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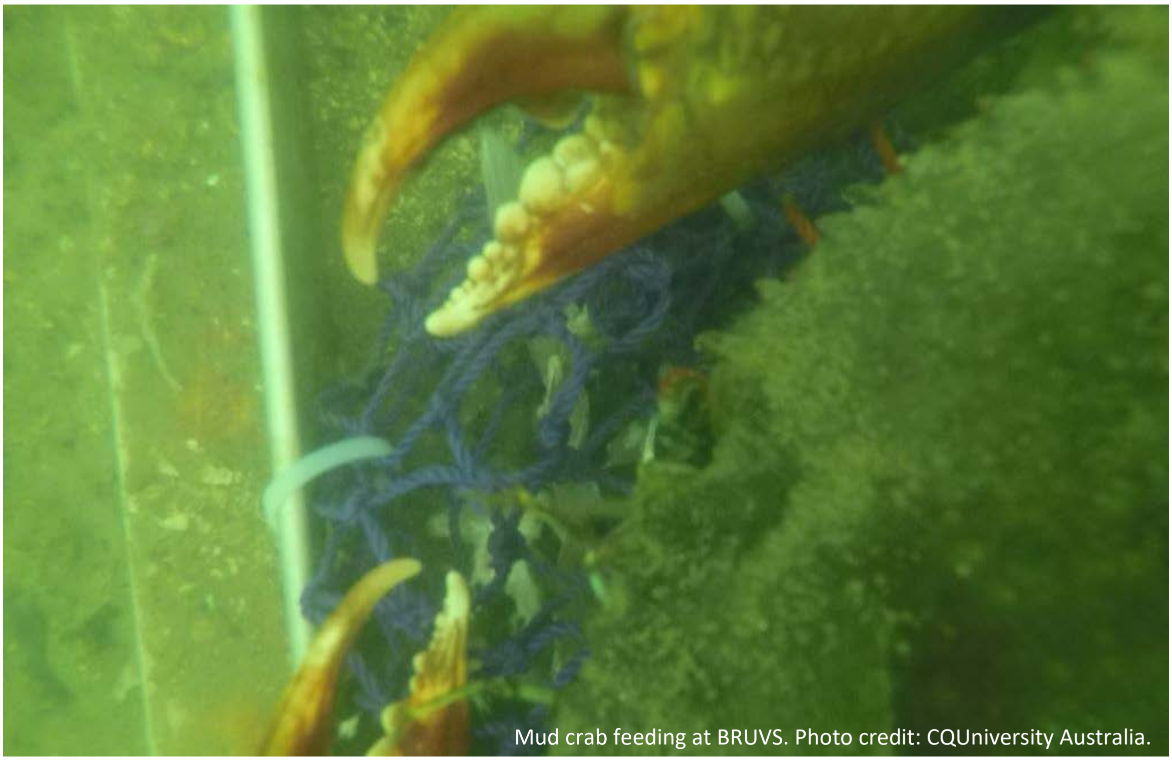
Mud Crab Indicators for the Gladstone Harbour Report Card: Project ISP015-2018


Mud crab feeding at BRUVS. Photo credit: CQUniversity Australia.

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The authors would like to take this opportunity to respectfully acknowledge the Traditional Owners of the land on which we live, work and learn, and pay our respects to the Elders, past, present and future for they hold the memories, the traditions, the culture and hopes of Indigenous Australia. In particular we pay our respects to the peoples on whose Country this research was carried out.

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

- General Fisheries Permit (Queensland Department of Agriculture and Fisheries; Permit Number 192151)
- Animal Ethics Approval (CQUniversity Animal Ethics Committee; Approval Number 20633)
- Authorisation for research in the Great Barrier Reef Marine Park (Approval Number G17/05-027)
- Field Work Risk Assessment (CQUniversity Occupational Health and Safety Unit)

Executive summary

Mud crabs (primarily *Scylla serrata*) are recreationally and commercially important species in Gladstone Harbour, as well as an iconic seafood item, with cultural value to Indigenous Australians. In 2017, CQUniversity proposed four mud crab measures for the Gladstone Harbour Report Card:

- Abundance
- Prevalence of rust lesions
- Sex ratio
- Biomass

Mud crab monitoring was conducted in seven Gladstone Harbour zones for the second consecutive year, over two sampling events in February and June 2018. Scores and grades were calculated using both data sets for the measures: Abundance, Prevalence of rust lesions, and Sex ratio for each of the seven recommended long term monitoring zones in Gladstone Harbour. The three measure scores were averaged to reach a Mud Crab Indicator score and grade for each zone, and for the Harbour.

Similar to 2017, the biomass measure of mud crab condition was not able to be scored in 2018, due to insufficient historical weight data for mud crabs in Gladstone Harbour to develop a benchmark. Once three years of monitoring data are available in 2019, this indicator will also be able to be scored. In 2018, additional data on mud crab condition were collected using the Australian Industry Live Mud Crab Grading Scheme (available at: https://www.c-aid.com.au/wp-content/uploads/Live-Mud-Crab_Grading-Scheme_WEB.pdf). This scheme may also be used in future years for understanding mud crab condition.

In June 2018, additional sampling was undertaken at Eurimbula Creek, 20 km south of Rodds Bay. The creek is within Queensland Government fisheries regulated waters with no crabbing permitted, and was proposed as a comparison site to crabbed zones in Gladstone Harbour, particularly for the Sex ratio measure. Further sampling will be conducted at Eurimbula in February 2019. The mud crab indicator grades for 2018 are provided below.

Zone	Abundance (CPUE)	Prevalence of rust lesions	Sex ratio*	Biomass	Zone score 2018
1. The Narrows	1	1	0	NC	0.67
2. Graham Creek	0.3	1	0.03	NC	0.44
4. Boat Creek	0.25	1	0.29	NC	0.51
5. Inner Harbour	0.52	1	0.02	NC	0.52
6. Calliope Estuary	0.47	1	0.11	NC	0.52
7. Auckland Inlet	0	NC	NC	NC	NC
13. Rodds Bay	0.2	0.90	0.06	NC	0.39
Harbour Average					0.51

* Sex ratio based on legal size limits.

NC = not calculable in 2018.

The scores and grades for the mud crab indicator reflect the variety of pressures on mud crabs in Gladstone Harbour, including commercial fishing, recreational fishing and environmental/habitat condition and over short time periods, and are also potentially influenced by biological variability. Low overall scores (D) were recorded at two of the seven zones: Graham Creek and Rodds Bay. Rodds Bay also scored a D in 2017. No grade has been calculated for Auckland Inlet, as only four mud crabs were caught across the two sampling periods. The small sample size (< 5) means it is not appropriate to calculate grades for this site, except for the Abundance measure. The Narrows was graded B in 2018 and also in 2017.

Low grades for Abundance of mud crabs (E) were recorded in Auckland Inlet and Rodds Bay. Graham Creek, Boat Creek and Calliope Estuary were all graded D and the Narrows was graded A. In 2017, Abundance was high (A) at Boat Creek and Inner Harbour. There are many factors that can influence the catchability of mud crabs, so the lower grade is not necessarily a cause for concern on the basis of a single year. In light of this, the decision was taken to allow Abundance to be scored based on a moving average technique, of the average of the 75th percentile of scores for current and previous sampling years, up to 10 years. This allows the Abundance benchmark to move in response to annual harbour-wide changes in catchability and abundance which are more likely to be natural variations. Some of the possible (non-anthropogenic) factors affecting catchability of mud crabs include moult state of crabs, reproductive cycles, lunar and diel cycles, temperature and water motion.

The Prevalence of rust lesions measure scored an A grade in all zones for which it was calculated in 2018, indicating very low prevalence of this abnormality during the sampling period. Rust lesions have previously been recorded at a much higher prevalence in Gladstone Harbour than reported here (average of 1% in February 2018 and 3% in June 2018 across all harbour zones, compared to up to 37% of mud crabs reported by a study conducted in 2012).

Sex ratios tended towards high proportions of female mud crabs, reflecting the sex-based fishery operating in Queensland. In 2018 the Sex ratio benchmark has been updated in light of new information from an unpublished Australian study of mud crab sex ratios in unfished estuaries. As 2018 is only the second year that the mud crab indicator has been scored, the results presented here should still be considered a pilot study. Data from the no-crabbing area (Eurimbula Creek) is currently being collected, and more information is becoming available to support further development of benchmarks and worst case scenarios (WCSs). The accuracy and reliability of the mud crab grades may improve as more data are collected and all indicators, including biomass, are able to be calculated. A key recommendation of this report is that a review of all measures, benchmarks, sampling zones and times, and the weighting of each measure, is undertaken in 2019, once three years of monitoring data are available.

List of Acronyms

Acronym	Definition
CPUE	catch per unit effort
CW:BW	carapace width: body weight
DAF	Queensland Department of Agriculture and Fisheries
DDT	Dichlorodiphenyltrichloroethane
DNA	deoxyribonucleic acid
FRDC	Fisheries Research and Development Corporation
GHHP	Gladstone Healthy Harbour Partnership
ISP	Independent Science Panel
LTMP	Fisheries Queensland Long Term Monitoring Program for the Mud Crab Fishery
NA	Not available
NC	Not calculable
NSW	New South Wales
NT	Northern Territory
ppt	parts per thousand
RNA	ribonucleic acid
SC	selection criteria
SD	standard deviation
SOI	Southern Oscillation Index
USA	United States of America
WCS	worst case scenario

Contents

Introduction

The mud crab (*Scylla* spp.) is a key fisheries product in Africa, Asia, Australia and the South Pacific (FAO, 2017). In Queensland, the 2015 total catch was approximately 1,000 tonnes worth AUD18.7 million (DAF, 2017). Mud crabs (primarily *Scylla serrata*, and potentially *S. olivacea*) are a recreationally and commercially important species in Gladstone Harbour, as well as an iconic seafood item, with cultural value to Indigenous Australian peoples (Brewster, 2015). As a result of their commercial importance to the fishing and aquaculture industries, the biology, ecology and biochemistry of mud crabs is relatively well known. The green mud crab, *S. serrata*, has previously been suggested as a potential biomonitoring species in tropical coastal marine environments of Australia (Negri *et al.*, 2009; van Oosterom *et al.*, 2010).

Because of the importance of mud crabs to the local community and stakeholders, the GHHP Independent Science Panel (ISP) selected mud crabs as an important indicator species to assess the health of Gladstone Harbour, and nominated candidate indicators for Gladstone Harbour including: size distribution of adult mud crabs; abundance of adult mud crabs; and visual health assessment of adult mud crabs (McIntosh *et al.*, 2014). The Gladstone Healthy Harbour Partnership (GHHP) commissioned CQUniversity to develop mud crab indicators for the Gladstone Harbour Report Card (Project ISP015-2017). CQUniversity conducted a review and analysis of potential mud crab indicators, and proposed four mud crab measures to be incorporated into the mud crab indicator for the Gladstone Harbour Report Card:

- Abundance (catch per unit effort – CPUE)
- Prevalence of rust lesions
- Sex ratio
- Biomass.

Ideal indicator species are sedentary, easily identifiable, abundant, long-lived, available to sample throughout the year, large enough to provide sufficient tissue for analysis, resistant to handling stress and tolerant to environmental variations in physicochemical parameters (Rainbow, 1995). To be regionally relevant, the species should also be naturally occurring in the environment to be tested. With the exception of when females are migrating to spawn, *Scylla* spp. exhibit a relatively small foraging range, meaning that compared to finfish, mud crabs may provide better detail of spatial and temporal pollution impacts (Hyland *et al.*, 1984; van Oosterom *et al.*, 2010; Alberts-Hubatsch *et al.*, 2016). The relative ease of capture and recapture of mud crabs makes them a suitable candidate as an indicator species.

Abundance

Abundance of mud crabs caught during standardised independent monitoring gives an estimate of changes in total abundance and population size (Dumas *et al.*, 2012; Meynecke *et al.*, 2012; Alberts-Hubatsch *et al.*, 2016) of mud crabs through time and between areas. Discrepancies in abundance can be due to capture technique, sampling areas and sampling times, so it is necessary to minimise monitoring variation in seasonality, catch and sampling techniques. Consistent methodologies are employed during each catch period, to ensure opportunity for direct comparison of CPUE and abundance at different zones and between years. Abundance can reflect a wide variety of natural and anthropogenic impacts on a population (Alberts-Hubatsch *et al.*, 2016). Factors influencing abundance of mud crabs may include localised and regional fishing pressure, habitat availability and habitat condition, availability of food and proximity to suitable nursery grounds for the settlement of mud crab megalopae and metamorphosis to immature crabs.

