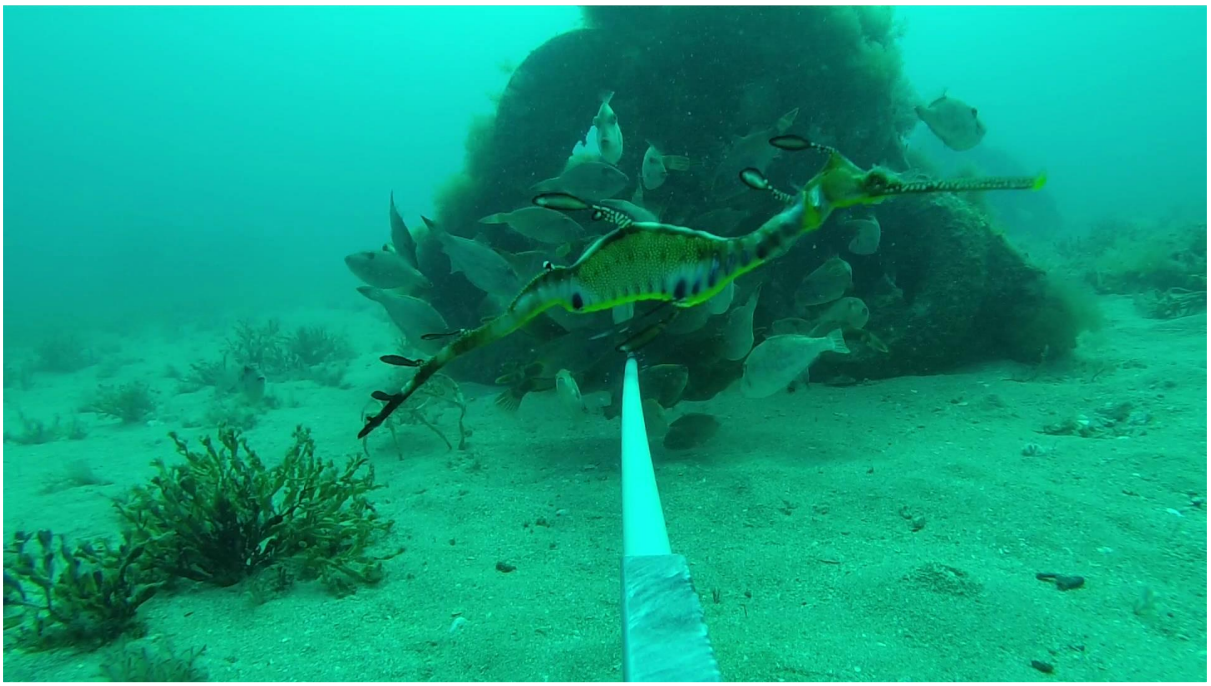
Investigating the Anthropocene influence on temperate fish assemblages through Baited Remote Underwater Video Stations (BRUVS)



Sasha Whitmarsh

Thesis

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College of Science and Engineering, Flinders University, Adelaide, Australia

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Contents

Thesis summary

The ever increasing human population and subsequent pressure this places on the marine environment has been the focus of studies for many years, with effects from some potential stressors receiving widespread attention, e.g. climate change. The global use of our marine resources has led to various detrimental effects including population declines, local species extinctions, and a loss of ecosystem services, with these events now being referred to as Anthropocene defaunation. Fish assemblages play a key role in the functioning of many marine ecosystems and are an integral part of most human societies through fishing or recreational activities (e.g. snorkelling/diving). However, research has often focused on commercial, tropical, or reef species, while temperate areas, soft sediment habitats, and non-commercially targeted species have typically been less often investigated. The objective of my thesis is to describe such under-studied fish assemblages and assess the effects of increasing anthropogenic activities in South Australia through use of a contemporary non-extractive monitoring method. I conducted over 600 deployments using Baited Remote Underwater Video Stations (BRUVS) to answer three broad aims: 1) describe temperate fish communities and their natural variations; 2) determine how fish assemblages respond to differing anthropogenic stressors; and 3) investigate how BRUVS assess fish assemblages and ways it may be improved as a method. Through a thorough literature review, I was able to highlight variations about the BRUVS method that cause discrepancies across studies, and the potential impacts of underreporting such variation. From my studies, I found a wide variety of fish assemblages inhabiting South Australian waters, which were often site-specific and varied by habitat type, season, and across years. My anthropogenic case-studies showed varying levels of influence from potential stressors and began an understanding of cumulative stressors, which has previously been difficult to untangle. Specifically, I found some areas were responding to early protection from fishing with increases in the abundance of certain fisheries species; no evidence of an influence of brine outfall from desalination processes; limited influence from bait and berley input from tourism activities at the Neptune Islands group; and influences from effluent and oyster leases on the fish assemblages observed in Coffin Bay. I was also able to determine the impact of additional viewpoints on the abundance and assemblages observed and use these additional viewpoints to reduce some of the biases inherent with using BRUVS in a traditional manner. My results are relevant to future studies

using BRUVS to provide a comprehensive species list and to quantify nighttime fish assemblages. Findings from my thesis will also enable a better understanding of fish assemblages in temperate Australia with a view to inform management decisions.

Declaration

I certify that this thesis does not incorporate without acknowledgment any material previously submitted for a degree or diploma in any university; and that to the best of my knowledge and belief it does not contain any material previously published or written by another person except where due reference is made in the text.

Author contributions, permits, and funding

The following outlines the author contributions for each data chapter:

2. Whitmarsh SK, Fairweather PG, Huveneers C (2017) What is Big BRUVver up to? Methods and uses of baited underwater video. Rev Fish Biol Fish 27: 53-73

Sasha K. Whitmarsh (SKW) Peter G. Fairweather (PGF) and Charlie Huveneers (CH) conceived and designed the study. SKW collected and analysed the data. PGF and CH advised on the data analysis. SKW wrote the manuscript. PGF and CH contributed to revising the manuscript.

3. Whitmarsh SK, Fairweather PG, Huveneers C (in prep) Multi-scale analysis of temporal changes in fish assemblages.

SKW, PGF and CH conceived and designed the study. SKW and CH collected the data and SW analysed the data. PGF and CH advised on the data analysis. SKW wrote the manuscript. PGF and CH contributed to revising the manuscript.

5. Whitmarsh SK, Huveneers C, Fairweather PG (2018) What are we missing? Advantages of more than one viewpoint to estimate fish assemblages using baited video. R Soc Open Sci 5: 171993

SKW, PGF and CH conceived and designed the study. SKW collected and analysed the data. PGF and CH advised on the data analysis. SKW wrote the manuscript. PGF and CH contributed to revising the manuscript

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I have had the utmost pleasure to have had two excellent supervisors throughout my PhD. While many forms prescribe a primary and associate supervisor, it seemed unfair to me for much of my time about whom I would place where. And it is also difficult to decide who to thank first here, but as I always hated as a child we will go in alphabetical order.

Throughout the time I have known Peter Fairweather I have valued and respected his advice and opinions and, during my PhD it has been no different. His immense amount of knowledge on just about every subject helped me enormously, but particularly I have valued his advice about experimental design and statistics. While not always the most forthcoming in a straight answer, his ability to help guide and develop my critical thinking has made me into a better scientist. "If it was easy everyone would do it'" and "just because other people do it that way doesn't mean it's right" these are the phrases he would say in our weekly meetings and I interpret them as follows, do not expect ecology to be simple and do not blindly choose the easiest path. I would like to thank Peter for all he has done for me, there are many people without whom this PhD would not have been possible but perhaps foremost of those is Peter.

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and encouraging all the things I ever took an interest in. Thank you for never doubting me.

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And now a short poem about BRUVS:

A fish, a fish I cried with glee
And marked it down triumphantly.
A MaxN of one is nice to see
When there's been nothing previously.
Oh look more fish, I'll note them down
And increase my count to ensure it's sound.
But then there's more,
They're everywhere.
New species, more sightings,
The fish are all biting.
The time is dragging and my patience is lagging.
Oh look a fish, it fills me with dread
But at last an hour, now I can go to bed.

Chapter 1

General Introduction

Anthropogenic influences on ecosystems

Exponential human growth, industrialisation, and technological advances have increasingly put pressure on the earth's resources and ecosystems (Vitousek et al. 1997; Halpern et al. 2008a; Ceballos et al. 2015). The comparatively rapid timescale at which such changes have been taking place has led to little time for species to adapt and also to the loss of species biodiversity and local population declines. The link to human activities has led to the term "Anthropocene defaunation" being used to denote the rapid biodiversity changes recently observed (Dirzo et al. 2014). Estimates indicate that present extinction rates may be up to 100 times higher than the extinction rates over the last century (Ceballos et al. 2015), with significant declines in the abundance of both vertebrate and invertebrate populations suggesting that a sixth mass extinction is already under way (Dirzo et al. 2014). In the marine realm, human activities have altered ecosystems and led to global and regional declines in abundance and biodiversity (McCauley et al. 2015). In some extreme cases, such declines have resulted in species loss or ecological extinction with species being unable to fulfil their roles within the ecosystem, leading to substantial alteration to the structure and functioning of the environment (McCauley et al. 2015).

Urbanisation, invasive species, climate change, habitat loss, pollution, and overfishing have all been identified as environmental stressors linked to increasing anthropogenic activities (Worm et al. 2006; Ceballos et al. 2015; McCauley et al. 2015; Robbins et al. 2017). The magnitude of the impact of these stressors are also inter-connected and can have synergistic effects. For example, urbanisation can lead to a loss of habitat, increased nutrient pollution and eutrophication, an increase in the use of waterways by humans (i.e. for boating, diving, fishing use), increased light and/or sound pollution, and facilitate the spread of exotic species. These often have negative effects on species abundance, diversity, and ecosystem function (Lee et al. 2006; Bulleri & Chapman 2010; Dafforn et al. 2015). While some of these stressors are well known and have received ongoing research attention (e.g. climate change), there remains many which have been less focused on (e.g. light and sound pollution). Few studies also consider cumulative impacts despite knowledge of their potential interactive effects (Halpern et al. 2008b).

The importance of understanding fish assemblages

Marine fishes are ecologically and economically important around the globe. Ecologically, large teleost fishes and many shark species serve as top predators, occupying high positions within food webs, and are thought to exert significant influence on the structure and function of ecosystems. Studies of predator-prey interactions have shown that changes in predator abundance have resulted in large cascading changes in lower trophic species (Ruppert et al. 2013). The large variety of niches occupied and multitude of dietary preferences of fishes enable further regulation of food chains (Holmlund & Hammer 1999). Many fish species play an important role in coupling and stabilising disparate ecosystems due to their movement patterns and allow the transport of nutrients across spatial boundaries (Holmlund & Hammer 1999). Fishes also serve to regulate ecosystem resilience by providing services such as herbivory in coral reef areas (Hughes et al. 2005), playing an important role in the distribution of bottom substrates and sedimentary processes, and affecting the recycling of nutrients and carbon fluxes within and between the water and atmosphere (Holmlund & Hammer 1999).

Fish and fisheries are likewise economically important, with wild-caught fisheries contributing ~90 million tonnes annually to global fishing economies (FAO 2016). For the past 50 years, annual fish consumption per capita has doubled from an average of 10 kg to more than 20 kg (FAO 2016) and it is estimated that over 56 million people were engaged in primary fisheries activities in 2014 (FAO 2016).

Due to this economic importance of commercially-targeted species, many studies focus on those species considered more valuable, leading to a comparative paucity of information on non-commercial species and community-level assemblages more broadly (Pope et al. 2000). Along with these economic reasons, some areas are also more difficult to study due to logistics and hence financially costly for researchers as they can be harder to access. This has led to vast amounts of knowledge being available about well-studied species and areas (e.g. commercial species, reef areas, tropical regions) but leaves knowledge gaps for the less-studied species and areas (e.g. non-targeted species, nocturnal assemblages, soft-sediment habitats, and temperate or polar

regions). Without continued research into fish and their community structures, knowledge of interactions between species and their environment may remain unclear.

There is also a demonstrated need for continued monitoring and baseline knowledge for many specific areas to track changes over time (Halpern & Warner 2002; Gerber 2005; Stuart-Smith et al. 2017). Areas that are protected or have significant ecological or commercial interest are typically monitored more frequently (Whitmarsh et al. 2017). Studying assemblages can also lead to increased knowledge of ecosystem function and resilience, which is important to predict the effects of future environmental change (Stuart-Smith et al. 2017; Wu et al. 2017). A strong public connection with fish exists especially those charismatic in nature or of recreational and commercial fishing interest, this leads to fishes also having high socio-economic value in many countries worldwide. Tourism from coral-reef ecosystems alone is worth US$36 billion per year (Spalding et al. 2017) and shark-based tourism in Australia worth AUD$47 million per year (Huveneers et al. 2017).

How fish assemblages can be monitored and assessed

A range of destructive and non-destructive methods are currently available to assess fish assemblages. Destructive and extractive methods are often more traditional ones such as hook and line fishing, netting, traps and trawling, along with other techniques such as using rotenone to poison fishes. Such methods can be fine-tuned to target specific fish species (e.g. hook and line fishing and traps) or be used to sample a wide range of species (e.g. trawling and poisoning). However, destructive and extractive techniques are often not suitable for use in all areas and may be avoided by researchers seeking better alternatives. Non-destructive and non-extractive alternatives to these traditional techniques have been growing in popularity, particularly since the implementation of video technology from the 1980's onwards (Murphy & Jenkins 2010; Mallet & Pelletier 2014). Commonly applied non-destructive techniques used to assess fish assemblages include Underwater Visual Census (UVC) and Baited Remote Underwater Video Stations (BRUVS). UVC is often used in shallow areas (suitable for divers) that have clear visibility and is popular in coral-reef locations (Colton & Swearer 2010). Divers' depth limitation and increasing WHS restrictions has resulted in the use of divers becoming prohibitive for sampling many areas.

Baited Remote Underwater Video Stations (BRUVS) are a commonly-used alternative method to assess fish populations (see Chapter 2 for literature review about using BRUVS). Briefly, they consist of one or more camera(s) attached to a frame with bait present to attract fish and other mobile fauna into the field of view for later analysis. The benefits to using BRUVS include: (1) having a non-destructive approach suitable in marine protected areas or other sensitive locations; (2) yielding a permanent, archivable record; (3) having the ability to record a diverse range of species (including both targeted and non-targeted species); and (4) is a relatively cheap and easy-to-use method. They can be used in more areas compared to UVC and are often deemed a more suitable choice because of the depth limitations and diver biases of UVC (Colton & Swearer 2010; Lowry et al. 2012). Although BRUVS are used by the South Australian government to monitor the new marine parks (described below), few studies have used BRUVS in South Australia [but see Svane and Barnett (2008); Svane et al. (2008); Whitmarsh et al. (2014); and Kelaher et al. (2014)].

Study region and related anthropogenic activities

South Australia is situated on the central-southern coast of Australia. It is a temperate area with 5,067 km of coastline extending from the rugged South East coast to the steep cliff faces of the Great Australian Bight (GAB; Geoscience Australia 2017). Major features of the coastline include the aforementioned GAB, the Bonney Upwelling region, two gulfs (Gulf St Vincent and Spencer Gulf) that act as large inverse estuaries, the mouth of the River Murray (Australia's largest river system) which consists of the Murray estuary, Coorong and Lower Lakes, and Kangaroo Island (Australia's third largest island). South Australian waters are situated at the confluence of two major current systems, the Leeuwin Current (from the west) and the Flinders Current (from the east), resulting in the transport of nutrients and organisms from both directions of Australia (Kämpf et al. 2004). This, along with the relative isolation and long southern facing coastline and a diverse range of habitats, helps promote the high rate of endemism that is evident in South Australia (Edyvane 1999; Phillips 2001) with approximately 85 % of South Australian marine species considered endemic to the region (Government of South Australia 2004). The range of habitats includes rocky reefs, deep-water sponge gardens, extensive seagrass meadows, sand and soft-sediment

areas, kelp forests, mangroves and saltmarshes, high-energy wave-exposed coastlines and shallow, sheltered lagoons and bays.

Over the last few decades, there have been significant opportunities for industrial and urban expansion in South Australia. This has led to an increased use of its marine environment, primarily close to populated regions or resource centres such as the two gulfs and Eyre Peninsula. Examples of these human activities include oil and gas exploration, commercial fisheries and aquaculture, desalination plants, aquaculture activities, tourism, and protection in form of marine parks. More specifically, exploration for oil and gas, and flow-on effects from mining operations (such as construction and maintenance of large ports) have resulted in substantial shipping and dredging activities (Doubleday et al. 2017). Commercial wild-fisheries (e.g., southern rock lobster, prawn, snapper, garfish, King George whiting, abalone, crab, sardine) and aquaculture (e.g., southern bluefin tuna, yellowtail kingfish, abalone, oyster, mussel) provide important economic returns to the State and are expanding (PIRSA 2014; 2015). There are also many recreational uses of the marine environment within South Australia with activities ranging from beach and seashore trampling, scuba-diving, snorkelling and swimming, shore and boat-based recreational fishing, and wildlife tourism such as swim-with-sea-lions, swim-with-dolphins, swim-with-tuna ventures, and white shark cage-diving. The increasing concern about the effects of such activities on the marine environment has prompted studies about the potential effects of these anthropogenic threats e.g. the Adelaide Coastal Waters Study (Fox et al. 2007) and the Spencer Gulf Ecosystem and Development Initiative (Doubleday et al. 2017, Robbins et al. 2017). While these studies have assessed potential effects through modelling and risk management frameworks, few studies have assessed what the effects were or described how to assess such effects.

Alongside the increasing use of the South Australian coastline, the South Australian government contributed towards Australia's commitments made under the 2002 World Summit on Sustainable Development to effectively protect 20–30 % of each marine habitat by 2012 (IUCN 2003). As a result, Australia implemented a plan for a National Representative System of MPAs (NRSMPA) in 1998 after signing up to the Convention on Biological Diversity (CBD) in 1993. In response to increasing usage, the South

Australian Government gazetted 19 new marine parks covering 44 % of state waters, with 6 % of the area designated as sanctuary or 'no-take' zone. Fishing restrictions (and all other management actions) within the parks were put into effect from October 2014 but some sanctuary zones were already established prior to the creation of the marine parks, from as early as 1971. Apart from those no-take or sanctuary zones, the parks also feature habitat protection zones (designed to limit damage to the seabed) and restricted access zones (no unauthorized entry) as well as general managed use zones which offer only little additional protection to areas outside a park.

Thesis aims

This study was initiated because of 1) the relatively understudied nature of some temperate fish assemblages, particularly those from understudied habitats (e.g. soft-sediment areas) in South Australia; 2) the need for effective methods of monitoring and studying fish assemblages; and 3) increased information required in the face of a changing marine environments and increasing anthropogenic pressures. Thus, the overall objective of my thesis is to describe so-far under-studied fish assemblages and assess the effects of increasing anthropogenic activities in South Australia through a contemporary non-extractive monitoring method.

To achieve this overall objective, I aim to:

1) Describe temperate fish communities and their natural variations;
2) Determine how fish assemblages respond to differing anthropogenic stressors; and
3) Investigate how BRUVS assess fish assemblages and ways it may be improved as a method.

To fulfil each aim, I have compiled four thesis chapters (excluding this introductory chapter [1] and a general discussion chapter [6]), each with specific goals which link to an aim and can be visualised in Figure 1.1.

Structure

Chapter 1 is a brief introductory chapter providing background information on the major thesis themes and outlines the overall thesis objective and structure. I have kept

this chapter brief as further introductory material can be found within each chapter as well as extensive information in a literature review on BRUVS in Chapter 2 (see below).

Chapter 2 is a literature review about the methodological uses of BRUVS. It has been published in *Reviews in Fish Biology and Fisheries* and provides detailed information on the methodological aspects of using baited underwater video as well as highlights how this method has been used in the past and current novel approaches. This chapter feeds into Aim 3 (Figure 1.1).

Chapter 3 is a multi-scale analysis of the temporal variations of fish assemblages. It assesses temporal change over short and long-term timescales in multiple habitat types with goals that feed into Aim 1 and Aim 3 (Figure 1.1). This chapter is currently in preparation to be submitted.

Chapter 4 assesses the effects of differing anthropogenic stressors on fish assemblages and comprises four case studies in different regions of South Australia, including protected areas in the Upper Gulf St Vincent, effluent output in the lower Gulf St Vincent, cumulative stressors including aquaculture and fishing in Coffin Bay, and bait and berley input due to shark-cage diving at the Neptune Islands. Goals from this chapter feed into Aim 1 and Aim 2 (Figure 1.1).

Chapter 5 uses a novel approach to test the potential fixed viewpoint bias of BRUVS by assessing the impact of additional cameras on the abundance and diversity metrics obtained. This chapter has been published in *Royal Society Open Science*. The goals from this chapter feed into Aim 3.

Chapter 6 discusses the major findings and synthesises the results of Chapters 2–5. It highlights the management implications of my research and identifies areas of future research.

The focus for this thesis was on the assemblages of fish and other nektonic animals observed from BRUVS. As such these studies can be considered community ecology and hence the data sets were multivariate (i.e. multiple species) in nature, although, a few

particular species of fish were analysed separately for specific questions. Hence, the key attributes being analysed in each data chapter were the assemblages' taxonomic composition (species presence/absence) and relative abundances (collated MaxN values for each species seen). Most data analysis was done using the software PRIMER v7 with the PERMANOVA+ add-on (Anderson et al. 2008; Clarke and Gorley 2015) although other programs were used as needed for other analyses and datasets. The consistency of this approach, especially when done using software that is very flexible in the designs that could be tested, has few limiting assumptions, and is mostly based on randomisation (i.e. distribution-free) testing, was seen as an advantage over any approach that was less systematic and might vary radically from chapter to chapter.

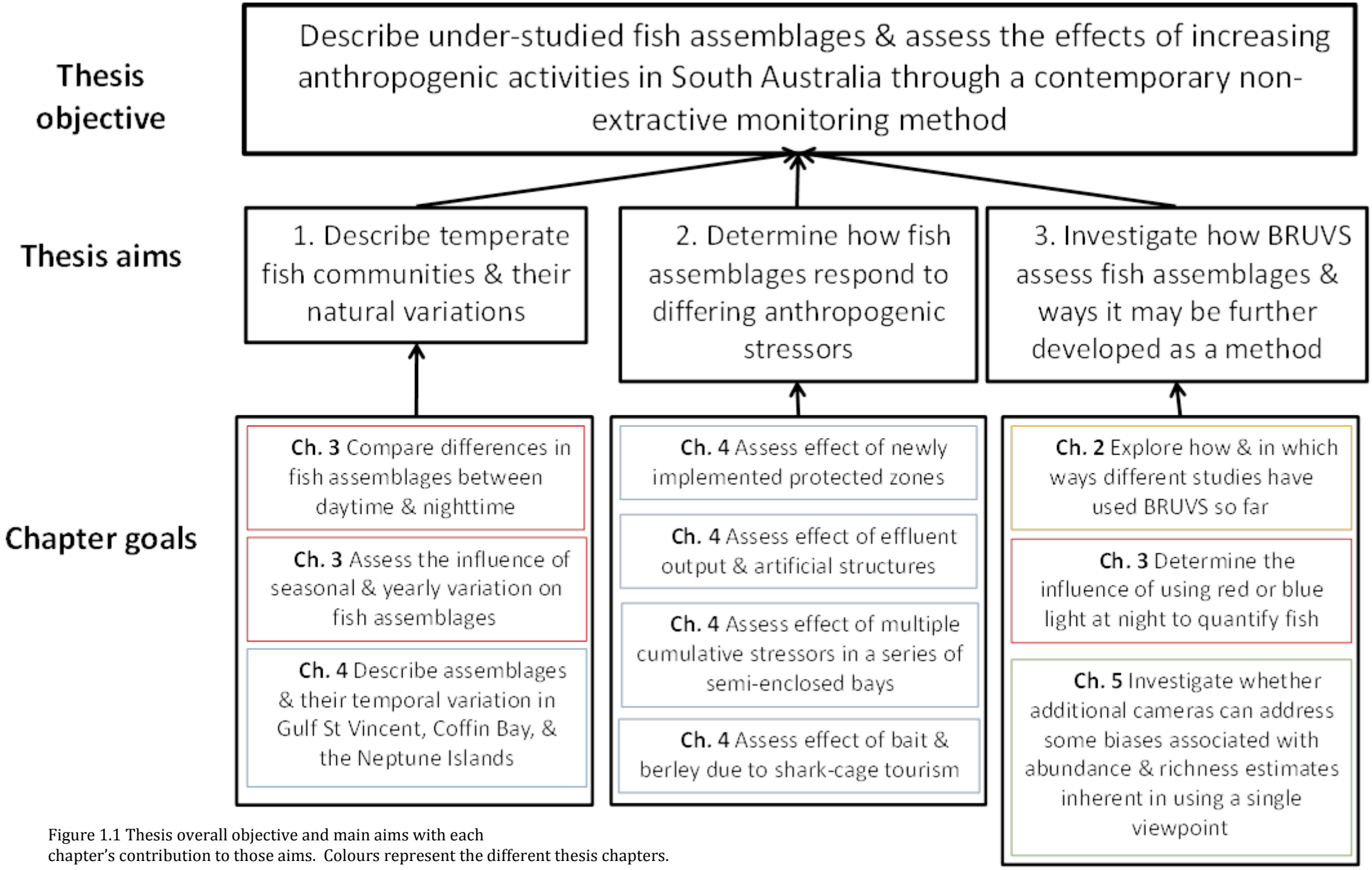

Figure 1.1 Thesis overall objective and main aims with each chapter's contribution to those aims. Colours represent the different thesis chapters.

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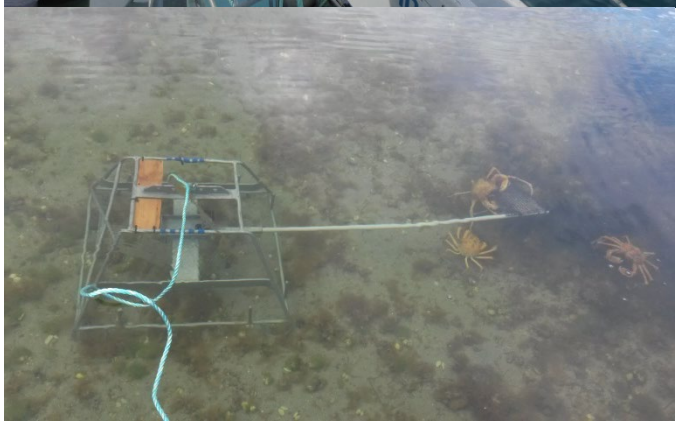

Chapter 2

What is Big BRUVver up to? Methods and uses of baited underwater

Sasha K. Whitmarsh
Peter G. Fairweather
Charlie Huveneers

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Abstract

Baited Remote Underwater Video Stations (BRUVS) is a popular technique to assess mobile nektonic and demersal assemblages, particularly for fish communities. The benefits of using BRUVS have been well documented, with their non-destructive and non-extractive nature, ease to replicate, relatively-cheap personnel costs, and low risk to personnel often cited. However, there is a wide variability in the set-up, experimental design, and implementation of this method. I performed a literature review of 161 peer-reviewed studies from all continents published from 1950 to 2016 to describe how BRUVS has been used by quantitatively assessing 24 variables, including camera set-up and orientation, soak time, bait quantity, type and preparation method, habitat and depth deployed in, and number of replicates used. Such information is critical to gauge the comparability of the results obtained across BRUVS studies. Generally, there was a wide variety in the location, deployment method, bait used, and for the purpose that BRUVS was deployed. In some studies, the methods were adequately described so that they included information on the 24 variables analysed, but there were 34 % of studies which failed to report three or more variables. I present a protocol for what minimal information to include in methods sections and urge authors to include all relevant information to ensure replicability and allow adequate comparisons to be made across studies.

Introduction

Information about marine ecosystems is becoming increasingly sought after as the understanding of their importance in ecosystem services, global processes, and economies increases (Costanza et al. 1997). For many of these services, fish and other nekton are particularly important and have been the main focus of several studies (e.g. Holmund and Hammer 1999; Worm et al. 2006). Such studies have highlighted the need for methods which are capable of sampling a large portion of the population or community, are non-extractive, and allow for simultaneous counts of multiple taxa. There is also a growing desire for more behavioural data about fish species, along with less destructive methods suitable for protected areas, and for methods that are cheap, repeatable, and comparable. Baited underwater video (for the purpose of this review referred to as Baited Remote Underwater Video Stations or BRUVS) is a popular

technique to assess mobile nektonic and demersal assemblages, particularly for fish communities and fits the above criteria.

BRUVS have been compared to many other commonly-used techniques for assessing fish assemblages with the most common comparison being between BRUVS and Underwater Visual Census (UVC) (e.g. Stobart et al. 2007, Colton and Swearer 2010, Lowry et al. 2012) or Diver Operated Video (DOV) (Watson et al. 2005, Langlois et al. 2010, Watson et al. 2010). Other comparisons include: BRUVS vs. baited traps (Harvey et al. 2012a; Wakefield et al. 2013, Langlois et al. 2015); vs. angling (Willis et al. 2000, Langlois et al. 2012a; Gardner and Struthers 2013); vs. trawling (Cappo et al. 2004); vs. seine netting (Whitmarsh 2012); vs. longline surveys (Brooks et al. 2011, Santana-Garcon et al. 2014a; McLean et al. 2015); and vs. Automated Underwater Vehicles (AUV) and towed video (Seiler 2013). These studies show that BRUVS are a useful tool with many benefits compared to more traditional techniques. Nevertheless, as each study has aims that vary, the appropriate method to use should be selected on a case-by-case basis (see Murphy and Jenkins (2010) or Mallet and Pelletier (2014) for a review of the benefits and biases of these methods in relation to BRUVS).

Over the last 15 years, as the available technology improved and the aims of studies using this equipment have broadened, the methods used when deploying BRUVS have progressively increased in variety. Factors that can vary from study to study include: the number and orientation of cameras; soak time (i.e. the amount of time the unit is left underwater); habitat(s) sampled; depth ranges of deployments; and the number of replicates used. The bait used can differ in terms of type, quantity, and preparation method. The type of video metric (i.e. how fish and other nekton are counted or measured) can also be different across studies. Standardisation in the use of BRUVS has previously been attempted (Cappo et al. 2007) to allow for a better comparison across studies, but modified or novel approaches to this technology are continually arising, increasing variability in methods used. I propose that authors should ensure that they provide enough information to allow comparisons between the different BRUVS set-ups used, instead of attempting to reach a level of standardisation that might not be achievable. The overall purpose of this literature review is to explore how and in which ways different studies have used BRUVS. I hope to highlight: the need for a

comprehensive and descriptive method section; aspects which could be further investigated to improve the informational output of BRUVS; other unexplored applications of BRUVS; and ultimately suggest a protocol of information that authors should routinely include in the methods section.

Searches of the peer-reviewed literature were conducted up to 18/07/2016 using the keywords "baited and video" or "BRUVS", within Google Scholar, Scopus, Proquest (Aquatic Sciences and Fisheries Abstracts), and Biological Abstracts for the time period between 1950 and the search date. Searches returned between 59 and 497 hits across the various databases, with additional (10,000 +, mostly irrelevant) hits from Google Scholar for the "baited and video" search term. Papers were included in the analysis if bait was used in one or more replicates and if video footage was used rather than still images. A total of 161 studies were found (Appendix 1,Table 7.1), from which 24 variables of the study were extracted (Table 2.1). The purpose and novelty of the studies were also assessed.

Results and discussion

A comparison of methods used in baited video studies

Description of the study: when and where

Studies using baited videos began in the mid-nineties (Ellis and DeMartini 1995) and have increased over time (Figure 2.1A), with 33 studies published in 2015 and 13 (plus three in press) in the first half of 2016. The year 2007 appeared to be a breakthrough year for BRUVS studies going from 1 in 2006 to 8. This increase may in part be due to a workshop on baited video held at a national conference in Australia in 2006. The increase in BRUVS studies over time is likely to be due to an increased exposure of the method and its benefits, advances in technology, and the trend towards more affordable electronic equipment.

Table 2.1: A list of the 24 variables included in this review of baited studies that also will act as a protocol for factors to include in method sections. In addition to those listed I also suggest including the time of day the study was conducted and any additional items added to the system such as lights or current meters.

Variable	**Examples**	**# of studies reported in (% out of 161)**
When and where		
Year published	1996, 2006, 2016	100 %
Location study was conducted in	Adelaide, South Australia, Australia	100 %
Geographical area	Temperate, tropical, polar	100 %
Aquatic realm	Marine, estuarine, freshwater	100 %
Habitat type	Seagrass, rocky reef	97 %
About the video system		
Name of systems	BRUVS	96 %
Orientation of camera(s)	Horizontal, vertical (to substrate)	99 %
Number and type of cameras	1 or 2, GoPro Hero 3+, Panasonic HandyCam	99 %
Type of length measurement	Fork length using stereo-BRUVS	85 %
Max range visible	3 m, to bait bag	46%
Soak time	30 min, 60 min	98 %
Distance between reps	250 m, 500 m	65 %
About the bait		
Type	Sardines, *Sardinops sagax*	94 %
Quantity	500 g, 1000 g	84 %
Preparation method	Crushed, whole, chopped	84 %
Deployment method	mesh bag, perforated PVC bait container	82 %
About the deployment		
Minimum depth	3 m, 10 m	85 %
Maximum depth	50 m, 25 m	86 %
Variation in depth (range)	47 m, 15 m	82 %
About the sampling design and analysis		
Number of replicates	3, 6	93 %
Video metric used	*MaxN*, T1st, etc.	99 %
Software used	EventMeasure, VLC etc.	54 %
Taxa included	Teleost, Chondrichthyes, Cephalopoda, Crustacea	96 %
% to species level	75 % able to be identified to species level	55 %

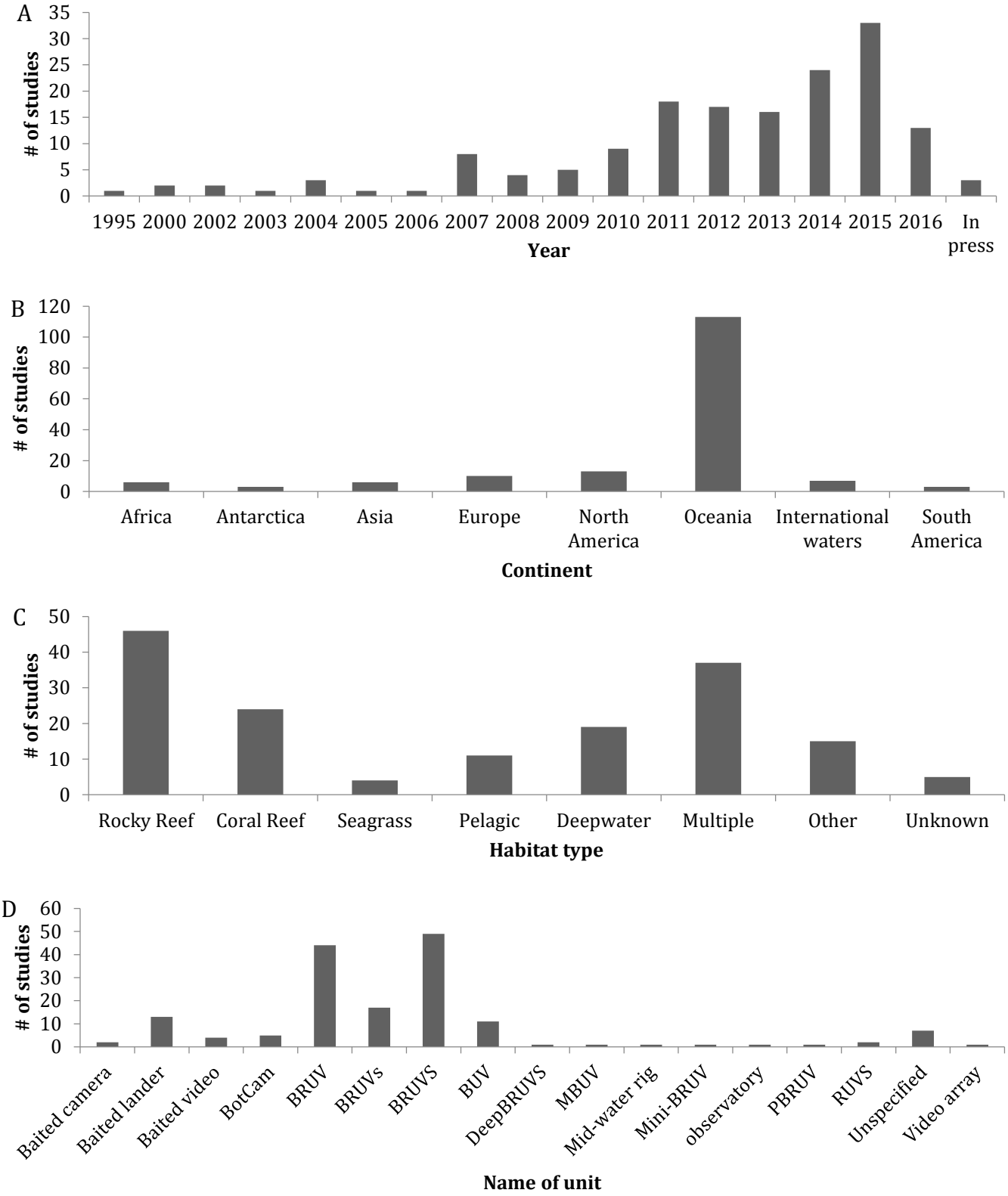

Figure 2.1: A) The frequency of BRUVS studies published by year until 18/07/2016. B) The continent or geographical realm in which each study was conducted. C) The habitat type in which BRUVS were deployed for the 161 studies assessed. The 'Multiple' category was used where more than one habitat type was studied and included some of the other habitat categories listed (except for pelagic and deep-water), as well as some included in the 'Other' category, such as bare sand. 'Deep-water (>100 m)' habitats included shelf slope, soft sediments and hard substrates. D) Frequency of the name given by the study's authors for the baited video unit from 161 studies assessed.

