0.025
ENDOSULFAN SULPHATE	mg/kg	< 0.74	< 0.059	< 0.025
ENDRIN	mg/kg	< 0.74	< 0.059	< 0.025
ENDRIN ALDEHYDE	mg/kg	< 0.74	< 0.059	< 0.025
HCH-a	mg/kg	< 0.74	< 0.059	< 0.025
HCH-b	mg/kg	< 0.74	< 0.059	< 0.025
HCH-d	mg/kg	< 0.74	< 0.059	< 0.025
HEPTACHLOR	mg/kg	< 0.74	< 0.059	< 0.025
HEPTACHLOR EPOXIDE	mg/kg	< 0.74	< 0.059	< 0.025
LINDANE (HCH-g)	mg/kg	< 0.74	< 0.059	< 0.025
METHOXYCHLOR	mg/kg	< 0.74	< 0.059	< 0.025
NONACHLOR cis	mg/kg	< 0.74	< 0.059	< 0.025
NONACHLOR trans	mg/kg	< 0.74	< 0.059	< 0.025
OXYCHLORDANE	mg/kg	< 0.74	< 0.059	< 0.025

Organics Laboratory Number		11KE8976	11KE8977	11KE8978	11KE8979	11KE8980	11KE8981
Client Reference		P11-7466-8	P11-7466-9	P11-7466-10	P11-7466-11	P11-7466-12	P11-7466-13
Collected Date		-	-	-	-	-	-
Sample Description	Units	Gill	Liver	Muscle	Gill	Liver	Muscle
Organophosphate Pesticides by GCMS							
AZINPHOS ETHYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
AZINPHOS METHYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
BROMOPHOS ETHYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CADUSAPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CARBOPHENOTHION	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CHLORFENVINPHOS E+Z	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CHLORPYRIFOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CHLORPYRIFOS ME	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CHLORPYRIFOS OXON	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
COUMAPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DIAZINON	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DIOXATHION	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
ETHION	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
ETHOPROPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
ETRIMIPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
FAMPHUR	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
FENCHLORPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
FENTHION ETHYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
FENITROTHION	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
MALATHION	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
METHIDATHION	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
MEVINPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PARATHION ETHYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PARATHION METHYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PIRIMIPHOS METHYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PHOSMET	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PROFENOPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PROTHIOPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PYRAZAPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
SULPROFOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TETRACHLORVINPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TERBUPHOS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012

Organics Laboratory Number	Units	11KE8982	11KE8983	11KE8984
Client Reference		P11-74903-2	P11-74903-1	P11-74903-3
Collected Date		-	-	-
Sample Description	Units	Gill	Liver	Muscle
Organophosphate Pesticides by GCMS				
AZINPHOS ETHYL	mg/kg	< 0.74	< 0.059	< 0.025
AZINPHOS METHYL	mg/kg	< 0.74	< 0.059	< 0.025
BROMOPHOS ETHYL	mg/kg	< 0.74	< 0.059	< 0.025
CADUSAPHOS	mg/kg	< 0.74	< 0.059	< 0.025
CARBOPHENOTHION	mg/kg	< 0.74	< 0.059	< 0.025
CHLORFENVINPHOS E+Z	mg/kg	< 0.74	< 0.059	< 0.025
CHLORPYRIFOS	mg/kg	< 0.74	< 0.059	< 0.025
CHLORPYRIFOS ME	mg/kg	< 0.74	< 0.059	< 0.025
CHLORPYRIFOS OXON	mg/kg	< 0.74	< 0.059	< 0.025
COUMAPHOS	mg/kg	< 0.74	< 0.059	< 0.025
DIAZINON	mg/kg	< 0.74	< 0.059	< 0.025
DIOXATHION	mg/kg	< 0.74	< 0.059	< 0.025
ETHION	mg/kg	< 0.74	< 0.059	< 0.025
ETHOPROPHOS	mg/kg	< 0.74	< 0.059	< 0.025
ETRIMIPHOS	mg/kg	< 0.74	< 0.059	< 0.025
FAMPHUR	mg/kg	< 0.74	< 0.059	< 0.025
FENCHLORPHOS	mg/kg	< 0.74	< 0.059	< 0.025
FENTHION ETHYL	mg/kg	< 0.74	< 0.059	< 0.025
FENITROTHION	mg/kg	< 0.74	< 0.059	< 0.025
MALATHION	mg/kg	< 0.74	< 0.059	< 0.025
METHIDATHION	mg/kg	< 0.74	< 0.059	< 0.025
MEVINPHOS	mg/kg	< 0.74	< 0.059	< 0.025
PARATHION ETHYL	mg/kg	< 0.74	< 0.059	< 0.025
PARATHION METHYL	mg/kg	< 0.74	< 0.059	< 0.025
PIRIMIPHOS METHYL	mg/kg	< 0.74	< 0.059	< 0.025
PHOSMET	mg/kg	< 0.74	< 0.059	< 0.025
PROFENOPHOS	mg/kg	< 0.74	< 0.059	< 0.025
PROTHIOPHOS	mg/kg	< 0.74	< 0.059	< 0.025
PYRAZAPHOS	mg/kg	< 0.74	< 0.059	< 0.025
SULPROFOS	mg/kg	< 0.74	< 0.059	< 0.025
TETRACHLORVINPHOS	mg/kg	< 0.74	< 0.059	< 0.025
TERBUPHOS	mg/kg	< 0.74	< 0.059	< 0.025

