

Project Plan SP 2019-031

Do marine reserves adequately represent high diversity cryptobenthic fish assemblages in a changing climate?

Marine Science

Project Core Team

Supervising Scientist	Shaun Wilson
Data Custodian	Shaun Wilson
Site Custodian	

Project status as of March 18, 2020, 4:07 p.m.

Pending project plan approval

Document endorsements and approvals as of March 18, 2020, 4:07 p.m.

Project Team	required
Program Leader	required
Directorate	required
Biometrician	required
Herbarium Curator	not required
Animal Ethics Committee	required

Do marine reserves adequately represent high diversity cryptobenthic fish assemblages in a changing climate?

Biodiversity and Conservation Science Program

Marine Science

Departmental Service

Service 6: Conserving Habitats, Species and Communities

Project Staff

Role	Person	Time allocation (FTE)
Supervising Scientist	Shaun Wilson	0.15
Research Scientist	Jordan Goetze	0.3
Research Scientist	Alan Kendrick	0.05

Related Science Projects

SP 2012-008 Marine Monitoring Program

Proposed period of the project

July 1, 2019 – June 30, 2022

Relevance and Outcomes

Background

Assemblages of marine fish provide key ecological functions and ecosystem services. Therefore, much effort is spent managing and monitoring fish in Western Australia's network of marine reserves. Monitoring currently focuses on large conspicuous fish, however small bodied species (<5cm) typically represent ~40% of all described species (Brandl et al. 2018). Moreover, many of these cryptobenthic reef fish (CRF) have not been described by science (Allen 2015) and their substantial contribution to current measures of fish diversity (a condition indicator for marine parks) and ecological significance are underestimated (Bellwood et al. 2019).

Short life spans of CRF suggest they are important conduits of energy transfer in marine food webs (Wilson 2004; Depczynski et al. 2007; Goatley and Bellwood 2016) and will respond more rapidly to environmental change and stressors than larger bodied counterparts (Bellwood et al. 2006). This is especially pertinent along the WA coastline, where marine heat waves have had a dramatic impact on coral (Moore et al. 2012), macroalgae (Wernberg et al. 2016) and seagrass (Arias-Ortiz et al. 2018) habitats, yet the implications for the high diversity CRF assemblages is equivocal. Furthermore, the frequency of marine heat waves is increasing (Hughes et al. 2018) and characterising the response of CRF to disturbance related changes in habitat may provide an indication of how larger bodied taxa and related ecological processes will react to climate related stressors over longer time frames (Wilson et al. 2019). Similarly, as CRF are prey of many longer-lived species targeted by WA fishers (Farmer and Wilson 2011), shifts in CRF assemblages may be indicative of trophic cascades due to changes in fishing pressure. Indeed, composition of CRF assemblages may be a valuable tool for monitoring predation and how it varies with respect to spatial management of fishing pressure in marine parks (Willis and Anderson 2003).

Standard visual methods of surveying fish are not suitable for monitoring CRF (Ackerman and Bellwood 2000), and whilst sampling with poisons is the preferred technique, this is not always appropriate, requires extensive time under water and is difficult to standardise among samples. Accordingly, this project will investigate an alternate method for assessing CRF assemblages which will involve the collection and analysis of water samples for fish DNA (eDNA), a non-invasive technique with the potential to detect cryptic and rare species with little effort in the field (Pikitch 2018). Standard poison and visual approaches to assessing fish assemblages will also

be undertaken to compare how measures of CRF assemblage vary among techniques. By collecting data from different habitats and management zones within marine parks it is possible to assess how effectively different techniques detect change in CRF due to spatial variation in environment or management. Results from these surveys will also help identify appropriate indicator species for future monitoring.

The study will initially determine which technique(s) best assess diversity and abundance of CRF in Ningaloo Marine Park. Appropriate techniques will then be used throughout the states network of marine reserves to assess how CRF assemblages vary among habitats and management zones.

Aims

The overarching aim of this project is to quantify the diversity and composition of CRF assemblages within Ningaloo Marine Park and gain an understanding of how CRF change due to shifts in fishing pressure and habitat composition caused by climate change. Thus the project will:

1. Develop appropriate methods for measuring and monitoring CRF assemblages
2. Compare CRF assemblages collected inside and outside of no-take sanctuary zones and
3. Compare CRF assemblages across a gradient of reef (habitat) types from high coral cover and complexity to reefs dominated by macroalgae with low complexity.

The project will initially take place in Ningaloo Marine Park, but may be extended to other parks after the first three stages are completed. This would provide a broader scale assessment of CRF assemblages from tropical to temperate biomes.