I found that BRUVS studies had been performed in all continents as well as within international waters (Figure 2.1B). Oceania was by far the most popular (70 %) region for studies to be conducted in, with Australia alone contributing 61 %. Other continents had generally fewer studies conducted there, especially Africa (6), Asia (6), and South America (3) along with Antarctica (3). Geographically, temperate areas were the main focus areas at 42 %; however, tropical and sub-tropical areas were still well-represented at 26 % and 35 %, respectively. Polar areas received less attention and were only investigated in 2 % of studies. The majority of the studies included in this review were exclusively in marine ecosystems (94 %), while 4 % were in estuarine and only 2 % in freshwater ecosystems (e.g. Ebner and Morgan 2013; Ebner et al. 2015).

The most common habitat in which BRUVS were deployed was reef areas, with coral and rocky reefs together accounting for 43 % of the habitats studied (Figure 2.1C). Studies in multiple habitats (23 %) also commonly used reef habitats as one of their sampled areas. Rocky reefs were more commonly sampled than coral reefs, which may be due to the prevalence of rocky reef areas within the temperate regions of Australia, where a large proportion of BRUVS studies are conducted. Pelagic (7 %; e.g. Rees et al. 2015) and deep-water (12 %; e.g. Collins et al. 2002) habitats were also studied. Seagrass and 'other' habitats were less common with only 2 % for seagrass (e.g. Whitmarsh et al. 2014) and 9 % for the 'other' category, which included soft sediments (e.g. Howarth et al. 2015) and restricted habitats such as intertidal rock pools (e.g. Harasti et al. 2014). The prevalence of use in reef habitats is most likely a factor of increased visibility through the water column compared to some other benthic (soft bottom) habitats. Reef areas are often home to commercially-targeted fish species and are prime areas for tourism such as snorkelling and diving, which makes these areas of high commercial interest. Ecologically, reef areas support a wide range of species and usually have high biodiversity (Malcolm et al. 2007) leading them to be targeted by researchers and managers.

Variables relating to the video system

The terminology surrounding BRUVS was widely variable, with the name of the unit falling into more than 17 categories (Figure 2.1D). Three very similar unit names dominated the literature: BRUV, BRUVS and BRUVs; all acronyms standing for Baited Remote Underwater Video Stations or Systems (Figure 2.1D). BRUV(S/s) as an acronym appears to have first been published by Cappo et al. (2001). Other common names include BUV (Baited Underwater Video) and more general names such as baited landers or baited video. Some authors have developed individual names for their systems such as DeepBRUVS (Marouchos et al. 2011) and BotCam (Merritt et al. 2011). Generally, having multiple names can be a problem because it leads to confusion and allows for ambiguity about the method. Multiple names also make literature searches more difficult and may confuse non-specialists. The name BRUVS has been trademarked by AIMS, but it is not linked to any patent of the design and AIMS does not enforce the use of the trademarked name in peer-reviewed publications (M. Cappo, pers. comm.). Since variations on BRUV(S/s) are the most commonly published names for this method, I urge that a standard form of this name be chosen and used. I am recommending BRUVS as this name has most prevalent use (Figure 2.1D).

The orientation of the camera(s) is an important aspect to consider when setting up the BRUVS arrangement. The majority (85 %) of BRUVS set-ups used a horizontal camera arrangement, while 14 % had a vertical orientation pointing down towards the seafloor; the remaining (1 %) studies did not specify the camera orientation. The orientation of the camera can affect the number of organisms that can be observed or reliably identified. For example, Langlois et al. (2006) showed that a horizontal set-up recorded 14 species vs. four for a vertical set-up, with some species appearing shy of entering the vertical field of view, most likely due to the perceived confined space under the camera. A major benefit of vertical set-ups, however, is the ability to measure fish size with single cameras using the known fixed height above the substrate and a ruler to measure fish. Vertical BRUVS were used first in the early 2000's (e.g. Willis and Babcock 2000) but have had limited use across the years, with one third of vertical set-ups occurring in deep habitats. The prevalence of horizontal BRUVS is likely because of the increased field of view (depending on water clarity) and the ease of identification of many fish species from a side-on perspective.

BRUVS are predominantly used with one (single) or two (stereo) cameras. Single-BRUVS consist of one camera usually mounted directly behind or above the bait arm. Stereo-BRUVS consist of two cameras mounted at specific angles (usually 7–8°) to each side of the bait of the arm and are calibrated to allow for accurate fish measurements (see Harvey et al. 2002a for more details). Single-BRUVS are smaller, lighter, cheaper, and take less time to set-up (prior to and during field work) than stereo-BRUVS. Stereo-BRUVS take up more boat space, require more specialised gear for retrieval and may be a limiting factor for replicate numbers when using smaller vessels or make field costs higher by requiring more days in the field than single systems. The calibration of stereo-BRUVS also adds to preparation and analysis time. Based on my literature search, the majority of studies (60 %) used a single camera compared to only 36 % using stereo-BRUVS and four studies using a combination of both systems and one failing to specify the number of cameras used. It is likely that the prevalence of single-BRUVS is the result of their ease of use, affordability, and space constraints. Overall, the question of whether to use single- or stereo-BRUVS may come down to a number of factors (e.g. money, space, and time) but ultimately should be decided depending on whether there is a need for accurate length measurements to fulfil the proposed aims of the study.

Length measurements can be used to estimate biomass, gain an understanding of population and recuitment dynamics, and estimate fecundity (Ricker 1975). It can also be particularly useful in protected areas, where there is an expectation that fishing influences the size of fishes and that there will be a different size distribution in protected areas compared to unprotected areas (e.g. Watson et al. 2009). There were 65 studies that either used stereo-BRUVS or mentioned length as a variable for their study. Out of those 65, 38 % failed to present or use any of the estimated length data. Typically, studies which did present the length data were evaluating the use of length data under a range of circumstances, e.g. for precision (Merritt et al. 2011), with new technology (Letessier et al. 2015) or over different soak times (Misa et al. 2016), comparing lengths or biomass between protected and unprotected areas, or across different methods (e.g. Langlois et al. 2015), habitats (e.g. Fitzpatrick et al. 2012), or other impacts (e.g. seasons (McIlwain et al. 2011). Where length data were presented, it was most often measured using stereo-BRUVS (41 studies) and within those, 15 studies presented fork length data. There were 20 studies that estimated fish length using single cameras, most often

using a reference object or ruler within the field of view (e.g. known length of bait bag) to gauge fish length. Stereo-BRUVS (or single-BRUVS with the ability to accurately measure fishes [see below]) are necessary to answer specific questions where fish size is a critical variable, but where this information is not required and in 38 % of studies not even presented, I believe that sampling effort and the additional cameras required for stereo could be better spent on increasing replication.

One of the other benefits of using stereo-BRUVS over single-BRUVS is the ability to accurately measure maximum visibility. Such knowledge can be used to improve comparisons between studies where visibility is different. It can also be used to standardise the maximum distance up to which fishes are counted, but such standardisation requires a longer video processing time as once a distance threshold has been set, it would be necessary to ensure that only fishes within that distance are counted. Although a visibility measurement can be informative for any study, it was only mentioned in 36% of all studies (Figure 2.2A). While it is possible to restrict the distance within which fish are counted on single-BRUVS, it can be based on a subjective distance and commonly involves constricting the analysed field of view to quite small areas (e.g. only to the bait bag).

Studies have compared the accuracy of stereo-BRUVS vs. single-BRUVS and shown that the accuracy of fish length measurement using single-BRUVS deteriorated with distance from the measuring scale (± 2 m) and angle of view (>50°), while stereo gave a good estimate of length at a variety of angles and distances within 7 m (Harvey et al. 2002b). Some work has been done more recently to improve the accuracy of measurements taken from single-BRUVS, such as the development of mirrored surfaces allowing for a more exact positioning of fish in vertical set-ups, leading to more accurate measurements (Trobbiani and Venerus 2015). It is also possible to obtain accurate length data using on a known ratio of eye to head height predetermined for each fish species (Richardson et al. 2015). This method could be especially useful for targeted studies that are focusing on a few species only, as the proportion of eye size to head height has to be calculated for each fish species prior to BRUVS deployments. Developments such as these continue to improve BRUVS as a method and make it more accessible by providing ways to gain additional accurate information from single-BRUVS.

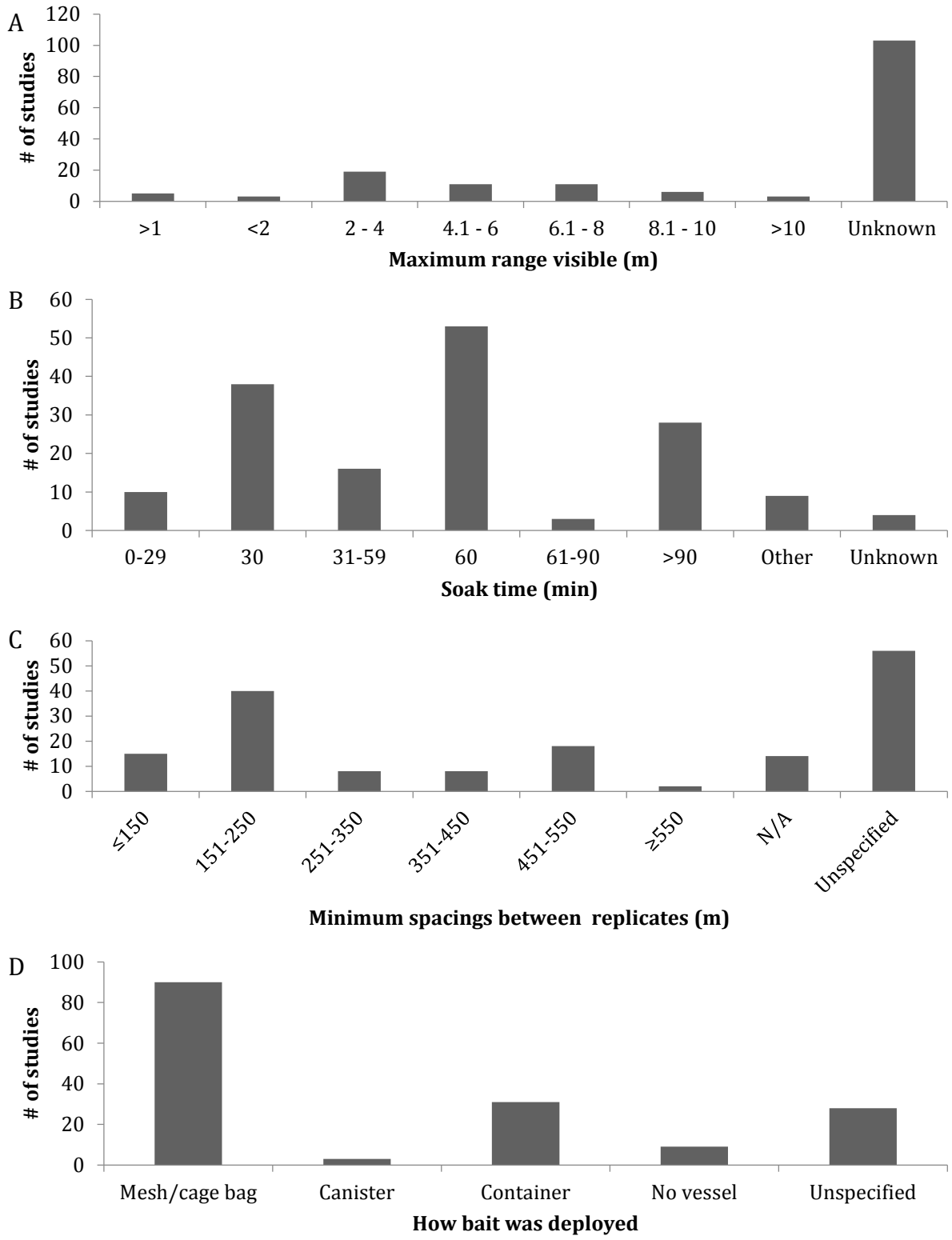

Figure 2.2: A) The maximum range visible from when viewing the BRUVS footage. B) The soak time for each of the 161 studies assessed that used a form of BRUVS. Studies in the 'Other' category included studies with multiple soak times and those which took periodic video clips over a larger time frame. C) The minimum space between replicates when being deployed within the field. N/A refers to studies which only had one replicate. D) Deployment method used for the bait. Containers were usually PVC pipe with holes to allow for plume dispersal, Canisters were used for timed releases often in deep-water habitats, and No vessel means no container was used and so the bait was physically attached to a section of the BRUVS.

Soak time differed greatly across studies (Figure 2.2B), with an apparent trimodal distribution. Peaks occurred around 30, 60, and >90 minutes. Few studies used times of less than 30 minutes, but 17 % of studies ran for more than 90 minutes, which often involved the use of additional power sources or extended batteries (e.g. Jamieson et al. 2006). Four studies (Gladstone et al. 2012; Santana-Garcon et al. 2014c; Harasti et al. 2015; Misa et al. 2016) specifically compared different soak times and found 60–90 minutes to be optimal for an estuarine environment (Gladstone et al. 2012), 120 minutes optimal for pelagic habitats (Santana-Garcon et al. 2014c), and 30 minutes was found to be sufficient in rocky reef habitats (Harasti et al. 2015). Misa et al. (2016) found shorter soak times (15 min) sufficient for snap-shot abundance estimates of Hawaiian bottomfish assemblages. Furthermore, some studies have included pilot studies of longer soak times to determine species and abundance accumulation curves, which justified a shorter time to be used subsequently in the main study (e.g. Unsworth et al. 2014).

Due to the variable and complex nature of currents (and hence bait plume modelling) and fish behaviour, the distance between replicates is often a contentious issue among BRUVS experts. There is considerable variation across the studies assessed in terms of the minimum distance between replicates (Figure 2.2C). Very few studies (only 2) had distances greater than 550 m, while 9 % (15 studies) had distances less than 150 m, with the minimum specified distance being 25 m (e.g. Colefax et al. 2016). Thirty-five percent of studies failed to mention the distance between replicates. There have been no studies investigating the impacts of replicate spacing on the assemblages observed. Distance between replicates is often used as a proxy for independence, with the hope that fish cannot swim or are not swimming between replicates. Such independence requirements avoid over-inflation of abundance by ensuring individuals are not double-counted on more than one replicate. The reasoning for different distances between BRUVS vary but are often based on hypothetical distances that fish may be able to swim between BRUVS within a given time frame (e.g. Ellis and DeMartini 1995). This can then lead to soak time becoming a factor for the appropriate minimum distance between replicates. However, based on the literature reviewed, I only found a weak positive correlation between soak time and distance between replicates (Figure 2.3A; Pearson correlation $p = 0.243$, 2-tailed probability = 0.020). It is likely that the

ideal distance between BRUVS will be variable and dependent on a number of factors including current speed and direction, influence of tides, time of day, and bait used. Fish behaviour is also likely to play a strong role in the assemblages observed and the recommended distance between replicates. Swimming speed, guild, schooling nature, shyness, interactions with other species, apparent hunger, and individual 'personality' (or behavioural syndrome, Sih et al. 2004) are aspects of behaviour that may affect fish assemblages and also whether fish are likely to be moving between replicates. There is, however, some evidence that even fish considered to be mobile might not move between replicates. For example, large more-mobile species such as smooth rays, *Dasyatis brevicaudata*, were only seen on a single replicate (out of 6) spaced 100 m apart (S Whitmarsh, unpublished data). While a greater distance between replicates is likely to reduce the chances of double-counting individuals, this may not always be possible. An example of this may be when investigating small isolated habitats (such as wrecks), when it may not be possible to space out replicates while still ensuring that the BRUVS are close enough to the target habitat. There is also a risk of spacing replicates too far apart and still expecting them to function as a replicate. Such an issue is more likely to occur in heterogeneous habitats. For example, if 6 replicates were spaced 500 m apart and arranged in a line (e.g. along a depth contour), the first and last replicate would be 3 km apart. This is far enough for other factors to have changed (e.g. wave exposure, current speed, wind direction, habitat). Without further studies investigating the impacts of spacing, I cannot recommend an optimal approach but I urge authors to carefully consider a distance that is logical based on the focus of the study, report the distance used, and explore the data collected to identify potential species that may have been double-counted.

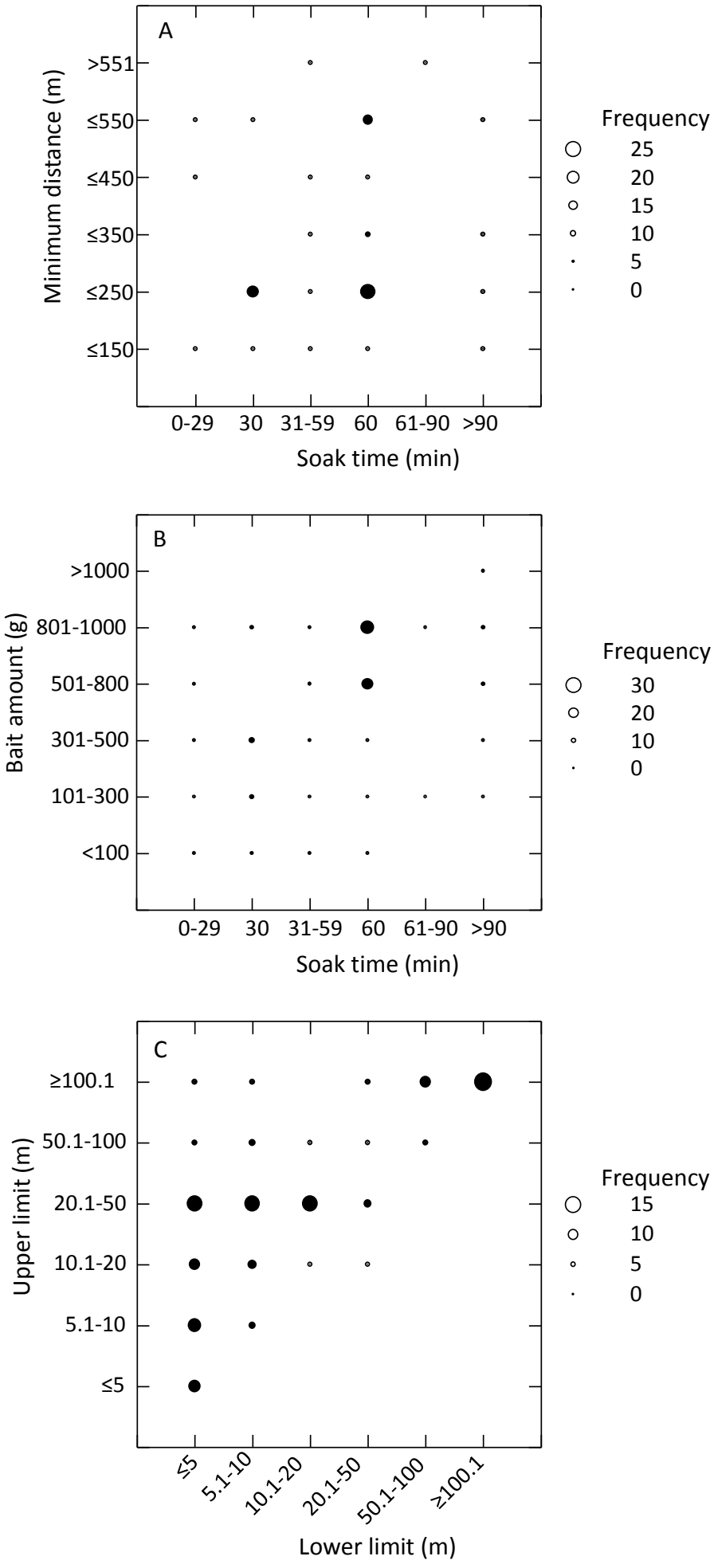

Figure 2.3: Soak time plotted against A) the minimum distance between replicates and B) the bait quantity, where the size of each dot represents the # of studies in each combination, as shown in legend. C) The lower and upper depth limits in which the BRUVS were deployed for the 161 studies that could be assessed. Excluding the 10 pelagic studies which were conducted mid-water and the 26 studies which failed to specify an upper (1), lower (4) or both limits (21).

Variables relating to the bait

The use of bait compared to unbaited systems has been specifically investigated by four studies (Harvey et al. 2007; Bernard and Götz 2012; Dorman et al. 2012; Hannah and Blume 2014). Bait increased the similarity between replicates providing better statistical power (Harvey et al. 2007; Bernard and Götz 2012; Dorman et al. 2012). Bait also increased the number of predatory and scavenging species, while not affecting the numbers of herbivorous and omnivorous fishes seen, and baited replicates were better able to detect changes between habitat types (Harvey et al. 2007; Bernard and Götz 2012; Dorman et al. 2012). Hannah and Blume (2014) showed that bait increased the abundance of deep-water demersal fishes by 47 % and lured the fish closer to the unit allowing for more accurate length measurements and species identification. Overall, these studies conclude that the benefits of using bait in marine environments appear to outweigh any perceived costs. There has been concern, however, about the ability of bait to attract fish from a large area potentially leading to inflated densities (Taylor et al. 2013). Variability in currents, winds, and turbidity across replicate deployments can lead to large changes in plume dispersal and significantly alter interpretations (Taylor et al. 2013). Studies rarely considered this factor and few studies implemented current measuring devices, such as current meters or drogues (Taylor et al. 2013).

Bait choice is often a well-discussed issue for all methods that require its use (e.g. longline fishing; Lokkeborg et al. 2014) and has also been investigated for BRUVS, with a number of studies specifically looking at the effects of bait type. Dorman et al. (2012) and Wraith et al. (2013) each investigated three different bait types. Dorman et al. (2012) compared sardines, cat food, and a vegetable mix with unbaited controls and found similar assemblages between these three bait types. Cat food, however, depleted rapidly and did not always last for the 60-minute deployment time. The vegetable mix was costlier, harder to use (due to having to mix the bait) and caused obscuration of the field of view, and consequently was not recommended by the authors. Wraith et al. (2013) compared amongst three marine baits, chopped sardine, chopped abalone viscera and crushed urchin, and found urchin to record significantly less fish abundance and species richness, and increased time of first arrival compared to the other two bait types. The bait type used also affected the feeding guilds observed with sardines attracting more generalist carnivores, zooplanktivores, and macroinvertebrate

carnivores, and being potentially more consistent at attracting herbivores than the other two types. Overall, the authors recommended using oily fish such as sardines. Walsh et al. (in press) also investigated three bait types, sardines, mussels and a locally available alternative to sardines (Australian salmon). Walsh et al. (in press) found similar results between the two fish species, while the mussels attracted more omnivorous species, but had a lower overall species diversity. Based on our literature search, the most common bait type was the Australian sardine, *Sardinops sagax*, although other species or sub-species of sardines were also commonly used. Sardines accounted for over 56 % of the bait types used (Figure 2.4) and were often also included as part of the mixed bait types and in some of the vegetable mixes used. Bait type was always marine-based (with the vegetable mixes containing fish oils), with the exception of chicken, which was used in specific studies to attract *Nautilis* spp., pig carcasses for attraction in the deep sea, silverside meat, and dough ('Other' category, Figure 2.4; Appendix 1,Table 7.1). The prevalence of sardines used in the collective literature appears to be supported by the above studies that compared bait types. Sardines are often said to be good as bait due to their oiliness, low cost, ready accessibility, and persistence within the bait bag (Dorman et al. 2012; Wraith et al. 2013). Although sardines can be easily accessed in some temperate regions such as Chile or Australia, in areas where sardines may be difficult to acquire such as in the tropics, I recommend using a similar oily fish that is readily accessible in that region, as per Walsh et al. (in press).

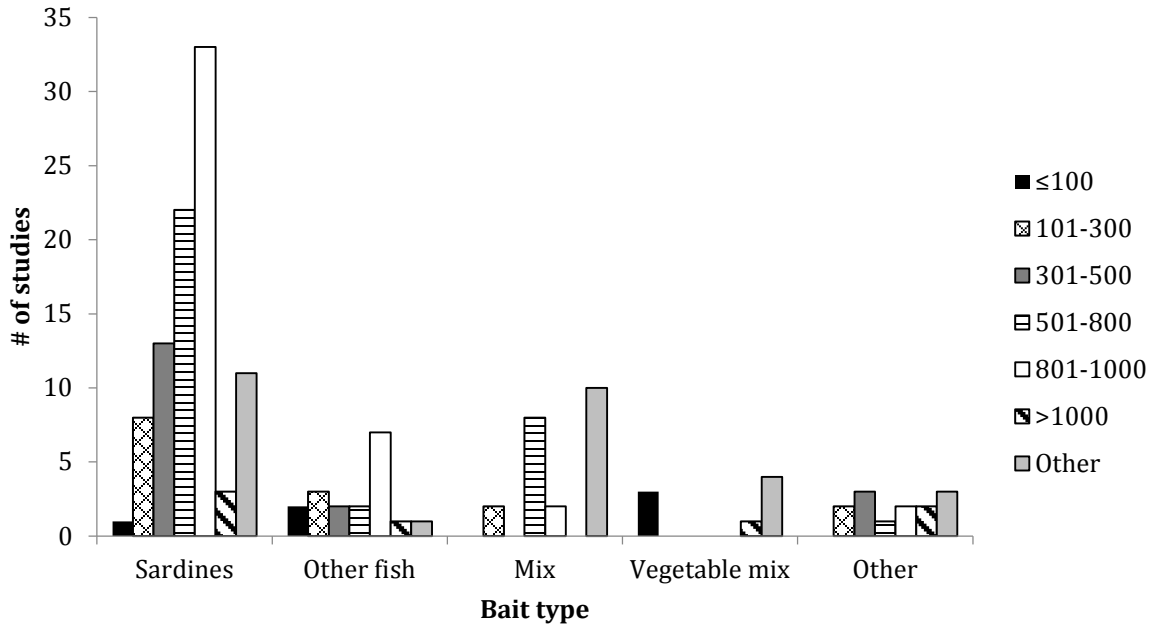

Figure 2.4: Bait type and quantity (g) for 161 studies that used a form of BRUVS. 'Vegetable mix' was composed of varying amounts of falafel mixed with fish oils. 'Mix' bait was composed of multiple components, usually fish and squid. The 'Other' bait type category includes baits such as commercial fish feeds and chicken. 'Sardines' were usually *Sardinops sagax* or *S. neopilchardus*. The unknown bait types are excluded. The 'Other' quantity category included those studies with variable amounts of bait, unknown quantities and those studies which only specified a whole number of fish.

Various quantities of bait have been used when deploying baited video ranging from 50 g to over 2 kg (Figure 2.4). The most common bait quantity was within the 801–1000 g category (with 27 % of studies; Figure 2.4), with a majority of these using approximately 1000 g (Appendix 1,Table 7.1); 501–800 g was the next most popular category with 20 % of studies using a quantity within this range (most commonly 800 g; Appendix 1,Table 7.1). Bait quantity was thus more varied across studies than the type or preparation method used (Figure 2.4). Only one study tested whether varying the quantity of bait affected the observed fish assemblages (Hardinge et al. 2013). There were no significant differences in fish diversity between 200, 1000, or 2000 g of bait but there were some individual species differences, with the moray eel, *Gymnothorax woodwardi*, being significantly more abundant with 2000 g of bait than with 200 g (Hardinge et al. 2013). The lack of differences in fish diversity may have been caused by the limited bait depletion (i.e. low bait predation) leading to fish being equally attracted to the baited video throughout the deployment regardless of the quantity of bait used (Harvey et al. 2007). There is, however, a need to spatially replicate the study in areas likely to have high bait depletion, such as those with high fish abundance or areas with different water temperatures that may affect the foraging rates of fishes. There was a variable positive correlation between bait quantity and soak time (Figure 2.3B; Pearson correlation $p = 0.221$, 2-tailed probability = 0.015), which only suggests a weak trend for the deployments with longer soak times (>90 min) to also use more bait (>1000 g). Overall, there appeared to be little consideration of the appropriate quantity of bait to choose and this remains an area for improvement and future research.

Despite the copious literature available on BRUVS, there appear to be no studies investigating the effects of the preparation method for the bait. Many authors assume that crushing the bait (particularly sardines) enables a more even plume dispersal (e.g. Watson et al. 2009), but it may be worthwhile for a future study to test this hypothesis. From the literature analysed, bait was prepared in a variety of ways, with crushing being the most common method (55 % of studies). Chopped and whole-bait preparations were less common at 15 % and 12 %, respectively, while 16 % of studies did not specify the way the bait was prepared (Table 2.1).

Deployment method for the bait also varied across studies (Figure 2.2D) with a majority of studies (55 %) using some form of mesh bag in which bait was likely to be somewhat accessible to taxa for feeding. The use of a perforated container (usually PVC) was the next most common method (19 %), which served to disperse the bait plume but restricted the access to taxa for feeding on the bait. Some studies (2 %) in the deep-water habitat choose a timed-release method using canisters to enable fresh bait to be released periodically. Six percent of studies used no form of vessel for bait deployment. The remaining 18 % of studies did not specify the bait deployment method. It is possible that the delivery method for the bait may influence assemblages observed as the ability to physically feed on the bait (more likely with a mesh bag) may lengthen the amount of time individuals remain around the BRUVS and hence inflate MaxN, or attract/deter other species. This area of study has not been investigated, but warrants future investigation.

Variables relating to the deployment

The depths at which baited video are deployed varied from very shallow (0.5 m) to deep-water (8074 m). Very shallow baited video studies were uncommon, with only 14 % of studies having the shallowest deployment depth class of less than 10 m (Figure 2.3C). Fifty-one percent of studies did not extend past 50 m in depth. Only nine studies sampled exclusively within the shallowest range (≤5 m), while there were 14 studies that sampled exclusively in the deepest depth range (>100 m). This shows a wide range of use for BRUVS and its applicability to a broad range of depths. The studies assessed had a narrow variation in depths sampled with 42 % sampling within a range of 20 m or less (Figure 2.5A) compared to 15 % spanning a range greater than 100 m. Failure to specify either a lower or upper depth limit resulted in 29 studies (18 %) having an unknown depth range.

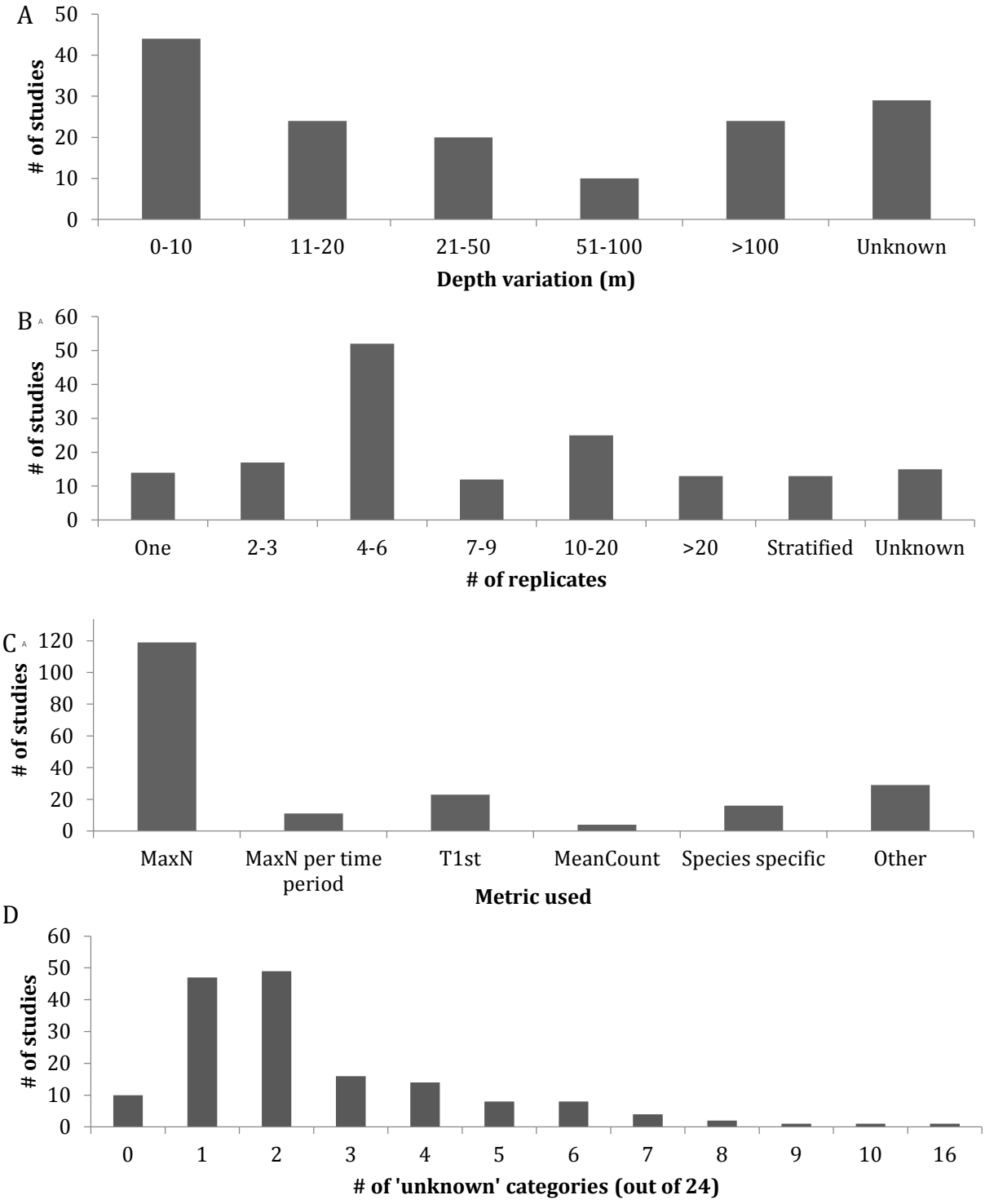

Figure 2.5: A) The variation (range) in metres between the lower and upper depths of the demersal BRUVS deployments for the 161 studies assessed (excluding the pelagic studies, as these did not have a normal depth variation; see pelagic BRUVS section). B) The number of replicates taken in each of the 161 studies assessed. Where a range was given, the upper value was used to assign the category here. Stratified indicates a sampling design which spans across a large area that conducted enough replicates to get good spatial coverage across particular strata such as habitat or depth (e.g. Moore et al. 2010). C) The metric used to assess the video footage from the 161 studies assessed. Studies were counted in more than one category where more than a single metric was used. Species-specific metrics included identifying individuals through the use of colouration or patterns. T1st is the time of first arrival for each species. 'Other' included metrics involving assessing behaviour of individuals, bait loss, habitat coverage, and abundance metrics other than *MaxN*. D) The number of categories (out of the 24 reviewed here) in which the methodology of that study was 'unknown' (i.e. not stated explicitly; *N* = 161 studies).

Variables relating to sampling design and analysis

The number of replicates used when deploying BRUVS was variable (Figure 2.5B). Thirty-two percent of the studies reviewed used four to six replicates at each location. I was unable to determine the number of replicates used in 9 % of the reviewed studies, while 9 % were unreplicated (all of which were in deep-water or 'other' habitats). These studies are likely to be un-replicated due to the large cost involved in sampling at such great depths. Some studies (8 %) also chose a stratified sampling design. This design type involves the deployment of BRUVS across a large area while ensuring that replicates are representative of all strata, e.g., habitats or depths, represented within that area (e.g. Moore et al. 2010).

Images from BRUVS videos can be measured in a variety of ways, depending on the aims of the study. The most common metric recorded was *MaxN* (the maximum number of a particular species seen in any one video frame across the duration of the video record). *MaxN* was used in some form, either over a set time period or across the whole video, in 81 % of the reviewed studies. *MaxN* can be used in conjunction with other metrics such as time of first arrival ($T1^{st}$) or time first fed (Figure 2.5C). However, this was done in less than 20 % of cases. *MaxN* can be modified slightly to include the maximum number seen over a set time period, e.g. 30 s, or can be estimated at specific intervals, e.g. every 5 min. Some studies (10 %) focused upon species-specific metrics such as identifying individual sharks (e.g. Bond et al. 2012; Ryan et al. 2015) or *Nautilis* spp. based on skin or shell colouration patterns (e.g. Dunstan et al. 2011). The 'Other' category includes observation of specific behaviour (e.g. Bailey et al. 2007), total species counts (e.g. Craig et al. 2011), and residence time (i.e. how long animals stayed at the bait; e.g. Smale et al. 2007) metrics.