Organics Laboratory Number		11KE8976	11KE8977	11KE8978	11KE8979	11KE8980	11KE8981
Client Reference		P11-7466-8	P11-7466-9	P11-7466-10	P11-7466-11	P11-7466-12	P11-7466-13
Collected Date		-	-	-	-	-	-
Sample Description	Units	Gill	Liver	Muscle	Gill	Liver	Muscle
Synthetic Pyrethroids by GCMS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
BIFENTHRIN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
BIORESMETHRIN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CYFLUTHRIN isomers	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CYHALOTHRIN isomers	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CYPERMETHRIN isomers	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DELTAMETHRIN isomers	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
FENVALERATE isomers	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
FLUVALINATE isomers	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PERMETHRIN isomers	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PHENOTHRIN isomers	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PIPERONYL BUTOXIDE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TETRAMETHRIN isomers	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TRANSFLUTHRIN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Others Pesticides by GCMS							
BENALAXYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
BENDIOCARB	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
BITERTANOL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CHLOROTHALONIL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DEET	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
FIPRONIL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
FURALAXYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
METALAXYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
OXADIAZON	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PIRIMICARB	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PROCYMIDONE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PROPARGITE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PROPICONAZOL isomers	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PROPOXUR	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TEBUCONAZOLE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TETRADIFON	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TRIADIMEFON	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TRIADIMENOL ISOMERS	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
VINCLOZALIN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012

