# A Fish Health Indicator for the 2019 Gladstone Harbour Report Card 

Final Project Report Project ISPO23-2018/19

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## Version history

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## Executive Summary

Fish are key biological indicators of environmental contamination in waterways including estuaries, as they are the dominant taxa by biomass, play a variety of important ecological roles, are continuously exposed throughout their life cycle, are readily identified and have high importance to the community.

When fish are exposed to contaminated water, they can be affected from the population level (numbers and diversity of fish species) down to biochemical impacts on single cells within individual fish. In 2018, Gladstone Healthy Harbour Partnership (GHHP) and the Fisheries Research and Development Corporation (FRDC) commissioned CQUniversity (CQUni) to investigate potential fish health indicators for the Gladstone Harbour Report Card and their suitability for adaptation across ports and estuaries of Northern Australia. The research identified and tested a range of potential indicators that were low-medium cost and complexity. The indicator that was found to be most suitable for the Report Card is a version of the Health Assessment Index (HAI), a composite metric that integrates observer evaluations of multiple organs and tissues. The premise of the HAI is that scores will cumulatively reflect the acute and chronic stressors present in the fish's environment, with poorer anatomical condition resulting in higher HAI scores, indicative of a more stressful environment.

In 2019, GHHP commissioned CQUni to continue to monitor fish health in Gladstone Harbour using methods developed in 2018 and provide scores and grades for the Gladstone Harbour Report Card.

Sampling was conducted across Gladstone Harbour and at two reference sites, in Spring 2018 and Autumn 2019. The primary aim of sampling was to collect the target fish taxa identified by GHHP and during the 2018 research project as priorities for further analysis: barramundi (Lates calcarifer), large mullet (sea mullet Mugil cephalus and diamondscale mullet Liza vaigiensis), barred javelin (Pomadasys kaakan) and blue catfish (Neoarius graffei). Bream (pikey bream Acanthopagrus pacificus and yellowfin bream Acanthopagrus australis) are also of interest to GHHP and were retained for laboratory analysis when caught.

During the two sampling events a total of 568 fish from 45 species were caught across Gladstone Harbour and both reference sites. The species that were caught at the most sites were: barred javelin, blue catfish, diamondscale mullet, blue threadfin, barramundi and sea mullet. The species caught in highest numbers were barred javelin, blue catfish, sea mullet, barramundi, diamondscale mullet and blue threadfin (Eleutheronema tetradactylum, not a target species).

All fish were measured, weighed, checked for abnormalities and released, except for target species which were humanely killed for further analysis. In total, 246 of the five target species groups were retained for health assessment ( 223 from Gladstone Harbour and 23 from reference sites).

HAI was calculated for each of the 223 fish from Gladstone Harbour that were assessed in Spring 2018 and Autumn 2019, by scoring and summing gross pathology scores for the following measures: skin, eyes, fins, gills, spleen, kidney, hindgut, liver, and parasite load. HAI is designed to be a used as a summed average for a sample population. Using this method, the Gladstone Harbour-wide HAI results (for nine measures) have been determined, by species.

| Taxa / <br> Measure | Barramundi <br> $\mathbf{( n = 2 3 )}$ | Bream <br> $(\mathbf{n}=\mathbf{9 )}$ | 1.74 | 1.11 | Barred Javelin <br> $\mathbf{( n = 6 3 )}$ |
| :--- | :--- | :--- | :--- | :--- | :--- |
| Skin | 2.61 | 0 | Blue Catfish <br> $\mathbf{( n = 4 8 )}$ | Mullet <br> $(\mathbf{n}=\mathbf{8 0})$ |  |
| Eyes | 0.43 | 0.32 | 1.88 | 0.75 |  |
| Fins | 0 | 0 | 0.48 | 0 | 1.13 |
| Gills | 1.30 | 0 | 0.63 | 1.49 | 1.00 |
| Spleen | 0 | 0 | 0 | 0.63 | 0.38 |
| Kidney | 1.30 | 0 | 0 | 1.25 | 2.63 |
| Hindgut | 13.91 | 0 | 0 | 9.38 | 5.63 |
| Liver | 14.17 | 13.33 | 0.32 | 0.21 | 0.50 |
| Parasites | 3.33 | 8.73 | 9.38 | 6.88 |  |
| HAI score | 35.22 | 23.33 | 0 | 0 | 4.00 |

Using a benchmark score of an average HAI of 10, and a pilot worst case scenario (WCS) score of an average HAI of 70 , example HAI scores and grades were calculated using a distance from the benchmark method. Scores and grades have been calculated using all data from Spring 2018 and Autumn 2019. Using GHHP's grading scale, grades for each species group were calculated, and an overall harbour score and grade determined by averaging the scores of the five species groups.

| Taxa / <br> Measure | Barramundi | Bream | Barred <br> Javelin | Blue Catfish | Mullet |
| :--- | :--- | :--- | :--- | :--- | :--- |
| Taxa score | Grade C <br> Score 0.58 | Grade B <br> Score 0.78 | Grade B <br> Score 0.77 | Grade C <br> Score 0.60 | Grade B <br> Score 0.73 |
| Overall <br> Harbour score | Grade B <br> Score 0.69 |  |  |  |  |

The primary considerations when determining confidence in HAI scores for 2018-19 are sample size and potential for interference by ecological characteristics of each species group.

Sample sizes of barramundi $(n=23)$ and bream $(n=9)$ were relatively low. Barramundi are also a particularly mobile fish species with tagging evidence of movements across many hundreds of kilometres. This means that a barramundi caught in Gladstone Harbour may have moved from elsewhere.

For these reasons, the confidence in scores for barramundi and bream are lower than for the other three species groups. Substantial numbers of barred javelin ( $n=63$ ), blue catfish ( $n=48$ ) and mullet ( $\mathrm{n}=80$ ) contributed to the scores for these species, providing greater confidence that the samples are representative of the wider population.

Based on the results of the 2018 pilot sampling year, seven recommendations have been provided for GHHP's consideration.

Recommendation 1: GHHP continues to monitor HAI of fish in Gladstone Harbour and at least one reference site.

Recommendation 2: Pilot baselines and methods should be reviewed if more localised information becomes available.

Recommendation 3: GHHP continues to monitor measurements required to calculate Fulton's K, HSI and GSI, to collate a long-term dataset.

Recommendation 4: If new research suggests some fluctuating asymmetry measures may be useful fish health indicators, these could be considered for future application in the report card.

Recommendation 5: GHHP considers testing for bioaccumulation of metals and other toxicants in collected fish tissue samples.

Recommendation 6: GHHP continues to conduct regionally stratified fish sampling across Gladstone Harbour.

Recommendation 7: GHHP continues to sample at least one reference site, at least once a year.

## Introduction

Fish are well recognised globally as key biological indicators of pollution in freshwater and marine environments, as they are continuously exposed to water-borne contaminants providing a direct measure of ecological consequences, are dominant taxa in terms of biomass, are relatively long-lived so the impacts of pollution accumulate over longer periods, and play various important ecological roles including within food webs (Van der Oost et al., 2003). Most fish species can be quickly identified in the field, even by non-experts, and there are relatively few species in comparison to invertebrates (Pidgeon, 2004), and the high socio-economic importance of fish generates a positive public response to environmental management. One of the challenges of using fish as biological indicators is their high mobility (Whitfield \& Elliot, 2002), which means species selection relative to the regional scale of the study is important.

In 2018, Gladstone Healthy Harbour Partnership (GHHP) and the Fisheries Research and Development Corporation (FRDC) commissioned an investigation into potential fish health indicators for the Gladstone Harbour Report Card and suitable for adaptation across ports and estuaries of Northern Australia. The research identified and tested a range of potential indicators that were lowmedium cost and complexity (Flint et al., 2018). Individual fish health was defined as structural and morphological health and functioning in terms of the physiology of the organism (Whitfield \& Elliott, 2002). The indicator that was found to be most suitable for immediate implementation in the Report Card is a version of the Health Assessment Index (HAI), first developed by Adams et al. (1993), and subsequently widely used and adapted, including by the Queensland Government during fish health investigations in Gladstone Harbour (Wesche et al., 2013). The HAI is a composite metric that integrates observer evaluations of parasite load as well as the condition of multiple organs and tissues, including skin, eyes, fins, gills, spleen, kidney, hindgut, and liver. The premise of the HAI is that scores will cumulatively reflect the acute and chronic stressors present in the fish's environment, with poorer anatomical condition resulting in higher HAI scores, indicative of a more stressful environment.

In 2019, GHHP continued the research project to pilot the fish health indicator in the 2019 Gladstone Harbour Report Card. The objectives of the 2019 research project were:

1. To continue to monitor fish health in Gladstone Harbour using the data collection and statistical methods developed in 2018; and
2. To provide fish health report card scores and grades for the 2019 Gladstone Harbour Report Card.

## Methods

## Permits and approvals

The following permits and approvals are in place for this research:

- General Fisheries Permit (Queensland Department of Agriculture and Fisheries; Permit Number 196040)
- Animal Ethics Approval (CQUniversity Animal Ethics Committee; Approval Number 20969)
- Authorisation for research in the Great Barrier Reef Marine Park (Approval Number G18/03029)
- Field Work Risk Assessment (CQUniversity OHS Unit)


## Sampling design

Sampling was conducted across Gladstone Harbour (Figure 1) and at a reference site at Baffle Creek (Figure 2) in both Spring 2018 and Autumn 2019, and also at Stanage Bay in Spring 2018 (Figure 3), see also Appendix 1. The sampling strategy in Gladstone Harbour was developed during 2018 and 2019 to achieve an approximately even spread of fish catch and effort between the northern, central and southern areas of the harbour, focusing on inshore and estuarine environments. The selection of sites, including reference sites, was described in detail in the 2018 research report (Flint et al., 2018).

The primary aim of sampling was to collect the target fish taxa identified by GHHP and during the 2018 research project (Flint et al., 2018) as priorities for further analysis: barramundi (Lates calcarifer), large mullet (sea mullet Mugil cephalus and diamondscale mullet Liza vaigiensis), barred javelin (Pomadasys kaakan) and blue catfish (Neoarius graffei). Bream (pikey bream Acanthopagrus pacificus and yellowfin bream Acanthopagrus australis) are also of interest to GHHP, due to their recreational fishing value, and were retained for analysis when caught.

Understanding the mobility of fish in Gladstone Harbour is an important consideration that was taken into account when confirming target species. Inshore and estuarine fish tagging studies have shown that some fish species including barramundi may travel long distances between capture and recapture events (e.g. Moore and Reynold (1982); Russell and Garrett (1988)). However some fish, including barramundi, may also stay resident in an area for prolonged periods (e.g. Russell and Garrett (1988); Meynecke, Poole, Werry, and Lee (2008)). Fish release and recapture tagging data for Stanage Bay, Gladstone and Baffle Creek were provided to CQUni by Infofish Australia and used to assess adult home ranges of potential target fish species (see Flint et al., 2018, Appendix 1). Because adult fish are generally highly mobile, it is more biologically relevant to pool data across the harbour instead of attempting to score fish health within each GHHP water quality zone.