MaxN can be used to assess the relative abundance of organisms. It is often considered a conservative estimate as more individual organisms may be present around the BRUVS but remain uncounted because they do not appear in the field of view at the same time. This relative abundance measure can be used to assess and compare spatio-temporal differences in aquatic assemblages. Saturation may, however, occur when a high number of individuals obscure the field of view to the point that additional individuals cannot be seen (Schobernd et al. 2014; Stobart et al. 2015). Such

saturation can result in the inability to detect differences between locations when fish abundance is high (Stobart et al. 2015) and results in *MaxN* being non-linearly related to true abundance (Schobernd et al. 2014). Recently, *MeanCount* (used in 2 % of studies; Figure 2.5C) has been suggested as an alternative to *MaxN* that can be linearly related to true abundance (Schobernd et al. 2014). *MeanCount* uses either systematically or randomly selected individual frames from across the video which are subsequently counted and then the mean is calculated. As the entirety of the video is not viewed, *MeanCount* has a tendency to over-inflate zero observations and is less precise than *MaxN* (Campbell et al. 2015; Stobart et al. 2015).

T1st is a measure of how fast species are first observed in the field of view. In some cases, there has been a negative correlation shown between *T1st* and *MaxN*, meaning that if a species where to arrive quickly to the bait, the species is often highly abundant (Stobart et al. 2015). *T1st* can also be used to infer the distance a species may have travelled to get to the BRUVS. However, as *T1st* is influenced by both the distance the fishes are away from the BRUVS as well as the behavioural response to the bait used (i.e. how attracted they are to the bait) which can vary between species, it can be difficult to disentangle what *T1st* is really showing.

The main types of software that were used to obtain the above metrics could be classified into 5 groups (Figure 2.6A): specialised software for viewing BRUVS videos such as EventMeasure (www.seagis.com.au; used in 34 % of studies) and the AIMS BRUVS software (no longer available; 12 %), generic media players or photo viewers (e.g. VLC, Adobe Photoshop; 8 %), software designed for measuring objects within photos (e.g. Visual Measurement System; 8 %), and other programs for further specific purposes (e.g. Hotspotter for identifying *Nautilis* spp.; 3 %). PhotoMeasure and EventMeasure were combined as a single category as PhotoMeasure has been superseded by the newer versions of EventMeasure. The use of a specialised software program designed for the viewing of BRUVS videos allows for considerable time-saving when processing videos. A high proportion of studies did not specify the software used for video analysis (46%).

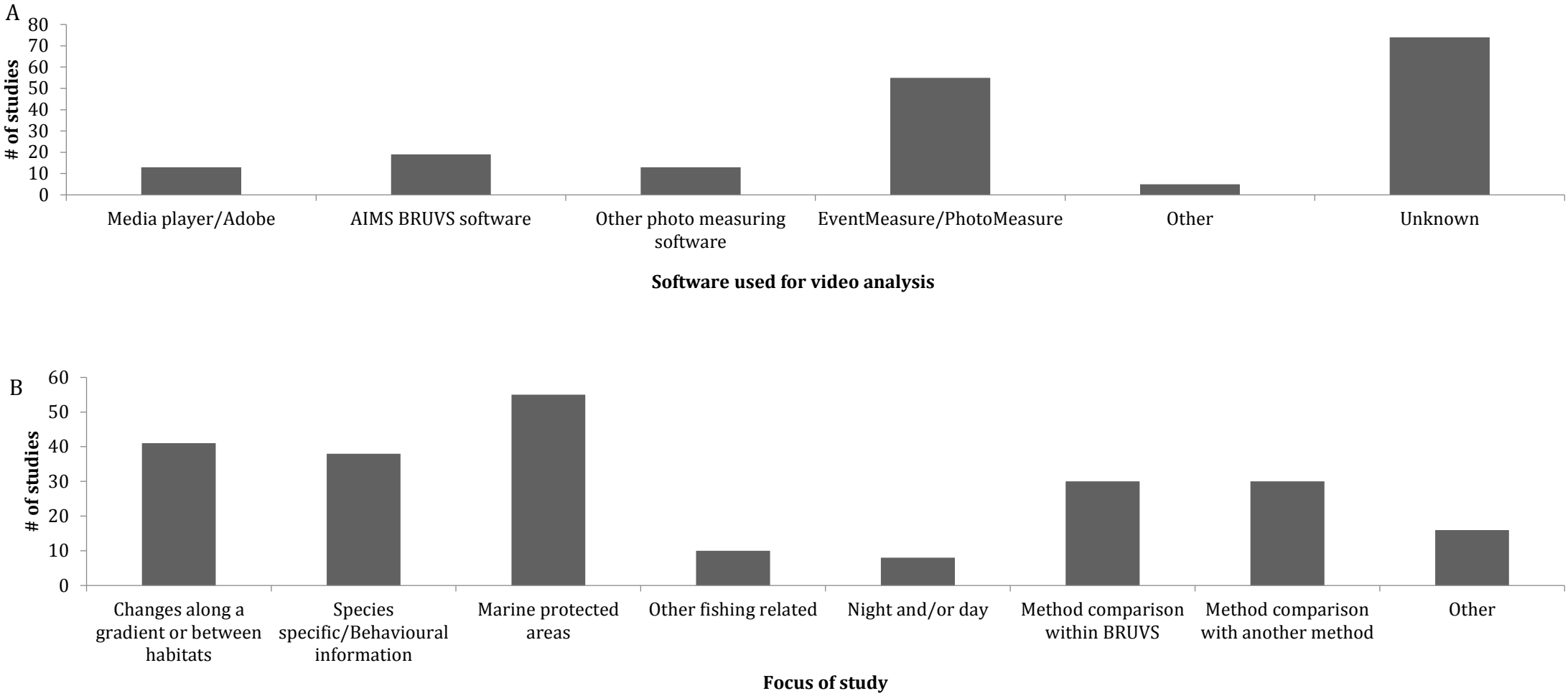

Figure 2.6: A) The software used to assess the videos. "Other photo measuring software' includes programs designed for measuring objects within photos (excluding PhotoMeasure which was combined with EventMeasure due to the dual function of EventMeasure in recent versions of the software) such as Visual Measurement System. The 'Other' category includes programs for more specific purposes such as Hotspotter which is used to identifying *Nautilis* spp. B) The focus of the study for each of the 161 studies analysed. Studies were counted in more than one column where they covered more than a single focus. The 'Other' category includes those which did not fit in any other category including artificial vs. natural reef assessments and other sorts of impacts.

Purpose of studies: What is BRUVS used for?

BRUVS has been used for answering a wide variety of scientific questions (Figure 2.6B; Appendix 1, Table 7.2). The most frequent reason (34 %) for deploying BRUVS was in relation to assessing the effects of marine protected areas (MPAs; e.g. Bornt et al. 2015; Coleman et al. 2015). The large number of studies using BRUVS to study MPAs is likely related to the non-destructive and non-extractive nature of BRUVS, making it a suitable alternative to more traditional methods. Studies that looked at particular species or behaviours (24 %; e.g. Denny et al. 2004; Gutteridge et al. 2011) and those which assessed changes in fish assemblages along a gradient or between habitats (25 %; e.g. Gomelyuk 2009; Langlois et al. 2012b) were the second- and third-most common study aims. Method comparisons both within BRUVS (e.g. different soak times; Gladstone et al. 2012) and between BRUVS and other methods (e.g. BRUVS vs. longlines; Brooks et al. 2011), were also popular with 19 % of studies choosing to focus upon within-BRUVS method comparisons and 18 % on comparisons with other methods. This perhaps reflects a view in many minds that BRUVS is still developing and their use needs justification. There were also studies that investigated day-to-day (Birt et al. 2012) or day-to-night (e.g. Svane et al. 2008) variation and variability in night-time (e.g. Fitzpatrick et al. 2013) assemblages, which accounted for 5 % of the total.

A majority of studies using BRUVS had a particular focus on fish assemblages, these being the nektonic organisms that most frequently come to the bait. However, a number of other organisms are also attracted to the baited units or can be seen by happenstance, particularly cephalopods and crustaceans, along with other mobile invertebrates, cetaceans, pinnipeds and aquatic birds (e.g. Whitmarsh et al. 2014). In my review, there were only 11 % of studies which counted all nektonic species seen on their videos, compared to 64 % that assessed teleost assemblages, and 60 % that assessed Chondrichthyes (Figure 2.7A). An additional 7 % of studies assessed a single or multiple specific fish species. Only six of the 161 studies analysed in this review had a focus on non-fish species (two on the cephalopod *Nautilis* (Dunstan et al. 2011; Barord et al. 2014), two on crustaceans (stone crabs in the Lithodidae family, Collins et al. 2002; and other decapod crustaceans, Jamieson et al. 2009), one on reptiles (three species of sea snakes, the olive sea snake, *Aipysurus laevis*, the spine-bellied sea snake, *Lapemis curtus*, and the ornate sea snake, *Hydrophis ocellatus*, Udyawer et al. 2014), and one on

buccinid gastropods (Aguzzi et al. 2012), with three out of these six being from deep-sea habitats. Aside from the traditionally teleost-focussed studies, in recent years studies focussing exclusively on chondrichthyans have begun to be published such as White et al. (2013), Rizzari et al. (2014) and Ryan et al. (2015).

I was able to determine the percentage of taxa putatively identified to species level in 65 % of studies (Figure 2.7B). Ten percent of all studies were able to identify all taxa to species level while only 2 % of studies had greater than 30 % unable to be identified to species level and none greater than 40 %. Generally, species that could not be identified to species level were small, cryptic or rare species, which is likely to result in a bias against such species. Visibility may also affect how well species are able to be identified. The type of organism targeted for the study can also affect rates of identification. For example, fish species can generally be reliably identified from video footage, but other smaller mobile animals e.g. crustaceans, echinoderms, and cephalopods can be more difficult to identify. Despite this, since these types of animals are generally less well-studied, any information gathered about them can be useful.

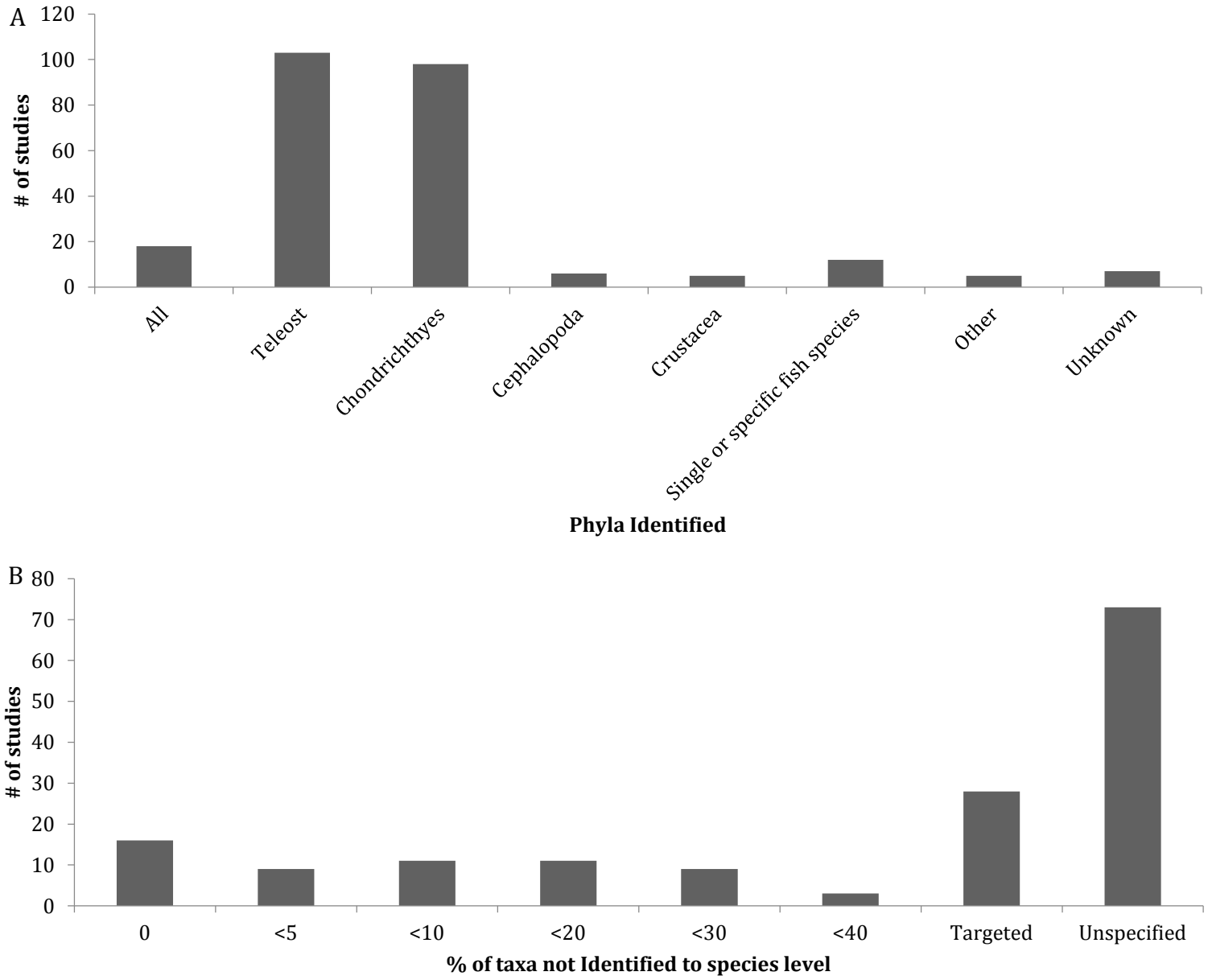

Figure 2.7**:** A) The phyla that were identified from each of the 161 studies in this review. All includes those studies which counted any mobile taxa able to be consistently recognised. Crustacea were most commonly decapods. The Other category included sea snakes, echinoderms, and one study in which no biota were identified (only the habitat type). B) The percentage of taxa unable to be identified down the species level for each study assessed (*n* = 160). The targeted category specifies those studies which focused on only a single or few specific species. Not shown is one study which assessed habitat only and as such this variable was not applicable.

A possibility for assisting newcomers to BRUVS and improving the ease of identification for existing persons could be for more routine and shared image archives. Mentions of image archives are not prominent within the published literature, but archives are likely to exist for many BRUVS teams and the sharing and concatenation of such archives would assist in ensuring the accuracy of identified species.

I also categorised each study as either standard or novel to highlight any unusual uses of BRUVS. For the purpose of this literature review, I define the standard use of BRUVS as follows (anything that did not fit into this category was considered 'novel'):

> *Daytime deployment on the seafloor, in subtidal, shallow (<120 m deep) habitat, single or stereo (2 cameras maximum) camera(s) facing towards the bait bag either horizontal or downwards, a single bait arm with a mesh bait bag attached, single use bait, and with a video length of no more than 90 min.*

Out of the 161 studies, 110 (68 %) were considered standard and 51 (32 %) novel. The main novel developments for BRUVS were the extensions into pelagic habitats, modification for deep-water deployments, and night-time uses.

Overall, only 6 % of the studies analysed had detailed method sections that stated all of the 24 main variables in this literature review. However, 60 % of the studies were only missing values for 2 or less of these categories (Figure 2.5D; Table 2.1). The most commonly unreported variables included the maximum visible range (reported in only 46 % of studies), the software used for analysis (54 %), the number of species able to be identified to species level (55 %) and the distance between replicates (65 %; Table 2.1).

Novel method development of BRUVS

Pelagic deployments

The use of BRUVS in the pelagic environment is a relatively recent development, with only two studies published using this method up to and including 2012 (Heagney et al. 2007; Robbins et al. 2011). Since 2012, it has increased in popularity with an additional nine studies using BRUVS in the pelagic environment (Letessier et al. 2013, Santana-Garcon et al. 2014a,b,c,d, Anderson and Santana-Garcon 2015, Bouchet and Meeuwig 2015, Rees et al. 2015, Scott et al. 2015). This method involves changing the focus of BRUVS from the traditional demersal setting to suspending the unit within the

water column to better sample pelagic fishes. The pelagic BRUVS are horizontally set up and usually allowed to float at a specific depth below the surface (e.g. 10 m; Heagney et al. 2007), as opposed to resting on or near the seafloor in standard use, although some studies set a specific distance above the substrate (e.g. 10 m above the bottom; Santana-Garcon et al. 2014a). Other major modifications include the use of additional floats, ropes, and weights to allow for a stable mid-water deployment. Recently, developments have been made to allow for a drifting pelagic set-up (Bouchet and Meeuwig 2015) that can cover broad stretches of ocean space that in that study had an average transect length of 4.9 km during a 165 min deployment.

Bait plume dispersal has been highlighted as a major factor that could affect the fish assemblages observed via pelagic BRUVS in particular due to the sparse and heterogeneous nature of fish assemblages within this environment (Heagney et al. 2007). Heagney et al. (2007) recommended the addition of a current meter to assist in determining the likely plume dispersal. Taylor et al. (2013) also recommended this or similar current-measuring devices to be used for benthic deployments. Furthermore, an increased soak time (Letessier et al. 2013) and replication (minimum of 8 in tropical environments) is needed to account for the highly heterogeneous distribution of pelagic species (Santana-Garcon et al. 2014c). There has also been evidence for additional attractants to be used alongside traditional bait in the pelagic environment, such as those based on sound (recordings of bait fish) and sight (metallic reflectors; Rees et al. 2015). Rees et al. (2015) compared these different attractant methods and found that the combination of all three attractants was more effective at attracting consistent numbers of fish than the individual components alone.

Six of the 11 studies using pelagic BRUVS (55 %) were focussed on developing and assessing the validity of the method, while, of the others, two looked at behaviour of a particular species (Robbins et al. 2011, Santana-Garcon et al. 2014b), one looked at the the impacts of artificial reefs (Scott et al. 2015), another used the data specifically to demonstrate a novel statistical analysis technique (Anderson and Santana-Garcon 2015) and one focussed upon using BRUVS to determine the effects of MPAs on pelagic species (Santana-Garcon et al. 2014d).

Deep-water deployments

While the use of still photography in the deep sea has occurred since the 1960s (Gage and Tyler 1991), the use of BRUVS in deep-water habitats has only begun in the last 14 years, with the first published articles appearing in 2002 (e.g. Collins et al. 2002, Yau et al. 2002). There are numerous challenges to using BRUVS within the deep sea that are not present in shallower environments, such as increased pressure resulting in the need for sturdier housing for the cameras, reduced light resulting in the need for external lighting sources (and consequently powered by batteries), reduced diversity and abundance resulting in a need for longer soak time and potentially more replication being necessary, which is also compounded by the long descent time from the surface. There are also depths where ropes and surface floats become impractical leading to the need for remote release mechanisms to allow gear recovery. Some deep-water studies also used larger baits, such as pig carcasses, and leave them out for extended periods (days-months; Anderson and Bell 2014). The additional cost for the these features along with increased general field costs associated with working in deep-water habitats means that sampling becomes very expensive, which could be a reason why there is little to no replication with deep-water studies (70 % with none or unknown) and also why 60 % have a soak time longer than 90 minutes (e.g. Bailey et al. 2007).

Night-time deployments

The optimisation of BRUVS for use specifically at night has begun recently with studies such as Fitzpatrick et al. (2013), although the use of BRUVS in deep-water habitats has occurred for a longer period and has some of the same challenges (e.g. use of lights). To observe the impact on fish assemblages, Fitzpatrick et al. (2013) examined three different light colours (red, white, and blue) in a range of habitats both inside and outside protected areas. They found that each light affected fish assemblages differently and suggested that this was most likely due to differences in fish behaviour or physiology towards different light sources. The wavelength of red light (620–630 nm), like that of infrared (<700 nm), is below the spectrum that fish are sensitive to but is rapidly attenuated in the water column compared to white and blue light, which can be seen for a greater distance but may attract or disturb some species. Fitzpatrick et al. (2013) found that red light sampled the highest abundance of fish of the three light colours and was particularly good at sampling non-commercial species; however, it

illuminated the smallest area due to the attenuation of red light in seawater. White and blue light sampled similar fish assemblages but had higher abundances of some commercially-targeted species such as snapper, *Chrysophrys auratus*, compared to red, and also illuminated a greater area. The authors recommended further studies into the impacts of light colour on fish assemblages. These results are somewhat different from those found from another study by Harvey et al. (2012b), where white light sampled a greater number of individual fish compared to red but was not able to distinguish between six different benthic habitat types as well as the red light could.

Another study used infrared light to assess nocturnal fish assemblages and compared these results to those from UVC (Bassett and Montgomery 2011). A higher abundance of olfactory specialists, species which rely heavily on sense of smell (e.g. yellow moray eels, northern conger eels, southern bastard cod) were observed from infrared BRUVS compared to UVC, and these species consistently arrived at the bait quicker than non-olfactory specialists. Studies have also used BRUVS to compare assemblages between day and night (e.g. Svane et al. 2008; Svane and Barnett 2008), and found that BRUVS can effectively discern changes between day-time and night-time behaviours, such as an increased consumption of bait at night.

Other innovations

Other novel uses of BRUVS include the development of 'miniBRUVS' for use in rockpool environments (Harasti et al. 2014), which is also the only intertidal use of BRUVS that has occurred so far. This development was successfully used to assess the abundance and distribution of a threatened and otherwise hard-to-study rockpool-specialist fish, the black rock-cod, *Epinephelus daemelii*.

Optimisation vs. standardisation: developing a protocol for reporting methods

Optimisation is the trialling of different variables to ensure the best use of resources (time, effort and money) to deliver benefits (e.g. detect increased abundance or diversity or maximise ability to discriminate between factors). There have been several studies that have focussed on the optimisation of BRUVS (e.g. Gladstone et al. 2012; Harasti et al. 2015), with all studies falling into the method development within BRUVS (19 %) considered as working towards optimisation. However, few studies have

compared method optimisation between locations or habitats. Different areas even within similar habitat types, such as temperate reefs, still seem to display different values for each optimal scenario, such as seen in a study by Harasti et al. (2015), which showed that, in Eastern Australia, the *MaxN* for many reef species occurred within 12.5 min making a soak time of 30 min quite practical. In contrast, *MaxN* in South Australia took longer to be reached (30–40 min; Whitmarsh et al. unpublished data) meaning that a soak time of 60 min is more applicable. Both studies used similar methods with the exception that Harasti et al. (2015) used slightly more bait at 1000 g compared to 800 g. Generally, I urge caution when assuming optimal scenarios still apply in different areas or habitats and advise authors to conduct their own pilot studies if possible.

In general, it is easy to deviate from the 'standard' use of BRUVS to tailor to specific objectives such as studies of *Nautilis* sp. using chicken as bait with a soak time of 12 h (e.g. Barord et al. 2014) or modifying the system to work in small rock-pool environments (e.g. Harasti et al. 2014). There is, however, no consensus about whether it is better to tailor the method to each specific scenario being tested or to strive towards standardisation to better enable valid comparisons across studies. The goal of standardisation of BRUVS as a method may be worthwhile but is ultimately, I believe, unachievable and may in fact negatively impact novel developments and methodological breakthroughs. Currently, if comparisons amongst studies are attempted, some authors fail to specify enough details in their paper's methods section for the differences to be accurately accounted for. I suggest a standard protocol of what information to be included within the literature (Table 2.1), rather than a standard protocol for use.

Future directions

I have identified some gaps in the current knowledge base such as the effects of distance between replicates, bait amount, preparation, and deployment method, continued lack of studies accounting for plume effects and using current meters, further impacts of light colours on nocturnal or deep fish assemblages, appropriate soak times under a range of habitats and conditions, and the appropriate numbers of replicates to account for the variable nature of fish assemblages.

One key aspect of method deployment not often covered in the literature is the effect of bait preparation on fish assemblages observed. Although it is unlikely that large differences in assemblages would be observed from using chopped vs. crushed sardines, it is reasonable to assume that some differences may result from a comparison of whole vs. crushed sardines, if there is any increased areal coverage of plume dispersal coming from crushed bait.

Future research using BRUVS could focus on gaining additional data from the video metrics in addition to *MaxN.* For example, behavioural data could enhance our knowledge of how species interact with themselves, other species, and bait, while oceanographic data (e.g. temperature, salinity) through attachment of sensors to the unit would provide a way to investigate the influence that these factors have on fish and other nekton. A more formal description of habitat features seen from the images and better use of fish arrival or departure times and hence length of stays could also increase our knowledge of fish assemblages. There is also scope to increase the use of BRUVS outside of reef areas, with some studies showing that it is an effective method for soft-bottom (Gladstone et al. 2012), seagrass (Whitmarsh et al. 2014), pelagic (Rees et al. 2015), and deep-water (D'Onghia et al. 2015a, b) environments. The other major area for potential growth in BRUVS is to focus on other nektonic species rather than fishes. Combinations of different unit designs and bait may enable BRUVS to be tailored to any number of mobile species including cephalopods, marine birds, marine mammals, marine reptiles, crustaceans, and other benthic mega-invertebrates (e.g. sea stars, sea cucumbers and large gastropods).

Conclusion

Overall, BRUVS is a widely-used method for assessing nektonic assemblages and their behaviour. This review shows the robust and flexible nature of BRUVS and its widely applicable uses from cataloguing the behaviour of particular species to broader changes in mobile communities within a wide variety of depths and habitats. Its use over the last two decades has led to further developments to the method, including the introduction of stereo-BRUVS, pelagic BRUVS, and night-time BRUVS. Several studies have also focused on optimising or standardising the use of BRUVS. To enable more accurate comparisons across studies while still allowing novel and specialised use, I

recommend a protocol that authors can follow to allow sufficient detail to be included in methods sections.

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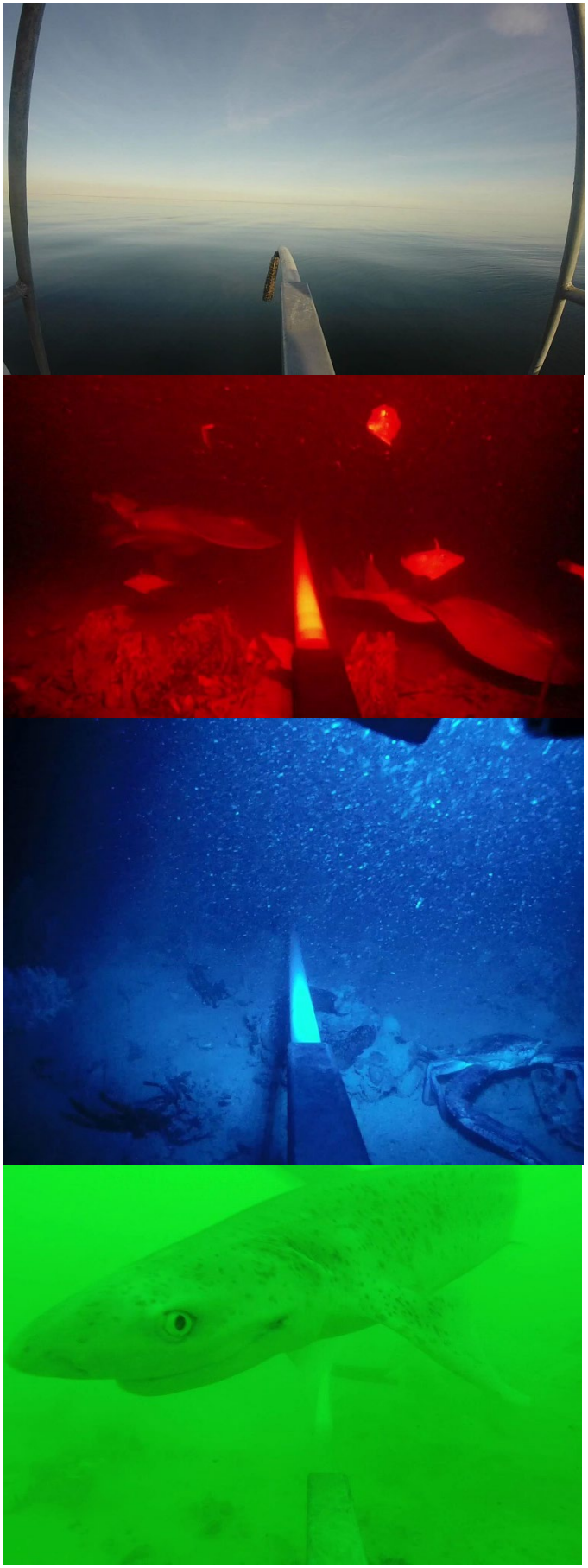

Chapter 3

Multi-scale analysis of temporal changes in fish assemblages

Sasha K. Whitmarsh

Peter G. Fairweather

Charlie Huveneers

Abstract

Without adequate knowledge of how fish assemblages change over time, assessing the diversity and abundance of species can be misleading and without context. This study aims to assess how fish assemblages change over multiple time-scales ranging from diel to yearly variations across a range of habitats within South Australia including those often understudied, i.e. soft sediment and seagrass. Fish assemblages at night were observed using red and blue light colours to determine which colour is most appropriate. Fish assemblages were sampled using standardised Baited Remote Underwater Video Stations (BRUVS) and the results from 200 replicate deployments across five sampling periods spanning three years (2013–2016) were analysed. The results showed no difference in assemblages observed at night with either red or blue light, but distinct and significant differences for daytime versus nighttime assemblages, and varying results by site according to season and year sampled. These results highlight the need for studies to consider temporal variation in their design to account for this dynamic nature of assemblages. Sampling should be conducted across seasons and at night to get a more comprehensive assessment of abundance and diversity of a given area.

Introduction

There is an increasing need to understand and quantify fish assemblages to ensure robust conservation and management practices (Gerber 2005; Stuart-Smith et al. 2017). However, without knowledge of how assemblages change over different time scales, they cannot be accurately and comprehensively assessed. Knowing the distribution, abundance, and movement patterns of marine species can help inform fisheries management practices and the management of protected areas. Knowledge of temporal variation also allows for a greater understanding of the dynamic nature of such communities. For example, temporal variation in fish abundance drives increases in seasonal effort in many fisheries worldwide (Pet-Soede et al. 2001) and variations in total allowable catch between years (Karagiannakos 1996). With an increased understanding of how assemblages change over different timescales, particularly between seasons and years, and under inherent environmental variability, scientists are better able to understand changes observed from monitoring programs. These programs may run over short periods and provide snapshots of abundance and

diversity or be ongoing and continuous long-term studies such as those proposed and executed in many marine protected areas (MPAs). Many such monitoring programs for MPAs conduct surveys over varying timescales from months (e.g. Edgar and Barrett 1997) to years (e.g. Stuart-Smith et al. 2017) apart and are usually conducted during the daytime and at particular seasons, where overlap into adjacent seasons may occur due to inclement weather or other logistical issues. Thus, understanding the temporal variation in the assemblages being studied can be key to correctly interpreting changes which may have occurred between surveys.

Though many studies have investigated changes of fish assemblages across different locations (e.g. Bell and Westoby 1986; Babcock et al. 1999; Willis et al. 2000; McLean et al. 2016), fewer have focused on temporal variations (but see Young 1981; Mazumder et al. 2005; Willis et al. 2006). Temporal changes can occur over varying time scales, from short-term diel variations (i.e. changes within a day such as between day and night; Harvey et al. 2012a; Myers et al. 2016) to longer-term seasonal or yearly changes of abundance and diversity (Lehodey et al. 2006; Olsson et al. 2012). Although variation in fish assemblages within and between days is common (Birt et al. 2012; Aguzzi et al. 2013), a lack of diel variation has also been suggested in some habitats (Willis et al. 2006). A complete understanding of diel variation in fish assemblages requires fishes to also be sampled at night (Helfman 1986; Bassett and Montgomery 2011). Fish assemblages at night are generally much less studied than daytime assemblages, with less than 5 % of studies (out of 161) that used baited video reporting nighttime sampling (Whitmarsh et al. 2017). The habitat use by different taxa can vary between day and night (Johnson and Covich 2000; Azzurro et al. 2007), with some species performing critical ecosystem functions at night. It is therefore important to understand nighttime fish assemblages to assess the effects of anthropogenic stressors that may have more influence at night (e.g. artificial light).

Studies at night have used different colours of light to illuminate fishes and compare day versus night assemblages (Svane and Barnett 2008; Svane et al. 2008; Harvey et al. 2012a; Harvey et al. 2012b; Harvey et al. 2012c). Different light colours can, however, result in dissimilar assemblages (e.g. Fitzpatrick et al. 2013), due to the spectral characteristics of each light colour, their attenuation in the water, and visual sensitivity

of species (Fitzpatrick et al. 2013). Red light (wavelength = 620 nanometres and above) is attenuated quickly within the water column leading to a small illuminated area compared to other colours such as white or blue (Fitzpatrick et al. 2013). Red light is also thought to be above the visual threshold for many nektonic species and thus does not interfere with the behaviour of species, unlike blue or white light, which may variously attract or scare away species (Marchesan et al. 2005; Widder et al. 2005).

Previous studies assessing seasonal and yearly variations have found differences in abundances and diversity of fish assemblages between seasons and years (e.g. Thompson and Mapstone 2002; Mazumder et al. 2005; McIlwain et al. 2011). Thompson and Mapstone (2002) sampled coral-reef fish assemblages over daily, monthly, and annual time scales to begin understanding the effect of sampling error in long-term monitoring studies and found that inter-annual variation was mostly accounted for by daily variability and did not reflect a true change in abundance. This shows the complexity of understanding temporal variations in fish assemblages and implies that studies need to be conducted across multiple timeframes to ensure a comprehensive understanding of fish assemblages.

The use of Baited Remote Underwater Video Stations (BRUVS) is now a common method of daytime sampling for demersal assemblages (Murphy and Jenkins 2010) and also provides a safe and non-extractive solution for assessing nighttime assemblages (Whitmarsh et al. 2017). I aim to assess temporal changes of fish assemblages in South Australian waters over varying time scales using BRUVS. Specifically, I will: assess whether using different light colours affects temperate fish assemblages; determine the differences in assemblages between fish species observed during the day compared to nighttime; and assess the influence of seasonal and yearly variations on fish assemblages. To assess the generality of the results across sites and habitats, I compared assemblages from six sites in Gulf St Vincent, South Australia, across four habitat types and two protection levels, and over a three-year period.

Methods

Sampling locations

The six sampling sites were chosen from locations within Gulf St Vincent (35° S, 138° E), South Australia, to represent different habitats and protection levels: Long Spit (unprotected shallow seagrass at 7 m depth; S34.56461 E138.22672); Zanoni (a historical shipwreck protected since 1983 at 18 m; S34.51163 E138.06368); Barge (unprotected shipwreck at 17 m; S34.52841 E138.06356); and Near Zanoni (unprotected soft sediment at 11–15 m deep; S34.51496 E138.08525); Aldinga Inside (protected reef since 1971; S35.27360 E138.43265); Aldinga Outside (unprotected reef; S35.24522 E138.45937). Depths at Aldinga Reef ranged from 6 to 18 m.

BRUVS set-up

Deployments were made using single BRUVS set-up similar to Whitmarsh et al. (2014) that consisted of Go-Pro Hero 3+ Silver Edition cameras mounted horizontally. Cameras were set to record 1080p wide angle video. BRUVS were baited with 800 g of crushed sardines (*Sardinops sagax*) per deployment and left for a soak time of 60 minutes on the seafloor. Due to the small size of the exclusion zone at the Zanoni and to keep a close proximity to the wrecks, deployments at the Zanoni, Barge, and Near Zanoni sites could not be spaced the recommended 200 m apart (Malcolm et al. 2007), and so were spaced approximately 100 m apart (similar to some other previously published studies e.g. Unsworth et al. 2014; Ebner et al. 2015; Griffin et al. 2016). Deployments at Long Spit and Aldinga Reef were spaced at least 200 m apart. I deployed the six replicates at each site simultaneously to minimise potential movements of fish between replicates and thus chances of double-counting individuals twice in a single day. While it is possible for individuals to have travelled between replicates within the 60-min soak time, larger, mobile species that are more likely to travel among BRUVS (e.g. smooth rays, *Dasyatis brevicaudata*, and flathead, *Platycephalus* spp.) were seen at only one BRUVS unit at respective sites, which suggests minimal movement of individuals occurred between replicates. Other species and individuals which could have potentially travelled between replicates (e.g. Port Jackson sharks, *Heterodontus portusjacksoni,* snapper, *Chrysophrys auratus*, and fiddler rays, *Trygonorrhina dumerilii*) were checked for the times they occurred and any identifying marks to indicate possible movements between replicates. I found no evidence for movement of individuals

between replicates being detected, with the exception of a singleton white shark, *Carcharodon carcharias,* observed during winter on multiple replicates which was subsequently removed and counted as only one sighting for its first appearance. Previous studies using BRUVS assessing juvenile white sharks (Harasti et al. 2016) and using vertically close-spaced benthic and pelagic BRUVS (Clarke et al. unpublished data) similarly found no evidence of movement between replicates.