Organics Laboratory Number		11KE8982	11KE8983	11KE8984
Client Reference		P11-74903-2	P11-74903-1	P11-74903-3
Collected Date		-	-	-
Sample Description	Units	Gill	Liver	Muscle
Synthetic Pyrethroids by GCMS	mg/kg	< 0.74	< 0.059	< 0.025
BIFENTHRIN	mg/kg	< 0.74	< 0.059	< 0.025
BIORESMETHRIN	mg/kg	< 0.74	< 0.059	< 0.025
CYFLUTHRIN isomers	mg/kg	< 0.74	< 0.059	< 0.025
CYHALOTHRIN isomers	mg/kg	< 0.74	< 0.059	< 0.025
CYPERMETHRIN isomers	mg/kg	< 0.74	< 0.059	< 0.025
DELTAMETHRIN isomers	mg/kg	< 0.74	< 0.059	< 0.025
FENVALERATE isomers	mg/kg	< 0.74	< 0.059	< 0.025
FLUVALINATE isomers	mg/kg	< 0.74	< 0.059	< 0.025
PERMETHRIN isomers	mg/kg	< 0.74	< 0.059	< 0.025
PHENOTHRIN isomers	mg/kg	< 0.74	< 0.059	< 0.025
PIPERONYL BUTOXIDE	mg/kg	< 0.74	< 0.059	< 0.025
TETRAMETHRIN isomers	mg/kg	< 0.74	< 0.059	< 0.025
TRANSFLUTHRIN	mg/kg	< 0.74	< 0.059	< 0.025
Others Pesticides by GCMS				
BENALAXYL	mg/kg	< 0.74	< 0.059	< 0.025
BENDIOCARB	mg/kg	< 0.74	< 0.059	< 0.025
BITERTANOL	mg/kg	< 0.74	< 0.059	< 0.025
CHLOROTHALONIL	mg/kg	< 0.74	< 0.059	< 0.025
DEET	mg/kg	< 0.74	< 0.059	< 0.025
FIPRONIL	mg/kg	< 0.74	< 0.059	< 0.025
FURALAXYL	mg/kg	< 0.74	< 0.059	< 0.025
METALAXYL	mg/kg	< 0.74	< 0.059	< 0.025
OXADIAZON	mg/kg	< 0.74	< 0.059	< 0.025
PIRIMICARB	mg/kg	< 0.74	< 0.059	< 0.025
PROCYMIDONE	mg/kg	< 0.74	< 0.059	< 0.025
PROPARGITE	mg/kg	< 0.74	< 0.059	< 0.025
PROPICONAZOL isomers	mg/kg	< 0.74	< 0.059	< 0.025
PROPOXUR	mg/kg	< 0.74	< 0.059	< 0.025
TEBUCONAZOLE	mg/kg	< 0.74	< 0.059	< 0.025
TETRADIFON	mg/kg	< 0.74	< 0.059	< 0.025
TRIADIMEFON	mg/kg	< 0.74	< 0.059	< 0.025
TRIADIMENOL ISOMERS	mg/kg	< 0.74	< 0.059	< 0.025
VINCLOZALIN	mg/kg	< 0.74	< 0.059	< 0.025

Organics Laboratory Number		11KE8976	11KE8977	11KE8978	11KE8979	11KE8980	11KE8981
Client Reference		P11-7466-8	P11-7466-9	P11-7466-10	P11-7466-11	P11-7466-12	P11-7466-13
Collected Date		-	-	-	-	-	-
Sample Description	Units	Gill	Liver	Muscle	Gill	Liver	Muscle
Herbicides by GCMS							
AMETRYN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
ATRAZINE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
BROMACIL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
CARBARYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
DICLOFOP METHYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
FLUAZIFOP BUTYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
HALOXYFOP 2-EtOEt	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
HALOXYFOP METHYL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
METOLACHLOR	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
MOLINATE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
OXYFLUORFEN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PENDIMETHALIN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PROMETRYN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PROPANIL	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
PROPAZINE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
SIMAZINE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TERBUTRYN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TERBUTHYLAZINE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TRIALLATE	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
TRIFLURALIN	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012

Organics Laboratory Number		11KE8982	11KE8983	11KE8984
Client Reference		P11-74903-2	P11-74903-1	P11-74903-3
Collected Date		-	-	-
Sample Description	Units	Gill	Liver	Muscle
Herbicides by GCMS				
AMETRYN	mg/kg	< 0.74	< 0.059	< 0.025
ATRAZINE	mg/kg	< 0.74	< 0.059	< 0.025
BROMACIL	mg/kg	< 0.74	< 0.059	< 0.025
CARBARYL	mg/kg	< 0.74	< 0.059	< 0.025
DICLOFOP METHYL	mg/kg	< 0.74	< 0.059	< 0.025
FLUAZIFOP BUTYL	mg/kg	< 0.74	< 0.059	< 0.025
HALOXYFOP 2-EtOEt	mg/kg	< 0.74	< 0.059	< 0.025
HALOXYFOP METHYL	mg/kg	< 0.74	< 0.059	< 0.025
METOLACHLOR	mg/kg	< 0.74	< 0.059	< 0.025
MOLINATE	mg/kg	< 0.74	< 0.059	< 0.025
OXYFLUORFEN	mg/kg	< 0.74	< 0.059	< 0.025
PENDIMETHALIN	mg/kg	< 0.74	< 0.059	< 0.025
PROMETRYN	mg/kg	< 0.74	< 0.059	< 0.025
PROPANIL	mg/kg	< 0.74	< 0.059	< 0.025
PROPAZINE	mg/kg	< 0.74	< 0.059	< 0.025
SIMAZINE	mg/kg	< 0.74	< 0.059	< 0.025
TERBUTRYN	mg/kg	< 0.74	< 0.059	< 0.025
TERBUTHYLAZINE	mg/kg	< 0.74	< 0.059	< 0.025
TRIALLATE	mg/kg	< 0.74	< 0.059	< 0.025
TRIFLURALIN	mg/kg	< 0.74	< 0.059	< 0.025