Expected outcome

The project will determine appropriate techniques for monitoring CRF, comparing assemblages in fished and unfished areas to assess how spatial variation in fishing pressure influences diversity of a KPI ecological value. Thus, the study will provide an indication of marine reserve effectiveness for conserving diversity and a key ecological process. Comparing CRF assemblages in different habitats and levels of habitat quality (e.g. % coral cover) will explore how changes in habitat due to climate related disturbances and pressures may affect fish assemblages. Findings will therefore determine the utility of CRF as indicators of environmental change and adequacy of management zones in marine reserves for capturing fish diversity and important ecological processes. This quantitative approach will provide a basis for assessing the diversity and ecological value of CRF relative to larger bodied fishes throughout the states network of marine reserves.

Knowledge transfer

The project will develop indicators and methodologies that can improve the monitoring of fish assemblages in the states network of marine reserves by the marine science program. DBCA researchers will learn methods for eDNA sampling, which has shown great potential as a rapid and non-destructive monitoring tool for a broad range of marine life. Improved understanding of the diversity and habitat associations of fishes will help with conservation planning and projected changes to biodiversity due to climate induced shifts in habitat. Findings from the project will be disseminated within DBCA via information sheets, meetings and presentations. As results will also be of interest to international researchers and conservationists, they will be published in peer reviewed journals, presented at conferences and circulated through social media.

Tasks and Milestones

Milestone/task	When
Collect samples with ichthyocide, UVC, eDNA	March 2020
ID and analyse samples	March-August 2020
Analyse of data, write up results of method comparison	August 2020- 2021
Analyse data, write up results on effects of no-take areas	Feb 2021-July 2021
Use appropriate method to sample across different habitats	Feb 2022-Feb 2023

Analyse samples	Feb 2022-July 2023
Write up results comparing samples spatially	Jul 2023-Dec 2023

References

- Ackerman JL, Bellwood DR (2000) Reef fish assemblages: a re-evaluation using enclosed rotenone stations. *Mar Ecol-Prog Ser* 206:227–237
- Allen GR (2015) Review of Indo-Pacific coral reef fish systematics: 1980 to 2014. *Ichthyol Res* 62:2–8
- Arias-Ortiz A, Serrano O, Masqué P, Lavery PS, Mueller U, Kendrick GA, Rozaimi M, Esteban A, Fourqurean JW, Marbà N (2018) A marine heatwave drives massive losses from the world's largest seagrass carbon stocks. *Nat Clim Change* 8:338
- Bellwood DR, Hoey AS, Ackerman JL, Depczynski M (2006) Coral bleaching, reef fish community phase shifts and the resilience of coral reefs. *Glob Change Biol* 12:1587–1594
- Bellwood DR, Streit RP, Brandl SJ, Tebbett SB (2019) The meaning of the term 'function' in ecology: A coral reef perspective. *Funct Ecol* 0: +
- Berry TE, Osterrieder SK, Murray DC, Coghlan ML, Richardson AJ, Grealy AK, Stat M, Bejder L, Bunce M. (2017). DNA metabarcoding for diet analysis and biodiversity, a case study using the endangered Australian sea lion (*Neophoca cinerea*). *Ecol Evol*, 7, 5435–5453.
- Brandl SJ, Goatley CH, Bellwood DR, Tornabene L (2018) The hidden half: ecology and evolution of cryptobenthic fishes on coral reefs. *Biol Rev*
- Deagle BE, Tollit DJ. (2007). Quantitative analysis of prey DNA in pinniped faeces, potential to estimate diet composition? *Conserv Genet*, 8, 743–747.
- Depczynski M, Bellwood DR (2004) Microhabitat utilisation patterns in cryptobenthic coral reef fish communities. *Mar Biol* 145:455–463
- Depczynski M, Fulton CJ, Marnane MJ, Bellwood DR (2007) Life history patterns shape energy allocation among fishes on coral reefs. *Oecologia* 153:111–120
- Farmer BM, Wilson SK (2011) Diet of finfish targeted by fishers in North West Australia and the implications for trophic cascades. *Environ Biol Fishes* 91:71–85
- Fisher R, Wilson SK, Sin TM, Lee AC, Langlois TJ (2018) A simple function for full-subsets multiple regression in ecology with R. *Ecol Evol* 8:6104–6113
- Fulton CJ, Abesamis RA, Berkström C, Depczynski M, Graham NAJ, Holmes TH, Kulbicki M, Noble MM, Radford BT, Tano S, Tinkler P, Wernberg T, Wilson SK (2019) Form and function of tropical macroalgal reefs in the Anthropocene. *Funct Ecol* 0:
- Goatley CHR, Bellwood DR (2016) Body size and mortality rates in coral reef fishes: a three-phase relationship. *Proc R Soc B* 283:20161858
- Graham NAJ, Jennings S, MacNeil MA, Mouillot D, Wilson SK (2015) Predicting climate-driven regime shifts versus rebound potential in coral reefs. *Nature* 518:94–97
- Hughes TP (1994) Catastrophes, phase shifts, and large-scale degradation of a Caribbean coral reef. *Science* 265:1547–1551
- Hughes TP, Anderson KD, Connolly SR, Heron SF, Kerry JT, Lough JM, Baird AH, Baum JK, Berumen ML, Bridge TC (2018) Spatial and temporal patterns of mass bleaching of corals in the Anthropocene. *Science* 359:80–83
- Hughes TP, Graham NAJ, Jackson JBC, Mumby PJ, Steneck RS (2010) Rising to the challenge of sustaining coral reef resilience. *Trends Ecol Evol* 25:633–642
- Kozioł A, Stat M, Simpson T, Jarman S, DiBattista JD, Harvey ES, Marnane M, McDonald J, Bunce M (2018) Environmental DNA metabarcoding studies are critically affected by substrate selection. *Mol Ecol Resour*
- Moore JAY, Bellchambers LM, Depczynski MR, Evans RD, Evans SN, Field SN, Friedman KJ, Gilmour JP, Holmes TH, Middlebrook R, Radford BT, Ridgway T, Shedrawi G, Taylor H, Thomson DP, Wilson SK (2012) Unprecedented Mass Bleaching and Loss of Coral across 12° of Latitude in Western Australia in 2010–11. *PLoS ONE* 7:e51807
- Mouillot D, Graham NAJ, Villéger S, Mason NWH, Bellwood DR (2013) A functional approach reveals community responses to disturbances. *Trends Ecol Evol* 28:167–177
- Pikitch EK (2018) A tool for finding rare marine species. *Science* 360:1180–1182
- Rodemann JR, Brandl SJ (2017) Consumption pressure in coastal marine environments decreases with latitude and in artificial vs. natural habitats. *Mar Ecol Prog Ser* 574:167–179