Field sampling

Sampling was conducted over four sampling trips (Table 3.1). The first trip was from 15–16th of January in 2013 during the austral summer. The next trip occurred from the 3–18th of September in 2014 in the austral winter. Sampling in the austral summer in 2015 occurred from 18–30th of March 2015 but daytime footage was lost due to technical faults with the electronic storage of the videos. Sampling was, therefore, repeated in the austral summer of 2016 from the 31st of January to the 23rd of March. With the exception of 2013, sampling was conducted at all six sites with six replicate deployments taken per site as this is a standard number of deployments for many BRUVS studies (Whitmarsh et al. 2017). In 2013, the Barge was not sampled and the two Zanoni sites had only three replicate deployments taken. All daytime sampling occurred in full daylight hours at least one hour after sunrise and concluded at least 1 hour before sunset. For nighttime deployments, BRUVS were not deployed until full darkness (at least 1 hour after sunset) to avoid crepuscular activity changes in fish assemblages (Myers et al. 2016). Light units were mounted above the cameras shining towards the bait arm and bag, and were the same as those used in Fitzpatrick et al. (2013). At each site, six replicate BRUVS deployments were made using either red light (620–630 nm) or blue light (450–465 nm). Each replicate was randomly allocated a light colour, with half of the total replicates being blue and half red.

Table 3.1: The number of replicates usable at each site for each sampling trip. Note: no replicates were taken at the Barge in 2013 and the nighttime Aldinga replicates were lost due to hard drive failure.

Year	**Season**	**Time of day**	**Site**						
			Aldinga Inside	Aldinga Outside	Barge	Long Spit	Zanoni	Near Zanoni	**Total**
2013	Summer	Day	6	6	0	6	3	3	**24**
2014	Winter	Day	6	6	6	6	6	6	**36**
2015	Summer	Night	0	0	9	10	11	12	**42**
2016	Summer	Night	11	7	12	12	12	11	**65**
2016	Summer	Day	4	6	5	6	6	6	**33**
		Total	**27**	**25**	**32**	**40**	**38**	**38**	**200**

Video annotation

Due to toppling over in strong currents, equipment failure, and outside interference of deployed BRUVS units, sixteen replicates had to be excluded from analysis, leaving a final $N = 200$. The included replicates were spread across the sites and sampling times. Videos were analysed using EventMeasure software (www.seagis.com.au). Videos were processed for fish abundance and diversity as per Whitmarsh et al. (2014). *MaxN*, the maximum number of individuals (for each species or taxon) observed in a single frame throughout the duration of a single replicate, was used as a conservative measure of relative abundance.

Data analysis

Analysis was carried out using PRIMER v.7/PERMANOVA+ software (Anderson et al. 2008; Clarke and Gorley 2015). Different levels of transformation (square root, fourth root, and dispersion weighting) were viewed via shade plots as a guide to find the most appropriate transformation (Clarke et al. 2014). Standard dispersion weighting (Clarke et al. 2006) by the factor Site was chosen and used to combat over-emphasis from the schooling nature of particular species.

Red vs. blue light analysis

Using the Bray-Curtis similarity matrix, transformed community data were portrayed visually as non-metric Multi-Dimensional Scaling (nMDS) and Canonical Analysis of Principal co-ordinates (CAP) ordination plots. Multivariate PERMutational ANalyses Of VAriance (PERMANOVA) were used along with CAP to test the difference in fish assemblages observed between the light colours. Factors analysed included Year (2 levels, a random factor), Light Colour (2 levels, fixed), and Site (6 levels, random). Pairwise tests were further used to assess significance of levels within factors and interactions.

Day vs. night analysis

Similar to red vs. blue light analysis, day and night assemblages were viewed using nMDS and CAP ordination plots, with PERMANOVA and CAP also being used to test for differences between factors along with pairwise tests for further interpretation. The factors analysed included Time of Day (2 levels, fixed) and Site (6 levels, random).

SIMilarity PERcentages (SIMPER) analysis was used to assess species contributions to the observed assemblages and differences across factors.

Seasonal and yearly analysis

A dummy variable was added when calculating the Bray-Curtis similarity matrix for these analyses due to the overall sparse data matrix and some replicates being very distinct from the rest (Anderson et al. 2008). High stress values (>0.2) for the nMDS for the yearly and seasonal daytime data excluded this from being used; however, CAP ordination plots and analyses were used to view and test differences between sampling trips along with PERMANOVA analysis. The factors analysed were Year (3 levels, random), Season (2 levels, fixed), and Site (6 levels, random). I chose Season to be a fixed factor due to summer and winter being the most extreme seasonal comparisons and producing the largest potential effect size. Pairwise tests were again used to assess significant factors and interactions. SIMPER was used to assess species driving similarity within sampling times as well as calculating dissimilarities between groups.

Coefficients of variation (calculated from the total number of individuals per replicate using the standard deviation divided by the mean) and PERMDISP values (a value showing the multivariate dispersion of the data) were calculated to assess the relative variability of fish assemblages.

Results

Red vs. blue light assemblages

In total, 52 species were observed at night (36 and 44 species in 2015 and 2016, respectively). Similar numbers of species were observed from each light colour in 2015 with 23 species observed using red light and 27 using blue. More species were observed on blue light in 2016 with 40 species compared to 29 for red light. Mean number of individuals (± SE) per replicate were similar within years for each light colour (2015 blue = 19.3 ± 4.0 [n = 22], red = 21.3 ± 3.4 [n = 20]; 2016 blue = 15.1 ± 2.3 [n = 34], red = 16.3 ± 2.6 [n = 31]), with 2016 observing slightly fewer individuals per replicate than 2015. Blue light observed a higher number of unique species overall than red light, however, the majority of species (53 %) were seen on both light colours (Figure 3.1a) and total numbers (red = 934 vs. blue = 940) were similar for each colour.

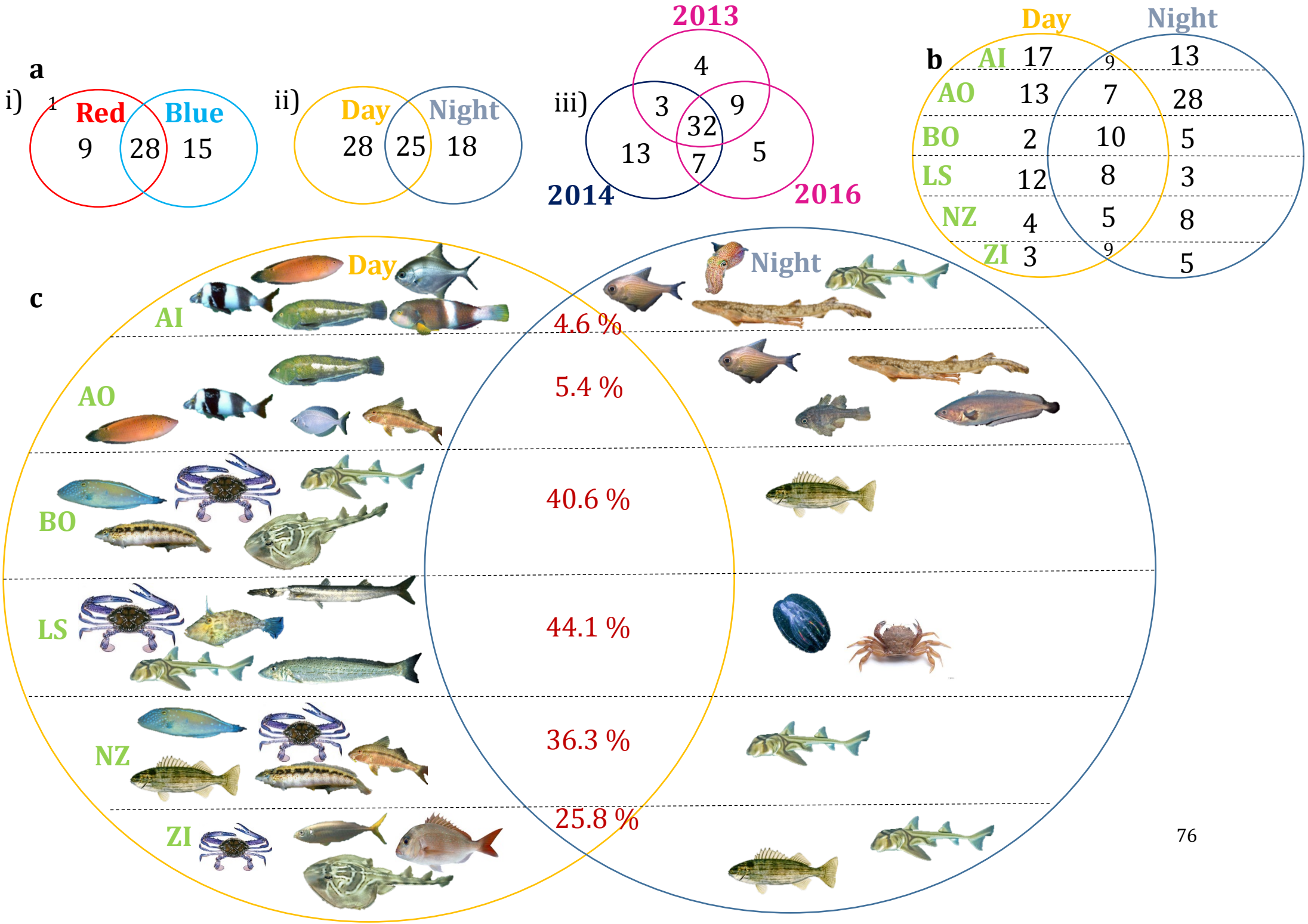
a
i)
Red
Blue
1
9
28
15
ii)
Day
Night
28
25
18
iii)
2013
4
3
9
32
13
7
5
2014
2016
b
Day
Night
AI 17 9 13
AO 13 7 28
BO 2 10 5
LS 12 8 3
NZ 4 5 8
ZI 3 9 5
c
Day
Night
AI
AO
BO
LS
NZ
ZI
4.6 %
5.4 %
40.6 %
44.1 %
36.3 %
25.8 %

Figure 3.1: Venn diagrams showing a) the number of species observed for i) red and blue light assemblages at nighttime; ii) daytime and nighttime assemblages from 2016; and iii) daytime assemblages from each year sampled (with dark blue indicating winter assemblages and pink indicating summer assemblages), b) the number of species by site for daytime, both and nighttime assemblages from 2016 and c) similarities of the assemblages from SIMPER for 2016 daytime and nighttime assemblages with the red percentages indicating the overall similarity between pairs and fish icons representing the 5 species contributing to the largest dissimilarity in assemblages or for nighttime assemblages were in the top 70 % of contributors and had the higher abundance at night. AI = Aldinga Reef Inside, AO = Aldinga Reef Outside, BO = Barge Outside, LS = Long Spit, NZ = Near Zanoni, and ZI = Zanoni Inside

Considering there was no significant effect of year and light colour, data from both years were combined and no significant differences were again detected between the two light colours (Table 3.2a). There were differences detected for the interaction between site and light colour (Figure 3.2a, Table 3.2a), but pairwise tests were unable to differentiate at which sites the differences occurred (Table 3.2b). CAP analysis was also unable to detect significant differences between assemblages observed from red and blue light (Figure 3.3a, trace and delta statistics $p = 0.114$) with a low allocation success rate of 56.2 %. Data from both light colours were, therefore, pooled in further analyses when comparing daytime and nighttime assemblages.

Daytime vs. nighttime assemblages

Sixty-seven species were observed during daytime, which was more than during nighttime (44 spp.). Higher numbers of individuals were also observed during the day than at night (day = 2,771 vs. night = 1,022) along with a higher number of unique species (Figure 3.1a). The numbers of species observed varied across sites with higher numbers at Aldinga compared to the other areas (Figure 3.1b). Numbers of unique and shared species were also variable across sites and times of day (Figure 3.1b). Aldinga Outside and Near Zanoni were the only sites with a higher number of species that were unique to night assemblages over daytime assemblages (Figure 3.1b).

Table 3.2: PERMANOVA results for the analysis of light colour at nighttime with a) the model testing Year (2 levels, random), Site (6 levels, random), Light Colour (2 levels, fixed) and b) the pairwise tests for each site by light colour and year. Monte Carlo *p* values (*P*(MC)) were used when unique permutations were low. Unless otherwise specified unique permutations (perms) ranged from 973 to 999. Bold entries were significant

a			**Combined**		**2015 only**		**2016 only**	
Source	**df**	**MS**	**Pseudo-*F***	***P*(perm)**	**Pseudo-F**	***P*(perm)**	**Pseudo-*F***	***P*(perm)**
Ye	1	6814.8	1.62	0.2615				
Si	5	23808	5.87	**0.0011**	9.95	**0.001**	9.61	**0.001**
Li	1	3258	1.26	0.3587	0.82	0.571	1.76	0.187
YexSi	3	4215.1	2.48	**0.0002**				
YexLi	1	1188	1.30	0.3227				
SixLi	5	2570.3	2.68	**0.0208**	1.23	0.236	1.08	0.329
YexSixLi	3	909.4	0.54	0.9756				
Res	85	1698						

b	**Red vs. Blue**			**2015 vs. 2016**	
Site	**t**	***P*(MC)**	**Unique perms**	**t**	***P*(perm)**
Aldinga Inside	0.67	0.7765	330	-	-
Aldinga Outside	1.38	0.1576	35	-	-
Barge	0.95	0.543	6	1.12	0.271
Long Spit	1.33	0.2931	6	1.65	**0.023**
Zanoni Inside	2.11	0.0647	6	2.19	**0.001**
Near Zanoni	1.78	0.0908	6	2.02	**0.001**

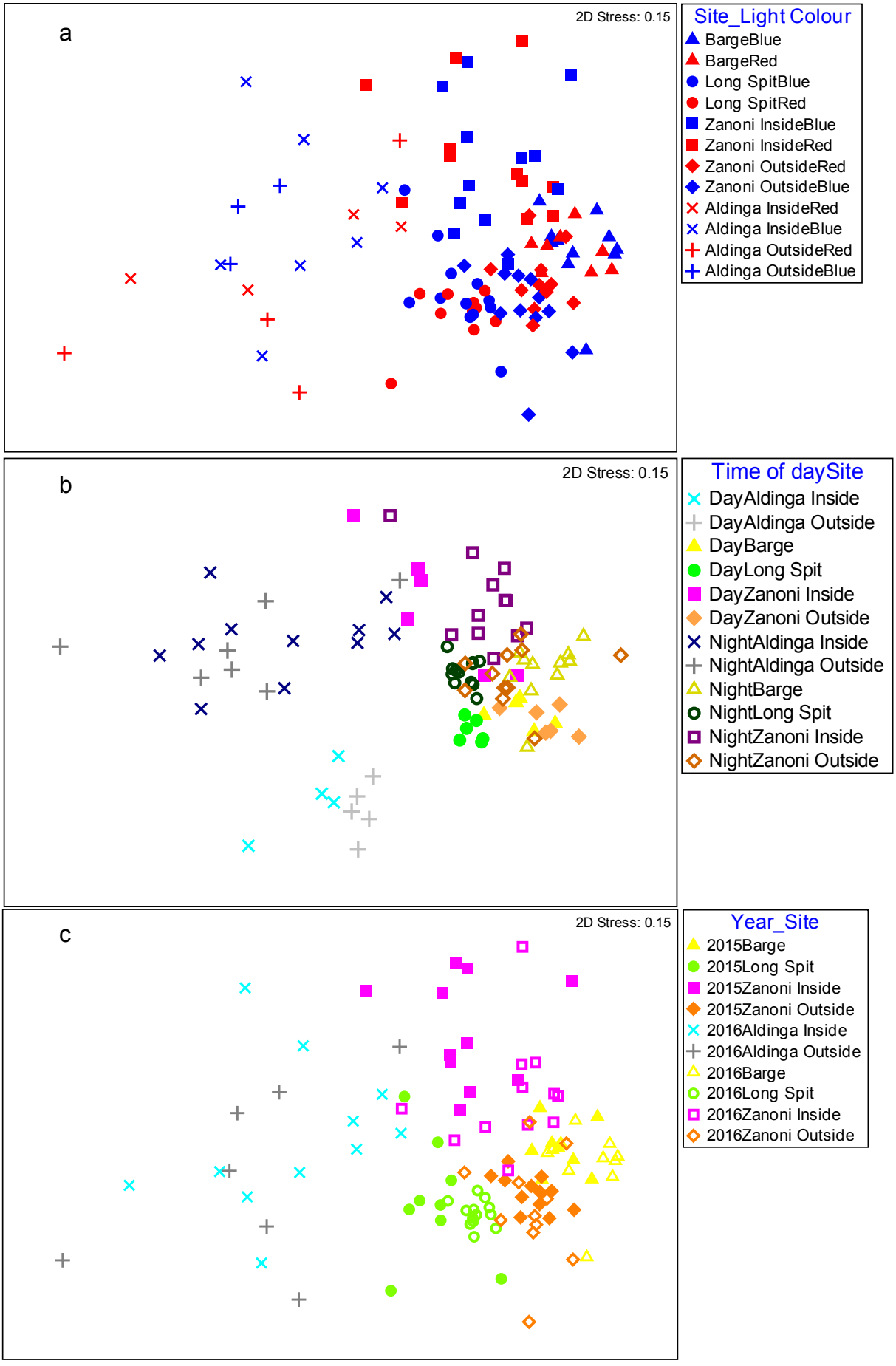

Figure 3.2: nMDS ordination plot showing influences of: a) Light Colour by Site for 2015 and 2016 data of nighttime assemblages; b); Time of Day by Site for 2016 data; and c) the interaction of Year and Site for 2015 and 2016 nighttime assemblage data

Overall, there was a significant difference between daytime and nighttime assemblages as well as a significant interaction between time of day and site (Figure 3.2b, Table 3.3). Pairwise tests indicated highly significant differences at all sites between daytime and nighttime assemblages ($p < 0.002$). CAP analysis also showed significant differences between daytime and nighttime assemblages (Figure 3.3b, trace and delta statistics $p = 0.001$) with a high allocation success rate of 92.7 %.

Average similarity between day and night assemblages was relatively low at 17.8 % with daytime assemblages characterised by a higher abundance of blue swimmer crabs *Portunus armatus*, Port Jackson sharks *Heterodontus portusjacksoni*, Degen's leatherjackets *Thamnaconus degeni*, fiddler rays *Trygonorrhina dumerilii*, and snapper *Chrysophrys auratus* along with 11 other species (Table 3.4). Nighttime assemblages had higher abundances of only two (trumpeter *Pelates octolineatus* and bullseye *Pempheris multiradiata*) of the 18 influential species identified from SIMPER analysis (Table 3.4). Species characterising diel differences were more variable between sites with Bray-Curtis similarities between day/night assemblages ranging from 4.6 % at Aldinga Inside to 44.1 % at Long Spit (Figure 3.3c). At four out of six sites, *P. armatus* was still a top-5 contributing species for daytime assemblages, while *Th. degeni* and *Tr. dumerilii* were contributors at two out of six sites (Figure 3.3c). Conversely, *H. portusjacksoni* was a daytime contributor at two out of six sites but a nighttime contributor for another three sites (Figure 3.3c). Generally, there were fewer species identified from the SIMPER output that had higher abundances at night than during the day (Figure 3.3c).

Table 3.3: PERMANOVA results from the comparison of daytime and nighttime data from 2016 using the factors Time of day (2 levels, fixed), and Site (6 levels, random) and the pairwise tests for each site by Time of Day. Unique permutations ranged from 568 to 999. Significant values are shown in bold.

Source	**df**	**MS**	**Pseudo-*F***	***P*(perm)**
Ti	1	26017	3.57	**0.022**
Si	5	20245	12.54	**0.001**
TixSi	5	7369.2	4.56	**0.001**
Res	84	1614.4		

Day vs. night		
	t	***P*(perm)**
Aldinga Inside	2.2714	**0.001**
Aldinga Outside	2.3505	**0.001**
Barge	2.9138	**0.001**
Long Spit	3.7369	**0.001**
Zanoni Inside	2.1142	**0.002**
Near Zanoni	2.4373	**0.001**

Table 3.4: SIMPER result from daytime vs. nighttime dispersion weighted assemblage data from 2016 for all sites combined. Average similarity between the two times of day was 17.77 %. Consistent indicators (indicated by a diss/SD greater than 1) are shown in bold.

Species	Average abundance (*MaxN*/video)			Average dissimilarity	Diss/ SD	% contribution	Cumulative %
	Daytime		Nighttime				
Portunus armatus	1.94	>	1.38	10.84	**1.22**	13.2	13.2
Heterodontus portusjacksoni	1.19	>	1.06	7.28	**1.18**	8.8	22.0
Pelates octolineatus	0.51	<	0.67	5.41	0.81	6.6	28.6
Thamnaconus degeni	0.71	>	0.04	4.81	0.71	5.8	34.5
Trygonorrhina dumerilii	0.69	>	0.14	4.32	0.89	5.3	39.7
Chrysophrys auratus	0.44	>	0.07	3.73	0.48	4.5	44.2
Parapercis haackei	0.51	>	0.02	3.18	0.71	3.9	48.1
Upeneichthys vlamingii	0.53	>	0.01	2.63	0.74	3.2	51.3
Austrolabrus maculatus	0.44	>	0.00	2.10	0.55	2.5	53.9
Parequula melbournensis	0.34	>	0.08	2.02	0.51	2.5	56.3
Sillaginodes punctatus	0.34	>	0.03	1.83	0.57	2.2	58.6
Scobinichthys granulatus	0.38	>	0.00	1.83	0.48	2.2	60.8
Notolabrus parilus	0.34	>	0.02	1.67	0.53	2.0	62.8
Trachurus novaezelandiae	0.19	>	0.06	1.45	0.45	1.8	64.6
Scorpis aequipinnis	0.22	>	0.03	1.32	0.35	1.6	66.2
Pseudocaranx spp.	0.23	>	0.01	1.32	0.45	1.6	67.8
Cheilodactylus nigripes	0.28	>	0.00	1.30	0.49	1.6	69.4
Pempheris multiradiata	0.00	<	0.20	1.23	0.36	1.5	70.9

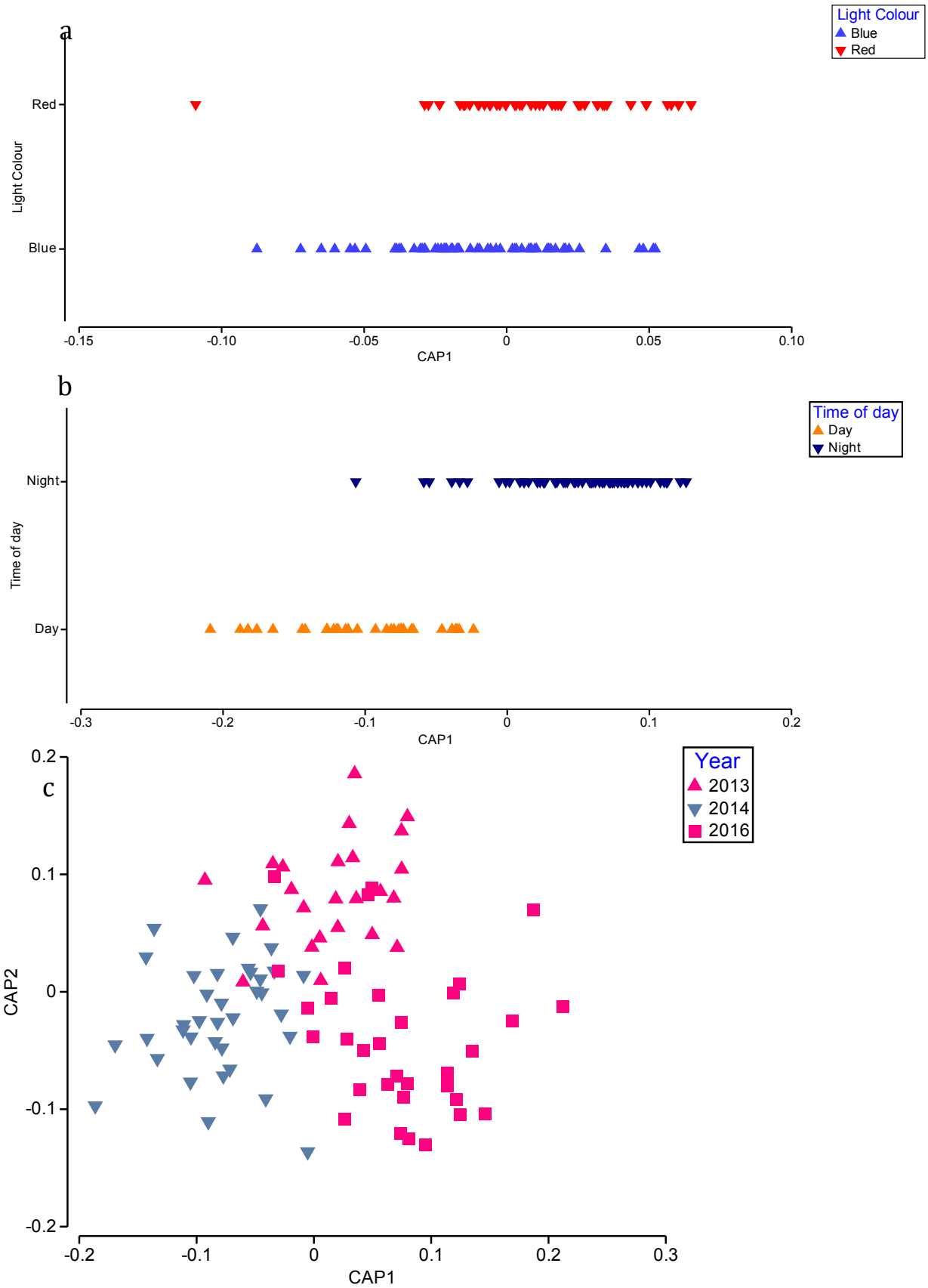

Figure 3.3: CAP ordination plots of a) the factor Light Colour (for *m* = 7 axes) for both 2015 and 2016 combined; b) the Time of Day factor (*m* = 5 axes) and c) Year factor for daytime assemblages only (*m* = 12 axes)

Seasonal and yearly assemblages

For all daytime assemblages, 5,317 individuals were observed across 73 species. Similar species numbers were observed across years and seasons with 48 species observed in the summer of 2013, 55 in winter of 2014, and 53 in summer of 2016. However, summer of 2016 had a much higher number of mean individuals (± SE) per replicate (83.9 ± 13.0) than winter of 2014 (40.1 ± 6.2) or summer of 2013 (45.8 ± 10.4). Only 43 % of species were observed across all daytime sampling periods (Figure 3.1a) with the winter sampling period in 2014 recording the highest number of unique species (17 %). The two summer sampling periods had the highest number of shared species out of the three combinations (Figure 3.1a).

For the assemblages observed, PERMANOVA tests showed that Year was a significant factor along with Site and the Ye x Si interaction; however, Season as a main effect was not significant although Site and Si x Se interaction were (Table 3.5). CAP analysis also showed significant differences (trace and delta statistics $p = 0.001$) between the years and seasons (Figure 3.3c) and had a relatively high allocation success rate (75 %). Pairwise tests indicated that sites at Aldinga Reef, particularly the Outside site, were less influenced by year and season than the more northern sites (Table 3.5). Seasonality appeared to affect Zanoni Inside the most because this site was not significantly different for the summer years (Table 3.5) but also had the highest dissimilarity for the winter comparative years (Figure 3.4). Contrasting to this, other sites such as Long Spit and Near Zanoni appeared to be more affected by yearly variation, particularly differences between summer of 2013 and the other years (Figure 3.4).

Assemblages at Long Spit were consistently dominated by Port Jackson sharks, *Heterodontus portusjacksoni*, across all three sampling times, although similarity within groups was higher in the later sampling times (Table 3.6). Lower similarity and more influence from leatherjackets, *Th. degeni*, and grubfish, *Parapercis haackei*, characterised the winter assemblage at the Zanoni compared to larger within group similarities and snapper, *C. auratus,* dominating assemblages for the summer years (Table 3.6). Assemblages at Near Zanoni were more variable across sampling times with only one species found consistently in winter of 2014 and summer of 2016; however,

within-group similarities were often higher showing less variability among replicates during each sampling trip (Table 3.6). Within-group similarity and influences of the leatherjacket, *Th. degeni,* were high for both sampling occasions of the Barge (Table 3.6). Three sites in 2016 were characterised by a consistent abundance of blue crabs, *P. armatus*, at the Barge, Long Spit and Near Zanoni.

For nighttime assemblages, significant yearly variation was observed at Long Spit, Near Zanoni and Zanoni, with no difference detected at the Barge (Figure 3.2c, Table 3.2) and no comparison to be made for the Aldinga sites due to no recorded data for 2015.

Variance in assemblages

Overall, CV values were high with the 2015 nighttime assemblages having the lowest value (84.4 %). Summer 2016 had the lowest level of variance out of the three sampling seasons for daytime assemblages with a CV value of 89.1 % and a PERMDISP value of 54.2 compared to 93 % and 58.6 for winter 2014 and 111.4 % and 61.3 for Summer 2013. Comparisons among seasons indicated more similarity between summer seasons with a CV of 104.2 % and PERMDISP value of 60.3 for the summer 2013 and 2016 sampling sessions combined compared to 128.8 % and 129.5 % for winter 2014 and summer 2013 combined and winter 2014 and summer 2016, respectively.

Table 3.5: PERMANOVA results for the daytime assemblage data from summer 2013, winter 2014 and summer 2016 testing the factors a) Season (2 levels, fixed) and b) Year (3 levels, random) along with Site (6 levels, random) along with their respective pairwise tests of the significant interactions showing differences for each site. Unique permutations ranged from 84 to 999. Significant values are shown in bold.

a			**Season**		**b**			**Year**	
	df	**MS**	**Pseudo-*F***	***P*(perm)**		**df**	**MS**	**Pseudo-*F***	***P*(perm)**
Se	1	10128	1.98	0.118	**Ye**	2	9252.7	1.93	**0.023**
Si	5	215551	13.39	**0.001**	**Si**	5	20644	4.26	**0.001**
SexSi	5	5155.8	3.20	**0.001**	**YexSi**	9	4899.5	3.59	**0.001**
Res	81	1609.5			**Res**	76	1365.3		

	Summer vs. winter		**2013 vs. 2014**		**2013 vs. 2016**		**2014 vs. 2016**	
	t	***P*(perm)**	***t***	***P*(perm)**	***t***	***P*(perm)**	***t***	***P*(perm)**
Aldinga Inside	1.44	**0.023**	1.31	0.054	0.79	0.864	1.20	0.175
Aldinga Outside	1.39	0.057	1.26	0.089	0.84	0.627	1.30	0.084
Barge	2.81	**0.003**	-	-	-	-	2.81	**0.003**
Long Spit	1.97	**0.009**	2.70	**0.004**	3.34	**0.004**	2.90	**0.005**
Near Zanoni	2.61	**0.001**	3.89	**0.011**	3.40	**0.014**	3.78	**0.002**
Zanoni Inside	2.13	**0.012**	2.12	**0.014**	1.69	0.073	1.87	**0.019**

Table 3.6: SIMPER analysis for YearSeason showing similarity values of assemblages categorised within groups and the species driving the similarities. Aldinga Reef sites are not shown due to weaker differences between seasons and years. Sim/SD shows the similarity divided by the standard deviation, with consistent indicators of each group having values above 1.00 and are shown here in bold. Avg. sim. is the average similarity within a group.

Location	Summer 2013				Winter 2014				Summer 2016			
	Avg. sim. (%)	**Species**	**Avg. abundance**	**Sim/SD**	**Avg. sim. (%)**	**Species**	**Avg. abundance**	**Sim/SD**	**Avg. sim. (%)**	**Species**	**Avg. abundance**	**Sim/SD**
Barge	-				72.0				64.4			
		-	-	-		*Th. degeni*	6.81	**5.75**		*Th. degeni*	5.98	**1.67**
										Po. armatus	1.47	**4.59**
Long Spit	32.0				56.5				60.1			
		H. portusjacksoni	1.50	0.75		*H. portusjacksoni*	2.17	**2.02**		*H. portusjacksoni*	3.50	**3.96**
		Pe. octolineatus	0.19	0.71		*Neo. balteatus*	1.33	**4.81**		*Po. armatus*	2.10	**5.53**
										S. granulatus	1.50	**1.18**
Near Zanoni	48.0				64.2				59.4			
		H. portusjacksoni	3.67	**6.38**		*Nec. integrifrons*	2.39	**2.15**		*Th. degeni*	5.48	**3.34**
						L. gaimardii	1.83	**3.72**		*Po. armatus*	1.75	**2.87**
						Th. degeni	1.73	**1.90**		*Pe. octolineatus*	2.08	0.87
Zanoni Inside	54.0				27.2				40.4			
		C. auratus	3.85	**4.17**		*Th. degeni*	1.80	0.81		*C. auratus*	1.99	0.86
		Tr. novaezelandiae	2.36	**3.60**		*Pa. haackei*	0.70	0.66		*H. portusjacksoni*	1.17	**1.28**

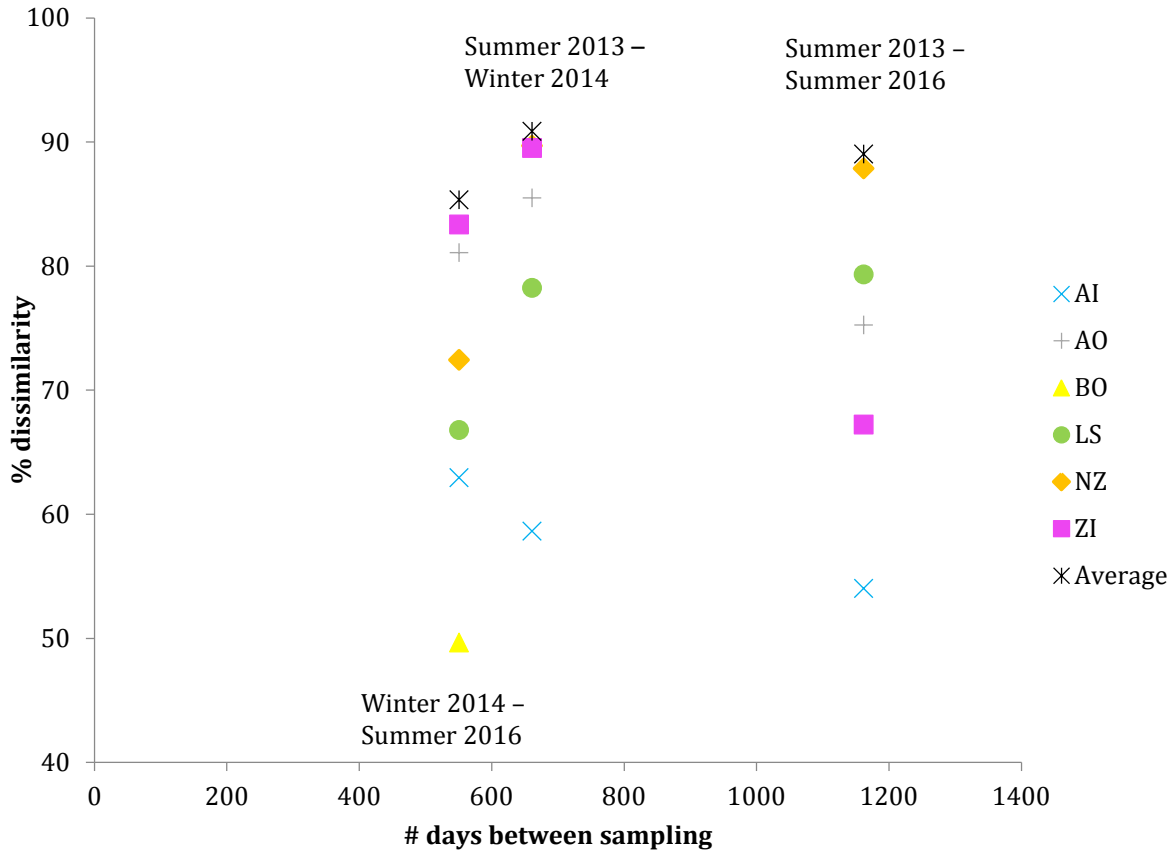

Figure 3.4: Scatterplot showing the dissimilarity of a site against the time interval between sampling periods, with the average indicating the average of all sites combined

Discussion

The fish assemblages observed showed variable changes over the different temporal scales tested. There were strong differences between assemblages observed during the daytime versus nighttime, with variable results among sites for the differences between years and seasons. The sites sampled showed distinct assemblages during all sampling periods likely driven by differences in habitat and site-specific attributes such as protection status.