## Field sampling methods

Field collections of fish were undertaken using $3 \times 50 \mathrm{~m}$ long gill nets with stretched mesh sizes 4.5", $6^{\prime \prime}$ and $8^{\prime \prime}$. A fourth gill / ring net of 110 m length, $2.13^{\prime \prime}$ stretched mesh size was used at some sites to supplement catch. Gears were deployed in areas and at times when the chances of catching these target species were maximised, and bycatch minimised.

Field sampling was undertaken during Spring 2018 (September / October) and Autumn 2019 (April) (Appendix 2). Step-by-step details of sampling procedures are described in Flint et al. (2018). In summary, at each sampling location nets were deployed, details of deployment (including time and
location) were recorded as well as physicochemical measurements (including temperature ( ${ }^{\circ} \mathrm{C}$ ), dissolved oxygen (\% and $\mathrm{mg} / \mathrm{L}$ ), electrical conductivity ( $\mu \mathrm{S} / \mathrm{cm}$ ), pH , turbidity (NTU), total dissolved solids (TDS; mg/L), oxidation reduction potential (ORP; mV) and salinity (ppt), see Appendix 2). Nets were soaked for approximately 30 minutes during each deployment, and several deployments of nets occurred at each site throughout each approximately 10-hour long sampling day. Depending on catch rates and travel times, either one or two different sites were sampled each day.

Captured fish were assigned a unique identifier code and either processed immediately or placed into an aerated swim tank to be kept alive until on-board processing. Teleost fish were photographed, measured and weighed, and the skin, fins and eyes were examined for abnormalities, parasites, lesions or erosion on board. Cartilaginous fishes (sharks and rays) were recorded and photographed but were not handled except to ensure their safe removal from the net and live release. Non-target fish were released alive, while target species were retained at each site and euthanized for laboratory analysis. Immediately following euthanasia, gill arch samples were collected and fixed in $10 \%$ formalin. Non-target fish that died during capture ( $n=24$ ) were also retained for future laboratory analysis. All retained fish were individually bagged with their unique identifier tag and placed in an ice slurry for return to the laboratory as soon as possible on the same day.


Figure 1: Spring 2018 (red) and Autumn 2019 (blue) sampling locations across Gladstone Harbour.


Figure 2: Spring 2018 (red) and Autumn 2019 (blue) sampling locations within the Baffle Creek reference site.


Figure 3: Spring 2018 (red) sampling locations within the Stanage Bay reference site.

## Laboratory methods

Retained fish from all sites were returned to the lab at CQUniversity's Gladstone Marina Campus for same day mid-level pathological examination as described by Cowled (2016). Pathological examination also included the dissection of organs and fixation in $10 \%$ formalin for later histopathological analysis, if required.

Each fish was dissected by a team of two researchers, using the following protocol:

1. Remove fish from icy slurry and extract from sample bag. Retain fish identification label. Set aside contents of sample bag in a plastic container for parasite analysis.
2. Take photographs of whole fish (both sides), each fin, each eye, each gill (operculum lifted open). Include fish identification label in every photo. Throughout dissection, photograph any abnormalities in the same manner.
3. Measure and weigh the fish. Lab weights are more accurate than field weights as a more accurate balance can be used on the flat, motionless lab benches (in comparison to on a boat).
4. Measure the diameter of each eye of the fish using stainless steel Vernier callipers. Examine the eye and record any abnormalities, redness, cloudiness or other damage.
5. Examine the skin and fins and record any abnormalities, lesions or erosion. Record general condition of the fish and abdomen. Expose a portion of muscle by removing skin using a stainless steel scalpel and record any abnormalities.

- Excise a 1 cm cube of skin/muscle and place in a vial containing $10 \%$ formalin.
- Excise a sample of muscle and place in a vial to freeze.

6. Remove the operculum (gill cover) using stainless steel scissors or scalpel and examine the gills. Record any abnormalities. Remove the non-dissected gill (one side was dissected on board the boat, as described above in field methods) and set aside in a petri dish of seawater for parasite analysis.

- 1 cm gill sample taken on boat immediately after fish death, placed in a vial containing $10 \%$ formalin.
- Excise remaining gill filaments from the same side as gill sample was taken, and place in a vial to freeze.

7. Open the body cavity of the fish by cutting anteriorly along the length of the fish from the vent towards the head, using stainless steel filleting knives, scissors and snips. Smaller fish were opened using a stainless steel scalpel.
8. Check the skin, gills, viscera and organs for visible parasites, and record scores for the Health Assessment Index (Table 1).
9. Expose organs and examine in line with standard protocols required for the Health Assessment Index (Table 1). Record assessments and/or scores for liver, kidney, gonad, spleen, heart, mesentary fat, hindgut and bile. Dissect out pyloric caeca and intestines and set aside in a plastic container of saline solution for parasite analysis.

- Weigh liver
- Weigh gonad
- Excise a 1 cm cube of liver and kidney and place in a vial containing $10 \%$ formalin.
- Excise a sample of liver, kidney, spleen and gonad, and place in a vial to freeze.

10. Freeze carcass of fish for later disposal. Thoroughly clean bench and instruments before commencing next dissection.

Table 1: Variables and substituted values used in the Health Assessment Index for this project (source: Wesche et al., 2013).

| Variable | Variable condition | Field Designation | Substituted Value |
| :---: | :---: | :---: | :---: |
| Fins | No active erosion | F0 | 0 |
|  | Light active erosion | F1 | 10 |
|  | Severe active erosion | F2 | 20 |
| Spleen | Normal: black, very dark red or red | B | 0 |
|  | Normal: granular, rough appearance | G | 0 |
|  | Nodular, containing fistulas or nodules | D | 30 |
|  | Enlarged | E | 30 |
|  | Other: aberration not fitting any above | OT | 30 |
| Hindgut | Normal, no inflammation or reddening | 0 | 0 |
|  | Slight inflammation or reddening | 1 | 10 |
|  | Moderate inflammation or reddening | 2 | 20 |
|  | Severe inflammation or reddening | 3 | 30 |
| Kidney | Normal: firm, dark, flat | N | 0 |
|  | Swollen: enlarged or swollen | S | 30 |
|  | Mottled: gray discolouration | M | 30 |
|  | Granular in appearance and texture | G | 30 |
|  | Urolithiasis or nephrocalcinosis | U | 30 |
|  | Other: aberration not fitting any above | OT | 30 |
| Skin | Normal: no aberration | SK0 | 0 |
|  | Mild skin aberrations | SK1 | 10 |
|  | Moderate skin aberrations | SK2 | 20 |
|  | Severe skin aberrations | SK3 | 30 |
|  | Extensive redness as a rash. Scales intact | SK4 | 40 |
| Liver | Normal: solid red or light red color | A, B | 0 |
|  | 'Fatty' liver, 'coffee with cream' colour | C | 30 |
|  | Nodules or cysts in liver | D | 30 |
|  | Focal discolouration | E | 30 |
|  | General discolouration | F | $30$ |
|  | Other: deviation not fitting any above | OT | 30 |
| Eyes | No aberration, good, clear eyes | E0 | 0 |
|  | Fresh haemorrhage (eg net damage) | EOa | 0 |
|  | Opaque eye (one or both) | E1 | 30 |
|  | Cloudy and swollen, red or haemorrhaging | E2 | 30 |
|  | Ruptured (one or both) | E3 | 30 |
| Gills | Normal: no apparent aberrations | N | 0 |
|  | Frayed, ragged appearance | F | 30 |
|  | Clubbed, swelling of tips | C | 30 |
|  | Marginate: light discoloured margin | M | 30 |
|  | Pale, very light colour | P | 30 |
|  | Other | OT | 30 |
| Parasites | No observed parasites | P0 | 0 |
|  | Few observed parasites | P1 | 10 |
|  | Moderate parasite infestation | P2 | 20 |
|  | Numerous parasites | P3 | 30 |

## Calculating fish condition measures

Health Assessment Index (HAI, original method developed Adams et al. (1993); modified by Wesche et al. (2013)) scores for the organs of each fish were recorded based on the gross pathological data collected during fish dissections. Total HAl score for each individual fish was calculated (sum of all organ scores) and then the average of the scores was calculated for each fish taxa, across the harbour. Barramundi, blue catfish and barred javelin are reported as individual species. The species groups bream and mullet both include two species, pooled due to their similar ecological characteristics and to allow for higher sample sizes.

Other fish condition measures including Fulton's condition factor ( K ), Hepatosomatic index (HSI), and Gonadosomatic index (GSI), were opportunistically calculated for each fish. Calculations used were as follows:

Fulton's condition factor:
$K=100 *\left(W / L^{3}\right)$
where: $\mathrm{W}=$ wet body weight (g); L = total length (cm)
Hepatosomatic index:
$H S I=100^{*}(H / W)$
where: $\mathrm{H}=$ wet liver weight (g); $\mathrm{W}=$ wet body weight (g)
Gonadosomatic index:
GSI $=100^{*}(\mathrm{G} / \mathrm{W})$
Where: $\mathrm{G}=$ wet gonad weight (g); $\mathrm{W}=$ wet body weight (g)

## Statistical analytical methods

Results for each fish health measure were graphed to visually compare differences between seasons and species.

Formal statistical tests to compare fish health measures between seasons were done for each target species. Analyses were done using PERMANOVA (Permutational Analysis of Variance, conducted in PRIMER 7 + PERMANOVA software package), which is a non-parametric approach that closely approximates standard parametric analysis of variance when considering univariate data (as used herein), but is a statistical method that accommodates uneven replication and is robust to departures from non-normality of data.

## Results

## Fish catches

During the Spring 2018 and Autumn 2019 sampling events a total of 568 fish from 45 species were caught across Gladstone Harbour and the reference sites (Table 2). The species that were caught at the most sites were: barred javelin, blue catfish, diamondscale mullet, blue threadfin, barramundi and sea mullet. The species caught in highest numbers were barred javelin, blue catfish, sea mullet, barramundi, diamondscale mullet and blue threadfin (Eleutheronema tetradactylum, not a target species) (Figure 4). In total, 246 of the five target species groups were retained for health assessment, from all sampling sites.

Table 1: Fish species (listed by common name) and abundance at Gladstone Harbour (divided by GHHP zones) and two reference sites. Site R2 (Stanage Bay) and zones 3, 12 and 13 were not sampled in Autumn 2019 due to a change in sampling strategy. White $=0$; blue $=1-5$; orange $=6-10 ;$ green $=$ 10+ specimens. Common names of target species retained for further analysis are shaded grey. Species names provided in Appendix 3. Site R1 = Baffle Creek; R2 = Stanage Bay.




Figure 4: Total number of each fish species caught in Spring 2018 and Autumn 2019 combined.