A review of the literature provided information from Australia and elsewhere on factors affecting mud crab abundance and distribution. The abundance and distribution of *Scylla* spp. in the Philippines was measured using baited lift nets and cylindrical bamboo traps (Walton *et al.*, 2006). The cylindrical bamboo traps were considered the best estimation of catch per unit effort (CPUE), as the method maintained constant effort (Walton *et al.*, 2006). *S. olivacea* dominated all *Scylla* spp. caught, with no correlations found between CPUE, salinity, temperature or tidal variation (Walton *et al.*, 2006). A variation in CPUE was detected between years, related to a decline in the relative abundance of *S. olivacea* in the area, suggesting variations in recruitment and increased fishing pressures (Walton *et al.*, 2006). In a similar study, abundance of *S. olivacea* was used as an indicator of mangrove habitat ecological function. Abundance was also estimated using CPUE from an experimental standardised trapping grid (Walton *et al.*, 2007). While a similar decrease in CPUE of *S. olivacea* was observed, abundance of other crab species in the area increased, suggesting that the area was still a suitable habitat, but that fishing pressures and recruitment limitations had impacted on the abundance of *S. olivacea* (Walton *et al.*, 2007).

Climate has been shown to impact the abundance of mud crabs so there is also potential for this indicator (Meynecke *et al*., 2012) to be used to monitor climate effects on mud crabs in the longer term, as well as in overall fishery management decisions. Exposure studies of juvenile *S. serrata* collected from NT, Australia demonstrated their survivability and tolerance of temperatures ranging from 25-35°C, with survival ranging from 94-98%. However, exposure of the crabs to a temperature of 20°C resulted in only a 36% survival rate (Ruscoe *et al.*, 2004), indicating poor tolerance to colder temperatures. *S. serrata* is more tolerant of lower temperatures than the black mud crab *S. olivacea* (Keenan *et al.*, 1998). Expected global warming and rising sea temperatures in the future may benefit some *S. serrata* populations, which tend to thrive in warmer waters. However, global warming is also predicted to increase the occurrence of extreme La Niña events (Cai *et al.*, 2015), and the associated higher rainfall, lower water salinity and ocean acidification could potentially be detrimental to *S. serrata* populations and contribute to habitat degradation.

In Australia, *S. serrata* tends to be dominant in mangrove habitats which typically have salinities around 34 ppt (Meynecke *et al.*, 2012). However, such populations are also vulnerable to flooding events, which reduces water salinity and in turn results in increased mortality (Meynecke *et al.*, 2012). Whilst previous studies have demonstrated that *S. serrata* does not survive in very low salinity (<5 ppt) (Chandrasekaran and Natarajan, 1994; Ruscoe *et al.*, 2004), as an estuarine species *S. serrata* is able to tolerate a reasonably wide range of salinities, with Williams and Hill (1982) reporting that salinity (ranging from 24-35 ppt) was not correlated with numbers caught. Ruscoe *et al.* (2004) also reported that survival of juvenile *S. serrata* was not significantly different in treatment groups exposed to water between 5-40 ppt.

Prevalence of rust lesions

In 1994, the first records of “rust spot” shell lesions in mud crabs were reported by commercial fishers in Gladstone Harbour (Andersen and Norton, 2001). The disease is not infectious and may possibly be related to inhibition of calcium uptake following sublethal copper exposure although this has not yet been experimentally confirmed (Andersen and Norton, 2001). The disease was studied extensively in the early 2000’s and data on prevalence of the disease in mud crab populations are available for Gladstone Harbour. The disease has also been reported from the Fitzroy River and (with much lower prevalence) from Moreton Bay, Ayr (Andersen and Norton, 2001) and Stanage Bay (Dennis et al. 2016).

Rust spot lesions initially start as an orange discolouration on the crab’s carapace, before progressing to penetration and degradation of the affected carapace area (‘perforation’), with resultant exposure of the soft tissues beneath the carapace (Andersen *et al.*, 2000; Andersen and Norton, 2001). This disease has the potential to damage the mud crab industry around Gladstone Harbour, making it a potential indicator of high interest to members of the public. A method to objectively measure and grade the severity of lesions on crab shell carapace was developed and described by Andersen *et al.* (2000), and is based on the size of the lesion, and whether the carapace has been perforated.

In 2011-12, there were reported increases in the incidence of disease in finfish and crabs in Gladstone Harbour. A veterinary analysis conducted by Dennis *et al.* (2016) in early 2012 found rust spot lesions on 37% of *S. serrata* caught in Gladstone Harbour. This is compared with a prevalence of 14% of the crabs caught in Stanage Bay and 7% of the crabs caught in Rodds Bay and Turkey Beach during the same period (Dennis *et al.*, 2016). Prevalence of rust spot lesions measured during the Dennis *et al.* (2016) study was greater than previously recorded prevalence in the same region. Andersen *et al.* (2000) recorded a rust spot prevalence of 22% from Gladstone Harbour in the late

1990s. The 2017 CQUniversity monitoring project found a low prevalence of rust spot lesions in the Gladstone Harbour population (ranging from 2% at the Narrows to 15% in Auckland Inlet, with a harbour average of 7.8%).

Due to the fact that rust spots are not continuously observed in the Gladstone Harbour region, the prevalence of rust spot at any given time is an overall indicator of environmental state. There is potential for rust spot to impact on the local fishery, as it impacts on the seafood 'grade' of crabs. Recording the presence of rust spot is a relatively straight forward and non-destructive monitoring tool. A worst case scenario (WCS) was determined using the results of previous studies in which prevalence was found to be higher than background levels (e.g. Dennis et al. 2016).

Sex ratio

Recreational and commercial fishing are major factors driving the management of Queensland's *S. serrata* population, and exert pressure on fished stocks. In Queensland, only male mud crabs over 150 mm carapace width (measured across the ninth posteriolateral spines, referred to as 'spine width' in this report) may be retained by either commercial or recreational fishers. Female mud crabs, and any mud crabs under 150 mm may not be retained. Identifying the gender of most crabs is quick and easy, and in areas where a sex-based fishery is enforced, changes in the ratio of males to females may be indicative of a change in fishing pressure (Heasman, 1980; Williams and Hill, 1982; Pillans *et al.*, 2005; Alberts-Hubatsch *et al.*, 2016). In unfished populations of *S. serrata*, the male:female ratio can be as high as 3:1 (Alberts-Hubatsch *et al.*, 2016). A study of marine reserves in Moreton Bay, South East Queensland, identified a male:female sex ratio of 2:1 in mature crabs (>150 mm) (Pillans *et al.*, 2005). Earlier fisheries research found a male:female ratio of only 1:3 in a heavily exploited region of southern Moreton Bay, and a ratio of 1.2:1 in the lightly exploited Pumicestone Passage (Hill, 1984). More recent research in protected estuaries in northern NSW found a gender ratio of approximately 2 males to each female mud crab (regardless of size) (Butcher, 2004). Shifts in sex ratio not only have implications for dynamics of the crab population and reproductive success, but may also influence ecosystem processes due to the different behaviours of the sexes. For example, male, rather than female mud crabs create burrows (Bonine *et al.*, 2008) which may aid in the process of bioturbation; and female crabs often migrate offshore to spawn (Knuckey, 1999).

Besides fishing pressure, another influence on the sex ratio of crabs is seasonal changes, and this needs to be considered when interpreting indicator scores. For example, the mud crab population in Princess Charlotte Bay in northern Queensland was dominated by females only during August (Hill, 1984). In observations on the mature Portunid crab *Callinectes sapidus* in a tributary of Chesapeake Bay, Harding and Mann (2010) found a dominance of male crabs when day lengths were above 13.4 hrs. However, as both day length and water temperature decreased, female crabs were up to four times more abundant than males, as males migrated to up-river habitats. In *Scylla* spp. from the Malaysian coast male:female sex ratios were found to change during the monsoon season when females migrate for spawning (Ikhwanuddin *et al.*, 2011). Similar results were found between December and February in the Northern Territory (Knuckey, 1999). In other areas no temporal patterns were observed, for example in a New Caledonian study of *S. serrata* populations, male dominance was apparent in all sampling periods, with proportions ranging from 50-100% of the population (Dumas *et al.*, 2012).

Dumas *et al.* (2012) also found a spatial difference in the ratio of males to females, with male crabs significantly more likely to be found inside burrows (86.1% of catch), compared to 74.4% of catch caught outside burrows (Dumas *et al.*, 2012). Spatially, females were significantly more abundant in coastal mangroves (Dumas *et al.*, 2012). Tagging experiments to assess habitat movement of *S. serrata* in Moreton Bay (Hyland *et al.*, 1984) from 1976 to 1981 identified two types of movement, one of which was sex specific. In an area with direct sea access, males and females moved similar distances, but in a channel environment, female movement (mean 6.6 km) was significantly greater than male movement (mean 3.7 km) (Hyland *et al.*, 1984).

Finally, the sex of the crab may also affect the probability of capture and recapture. Williams and Hill (1982) showed differences in the vulnerability to capture among *S. serrata* of different sizes, with vulnerability increasing strikingly for males over 130 mm and females over 140 mm. In 1980, Heasman found a difference between the capture of different sexes of moulting sub adults of *S. serrata* in Moreton Bay. The study found that recently moulted females less than 150 mm were not captured in commercial crab pots, whilst recently moulted males of down to 120 mm were captured (Heasman, 1980). The reason proposed was that behavioural differences between the sexes and among size classes led to unequal vulnerability to capture.

Biomass

Similar to abundance, biomass can be an important indicator of both species and ecosystem health (Alberts-Hubatsch *et al.*, 2016). The carapace width to body weight ratio (CW:BW) is an indicator of health in mud crabs, with high ratios indicating high ecosystem productivity and food densities (Bolger and Connolly, 1989; Ikhwanuddin *et al.*, 2011; Grubert *et al.*, 2012). It is simple to measure and non-destructive. This measure can be compared across zones as an indicator of ecosystem productivity and can be used to estimate the amount of obtainable edible meat from an individual, or a sample of mud crabs. From a fisheries perspective, biomass is likely to give a comparative estimation of condition of mud crabs across zones, and to signal changes to mud crab health and ecosystem productivity through time. There are no historical data with which to determine a benchmark for this indicator, so a relative scoring system was developed as an interim measure, using current data, for future consideration (Flint *et al*., 2017). It is important to note that biomass will vary significantly between summer (when mud crabs moult more regularly) and winter (when mud crabs moult less often, and tend to be 'fuller' and heavier for their size). Missing limbs would also affect the ratio.

A study in New Caledonia conducted by Dumas *et al.* (2012), found that environmental and fishing variables were explained 14.9% of the size variability of *S. serrata* (Dumas *et al.*, 2012). Mangrove type was found to be a major predictor for crab size with larger crabs in coastal areas, compared to estuarine areas (Dumas *et al.*, 2012). Exposure studies of juvenile *S. serrata* collected from NT, Australia observed maximum growth rates at a water temperature of 30°C (Ruscoe *et al.*, 2004). As there have been no comparative studies conducted on the three different continents where *S. serrata* reside, it is not known whether size and biomass variations reported in the literature are due to sampling methods, genetic diversity or phenotypic plasticity (Alberts-Hubatsch *et al.,* 2016).

Objectives

The overall objectives of this project are to:

1. Monitor mud crabs across the long term monitoring sites in Gladstone Harbour using the methods developed by Flint et al. (2017).

2. Provide report card grades and scores for the 2018 Gladstone Harbour Report Card.

Methods

Field methods

Two mud crab surveys were undertaken in 2018 (Table 1), representing a summer (warm, wet season) and winter (cool, dry season) sample. The seven monitoring sites (Figure 1) were previously chosen through a quantitative selection process (Flint *et al.*, 2017).

Table 1: Gladstone zones/sites sampled during February and June 2018.

Zone/site	Survey 1	Survey 2
Zone 1: Narrows	27 February	24 June
Zone 2: Graham Creek	27 February	24 June
Zone 4: Boat Creek	26 February	23 June
Zone 5: Inner Harbour	25 February	22 June
Zone 6: Calliope Estuary	26 February	23 June
Zone 7: Auckland Inlet	25 February	22 June
Zone 13: Rodds Bay	28 February	25 June

Sampling dates and times were determined by tidal cycles. Surveys were conducted on dates when low tide was between 10.30am and 3.00pm. Pots were set at least three hours before the low tide, and collected at least two hours after the low tide, resulting in soak times of at least five hours per pot. To comply with Animal Ethics Approval pots were placed so that they would still be submerged at low tide (preventing exposure mortality of any fish caught in the pots). Pots were placed as close as possible to mangrove habitats within this parameter.