In contrast to previous studies (e.g. Harvey et al. 2012a,c; Fitzpatrick et al. 2013), light colours did not significantly affect the fish assemblage observed in my study. This disparity in outcomes might be due to the differences in visibility, small-scale spatial and temporal differences between study sites, and the different species observed between studies. It is unlikely to be just due to light characteristics (i.e. light wavelength or luminosity) because the same lights were used in the present study, Harvey et al. (2012a), and Fitzpatrick et al. (2013). As single BRUVS were used in the present study, I was unable to accurately measure the field of view (cf. stereo BRUVS). Visibility was, however, relatively poor (~3 m) during the nighttime sampling. As a result, the difference in light penetration and area illuminated between red and blue lights might have been reduced, contrary to a previous study, where the discrepancy in nighttime fish assemblages is suggested to be affected by differences in light penetration between colours (Fitzpatrick et al. 2013). A comparison of the light penetration using different light colours under various visibility conditions could determine whether light penetration and visibility affect the influence of light colours on observed fish assemblages.

Comparisons across studies also indicate that species and individuals may act differently under different environmental conditions. For example, out of the 18 species seen in both the present study and Harvey et al. (2012a) during the nighttime, seven species were observed under similar lighting conditions, four species were observed at night for the current study but during the day for Harvey et al. (2012a), four species were observed for a single light colour during the present study but on both colours for Harvey et al. (2012a) and the remaining three species were observed under either opposite light colours, or opposing times of the day. These behavioural differences

amongst species may have contributed to the contrasting results between studies assessing differences among light colours.

The strongest significant differences between assemblages were observed between daytime and nighttime. All sites showed distinct assemblages for each time of day, with typical changes in assemblages occurring from diurnally-active fish species, such as wrasses and leatherjackets, to those typically nocturnal, such as cat sharks, bulls eye fishes, and bobtail squid. Only two sites (Aldinga Inside and Long Spit) had a higher number of species during the day compared to nighttime, which is an unexpected trend as most studies do not report higher biodiversity for nighttime fish assemblages (Harvey et al. 2012a; Aguzzi et al. 2013; Myers et al. 2016). The increased species richness at night may stem from habitat-related connectivity, where previous studies have shown nocturnal excursions of fish from other habitats into sand and seagrass areas (Kopp et al. 2007), such as those found at Long Spit and Near Zanoni (and to a lesser extent The Barge and Zanoni where the wrecks are surround by soft-sediment habitat). Particular species exhibited different patterns of abundance for the day and night depending on the site in which they were observed. For example, Port Jackson sharks had higher abundances during the daytime at the Barge and Long Spit, but higher numbers at night for Aldinga Inside, Near Zanoni and Zanoni. These differences may be related to the nocturnal foraging strategies of *H. portusjacksoni* (Powter and Gladstone 2009). Thus, my study highlights the different nocturnal assemblage, which is not captured through traditional daytime sampling and may show important changes over seasons and years that is not present when assessing only daytime assemblages.

Fish assemblages changed distinctly across years and seasons. Due to logistical constraints and equipment failure I was unable to conduct sampling across the summer and winter seasons within a single year and thus cannot attribute with certainty which effects are the result of seasonal or yearly variation among assemblages. Continued sampling of seasonal and yearly variation may begin to tease apart such effects and further our understanding of how fish assemblages change over time for these places and habitats. Indicators of similarity and variance suggest that different sites responded differently to seasonal and yearly changes, with stronger effects of yearly variation seen at Long Spit and Near Zanoni compared to stronger seasonal influences at Zanoni. The

reef sites of Aldinga appeared to show the least difference between sampling times which could indicate a relatively stable assemblage, which is not often observed for rocky reef areas (e.g. Henriques et al. 2013). The differences among sampling times for Long Spit and Near Zanoni are in part driven by the decreased abundance of *Portunus armatus* during the 2013 summer. In 2013, there was a voluntary closure of the blue crab fishery within South Australia due to declining catch rates (Beckmann and Hooper 2017), which coincides with the minimal observations made by my study. The seasonal differences at the Zanoni are likely driven by the high abundance and spawning aggregations of snapper *C. auratus* in late spring and summer (Fowler et al. 2017). The presence of the white shark, *Ca. carcharias*, at the Zanoni during winter might have negatively influenced snapper abundance. Indeed, large predators can influence the abundance and composition of fish assemblages observed via baited video methods (Klages et al. 2014). Large predators can also illicit direct and indirect responses in prey species, often referred to as creating a landscape of fear (Wirsing et al. 2007; Wirsing et al. 2008). Other factors driving differences in assemblages between years may include environmental factors such as heatwaves, fish kills due to pathogens and a general shift in assemblages throughout time (e.g. due to a warming ocean or El Nino cycles; Lehodey et al. 2006; Olsson et al. 2012). Meanwhile, seasonal variation in fish assemblages can often be driven by changes in water temperature and other physical factors including changing salinity and current regimes (Olsson et al. 2012). Additionally, fish may move to different areas for foraging preferences or for breeding and spawning strategies (Henriques et al. 2013). Juvenile and adult fish often exhibit different movement patterns that may change at different times of the year due to growth or seasonal environmental changes (Fowler and Booth 2013).

Conclusion

I found fish assemblages to be highly variable and thus require sampling on many occasions to truly understand abundance and diversity within an area. Contrary to previous studies, there was no difference in fish assemblages under red and blue light. This suggests that the effects of light colours can be nil to variable and need to be carefully considered when designing a study aimed at sampling nocturnal fish assemblages. Daytime and nighttime assemblages were the most different with a typical shift from diurnally active to nocturnally active species seen at most sites. Yearly and

seasonal patterns were variable across sites and highlighted the need for multiple sampling trips to each site to get a comprehensive overview of abundance and diversity. My results can be used to support management decisions related to monitoring or sampling, particularly for marine protected areas and other long-term studies that need to account for variability in fish assemblages.

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Chapter 4

Exploring variations of anthropogenic stressor effects on fish assemblages: an analysis of four case studies

Abstract

The many types of anthropogenic stressors and their influence on marine ecosystems is an increasing area of research. However, some stressors receive greater research attention than others, leading to some gaps in our overall understanding. But stressors rarely operate in isolation of each other. Despite this, the cumulative effects of multiple stressors are studied much less often than single impacts, in part because they can be more complex and difficult to untangle. Here I have chosen four case studies to assess the associations of different anthropogenic stressors and fish assemblages, using BRUVS as the tool for these assessments. Due to a paucity of research in certain ecosystems and areas (i.e. temperate seagrass and soft-sediment habitats), I have tried to target such locations where possible and appropriate. The four case studies I have chosen variously assessed the effects of marine protected areas (#1), disposal of brine effluent from desalination (#2), bait and berley input from tourism activities (#3), and multiple cumulative stressors within a shallow embayment (#4). The results from these four studies varied with different stressors having different associations with the assemblages. Marine protected areas showed some association with fish assemblages despite the short time-frame of their enforcement (months to years). In contrast, minimal association was seen from desalination brine, while bait and berley was shown to have impacts only on certain species. Multiple anthropogenic stressors in the form of proximity to effluent (i.e. run-off, groundwater, and stormwater) input and distance to oyster leases were shown to be associated with the fish assemblages in the shallow embayment along with some other environmental variables. Overall, results from these case studies have expanded our knowledge of their effects in less-studied temperate habitats and will have ongoing use for management decisions.

Introduction

Coastal regions in particular are at threat from climate change, recreational and commercial fishing, aquaculture practices, pollution, and coastal urbanisation (Lotze et al. 2006; Halpern et al. 2008b; Robbins et al. 2017). These stressors put increased pressures on marine communities including fish assemblages, which have shown declines in abundance and diversity (Myers and Worm 2003; Worm et al. 2006; McCauley et al. 2015), e.g. due to fishing practices (Halpern and Warner 2002; Lester et

al. 2009), urbanisation (Vargas-Fonseca et al. 2016), and pollution (Islam and Tanaka 2004).

In nature, most assemblages are subject to multiple potential stressors. Despite the many studies focusing on anthropogenic stressors (particularly fishing pressure), few studies have attempted to consider, measure, and tease apart the cumulative effects of multiple stressors on fish assemblages (Crain et al. 2008). Understanding how potential stressors can interact is particularly important for ecosystem management with stressors able to have cumulative, synergistic or antagonistic effects in different situations (Crain et al. 2008; Halpern et al. 2008a). Previous research on cumulative stress in marine systems has found that stressors often act synergistically resulting in outcomes that are worse than for single stressors alone (Crain et al. 2008). Crain et al. (2008) also found that the majority of studies assessing stressors in marine ecosystems were conducted in lab-based experiments (73 %) while less than 10 % were conducted in the field. They also highlighted large gaps in the research for particular stressors including fishing impacts (Crain et al. 2008). Without knowledge of how different potential stressors affect fish assemblages but also might interact together, it can be difficult to properly manage areas or fish populations.

Along with the development of a statewide marine park network (see Chapter 1 for more details), South Australia has been experiencing increased use of coastal areas due to mining and industrial expansion, shipping and port expansion, commercial and recreational fishing, aquaculture practices, and tourism activities such as shark-cage diving (Doubleday et al. 2017; Huveneers et al. 2017; Robbins et al. 2017). The diversity of potential anthropogenic stressors, their proximity to urban centres, and the relative understudied nature of many areas within South Australia has made it an ideal place to tease apart the influence of such stressors on fish assemblages.

Baited Remote Underwater Video Stations (BRUVS) are becoming a common way to investigate fish assemblages, including the influence of stressors upon them (Whitmarsh et al. 2017). Previous studies have used BRUVS to assess the effects of marine protected areas (e.g. Kelaher et al. 2014; Whitmarsh et al. 2014), fishing or aquaculture impacts (e.g. Lindfield et al. 2014; Tanner and Williams 2015), urbanisation

(e.g. Vargas-Fonseca et al. 2016), and offshore wind farms (e.g. Griffin et al. 2016); but see Chapter 2 for a more thorough review. The nature of BRUVS as a method has made it an ideal tool to assess fish assemblages in many scenarios because it is non-extractive, easily repeatable, can be used in a range of depths and habitat types, and can sample a large proportion of nektonic species.

The aim of this chapter is to assess the impact of various anthropogenic influences on South Australian fish assemblages using BRUVS. I plan to do this through four distinct case studies each focusing on different areas and stressors. These case studies are: 1) the influence of newly-implemented sanctuary zones on the fish assemblages observed in seagrass and soft-sediment habitats; 2) the influence of desalination brine effluent on fish assemblages near metropolitan Adelaide; 3) the influence on fish assemblages of bait and berley input from shark-cage diving at the Neptune Islands, South Australia; and 4) the influence of recreational fishing pressure, aquacultural practices, and proximity to effluent (run-off, groundwater, and stormwater) sources on fish assemblages within Coffin Bay, South Australia.

Case study #1

Marine protected areas of the Upper Gulf St Vincent: Choose your zone adventure

Background

There has been widespread general acceptance, and proliferation, of marine protected areas (MPA; specifically no-take or sanctuary zones) worldwide in an attempt to conserve marine biodiversity (Halpern and Warner 2002; Lester et al. 2009; Stewart et al. 2009; Lubchenco and Grorud-Colvert 2015). However, despite studies demonstrating strong ecological benefits from the protection provided by MPAs (Lester et al. 2009; Edgar et al. 2014; Roberts et al. 2017), there has also been concern that many MPAs, through ill design, either fail to meet their own objectives, provide no benefits for marine ecosystems, or exist merely as 'paper parks' (Agardy et al. 2003; Chaigneau and Brown 2016; Pendleton et al. 2017).

The number of MPAs worldwide has been steadily increasing since the 1970s, with a large increase in the coverage of protected areas in the last decade (Lubchenco and Grorud-Colvert 2015). Governments are under increasing pressure to justify their policies by attempting to prove the value of MPAs over short timescales such as those aligning with election cycles (e.g. 3 or 4 years). It is therefore important to consider what initial benefits protection may have and to specify the timeframe over which changes are expected to occur (Claudet et al. 2008; Vandeperre et al. 2011; Edgar et al. 2014). Most studies have focused on investigating the effects of protection over longer (> 3–10 years) timescales (e.g. Willis 2001; Stobart et al. 2009; Malcolm et al. 2015) and found that the changes increase with time since protection (Halpern and Warner 2002; Claudet et al. 2008; Vandeperre et al. 2011). However, contention still exists about the timeframe over which changes become observable and which species are most likely to be affected first (Russ and Alcala 2004; Babcock et al. 2010).

The expected effects from protection will depend on factors like the species home range sizes, life-history characteristics, and habitat preferences (Kramer and Chapman 1999; Molloy et al. 2009). While some species can increase in abundance and size under protection (Halpern and Warner 2002; Lester et al. 2009; Malcolm et al. 2015), others may decline or not be affected (e.g. McLaren et al. 2015). Depending on the mobility of adults and the reproductive patterns of species, those benefitting from protection may exhibit spillover, where adjacent areas also show benefits despite not being protected (McClanahan 2000; Halpern et al. 2009b; Stobart et al. 2009).

With South Australia having recently implemented a network of new MPAs (fully enforced since October 2014; see Chapter 1 for more details), it is an ideal location to investigate the initial effects of protection and also has the benefit of providing MPAs in less-well-studied habitats than reefs, such as shallow seagrass areas. Thus, the aim of this study was to assess and compare newly-implemented no-take areas to nearby control areas in shallow seagrass habitats. I hypothesise that there will be differences between protected and unprotected sites with an increase in the abundance of targeted species in protected sites compared to unprotected sites. This effect should strengthen over time with greater differences observed in later years of sampling compared to earlier ones. Individual areas may also respond differently to protection depending on

factors such as the assemblage present, previous fishing intensity, likelihood of illegal fishing activity, and the adjacent protection types (zones).

Methods

Study sites

Sampling took place in Upper Gulf St Vincent, South Australia. Gulf St Vincent is a large, inverse estuary of relatively shallow depth (<30 m). At the top (north) of the gulf, salinities can be as high as 42 in the summer months, with sea surface temperatures over 22°C (de Silva Samarasinghe et al. 2003). Two no-take MPAs (hereafter referred to as sanctuary zones or SZ) located in the 'Upper GSV Marine Park' were selected for this study: Clinton Wetlands SZ-1 and Upper Gulf St Vincent SZ-2 (Figure 4.1). Both areas are shallow seagrass habitat with Clinton Wetlands SZ being slightly shallower with an approximate depth of 3 m (referred hereafter as Area 1), while Upper Gulf St Vincent SZ is deeper at 7 m (referred to as Area 2). Two sites were selected to be studied within each SZ along with two paired control sites outside of the MPAs resulting in a total of eight sites (Figure 4.1). The control sites were situated in Habitat Protection Zones (HPZ), which allow most fishing but are prohibitive of activities which may damage the benthos (e.g. trawling). These areas had not been sampled prior to marine-park implementation and enforcement.

Field sampling

This study started in the austral summer (January 2015) three months after enforcement began and was repeated one year later (January 2016). Stereo-BRUVS (Harvey and Shortis 1995) were used to observe the fish assemblages to record abundance and diversity. Each BRUVS unit consisted of two Panasonic HC-V700 cameras set to record in 1080p in 50 fps with a wide field of view. At each site, six replicate deployments were taken but, due to weather and logistical issues, only Area 2 could be resampled in 2016. One control site within Area 2 was also able to have only four replicates taken due to inclement weather. Thus, the total number of replicates was $N = 68$. BRUVS were deployed during daylight hours for 60 min on the seafloor before retrieval and were baited with 500 g of minced sardines (*Sardinops sagax*). BRUVS were spaced at least 250 m apart (Malcolm et al. 2007).

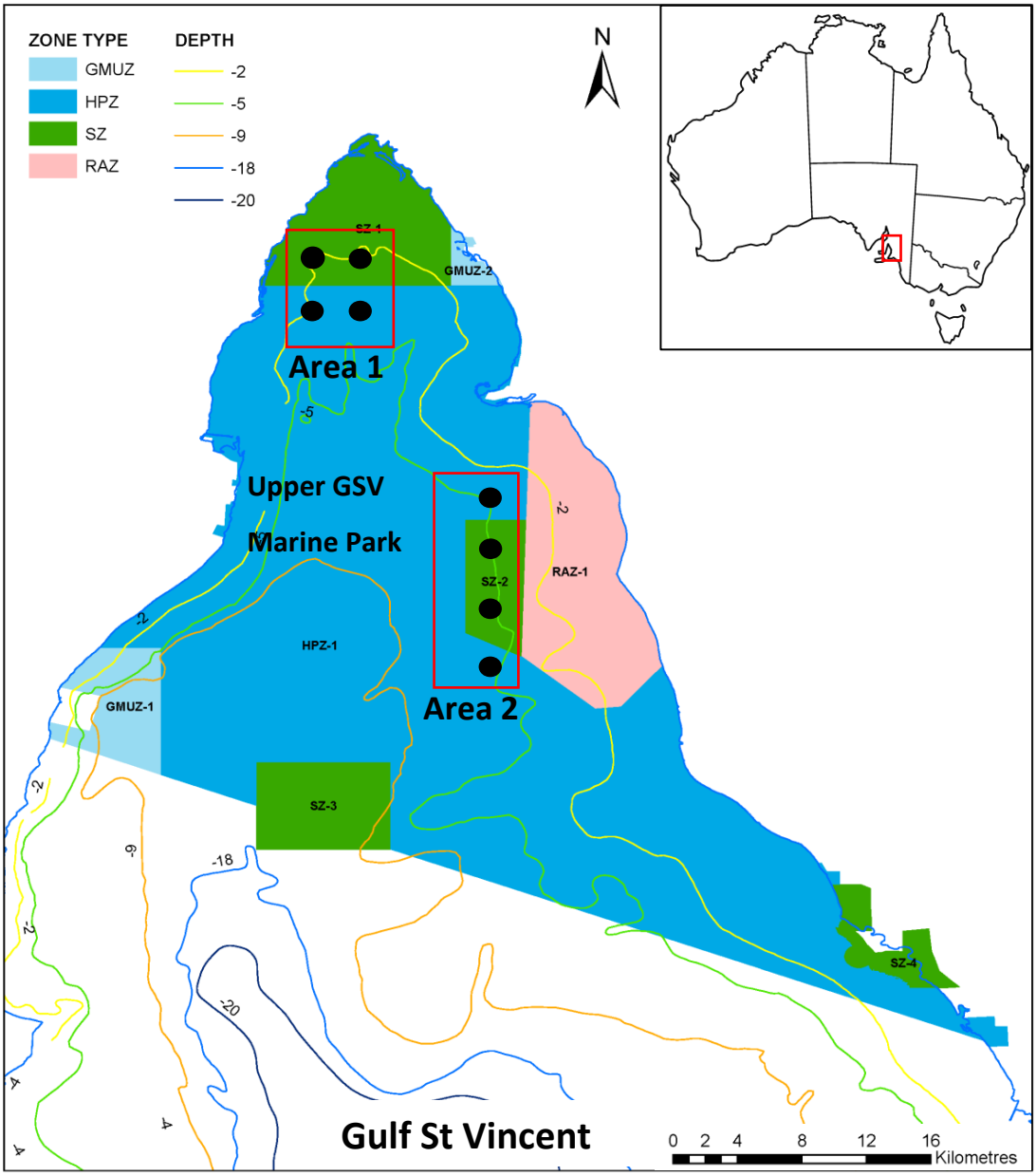

Figure 4.1: Map showing the Upper Gulf St Vincent (GSV) Marine Park with the areas sampled shown by the red rectangles and the sampling sites shown by black dots. Different protection levels are shown with the lowest protection being General Managed Use Zone (GMUZ), followed by Habitat Protection Zone (HPZ), with Sanctuary Zone (SZ) and Restricted Access Zones (RAZ) having the highest level of no-take protection. Bathymetry in metres is also displayed by the coloured contour lines. Source: Department of Environment, Water and Natural Resources.

Video processing

Videos were analysed using the specialised SeaGIS *EventMeasure* software (SeaGIS Pty. Ltd., Bacchus Marsh, Victoria, Australia; www.seagis.com.au/event.html). On each replicate, taxa were identified to the finest taxonomic level possible (Kuiter 1996; Gomon et al. 2008) and counted using the relative abundance measure, *MaxN*. *MaxN* is the maximum number of individual fish (for each species or taxon) observed in a single frame throughout the deployment duration. *MaxN* is thus a conservative estimate of abundance, particularly where large numbers of fish are present or a lot of turnover of individuals occurs during the video (Priede et al. 1994; Ellis and DeMartini 1995; Willis et al. 2000). Due to issues with water quality, fish length measurements were unable to be taken for some replicates and were thus excluded from all analysis. These visibility issues resulted in the loss of the extremities of some species which are critical for accurate measurements to be taken. The visibility did not present many difficulties for the identification of species, as the nature of the video footage allowed for movement and key identifiable features (e.g. General Impression of Size and Shape or GISS; O'Brien et al. 2006) to be easily recognised for the relatively limited assemblage (28 teleost species) observed. Most species were easily recognisable but, if taxa were not able to be reliably identified to species level, then they were grouped into genus or family, e.g. two trevally species could not be so differentiated and thus were grouped into *Pseudocaranx* spp.

Statistical analysis

Statistical analyses were conducted in PRIMER v7 (Clarke and Gorley 2015) and PERMANOVA+ (Anderson et al. 2008). A total of 68 replicates were used for the analysis, 35 from protected sites and 33 from unprotected sites.

Analysis was undertaken on two separate but overlapping data subsets as the lack of re-sampling Area 1 in 2016 prevented testing for the effects of Year and Area simultaneously. Univariate total abundance per replicate (all species summed) were analysed using PERMANOVA with a Euclidean resemblance matrix to test for differences using the two data subsets. The first data subset was those samples from Area 2 only and allowed for testing the factors Year (fixed factor, two levels) and Protection status (fixed factor, two levels). The second data subset was selected using

only 2015 data and tested for differences between Area (fixed factor, two levels), and Protection status (fixed factor, two levels).

Similar PERMANOVA analyses were done using the multivariate assemblage data with a Bray-Curtis resemblance matrix. Before the matrix was constructed, abundance data were transformed using dispersion weighting by site. Dispersion weighting is used to accommodate the schooling nature of certain fish species dominating calculations (Clarke et al. 2006a). For significant factors, pairwise tests were used to further discern where differences occurred. The discriminant function test of Canonical Analysis of Principal coordinates (CAP) was also used on the full data set to test for differences due solely to a composite factor between Year, Area, and Protection, and the CAP constrained-ordination plot was used to visualise differences among such groups. Allocation success rates were also used to assess how well the samples fit each group. SIMilarity PERcentage (SIMPER) analyses were used to determine those species contributing most to similarities within protection types and those which best indicated differences among them.

Species were grouped into categories based on fishing status. Species which are primarily targeted by recreational or commercial fishers were classified as 'targeted', species less-commonly targeted or those caught as by-catch but typically kept by fishers were grouped into the 'by-product' category, while those species that are not targeted by fishers and typically released or discarded were classified as 'not-sought' (Appendix 2A, Table 7.3). Species abundance within each group was analysed using the same univariate analysis as the total abundance data described above but with a simplified PERMANOVA design assessing only for differences with Protection Status in order to maximise power for the analysis. This design was used as the interactions between Year and Area with Protection were not-significant. Each group was also analysed using SIMPER to determine species characterising each fisheries category and protection status.

Results

A total of 2,603 individuals were observed across 38 taxa (Appendix 2A,Table 7.3) for both years sampled; this included six chondrichthyans, one cephalopod, and three

crustaceans, with the remaining 28 taxa comprising teleost fish species. The average number of individuals seen per replicate was 40.0 (± 5.3) for 2015 and 34.5 (± 7.0) for 2016. Similar numbers of individuals were observed between protected and unprotected sites for both years combined (Protected = 39.8 ± 4.2 vs. Unprotected = 36.6 ± 7.6), with no significant difference between the total number of individuals for Year and Protection, or Area and Protection (all PERMANOVA *p* values > 0.696), as well as their respective interactions.

The overall fish assemblage differed significantly between Areas and Protection (for 2015 data) as well as between Years and Protection (for Area 2 data), with significant interactions between Area and Protection (Table 4.1). Although there were no differences in fish assemblages between protected and unprotected sites in Area 1, fish assemblages varied by protection status in Area 2 (Table 4.1). Distinct separation between Areas and Years can be observed via CAP (Figure 4.2, trace and delta statistics $p = 0.001$) and allocation success rate was high overall at 70.5 % correct; however, the unprotected sites in Area 2 had the highest misclassification rates for a group at 50 % each. Replicates from 2015 in the unprotected Area 2 were misclassified to the corresponding protected site (3 replicates misclassified), the 2016 unprotected Area 2 site (1 replicate) and the 2016 protected Area 2 site (2 replicates), while the replicates from 2016 in the unprotected Area 2 were misclassified to the corresponding protected site (3 replicates) and the 2015 unprotected Area 2 site (2 replicates).

The species contributing most to the similarities between groups varied by Area and Year, with Area 1 characterised by toadfish *Torquigener pleurogramma* and trumpeter *Pelates octolineatus*, and Area 2 characterised by blue swimmer crab *Portunus armatus,* Port Jackson shark *Heterodontus portusjacksoni,* and rough leatherjacket *Scobinichthys granulatus* (Figure 4.2; Appendix 2A, Table 7.4). Further analysis of the SIMPER results showed that at both areas in 2015 eight species were collectively contributing 70 % of the dissimilarity between protected and unprotected sites, compared to 11 species in 2016 (Table 4.2). Six species were common contributors to both years but only two species (*Neoodax balteatus* and *Torquigener pleurogramma*) showed different trends between protection status and years (Table 4.2).

Table 4.1: PERMANOVA results for the multivariate fish assemblage data showing the two approaches based on year or area along with the pairwise tests for any significant interactions. Unique permutations ranged from 996 to 999 per test. Significant values are shown in bold.

Data subset	Source	df	MS	Pseudo-*F*	*p*(perm)	Protected vs. unprotected		
						Group	*t*	*p*(perm)
2015 only	**Area**	1	37185	28.95	**0.001**			
	Protection	1	4284.1	3.34	**0.001**			
	Ar x Pr	1	3172.7	2.47	**0.009**	**Area 1**	1.66	0.053
	Res	42	1284.2			**Area 2**	1.72	**0.003**
Area 2 only	**Year**	1	9818.7	6.36	**0.001**			
	Protection	1	5907.4	3.82	**0.001**			
	Ye X Pr	1	2487.7	1.61	0.078			
	Res	41	1544.8					

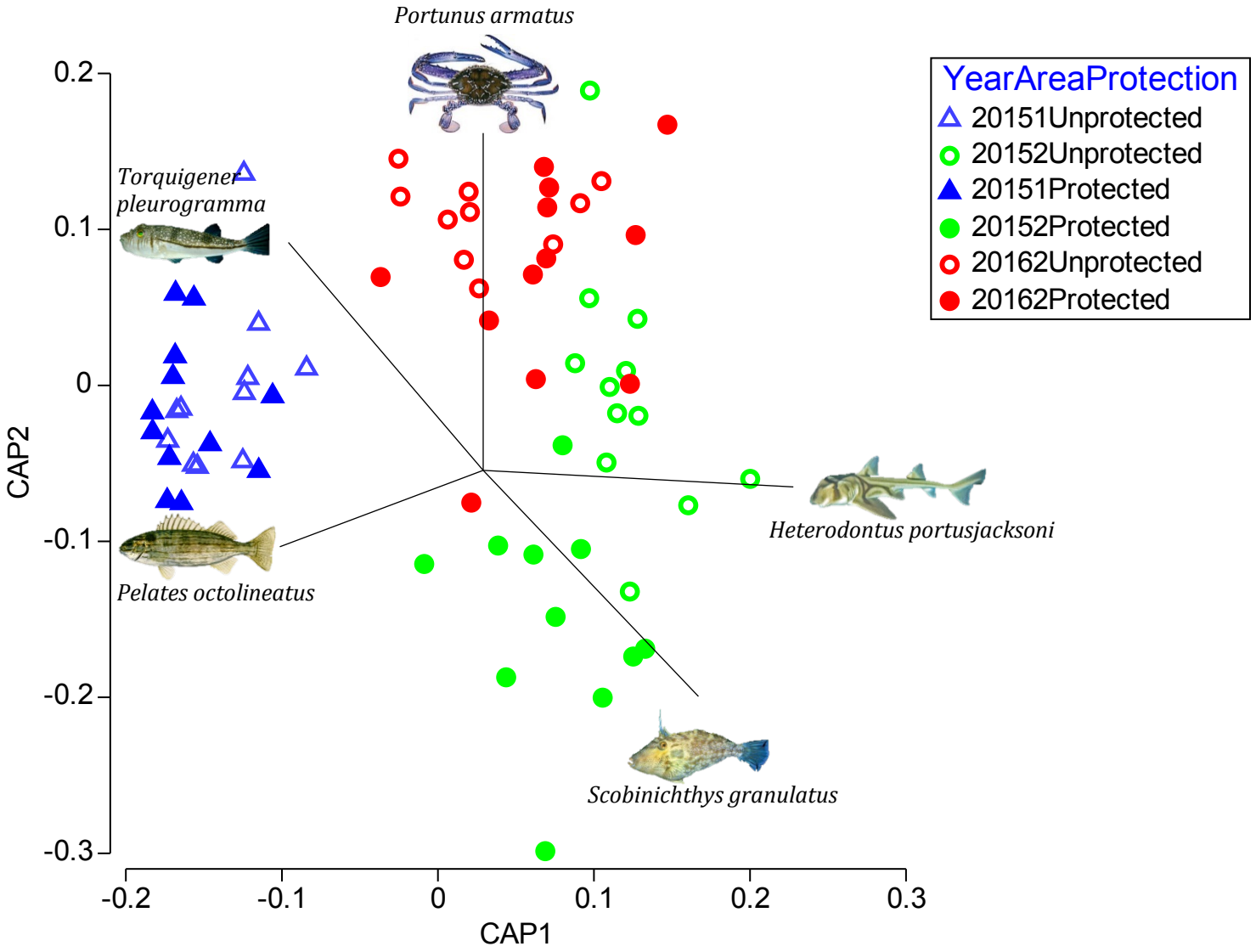

Figure 4.2: CAP constrained-ordination plot showing the first two PCO axes (out of *m* = 9) for the combined factor of Year, Area and Protection. Fish pictogram (Appendix 2A, Table 7.3) overlays show the dominant species contributing the most to the similarity for each group according to SIMPER analysis (see Appendix 4A, Table 7.4 for a full list).

Table 4.2: SIMPER results for the dissimilarity between protected and unprotected zones for both 2015 and 2016. Diss/SD shows the dissimilarity divided by the standard deviation and values above 1 are thought to be consistent indicators and hence are shown in bold. % contribution is the percent in which that species contributes to the overall dissimilarity between groups. The average dissimilarity between groups in 2015 was 64 % and for 2016 was 51 %.

Species	**Fisheries group**	**Average abundance**			**Average dissimilarity**	**Diss/SD**	**% contribution**
		Unprotected		**Protected**			
2015							
Scobinichthys granulatus	By-product	1.25	<	2.18	7.36	**1.34**	11.52
Portunus armatus	Targeted	1.58	>	0.82	5.12	**1.46**	8.02
Heterodontus portusjacksoni	Not-sought	1.83	>	2.09	5.07	**1.31**	7.95
Neoodax balteatus	Not-sought	1.67	~	1.64	5.00	**1.12**	7.83
Acanthaluteres spilomelanurus	By-product	1.05	>	0.68	4.95	0.98	7.76
Pelates octolineatus	By-product	0.16	<	1.14	4.16	**1.11**	6.51
Chrysophrys auratus	Targeted	0.05	<	1.05	3.85	0.78	6.03
Torquigener pleurogramma	Not-sought	0.68	<	0.81	3.64	**1.03**	5.70
Sillaginodes punctatus	Targeted	0.66	>	0.07	2.59	0.57	4.05
Meuschenia scaber	By-product	0.56	>	0.15	2.55	0.73	3.99
Sepioteuthis australis	Targeted	0.25	<	0.55	2.39	0.80	3.74
2016							
Pelates octolineatus	By-product	1.41	<	2.24	8.31	**1.22**	16.27
Portunus armatus	Targeted	3.60	>	3.17	5.49	**1.16**	10.74
Neoodax balteatus	Not-sought	1.00	<	1.33	4.65	**1.13**	9.11
Torquigener pleurogramma	Not-sought	1.23	>	0.17	4.45	0.95	8.71
Heterodontus portusjacksoni	Not-sought	1.70	<	2.42	3.97	**1.06**	7.77
Scobinichthys granulatus	By-product	0.50	<	1.33	3.96	**1.03**	7.76
Upeneichthys vlamingii	By-product	0.90	>	0.33	3.44	0.82	6.74
Arripis georgianus	Targeted	0.08	<	0.61	2.38	0.78	4.66

Of the 38 taxa observed, we classified nine as targeted species, 17 as by-product species, and 12 as not-sought species (Appendix 2A, Table 7.3), equating to 13 %, 55 %, and 32 %, respectively, of the total individuals observed. The mean number of individuals per replicate ranged from 5.1 ± 0.6 for targeted species to 26.6 ± 3.3 for by-product species (Figure 4.3) with significant differences in abundance observed only for by-product species between protected and unprotected sites (Pseudo-F = 8.3, p = 0.014), but not for targeted or not-sought species (p > 0.257; Figure 4.3). Trumpeter *P. octolineatus*, contributed most (43 %) to the dissimilarity between the two levels of protection status for by-product species (with higher abundance in protected sites), above *S. granulatus* (17.5 %; higher abundance in protected sites), red mullet *Upeneichthys vlamingii* (9.2 %; higher abundance in unprotected sites), and bridled leatherjacket *Acanthaluteres spilomelanurus* (8.0 %; higher abundance in unprotected sites).

Synthesis

My results showed that some places exhibited differences between protected and unprotected areas despite the young age of the SZ involved, supporting my main hypothesis. However, the species mostly driving this difference were not species that are typically targeted commercially or recreationally. The effects of protection also varied between areas, with one location showing no significant differences between protected and unprotected sites for the first year of sampling compared to large differences in fish assemblages between protection levels for Area 2 in both years. The species being affected by protection also changed over time (see Figure 4.2) but effects were limited for targeted species even during the second year of sampling. Overall, this supports my third hypothesis that the effect of protection varies according to fish assemblages or location differences e.g. previous fishing intensity, adjacent protection types, and likelihood of illegal fishing. In general, however, it is still unclear whether the effects observed are attributable to protection or are just differences inherent between the SZ and HPZ sites tested. Without comprehensive before data, which is lacking for these areas, it is difficult to tease apart such factors. The data collected by this study can, however, be used as baseline for future studies to continue monitoring the effects of protection for these areas, particularly due to the sampling being conducted soon (~ three months) after enforcement began.

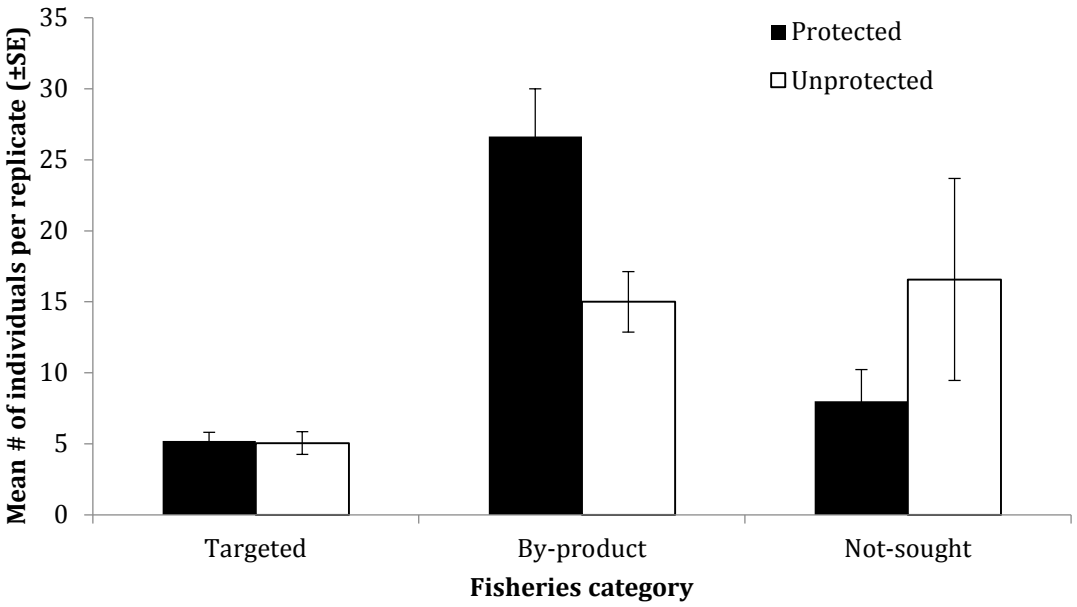

Figure 4.3: Mean (± SE) number of individuals per replicate for each category of species according to their fisheries status for both protected and unprotected areas. Total *N* = 68.