Organics Laboratory Number		11KE8976	11KE8977	11KE8978	11KE8979	11KE8980	11KE8981
Client Reference		P11-7466-8	P11-7466-9	P11-7466-10	P11-7466-11	P11-7466-12	P11-7466-13
Collected Date		-	-	-	-	-	-
Sample Description	Units	Gill	Liver	Muscle	Gill	Liver	Muscle
POLYNUCLEAR AROMATIC HYDROCARBONS by GCMS							
Naphthalene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Acenaphthylene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Acenaphthene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Fluorene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Phenanthrene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Anthracene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Fluoranthene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Pyrene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Benz[a]anthracene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Chrysene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Benzo[b+k]fluoranthene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Perylene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Benzo[a]pyrene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Benzo[e]pyrene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Indeno[1,2,3-cd]pyrene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Dibenz[a,h]anthracene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
Benzo[ghi]perylene	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012

Note : Spike recovery for Anthracene and Benzo[a]pyrene were lower than expected.

Organics Laboratory Number		11KE8982	11KE8983	11KE8984
Client Reference		P11-74903-2	P11-74903-1	P11-74903-3
Collected Date		-	-	-
Sample Description	Units	Gill	Liver	Muscle
POLYNUCLEAR AROMATIC HYDROCARBONS by GCMS				
Naphthalene	mg/kg	< 0.74	< 0.059	< 0.025
Acenaphthylene	mg/kg	< 0.74	< 0.059	< 0.025
Acenaphthene	mg/kg	< 0.74	< 0.059	< 0.025
Fluorene	mg/kg	< 0.74	< 0.059	< 0.025
Phenanthrene	mg/kg	< 0.74	< 0.059	< 0.025
Anthracene	mg/kg	< 0.74	< 0.059	< 0.025
Fluoranthene	mg/kg	< 0.74	< 0.059	< 0.025
Pyrene	mg/kg	< 0.74	< 0.059	< 0.025
Benz[a]anthracene	mg/kg	< 0.74	< 0.059	< 0.025
Chrysene	mg/kg	< 0.74	< 0.059	< 0.025
Benzo[b+k]fluoranthene	mg/kg	< 0.74	< 0.059	< 0.025
Perylene	mg/kg	< 0.74	< 0.059	< 0.025
Benzo[a]pyrene	mg/kg	< 0.74	< 0.059	< 0.025
Benzo[e]pyrene	mg/kg	< 0.74	< 0.059	< 0.025
Indeno[1,2,3-cd]pyrene	mg/kg	< 0.74	< 0.059	< 0.025
Dibenz[a,h]anthracene	mg/kg	< 0.74	< 0.059	< 0.025
Benzo[ghi]perylene	mg/kg	< 0.74	< 0.059	< 0.025

Note : Spike recovery for Anthracene and Benzo[a]pyrene were lower than expected.