At each site and sampling event, 20 heavy duty 4-entry round collapsible crab pots were set a minimum of 100 m apart. The exception to this was Zone 4 Boat Creek, as only 15 pots can be accommodated in this smaller system. Collapsible crab pots were purchased from a local tackle store, as they are easy to transport, assemble on the vessel and replace (Fisheries Queensland, 2009). Each pot was baited with one large sea mullet (*Mugil cephalus*) head, and all floats were attached with 10 m ropes and marked with researcher contact details and the Fisheries Queensland research permit number. Every float had a unique identifying number to allow any missing pots to be identified quickly during retrieval. The opening of each pot was secured with a cable tie, so that if crabs were removed by others this could be detected and recorded on retrieval.

At each sampling site, the following information was recorded:

- Zone and site name;
- GPS location;
- Date;
- Set time and retrieval time for each uniquely identified pot;
- The total number of animals of each species caught in every pot, and the sex of all mud crabs caught; and
- Water quality parameters (temperature, dissolved oxygen, conductivity, pH, turbidity, total dissolved solids, oxidation reduction potential and salinity) measured using a YSI ProDSS Multiparameter Sampling Instrument, recorded once before setting the first pot and once after retrieving the final pot.

For every mud crab captured at each site, the following information was recorded:

- Species;
- Sex;
- Carapace width (notch width) (mm);
- Weight (g);
- Abnormalities: type, body location, dimensions of rust spot lesions, grade of rust spot lesions (source Andersen, 2003; Appendix 1).

All bycatch species (including blue swimmer crabs, fish and other crabs) were also recorded. Blue swimmer crabs were weighed, measured, and checked for abnormalities before release. All catch was released alive at the site of capture. Used baits were kept on board the vessel for later disposal on land, and not discarded at the sampling site, to reduce interference with commercial and recreational mud crabbers in the area.

Eurimbula Creek methods

Sampling was also conducted at Eurimbula Creek, located south of Rodds Bay in the Baffle Catchment, on 26 June 2018. The field methods used at Eurimbula Creek were identical to field sampling at each Gladstone Harbour zone, described above. As the creek is within fisheries regulated waters prohibiting crabbing (map available at: https://www.daf.qld.gov.au/__data/assets/pdf_file/0004/87052/Eurimbula-Creek-FRW-162.pdf), data from Eurimbula will be used as a comparison to crabbed zones in Gladstone Harbour. Full analysis of the Eurimbula data will be provided in 2019, once two seasons of sampling have been conducted.

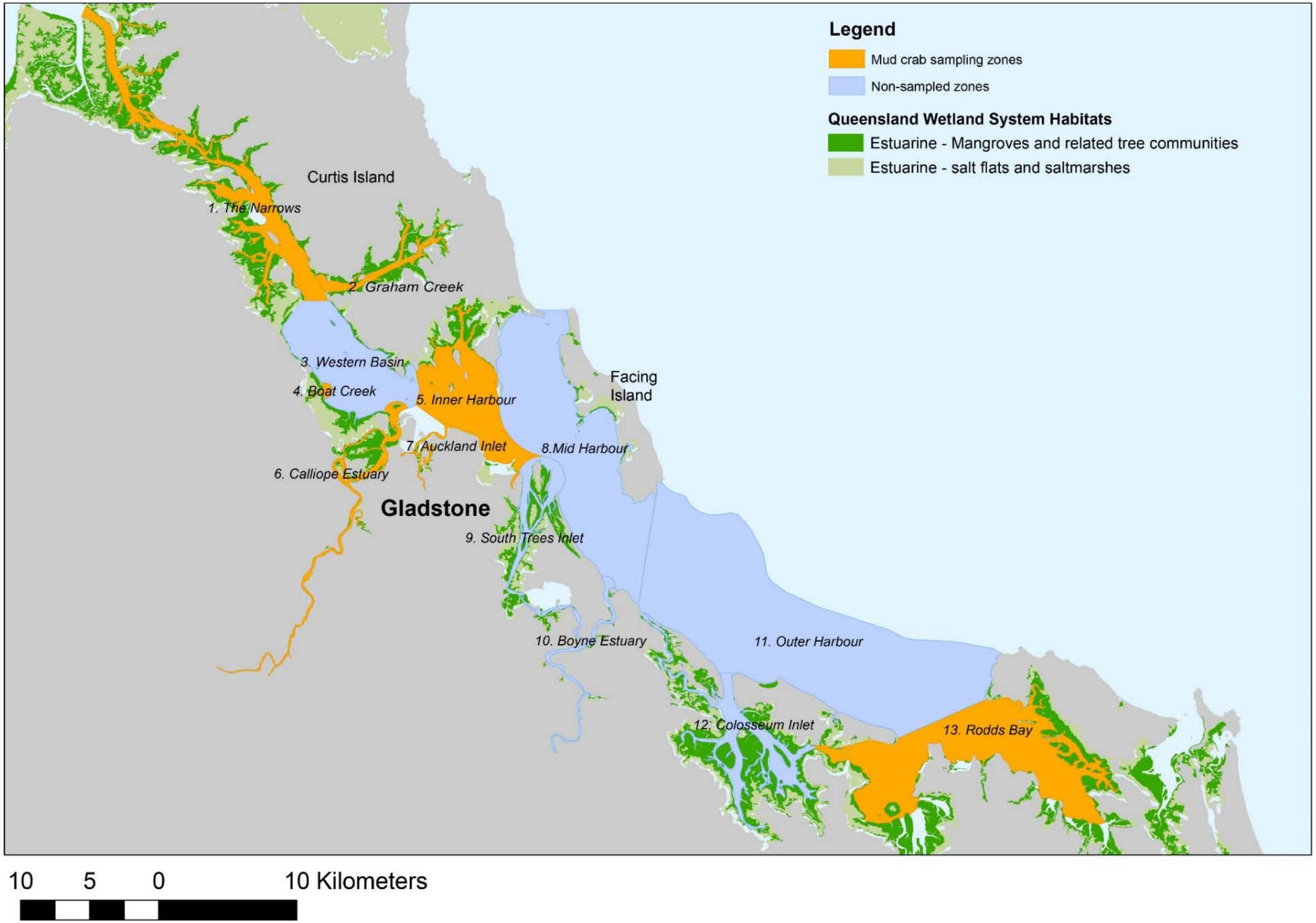


Figure 1: Map of the Gladstone Harbour zones showing recommended long-term mud crab monitoring sites.

Data analysis

Data from the first (25-28 Feb 2018) and second (22-25 Jun 2018) field sampling events were analysed separately and together. Exploratory analyses included descriptive statistics, for example distribution plots (kernel density), and box plots for visual comparisons of differences and variance around the mean. Welch's unequal variances t-tests were used to test the hypothesis that two populations (of different sample sizes and variances) have equal means.

All analyses were conducted in R version 3.4.1 (https://www.r-project.org/).

Scoring, grading and aggregation

The mud crab measures were calculated for each Zone, as follows:

- **Abundance** (CPUE) in each zone

$$= \frac{\text{(total number of mud crabs)}}{\text{(number of pots set)}}$$

- **Prevalence of rust lesions** in each zone

$$= \frac{\text{(number of crabs with rust lesions)}}{\text{(number of crabs assessed for rust lesions)}}$$

- **Biomass** as a condition measure of individual male and female mud crabs, calculated for each mud crab and then averaged for each zone (where width is measured in mm and weight in g)

$$= \frac{\text{carapace (notch) width}}{\text{body weight}}$$

- **Sex ratio** based on oversize mud crabs, for each zone

$$= \frac{\text{(number of male mud crabs > 150 mm)}}{\text{(number of female mud crabs > 150 mm)}}$$

The formulae provided in Table 2 were used to score the mud crab measures, comparing each index value against a pilot benchmark and a worst case scenario (WCS) value. Using this method, index values worse than the WCS score a 0, while index values better than the benchmark score a 1 and all other index values range between these bounds. The method for determining benchmark and WCS values for each measure is described by Flint et al. (2017), with two revisions accounting for new information in 2018. A similar approach to scoring between best case and worst case thresholds is taken for environmental indicators in the South East Queensland Report Card (Healthy Waterways, 2007) and the Fitzroy Basin Report Card (Flint et al., 2017). The Gladstone Harbour Report Card grading system is provided in Table 3.

For one of the four recommended measures, biomass (CW:BW), calculated separately for male and female mud crabs, the only available data on which to base scoring was collected by this project during 2017 and 2018. For this reason, Flint et al. (2017) recommended that the measure is retained, but not scored, until at least three years of mud crab monitoring data are available to calculate baselines.

Sex ratio of legal-sized crabs (> 150 mm carapace spine width, which is equivalent to a crab with 143 mm notch width) is calculated against a 'minimally disturbed' benchmark from the international literature. In 2017 a conservative ratio of 3:1 (*sec.* Alberts-Hubatsch *et al.*, 2016) was used as the benchmark for Sex ratio (Flint *et al.*, 2017). In 2018 an unpublished thesis describing sex ratios in unfished Australian estuaries was made available (Butcher, 2004). The sex ratio in the thesis, 2 males:1 female, was equivalent to results in a separate study from an uncrabbed area in Moreton Bay (Pillans et al., 2005). As a result of this information, the benchmark for Sex ratio has been updated to 2:1 for 2018. In 2019, when the full set of data are available for an unfished region in Central Queensland (Eurimbula Creek, which is located approximately 20 km south of Rodds Bay), this benchmark will again be reviewed.

The benchmark for the Abundance measure has also been updated in 2018. In 2017, the 75th percentile of the 2017 scores was used as the benchmark. In 2018, a moving average of the 75th percentile of this year (2018) and previous years (2017) scores has been applied. It is recommended that this approach continues in 2019 and beyond, using a 10 year moving average methodology.

Table 2: Pilot benchmarks and scoring method for each of the four recommended measures. NC = not calculable.

Measure	Benchmark	Worst case scenario	Method of calculation
	Selected	**Selected**	
Abundance (CPUE)	2017: 3.5 crabs/pot (75th %ile of 2017 scores) 2018: 2.5 crabs/pot (moving average of 75th %ile of 2017 and 2018 scores) 2019+: Moving average of 75th %ile of scores for current and previous years, up to 10 years	0.25	The function used to calculate scores for abundance is: 1-((x-B)/(WCS-B)) Where: x = recorded CPUE B = benchmark (2.5) WCS = worst case scenario (0.25)
Prevalence of rust lesions	4% = 0.04	35% = 0.35	The function used to calculate scores for prevalence is: 1-((x-B)/(WCS-B)) Where: x = recorded prevalence B = benchmark (0.04) WCS = worst case scenario (0.35)
Biomass	NC	NC	No data are available for weight of mud crabs in Gladstone Harbour until 2017 and 2018. This measure is recommended for inclusion in the Report Card, but should not be scored until at least three years of mud crab weight data are available to calculate benchmarks. More data from the unfished area (Eurimbula Creek) will also provide a good comparison.
Sex ratio	2017: 3 (based on unfished tropical mud crab populations from the literature, Alberts-Hubatsch et al., 2016) 2018: 2 (based on new information from Australian estuaries) 2019+: ratio from an unfished Central Queensland population at Eurimbula	0.25	The function used to calculate scores for sex ratio is: 1-((x-B)/(WCS-B)) Where: x = recorded sex ratio (M:F) B = benchmark (2) WCS = worst case scenario (0.25)

Table 3: Gladstone Harbour Report Card grading scale (Source: GHHP, 2015).

Score	Grade
>=0.85	A
>=0.65, <0.85	B
>=0.5, <0.65	C
>=0.25, <0.5	D
0, <0.25	E

Results

Results of mud crab sampling in Gladstone Harbour, February and June 2018

Abundance and size

A total of 175 mud crabs were caught in the seven Gladstone Harbour zones in February 2018. Of these, 62 were male and 111 were female. A total of 163 mud crabs were caught across all Gladstone Harbour zones sampled in June 2018 including 67 males, 95 females, and one crab for which the sex was not recorded (excluded from further analysis).

The average size of mud crabs caught in February 2018 was 142.53 mm carapace notch width (Table 4), smaller than crabs caught in June 2018 (average 150.37 mm carapace notch width, Table 4) or in June and July 2017 (149.16 mm and 155.00 mm respectively). Smaller crabs were also caught in summer during monitoring conducted by the Fisheries Queensland Long Term Monitoring Program (LTMP; also provided in Table 4 for comparison; more details of crabs caught in the LTMP are provided by Flint et al., 2017).

Results of Welch Two sample t-tests found that females caught in February 2018 were significantly larger than males caught in February ($t = -4.9439$, $df = 97.055$, $p < 0.001$; Figure 2), and females caught in June 2018 were also significantly larger than males caught in June ($t = -7.1795$, $df = 138.17$, $p < 0.001$; Figure 3).

The largest average mud crab size in February 2018 was recorded for Graham Creek (mean notch width 181 mm) and the smallest for Calliope Estuary (59 mm) (Figure 4), while the largest average mud crab size in June 2018 sampling was recorded for Auckland Inlet (mean notch width 164.00 mm, but with only two crabs recorded) and the smallest again at Calliope Estuary (141.00 mm) (Figure 5). In 2017 samples the smallest average mud crab size was recorded at Boat Creek.