Contrary to my expectation, by-product species drove most of the differences between protected and unprotected sites, although similar results have been observed in other studies (e.g., McLaren et al. 2015). McLaren et al. (2015) assessed the effects of several small sanctuary zones (<2 km^2) in Western Australia and found that they were most beneficial to small-bodied by-catch species because those species typically had small home ranges wholly within the confines of the sanctuary zone. Large-bodied, commonly-targeted fish species showed little difference as they were more likely to move outside the boundaries of the protected zones (McLaren et al. 2015). Although the sanctuary zones assessed in the present study are moderately sized (approximately 23–45 km^2) and larger than the sanctuary zones studied in McLaren et al (2015), the similar results might be due to the differences between habitat types studied. Fish species that inhabit seagrass areas are likely to have larger home ranges than those from reef areas, such as the ones studied by McLaren et al. (2015), which may explain the lack of difference between protection levels for targeted species even though the SZ in SA are moderately sized. Not-sought species also showed some differences between zones, but were not significant due to large variations in abundances. It is likely that the young age of the sanctuary zones assessed in the present study also contributed to the non-significant outcome for targeted species.

There was a distinctive change in the assemblages observed for Area 2 between the years sampled. In particular, the species driving the differences between protected and unprotected sites changed. There were more targeted species contributing to the differences between protected and unprotected sites (e.g. snapper *Chrysophrys auratus*, King George whiting *Sillaginodes punctatus*, and southern calamari squid *Sepioteuthis australis*) in 2015 than in 2016. *Chyrsophrys auratus* and *Se. australis* showed the typical pattern of higher abundance in protected sites but the opposite trend was observed for the highly valued *Si. punctatus*. Snapper species have been shown to benefit greatly from protection in other areas (e.g. Denny et al. 2004; Malcolm et al. 2015), so the decline in abundance between 2015 and 2016 may have been a result of stock declines that were shown to decrease between 2015 and 2016 for the upper Gulf St Vincent (Steer et al. 2018). Other reports have shown a mixed response of *Si. punctatus* to protection status with other seagrass SZ showing higher abundances in unprotected areas compared to protected but this trend was not consistent across years (DEWNR

2017). Differences revealed in fish assemblages could also be attributed to variation in life-history characteristics among these species. Squid *Se. australis* form mating aggregations in which internal fertilisation of eggs occur, while whiting *Si. punctatus* and snapper *C. auratus* also form schools, but are serial spawners and thus, probably have greater dispersal capabilities than *Se. australis*. The localisation and rapid growth of *Se. australis* may be favourable for taking advantage of small and recently-declared protected areas, which may take time to be colonised by species which have longer maturation and larval dispersal phases. The use of seagrass areas by predominantly juvenile *Si. punctatus* (Jenkins and Wheatley 1998), may also have been contributing to the differences observed with protection from fishing affecting adult fish more often than juveniles.

The observed differences between protected and unprotected sites for Area 2 but not Area 1 may be due to the assemblages inherent within the area or specific location effects. There were distinct assemblages observed between Area 1 and Area 2, with Area 1 having fewer species contributing to such differences (see Appendix 2B, Table 7.4). It is possible these species observed were less likely to benefit from protection or that this area may take longer to show effects from protection compared to Area 2. The two main species contributing to differences for Area 1 were trumpeter *P. octolineatus* and toadfish *T. pleurogramma* categorised as a by-product and not-sought species, respectively. While by-product species showed significant differences between protection levels for Area 2, it is unclear why these differences were not observed for Area 1. Considering the result for Area 1 was close to being significant, this area may have showed more distinct differences between protected and unprotected sites if it was also sampled in 2016. Locational differences may also have contributed to the different outcome for each Area. The Area 2 SZ is adjacent to another long-standing protected area (see RAZ-1 in Figure 4.1) that was established by the military in the 1940s as a restricted access (no-go) zone prohibiting not only removal of organisms but also any entry to the area. The close proximity of an enforced, long-standing highly-protected zone might have led to some spillover benefits to the adjacent SZ in Area 2. Although spillover can contribute to increases in abundance of species responding positively to protection up to 800 m outside the zone boundary (Halpern et al. 2009b; Stobart et al. 2009), other studies have found at best limited evidence of this occurring

(e.g. Edgar et al. 2004). Thus, future research investigating fish movement and larvae dispersal regimes are needed to test whether spillover is occurring. Similarly, before the implementation of the MPA, Area 1 was subject to commercial and recreational fishing efforts more so than Area 2. This area was also the subject of considerable debate amongst stakeholders in relation to the placement of the SZ during the development of SA's Marine Park network (Kosturjak et al. 2015). A lack of community support for this SZ is likely to drive illegal fishing within the sanctuary (Pita et al. 2011). Considering the proximity of this SZ to the nearest boat ramp and township (Port Wakefield), this area may experience more illegal fishing activity than Area 2, which is further away and is more widely accepted by the community.

To further assess the implications of protection on these areas, it is crucial to continue monitoring over longer time periods. Sampling could perhaps also be timed to coincide with better water visibility to allow the measurement of fish lengths. This metric would be an important indicator of the demographic effects of protection on fish assemblages as many studies show an increase in lengths and hence biomass as a result of protection (Edgar and Stuart-Smith 2009; Lester et al. 2009; Watson et al. 2009). The likely effects of protection on shallow seagrass communities in this region could also be studied by comparing the newly-formed SZ to those that are longer-standing such as the nearby RAZ. Sampling was planned for this zone but could not be undertaken due to permitting restrictions. If sampling could be undertaken within the RAZ then it may prove beneficial to know what impacts protection may have in shallow seagrass areas and how the new protection zones may respond.

Conclusion

Overall, these results highlight the need for prior baseline data to ensure any differences observed in future MPAs can be attributable to protection and not inherent differences between impact and control areas. We did not find an expected increase in the abundance of all targeted species in protected sites but have shown that protection may benefit species other than those directly targeted by fishing. Studies that only focus on targeted species may miss key results and fail to show benefits that protection may offer. These results can now provide the foundation for future studies to better assess the influence on protection on these areas and assess future changes.

Case study #2

Effects of effluent output on fish assemblages: Below the brine

Background

Rising human population and anthropogenic activity has resulted in an increased number of urban, agricultural, and industrial effluents (McKinley and Johnston 2010). The sources of effluent vary from wastewater (including sewage) treatment plants, stormwater drains, diffuse agricultural run-off, and industrial outfalls such as from power or desalination plants. The effluents can have notable effects on marine ecosystems including increased nutrient loads, or turbidity, salinity and heavy metal concentrations, which can lead to eutrophication, anoxic conditions, habitat loss, loss of ecosystem function, and reduced ecosystem health (Islam and Tanaka 2004). These impacts can affect all levels of the ecosystem from the benthos to large nektonic (transitory) animals such as marine mammals (Islam and Tanaka 2004). For fish assemblages, effluent output has been shown to cause changed reproductive physiology, bioaccumulation of heavy metals, reduced fish health, and decreased abundance and diversity (Neuman and Karås 1988; Islam and Tanaka 2004; Vajda et al. 2008; McKinley and Johnston 2010; Roberts et al. 2010).

Coastal desalination plants are becoming common as a solution to mitigate freshwater shortages (Roberts et al. 2010). Desalination plants convert water taken from nearby marine sources to freshwater using either membranes or thermal methods. The concentrated brine that results from this process is then typically released back into the ocean (El Saliby et al. 2009). Desalination plants exist in many countries, but are particularly prevalent in the arid Middle East and continue to be built in Europe, USA, and Australia to combat water shortages and ensure water security in the future (Roberts et al. 2010). Previous studies about the effects of desalination brine on marine life have found decreases in echinoderm, coral, plankton, and fish abundances, along with decreases in infaunal and sessile invertebrate assemblages (Roberts et al. 2010). These decreases may have been a result of the increased salinity, temperature, or concentration of anti-fouling compounds such as copper, although there also exists some contention about the validity of some results due to a lack of methodological detail

and/or experimental design rigor (Roberts et al. 2010). Advances in technology have, however, also reduced the risks of impacts from newer plants through the use of more effective plume dispersal engineering, reduced emission of anti-fouling compounds, and lessened effects of temperature change when using membrane methods for extraction rather than thermal ones (Lattemann and Höpner 2008; El Saliby et al. 2009; Roberts et al. 2010).

South Australia recently built a 100 GL/year capacity desalination plant along the Adelaide metropolitan coastline that has been operational since 2011 (Kämpf and Clarke 2013). This plant was built in response to Adelaide's growing water needs during the last drought (Dijk et al. 2013) and underwent rigorous planning and design to ensure minimal expected environmental impact (SA Water 2008). The plant uses reverse osmosis processes to convert saltwater to freshwater by passing it through a membrane. The plant sits 20 km south of the Adelaide city centre on the shore of Gulf St Vincent, South Australia with the intake pipe and outfall diffuser at 1.5 and 2 km from shore, respectively. To adequately disperse the brine plume at all discharge volumes, an artificial structure housing state-of-the-art 'duck-billed' diffusers was constructed, which in addition to the intake pipe increased the available reef habitat of the area (SA Water 2008). To prevent vessels from anchoring or from fish gear entangling on these structures, an exclusion zone (covering approximately 0.5 km^2) was placed around the area. As part of a government-mandated licensing agreement, environmental impact assessments must be conducted, with fish assemblages currently requiring monitoring every three years. Previous assessments of the fish assemblages showed minimal changes around the effluent output, but some of these results were debated because of the possible inadequacy of the chosen reference (no impact) sites (Barbara 2016). As a result, a new sampling design was used for the latest round of assessments in 2015, which forms the basis of this case study. Along with the brine discharge, the outfall area also exhibits other factors that may influence fish assemblages such as the artificial nature of the physical outfall-pipe structure and the protected status of the area.

Structures built to support outfalls form artificial reefs on otherwise soft bottoms with seagrass and algae. Some artificial structures have been shown to have higher fish abundance and diversity at artificial reefs compared to natural reefs (Folpp et al. 2013),

while other studies showed the opposite effect (Carr and Hixon 1997). It can often be difficult to find directly comparable areas for artificial reefs due to differences in reef age, structure, profile, and habitat characteristics (e.g. depth and exposure). Due to the exclusion zone surrounding this area, fish assemblages near the artificial structures may also be influenced by the protection afforded from the exclusion zone. More information about the effect of protection on fish assemblages is provided in Case study # 1.

The aim of this case study is to determine the effects of the desalination brine output and infrastructure on the surrounding fish assemblages. Specifically, I shall compare the outfall area to four control areas with two of these areas being artificial and two natural. One of the selected natural reef areas is also protected for comparison to the protected outfall area. No artificial protected area was able to be compared as there were not any of a similar size, depth, age, and material found within the geographic area that would allow for a valid comparison. Thus, I am able to compare between impacted and control sites, artificial and natural reef types, and between protected and unprotected sites, and assess relative changes in fish assemblages across these comparisons. My hypothesis was that the difference between impact and control sites would be greater than in the other between-site comparisons if effluent were having a significant effect on the fish assemblages.

Methods

Sampling sites

All five sites were located within Gulf St Vincent, South Australia (Figure 4.4). The Adelaide Desalination Plant (ADP) site is the impacted site and is located within the existing exclusion area (established 2009) that contains the intake and outfall pipes. The inlet and outlet areas provide an artificial reef-like substratum within a predominantly sand/seagrass habitat that is approximately 20 m deep. The control sites consist of the remaining four sites: (1) Port Noarlunga Reef is a natural high-profile rocky reef that has been protected from fishing since 1971. With the reef structure situated parallel to the coastline in approximately 10–20 m of water, it is a popular area for recreational activities such as swimming, snorkelling, and diving; (2) Noarlunga Tyre Reef is an artificial reef constructed from pyramid-shaped structures made out of car tyres that were placed at a depth of approximately 18 m alongside these tyre

structures also exists two wrecks, the Seawolf and the Lumb (the most recent addition scuttled in 2002), the surrounding habitat is soft-sediment; 3) Seacliff Reef is a lower-profile natural reef, situated in approximately 10 m of water surrounded by seagrass and soft-sediment habitats; and (4) Glenelg Tyre Reef is constructed from similar material to the Noarlunga Tyre Reef, is situated in approximately 20 m of water and was built around 1980. All Noarlunga Tyre Reef, Seacliff Reef, and the Glenelg Tyre Reef are popular recreational fishing sites. All sites were within 13 km of the ADP. Sites were further classified by the geographical distribution with southern sites being the ADP, Port Noarlunga Reef and Noarlunga Tyre Reef all located to the south of the coastal point at Hallett Cove. Northern sites were Glenelg Tyre Reef and Seacliff Reef, located north of Hallett Cove.

BRUVS deployments

Fish assemblages were investigated using Baited Remote Underwater Video Systems (BRUVS). BRUVS were set up with high-definition GoPro Hero 3+ Silver video cameras on metallic frames. These cameras were selected due to their relative low cost, ability to record in high definition, long battery life, wide-angle viewing, and image quality in low light conditions. Single, horizontal set-ups were used because fish length measurement was not required for this study.

The units were baited with 500 g of crushed sardines (*Sardinops sagax*). Six replicates were performed at each site. A minimum of 50 m separated each deployment but 67 % of the deployments were separated by at least 100 m to reduce the likelihood of bait plume interactions between units deployed concurrently. Attempts were made to optimise distances between deployments while still being close enough to the relevant reef structures under investigation. The small size of the reefs being investigated in this study hindered our ability to further space out replicates. Further reductions in bait plume overlap were achieved by only deploying three simultaneous replicates at any one site on each day. Units were set to continuous recording and deployed for a minimum of one hour before retrieval. Videos were processed as per Case study #1.

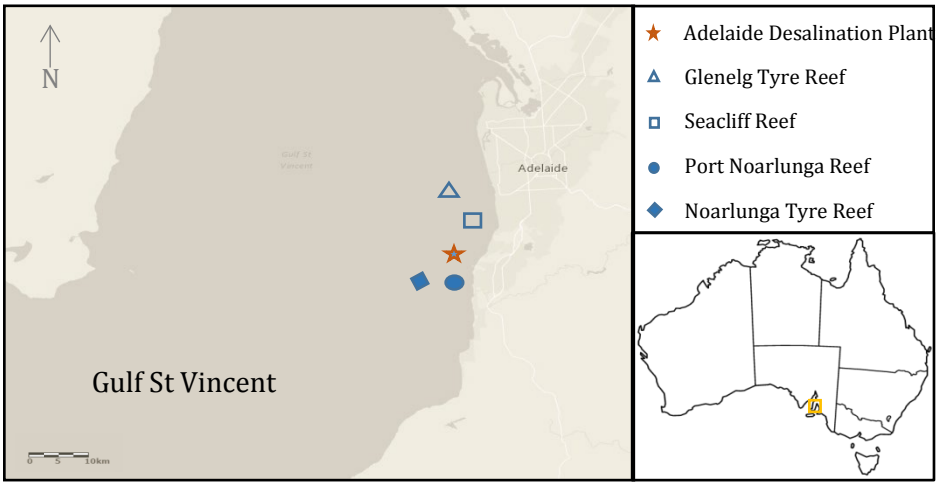

Figure 4.4: A map of the study sites within Gulf St Vincent, South Australia, showing the impact site, Adelaide Desalination Plant, as a red star and the control sites in blue. Northern sites are indicated by unfilled symbols, while southern sites are filled.

Data analysis

Statistical analyses were conducted in PRIMER v7 (Clarke and Gorley 2015) and PERMANOVA+ (Anderson et al. 2008). A total of 60 replicate deployments were available for analysis, with 30 each from the spring and autumn seasons. Univariate total abundances per replicate were used to test for differences between seasons and sites with the following design, Season (fixed factor, two levels) and Site (fixed factor, five levels).

Multivariate data were transformed using dispersion weighting by site to account for the variable schooling nature of some fish species (Clarke et al. 2006a). To test for differences in assemblages between sites and seasons, the same PERMANOVA design was used as above with the Bray-Curtis resemblance measure between samples. Pairwise tests were used to further investigate significant factors. SIMilarity PERcentage (SIMPER) analyses were used to determine the similarity between groups and which species were driving any observed differences. Distance to centroids for each pair of sites was calculated and then averaged for those comparing ADP to other sites and then for the rest of the site comparisons which did not involve the ADP. Bootstrap averages (run 100 times) were calculated and used to construct a non-metric MultiDimensional Scaling (MDS) ordination plot showing differences among sites within seasons. To further test for differences between assemblages for groups of sites, planned comparisons using PERMANOVA analysis were conducted on one vs the rest groupings (e.g. ADP vs Glenelg Tyre Reef, Noarlunga Tyre Reef, Seacliff Reef, and Port Noarlunga Reef).

To assess the influence of the factors Geographic location (north [two levels] vs south [three levels]), Type (artificial [three levels] vs natural [two levels]) and Protection (protected [two levels] vs unprotected [3 levels]), the discriminant function test of Canonical Analysis of Principal coordinates (CAP) was used. Allocation success rates were then compared to assess the discriminatory power of each factor.

Results

Sixty-three species and 6,918 individuals were observed across the five sites and two seasons. A broad range of taxa were observed including 53 teleosts, three

chondrichthyans, two cephalopods, and five decapod crustaceans. Spring and autumn surveys had similar numbers of species (57 vs 47, respectively) and broadly similar numbers of species were also seen at each site, ranging from 32 in Port Noarlunga to 40 at the ADP. The total number of fish per replicate ranged over an order of magnitude from 33 ± 13.7 in the spring for Seacliff to 300 ± 23.8 in the spring for Noarlunga Tyre Reef (Figure 4.5). The number of individuals varied between Season (Pseudo-F = 4.47, p = 0.037) and Site (Pseudo-F = 25.01, p = 0.001), with seasonal differences also changing between sites (Pseudo-F = 7.30, p = 0.001). Pairwise tests indicated that Noarlunga Tyre Reef was the only site driving the differences observed between Seasons (t = 6.65, p = 0.001) with the remaining sites not significantly different between seasons (p > 0.05; Figure 4.5). Spring had the higher number of individuals compared to autumn at Noarlunga Tyre Reef (300 ± 23.8 vs. 122 ±12.1; Figure 4.5).

Similarly to numbers of individuals, fish assemblages were significantly different between seasons (Pseudo-F = 4.98, p = 0.001) and sites (Pseudo-F = 7.71, p = 0.001), and so was the interaction between Season and Site (Pseudo-F = 3.18, p = 0.001). Comparisons between sites within season showed significant differences between all pairs of sites in spring and for all pairs in autumn, except ADP vs. Noarlunga Tyre Reef (Table 4.3). Dissimilarity values between sites supported the paired comparison and were often higher for spring than autumn with the highest dissimilarity values occurring between Port Noarlunga Reef and each of Noarlunga Tyre Reef, Glenelg, and Seacliff (Table 4.4). The sites most similar to the ADP (lowest dissimilarity) were the other two artificial reefs, Noarlunga Tyre Reef and Glenelg (Table 4.4), indicating substratum type was important in characterising fish assemblages. The degree of difference in fish assemblages between ADP and the control sites was consistent with the variation between the four control sites, with the average distance to centroids for pairs of sites being 48.6 for those involving the ADP and 50.8 for pairs not involving the ADP. This can also be visualised using a bootstrapped averages MDS plot, where the centroid and 95 % confidence ellipse for the ADP lies nearby to other control locations (Figure 4.6). Port Noarlunga Reef appears to be the least similar site compared to the others. Planned comparison analysis showed all singular sites were highly significantly different from the other sites combined (p = 0.001 in all cases).

Table 4.3: Pairwise PERMANOVA results from the interaction of the factors Season and Site showing the comparison between seasons. Values in bold indicate significant differences. Unique permutations ranged from 400 to 557 per pairwise test.

	Spring vs. Autumn	
Site	**t**	***p(perm)***
ADP	1.18	0.195
Glenelg	1.60	**0.028**
Noarlunga Tyre Reef	2.42	**0.003**
Port Noarlunga Reef	2.52	**0.008**
Seacliff	1.48	**0.012**

Table 4.4: Pairwise PERMANOVA results from the interaction of Season and Site showing the comparison between sites and the dissimilarity values from SIMPER. Values in bold indicate significant differences. Unique permutations ranged from 399 to 784 per pairwise test.

Groups	**Spring**			**Autumn**		
	t	***p(perm)***	**Dissimilarity %**	***t***	***p(perm)***	**Dissimilarity %**
ADP vs. Glenelg	2.23	**0.001**	78.3	2.05	**0.002**	75.2
ADP vs NTR	2.76	**0.004**	67.8	1.29	0.089	66.6
ADP vs. PNR	3.34	**0.005**	89.4	1.79	**0.008**	81.0
ADP vs. Seacliff	2.59	**0.004**	88.4	2.16	**0.001**	85.4
Glenelg vs. NTR	2.92	**0.002**	84.9	2.12	**0.011**	74.5
Glenelg vs. PNR	2.82	**0.001**	91.5	2.48	**0.004**	90.8
Glenelg vs. Seacliff	1.67	**0.003**	79.4	1.68	**0.013**	71.9
NTR vs. PNR	4.00	**0.003**	91.8	1.57	**0.019**	75.7
NTR vs. Seacliff	2.79	**0.001**	85.6	2.04	**0.005**	81.4
PNR vs. Seacliff	2.62	**0.003**	90.8	2.16	**0.006**	91.2

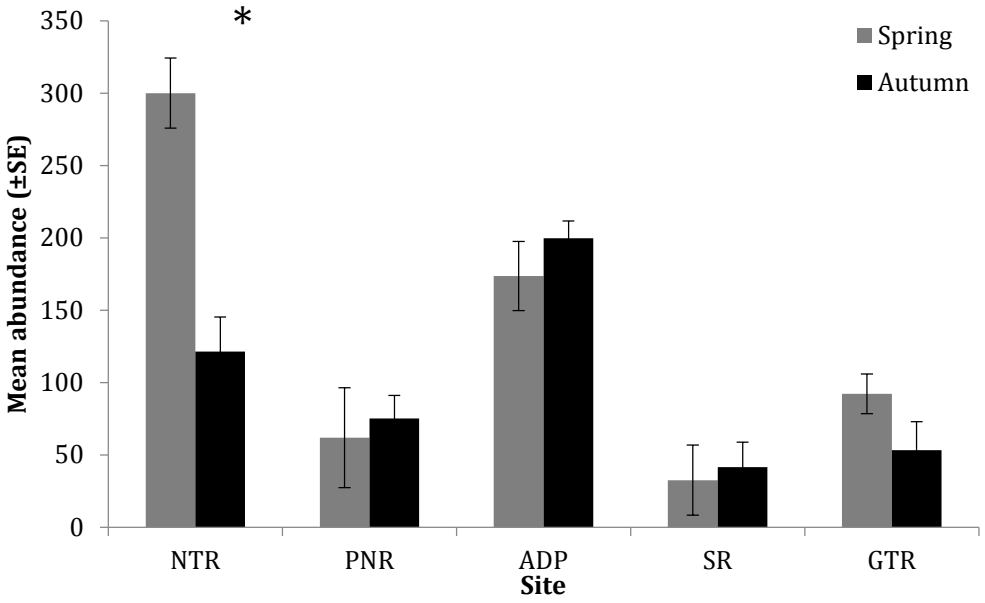

Figure 4.5: Mean abundance ± SE of total individuals per site for both spring and autumn surveys. ADP = Adelaide Desalination Plant, GTR = Glenelg Tyre Reef, NTR = Noarlunga Tyre Reef, PNR = Port Noarlunga Reef, SR = Seacliff Reef.* indicates a significant difference observed via univariate PERMANOVA pairwise tests. *N* = 60. Sites are ordered from the most southern to northern.

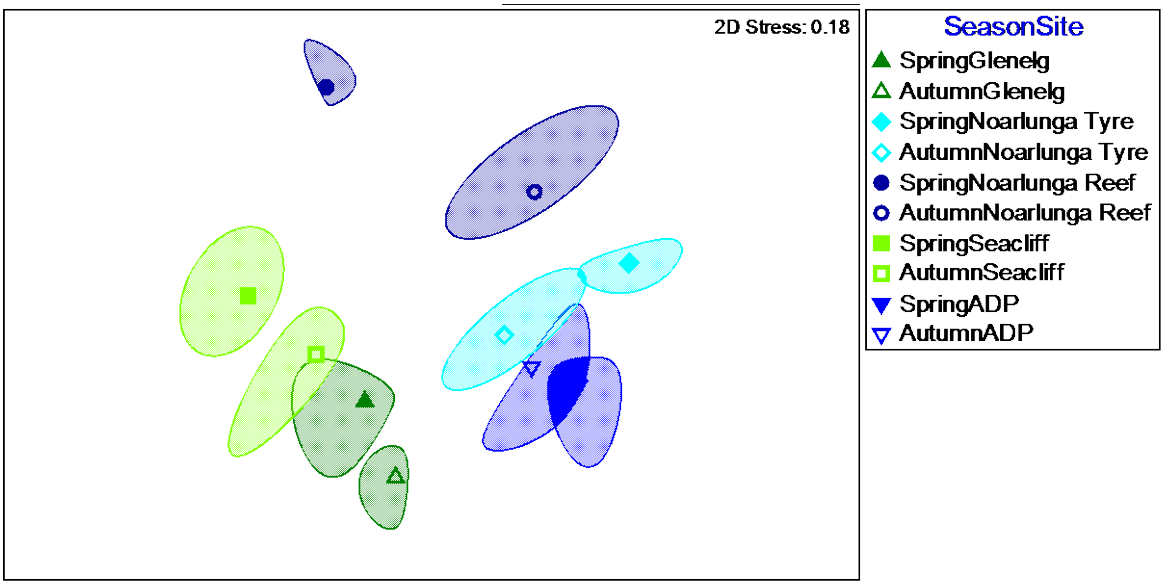

Figure 4.6: A non-metric MDS plot calculated using bootstrapped averages and showing the resulting centroid and 95 % confidence ellipses for each site within each season (n = 100 bootstraps). Southern sites are shown in blue, while northern sites are shown in green.

Pairwise tests indicated significant differences between Seasons for all sites but the ADP, with the level of dissimilarity and species driving the differences between seasons varying across sites (Figure 4.7; Table 4.4). The giant spider crab *Leptomithrax gaimardii* had consistently higher abundance during spring for the three sites where it was present. At other sites, several species drove changes between seasons, e.g. rough leatherjacket *Scobinichthys granulatus*, snapper C*hrysophrys auratus*, and silver drummer *Kyphosus sydneyanus* (Figure 4.7). The leatherjacket *Thamnaconus degeni* had the single highest % contribution to dissimilarity of any one species across all sites (28.5 %) with a large difference in relative average abundance at Noarlunga Tyre Reef (7.3 in spring vs. 1.6 in autumn).

Geographic location, Type, and Protection status were all significantly different in CAPs (trace and delta $p = 0.001$; Appendix 2B, Figure 7.1). Geographic location was the factor that best explained the differences in fish assemblages as it had the lowest misclassification error of 1.7 %, with only one sample being misclassified. Type had a higher misclassification error of 3.3 % with two samples being misclassified, and Protection had the highest misclassification error (10 %) with six samples misclassified. The combined factor of Type and Protection was also analysed using CAP and likewise showed significant differences (trace and delta $p = 0.001$). This combined factor had a similar misclassification error to that of Protection alone (10 %) with six samples misclassified.

Synthesis

Overall, BRUVS surveys at the five sampled sites revealed different fish abundances and diversity. These sites varied in their level of protection from recreational activities (e.g. anchoring, fishing), type of habitat (natural vs. artificial), and impact level. However, differences in fish assemblages at the ADP (impacted site) were not larger than differences between the four control sites and assemblages at the ADP were most similar to other artificial sites (i.e. NTR and GTR). Comparisons based on other factors (i.e. level of protection, natural vs. artificial) were also similarly different from the impact vs. control sites comparison, rejecting my hypothesis.

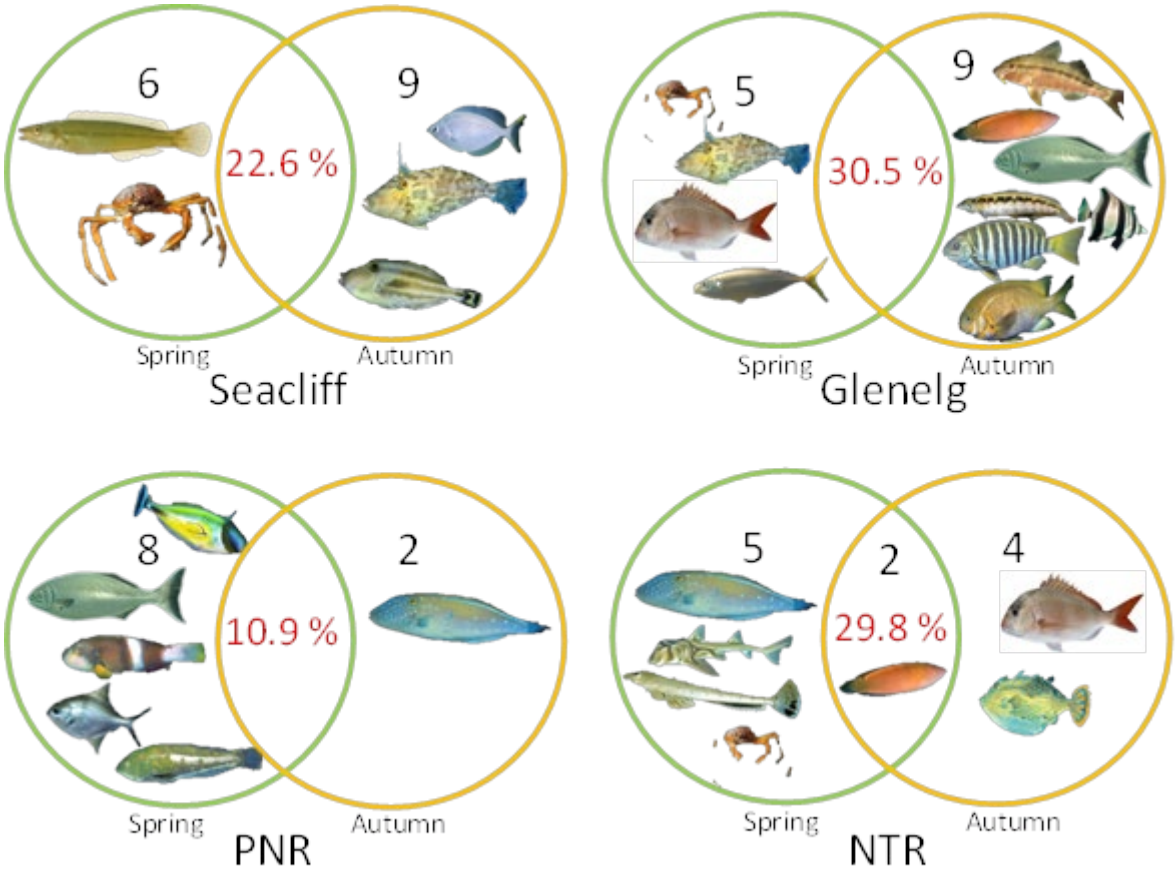

Figure 4.7: Venn diagrams displaying the results from SIMPER analysis of Season and Site factors. Numbers within the overlapping area in red show the % similarity between seasons, while the black numbers indicate the number of species with a higher average abundance during that season. Fish pictograms (Appendix 4, Table 7.15) represent species considered significant indicators (Diss/SD >1), with a higher abundance in the season they are placed in. Fish pictograms are ordered with the top fish having the highest % contribution to the dissimilarity between seasons. PNR = Port Noarlunga Reef, NTR = Noarlunga Tyre Reef. ADP is not shown as there was not a significant difference between seasons for this impacted site (see Table 4.3).

The ADP was not significantly negatively affected by brine effluent output/infrastructure because fish abundance and diversity was high at this site and in comparable numbers to the control sites. The artificial structure of the diffuser may be providing additional habitat for fish species, which are also protected through the exclusion zone. Both of these factors have been shown to be associated with increases in fish abundance and diversity (Wilhelmsson et al. 2006; Lester et al. 2009). Prior to the plant, habitat of the region was dominated with soft sediment with a small 1.5 km^2 low-profile reef inshore, without any high-profile reef structures (SA Water 2008). The change of habitat and relief stemming from the diffuser likely drove the change in fish community from species dominating soft sediments (e.g. silverbelly *Parequula melbournensis* and gobies Gobiidae spp.; SA Water 2008) to common reef species (e.g. *Chrysophrys auratus* and many leatherjacket species in the Monacanthidae family; present study). Pre-plant surveys also found low species richness in the areas now occupied by the intake and diffuser, with 6–20 species recorded depending on the area and season sampled (SA Water 2008) compared to 40 species in the present study. Another survey conducted as the plant was nearing completion was conducted in 2009/10 (Colella et al. 2010) but is difficult to directly compare to the present study due to a change in the sites sampled. The previous survey did not sample the area likely to be directly influenced by the brine plume and also compared soft sediment habitats and nearshore reef areas, which the present study did not.

The nature of the ADP exclusion zone is expected to provide some protection to resident fish species (Willis 2001; McLaren et al. 2015). It is likely to be particularly beneficial to species with small activity spaces and those often targeted by fishers such as *C. auratus*, which was the 2nd-most abundant species at the ADP site. The ADP is also well-enforced through continuous monitoring by security cameras reducing illegal fishing, with overseas studies showing well-enforced protected areas have increased benefits compared to those poorly enforced (Guidetti et al. 2008). It is also prohibited for vessels to anchor within the ADP exclusion zone, which may afford the seabed additional protection compared with other areas in which vessels commonly anchor e.g. Port Noarlunga Reef. The other protected area sampled in this study, Port Noarlunga Reef, is also a reasonably unique situation in that it is heavily used by beach goers for recreational activities such as scuba-diving and snorkelling, along with kayaking, stand-

up paddle boarding, and fishing from the jetty (allowed to within 50 m of the reef). It appears that protection may still be having a significant effect at Port Noarlunga Reef, however, because it has the most unique assemblage with the highest dissimilarity values compared to these other sites.

Seasonal variations occurred at all sites except the ADP, which did not differ significantly by season. Seasonal variations in diversity and abundance are common for temperate fish assemblages (Lehodey et al. 2006; Olsson et al. 2012; see Chapter 3). The lack of seasonal variation at the ADP site is due to consistent abundances of key resident species such as snapper *C. auratus* and Degen's leatherjacket *Thamnaconus degeni,* which had fluctuating abundances at other sites (e.g. Glenelg, Port Noarlunga Reef, and Noarlunga Tyre Reef). The reasons for these consistent abundances are currently unknown but it is likely related to favourable environmental conditions and food availability throughout both seasons. The highly enforced nature of the exclusion zone also likely protects resident fish more consistently than at other sites with lower levels of enforcement.

Assemblages observed at the study sites appeared to show highly significant clustering by location with the southern sites (Port Noarlunga Reef, Noarlunga Tyre Reef, and ADP) versus the northern sites (Seacliff Reef and Glenelg Tyre Reef). The major species associated with this difference between locations was Degen's leatherjacket *Thamnaconus degeni*, which was highly abundant at the southern sites and comparatively absent from the northern sites. The reasons for this difference are currently unknown, but are unlikely to be due to any major changes in environmental conditions as the sites are all reasonable close together and experience similar conditions. Other taxa have exhibited similar clustering by north and south, with bottlenose dolphins *Tursiops* sp. in the same region also showing distinct social groupings between the northern and southern extent of the Adelaide metropolitan region (Zanardo et al. 2017). These groups of dolphins are potentially feeding on distinct food guilds, which is supported by the evidence of different fish assemblages.

Fish assemblages were also affected by substratum type sampled, with significantly different clusters observed for assemblages between artificial and natural reefs.

Artificial reef sites had lower dissimilarity values when paired with other artificial reef sites compared to natural ones. Like other studies, I also found increased abundance for some artificial sites compared to natural areas (Arena et al. 2007; Folpp et al. 2013). Seacliff Reef was a natural site that had low abundance and diversity compared to the other sites. This site also had the lowest profile reef which was patchier than other sites such as Port Noarlunga, these habitat features may have contributed to the lower abundance and diversity. Previous research has shown relief may significantly affect fish assemblages, with studies showing increased abundance (Wilhelmsson et al. 2006) and diversity (Gratwicke and Speight 2005) for areas with high relief compared to low relief.

Assessment of fish assemblages at multiple locations with desalination brine effluents would be necessary to determine whether the findings from this study can be generalised or are merely site-specific. The areas around the desalination plant and more broadly in Gulf St Vincent also lack deep, high-profile rocky reef areas and protected artificial reefs. The ability to sample such areas suitable as 'control sites' would enable better comparisons to the ADP site. Changes in sampling design between studies also make it difficult to compare results to previous years which would be key to identifying changes over time. Future monitoring planned for this area should continue with the current sampling design and should monitor for changes over time.