## Fish condition measures

The average Health Assessment Index scores of fish species, across all sites, varied between seasons (Figure 5). The HAI score is scored as a subtractive measure, such that a score of 0 is ideal (all assessed organs appear normal) and higher scores equate to more abnormalities (up to a maximum score of 270). The lowest (best) scores were recorded for diamondscale mullet and blue catfish caught in Spring 2018, and the highest (worst) scores were recorded for blue catfish in Autumn 2019, sea mullet in both seasons, and barramundi in Spring 2018. Permanova analysis (Appendix 4) revealed that barred javelin was the only species for which the difference between seasons was significant $(P=0.015)$ to the $p<0.05$ level. While there was a visible difference in scores for blue catfish, this species was added as a target species for the Autumn 2019 sampling event so the number of blue catfish analysed varied greatly between seasons ( $n=3$ in Spring 2018 vs $n=47$ in Autumn 2019), so the difference in scores was not significant.


Figure 5: Health Assessment Index scores by species and season. White bars = Spring 2018. Black bars = Autumn 2019. "Bream" includes both pikey bream and yellowfin bream. An '*' indicates PERMANOVA detected a significant difference between sampling seasons for that species. Sample numbers, Spring 2018: barramundi $n=17$, barred javelin $n=31$, blue catfish $n=3$, bream $n=6$, diamondscale mullet $n=18$, sea mullet $n=13$. Sample numbers, Autumn 2019: barramundi $n=11$, barred javelin $n=36$, blue catfish $n=47$, bream $n=3$, diamondscale mullet $n=4$, sea mullet $n=57$.

Fulton's condition factor (K) did not vary greatly between seasons, and the only significant difference was for blue catfish (Figure 6; $\mathrm{P}=0.045$ ), although the result needs to be treated with caution due to the difference in sample size between seasons.

Hepatosomatic index (HSI) was higher in bream caught in Spring 2018 than Autumn 2019 (Figure 7), but the difference was not significant. The only significant difference in HSI between seasons was recorded for sea mullet, which showed significantly higher HSI scores in Spring 2018 than Autumn 2019 ( $P=0.01$ ).


Figure 6: Fulton's Condition factor (K) scores by species and season. White bars = Spring 2018. Black bars = Autumn 2019. "Bream" includes both pikey bream and yellowfin bream. An '*' indicates PERMANOVA detected a significant difference between sampling seasons for that species. Sample numbers, Spring 2018: barramundi $n=17$, barred javelin $n=31$, blue catfish $n=3$, bream $n=6$, diamondscale mullet $n=18$, sea mullet $n=13$. Sample numbers, Autumn 2019: barramundi $n=11$, barred javelin $n=36$, blue catfish $n=47$, bream $n=3$, diamondscale mullet $n=4$, sea mullet $n=57$.


## Species

Figure 7: Hepatosomatic Index (HSI) scores by species and season. White bars = Spring 2018. Black bars = Autumn 2019. "Bream" includes both pikey bream and yellowfin bream. An '*' indicates PERMANOVA detected a significant difference between sampling seasons for that species. Sample numbers, Spring 2018: barramundi $n=17$, barred javelin $n=31$, blue catfish $n=3$, bream $n=6$, diamondscale mullet $n=18$, sea mullet $n=13$. Sample numbers, Autumn 2019: barramundi $n=11$, barred javelin $n=36$, blue catfish $n=47$, bream $n=3$, diamondscale mullet $n=4$, sea mullet $n=57$.

Average GSI of female barramundi was higher for females caught in Spring than Autumn (Figure 8A) which is not surprising as barramundi spawn in early summer. The difference could not be statistically tested as only one female was caught in each sampling period. There was no significant difference in GSI of male barramundi between seasons. Similarly, there was no significant seasonal difference in GSI of male or female barred javelin (Figure 8B), diamondscale mullet (Figure 8C) or sea mullet (Figure 8D). However, GSI of female blue catfish (Figure 9A) and bream (Figure 9B) differed significantly between seasons ( $P=0.001$ and $P=0.26$, respectively).


Figure 8: Gonadosomatic Index (GSI) scores of male and female fish by season. White bars = female. Black bars = male. A) Barramundi. Samples numbers Spring 2018: male $n=16$, female $n=1$; Autumn 2019: male $n=8$, female $n=1$. B) Barred javelin. Sample numbers Spring 2018: male $n=4$, female $n$ = 27; Autumn 2019: male $n=8$, female $n=27$. C) Diamondscale mullet. Sample numbers Spring 2018: male $n=3$, female $n=15$; Autumn 2019: male $n=1$, female $=3$. D) Sea mullet. Sample numbers Spring 2018: male $n=1$, female $n=12$; Autumn 2019: male $n=6$, female $=48$.


Figure 9: Gonadosomatic Index (GSI) scores of male and female fish by season. White bars = female. Black bars = male. "Bream" includes both pikey bream and yellowfin bream. An '*' indicates PERMANOVA detected a significant difference between sampling seasons for that species. A) Blue catfish. Sample numbers Spring 2018: male $n=1$, female $n=1$; Autumn 2019: male $n=10$, female = 33. B) Bream. Sample numbers Spring 2018: male $n=1$, female $n=4$; Autumn 2019: male $n=0$, female $n=3$.

## 2019 Fish Health Indicator Results for Gladstone Harbour

In 2018, HAI was identified as the most appropriate fish health indicator for immediate implementation in the Gladstone Harbour Report Card (Flint et al., 2018). The metric requires gross pathological analysis during dissection and produces a composite metric that integrates evaluations of the condition of multiple organs and tissues. The premise of the index is that scores will cumulatively reflect the acute and chronic stressors present in the fish's environment, with poorer anatomical condition resulting in higher HAI scores and thus indicative of a more stressful environment. The version of the HAI used in this study was also used by Wesche et al. (2013) during the fish health investigation in Gladstone Harbour in 2011-2012.

## Measures and baselines

HAI was calculated for each of the 223 fish from Gladstone Harbour that were assessed in Spring 2018 and Autumn 2019, by scoring and summing gross pathology scores for the following measures: skin, eyes, fins, gills, spleen, kidney, hindgut, liver, and parasite load. The best possible score for each measure, and in total, is 0 . Any increase from a score of 0 indicates the identification of gross pathologies visible during a routine necropsy dissection. The highest (worst) score for each individual measure is 30 .

The HAI is designed to be a used as a summed average for a sample population (Adams et al., 1993). Using this method, the Gladstone Harbour-wide HAI results (nine measures) were determined, by species (Table 3). Reference site data were excluded from these calculations and are provided in Table 4 for comparison. Average HAI scores for reference sites in the present study ranged from 14
to 70, while scores for Gladstone Harbour ranged from 23.3 (bream) to 35.2 (barramundi).
Reference site results should be treated with caution, due to the low sample sizes. For example, the HAI score of 70 for blue catfish is an average of two catfish which scored 10 and 130.

Table 3: Average measure and HAI total scores, calculated for fish caught in Gladstone Harbour in Spring 2018 and Autumn 2019 combined. Individual scores for each organ range from 0-30. Total individual HAl scores range from 0-270. The category "Bream" includes pikey bream and yellowfin bream. The category "Mullet" includes sea mullet and diamondscale mullet.

| Taxa / Measure | Barramundi $(n=23)$ | Bream $(n=9)$ | Barred Javelin $(n=63)$ | Blue Catfish $(n=48)$ | Mullet $(\mathrm{n}=80)$ |
| :---: | :---: | :---: | :---: | :---: | :---: |
| Skin | 1.74 | 1.11 | 0.32 | 1.88 | 0.75 |
| Eyes | 2.61 | 0 | 0.48 | 0 | 1.13 |
| Fins | 0.43 | 0 | 0.63 | 1.49 | 1.00 |
| Gills | 0 | 0 | 0 | 0.63 | 0.38 |
| Spleen | 1.30 | 0 | 0 | 1.25 | 2.63 |
| Kidney | 0 | 0 | 0 | 9.38 | 5.63 |
| Hindgut | 1.30 | 0 | 0.32 | 0.21 | 0.50 |
| Liver | 13.91 | 13.33 | 8.73 | 9.38 | 6.88 |
| Parasites | 14.17 | 3.33 | 0 | 0 | 4.00 |
| HAl score | 35.22 | 23.33 | 23.81 | 34.17 | 26.38 |

Table 4: Average measure and HAI total scores, calculated for fish caught in reference sites in Spring 2018 (Stanage Bay and Baffle Creek) and in Autumn 2019 (Baffle Creek) combined. Individual scores for each organ range from 0-30. Total individual HAI scores range from 0-270. The category "Bream" includes pikey bream and yellowfin bream. The category "Mullet" includes sea mullet and diamondscale mullet.

| Taxa / <br> Measure | Barramundi <br> $(\mathbf{n}=\mathbf{5 )}$ | Bream <br> $(\mathbf{n}=\mathbf{0})$ | Barred Javelin <br> $\mathbf{( n = 4 )}$ | Blue Catfish <br> $\mathbf{( n = 2 )}$ | Mullet <br> $\mathbf{( n = 1 2 )}$ |
| :--- | :--- | :--- | :--- | :--- | :--- |
| Skin | 0 |  | 0 | 0 | 0.83 |
| Eyes | 0 | 0 | 0 | 0 |  |
| Fins | 0 |  | 0 | 5.00 | 1.67 |
| Gills | 0 | 0 | 0 | 0 |  |
| Spleen | 0 | 0 | 15.00 | 5.00 |  |
| Kidney | 0 | 0 | 15.00 | 7.50 |  |
| Hindgut | 0 | 0 | 0 | 0 |  |
| Liver | 6.00 |  | 22.50 | 15.00 | 9.17 |
| Parasites | 8.00 |  | 5.00 | 10.00 | 0 |
| HAI score | 14.00 | ND | 27.50 | 70.00 | 31.67 |

Benchmark: The natural individual fish benchmark for HAI is $0-$ no observable pathologies. However, the average HAI of a large sample of a fish population is unlikely to consistently be maintained at 0 . Even in a pristine environment, fish may have skin abrasions, parasites (most animals have parasites) and slight fin erosion, including as a result of capture in a net. In this study, a score of 0 was achieved by a total of 70 of the 223 fish assessed from Gladstone Harbour, including three barramundi, 14 barred javelin, 15 blue catfish, 3 bream and 35 mullet. Five of the 23 fish
assessed from reference sites achieved a score of 0 , including one barramundi and four mullet. The prevalence of non-0 scores in the reference sites suggests attaining a population level score of 0 in a wild population of fish is unlikely. Instead, a pilot benchmark of an average HAI of $\mathbf{1 0}$ is proposed.

Worst Case Scenario: The maximum total score for an individual fish, using the nine measures tested, is 270 (see Table 1). The level of deviation from normal variation that would constitute a biological tipping point, beyond which a fish population is severely diseased must be derived from other studies.