Table 4. Notch width (in mm) of mud crabs caught in February 2018, June 2018 and by the LTMP in the period 2001-2009.

	FULL SAMPLE			MALES			FEMALES		
	February 2018 data	**June 2018 data**	**Historical data LTMP (2001-2009)**	**February 2018 data**	**June 2018 data**	**Historical data (2001-2009)**	**February 2018 data**	**June 2018 data**	**Historical data (2001-2009)**
Mean	142.53	150.37	145.45	131.92	138.88	135.12	148.45	158.43	151.67
Standard deviation	20.79	19.39	20.74	23.18	17.38	18.65	16.72	16.61	19.43

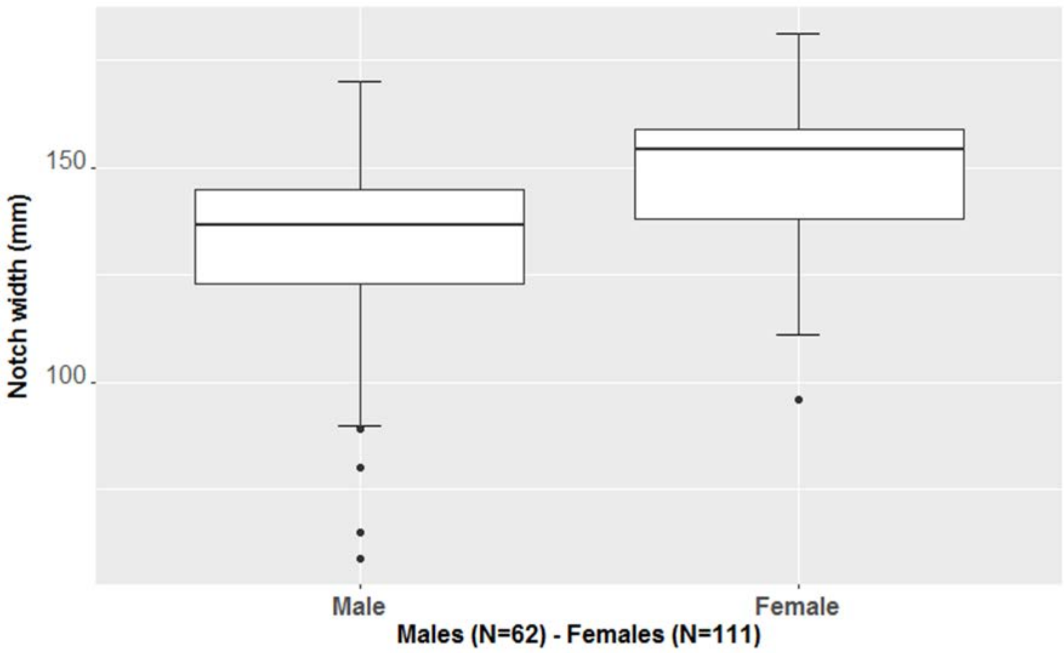


Figure 2: Notch width (mm) distribution of male and female mud crabs caught in February 2018.

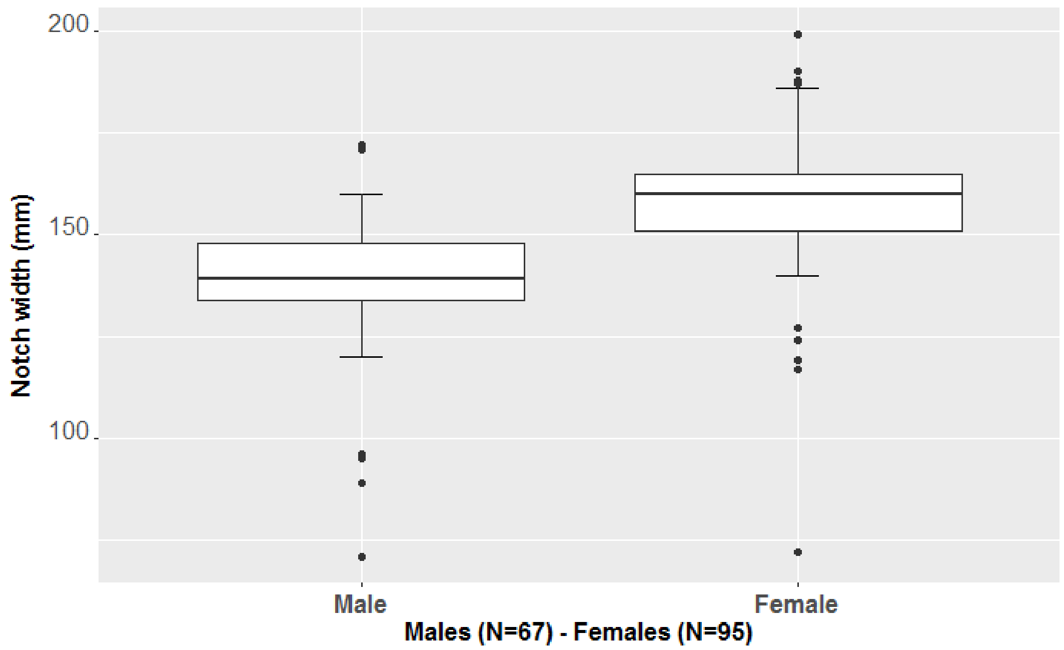


Figure 3: Notch width (mm) distribution of male and female mud crabs caught in June 2018.

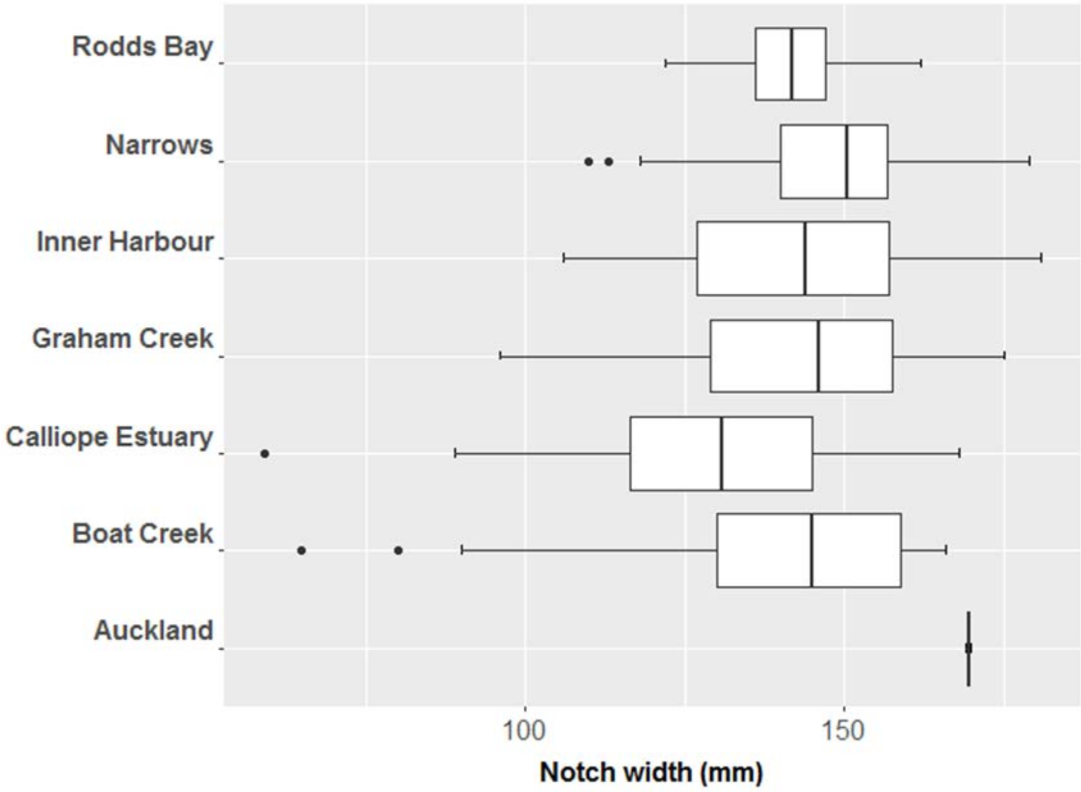


Figure 4: Notch width (mm) of mud crabs caught in February 2018, by zone.

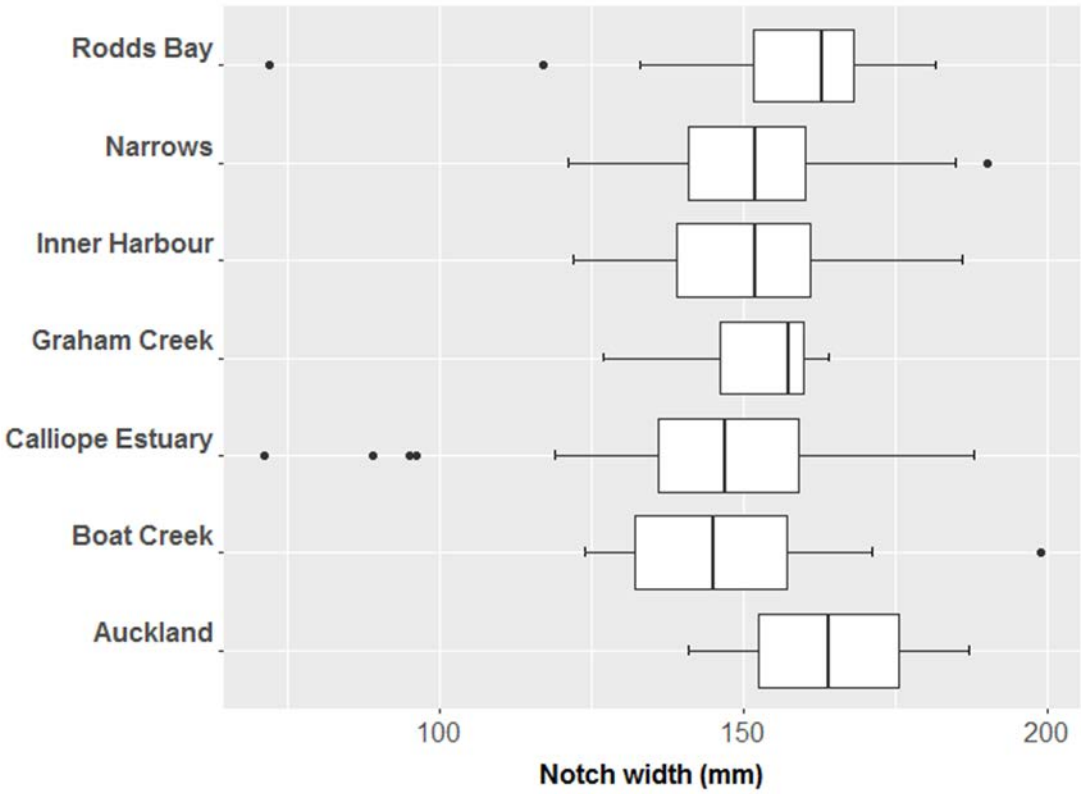


Figure 5: Notch width (mm) of mud crabs caught in June 2018, by zone.

The average weight of mud crabs caught in February 2018 was 715.47 g, and in June 2018 was 762.28 g, similar to the averages of 718.44 g and 741.28 g reported in June and July 2017 respectively (Table 5, Figures 6 and 7). Unlike 2017, results of a Welch Two sample t-test found that there was no significant difference between weight of females and weight of males caught in February 2018 ($t = -0.71441$, $df = 83.802$, $p = 0.477$), however there was a significant difference in the weight of female and male crabs in June 2018 at the $p < 0.05$ level ($t = -2.1234$, $df = 104.65$, $p = 0.036$).

The zone with the highest average mud crab weight in February 2018 was Auckland Inlet (mean weight 1691.00 g) however only two mud crabs were caught at this zone so the weight is biased by a small sample size effect. The next highest average mud crab weight was recorded at Bot Creek (748.69 g) (Figure 8). In June 2018 the zone with the highest average mud crab weight was Rodds Bay (811.05 g) (Figure 9).

Table 5: Weight (g) of mud crabs caught in February 2018.