The planning and design of the desalination plant (i.e. initial environmental modelling and ground-truth monitoring, along with innovations such as duck-billed diffusers used to disperse the brine discharge) likely resulted in the changes in fish assemblages being reduced. As such the changes observed were more similar to those linked with exclusion zones or artificial reefs. How such changes compare to other desalinisation plants is difficult to assess. Desalination plants that have been in operation for a long period or those situated in different environmental conditions are unlikely to provide good comparisons. In addition, many studies investigating the effects of desalination-plant effluents are not published or only exist as confidential reports. I could not, for instance, gain access to reports for studies conducted on similar desalination plants within Australia (e.g. Sydney and Southern Seawater (south of Perth)) for comparison.

Conclusion

The results showed no negative impacts (e.g. reduced fish abundance or diversity) as a result of desalination brine discharge at the ADP site. Such outcomes are likely due to the implementation of engineering methods to discharge the brine plume (e.g. the duck-bill diffuser). The artificial structure constructed around the discharge outlet provides habitat for fish assemblages and serves to attract fish into the area, and are then subsequently protected by the exclusion zone. Overall, all sites were distinct and showed variability among seasons.

Case study #3

The Neptune Islands group: A garden of berley delights

Background

Wildlife tourism is now a popular activity globally (Orams 2002; Cisneros-Montemayor et al. 2013; Huveneers et al. 2017; Trave et al. 2017) with the opportunity to view species in their natural environment sometimes being the primary driver for people's trip planning and destination choice (Apps et al. 2016). In the marine realm, wildlife tourism activities are often focussed on charismatic megafauna, which include dolphin swims, whale watching, shark diving, and other diving, swimming, or viewing with turtles, pinnipeds and large fish (Trave et al. 2017). While many wildlife tourism activities are promoted as being passive recreation or eco-friendly and may generate revenues to the regions where they are undertaken, there has been continued investigation and concern into the direct or indirect impacts of such activities (Orams 2002; Trave et al. 2017; Patroni et al. 2018).

Shark-based tourism is increasing in popularity worldwide (Cisneros-Montemayor et al. 2013; Gallagher et al. 2015). In Australia, there are four major tourism industries that focus on shark-diving: snorkelling with whale sharks off Ningaloo Reef (Western Australia); cage-diving with white sharks off Port Lincoln (South Australia); diving with grey nurse sharks off the coast of New South Wales; and diving with reef sharks at Osprey Reef (Queensland) (Huveneers et al. 2017). The Australian shark-diving industry generates $47 M per year (Huveneers et al. 2017), with white shark tourism at

the Neptune Islands generating the 2nd-largest revenue. Like many other shark-and ray-based tourism ventures (e.g. Brunnschweiler et al. 2014), the white shark cage-diving industry uses bait and berley (a mix of minced southern bluefin tuna *Thunnus maccoyii* products) to attract sharks close to the vessels.

Three operators are licensed to conduct cage-diving activities at the Neptune Islands; however, only two are licensed to use bait and berley. New licence conditions from 1 July 2017 limit the amount of bait and berley to 100 kg per day and introduced a 15-minute penalty during which bait and berley cannot be used every time a bait is consumed by a white shark. Thus, the maximum amount of bait and berley which can be released each day is 200 kg. White shark cage-diving is restricted to the Neptune Islands group (S 35.2751, E 136.0870), with 90 % of the trips occurring at the North Neptune Islands group and the remaining trips conducted 10 km away at the South Neptune Islands group. The North Neptune Islands group is usually the preferred site because it is closer to Port Lincoln than the South Neptune Islands group and typically has more sharks. Nevertheless, operators go to the South Neptune Islands group about 10 % of the time to reduce competition for sharks and when shark activity at the North Neptune Islands group is minimal (Huveneers, unpublished data).

Studies investigating the effects of tourism on sharks and rays have shown that such activity can have a range of effects including changes in seasonality, residency or abundance (Meyer et al. 2009; Bruce and Bradford 2013; Brunnschweiler et al. 2014), space use (Corcoran et al. 2013; Huveneers et al. 2013), vertical activity (Fitzpatrick et al. 2011; Huveneers et al. 2013), and physiological impact (Semeniuk et al. 2009). Nearly all of the studies investigating the effects of marine wildlife tourism have principally focused on the species that tourism operators target for customer satisfaction, so understanding the effects on non-focal species remains limited (but see Rizzari et al. 2017). At the Neptune Islands group, several studies have investigated the residency and fine-scale behaviour of white sharks in response to cage-diving activities (Bruce and Bradford 2013; Huveneers et al. 2013) but no published studies have investigated the effects of cage-diving on fish assemblages.

While white sharks attempt to consume the unminced baits, the minced berley is not consumed by the sharks because the particle size is too small for consumption. However, other species are seen feeding on berley and baits. Shark-cage diving operators at the Neptune Islands have observed several fish species feeding on the bait and berley (e.g. trevally *Pseudocaranx* spp., horseshoe leatherjackets *Meuschenia hippocrepis,* and yellowtail kingfish *Seriola lalandi*) and have noticed behavioural change, e.g. demersal species coming (from ~25 m deep) to the surface to feed or species being attracted to the boat even before berley is released (A. Fox pers. comm.). An investigation of the Neptune Islands fish assemblages is therefore required to assess the potential impacts of the cage-diving industry and the use of bait and berley on local fish communities.

Due to the presence of white sharks at the Neptune Islands making underwater visual census dangerous, it was ideal to use baited underwater video to assess the fish assemblages within the area. Two other isolated islands (Dangerous Reef and Liguanea Island) were selected as control locations as they have similar habitats, are in proximity to the Neptune Island group, but don't have routine bait and berley input. These locations were also chosen because of their differing levels of protection; Dangerous Reef is a sanctuary 'no-take' zone but fishing is allowed around Liguanea Island, which compare, respectively, to the North Neptune Islands group (with a sanctuary 'no-take' zone) and the South Neptune Islands group (fishing permitted). Thus, the aims of this project were to assess the impact of bait and berley input on fish assemblages at both Neptune Island locations compared to control locations with no routine berley input. Specifically, my hypotheses are that berley input will significantly alter the fish assemblages at the North Neptunes Islands group, and show an increase in abundance for species seen to feed on the bait and berley, compared to control sites or sites with lower bait and berley input (i.e. South Neptunes Islands group).

Methods

Sampling sites

The Neptune Islands are a series of two island groups, 70 km from Port Lincoln, South Australia. In addition to the frequent white shark visitors, the islands are home to a sizeable breeding population of long-nosed fur seals *Arctocephalus forsteri* and a

smaller breeding population of Australian sea lions *Neophoca cinerea* (Goldsworthy and Page 2009). North Neptune Islands group (henceforth called North Neptunes; S 35.2342, E 136.0656) was declared a sanctuary 'no-take' zone in 2014, prohibiting any extractive activities. The South Neptune Islands group (henceforth called South Neptunes; S 35.3375, E 136.1199) is situated approximately 10 km south from North Neptunes and is not protected with any no-take area. Liguanea Island (S34.9895, E 135.6214) is 3.5 km from the mainland of Australia and approximately 35 km from Port Lincoln. It is also home to a large breeding population of *A. forsteri* and smaller breeding colony of *N. cinerea* (Goldsworthy and Page 2009). Liguanea Island also hosts an aggregation of white sharks (Robbins et al. 2015). Dangerous Reef (S 34.8156, E 136.2125) is located approximately 35 km east of Port Lincoln and is home to the largest breeding colony of Australian sea lions (Goldsworthy and Page 2009). The island itself has been protected for more than 100 years; however, previous shark-cage diving and fishing efforts occurred at this location throughout the 1980s until protection was declared in 1989 for nearshore waters and then berleying for the sporadic cage-diving visits ceased in 2002 (Robbins et al. 2015). A larger sanctuary 'no-take' zone, extending further offshore than the one declared in 1989, became enforced from 2014.

Experimental design

Sampling occurred in January 2016 and 2017 over two 7-day periods. In 2016, all four locations were visited with six replicate deployments undertaken at each location, totalling 24 (Appendix 2C, Table 7.5). Seven additional deployments were done at North Neptunes but not at other sites due to logistical constraints, thus increasing the number of deployments at this one site to 13. Six shallower deployments (5–10 m deep) were also undertaken at North Neptunes for comparisons with the initial deeper ones (20–35 m). This 2016 sampling design allowed for comparisons among: the four Locations; the Habitats observed on the deployments (e.g. deep seagrass, deep or shallow reef, deep sand); their Protection status (protected [i.e North Neptunes and Dangerous Reef] vs. unprotected [i.e. South Neptunes and Liguanea Island]); and their Impact level (high [North Neptunes] vs. low [South Neptunes] vs. control [Dangerous Reef and Liguanea Island]).

Preliminary visual observation of the 2016 deployments showed that the habitats and substrata encountered were highly variable within and amongst the different sites, thus it was decided that a recast sampling design was to be implemented in 2017. The new design consisted of 18 replicates per location, with six targeting sand habitat, six targeting reef or seagrass (seagrass habitat was not available at all sites, thus this habitat was combined with reef when considering the initial deployments but later separated for analysis), and six deployed approximately 5 m below the surface to sample the pelagic habitat (average site depth was about 30 m, thus allowing about 25 m to the seafloor for each replicate). Logistical constraints additionally prohibited any visit to Liguanea Island during the 2017 sampling period. This 2017 sampling allowed for the following comparisons among: the three Locations; the Habitats observed on the deployments (all deep; seagrass, reef, sand, and pelagic); their Protection status (protected [i.e. North Neptunes and Dangerous Reef] vs. unprotected [i.e. South Neptunes]); and their Impact level (high [North Neptunes] vs. low [South Neptunes] vs. control [Dangerous Reef]).

BRUVS deployments

Fish assemblages were observed using Baited Remote Underwater Video Stations (BRUVS). Each benthic BRUVS unit consisted of a GoPro Hero 3+ Silver edition camera mounted within a metal frame to which a metal bait arm and mesh bait bag were attached. The pelagic BRUVS consisted of a wooden board with camera and bait arm set below the surface and anchored to the seafloor to prevent drifting. BRUVS units were baited with 500 g of minced sardines. Cameras were set to record in 1080p at 60 fps with a wide field of view. BRUVS were left to soak for 60 min before retrieval. Units were spaced a minimum of 250 m apart. Videos were processed as per Case study #1 in this chapter.

Data analysis

Despite best efforts to deploy replicates based on habitat type in 2017, some replicates were still in undesired or unintended habitats. A breakdown of replicates by year and habitat type can be seen in Appendix 2C, Table 7.5. There were some replicates which could not be included in the analysis due to the camera being obscured by high levels of macroalgae, leaving a final *N* of 87 replicates.

Statistical analyses were conducted in PRIMER v7 (Clarke and Gorley 2015) with PERMANOVA+ (Anderson et al. 2008). To compare total abundance among levels of the factors Locations and Habitat types, the total number of individuals per replicate was calculated and used to construct a Euclidean distance resemblance matrix. To broadly assess how abundance differed between locations and habitats, two such univariate PERMANOVA analyses were performed with the design of either Location (fixed factor, four levels) or Habitat type (fixed factor, four levels). I tested each factor individually to increase the power as some habitats were sparsely sampled by year or location and I wished to only make broad conclusions about fish abundances compared to the more specific, targeted analyses below. For significant factors pairwise tests were used to identify differences between individual pairs of levels.

Due to the differences in sampling design between years, further analyses were conducted separately for each yearly data set. To account for the schooling behaviour of certain fish species, multivariate assemblage data were transformed using dispersion weighting by location (Clarke et al. 2006a). Bray-Curtis similarity matrices were constructed and used to run PERMANOVA tests with Location and Habitat type considered as fixed factors. A dummy variable of 1 was added to the 2017 data to account for the pelagic replicates that observed no individuals (Clarke and Gorley 2015). Non-metric MultiDimensional Scaling (MDS) ordination plots were used to show patterns of similarity among Locations and Habitat types. The discriminant function test of Canonical Analysis of Principal coordinates (CAP) was used to test the factors of Protection status and Impact level. These analyses were used to asses these factors alone to determine whether they had any influence on the assemblages observed. SIMilarity PERcentage (SIMPER) analyses were used to determine the similarity between groups and which species were driving observed differences.

To assess for an influence on key species that have been observed to feed on the bait and berley univariate analyses were conducted on the trevally *Pseudocaranx* spp. and horseshoe leatherjeacket *Meuschenia hippocrepis* testing for the factors Year (Random factor; two levels) and Location (Fixed factor; four levels). Habitat was not included in this design to increase the power and generality of the analysis.

Results

General fish assemblage structure

Seventy-eight species were observed across all deployments, of which 10 were chondrichthyans, eight were invertebrates, and two were marine mammals. The remaining 58 species were teleost fish species which accounted for 96.4 % of the total abundance (3,706 observations), with chondrichthyans comprising the next largest portion at 2.7 % of individuals. A similar average total abundance per replicate was observed across years with an average of 43.7 ± 5.5 per BRUVS for 2016 vs. 41.8 ± 4.5 for 2017. Similar numbers of individuals per replicate were also observed among locations (Figure 4.8A; Pseudo-F = 1.59, p = 0.18). Differences in abundance among habitats was observed (Figure 4.8B; Pseudo-F = 8.13, p = 0.001), with significant differences observed between reef habitat and both pelagic (t = 4.40, p = 0.001) or sand (t = 2.89, p = 0.008; all other p > 0.053), with reef having higher abundances in both instances.

Dangerous Reef had the highest number of species unique to any location (16 species) compared to 10 at North Neptunes, four at South Neptunes, and none for Liguanea Island. In total, 14 species were only sighted on a single occasion including rarely seen species such as the silver spot *Threpterius maculosus*, and species which may be undergoing range extensions such as the banded morwong *Cheilodactylus spectabilis* and the footballer sweep *Neatypus obliquus* (www.ala.org.au). The white shark, *Carcharodon carcharias*, target species for wildlife tourism in the area, was also only observed on a single replicate but this may be due to selectively choosing times of expected low shark presence for sampling to prevent sharks tampering with the gear.

2016 fish assemblages

The fish assemblages observed at each Location and Habitat type showed significant differences during 2016 (Figure 4.9A; Table 4.5A). Liguanea Island appeared to be the least distinct location, with no significant difference observed between this location and South Neptunes (Table 4.5B). All other pairs of locations were significantly different (Table 4.5B). Reef and sand habitat types were observed to be significantly different from each other, while seagrass sites showed no significant difference (Table 4.5C), likely due to the low sample size for seagrass replicates.

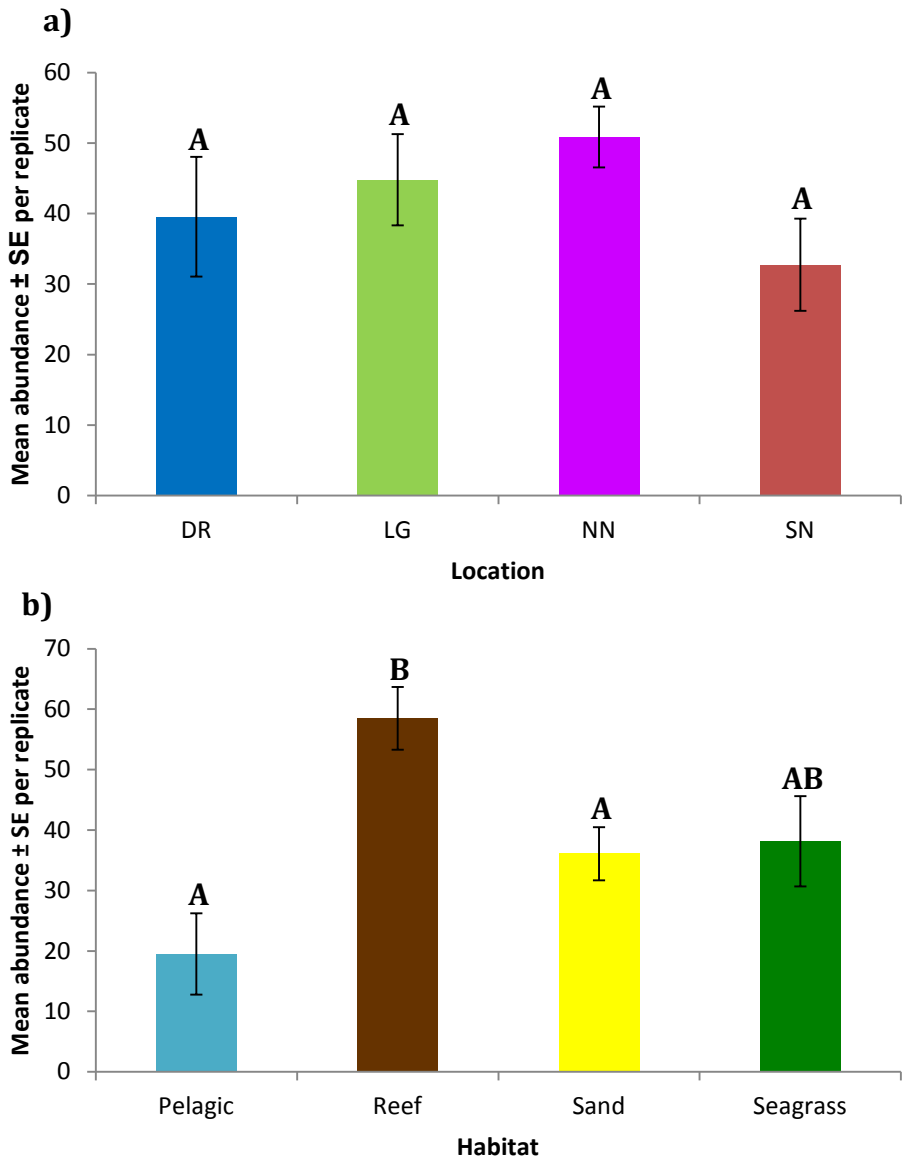

Figure 4.8: Mean abundance per replicate for all species and years combined for a) each location and b) each habitat type. DR = Dangerous Reef, NN = North Neptunes, SN = South Neptunes, LG = Liguanea island. Uppercase letter indicate significant differences assessed using univariate PERMANOVA.

Table 4.5: 2016 fish assemblage data: PERMANOVA analysis for A) the main test assessing the factors Location and Habitat type, B) the pairwise test results for Location, and C) the pairwise test results for the Habitat type factor. Values in bold indicate significant differences. Unique permutations ranged from 168 to 999.

A) Source	**df**	**MS**	**Pseudo-*F***	***p*(perm)**
Location	3	6188.5	3.51	**0.001**
Habitat type	3	3609.6	2.05	**0.001**
Lo x Ha	3	2338.1	1.32	0.109
Res	25	1764.4		

B) Location Pairs	***t***	***p*(perm)**
Dangerous Reef, Liguanea Island	2.03	**0.008**
Dangerous Reef, North Neptunes	2.11	**0.001**
Dangerous Reef, South Neptunes	1.76	**0.006**
Liguanea Island, North Neptunes	1.44	**0.025**
Liguanea Island, South Neptunes	1.25	0.182
North Neptunes, South Neptunes	1.66	**0.003**

C) Habitat type Pairs	***t***	***p*(perm)**
ReefDeep, SandDeep	1.71	**0.007**
ReefDeep, ReefShallow	1.52	**0.005**
ReefDeep, SeagrassDeep	1.27	0.122
SandDeep, ReefShallow	1.45	**0.029**
SandDeep, SeagrassDeep	1.02	0.407
ReefShallow, SeagrassDeep	1.34	0.129

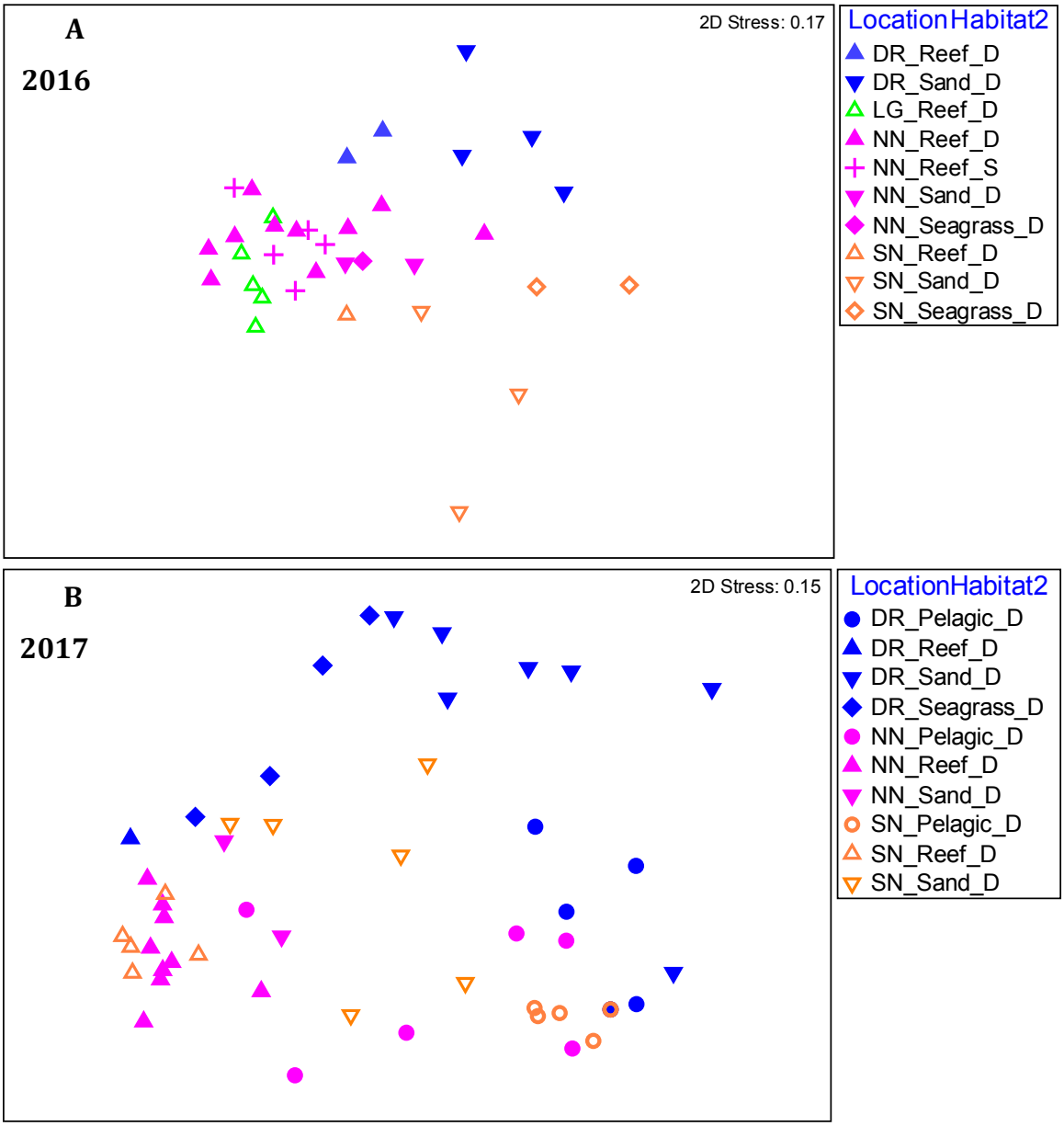

Figure 4.9: nMDS ordination plot showing the location and habitat type for each BRUVS deployment (see Appendix 2C, Table 7.5 for the available habitat types in each location) for a) 2016 (*n* = 35) and b) 2017 (*n* = 52). DR = Dangerous Reef (blue symbols), NN = North Neptunes (red), SN = South Neptunes (orange), LG = Liguanea island (green), a D indicates Deep deployments while S indicates Shallow deployments. Habitats were pelagic (circle), reef (upward triangle or cross), sand (downward triangle), or seagrass (diamond).

Protection status appeared to have an effect on the fish assemblages observed (Figure 4.10A) with CAP analyses showing a significance difference between protected and unprotected sites (trace and delta $p = 0.001$) with a high allocation success rate of 97 %. Of the top 10 species contributing to the differences between levels of protection, only two were fisheries targeted and only one of these species, *Pseudocaranx* spp., had a higher abundance in protected areas (Appendix 2C, Table 7.8). Clear separation among groups can also be seen when considering locations in terms of impact level (Figure 4.11A), with significant differences detected (trace and delta $p = 0.001$) and a relatively high allocation success rate of 83 %. However, the split of groups along the first axis represents differences between high and low impact levels rather than between impacted and control sites.

Trevally *Pseudocaranx* spp. contributed the most to the difference between the control and high impact groups, with a higher average abundance observed at the control locations compared to the impact locations (Figure 4.11A). Horseshoe leatherjackets *Meuschenia hippocrepis*, blue-throated wrasse *Notolabrus tetricus*, and senator wrasse *Pictilabrus laticlavius* also contributed to differences between impact groups with a higher abundance observed at the high impact locations, while eagle rays *Myliobatis tenuicaudatus* and red mullet *Upeneichthys vlamingii* were contributing to the differences for the low-impact locations.

2017 fish assemblages

Similar to 2016, the fish assemblages observed in 2017 were also significantly different among Locations and Habitat types (Table 4.6). Additionally, the interaction between Location and Habitat type was also significant in this second year. However, the MDS ordination plot showed groups were less distinct than those for 2016 (Figure 4.9B). There were more differences between fish assemblages by habitat type within each location than by similar habitat types between locations (Table 4.6). Pelagic habitats differed the most amongst locations with differences observed between South Neptunes and both North Neptunes and Dangerous Reef while reef habitats were not significantly different across any locations (Table 4.6).

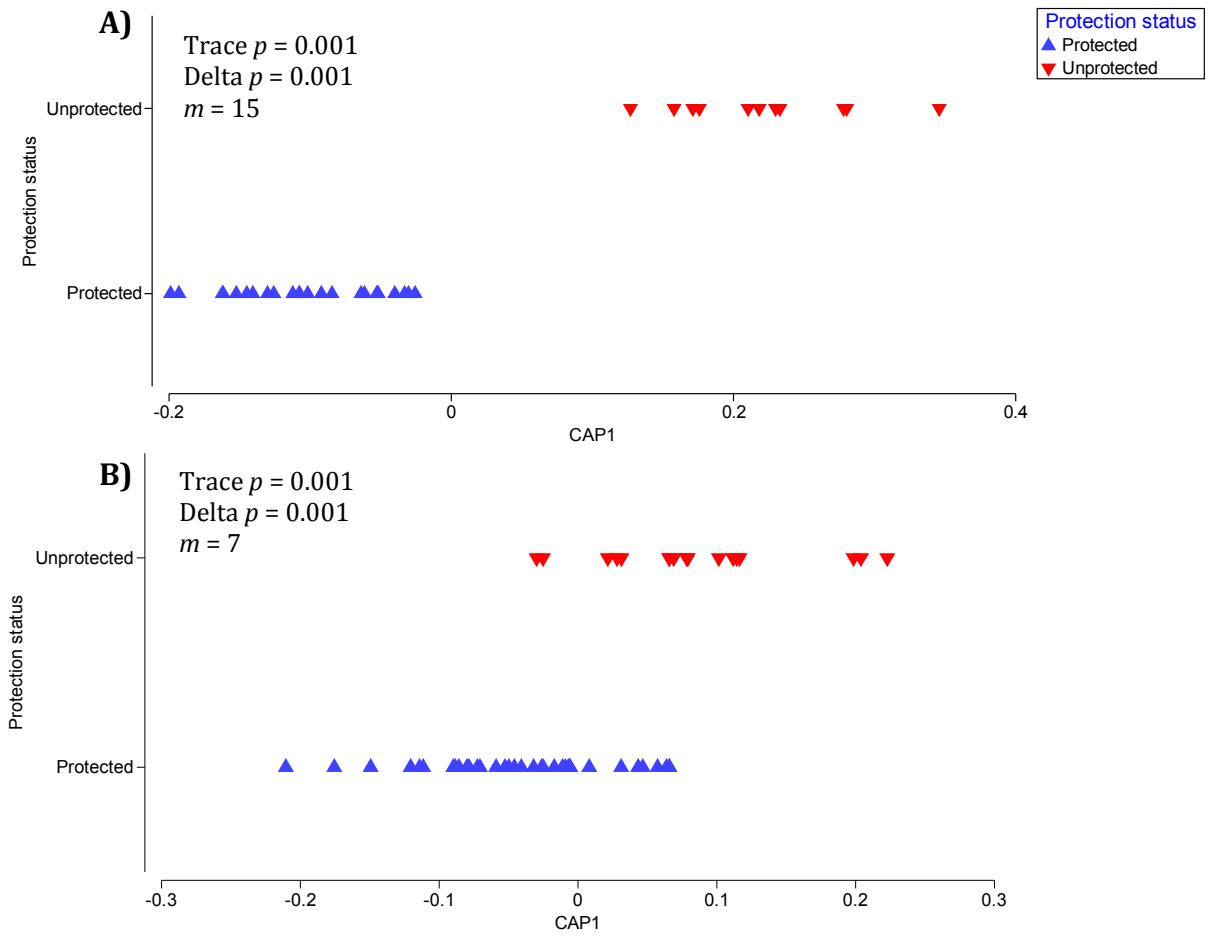

Figure 4.10: CAP constrained ordination plots showing the 1st PCO axis for the factor of Protection for A) 2016 (n = 35) and B) 2017 (n = 52) for the BRUVS deployments conducted at the Neptune Islands and surrounding control areas. m = number of axes used. Significance values for tests of the factor Protection are also given. Allocation success rate for 2016 was 97 % and for 2017 was 81 %.

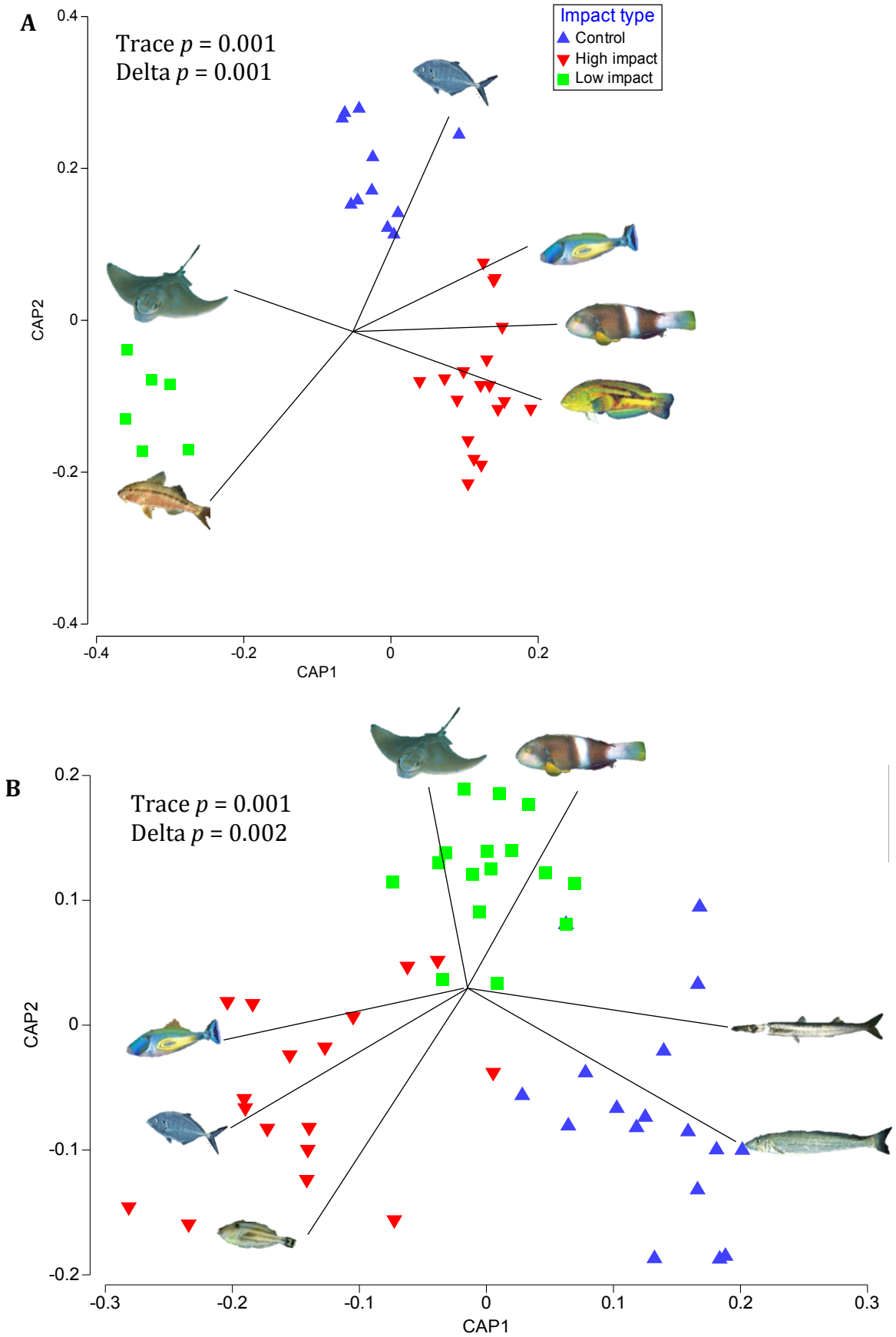

Figure 4.11: CAP ordination plots showing Level of Impact (Control = Liguanea Island and Dangerous Reef, Low = South Neptunes, High = North Neptunes) for fish assemblages in A) 2016 (*m* (# of axes) = 15) and B) 2017 (*m* = 18). Fish pictograms (Appendix 4, Table 7.15) represent the species contributing most to similarity within groups and dissimilarity between groups according to SIMPER analysis Appendix 2C, Table 7.6 and Table 7.7). Allocation success rate for 2016 was 83 % and for 2017 was 73 %

Table 4.6: 2017 fish assemblage data: PERMANOVA analysis for A) the main test assessing the factors Location and Habitat type, and the pairwise test results for the interaction of Location and Habitat type showing , B) tests between Locations and C) tests between Habitat types. Values in bold indicate significant differences.

A) Main test	**df**	**MS**	**Pseudo-*F***	***p*(perm)**	**perms**
Location	2	4962.6	3.15	0.001	999
Habitat type	3	14135	8.98	0.001	995
Lo x Ha	4	2506.4	1.59	0.011	997
Res	42	1574.7			

B) Location by Habitat	**Pelagic**			**Reef**			**Sand**		
Pairs	***t***	***p*(MC)**	**perms**	***t***	***p*(MC)**	**perms**	***t***	***p*(MC)**	**perms**
Dangerous Reef, North Neptunes	1.6	0.059	305	1.31	0.158	10	1.51	0.053	36
Dangerous Reef, South Neptunes	1.78	0.049	116	1.15	0.32	6	1.77	0.007	751
North Neptunes, South Neptunes	2.07	0.01	302	1.14	0.255	799	1.07	0.382	28

C) Habitat by Location	**Dangerous Reef**			**North Neptunes**			**South Neptunes**		
Pairs	***t***	***p*(MC)**	**perms**	***t***	***p*(MC)**	**perms**	***t***	***p*(MC)**	**perms**
PelagicDeep, ReefDeep	1.90	0.038	6	2.84	0.001	897	5.06	0.001	305
PelagicDeep, SandDeep	1.97	0.005	684	1.42	0.148	28	2.94	0.001	313
PelagicDeep, SeagrassDeep	2.59	0.005	152						
ReefDeep, SandDeep	1.29	0.149	8	1.68	0.023	55	2.24	0.007	409
ReefDeep, SeagrassDeep	1.11	0.407	5						
SandDeep, SeagrassDeep	1.48	0.059	316						

Protection status had less distinct clusters in 2017 than in 2016 (Figure 4.10B) but still showed significant differences between protected and unprotected locations (trace and delta $p = 0.001$) and had high allocation success (81 %), but lower than in 2016. Three of the top 10 species contributing to the dissimilarity between protection levels were fisheries targeted and all had a higher abundance in the protected areas (Appendix 2C, Table 7.8). Differences between impact level were also less distinct in 2017 (Figure 4.11B) but were significantly different among groups (trace and delta $p = 0.001$ and $p = 0.002$, respectively) and had a good allocation success rate of 73 %. It is also noteworthy that the split along the primary axis of the ordination plot was between impact and control locations contrary to the 2016 ordination plot which was between impact levels (Figure 4.9B and Figure 4.11B).

Aside from the eagle ray *M. tenuicaudatus,* which was a key contributor in driving differences between low-impact and high-impact locations in both 2016 and 2017, many of the species driving the differences between impact types differed between the two sampled years (Figure 4.11B). In 2017, control locations were characterised by high abundances of snook *Sphyraena novaehollandiae* and King George whiting *Sillaginodes punctatus* compared to high abundances of *M. hippocrepis*, *Pseudocaranx* spp., and six-spined leatherjackets *Meuschenia freycineti* at high-impact locations (Figure 4.11B).