During the 2011-2012 fish health investigation in Gladstone Harbour, the highest reported Harbour-wide HAI score was 38.8, recorded in the upper Boyne Estuary by Wesche et al. (2013). This score was for all the fish species collected, the report did not detail HAI scores for all individual fish species, although barramundi scores were reported separately. Again for all fish species considered, adjusted means of HAI score identified by Wesche et al. (2013) were 18.3 for fish without a clear disease diagnosis during field assessment, vs. 31.0 for fish with a field diagnosis of diseased, a difference that was significant at the p < 0.01 level (Wesche et al. (2013), Appendix B, pages 108109).

For barramundi, the adjusted mean HAl score was 16.6 for fish without a clear disease diagnosis during field assessment, vs. 32.6 for fish with a field diagnosis of diseased, a difference that was again significant at the $p<0.01$ level (Wesche et al., 2013; Appendix B, page 116). The locations with the highest HAI scores for barramundi were the Upper Boyne Estuary (Trip 147.7 and Trip 2 48.5) and the Lower Boyne Estuary (Trip 141.8 and Trip 2 30.0) (Wesche et al., 2013). Differences in mean barramundi HAI between locations were significantly different at the $p<0.01$ level (Wesche et al., 2013; Appendix B, page 117). Based on the results of Wesche et al. (2013), a possible WCS for average HAI is between 40.0 and 50.0 , however it is not clear from the results that this example represents a "worst case".

In studies from other regions, worst cases can be identified from areas that are known to be polluted. The original HAI developed by Adams et al. (1993) was used to assess the health of fish a range of field sites in the United States of America, including a reservoir contaminated by polychlorinated biphenyls. The original HAI assesses not only the nine measures used in this study, but also five other measures including the thymus, pseudobranch and three blood variables. Therefore, the highest (worst) possible score for a fish using the original HAI is 420, compared with the highest possible score of 270 for the modified HAI used in this study. To compare studies that use the original HAI with this study, it is necessary to compare only the maximum score for the organs used here. Adams et al. (1993) calculated maximum HAl scores of 79 in the worst-scoring reservoir, and 74 in the reservoir containing PCBs. These scores equate to proportionate scores of 50.79 and 47.57 , respectively, when using the modified HAI with the lower possible maximum.

Watson et al. (2012) tested Adams et al.'s (1993) HAI at dams and rivers in South Africa, including the polluted Loskop Dam and Mamba River. The highest average HAI scores determined from these sites were 113.8 and 108.0, in fish from Loskop Dam. Proportionate to the maximum score that could have been achieved using the modified HAI method used here, these scores equate to 73.2 and 69.4 respectively.

Based on these results from other studies, we suggest a pilot WCS score of an average HAI of $\mathbf{7 0}$.

## Scoring the HAI

Using a benchmark score of an average HAI of 10, and a pilot WCS score of an average HAI of 70, example HAI scores and grades were calculated using a distance from the benchmark method, as is used for similar ecological indicators including South East Queensland Report Card (Healthy Land \& Water, 2017), the Fitzroy Basin Report Card (Flint et al., 2017B) and for GHHP's Mud Crab Indicator (Flint et al., 2017A).

Scores and grades were calculated using data from Spring 2018 and Autumn 2019 (Table 5).
The distance from the benchmark function used is as follows:
Calculated score $=1-((x-B) /(W C S-B))$
Where:
$x=$ recorded value
$B=$ benchmark
WCS = worst case scenario

For reference, the HAI score break points when using the proposed benchmark and WCS, can be calculated for each report card grade as follows:

A = average HAI of 0-19
B = average HAI of 20-31
C = average HAI of 32-40
D = average HAI of 41-55
E = average HAI of 56+

Table 5: Calculation of HAI scores and grades for Gladstone Harbour using data from Spring 2018 and Autumn 2019 combined.

| Species <br> Average <br> HAI | Benchmark | WCS | Calculated score |  |
| :--- | ---: | ---: | ---: | ---: |
| Barramundi | 35.22 | 10 | 70 | 0.58 |
| Bream | 23.33 | 10 | 70 | 0.78 |
| Barred javelin | 23.18 | 10 | 70 | 0.77 |
| Blue catfish | 34.17 | 10 | 70 | 0.60 |
| Mullet | 26.38 | 10 | 70 | 0.73 |

Using GHHP's grading scale, grades for each species group were calculated (Table 6), and an overall harbour score and grade determined by averaging the scores of the five species groups.

Table 6: Fish Health Indicator scores and grades for the 2019 Gladstone Harbour Report Card.

| Taxa / <br> Measure | Barramundi | Bream | Barred <br> Javelin | Blue Catfish | Mullet |
| :--- | :--- | :--- | :--- | :--- | :--- |
| Taxa score | Grade C <br> Score 0.58 | Grade B <br> Score 0.78 | Grade B <br> Score 0.77 | Grade C <br> Score 0.60 | Grade B <br> Score 0.73 |
| Overall <br> Harbour score | Grade B <br> Score 0.69 |  |  |  |  |

## Confidence in scores

The primary considerations when determining confidence in HAI scores for 2018-19 are sample size and potential for interference by ecological characteristics of each species group.

Sample sizes of barramundi $(n=23)$ and bream $(n=9)$ were relatively low. Also, as discussed in detail in the 2018 report (Flint et al., 2018), barramundi are a particularly mobile fish species with tagging evidence of movements across many hundreds of kilometres. This means that a barramundi caught in Gladstone Harbour may have moved from elsewhere.

For these reasons, the confidence in scores for barramundi and bream are lower than for the other three species groups. Substantial numbers of barred javelin ( $n=63$ ), blue catfish ( $n=48$ ) and mullet ( $\mathrm{n}=80$ ) contributed to the scores for these species, providing greater confidence that the samples are representative of the wider population.

## Discussion of 2018-19 results and recommendations

The 2018 fish health indicator sampling and analysis identified several fish health indicators that are particularly promising for further analysis and possible inclusion in the Gladstone Harbour Report Card. The most suitable indicator for immediate implementation was the HAI. HAI scores and grades have been calculated for the 2019 Gladstone Harbour Report Card using data collected in Spring 2018 and Autumn 2019. The benchmark value is 10, and a pilot WCS baseline of 70 has been proposed using best available information for different fish species and regions. Using these values, the calculated scores show satisfactory (C) to good (B) fish health in Gladstone Harbour across five species groups, and an overall harbour grade of $B$.

If information becomes available in future to improve the pilot WCS to be species and regionspecific, this should be taken into account by GHHP.

In 2018-19 two sampling events were conducted, to provide information on any differences in fish health that related to season (Spring pre-wet season vs Autumn post-wet season). The only significant difference in HAI between seasons was for barred javelin, which scored better (i.e. lower) in Spring 2018 than in Autumn 2019. For other species, season does not appear to be a big driver of HAI , at least during this sampling year.

Recommendation 1: GHHP continues to monitor HAI of fish in Gladstone Harbour and at least one reference site.

## Recommendation 2: Pilot baselines and methods should be reviewed if more localised information becomes available.

Several other condition measures can be calculated using the data collected during GHHP's fish health monitoring in Gladstone Harbour. The condition measures Fulton's K, HSI and GSI are biologically variable which makes establishment of scientifically defensible baselines difficult in the short term. For example, all three measures are affected by reproductive status of the fish. Fluctuating asymmetry of eye diameter has also been monitored but the lack of information on 'normal' levels of asymmetry in Australian inshore species rule out this potential indicator. A CQUni Masters student is currently investigating the utility of a variety of fluctuating asymmetry measures as indicators of fish health for Queensland's inshore fish species.

All four of these condition metrics can be rapidly measured during dissections, so while they may not yet be useful indicators for the Report Card, it is worthwhile continuing to collect data from future samples to establish a long time series. Following the Gladstone fish health investigation in 20112012, Wesche et al. (2013) reported significantly lower condition factors of barramundi from Gladstone harbour than from reference sites (at the $p<0.05$ level), and barramundi from Gladstone also had significantly higher proportions of sunken abdomens and lower levels of mesentery fat.
During events such as that experienced in 2011-2012, noticeable changes in condition measures are more likely.

Recommendation 3: GHHP continues to monitor measurements required to calculate Fulton's K, HSI and GSI, to collate a long-term dataset.

## Recommendation 4: If new research suggests some fluctuating asymmetry measures may be

 useful fish health indicators, these could be considered for future application in the report card.As discussed in the 2018 fish health research project (Flint et al., 2018), bioaccumulation of toxicants in fish tissues may also be a useful indicator for future consideration. While bioaccumulation only
becomes an indicator of fish health at levels that cause the initiation of detoxification mechanisms and tissue damage (Whitfield \& Elliott, 2002), it also provides information on the bioavailability of toxicants in the environment and is an important consideration for fish that are consumed by people. Bioaccumulation is regarded as an integrative measure and an indicator of exposure of organisms to toxicants in polluted ecosystems. Metals are not metabolised by organisms, and therefore, bioaccumulation of metals and metalloids is of particular value (Luoma \& Rainbow, 2005). Therefore, tissue samples were collected from dissected fish and stored to allow future testing for bioaccumulated metals and other toxicants.

## Recommendation 5: GHHP considers testing for bioaccumulation of metals and other toxicants in collected fish tissue samples.

In Spring 2018, target species were barramundi, bream (including pikey bream and yellowfin bream), large bodied mullet (including diamondscale mullet and sea mullet) and barred javelin. In Autumn 2019, blue catfish were also retained providing more information on demersal scavengers. As discussed in the 2018 research report, bream are not frequently caught in gill nets, and the total number of bream caught in 2018-19 was 9 . Other options that could be considered if GHHP wishes to increase the sample size for bream, are to include targeted hook and line fishing for bream in the monitoring program, or to link with other GHHP projects to provide bream from another source.

A fish movement analysis was conducted as part of the 2018 research project (Flint et al., 2018, Appendix 1), and the range and the average movements of a variety of recreationally caught inshore and estuarine fish species were compared from tag-recapture data provided by the SunTag recreational fishing tagging program. Barramundi are a wide-ranging fish species and can move many hundreds of kilometres between tagging and recapture. There are some issues with the interpretation of any identified health issues for this species, as it may be difficult to determine how long the fish has been resident in the area of capture. Some other inshore species, including bream and barred javelin, are all more resident, so continued monitoring of these taxa is recommended. Large mullet and blue catfish can also travel long distances, but no local tagging records are available for these species because they are not normally targeted by recreational fishers.

During the Spring 2018 and Autumn 2019 sampling events, a range of other inshore and estuarine fish species were captured incidentally. Of the species captured, barramundi, blue catfish, barred javelin, diamondscale mullet are demersal or benthic species that are likely to be in closer contact with pollutants accumulated in sediments, making them useful indicator species (Cowled, 2016). Other demersal and benthic species were caught, but in much smaller numbers. Based on these results, barred javelin and blue catfish were added to the list of target species during 2018-19.