	FULL SAMPLE		MALES		FEMALES	
	February 2018 data	June 2018 data	February 2018 data	June 2018 data	February 2018 data	June 2018 data
Mean	715.47	762.28	693.06	712.16	727.99	797.63
Standard deviation	266.36	238.09	353.94	289.20	202.54	187.93

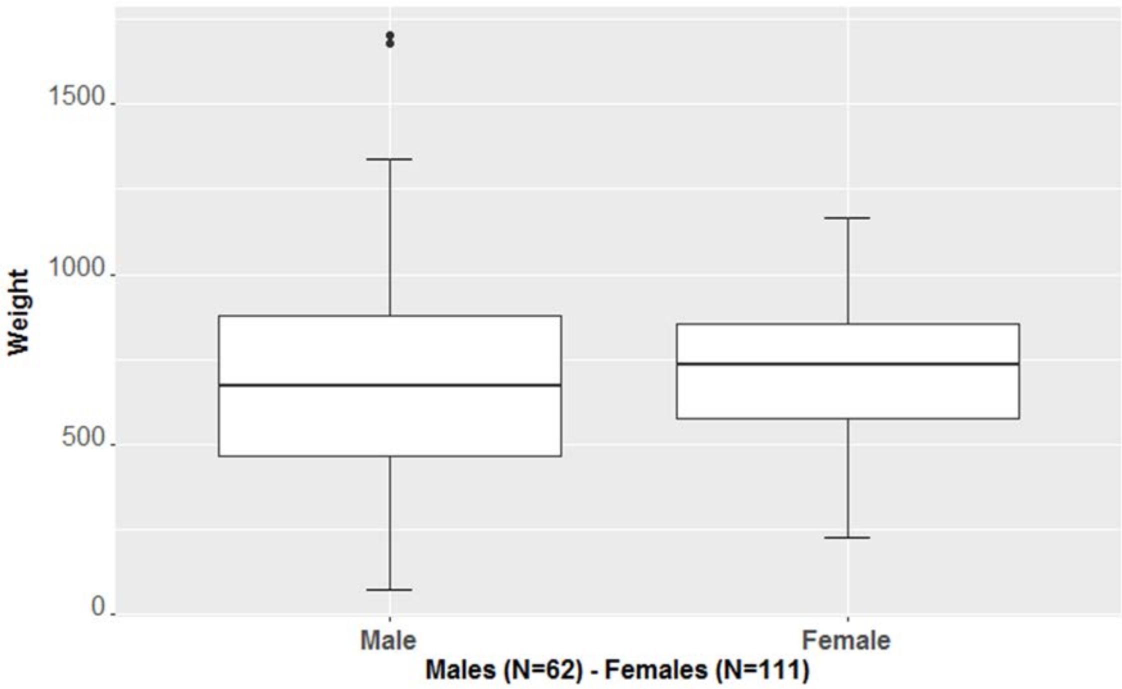


Figure 6: Weight (g) of male and female mud crabs caught in February 2018.

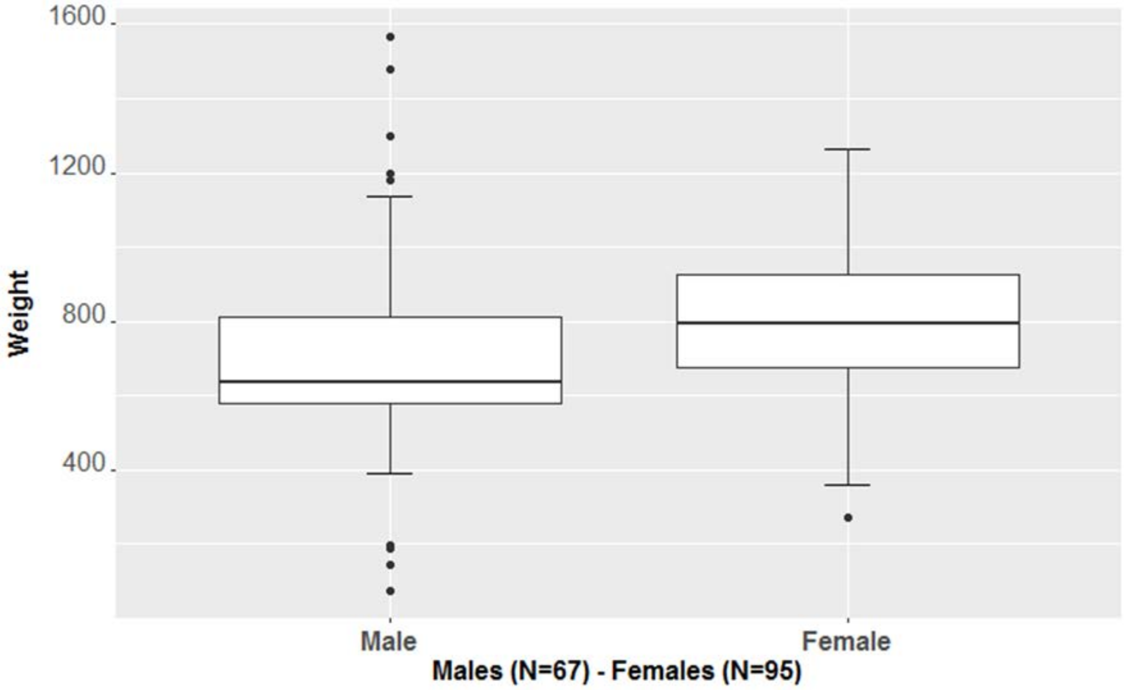


Figure 7: Weight (g) of male and female mud crabs caught in June 2018.

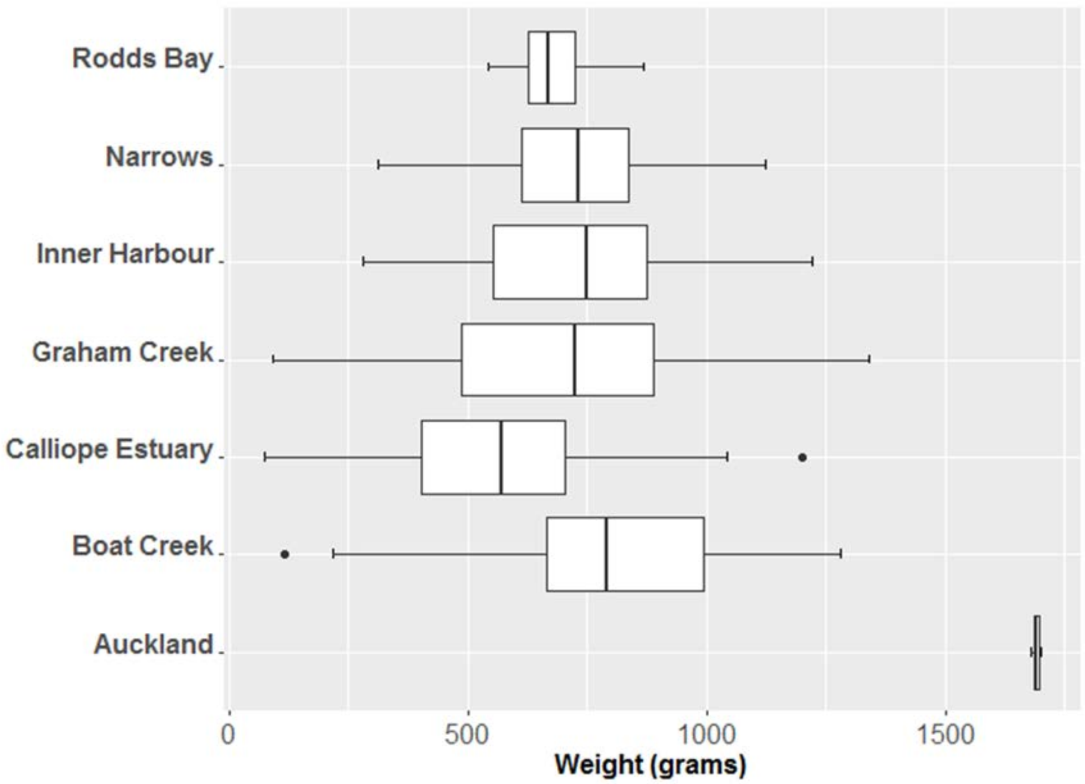


Figure 8: Weight (g) of mud crabs caught in February 2018, by zone.

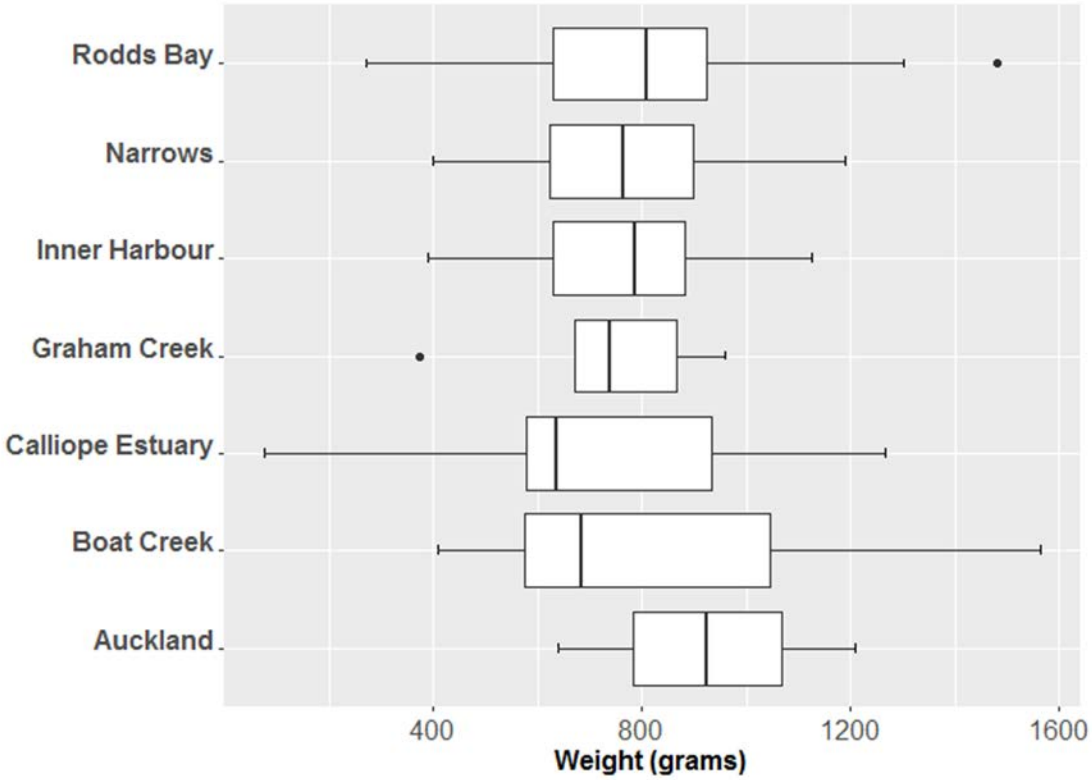


Figure 9: Weight (g) of mud crabs caught in June 2018, by zone.

In February 2018 total catch per unit effort (CPUE) was again highest at the Narrows (3.5) and lowest at Auckland Inlet (0.1) (Table 6, Figure 10). CPUE was also highest at the Narrows in June 2018 (3.0) and lowest at Auckland Inlet (0.1) (Table 6, Figure 11). CPUE at the highest catch zone, the Narrows, was lower in both 2018 samples than in June 2017 (4.62) but higher than in July 2017 (2.75).

Table 6: Catch per unit effort in February 2018 and June 2018, by zone.

Zone	**Zone name**	**February 2018 data**			**June 2018 data**		
		# Pots deployed	**Total # mud crabs caught**	**CPUE**	**# Pots deployed**	**Total # mud crabs caught**	**CPUE**
1	Narrows	20	70	3.5000	20	60	3.0000
2	Grahams Creek	20	28	1.4000	20	11	0.5500
4	Boat Creek	16	13	0.8215	17	15	0.8824
5	Inner Harbour	20	29	1.4500	20	28	1.4000
6	Calliope Estuary	20	23	1.1500	20	29	1.4500
7	Auckland Inlet	20	2	0.1000	20	2	0.1000
13	Rodds Bay	20	10	0.5000	20	19	0.9500
	Harbour wide average			**1.3**			**1.2**

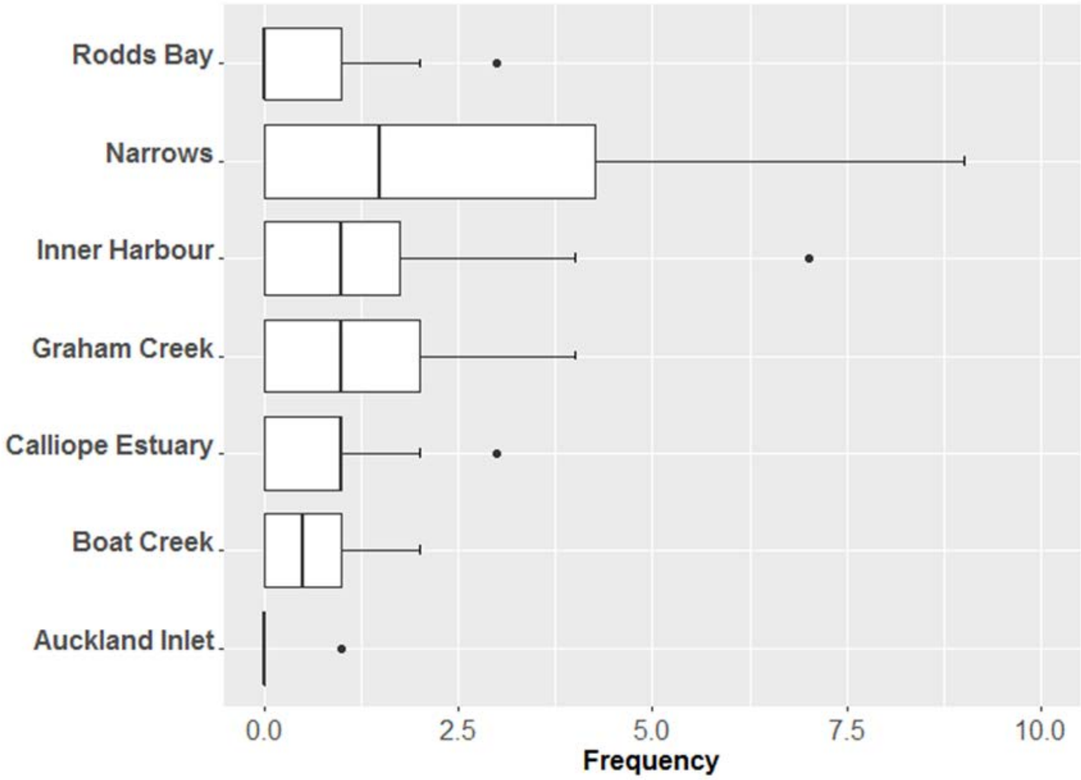


Figure 10: Number of mud crabs in each pot set in February 2018, by zone.