Organics Laboratory Number		11KE8976	11KE8977	11KE8978	11KE8979	11KE8980	11KE8981
Client Reference		P11-7466-8	P11-7466-9	P11-7466-10	P11-7466-11	P11-7466-12	P11-7466-13
Collected Date		-	-	-	-	-	-
Sample Description	Units	Gill	Liver	Muscle	Gill	Liver	Muscle
Polychlorinated Biphenyls(PCB's) by GCMS							
IUPAC 1 2-chlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 5 2,3-dichlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 18 2,2', 5-trichlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 28 2,4,4'-trichlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 31 2,4',5-trichlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 44 2,2',3,5'-tetrachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 52 2,2',5,5'-tetrachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 66 2,3',4.4'-tetrachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 77 3,3',4,4'-tetrachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 81 3,4,4',5-tetrachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 87 2,2',3,4,5'-pentachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 101 2,2',4,5,5'-pentachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 105 2,3,3',4,4'-pentachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 110 2,3,3',4',6-pentachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 114 2,3,4,4',5-pentachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 118 2,3',4,4',5-pentachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 123 2',3,4,4',5-pentachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 126 3,3',4,4',5-pentachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 138 2,2',3,4,4',5'-hexachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 141 2,2',3,4,5,5'-hexachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 149 2,2',3,4',5',6-hexachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 151 2,2',3,5,5',6-hexachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 153 2,2',4,4',5,5'-hexachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 156 2,3,3',4,4',5-hexachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 157 2,3,3',4.4',5'-hexachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 167 2,3',4,4',5,5'-hexachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 169 3,3',4,4',5,5'-hexachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 170 2.2',3,3',4,4',5-heptachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 180 2,2',3,4,4',5,5'-heptachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 183 2,2',3,4,4',5',6-heptachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 189 2,3,3',4,4'.5.5'-heptachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 194 2,2',3,3',4,4',5,5'-octachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012
IUPAC 206 2,2',3,3',4,4',5,5',6-nonachlorobiphenyl	mg/kg	< 0.031	< 0.16	< 0.012	< 0.028	< 0.13	< 0.012

Organics Laboratory Number		11KE8982	11KE8983	11KE8984
Client Reference		P11-74903-2	P11-74903-1	P11-74903-3
Collected Date		-	-	-
Sample Description	Units	Gill	Liver	Muscle
Polychlorinated Biphenyls(PCB's) by GCMS				
IUPAC 1 2-chlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 5 2,3-dichlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 18 2,2', 5-trichlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 28 2,4,4'-trichlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 31 2,4',5-trichlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 44 2,2',3,5'-tetrachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 52 2,2',5,5'-tetrachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 66 2,3',4.4'-tetrachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 77 3,3',4,4'-tetrachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 81 3,4,4',5-tetrachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 87 2,2',3,4,5'-pentachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 101 2,2',4,5,5'-pentachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 105 2,3,3',4,4'-pentachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 110 2,3,3',4',6-pentachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 114 2,3,4,4',5-pentachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 118 2,3',4,4',5-pentachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 123 2',3,4,4',5-pentachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 126 3,3',4,4',5-pentachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 138 2,2',3,4,4',5'-hexachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 141 2,2',3,4,5,5'-hexachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 149 2,2',3,4',5',6-hexachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 151 2,2',3,5,5',6-hexachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 153 2,2',4,4',5,5'-hexachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 156 2,3,3',4,4',5-hexachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 157 2,3,3',4.4',5'-hexachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 167 2,3',4,4',5,5'-hexachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 169 3,3',4,4',5,5'-hexachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 170 2.2',3,3',4,4',5-heptachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 180 2,2',3,4,4',5,5'-heptachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 183 2,2',3,4,4',5',6-heptachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 189 2,3,3',4,4'.5.5'-heptachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 194 2,2',3,3',4,4',5,5'-octachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025
IUPAC 206 2,2',3,3',4,4',5,5',6-nonachlorobiphenyl	mg/kg	< 0.74	< 0.059	< 0.025

Organics Laboratory Number		11KE8976	11KE8977	11KE8978	11KE8979	11KE8980	11KE8981
Client Reference		P11-7466-8	P11-7466-9	P11-7466-10	P11-7466-11	P11-7466-12	P11-7466-13
Collected Date		-	-	-	-	-	-
Sample Description	Units	Gill	Liver	Muscle	Gill	Liver	Muscle
Mass of Sample Analysed	g	6.5	1.3	16.8	7.1	1.6	16.1