The fish movement analysis conducted during the 2018 research project also detected high transience of fish between different areas within Gladstone Harbour. As such, the ISP elected to report scores of fish health at the harbour-wide scale. Because fish health scores are reported on a harbour-wide scale, the ISP also decided for 2019 to amend the sampling design to allow for higher catches with lower levels of effort (i.e. spend time fishing in zones with a high probability of target species catch). After this change in sampling strategy the catch per day of target species increased.

## Recommendation 6: GHHP continues to conduct regionally stratified fish sampling across

 Gladstone Harbour.In 2018, two reference sites were monitored to assist with the development of baselines for fish health measures, Stanage Bay and Baffle Creek. In 2019, the Stanage Bay site was removed. Fish from the reference sites did not appear to be in pristine condition, which may reflect local
environmental effects. Regardless, in order to continue to assess the condition of fish in Gladstone Harbour in a relative way, it would be beneficial for GHHP to continue to sample at least one reference site (although once a year may be enough), as a precaution against misinterpreting more widespread changes as localised impacts.

Recommendation 7: GHHP continues to sample at least one reference site, at least once a year.

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## Appendix 1: Details of all sampling locations and times

$\left.\begin{array}{|r|l|l|l|l|l|l|l|l|}\hline \begin{array}{l}\text { GHHP } \\ \text { Zone } \\ \text { Number }\end{array} & & \text { Survey } & \begin{array}{l}\text { Gill net } \\ \text { mesh } \\ \text { size } \\ \text { (inches) }\end{array} & & \text { Date } & \begin{array}{l}\text { Deploy } \\ \text { time }\end{array} \\ \hline \text { Soak } \\ \text { time } \\ \text { (h:mm) }\end{array}\right)$
$\left.\begin{array}{|r|l|l|l|l|l|l|l|l|}\hline \begin{array}{l}\text { GHHP } \\ \text { Zone } \\ \text { Number }\end{array} & \text { Location } & \text { Survey } & \begin{array}{l}\text { Gill net } \\ \text { mesh } \\ \text { size } \\ \text { (inches) }\end{array} & & \text { Date } & \text { Deploy } \\ \text { time }\end{array} \begin{array}{l}\text { Soak } \\ \text { time } \\ \text { (h:mm) }\end{array}\right)$

| GHHP <br> Zone <br> Number | Location | Survey | Gill net <br> mesh <br> size <br> (inches) | Date | Deploy time | Soak time (h:mm) | Latitude | Longitude |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 9 | South Trees Inlet | Sep-2018 | 6 | 20/09/2018 | 10:06 | 0:31 | -23.85333 | 151.29757 |
| 9 | South Trees Inlet | Sep-2018 | 6 | 20/09/2018 | 11:32 | 0:13 | -23.91723 | 151.29999 |
| 9 | South Trees Inlet | Sep-2018 | 4.5 | 20/09/2018 | 11:37 | 0:16 | -23.91797 | 151.30013 |
| 9 | South Trees Inlet | Sep-2018 | 4.5 | 20/09/2018 | 11:57 | 0:20 | -23.91617 | 151.29997 |
| 9 | South Trees Inlet | Sep-2018 | 6 | 20/09/2018 | 12:04 | 0:07 | -23.91661 | 151.29956 |
| 9 | South Trees Inlet | Sep-2018 | 4.5 | 20/09/2018 | 12:22 | 0:44 | -23.91273 | 151.29696 |
| 9 | South Trees Inlet | Sep-2018 | 6 | 20/09/2018 | 12:29 | 0:32 | -23.91320 | 151.29684 |
| 9 | South Trees Inlet | Sep-2018 | 4.5 | 20/09/2018 | 13:36 | 0:33 | -23.93966 | 151.30212 |
| 9 | South Trees Inlet | Sep-2018 | 6 | 20/09/2018 | 13:40 | 0:35 | -23.94043 | 151.30220 |
| 9 | South Trees Inlet | Sep-2018 | 2 | 20/09/2018 | 13:48 | 0:01 | -23.93977 | 151.30276 |
| 10 | Boyne River | Sep-2018 | 4.5 | 03/10/2018 | 5:39 | 0:37 | -23.97725 | 151.33092 |
| 10 | Boyne River | Sep-2018 | 6 | 03/10/2018 | 5:51 | 0:47 | -23.97683 | 151.32348 |
| 10 | Boyne River | Sep-2018 | 8 | 03/10/2018 | 6:06 | 0:39 | -23.97938 | 151.31917 |
| 10 | Boyne River | Sep-2018 | 4.5 | 03/10/2018 | 6:17 | 0:47 | -23.97725 | 151.33092 |
| 10 | Boyne River | Sep-2018 | 8 | 03/10/2018 | 6:46 | 0:39 | -23.97938 | 151.31917 |
| 10 | Boyne River | Sep-2018 | 6 | 03/10/2018 | 6:50 | 0:40 | -23.97965 | 151.31986 |
| 10 | Boyne River | Sep-2018 | 4.5 | 03/10/2018 | 7:20 | 0:38 | -23.97913 | 151.32087 |
| 10 | Boyne River | Sep-2018 | 8 | 03/10/2018 | 7:26 | 0:27 | -23.97938 | 151.31917 |
| 10 | Boyne River | Sep-2018 | 6 | 03/10/2018 | 7:47 | 0:33 | -23.97782 | 151.32123 |
| 10 | Boyne River | Sep-2018 | 8 | 03/10/2018 | 7:54 | 0:40 | -23.97938 | 151.31917 |
| 10 | Boyne River | Sep-2018 | 4.5 | 03/10/2018 | 7:59 | 0:41 | -23.97913 | 151.32087 |
| 10 | Boyne River | Sep-2018 | 6 | 03/10/2018 | 8:22 | 0:36 | -23.97782 | 151.32123 |
| 10 | Boyne River | Sep-2018 | 8 | 03/10/2018 | 8:35 | 0:28 | -23.97938 | 151.31917 |
| 10 | Boyne River | Sep-2018 | 4.5 | 03/10/2018 | 8:41 | 0:27 | -23.97913 | 151.32087 |
| 10 | Boyne River | Sep-2018 | 6 | 03/10/2018 | 8:59 | 0:31 | -23.97782 | 151.32123 |
| 10 | Boyne River | Sep-2018 | 8 | 03/10/2018 | 9:04 | 0:22 | -23.97913 | 151.32087 |
| 10 | Boyne River | Sep-2018 | 4.5 | 03/10/2018 | 9:20 | 0:31 | -23.97844 | 151.32091 |
| 10 | Boyne River | Sep-2018 | 8 | 03/10/2018 | 9:27 | 0:33 | -23.97913 | 151.32087 |
| 10 | Boyne River | Sep-2018 | 6 | 03/10/2018 | 9:31 | 0:17 | -23.97782 | 151.32123 |
| 10 | Boyne River | Sep-2018 | 4.5 | 03/10/2018 | 9:52 | 0:28 | -23.97844 | 151.32091 |
| 10 | Boyne River | Sep-2018 | 6 | 03/10/2018 | 9:49 | 0:36 | -23.97782 | 151.32123 |
| 10 | Boyne River | Sep-2018 | 8 | 03/10/2018 | 10:01 | 0:35 | -23.97913 | 151.32087 |
| 10 | Boyne River | Sep-2018 | 4.5 | 03/10/2018 | 10:21 | 0:35 | -23.97844 | 151.32091 |
| 10 | Boyne River | Sep-2018 | 6 | 03/10/2018 | 10:26 | 0:23 | -23.97782 | 151.32123 |
| 12 | Colosseum Inlet | Sep-2018 | 6 | 21/09/2018 | 7:51 | 0:40 | -24.06267 | 151.48302 |
| 12 | Colosseum Inlet | Sep-2018 | 4.5 | 21/09/2018 | 7:59 | 0:35 | -24.06336 | 151.48342 |
| 12 | Colosseum Inlet | Sep-2018 | 2 | 21/09/2018 | 8:18 | 0:09 | -24.05662 | 151.48118 |
| 12 | Colosseum Inlet | Sep-2018 | 6 | 21/09/2018 | 8:32 | 0:38 | -24.06267 | 151.48302 |
| 12 | Colosseum Inlet | Sep-2018 | 4.5 | 21/09/2018 | 8:35 | 0:38 | -24.06336 | 151.48342 |
| 12 | Colosseum Inlet | Sep-2018 | 2 | 21/09/2018 | 8:58 | 0:09 | -24.05225 | 151.46112 |
| 12 | Colosseum Inlet | Sep-2018 | 6 | 21/09/2018 | 9:11 | 0:21 | -24.06267 | 151.48302 |
| 12 | Colosseum Inlet | Sep-2018 | 4.5 | 21/09/2018 | 9:14 | 0:26 | -24.06336 | 151.48342 |
| 12 | Colosseum Inlet | Sep-2018 | 4.5 | 21/09/2018 | 9:51 | 0:59 | -24.07363 | 151.48430 |

$\left.\begin{array}{|l|l|l|l|l|l|l|l|l|}\hline \begin{array}{l}\text { GHHP } \\ \text { Zone } \\ \text { Number }\end{array} & \text { Location } & \text { Survey } & \begin{array}{l}\text { Gill net } \\ \text { mesh } \\ \text { size } \\ \text { (inches) }\end{array} & & \text { Date } & \text { Deploy } \\ \text { time }\end{array} \begin{array}{l}\text { Soak } \\ \text { time } \\ \text { (h:mm) }\end{array}\right)$
$\begin{array}{|l|l|l|r|l|l|l|l|l|}\hline \begin{array}{l}\text { GHHP } \\ \text { Zone } \\ \text { Number }\end{array} & \text { Location } & \text { Survey } & \begin{array}{l}\text { Gill net } \\ \text { mesh } \\ \text { size } \\ \text { (inches) }\end{array} & & \text { Date } & \text { Deploy } \\ \text { time }\end{array}$ Soak $\left.\begin{array}{l}\text { time } \\ \text { (h:mm) }\end{array}\right)$