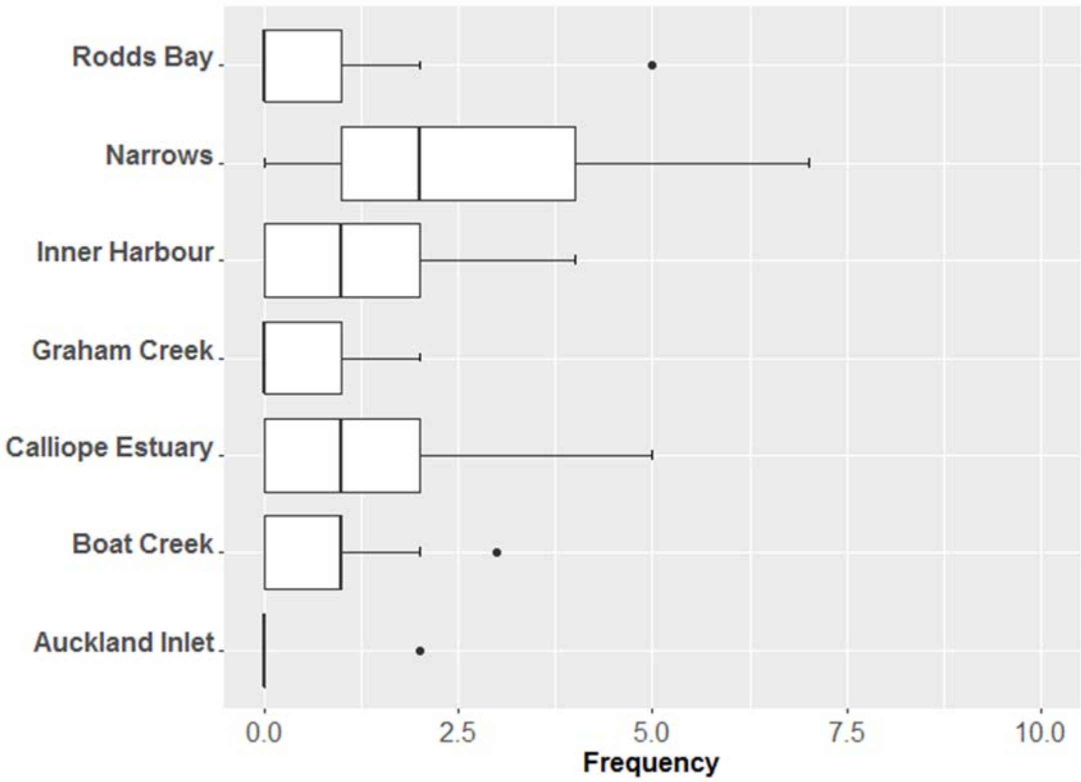


Figure 11: Number of mud crabs in each pot set in June 2018, by zone.

Sex ratio

In February and June 2018, many more oversize female crabs were caught than oversize male crabs. Sex ratios were low across the harbour in both February and June (Table 7). A total of 201 mud crabs over the legal size limit of 150mm carapace width were caught in 2018, and only 38 were male. The highest sex ratio by zone was from Rodds Bay in February (a ratio of 1), but from only four mud crabs. More oversized mud crabs were caught at Rodds Bay in June (17) and the sex ratio from this larger sample was 0.25.

Table 7: Sex ratios of mud crabs in February and June 2018, by zone. NC = not calculable.

Zone	Zone name	February 2018 data			June 2018 data		
		Males > 143mm notch width	Females > 143mm notch width	Sex ratio	Males > 143mm notch width	Females > 143mm notch width	Sex ratio
1	Narrows	3	44	0.07	3	39	0.08
2	Grahams Creek	4	10	0.40	1	7	0.14
4	Boat Creek	3	4	0.75	3	4	0.75
5	Inner Harbour	4	11	0.36	3	13	0.23
6	Calliope Estuary	2	4	0.50	5	12	0.42
7	Auckland Inlet	2	0	NC	0	1	0
13	Rodds Bay	2	2	1	3	12	0.25
	Harbour wide average			**0.51**			**0.27**

Rust lesions

Of the 173 mud crabs measured, weighed and checked for lesions in February 2018, only 4 had rust lesions and 169 did not. During February 2018, the percentage of crabs with lesions was highest at the Narrows (4.29%; from three mud crabs) and Graham Creek (3.7%; from one mud crab) and was zero at all other zones (Table 7). Prevalence of rust lesions was similarly low in June 2018, with only four of 162 mud crabs displaying lesions. During June 2018, the percentage of crabs with lesions was highest at Rodds Bay (11.76%; from two mud crabs), Boat Creek (7.69%; from one mud crab) and the Narrows (1.67%, from one mud crab) and was zero at all other zones (Table 8).

Table 8: Percentage of mud crabs caught in February and June 2018 with and without rust spot lesions, by zone.

Zone	Zone name	February 2018 data				June 2018 data			
		Mud crabs with lesions	Mud crabs without lesions	% without lesions	% With lesions	Mud crabs with lesions	Mud crabs without lesions	% without lesions	% With lesions
1	Narrows	3	67	95.71	4.29	1	59	98.33	1.67
2	Grahams Creek	1	26	96.30	3.70	0	10	100	0
4	Boat Creek	0	13	100	0	1	13	92.31	7.69
5	Inner Harbour	0	29	100	0	0	28	100	0
6	Calliope Estuary	0	23	100	0	0	29	100	0
7	Auckland Inlet	0	2	100	0	0	2	100	0
13	Rodds Bay	0	9	100	0	2	17	88.24	11.76
	Harbour wide average			**99%**	**1%**			**97%**	**3%**

Scores by zone

The mud crab data set used to score each selected zone for the 2018 Gladstone Harbour Report Card included combined data from both February and June 2018. Results for each Zone are provided in Table 9.

Table 9: Calculated index values for 2018, for each of the four recommended measures in each of the seven recommended long term monitoring sites. NC = not calculable (<5 crabs caught at zone in 2018).

Zone	Zone name	Abundance (CPUE)	Prevalence of rust lesions	Sex ratio	Biomass	
					Male	Female
1	Narrows	3.25	0.031	0.072	0.214	0.207
2	Graham Creek	0.975	0.027	0.294	0.345	0.221
4	Boat Creek	0.848	0.037	0.750	0.219	0.197
5	Inner Harbour	1.43	0	0.292	0.219	0.209
6	Calliope Estuary	1.30	0	0.438	0.310	0.231
7	Auckland Inlet	0.100	0	NC	0.140	0.155
13	Rodds Bay	0.725	0.071	0.357	0.194	0.217
	Harbour wide average	**1.23**	**0.02**	**0.37**	**0.23**	**0.21**

Indicator grades

Scores and grades for the three calculable mud crab measures for the 2018 Report Card are provided in Table 10. Scores > 1 and < 0 have been bounded by [1, 0] in line with GHHP standard methods (GHHP, 2014). A combined score for the Mud Crab Indicator has been calculated as the average of the three measure scores, and an overall grade is provided for each zone and for the Harbour. Only four mud crabs were caught in Zone 7 – Auckland Creek, two in February 2018 and two in June 2018. Given the very small sample size (< 5 mud crabs from 40 pots) it is not appropriate to calculate scores and grades for the prevalence of rust lesions or sex ratio measures in this zone.

Table 10: Scores and grades for mud crab measures and the mud crab indicator by Zone. NC = Not calculable.

Zone	Abundance (CPUE)	Prevalence of rust lesions	Sex ratio*	Biomass	Zone score 2018
1. The Narrows	1	1	0	NC	0.67
2. Graham Creek	0.3	1	0.03	NC	0.44
4. Boat Creek	0.25	1	0.29	NC	0.51
5. Inner Harbour	0.52	1	0.02	NC	0.52
6. Calliope Estuary	0.47	1	0.11	NC	0.52
7. Auckland Inlet	0	NC	NC	NC	NC
13. Rodds Bay	0.2	0.90	0.06	NC	0.39
Harbour Average					0.51

* Sex ratio based on legal size limits. NC = not calculable in 2018.

Eurimbula Creek

A total of 56 mud crabs were caught during a single day of sampling at Eurimbula Creek on 26 June 2018, using 20 crab pots deployed for at least five hours. There were 35 male mud crabs, 20 female mud crabs and one additional male mud crab which escaped through a hole in the pot and was omitted from further analysis.

Of the 55 mud crabs analysed, average size (notch width) was 145.16 mm for the full sample, 143.54 mm for males (which is above the legal size limit of 143 mm notch width / 150 mm spine width), and 148.00 mm for females. Results of a Welch Two sample t-test found that there was no significant difference between the size of females and males caught at Eurimbula Creek (t = -1.0389, df = 32.888, p = 0.306). Average weight of the full sample of mud crabs caught at Eurimbula was 773.64 g. The average male mud crab weight was 838.14 g while the average female weight was 660.75 g. A Welch Two sample t-test found that male mud crabs caught at Eurimbula were heavier than female mud crabs (t = 2.9166, df = 52.572, p = 0.005).

Catch per unit effort was 2.75 crabs per pot, and every pot contained at least one mud crab. In June 2018 the sex ratio of male:female crabs over the legal size limit of 150 mm carapace width (equivalent to 143 mm notch width) was 1.43. That is, there were 1.4 oversize male mud crabs caught for every one oversize female mud crab. This was higher than recorded in the Gladstone Harbour zones, where less oversize males than females were caught in June 2018 at all seven zones. Of the 55 crabs analysed from Eurimbula Creek, only one male mud crab had a small rust lesion, a prevalence of 1.82%, similar to the Gladstone Harbour average of 3% in June 2018.

Discussion

The overall grades for the Mud Crab Indicator for Gladstone Harbour in 2018 are as follows:

A: No zones.

B: Zone 1 – the Narrows.

C: Zone 4 – Boat Creek, Zone 5 – Inner Harbour, Zone 6 – Calliope Estuary,

D: Zone 2 – Graham Creek, Zone 13 – Rodds Bay.

E: No zones.

Not calculable (< 5 mud crabs caught): Zone 7 – Auckland Creek.

In comparison, the 2017 grades were:

A: Zone 5 – Inner Harbour;

B: Zone 1 – Narrows and Zone 4 – Boat Creek;

C: Zone 2 – Graham Creek;

D: Zone 6 – Calliope Estuary, Zone 7 – Auckland Inlet and Zone 13 – Rodds Bay; and

E: No zones.

A key difference between sampling in 2017 and 2018 was the inclusion of a 2018 summer sample conducted in February. The inclusion of both summer and winter sampling allows for the detection of seasonal changes in the local mud crab population. The change in sampling regime may have been a factor in the decline in grades experienced across the harbour, however natural variations in mud crab population dynamics and movements can also potentially influence results.