Stat, M, Huggett MJ, Bernasconi R, DiBattista JD, Berry TE, Newman SJ, Harvey ES, Bunce, M. (2017). Ecosystem biomonitoring with eDNA, metabarcoding across the tree of life in a tropical marine environment. *Scientific Reports*, 7, 12240.

Warwick RM, Clarke KR (1998) Taxonomic distinctness and environmental assessment. *J Appl Ecol* 35:532–543

Wernberg T, Bennett S, Babcock RC, Bettignies T de, Cure K, Depczynski M, Dufois F, Fromont J, Fulton CJ, Hovey RK, Harvey ES, Holmes TH, Kendrick GA, Radford B, Santana-Garcon J, Saunders BJ, Smale DA, Thomsen MS, Tuckett CA, Tuya F, Vanderklift MA, Wilson S (2016) Climate-driven regime shift of a temperate marine ecosystem. *Science* 353:169–172

Willis T, Anderson M (2003) Structure of cryptic reef fish assemblages: relationships with habitat characteristics and predator density. *Mar Ecol Prog Ser* 257:209–221

Wilson SK (2004) Growth, mortality and turnover rates of a small detritivorous fish. *Mar Ecol Prog Ser* 284:253–259

Wilson SK, Fisher R, Pratchett MS (2013) Differential use of shelter holes by sympatric species of blennies (Blennidae). *Mar Biol* 160:2405–2411

Wilson SK, Graham NAJ, Polunin NVC (2007) Appraisal of visual assessments of habitat complexity and benthic composition on coral reefs. *Mar Biol* 151:1069–1076

Wilson SK, Robinson JP, Chong-Seng K, Robinson J, Graham NAJ (2019) Boom and bust of keystone structure on coral reefs. *Coral Reefs* 38:625–635

Study design

Methodology

i) Method/management comparisons

The composition of CRF assemblages will be assessed by; ichthyocide collections, underwater visual surveys and collection of water/sediment for eDNA. CRF assemblages will be sampled at six sites inside sanctuary zones and six sites within recreational zones in the Ningaloo lagoon. Within each of the sites there will be 5-6 replicate survey areas over rubble dominated benthos. This type of benthos is common at Ningaloo, and typically has high diversity and abundance of CRF (Depczynski and Bellwood 2004).

Underwater visual census (UVC) will be used to assess the abundance of CRF relative to more mobile species, including potential predators of CRF, that may also influence their abundance. UVC will be undertaken using 5-6 point counts of 5m radius at each site, recording the size and abundance of all fish species encountered. Following visual assessment, small plots (~0.5m²) within the UVC survey area will be enclosed using a fine mesh and fish within the enclosed area will be collected using clove oil (Depczynski and Bellwood 2004). All CRF collected will be euthanised and preserved for identification.