| GHHP <br> Zone <br> Number | Location | Survey | Gill net <br> mesh <br> size <br> (inches) | Date | Deploy time | Soak time (h:mm) | Latitude | Longitude |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 2 | Graham Creek | Apr-2019 | 6 | 13/04/2019 | 8:43 | 0:36 | -23.70720 | 151.22509 |
| 2 | Graham Creek | Apr-2019 | 8 | 13/04/2019 | 8:51 | 0:34 | -23.70269 | 151.22471 |
| 2 | Graham Creek | Apr-2019 | 8 | 13/04/2019 | 9:26 | 0:24 | -23.70269 | 151.22471 |
| 2 | Graham Creek | Apr-2019 | 6 | 13/04/2019 | 10:07 | 0:27 | -23.72963 | 151.21591 |
| 2 | Graham Creek | Apr-2019 | 4.5 | 13/04/2019 | 10:18 | 0:27 | -23.73243 | 151.21653 |
| 2 | Graham Creek | Apr-2019 | 6 | 13/04/2019 | 10:35 | 0:21 | -23.72963 | 151.21591 |
| 2 | Graham Creek | Apr-2019 | 4.5 | 13/04/2019 | 10:46 | 0:32 | -23.73243 | 151.21653 |
| 2 | Graham Creek | Apr-2019 | 6 | 13/04/2019 | 11:39 | 0:31 | -23.74367 | 151.12940 |
| 2 | Graham Creek | Apr-2019 | 4.5 | 13/04/2019 | 11:50 | 0:11 | -23.74745 | 151.12663 |
| 5 | Inner Harbour | Apr-2019 | 4.5 | 10/04/2019 | 8:48 | 0:27 | -23.77141 | 151.24742 |
| 5 | Inner Harbour | Apr-2019 | 6 | 10/04/2019 | 8:54 | 0:06 | -23.76884 | 151.24595 |
| 5 | Inner Harbour | Apr-2019 | 6 | 10/04/2019 | 9:03 | 0:29 | -23.76928 | 151.24541 |
| 5 | Inner Harbour | Apr-2019 | 4.5 | 10/04/2019 | 9:16 | 0:29 | -23.77141 | 151.24742 |
| 5 | Inner Harbour | Apr-2019 | 6 | 10/04/2019 | 9:38 | 0:27 | -23.77172 | 151.24663 |
| 5 | Inner Harbour | Apr-2019 | 4.5 | 10/04/2019 | 10:21 | 0:29 | -23.78795 | 151.24326 |
| 5 | Inner Harbour | Apr-2019 | 6 | 10/04/2019 | 10:28 | 0:29 | -23.79358 | 151.24411 |
| 5 | Inner Harbour | Apr-2019 | 4.5 | 10/04/2019 | 10:51 | 0:29 | -23.78795 | 151.24326 |
| 5 | Inner Harbour | Apr-2019 | 6 | 10/04/2019 | 10:58 | 0:27 | -23.79358 | 151.24411 |
| 5 | Inner Harbour | Apr-2019 | 2 | 10/04/2019 | 11:02 | 0:25 | -23.79181 | 151.24008 |
| 5 | Inner Harbour | Apr-2019 | 4.5 | 10/04/2019 | 11:21 | 0:33 | -23.78795 | 151.24326 |
| 5 | Inner Harbour | Apr-2019 | 6 | 10/04/2019 | 11:26 | 0:37 | -23.79358 | 151.24411 |
| 5 | Inner Harbour | Apr-2019 | 6 | 10/04/2019 | 12:13 | 0:42 | -23.80719 | 151.25339 |
| 5 | Inner Harbour | Apr-2019 | 4.5 | 10/04/2019 | 12:19 | 0:39 | -23.80275 | 151.25533 |
| 5 | Inner Harbour | Apr-2019 | 2 | 10/04/2019 | 12:35 | 0:17 | -23.80398 | 151.25398 |
| 5 | Inner Harbour | Apr-2019 | 6 | 10/04/2019 | 12:56 | 0:28 | -23.80719 | 151.25339 |
| 6 | Calliope River | Apr-2019 | 4.5 | 11/04/2019 | 5:49 | 0:46 | -23.86752 | 151.18605 |
| 6 | Calliope River | Apr-2019 | 6 | 11/04/2019 | 6:05 | 0:15 | -23.86558 | 151.18707 |
| 6 | Calliope River | Apr-2019 | 6 | 11/04/2019 | 6:21 | 0:08 | -23.86552 | 151.18721 |
| 6 | Calliope River | Apr-2019 | 6 | 11/04/2019 | 6:48 | 0:07 | -23.85651 | 151.17735 |
| 6 | Calliope River | Apr-2019 | 6 | 11/04/2019 | 7:15 | 0:32 | -23.88801 | 151.19163 |
| 6 | Calliope River | Apr-2019 | 4.5 | 11/04/2019 | 7:34 | 0:25 | -23.89077 | 151.19606 |
| 6 | Calliope River | Apr-2019 | 6 | 11/04/2019 | 8:06 | 0:37 | -23.89160 | 151.19252 |
| 6 | Calliope River | Apr-2019 | 4.5 | 11/04/2019 | 8:00 | 0:25 | -23.89077 | 151.19606 |
| 6 | Calliope River | Apr-2019 | 4.5 | 11/04/2019 | 8:30 | 0:32 | -23.89242 | 151.19500 |
| 6 | Calliope River | Apr-2019 | 6 | 11/04/2019 | 8:57 | 0:11 | -23.89521 | 151.19356 |
| 6 | Calliope River | Apr-2019 | 4.5 | 11/04/2019 | 9:05 | 0:27 | -23.89242 | 151.19500 |
| 6 | Calliope River | Apr-2019 | 6 | 11/04/2019 | 9:10 | 0:30 | -23.89521 | 151.19356 |
| 6 | Calliope River | Apr-2019 | 6 | 11/04/2019 | 9:41 | 0:29 | -23.89521 | 151.19356 |
| 6 | Calliope River | Apr-2019 | 4.5 | 11/04/2019 | 9:49 | 0:38 | -23.89834 | 151.19704 |
| 6 | Calliope River | Apr-2019 | 4.5 | 11/04/2019 | 10:28 | 0:21 | -23.89834 | 151.19704 |
| 6 | Calliope River | Apr-2019 | 2 | 11/04/2019 | 10:31 | 0:09 | -23.89841 | 151.19726 |
| 7 | Auckland Creek | Apr-2019 | 6 | 10/04/2019 | 5:12 | 0:33 | -23.84976 | 151.24146 |
| 7 | Auckland Creek | Apr-2019 | 4.5 | 10/04/2019 | 5:28 | 0:29 | -23.85263 | 151.24132 |


| GHHP <br> Zone <br> Number | Location | Survey | Gill net <br> mesh <br> size <br> (inches) | Date | Deploy time | Soak time (h:mm) | Latitude | Longitude |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 7 | Auckland Creek | Apr-2019 | 6 | 10/04/2019 | 5:45 | 0:26 | -23.84976 | 151.24146 |
| 7 | Auckland Creek | Apr-2019 | 4.5 | 10/04/2019 | 5:57 | 0:08 | -23.85263 | 151.24132 |
| 7 | Auckland Creek | Apr-2019 | 6 | 10/04/2019 | 6:24 | 0:16 | -23.84911 | 151.23190 |
| 7 | Auckland Creek | Apr-2019 | 6 | 10/04/2019 | 7:09 | 0:25 | -23.84815 | 151.23040 |
| 7 | Auckland Creek | Apr-2019 | 4.5 | 10/04/2019 | 7:18 | 0:37 | -23.85094 | 151.23301 |
| 7 | Auckland Creek | Apr-2019 | 6 | 10/04/2019 | 7:35 | 0:29 | -23.84933 | 151.23199 |
| 8 | Mid Harbour | Apr-2019 | 6 | 16/04/2019 | 6:00 | 0:34 | -23.82688 | 151.33845 |
| 8 | Mid Harbour | Apr-2019 | 4.5 | 16/04/2019 | 6:09 | 0:28 | -23.82597 | 151.33980 |
| 8 | Mid Harbour | Apr-2019 | 6 | 16/04/2019 | 6:35 | 0:31 | -23.82688 | 151.33845 |
| 8 | Mid Harbour | Apr-2019 | 4.5 | 16/04/2019 | 6:38 | 0:36 | -23.82597 | 151.33980 |
| 8 | Mid Harbour | Apr-2019 | 6 | 16/04/2019 | 7:07 | 0:26 | -23.82688 | 151.33845 |
| 8 | Mid Harbour | Apr-2019 | 4.5 | 16/04/2019 | 7:29 | 0:34 | -23.82842 | 151.33796 |
| 8 | Mid Harbour | Apr-2019 | 6 | 16/04/2019 | 7:34 | 0:31 | -23.82688 | 151.33845 |
| 8 | Mid Harbour | Apr-2019 | 4.5 | 16/04/2019 | 8:04 | 0:31 | -23.82842 | 151.33796 |
| 8 | Mid Harbour | Apr-2019 | 6 | 16/04/2019 | 8:06 | 0:24 | -23.82688 | 151.33845 |
| 8 | Mid Harbour | Apr-2019 | 4.5 | 16/04/2019 | 8:42 | 0:32 | -23.82842 | 151.33796 |
| 8 | Mid Harbour | Apr-2019 | 6 | 16/04/2019 | 8:50 | 0:27 | -23.82492 | 151.33662 |
| 8 | Mid Harbour | Apr-2019 | 4.5 | 16/04/2019 | 9:15 | 0:39 | -23.82842 | 151.33796 |
| 8 | Mid Harbour | Apr-2019 | 6 | 16/04/2019 | 9:18 | 0:23 | -23.82492 | 151.33662 |
| 8 | Mid Harbour | Apr-2019 | 6 | 16/04/2019 | 10:05 | 0:26 | -23.81627 | 151.33422 |
| 8 | Mid Harbour | Apr-2019 | 4.5 | 16/04/2019 | 10:10 | 0:27 | -23.81434 | 151.33476 |
| 8 | Mid Harbour | Apr-2019 | 6 | 16/04/2019 | 10:32 | 0:34 | -23.81627 | 151.33422 |
| 9 | South Trees Inlet | Apr-2019 | 4.5 | 14/04/2019 | 5:59 | 0:10 | -23.95156 | 151.31870 |
| 9 | South Trees Inlet | Apr-2019 | 4.5 | 14/04/2019 | 6:28 | 0:29 | -23.94930 | 151.32901 |
| 9 | South Trees Inlet | Apr-2019 | 6 | 14/04/2019 | 6:34 | 0:25 | -23.94786 | 151.32797 |
| 9 | South Trees Inlet | Apr-2019 | 4.5 | 14/04/2019 | 6:58 | 0:28 | -23.94930 | 151.32901 |
| 9 | South Trees Inlet | Apr-2019 | 6 | 14/04/2019 | 7:00 | 0:24 | -23.94786 | 151.32797 |
| 9 | South Trees Inlet | Apr-2019 | 2 | 14/04/2019 | 7:02 | 0:13 | -23.94857 | 151.33109 |
| 9 | South Trees Inlet | Apr-2019 | 6 | 14/04/2019 | 7:25 | 1:00 | -23.94786 | 151.32797 |
| 9 | South Trees Inlet | Apr-2019 | 4.5 | 14/04/2019 | 7:27 | 0:47 | -23.94930 | 151.32901 |
| 9 | South Trees Inlet | Apr-2019 | 2 | 14/04/2019 | 7:58 | 0:10 | -23.94929 | 151.32674 |
| 9 | South Trees Inlet | Apr-2019 | 6 | 14/04/2019 | 8:53 | 0:33 | -23.94798 | 151.29132 |
| 9 | South Trees Inlet | Apr-2019 | 4.5 | 14/04/2019 | 9:11 | 0:23 | -23.94962 | 151.29274 |
| 9 | South Trees Inlet | Apr-2019 | 6 | 14/04/2019 | 9:27 | 0:16 | -23.94798 | 151.29132 |
| 9 | South Trees Inlet | Apr-2019 | 4.5 | 14/04/2019 | 9:35 | 0:02 | -23.94992 | 151.29220 |
| 9 | South Trees Inlet | Apr-2019 | 6 | 14/04/2019 | 10:00 | 0:15 | -23.91305 | 151.29693 |
| 9 | South Trees Inlet | Apr-2019 | 4.5 | 14/04/2019 | 10:29 | 0:36 | -23.89142 | 151.29376 |
| 9 | South Trees Inlet | Apr-2019 | 6 | 14/04/2019 | 10:34 | 0:21 | -23.89172 | 151.29348 |
| 9 | South Trees Inlet | Apr-2019 | 4.5 | 14/04/2019 | 11:06 | 0:14 | -23.89142 | 151.29376 |
| 9 | South Trees Inlet | Apr-2019 | 4.5 | 14/04/2019 | 11:33 | 0:28 | -23.87377 | 151.30492 |
| 9 | South Trees Inlet | Apr-2019 | 2 | 14/04/2019 | 11:39 | 0:31 | -23.87635 | 151.30491 |
| 9 | South Trees Inlet | Apr-2019 | 4.5 | 14/04/2019 | 12:02 | 0:29 | -23.87377 | 151.30492 |
| 9 | Mid Harbour | Apr-2019 | 4.5 | 16/04/2019 | 11:40 | 0:26 | -23.80292 | 151.28916 |