Zone 1 – the Narrows, scored a B in both 2017 and 2018. Grades for Zone 4 – Boat Creek and Zone 5 – Inner Harbour have both dropped in 2018, falling from a B and an A grade respectively to C grades. Much lower mud crab catches were achieved in both zones in 2018, and the grade for the Abundance measure has declined from an A in both zones in 2017 to a D and a C, respectively, in 2018. There are many factors that can influence the catchability of mud crabs, so the lower grade is not necessarily a cause for concern on the basis of a single year. Some of the possible (non-anthropogenic) factors affecting catchability of mud crabs include moult state of crabs, reproductive cycles, lunar and diel cycles, temperature and water motion (Knuckey, 1999). A more worrying trend would be a long term decline in numbers over several years, which may indicate impacts such as a reduction in suitable habitat or overfishing. It is currently too early in the development and grading of the Gladstone Harbour mud crab indicator to establish trends, but the incorporation of a 10-year moving average methodology in setting the Abundance benchmark from 2018 onwards will help to allow for these natural variations.

The grades reflect the wide variety of pressures on mud crabs in Gladstone Harbour, including commercial fishing, recreational fishing and environmental/habitat condition. In Zone 7 – Auckland Creek, only four mud crabs were caught across both sampling periods (February and June) in 2018. Given the very small sample size available for analysis (< 5 mud crabs from 40 pots), it is not appropriate to calculate scores and grades for the Prevalence of rust lesions or Sex ratio measures in this zone in 2018.

The Prevalence of rust lesions measure scored an A grade in all six zones for which it was calculated. This measure is based on a moderately-high confidence benchmark and WCS developed using research data published by Andersen and Norton (2001) and Dennis et al. (2016), and data collected in June 2017. Rust lesions have previously been recorded at a much higher prevalence in Gladstone Harbour than reported here, with up to 37% of mud crabs afflicted by rust spot lesions in a 2012 study (Dennis et al. 2016).

The Abundance measure was scored in 2018 with the benchmark of a moving average of the 75th percentile of abundance scores across the harbour in 2017 and 2018. A 10-year moving average benchmark will continue to be calculated for Abundance, to allow for natural variations in catchability and abundance while still providing for the analysis of trends through time. In the 2017 pilot year, the 75th percentile of the 2017 abundance scores was used as the benchmark. Historical data provided the Fisheries Queensland LTMP for the Mud Crab Fishery was not used in the development of the Abundance benchmark as it was collected in a slightly different way: sampling was conducted only in summer, sites included areas to the north of the Gladstone Harbour zones, different soak times were used and the crab pots were smaller. It is possible that these factors may have affected CPUE more than they affected the other measures. In 2017 and 2018, a catch rate of less than one crab in four pots has been used as a WCS. The WCS of 0.25 crabs/pot is based on assumed recreational fisher preferences of catching a crab (Flint et al., 2017). An alternative approach would be to apply commercial fisher preferences. Knuckey (1999) suggested crabbers in the Northern Territory cease crabbing when the saleable catch is <0.1 kg/pot.day, which in Queensland equates to about one oversized male crab in every 8-9 pots.

The 2017 grades were highest (A) at Zone 1 – Narrows, Zone 4 – Boat Creek and Zone 5 – Inner Harbour. The lowest grade (E) was recorded at Zone 6 – Calliope Estuary, Zone 7 – Auckland Inlet and Zone 13 – Rodds Bay. In 2018, using the moving average benchmark and the same WCS, Zone 1 – Narrows again scored an A, Zone 5 – Inner Harbour scored a C, Zone 2 – Graham Creek, Zone 4 – Boat Creek and Zone 6 – Calliope Estuary scored a D, and Zone 7 – Auckland Inlet and Zone 13 – Rodds Bay scored an E. Discrepancies in abundance can be caused by capture technique, sampling areas and sampling times, or by differences in crab distribution, growth or survival induced by habitat and environmental conditions (Alberts-Hubatsch *et al.*, 2016). During 2017 and 2018 sampling, possible variations due to catch and sampling techniques were controlled as much as possible. When these factors are controlled, abundance is a simple indicator of pressures such as habitat availability, extraction (fishing) and recruitment limitation, although natural biological variation can also be a factor. Using a moving average benchmark helps to account for natural variations in catchability and abundance in the mud crab population in Gladstone Harbour and allows for the long term assessment of trends in abundance over time.

The Sex ratio measure compares the number of male crabs over the legal size limit (150 mm carapace width, equivalent to 143 mm notch width) to female crabs over the (male) legal size limit. A benchmark for the sex ratio measure was established using international scientific literature from unfished mud crab populations in the 2017 pilot year. This benchmark was considered to be of low reliability and Flint et al. (2017) recommended that an unfished population from Central Queensland should sampled in 2018 to provide a reference guideline. GHHP has supported this recommendation by commissioning CQUniversity to sample mud crabs at Eurimbula Creek during June 2018 and February 2019. The results will inform a review of the Sex ratio benchmarks in 2019. In 2018, the Sex ratio benchmark was adjusted to account for a previously unavailable unpublished thesis from uncrabbed estuaries in northern New South Wales (Butcher, 2004). The results of the thesis aligned with results from a study in a small uncrabbed region in Moreton Bay, southern Queensland (Pillans et al., 2005). Both studies found a ratio of two male mud crabs to one female mud crab. As a result of this additional information, the Sex ratio benchmark has been adjusted down from 3 to 2 to calculate 2018 scores. The benchmark will be reviewed again in 2019 using the results from Eurimbula Creek. The WCS for the sex ratio measure remained the same in 2018, and is the 25th percentile of LTMP data. In 2017, all zones except Zone 5 – Inner Harbour (B) scored poorly on the Sex ratio measure. In 2018, all zones except Zone 4 (D) were graded poor (E).

In areas such as Queensland, where a sex-based fishery is enforced, changes in the ratio of males to females that can't be explained by biological factors such as spawning migrations, are likely to be indicative of a change in fishing pressure. It is also worth noting, as mentioned in Flint et al. (2017) that the pattern observed suggests that fishers are observing regulations regarding the release of females. Shifts in sex ratio caused by overfishing have implications for population dynamics and may also influence ecosystem processes through gender-biased behaviours such as burrow digging. There is a remaining knowledge gap as to the timing and population effect of the female spawning migration in Gladstone Harbour which would benefit from future research. If females were found to be absent from Gladstone Harbour during summer, as is the case elsewhere (e.g. Dec-Feb in the Northern Territory; Knuckey (1999)) then sampling outside of this period would provide a more stable population for long term monitoring. An early Queensland Government study of the Queensland mud crab fishery identified the period from March to May as the time with the highest catch of legal males per pot at Princess Charlotte Bay (northern Queensland) (Hill, 1984). In the present study, sex ratios were only slightly higher (i.e. more males) in February than in June.

The Queensland Government's QFish database provides data on commercial mud crab licenses, catch and effort. In 2014 there were 38 commercial mud crab licenses operating in Grid S30 (includes Gladstone Harbour), seven in Grid S31 (includes Colosseum Inlet) and 16 in Grid T31 (includes Rodds Bay). In 2014 these 61 licenses accounted for more than 16% of the total number of mud crab licenses in Queensland (total of 370) and more than 21% of the total mud crab catch (287 tonnes of a total 1,329 tonnes). Grid S30 has the largest number of licenses in a single Grid area after W37 in Moreton Bay (49 licenses) and equivalent to Grid R30 in the Fitzroy River delta (38 licenses), emphasising the importance of Gladstone Harbour to the Queensland fishery. Recreational and Indigenous catches are not as well understood as commercial catches. Recent estimates for Queensland suggest a combined take of mud crabs by recreational and Indigenous fishers of about 25% of the total harvest (Grubert *et al.*, 2015) but these estimates are not considered highly reliable.

As the 2017 and 2018 datasets contain the only available mud crab weight data from Gladstone Harbour, scores have not been calculated for Biomass (CW:BW) this year. Once three years of data are available, a scoring system can be developed in 2019. The relationship between carapace width and body weight can be used as a general indicator of condition, providing a measure of how well-fed an individual is, with high ratios typically indicating that an ecosystem has high productivity and food density. Other information collected during 2018 includes the seafood 'grade' of mud crabs, measured using the Australian Industry Live Mud Crab Grading Scheme (available at: https://www.c-aid.com.au/wp-content/uploads/Live-Mud-Crab_Grading-Scheme_WEB.pdf). It is recommended that this grading method is considered as a possible condition measure and/or combined condition measure for use with the biomass measure, in 2019.

The highest overall mud crab grade was recorded for Zone 1 – Narrows (B), while the lowest overall grades (D) were recorded for all other zones, except Zone 7 – Auckland Inlet which has not been scored due to a very small sample size (<5 mud crabs). The Harbour Average for 2018 is a D. As 2018 is only the second year this study has been conducted, the accuracy and reliability of the mud crab grades may improve as more data are collected and all measures, including biomass, are able to be calculated. It is recommended that a review of the benchmarks and WCSs used for each mud crab measure, as well as the weighting applied to measures, is conducted once three years of monitoring data are available, in 2019.

In addition to the four recommended measures, two potential indicators were identified by Flint et al. (2017) as potentially useful. Both would require additional research and/or monitoring costs. These two indicators are:

- Bioaccumulation of metals; and
- Recruitment to nursery grounds.

Both of these potential indicators would require research to develop confidence in baselines. Measuring recruitment to nursery grounds would be non-lethal but would involve extra costs because of the requirement for alternative monitoring techniques and sites, and the sampling effort required can be disproportionately high in comparison to catch using standard trapping techniques for adult crabs (Knuckey, 1999). Emerging technologies such as video sampling may be more cost-effective, but have not been previously trialled for image recognition of juvenile mud crabs so would require further research and development before implementation. High turbidity in some areas of Gladstone Harbour would also be an issue for video sampling.

Bioaccumulation requires lethal sampling of mud crabs to measure toxicant (e.g. metal) concentrations in the hepatopancreas, muscle tissue or gills, with additional costs for dissection and analysis. Bioaccumulation of toxicants can be a particularly relevant indicator for urban and industrialised areas such as ports, and is measured using established methods. Several studies have been conducted using crabs as bioindicators of heavy metal and toxicant bioaccumulation. The effect of heavy metals on natural populations of organisms needs to take into account tolerance in communities that have been exposed to particular toxicants over several generations (Chiarelli and Roccheri, 2014). The usefulness of an organism as a bioindicator of toxicant also needs to consider if the organism bioaccumulates the toxicant over a measurable time, or if the toxicant in question is regulated by the organism, with no significant bioaccumulation measurable (McPherson and Brown, 2001).

Scylla serrata has previously been shown to accumulate a range of contaminants including persistent organic pollutants (POPs) and metals (Mortimer, 2000). *S. serrata* were sampled in six estuaries between Brisbane and Cairns, Qld, for analysis of metals, metalloids and pesticides in muscle and hepatopancreas tissue (Mortimer, 2000). Most metals and metalloids were found to be preferentially sequestered to the hepatopancreas. *S. serrata* from industrialised areas such as the Brisbane River and Gladstone showed higher concentrations of Pb, Se and Sn than less impacted locations (Mortimer, 2000). More recently, *Scylla serrata* hepatopancreas samples from 11 Queensland rivers were analysed for pesticides and POPs with Dichlorodiphenyltrichloroethane (DDT) and dieldrin detected along with other banned organochlorines (Negri *et al.*, 2009). As the rivers sampled were similar to the Mortimer (2000) study, the authors compared results directly. Concentrations of DDT and dieldrin measured in the earlier Mortimer (2000) study were found to be around one order of magnitude greater than those measured in the Negri *et al.* (2009) study. Concentrations of DDT and dieldrin measured in the Negri *et al.* (2009) study were found to be too low to pose a threat to human health. Contemporary pesticides were also analysed in *S. serrata* hepatopancreas by Negri *et al.* (2009), however, only endosulfan and chlorpyrifos were detected. The authors noted that while other contemporary pesticides (e.g. diuron, atrazine, simazine etc.) do not appear to be bioaccumulated by *S. serrata*, this does not mean that there are no effects from them (Negri *et al.*, 2009).

Structural deformities of the gills, muscles and hepatopancreas have been observed in *S. serrata* due to metal toxicity, with the degree of damage correlated with elevated bioaccumulated metal concentration (Arockia Vasanthi *et al.*, 2014). Significant metal accumulation and histocytological lesions were evident in the mud crabs (Arockia Vasanthi *et al.*, 2014). Tissue and metal specific bioaccumulation was observed, with Cu, Pb, Cd and Mn highest in the hepatopancreas, lower in the gills and significantly lower in the muscle samples analysed, whereas Zn and Fe were highest in the gills, lower in the hepatopancreas and significantly lower in the muscles (Arockia Vasanthi *et al.*, 2014).

In Gladstone, elevated metal (As, Cr, Cu, Fe, Hg, Mn, Ni, U, Zn) concentrations have been reported from the hepatopancreas of *S. serrata* collected at two sites at Spillway Creek, in comparison to two additional sites in the same creek, and to sites in Wild Cattle Creek (Gladstone Harbour), Baffle Creek (south of Rodds Bay) and Ayr (North Queensland) (Andersen *et al.*, 2001). A subsequent study on *S. serrata* identified only Fe and Se of ten metals tested in hepatopancreas tissues (Andersen *et al.*, 2003), and both were recorded at concentrations below food safety guidelines.