To assess fish presence using environmental DNA, 5-6 replicate 1L water samples will be collected from the waters surface and 5-6 samples from just above the benthos at each site. Water samples will be filtered using 0.45µm membranes on the day of collection and filters frozen at -20°C. Water has been chosen as the medium for DNA collection as it tends to contain more fish DNA than other substrates (Kozioł et al. 2018). However as CRF are closely associated with the benthos we will also collect 5-6 sediment samples at each site using a 50ml syringe.

DNA extractions, including negative field controls, will be conducted in a dedicated DNA laboratory using automated sample preparation (QIAGEN QIAcube, Venlo, Netherlands). A single step metabarcoding approach using oligonucleotides targeting fish taxa will be used to amplify fish DNA (Stat et al. 2017) and will include both negative and positive laboratory controls. Resulting samples will be pooled (equimolar) into a library and sequenced using an Illumina MiSeq platform (Illumina; San Diego, USA). All data generated through sequencing will be filtered through a series of quality control steps prior to taxonomic assignment (Stat et al. 2017).

ii) Variation in CRF across habitats

CRF assemblages will be assessed across a range of habitats, from healthy coral reefs, with high structural complexity to reefs dominated by canopy forming macroalgae and low levels of hard structural complexity. The extremes of this range represent two prominent and ecologically important habitat types in tropical shallow waters (Fulton et al. 2019), as well as alternate stable states that evolve due to differing exposure to disturbance and anthropogenic stressors (Hughes et al. 2010). In particular, coral dominated reefs are highly susceptible to heat stress and extensive bleaching of corals following heat stress, or gradual erosion of reef resilience

due to fishing, disease and cyclones, can result in phase shifts to macroalgal dominated reefs (Hughes 1994; Graham et al. 2015). Understanding how CRF assemblages vary across this range of habitats will therefore give an indication of how fish assemblages may change due to climate and human related pressures. Identifying species unique to different habitat types will also be the basis of developing suitable indicators for monitoring the condition of CRF relative to environmental pressures. All samples will be collected using the technique identified from method comparisons as being most appropriate for collecting CRF. The habitat present within each site will be assessed using line intercept transects and structural complexity will be recorded using a combination of seascape and small scale measures relevant to fish of different body sizes (Wilson et al. 2007, 2013).

Statistical analyses

i) Method/management comparison

Data from each sample will be analysed to determine the number of species present and indices of; species, taxonomic (Warwick and Clarke 1998) and functional diversity (Mouillot et al. 2013) calculated for all replicates. How these measures of diversity differ with respect to the sampling methods (UVC, eDNA, ichthyocides), and management zones (sanctuary, general use) will be investigated using generalised additive mixed models. Method and management zones will be included in models as fixed factors and sites will be random factors. All combinations of variables will be considered in analyses and the best model will be identified using AIC and model weights (Burnham and Anderson 2010; Fisher et al. 2018). Differences in CRF assemblage composition among methods, and management zones will be examined using permutational analysis of variance.

ii) Variation in CRF across habitats

The influence of habitat on CRF assemblages will be assessed using a combination of GAMM (Generalised additive mixed models), DistLM (Distance based linear modelling) and PCO (Principal Coordinates Analysis). The number of individuals, species, taxonomic and functional diversity will be calculated for each replicate and used as response variables in GAMM, whilst habitat measures (e.g. coral cover, macroalgal cover, complexity) will be co-variables and site a random factor. All combinations of variables will be considered using a full subsets approach (Fisher et al 2018) and the best model for explaining variation in community measures will be based on the lowest AIC value and model weights (Burnham and Anderson 2010). Differences in CRF community composition will be examined using DistLM, with habitat measures used as predictor variables and findings viewed in multivariate space using PCO.

Biometrician's Endorsement

required

Data management

No. specimens

Herbarium Curator's Endorsement

not required

Animal Ethics Committee's Endorsement

required

Data management

The raw data will be stored on MSP T drive with a full back-up stored on an external hard drive. Additionally, a page will be created on the DBCA Confluence site that will provide links to the data sets (csv), R scripts, and output figures as appropriate. The metadata reports for each field trip will be uploaded onto Confluence under a newly created project linked to the SPP number.

Data custodian: Marine Science Program, Shaun Wilson

Budget

Consolidated Funds

Source	Year 1	Year 2	Year 3
FTE Scientist	0.5	0.5	0.5
FTE Technical	0.05	0.05	0.05
Equipment			7000
Vehicle			
Travel	5000	5000	5,000
Other			
Total	5000	5000	12000

External Funds

Source	Year 1	Year 2	Year 3
Salaries, Wages, Overtime	0.25 FTE	0.25 FTE	0.25 FTE
Overheads			
Equipment	12500		5000
Vehicle			
Travel	1000	1000	1000
Other	1000	1000	1000
Total	14500	2000	7000