$\begin{array}{|l|l|l|r|l|l|l|l|l|}\hline \begin{array}{l}\text { GHHP } \\ \text { Zone } \\ \text { Number }\end{array} & \text { Location } & \text { Survey } & \begin{array}{l}\text { Gill net } \\ \text { mesh } \\ \text { size } \\ \text { (inches) }\end{array} & & \text { Date } & \text { Deploy } \\ \text { time }\end{array}$ Soak $\left.\begin{array}{l}\text { time } \\ \text { (h:mm) }\end{array}\right)$

| GHHP <br> Zone <br> Number | Location | Survey | Gill net <br> mesh <br> size <br> (inches) | Date | Deploy <br> time | Soak <br> time <br> (h:mm) | Latitude | Longitude |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- |
|  | Baffle Creek | Apr-2019 | 2 | $17 / 04 / 2019$ | $11: 44$ | $0: 16$ | -24.52693 | 152.03395 |
|  | Baffle Creek | Apr-2019 | 2 | $17 / 04 / 2019$ | $12: 04$ | $0: 04$ | -24.52693 | 152.03395 |
|  | Baffle Creek | Apr-2019 | 4.5 | $17 / 04 / 2019$ | $11: 41$ | $0: 30$ | -24.52702 | 152.03351 |
|  | Baffle Creek | Apr-2019 | 4.5 | $17 / 04 / 2019$ | $12: 12$ | $0: 24$ | -24.52702 | 152.03351 |
|  | Baffle Creek | Apr-2019 | 6 | $17 / 04 / 2019$ | $12: 52$ | $0: 34$ | -24.52542 | 152.02697 |
|  | Baffle Creek | Apr-2019 | 4.5 | $17 / 04 / 2019$ | $13: 03$ | $0: 32$ | -24.52508 | 152.02640 |
|  | Baffle Creek | Apr-2019 | 6 | $17 / 04 / 2019$ | $12: 27$ | $0: 18$ | -24.52542 | 152.02697 |
|  | Baffle Creek | Apr-2019 | 6 | $17 / 04 / 2019$ | $12: 58$ | $1: 10$ | -24.51390 | 152.02254 |
|  | Baffle Creek | Apr-2019 | 4.5 | $17 / 04 / 2019$ | $14: 04$ | $0: 22$ | -24.51091 | 152.02100 |
|  | Baffle Creek | Apr-2019 | 6 | $17 / 04 / 2019$ | $14: 09$ | $0: 29$ | -24.51390 | 152.02254 |
|  |  |  |  |  |  |  |  |  |

Appendix 2: Site physicochemical data

Spring 2018

| Site | Zone | GHHP Zone <br> Number | Date/Time | Temp $\left({ }^{\circ} \mathrm{C}\right)$ | $\begin{aligned} & \text { DO } \\ & \text { (\%) } \end{aligned}$ | $\begin{aligned} & \text { DO } \\ & (\mathrm{mg} / \mathrm{L}) \end{aligned}$ | EC ( $\mu \mathrm{s} / \mathrm{cm}$ ) | pH | Turbidity (NTU) | TDS | ORP | Salinity (ppt) |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Western Basin | WB | 3 | 17/09/2018 8:36 | 22.2 | 94.7 | 6.66 | 55890 | 8.10 | 5.1 | 36336 | -182.9 | 37.17 |
| Calliope River | CR | 6 | 17/09/2018 13:23 | 23.1 | 98.9 | 7.05 | 48739 | 8.00 | 7.0 | 31684 | -179.0 | 31.88 |
| Graham Creek | GC | 2 | 18/09/2018 9:25 | 22.5 | 91.5 | 6.39 | 56092 | 7.80 | 3.3 | 36463 | -190.1 | 37.31 |
| Auckland Creek | AC | 7 | 18/09/2018 14:46 | 23.1 | 96.6 | 6.69 | 55699 | 7.70 | 10.0 | 36209 | -200.4 | 37.01 |
| Narrows | NW | 1 | 19/09/2018 9:51 | 22.2 | 83.4 | 5.85 | 56958 | 7.60 | 3.4 | 37027 | -188.1 | 37.96 |
| Inner Harbour | IH | 5 | 19/09/2018 14:24 | 25.3 | 99.4 | 6.63 | 55759 | 7.80 | 14.4 | 36241 | -197.2 | 37.01 |
| Mid Harbour | MH | 8 | 20/09/2018 9:39 | 22.3 | 94.5 | 6.66 | 54673 | 8.00 | 7.4 | 35623 | -181.7 | 36.27 |
| South Trees | STI | 9 | 20/09/2018 14:26 | 23.8 | 66.1 | 4.44 | 56304 | 7.80 | 6.1 | 36592 | -221.9 | 37.43 |
| Colosseum | RCI | 12 | 21/09/2018 8:42 | 22.0 | 83.9 | 5.89 | 56437 | 7.90 | 4.2 | 36887 | -195.4 | 37.79 |
| Boyne River | BR | 10 | 03/10/2018 11:03 | 23.8 | 87.4 | 6.12 | 49964 | 7.80 | 3.3 | 32487 | -165.5 | 32.76 |
| Baffle Creek | BC |  | 04/10/2018 12:48 | 23.8 | 105.5 | 7.32 | 52145 | 8.00 | 2.1 | 33921 | -168.4 | 34.35 |
| Rodds Bay | RB | 13 | 05/10/2018 11:55 | 23.5 | 100.1 | 6.89 | 55172 | 8.10 | 3.8 | 35801 | -181.4 | 36.61 |
| Stanage Bay | SB |  | 01/11/2018 13:00 | ND | ND | ND | ND | ND | ND | ND | ND | ND |

Autumn 2019

| Site | Zone | GHHP Zone Number | Date/Time | Temp ( $\left.{ }^{\circ} \mathrm{C}\right)$ | $\begin{aligned} & \text { DO } \\ & \text { (\%) } \end{aligned}$ | $\begin{aligned} & \mathrm{DO} \\ & (\mathrm{mg} / \mathrm{L}) \end{aligned}$ | $\begin{aligned} & \mathrm{EC} \\ & (\mu \mathrm{~s} / \mathrm{cm}) \end{aligned}$ | pH | Turbidity (NTU) | TDS | ORP | Salinity (ppt) |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Western Basin | WB | 3 | NS | NS | NS | NS | NS | NS | NS | NS | NS | NS |
| Calliope River | CR | 6 | 11/04/2019 10:38 | 25.9 | 79.6 | 5.18 | 58870 | 7.47 | 18.6 | 37644 | 170.0 | 38.59 |
| Graham Creek | GC | 2 | 13/04/2019 6:49 | 22.6 | 82.4 | 5.65 | 57175 | 7.65 | 8.9 | 38974 | -111.1 | 40.23 |
| Auckland Creek | AC | 7 | 10/04/2019 6:58 | 25.0 | 96.1 | 6.38 | 57433 | 7.72 | 4.5 | 37321 | 164.9 | 38.25 |
| Narrows | NW | 1 | 12/04/2019 12:34 | 24.5 | 105.5 | 7.06 | 57858 | 7.98 | 16.5 | 37611 | 156.3 | 38.61 |
| Inner Harbour | IH | 5 | 10/04/2019 12:25 | 25.8 | 101.3 | 6.64 | 58410 | 8.13 | 2.8 | 37389 | -143.3 | 38.31 |
| Mid Harbour (south) | MH | 8 | 16/04/2019 9:10 | 23.2 | 97.4 | 6.67 | 55260 | 8.09 | 8.2 | 37165 | 122.2 | 38.11 |
| Mid Harbour (middle) | MH | 8 | 16/04/2019 13:18 | 23.8 | 99.7 | 6.76 | 56071 | 8.43 | 7.8 | 37289 | 116.1 | 36.25 |
| South Trees | STI | 9 | 14/04/2019 9:23 | 22.4 | 66.1 | 4.5 | 58010.0 | 7.4 | 24.6 | 39665.0 | 168.7 | 41.0 |
| Colosseum | RCl | 12 | NS | NS | NS | NS | NS | NS | NS | NS | NS | NS |
| Boyne River | BR | 10 | 15/04/2019 5:39 | 23.4 | 88.3 | 6.48 | 38477 | 7.83 | 1.2 | 25663 | 110.3 | 25.26 |
| Baffle Creek | BC |  | 17/04/2019 7:08 | 23.7 | 88.2 | 6.03 | 54177 | 7.71 | 2.4 | 36004 | 153.5 | 36.78 |
| Rodds Bay | RB | 13 | NS | NS | NS | NS | NS | NS | NS | NS | NS | NS |
| Stanage Bay | SB |  | NS | NS | NS | NS | NS | NS | NS | NS | NS | NS |
| ND = No data, NS = not | mpled. |  |  |  |  |  |  |  |  |  |  |  |