Recommendations

The following additional recommendations are provided for the application of the mud crab indicator in future years. A review of the measures, and the benchmarks and WCSs used for each measure, should be conducted in 2019, when three years' worth of monitoring data are available.

1. **BIOMASS MEASURE:** As the 2017 and 2018 data represent the only available mud crab weight data from Gladstone Harbour, it is not yet possible to determine a benchmark and WCS. We recommend the benchmark and WCS for this measure are calculated once three years of mud crab monitoring data are available (in 2019). At this stage, it may also be useful to analyse mud crab seafood grade data collected during 2018 and 2019 using the Australian Industry Live Mud Crab Grading Scheme.
2. **SAMPLING UNFISHED POPULATIONS:** Sex ratios were scored in the 2017 pilot year against a benchmark of a 3:1 male to female ratio, based on unfished mud crab populations internationally (Alberts-Hubatsch et al., 2016) and in 2018 against a benchmark of 2:1 male to female ratio, based on previously unavailable

information from Australian estuaries in southern Queensland and northern New South Wales (Butcher, 2004; Pillans et al., 2005). This value may again be updated for local mud crab populations using data collected in a separate GHHP project sampling a nearby unfished population at Eurimbula Creek near Round Hill. Data from the unfished population collected during June 2018 and February 2019 will be used to review the Sex ratio measure in 2019.

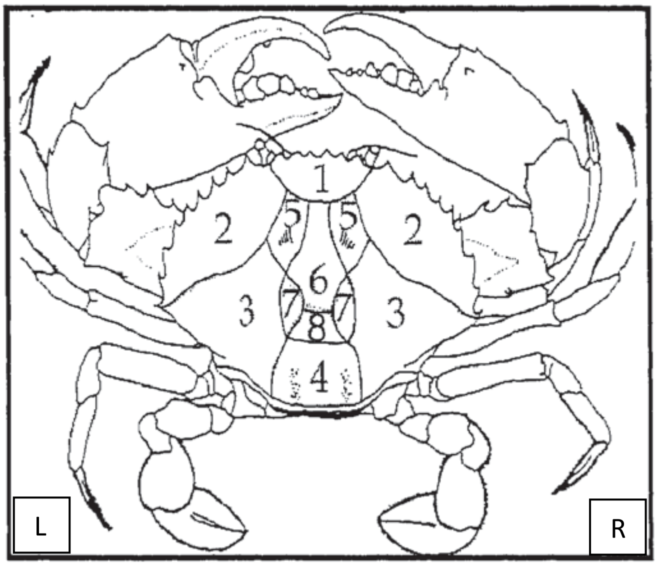
3. **REVIEW:** It is recommended that a review of the benchmarks and WCSs used for each mud crab measure, and weighting of the measures, is conducted once three years of monitoring data from Gladstone Harbour, and two samples from Eurimbula Creek, are available in 2019.
4. **OTHER MEASURES:** Bioaccumulation and nursery value both scored lower than the four selected measures based on higher costs associated with field sampling (Flint et al., 2017), but would potentially be useful to include if future budgets allowed. The bioaccumulation results would be of particular interest as an additional indication of water and sediment quality, and uses established methods.

Appendix 1: Mud crab rust spot lesion grading system (Source: Andersen, 2003)

Gross grading system for rust spot lesions in mud crabs.

Grade	Description
Grade 1	Non Perforated; < 5mm diameter
Grade 2	Non Perforated; ≥ 5mm diameter
Grade 3	Perforated cuticle (either partially or fully); < 5mm diameter
Grade 4	Perforated cuticle (either partially or fully); ≥ 5 mm diameter & < 20mm diameter
Grade 5	Perforated cuticle (either partially or fully); ≥ 20mm diameter

Numbered areas (locations) of the mud crab carapace to which lesions are allocated.



Legend

1. FrontaVorbital
2. Hepatic/epibranchial
3. Mesobranchial/metabranchial
4. Intestinal
5. Progastric
6. MesogastricImetagastric/ uro- gastric
7. Internal cuticular partition
8. Cardiac

(Adapted from McLaughlin, 1980).

Also note Left (L) or Right (R) side – when looking down on carapace.

References

Alberts-Hubatsch, H., Lee, S.Y., Meynecke, J.-O., Diele, K., Nordhaus, I., Wolff, M., 2016. Life-history, movement, and habitat use of Scylla serrata (Decapoda, Portunidae): current knowledge and future challenges. Hydrobiologia 763, 5-21.

Andersen, L., Lewis, S., Melzer, A., 2001. Fluoride and metals in Spillway Creek crustacea. Gladstone, Australia, p. 53.

Andersen, L., Norton, J., 2001. Port Curtis mud crab shell disease: nature, distribution and management. FRDC Project No. 98/210. Central Queensland University, Gladstone.

Andersen, L., Storey, A.W., Sinkinson, A., Dytlewski, N., 2003. Transplanted oysters and resident mud crabs as biomonitors in Spillway Creek. Gladstone, Australia, p. 30.

Andersen, L.E., Norton, J.H., Levy, N.H., 2000. A new shell disease in the mud crab *Scylla serrata* from Port Curtis, Queensland (Australia). Diseases of aquatic organisms 43, 233-239

Arockia Vasanthi, L., Muruganandam, A., Revathi, P., Baskar, B., Jayapriyan, K., Baburajendran, R., Munuswamy, N., 2014. The application of histo-cytopathological biomarkers in the mud crab Scylla serrata (Forskal) to assess heavy metal toxicity in Pulicat Lake, Chennai. Marine Pollution Bulletin 81, 85-93.

Bolger, T., Connolly, P.L., 1989. The selection of suitable indices for the measurement and analysis of fish condition. Journal of Fish Biology 34, 171-182.

Bonine, K.M., Bjorkstedt, E.P., Ewel, K.C., Palik, M., 2008. Population characteristics of the mangrove crab Scylla serrata (Decapoda: Portunidae) in Kosrae, Federated States of Micronesia: effects of harvest and implications for management. Pacific Science 62, 1-19.

Brewster, A., 2015. Giving this Country a Memory: Contemporary Aboriginal Voices of Australia. Cambria Press, New York.

Butcher, P.A., 2004. Mud crab (Scylla serrata) and marine park management in estuaries of the Solitary Islands Marine Park, New South Wales. University of New England, Armidale, Australia.

Cai, W., Wang, G., Santoso, A., McPhaden, M.J., Wu, L., Jin, F.-F., Timmermann, A., Collins, M., Vecchi, G., Lengaigne, M., England, M.H., Dommenget, D., Takahashi, K., Guilyardi, E., 2015. Increased frequency of extreme La Niña events under greenhouse warming. Nature Climate Change 5, 132-137.

Chandrasekaran, V.S., Natarajan, R., 1994. Seasonal abundance and distribution of seeds of mud crab *Scylla serrata* in Pichavaram Mangrove. Journal of Aquaculture in the Tropics 9, 343-350.

Chiarelli, R., Roccheri, M.C., 2014. Marine Invertebrates as Bioindicators of Heavy Metal Pollution. Open Journal of Metal 4, 93-106.

DAF, 2017. Mud Crab Fisheries. Queensland Department of Agriculture and Fisheries.

Dennis, M.M., Diggles, B.K., Faulder, R., Olyott, L., Pyecroft, S.B., Gilbert, G.E., Landos, M., 2016. Pathology of finfish and mud crabs Scylla serrata during a mortality event associated with a harbour development project in Port Curtis, Australia. Diseases of aquatic organisms 121, 173-188.

Dumas, P., Léopold, M., Frotté, L., Peignon, C., 2012. Mud crab ecology encourages site-specific approaches to fishery management. Journal of Sea Research 67, 1-9.

FAO, 2017. Scylla serrata. Rome.

Flint, N., Anastasi, A., De Valck, J., Chua, E., Rose, A., Jackson, E.L., 2017. Developing mud crab indicators for the Gladstone Harbour Report Card: Project ISP015-2017. CQUniversity Australia, Rockhampton, Australia.

GHHP, 2015. Technical Report, Gladstone Harbour Report Card 2015, GHHP Technical Report No.2. Gladstone, Australia.

Grubert, M., Johnson, D., Johnston, D., Leslie, M., 2015. Status of Australian Fish Stocks Report. Mud Crabs, Queensland East Coast.

Grubert, M.A., Phelan, M.J., Bird, M.H., 2012. Use of a Durometer to Differentiate Between Soft- and Hard-Shelled Mud Crabs (Scylla serrata). Journal of Aquatic Food Product Technology 21, 3-13.

Harding, J.M., Mann, R., 2010. Observations of distribution, size, and sex ratio of mature blue crabs, Callinectes sapidus, from a Chesapeake Bay tributary in relation to oyster habitat and environmental factors. Bulletin of Marine Science 86, 75-91.

Heasman, M.P., 1980. Aspects of the general biology and fishery of the mud crab Scylla serrata (Forskal) in Moreton Bay, Queensland.

Hill, B.J., 1984. The Queensland Mud Crab Fishery. In: Industries, Q.D.o.P. (Ed.). Queensland Government, Brisbane, Australia.

Hyland, S.J., Hill, B.J., Lee, C.P., 1984. Movement within and between different habitats by the portunid crab Scylla serrata. Marine Biology 80, 57-61.
Ikhwanuddin, M., Azmie, G., Juariah, H.M., Zakaria, M.Z., Ambak, M.A., 2011. Biological information and population features of mud crab, genus Scylla from mangrove areas of Sarawak, Malaysia. Fisheries Research 108, 299-306.
Keenan, C., Davie, P.J., Mann, D., 1998. A revision of the genus Scylla de Haan, 1833 (Crustacea: Decapoda: Brachyura: Portunidae). The Raffles Bulletin of Zool-ogy 46, 217-245.
Knuckey, I.A., 1999. Mud crab (Scylla serrata) population dynamics in the Northern Territory, Australia and their relationship to the commercial fishery. Northern Territory University, Darwin, Australia.
McIntosh, E.J., Poiner, I.R., Panel, I.S., 2014. Gladstone Harbour Report Card Framework recommendation. Gladstone Healthy Harbour Partnership, Gladstone, Queensland, p. 82.
McPherson, R., Brown, K., 2001. The bioaccumulation of cadmium by the Blue Swimmer Crab Portunus pelagicus L. Science of The Total Environment 279, 223-230.
Meynecke, J.-O., Grubert, M., Arthur, J.M., Boston, R., Lee, S.Y., 2012. The influence of the La Niña-El Niño cycle on giant mud crab (Scylla serrata) catches in Northern Australia. Estuarine, Coastal and Shelf Science 100, 93-101.
Mortimer, M.R., 2000. Pesticide and Trace Metal Concentrations in Queensland Estuarine Crabs. Marine Pollution Bulletin 41, 359-366.
Negri, A.P., Mortimer, M., Carter, S., Müller, J.F., 2009. Persistent organochlorines and metals in estuarine mud crabs of the Great Barrier Reef. Marine Pollution Bulletin 58, 769-773.
Pillans, S., Pillans, R.D., Johnstone, R.W., Kraft, P.G., Haywood, M.D.E., Possingham, H.P., 2005. Effects of marine reserve protection on the mud crab *Scylla serrata* in a sex-biased fishery in subtropical Australia. Marine Ecology Progress Series 295, 201-213.
Rainbow, P.S., 1995. Biomonitoring of heavy metal availability in the marine environment. Marine Pollution Bulletin 31, 183-192.
Ruscoe, I.M., Shelley, C.C., Williams, G.R., 2004. The combined effects of temperature and salinity on growth and survival of juvenile mud crabs (*Scylla serrata* Forskål). Aquaculture 238, 239-247.
van Oosterom, J., Codi King, S., Negri, A., Humphrey, C., Mondon, J., 2010. Investigation of the mud crab (Scylla serrata) as a potential bio-monitoring species for tropical coastal marine environments of Australia. Marine Pollution Bulletin 60, 283-290.
Walton, M.E., Le Vay, L., Lebata, J.H., Binas, J., Primavera, J.H., 2006. Seasonal abundance, distribution and recruitment of mud crabs (Scylla spp.) in replanted mangroves. Estuarine, Coastal and Shelf Science 66, 493-500.
Walton, M.E., Le Vay, L., Lebata, J.H., Binas, J., Primavera, J.H., 2007. Assessment of the effectiveness of mangrove rehabilitation using exploited and non-exploited indicator species. Biological Conservation 138, 180-188.
Williams, M.J., Hill, B.J., 1982. Factors influencing pot catches and population estimates of the portunid crab Scylla serrata. Marine Biology 71, 187-192.