Organics Laboratory Number		11KE8982	11KE8983	11KE8984
Client Reference		P11-74903-2	P11-74903-1	P11-74903-3
Collected Date		-	-	-
Sample Description	Units	Gill	Liver	Muscle
Mass of Sample Analysed	g	0.30	3.4	7.9

Queensland Government

Queensland **Health**

Forensic & Scientific Services

Enquiries to : Dr Ujang Tinggi
Telephone: (07) 3274 9144
Fax: (07) 3274 9186
Our Reference: 11PE148-156:UTa
Your Reference:

30 November 2011

TO: **Att:** Dr Roger Chong
Biosecurity Queensland
Department of Employment, Economic Development and Innovation
Health and Food Science Precinct
39 Kessels Rd, Coopers Plains, QLD 4108

Dear Sir,

AMENDED ANALYSIS REPORT

On 28/09/11 we received 9 samples of fish tissues from you for analysis.

Reason for submission

Determination of aluminium (Al), arsenic (As), barium (Ba), cadmium (Cd), chromium (Cr), nickel (Ni), copper (Cu), iron (Fe), zinc (Zn), silver (Ag), and selenium (Se).

Results of Analysis :

See Table 1 Elemental content of fish tissues on page 2.

Methodology:

The sample "as received" was homogenized and sub-sample was taken for analysis. The levels of heavy metals were analyzed by ICP-MS after microwave digestion (Method No. 12659). The analysis was carried out with appropriate standard reference materials as a quality control for the recoveries of elements, except for Cr, Ni, Ba, and Al where suitable reference materials are not available. However, the concurrent analysis of other elements gave acceptable recoveries which indicated method accuracy and precision.

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INTERNATIONAL CODE 61

Table 1. Elemental content (mg/kg, wet weight) of fish tissues

No.	Client ID	Description	Tissue	Elemental concentration (mg/kg)										
				Al	As	Ba	Cd	Cr	Ni	Cu	Fe	Zn	Ag	Se
1	P11-74663-1-009	Barramundi	Liver	<0.5	0.36	<0.1	0.03	<0.05	0.09	1.6	812	9.5	<0.1	0.57
2	P11-74663-2-0010	Barramundi	Muscle	<0.5	0.16	<0.1	<0.01	<0.05	<0.05	0.23	1.7	3.0	<0.1	0.20
3	P11-74663-3-0011	Barramundi	Gill	10.9	0.15	<0.1	<0.01	<0.05	<0.05	0.72	64	10	<0.1	0.32
4	P11-74663-4-0012	Barramundi	Liver	<0.5	0.50	<0.1	0.03	<0.05	<0.05	2.1	802	18	<0.1	0.78
5	P11-74663-5-0013	Barramundi	Muscle	<0.5	0.29	<0.1	<0.01	0.09	<0.05	0.25	3.1	3.1	<0.1	0.19
6	P11-74663-6-008	Barramundi	Gill	5.6	0.18	<0.1	<0.01	<0.05	<0.05	0.66	46	9.8	<0.1	0.27
7	P11-74903/1	Barramundi	Liver	<0.5	0.13	<0.1	0.05	<0.05	<0.05	3.4	203	14	<0.1	0.67
8	P11-74903/2	Barramundi	Gill	15.3	0.09	<0.1	<0.01	0.07	<0.05	0.74	43	12	<0.1	0.37
9	P11-74903/3	Barramundi	Muscle	<0.5	0.09	0.5	<0.01	<0.05	<0.05	0.42	3.8	4.2	<0.1	0.18

Note: The limits of reporting (LOR) for Fe and Al are 0.5 mg/kg. The LOR for Cr, Cu, Zn, As, Se and Ni are 0.05 mg/kg, and 0.1 mg/kg for Ba and Ag.

Yours faithfully,

U. Tinggi
(Analyst)

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