## Appendix 3: Species (scientific and common name) catch by zone and season

| Common name | Scientific name | Spring 2018 |  |  |  |  |  |  |  |  |  |  |  |  |  | Autumn 2019 |  |  |  |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | 1 | 2 | 3 | 5 | 6 | 7 | 8 | 9 | 1 | 1 | $\begin{aligned} & 1 \\ & 3 \end{aligned}$ | $\begin{aligned} & \mathrm{R} \\ & 1 \end{aligned}$ | $\begin{aligned} & \mathbf{R} \\ & \mathbf{2} \end{aligned}$ | Tota I | 1 | 2 | 5 | 6 | 7 | 8 | 9 | 1 | $\begin{aligned} & \mathrm{R} \\ & 1 \end{aligned}$ | Tota I |
| Barramundi | Lates calcarifer | 2 |  |  |  |  | 3 | 1 | 1 |  | 6 |  |  | 6 | 19 | 1 | 1 |  | 2 | 4 | 1 |  | 2 |  | 11 |
|  |  | 1 | 1 |  | 1 |  |  |  |  |  |  |  |  |  |  |  | 1 |  |  |  |  |  |  |  |  |
| Barred Javelin | Pomadasys kaakan |  | 4 | 2 | 3 | 1 | 9 | 9 | 1 |  |  | 1 | 4 |  | 55 | 2 | 6 | 4 | 7 |  | 7 |  |  |  | 36 |
| Batfish | Ephippidae |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  | 1 | 1 |
| Beach Salmon | Leptobrama muelleri |  | 2 | 6 |  |  |  |  |  |  |  |  |  | 1 | 9 | 3 | 1 | 6 |  |  |  | 1 |  |  | 11 |
| Blackspotted rockcod Blubber lip bream (Brown Sweetlips) | Epinephelus malabaricus |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  | 1 |  |  |  |  |  | 1 |
|  | Plectorhinchus gibbosus |  |  |  |  |  | 3 |  |  |  |  |  |  | 1 | 4 |  | 1 |  | 1 |  |  |  |  |  | 2 |
|  |  |  |  |  |  |  |  |  |  |  | 1 |  |  |  |  |  |  |  | 1 |  |  | 1 |  |  |  |
| Blue Catfish | Neoarius graeffei |  | 1 |  | 1 | 4 | 2 |  | 5 | 1 | 0 | 6 |  | 26 | 61 | 5 |  | 5 | 2 |  | 1 | 8 | 7 | 1 | 49 |
|  |  |  |  |  |  |  |  |  |  |  |  | 1 |  |  |  | 1 |  |  |  |  |  |  |  |  |  |
| Blue Threadfin | Eleutheronema tetradactylum | 2 | 1 | 5 | 3 |  | 3 | 9 |  |  | 2 | 1 |  |  | 36 | 0 | 4 | 1 | 2 |  | 6 | 5 |  |  | 28 |
| Blue tuskfish | Choerodon cyanodus |  |  |  |  |  |  |  |  |  |  |  |  | 1 | 1 |  |  |  |  |  |  |  |  |  |  |
| Bony bream | Nematalosa erebi |  |  |  |  |  |  |  |  |  |  | 1 |  |  | 1 |  |  |  |  |  |  |  |  |  |  |
| Bull Shark | Carcharhinus leucas |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  | 2 |  |  | 1 | 9 |  | 12 |
| Common Ponyfish | Leiognathus equulus |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  | 1 |  |  |  |  |  |  | 1 |
| Common Silverbiddy | Gerres subfasciatus |  |  |  |  |  |  |  |  |  | 1 |  |  |  | 1 |  |  |  |  |  |  |  |  |  |  |
| Diamond Scale Mullet | Liza vaigiensis |  | 4 | 3 | 7 | 1 | 1 | 1 | 3 |  |  |  |  | 1 | 21 |  |  |  |  |  | 2 | 2 |  |  | 4 |
| Dusky flathead | Platycephalus fuscus |  |  |  | 1 |  |  |  | 1 |  |  |  | 1 |  | 3 | 1 | 1 |  |  |  |  |  |  | 1 | 3 |
|  | Scomberoides |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| Giant queenfish | commersonnianus |  |  |  |  |  | 2 | 1 | 1 |  | 1 |  |  |  | 5 |  |  |  |  |  | 1 | 2 | 6 |  | 9 |
| Giant Trevally | Caranx ignoblis |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  | 1 | 1 | 1 | 3 |
| Giant Shovelnose Ray | Glaucostegus typus |  |  |  |  |  |  |  |  |  |  |  | 1 |  | 1 |  |  |  |  |  |  |  |  |  |  |
| Goldlined Rabbitfish | Siganus lineatus |  |  |  |  |  |  |  |  |  |  | 1 |  |  | 1 |  |  |  |  |  |  |  |  |  |  |
| Golden snapper | Lutjanus johnii |  |  |  |  |  |  |  |  |  |  |  |  |  |  | 1 |  | 2 |  |  |  |  |  |  | 3 |



## Appendix 4: Permanova analysis results

## Spring 2018 and Autumn 2019 comparison

Comparisons of fish health variables between each sampling season were made for each target species using PERMANOVA. Data were pooled across sampling zones to increase replication for individual tests, but to also provide a broader harbour-wide assessment of fish health. The following results were obtained.

## Health Assessment Index (HAI)

Analysis of HAI between seasons highlighted only a significant difference for Barred Javelin ( $\mathrm{P}=$ 0.015), where HAI was greater in April 2019 than September 2018.

| Species | Source of variation | df | MS | $F$ | $P$ |
| :--- | :--- | :---: | :---: | :---: | :---: |
| Barramundi | Season | 1 | 312.91 | 0.317 | 0.593 |
|  | Residual | 26 | 985.77 |  |  |
|  |  |  |  |  |  |
| Barred Javelin | Season | 1 | 1261.00 | 7.484 | 0.015 |
|  | Residual | 65 | 168.48 |  |  |
|  |  |  |  |  | 0.085 |
| Blue Catfish | Season | 1 | 3322.80 | 3.121 |  |
|  | Residual | 48 | 1064.80 |  |  |
|  |  |  |  |  |  |
| Bream | Season | 1 | 50.00 | 0.111 |  |
|  | Residual | 7 | 450.00 |  | 0.463 |
|  |  | 1 | 81.82 | 0.992 |  |
| Diamondscale Mullet | Season | 20 | 82.50 |  | 0.883 |
|  | Residual |  |  |  |  |
|  |  | 1 | 23.33 | 0.022 |  |
| Sea Mullett | Season | 68 | 1066.00 |  |  |
|  | Residual |  |  |  |  |

## Fulton's condition index

Analysis of Fulton's condition index between seasons highlighted only a significant difference for Blue Catfish ( $\quad(=0.045$ ), where Fulton's condition index was greater in April 2019 than September 2018.

| Species | Source of variation | df | MS | $F$ | $P$ |
| :--- | :--- | :---: | :---: | :---: | :---: |
| Barramundi | Season | 1 | 0.94 | 1.240 | 0.413 |
|  | Residual | 26 | 0.76 |  |  |
|  |  |  |  |  |  |
| Barred Javelin | Season | 1 | 0.09 | 2.020 | 0.189 |
|  | Residual | 65 | 0.05 |  |  |
|  |  |  |  |  |  |
| Blue Catfish | Season | 1 | 0.04 | 4.420 | $\mathbf{0 . 0 4 5}$ |
|  | Residual | 48 | 0.01 |  |  |


|  |  |  |  |  |  |
| :--- | :--- | :---: | :---: | :---: | :---: |
| Bream | Season | 1 | 0.09 | 1.732 | 0.218 |
|  | Residual | 7 | 0.06 |  |  |
|  |  |  |  |  |  |
| Diamondscale Mullet | Season | 1 | 0.05 | 0.650 | 0.375 |
|  | Residual | 20 | 0.08 |  |  |
|  |  |  |  |  | 0.456 |
| Sea Mullett | Season | 1 | 0.01 | 0.509 |  |
|  | Residual | 68 | 0.02 |  |  |

## Hepatosomatic index

Analysis of the hepatosomatic index between seasons highlighted only a significant difference for Sea Mullet ( $P=0.010$ ), where the hepatosomatic index was less in April 2019 than September 2018.

| Species | Source of variation | df | MS | $F$ | $P$ |
| :--- | :--- | :---: | :---: | :---: | :---: |
| Barramundi | Season | 1 | 0.20 | 3.220 | 0.087 |
|  | Residual | 26 | 0.06 |  |  |
|  |  |  |  |  |  |
| Barred Javelin | Season | 1 | 0.27 | 0.166 | 0.741 |
|  | Residual | 65 | 1.63 |  |  |
|  |  |  |  |  | 0.862 |
| Blue Catfish | Season | 1 | 0.00 | 0.026 |  |
|  | Residual | 48 | 0.05 |  | 0.605 |
|  |  |  |  |  |  |
| Bream | Season | 1 | 3.22 | 0.575 | 0.376 |
|  | Residual | 7 | 5.60 |  |  |
|  |  | 1 | 0.06 | 0.845 |  |
| Diamondscale Mullet | Season | 20 | 0.07 |  | 0.010 |
|  | Residual |  |  |  |  |
|  |  | 1 | 1.56 | 8.192 |  |
| Sea Mullett | Season | 68 | 0.19 |  |  |
|  | Residual |  |  |  |  |

## Gonadosomatic index

Analysis of the Gonadosomatic index between seasons and/or between sexes was possible for a subset of species $x$ sex combinations due to formal statistical analysis being inhibited by the sampling of one or fewer fish of a particular sex in a particular season. Where analyses were possible, a significant difference between the GSI of males and females was detected for Blue Catfish in the April 2019 samples ( $P=0.001$ ), with females exhibiting a greater GSI than Males. Meanwhile, female Bream exhibited greater GSI in September 2018 than in April 2019 ( $P=0.026$ ).

| Species | Source of variation | df | MS | $F$ | $P$ |
| :--- | :--- | :---: | :---: | :---: | :---: |
| Barramundi - Male | Season | 1 | 0.01 | 3.561 | 0.061 |
|  | Residual | 22 | 0.00 |  |  |


|  |  |  |  |  |  |
| :--- | :--- | :---: | :---: | :---: | :---: |
| Barred Javelin | Season | 1 | 3.01 | 1.969 | 0.152 |
|  | Sex | 1 | 5.85 | 3.823 | 0.064 |
|  | Season x Sex | 1 | 1.76 | 1.153 | 0.259 |
|  | Residual | 60 | 1.53 |  |  |
|  |  |  |  |  |  |
| Blue Catfish | Sex | 1 | 0.91 | 65.755 | $\mathbf{0 . 0 0 1}$ |
|  | Residual | 40 | 0.01 |  |  |
| Bream - Female |  |  |  |  | 0.026 |
|  | Season | 1 | 31.13 |  |  |
|  | Residual | 5 | 3.59 |  | 0.552 |
| Diamondscale Mullet | Season |  | 1 | 0.23 | 0.229 |
| - Female | Residual | 16 | 0.99 |  |  |
|  |  |  |  |  | 0.099 |
| Sea Mullet - Female | Season | 1 | 45.13 | 3.029 |  |
|  | Residual | 59 | 14.90 |  |  |