Key species

The mean abundance of *Pseudocaranx* spp. was variable across locations and between years (Figure 4.12A). Significantly higher abundances were observed for Dangerous Reef in 2016 compared to the other locations, whereas North Neptunes had the highest abundance in 2017 (Appendix 2C, Table 7.9). Total abundance of *M. hippocrepis* was highest for North Neptunes in both years, but mean abundance per replicate was significantly higher at Liguanea Island compared to the other sites (Figure 4.12B).

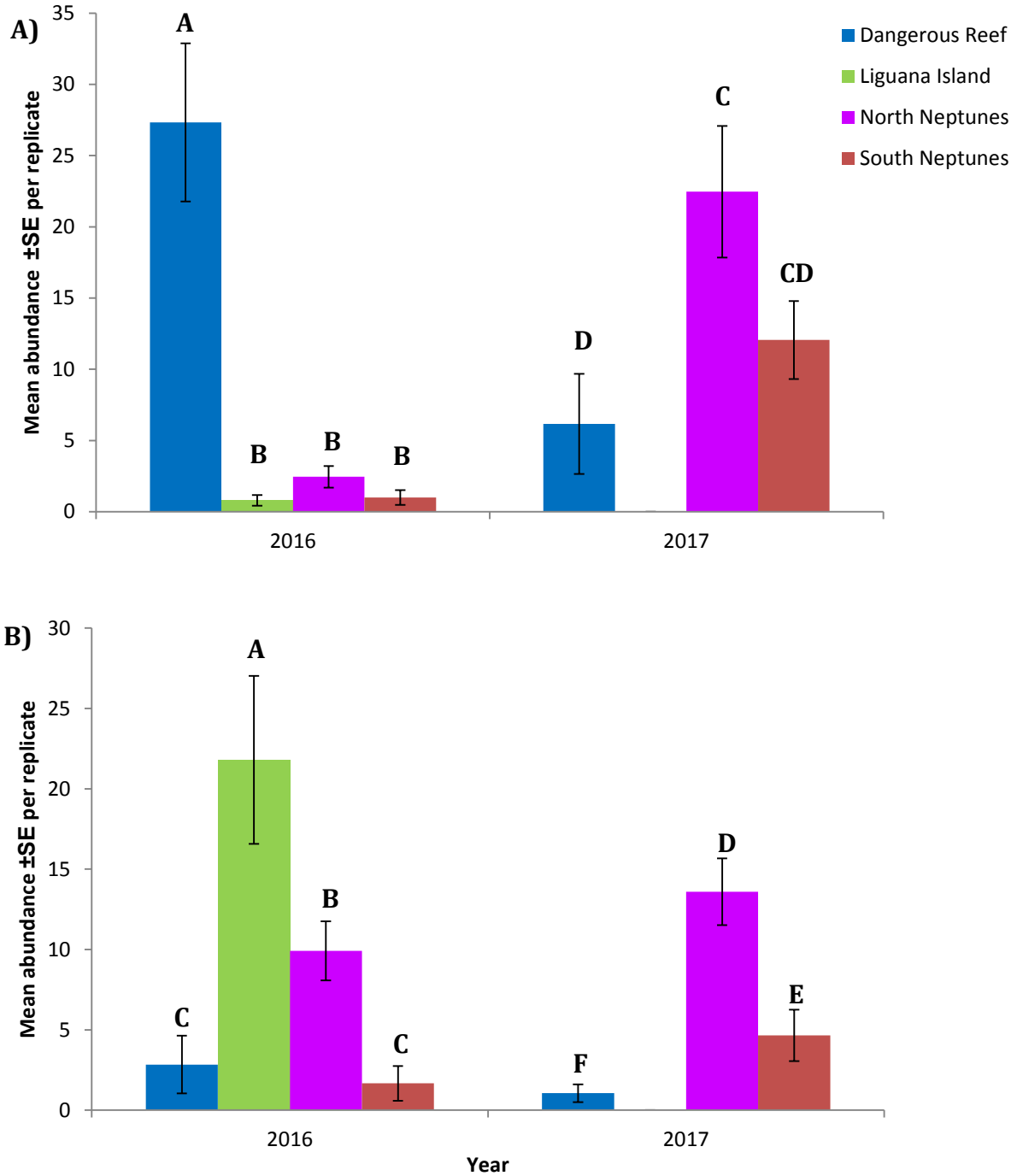

Figure 4.12: Mean abundance (± the standard error) per BRUVS deployment for A) trevally *Pseudocaranx* spp. and B) horseshoe leatherjacket *Meuschenia hippocrepis*. Note: Liguanea Island was not sampled in 2017. Uppercase letter indicate significant differences assessed using univariate PERMANOVA.

Synthesis

There is some evidence for an effect of bait and berley input at the North Neptunes but this was only consistently shown for one species, the horseshoe leatherjacket *Meuschenia hippocrepis*, which was influential for high impact locations (North Neptunes) in both sampling years and has been observed to feed on the bait and berley used during shark cage-diving activities. While fish communities varied across intensity of bait and berley, these differences were not consistent across years and the fish community at the location with high berley and bait input (North Neptunes) was not different to the control location (Dangerous Reef) for any of the three habitats sampled in 2017. Protection and habitat type significantly influences the structure and composition of assemblages in both years sampled.

My results show *Pseudocaranx* spp. and *M. hippocrepis* may be influenced by the bait and berley input from the shark-cage diving industry. While other studies have been conducted on the effects of provisioning during shark and ray tourism (e.g. Meyer et al. 2009; Semeniuk et al. 2009; Huveneers et al. 2013; Rizzari et al. 2017), less studies have focused on bony-fish assemblages (but see Trave et al. 2017; Patroni et al. 2018 for reviews). Based on those limited studies, provisioning can result in changes in fish abundance, species composition, behaviour, movement patterns, space use, and habituation to human presence (Cole 1994; Milazzo et al. 2005; Ilarri et al. 2008; Albuquerque et al. 2014). Observations have shown that trevally *Pseudocaranx* spp. actively congregate around the cage-diving boats, prolifically feed on the bait and berley used by the industry, and are observed in higher numbers at North Neptunes compared to South Neptunes (S. Whitmarsh, pers. obs.). Preliminary results from ongoing studies have shown that *Pseudocaranx* spp. from North Neptunes have fatty acid signatures reflecting the bait and berley used by the cage-diving industry and are significantly different to individuals from adjacent areas with less or no cage-diving tourism (L. Meyer pers. comm.). Thus, I expected that North Neptunes would have a higher abundance of *Pseudocaranx* spp. compared to the control locations due to the copious food available to this species from the cage-diving industry. While I found this to be the case in 2017, consistently higher abundances of *Pseudocaranx* spp. were observed at Dangerous Reef compared to North and South Neptunes in 2016. The schooling nature and heterogeneous distribution of *Pseudocaranx* spp., likely contributed to these

discrepancies between years. The aggregation of silver trevally, *Pseudocaranx georgianus*, around the cage-diving boats may have reduced the number of *Pseudocaranx* spp. being observed on the BRUVS footage (i.e. if individuals are conditioned to mill around the boats, they are not observed on the BRUVS, located further away). The perceived higher abundances of *Pseudocaranx* spp. at North Neptunes by the operators may therefore be either a true difference that cannot be adequately sampled by BRUVS positioned away from the operator vessels, or due to a perceived difference due to the individuals having changed their behaviour to aggregate around the vessels. Continued sampling is required to better assess trevally abundance when operators are absent, using small vessels to alleviate the potential bias from *Pseudocaranx georgianus* aggregating around the cage-diving vessels.

Meuschenia hippocrepis is the only other benthic species seen feeding on the bait and berley at the surface. *Meuschenia hippocrepis* generally consumes a large amount of algae and sponges (Jones 1992; Rodgers et al. 2013), which is different from the fish-based bait used by the cage-diving operators. However, studies have also found they are attracted to fish-based baits and are commonly caught as by-catch in the commercial rock-lobster fishery (Brock et al. 2007; Rodgers et al. 2013). It is likely that this species is an opportunistic scavenger and will consume carnivorous baits when available, and thus can be affected by the bait and berley used. This was supported by my study showing that *M. hippocrepis* abundance was highest at North Neptunes followed by South Neptunes and Dangerous Reef in 2017. The higher abundance of *M. hippocrepis* at Liguanea Island compared to other sites in 2016 might be related to the strong macroalgae-covered rocky reef preference exhibited by this species (www.fishesofaustralia.net.au), which dominated at Liguanea Island.

Protected and unprotected locations had significantly different assemblages in both years. Protection appeared to have a positive effect on some fisheries-targeted species with higher abundances observed in the protected areas for both years sampled (trevally *Pseudocaranx* spp., King George whiting *Sillaginodes punctatus*, and snook *Sphyraena novaehollandiae*; Appendix 2C, Table 7.8). Dangerous Reef, one of the protected locations, also had the highest diversity of unique species which may be a result of its protected status (protected in the nearshore waters since 1989) and the

lack of bait and berley input. Studies have also found that *M. hippocrepis* can respond positively to protection with significantly higher abundances at protected sites and an eight times increase in biomass compared to unprotected sites (Kleczkowski et al. 2008). This may explain the high abundances of this species at North Neptunes. A related species of trevally *Pseudocaranx dentex*, was shown to be highly mobile with large home ranges, particularly for individuals tagged at offshore islands (Afonso et al. 2009). If the species of trevally found at the Neptunes (*P. wrighti* and *P. georgianus*) are also so highly mobile, it is unlikely that they would be potentially showing influence of protection. However, it is possible that the aggregating behaviour they exhibit at North Neptunes may encourage less movement of individuals. Studies tagging individuals and tracking their movement would allow researchers to determine if the protected area is adequate to protect adult fishes. Other studies have also found a different trevally species (*Pseudocaranx dinjerra*) was the only studied species that did not show a difference in length between protected and unprotected areas (Watson et al. 2009). Research is currently ongoing investigating movement patterns and lengths of individuals from *Pseudocaranx georgianus* at the Neptune Islands group (L. Meyer, unpublished data) and will increase our understanding of the effects of protection on this species.

Sampling in such remote offshore island locations has many associated difficulties, i.e. there are little mapping data available to ensure deployments are conducted on desired habitats; locations are remote and can be costly to access; and control locations can be hard to find as each island group has its own associated environmental conditions and inherent stressors. The lack of control locations with similar levels of habitat types, exposure, size, and levels of protection impedes the ability to tease apart which factors are affecting fish assemblages. Previous studies have shown that incorporating multiple control locations may enable better detection of differences between locations and which factors are affecting these changes (Underwood 1994). Continued monitoring in the study locations and inclusion of more control locations would allow for ongoing trends and a better understanding of the natural variability of fish abundance to be understood. To ensure the cage-diving industry is sustainable and has the lowest possible impact, it may also be useful to conduct additional research. This research should focus on those species which are primarily affected (e.g.

Pseudocaranx spp., *M. hippocrepis*, and *S. lalandi*) to see how congregating around the shark-cage diving vessels may affect the survivorship of such assemblages, and to assess any flow-on effects that increased fish abundances may be having on other parts of the ecosystem.

Conclusion

Overall, trevally *Pseudocaranx* spp. and horseshoe leatherjacket *Meuschenia hippocrepis* may have been influenced by the bait and berley input from the shark-cage diving industry. These effects were, however, small and varied between years sampled. If the bait and berley were significantly affecting the overall fish community, we would expect that the assemblages observed among locations would show more distinct and consistent differences. My results were one of the few studies conducted on fish assemblages at offshore islands in temperate southern Australia. These results are the first quantitative assessment of fish assemblages at the Neptune Islands group and will provide baseline data for future research, particularly for assessing changes as a result of the cage-diving industry, but also to assess the effects of long-term protection at North Neptunes.

Case study #4

Cumulative stressors in a semi-enclosed bay: Why more is stress

Background

Research on anthropogenic stressors has been increasing globally, but studies often research only singular potential stressors (Crain et al. 2008).The possible influence of cumulative stressors is not often studied, particularly using field experiments (Vinebrooke et al. 2004; Crain et al. 2008). This is likely due to the difficulty in untangling the potential effects of each stressor. Of the research that has been conducted assessing cumulative stressors on fish assemblages, the focus has been primarily on tropical reef communities (e.g. Hughes and Connell 1999; Wilson et al. 2006), or lakes (e.g. Jennings et al. 1999; Christensen et al. 2006), with some studies considering whole-ecosystem impacts on large scales (e.g. Halpern et al. 2008a; Halpern et al. 2009a; Ban et al. 2010; Marta et al. 2012). More limited research on temperate reef

areas has found that multiple stressors (such as fishing and algal blooms) had complex effects impacting multiple trophic levels (Shears and Ross 2010). The complex and unanticipated influences that multiple anthropogenic stressors can have (Shears and Ross 2010) highlight the continued need for developing such approaches, especially through simultaneous assessment of a range of stressors and focussing upon their cumulative effects within real-life field situations. Thus, I have selected a small temperate bay with multiple potential anthropogenic stressors with modest intensities, to assess cumulative impacts in soft sediment and seagrass-dominated ecosystems.

Coffin Bay (S 34.6229, E 135.4714) is a series of bays situated within the western side of Eyre Peninsula, South Australia. It is a popular holiday destination, with a base population of 600–650 people swelling to over 4,000 during summer (Australian Bureau of Statistics, 2016). The area consists of a series of semi-enclosed, shallow bays (Figure 4.13) which cover approximately 125 km^2 (Saunders 2012) . The average water depth of Coffin Bay is 2.5 m, with 20 % of the area being less than 1 m deep (Strutton et al. 1996). The convoluted shoreline of Coffin Bay covers 120 km and consists of a variety of habitats including sandy beaches, sandflats, saltmarsh areas and rocky limestone ridges (Saunders 2012). Coffin Bay acts as an inverse estuary with salinities often higher within the bay than those of the adjacent open ocean (Strutton et al. 1996; Kämpf and Ellis 2014).

The 'Coffin Bay Oyster' is a highly sought-after aquaculture product comprising of Pacific oysters *Crassostrea gigas*. After the ruin of the natural oyster reefs in the late 1800's and early 1900's through systematic harvesting and dredging (Alleway and Connell 2015), Pacific oysters were introduced into Coffin Bay in 1969 (Pierce 2011). Commercial farm production began in the 1980's and recently (2014/2015) contributed $19.6 M directly into the South Australian economy with an additional $15.7 M produced in downstream and flow-on outputs from the oyster industry in Eyre Peninsula (Econsearch 2016). Oyster aquaculture on the Eyre Peninsula also contributes 204 full-time equivalent jobs (Econsearch 2016). The total lease area for oysters statewide is 940 ha with a majority of the Coffin Bay oysters grown in Kellidie Bay and Port Douglas Bay (Figure 4.13; Primary Industries and Regions South Australia 2014).

Semi-enclosed bays can often be more at risk for exposure to, and subsequent harm from, effluent outputs than open areas (Webster and Harris 2004). The long lag times for water exchange due to their small mouth openings can lead to the accumulation of pollutants from effluent sources (Strutton et al. 1996). There are various effluent outputs which flow into Coffin Bay including two small natural creeks that flow into Kellidie Bay during heavy rain events, groundwater seepage from Coffin Bay 'lens A' (Saunders 2012), run off from agricultural areas surrounding Kellidie Bay and Mount Dutton Bay, and outflows of stormwater from the Coffin Bay township and smaller settlements such as Little Douglas (Saunders 2012).

Due to the range of species that can be found in the varied sheltered habitats located within Coffin Bay (Saunders 2012), it is a popular recreational fishing spot where fishers target species such as King George whiting *Sillaginodes punctatus*, Australian salmon *Arripis truttaceus*, and yellowtail kingfish *Seriola lalandi*. Commercial fishers also operate within the region, targeting octopus and marine scalefish. Marine parks were declared in Coffin Bay in 2014 to provide some protection from the effects of such fishing pressure and other human activties, but their effects on the fish assemblages remain unknown.

Coffin Bay affords a unique opportunity to study the effects of a range of anthropogenic influences. The shallow, sheltered nature of the waters within Coffin Bay makes them ideal to sample using Baited Remote Underwater Video Stations (BRUVS). Thus, the aims of this study are: 1) to assess fish assemblages in the different bays around Coffin Bay; 2) to determine the influence on seasonality on assemblages; and 3) to determine the influence of the mix of anthropogenic stressors across those bays on fish assemblages in particular investigating proximity to oyster leases, effluent outputs, and fishing pressure.

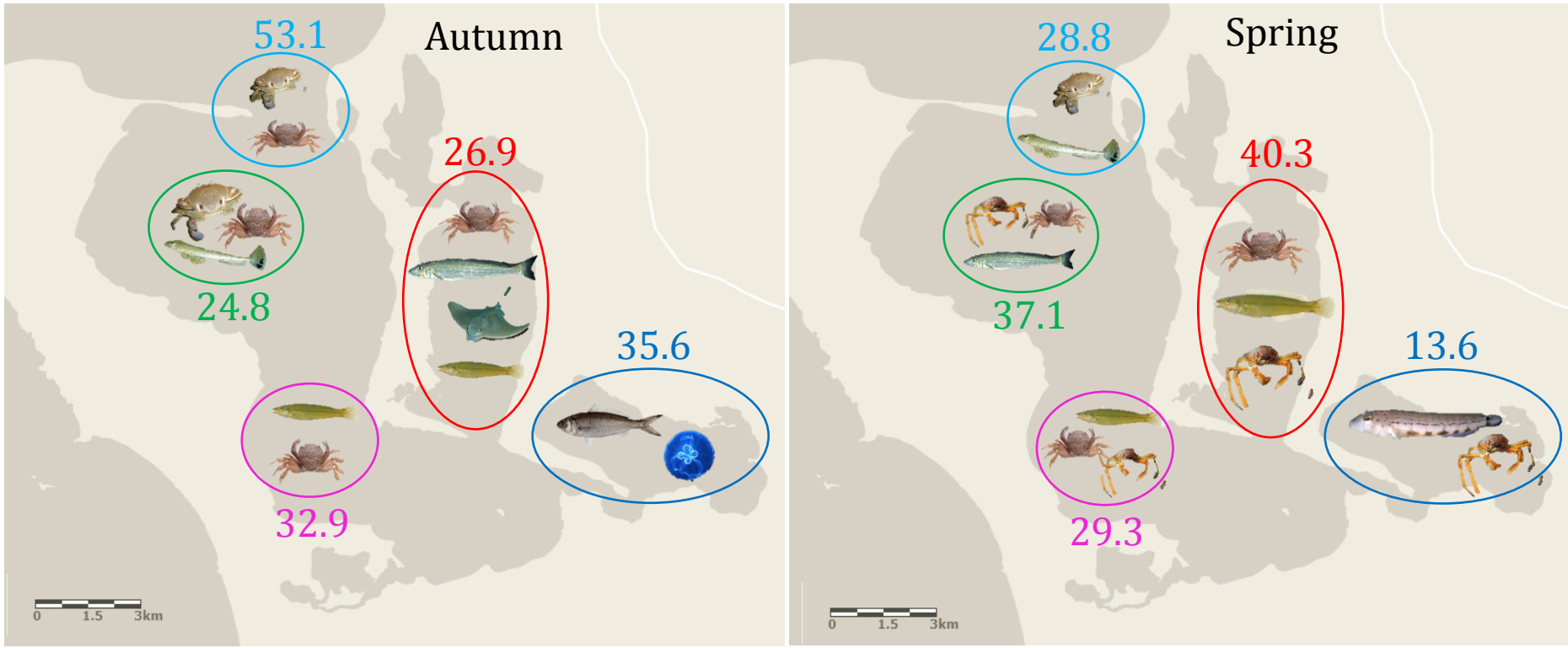

Figure 4.13: Species contributing most to the similarity within a site as shown by SIMPER (Appendix 2D, Table 7.10) with the numbers indicating the % similarity within that site for each season sampled. Pictograms can be linked to fish names Appendix 4, Table 7.15. Dark blue = Kellidie Bay, red = Mount Dutton, pink = Port Douglas South, green = Port Douglas Mid, light blue = Point Longnose.

Table 4.7: A summary of each of the habitats present in the sites sampled (see Figure 4.13), and scoring for the different anthropogenic stressors present within each site. See Appendix 2D, Figure 7.2 for more details of the habitats sampled within each site.

Site	**Habitats**			**Stressor scoring**			
	sand	algae	seagrass	**Oyster leases**	**Effluent input**	**Fishing pressure**	**Exploitation (i.e. not protected)**
Kellidie Bay	+	+		+	+++	++	-
Mount Dutton		+	+	-	+	+	-
Port Douglas South	+		+	-	+	+++	+
Port Douglas Mid	+	+		+	+	+++	+
Point Longnose	+			+	++	+++	+

Methods

Sampling sites

Five sampling sites were chosen within the inner region of Coffin Bay (Figure 4.13), Kellidie Bay, Mount Dutton, Port Douglas South, Port Douglas Mid, and Point Longnose. These sites were chosen to be representative of each bay within the inner region of Coffin Bay and reflect the different levels of anthropogenic influences within each area (Table 4.7).

Sampling design

Sampling was conducted across two trips within two consecutive days in different seasons, in April for the autumn sampling period and September for the spring sampling period. At each site, six replicate deployments were made using single BRUVS, resulting in $N = 60$ (30 per season). Within Kellidie Bay and Mount Dutton, three replicates were conducted in protected areas and three in unprotected areas. Thus, combinations of the following factors could be analysed: Season (fixed factor; two levels); Site (fixed factor; five levels); and Protection Status (fixed factor; two levels).

Data collection

Each BRUVS unit consisted of a GoPro Hero 3+ Silver edition camera mounted within a metal frame to which a bait arm and mesh bait bag were attached. BRUVS units were baited with 500 g of minced sardines. Cameras were set to record in 1080p at 60 fps with a wide field of view. BRUVS were deployed on the seafloor and left to soak for 60 min before retrieval. Replicate deployments were spaced at a minimum 250 m apart. Videos were processed as per Case study #1 in this chapter. Extra image analysis was also conducted using BenthoBox (www.benthobox.com) to record visibility and habitat type for each deployment. BenthoBox allowed different configurations of points to be overlaid onto the uploaded images. Different variables can then be attributed to the whole image or points within each image. My images were analysed using a 20 point grid overlay set onto each still image that was taken from the start of each deployment (once the BRUVS had settled). For each grid point the broad habitat type (i.e. Macroalgae, Seagrass, or Unconsolidated [i.e. either sand or mud]) was assessed. For grids that contained multiple habitat types, the dominant type was recorded. Grids which were dominated by open water were excluded from analysis and the remaining

habitat types were calculated up to 100 %. A score was also given to each grid for a relief measure, with values between 1 and 5 (based on Wilson et al. 2007), 5 having the highest relief. Visibility was visually estimated for the whole image.

Environmental variables were collected during separate surveys within the same month of sampling to get measurements of temperature, salinity, and pH (see Passadore 2017 for more details). When no measurements were taken in the immediate vicinity (~25 % of deployments), an average of the three closest measurements was used. Assessments of anthropogenic influences were undertaken using observations during sampling (i.e. for fishers present) and using local expert knowledge. Each deployment was given an ordinal value for fishing intensity (never, rarely, occasional, often, heavy) and effluent proximity (furthest, far, near), which was subsequently standardised into a numerical range (0, 5, 15, 30, 50 and 0, 15, 50, respectively). For distance to oyster lease, measurements were taken from Google Earth using the GPS location of the deployment to determine distances (m) to the nearest oyster lease visible on Google Earth. Thus, each BRUVS deployment had a measurement for the habitat variables, visibility, depth, temperature, salinity, pH, distance to oyster lease, and standardised numerical values for fishing intensity and effluent input (Table 4.8).

Statistical analysis

Statistical analyses were conducted in PRIMER v7 (Clarke and Gorley 2015) and PERMANOVA+ (Anderson et al. 2008). Univariate total abundance per replicate (all species summed) were analysed using PERMANOVA with a Euclidean resemblance matrix to test for differences due to factors Seasons and Sites. Multivariate assemblages were viewed first using shade plots (Clarke et al. 2014) before a transformation by dispersion weighting (by Site) was applied (Clarke et al. 2006b). This transformation was used to account for the schooling nature of some fish species and down-weighed the importance of highly abundant species. A Bray-Curtis resemblance matrix was then constructed incorporating a dummy variable of one to account for sparseness and an outlying replicate which observed only one species (Australian anchovy *Engraulis australis*) that was not seen on any other deployment. PERMANOVA was then used to test for differences for the factors Season and Site. Metric Multi-Dimensional Scaling (MDS) ordination plots constructed using bootstrapped averages (run 120 times; Clarke

and Gorley 2015) for the factor Site were used to visualise patterns in the data. Regular non-metric MDS plots had stress levels over the accepted limit of 0.2 (Clarke and Gorley 2015) and thus could not be used. Significant results from the PERMANOVA analysis were then expanded upon using pairwise tests done with species contributing most to the similarities within sites investigated using SIMPER analysis. To test for a relationship between replicate similarity and geographic proximity, a RELATE analysis was conducted using the multivariate assemblage matrix and a Euclidean distance matrix based on latitude and longitude for each replicate. The influence of Protection Status on fish assemblages was investigated using a subset of the multivariate fish assemblages incorporating only results from Kellidie Bay and Mount Dutton (where protected and unprotected areas were sampled). A PERMANOVA analysis was conducted on a reduced Bray-Curtis resemblance matrix (still including a dummy variable) for only deployments from the Kellidie Bay and Mount Dutton sites (n = 24) for the factors Season, Site and Protection Status (nested within Site). Patterns in the data were then visualised using nMDS ordination plots.

The influence of the anthropogenic stressors and environmental variables were tested using Distance based Linear Models (DistLM; McArdle and Anderson 2001). This approach seeks to analyse and model the relationship between one or more predictor variables and the resemblance matrix of a desired biotic dataset and can fit variables individuals or in groups (Anderson et al. 2008). My analysis was run on both the full multivariate assemblage matrix and for the univariate total abundance matrix alone, and was used to identify which of the variables (Table 4.8; or a combination of them) explained the most variation in the assemblages. Draftsman plots were first viewed to assess for potential inter-correlations between environmental variables; with a strong negative correlation (> - 0.7) between % unconsolidated and relief being observed, so that % unconsolidated was excluded from further analysis. Combinations of variables were tested using the "step-wise" selection method with the AIC (Akaike Information Criterion) being used to select the final model (Anderson et al. 2008). The step-wise selection method was used as it combines the 'forward' selection method which starts with a null model and adds variables at each step until the selection criterion no longer improves along with a possible backward elimination which allows for variables to be removed. This method creates a parsimonious model that incorporates the 'best'

combination of variables (Anderson et al. 2008). This 'best' combination of variables was then plotted as vector overlays over the multivariate assemblage using a constrained ordination plot created using a distance-based redundancy analysis (dbRDA) performed with the best model (Anderson et al. 2008).

Results

A description of fish assemblages in Coffin Bay

A variety of animals were observed on the BRUVS deployed throughout both seasons of sampling in Coffin Bay including cnidarians (3 species), cephalopods (2), decapods (3), echinoderms (1), marine mammals (1), aquatic birds (1), along with fish species comprising of teleosts (29) and chondrichthyans (3), totalling 43 taxa. For ease of reference I will refer to the above mixed assemblages as a 'fish assemblage' throughout. Across both seasons, 1,734 individuals were observed with more individuals observed in autumn (1027) than spring (707). Similar numbers of individuals and species were observed at each site (Figure 4.14) with no significant differences observed for abundance among sites (Pseudo-F = 1.51, p = 0.187) or between seasons (Pseudo-F = 0.11, p = 0.741) or for their interaction (Pseudo-F = 1.49, p = 0.219). The most abundant species observed was the weedy whiting *Neoodax balteatus*, followed by the combined groups of Australian salmon and herring *Arripis* spp., and trevally *Pseudocaranx* spp. Three crab species (*Ovalipes australiensis*, *Nectocarcinus integrifrons* and *Leptomithrax gaimardii*) were also included in the top six most abundant species. *Arripis* spp., *N. integrifrons* and *N. balteatus* were also the most prevalent species appearing in 65 %, 63 %, and 55 % of deployments, respectively.

Table 4.8: Summary of the 11 environmental variables included in the DistLM analysis.

Variable	Shorthand	Units	How it was measured
% Seagrass	% S	Percentage	From BRUVS field of view using 20 point grid overlay
% Macroalgae	% M	Percentage	From BRUVS field of view using 20 point grid overlay
Relief	R	Standardised range	From a scale of 0 to 5 from BRUVS field of view using 20 point grid overlay
Visibility	Vis	m	Estimated from BRUVS field of view
Depth	D	m	Recorded during BRUVS deployments (from boat sounder)
Temperature	T	˚C	Using temperature and salinity probe
Salinity	S	ppm	Using temperature and salinity probe
pH	pH		Using temperature and salinity probe
Distance to oyster lease	DOL	m	Calculated from Google Earth
Fishing intensity	FI	Standardised range	Using categories subsequently standardised numerically based on expert opinion
Effluent input	E	Standardised range	Using categories subsequently standardised numerically based on expert opinion

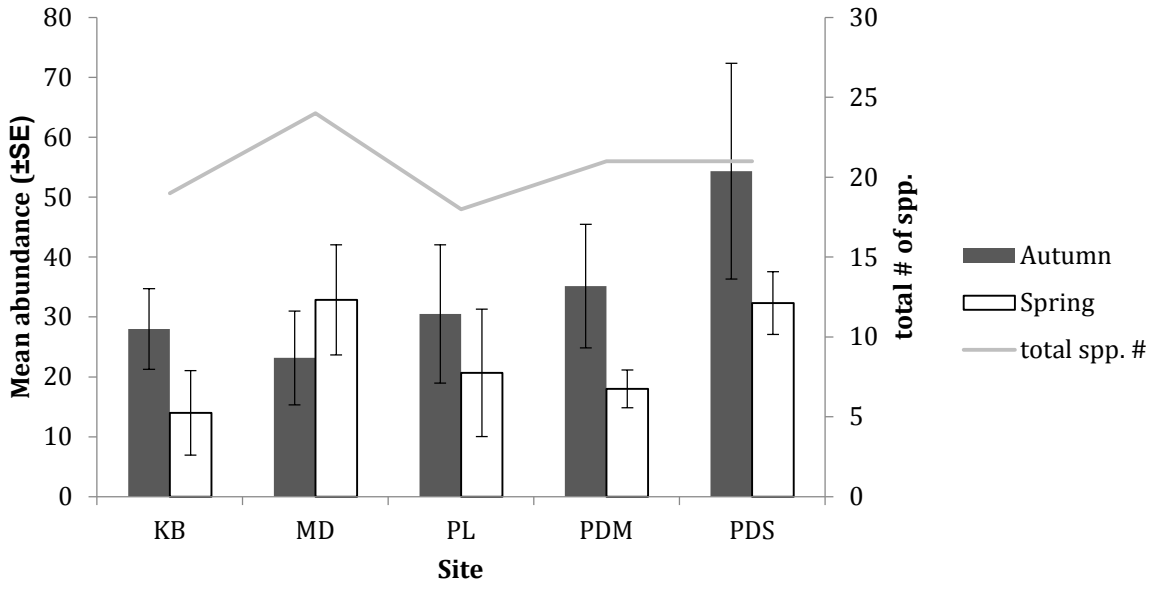

Figure 4.14: The mean number of individuals (±SE) observed for each site sampled by season, alongside the total number of species for both seasons combined. KB = Kellidie Bay, MD = Mount Dutton, PL = Point Longnose, PDM = Port Douglas Mid, PDS = Port Douglas South.

Site and season variability amongst assemblages

Distinct assemblages were observed among sites (Figure 4.15; PERMANOVA Pseudo-F = 5.43, p = 0.001) and seasons (PERMANOVA Pseudo-F = 5.36, p = 0.001) but there was no significant interaction between the two factors (PERMANOVA Pseudo-F = 1.34, P = 0.072). All sites exhibited significant differences between pairs with the exception of Point Longnose and Port Douglas Mid (Figure 4.15; Table 4.9). There was a significant relationship between replicate similarity and geographic proximity of replicates (*Rho* = 0.322, p = 0.001). Species influencing the similarity within sites changed across the seasons, with more spider crabs *L. gaimardii* present in spring versus autumn for four out of five sites (Figure 4.13; Appendix 2D, Table 7.10). Kellidie Bay, Mount Dutton, and Port Douglas Mid appeared to be the most affected by seasonality as these sites had the biggest change of species influencing similarity (Figure 4.13; Appendix 2D, Table 7.10). Point Longnose in autumn had the highest similarity value indicating that these replicate deployments had the most consistent assemblages. Protection was not observed to have a significant effect on assemblages for either site or season (Figure 4.16; Table 4.10).

The apparent influence of anthropogenic stressors

Seven of the 11 environmental variables included in the analysis were identified as part of the best model to explain the variation for the multivariate fish assemblage (effluent, visibility, relief, % seagrass habitat cover, pH, distance to oyster lease, and salinity), accounting for 37.3 % of the total variation. Assemblages within Kellidie Bay appeared to be more affected by many of the selected environmental variables, particularly for the autumn sampling period (Figure 4.17), due to its separation from the other sites in the dbRDA ordination plot. The first two primary axes of the dbRDA explained 63 % of the variation accounted for in the best multivariate model, and thus 23 % of the total variation (Figure 4.17; Appendix 2D, Table 7.11). The variables distance to oyster lease and effluent were the most strongly correlated with each of the first two axes, respectively (Table 4.11; Figure 4.17). Similarly % seagrass, relief, and salinity were also strongly correlated with the axes, while pH and visibility, were less strongly correlated (Figure 4.17). In contrast DistLM showed that total abundance was not affected by environmental variables, with the best models explaining only small amounts of the variation (3.5 %), thus no further analyses were conducted with these data.

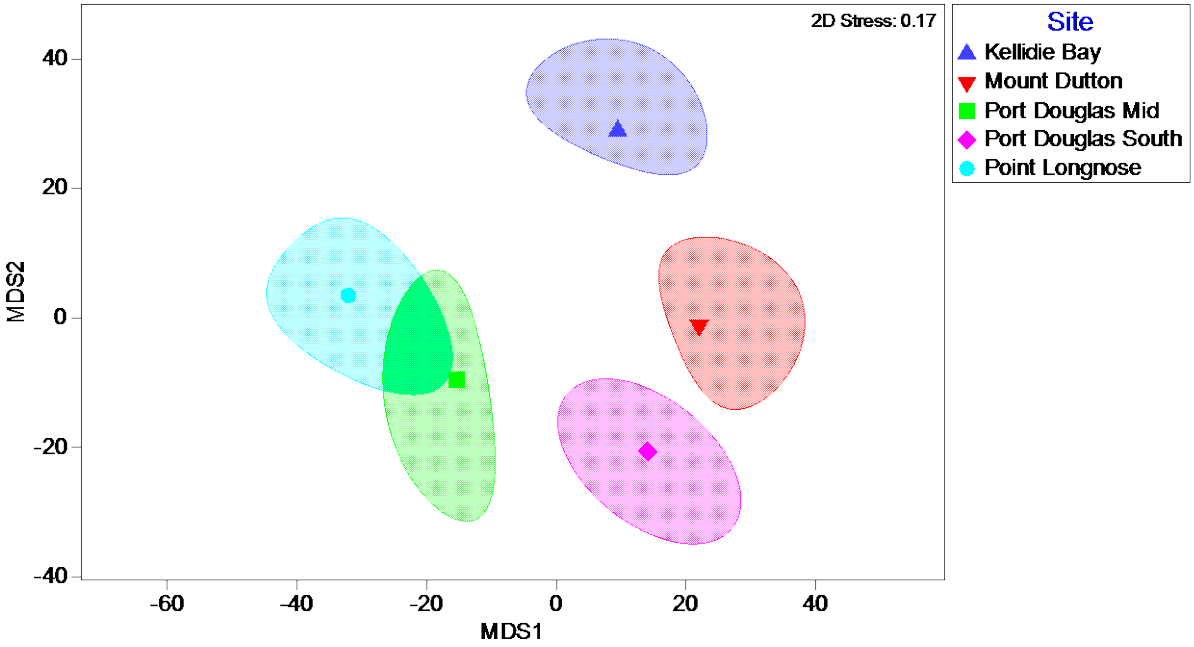

Figure 4.15: Metric MDS plot showing the bootstrap averages for each site sampled for both seasons combined. Coloured points are the centroids and the ellipses show the variation from 120 runs.

Table 4.9: Pairwise tests for the factor Site based on the multivariate fish assemblage observed at Coffin Bay in both spring and autumn. Significant values are shown in bold. Unique permutations ranged from 997 – 999. Sites are indicated by their initials.

Pairs	***t***	***p*(perm)**
KB vs. MD	2.07	**0.001**
KB vs. PL	2.89	**0.001**
KD vs. PDM	2.44	**0.001**
KB vs. PDS	2.33	**0.001**
MD vs. PL	3.14	**0.001**
MD vs. PDM	1.99	**0.001**
MD vs. PDS	1.54	**0.011**
PL vs. PDM	1.48	0.074
PL vs. PDS	2.77	**0.001**
PDM vs. PDS	2.08	**0.001**

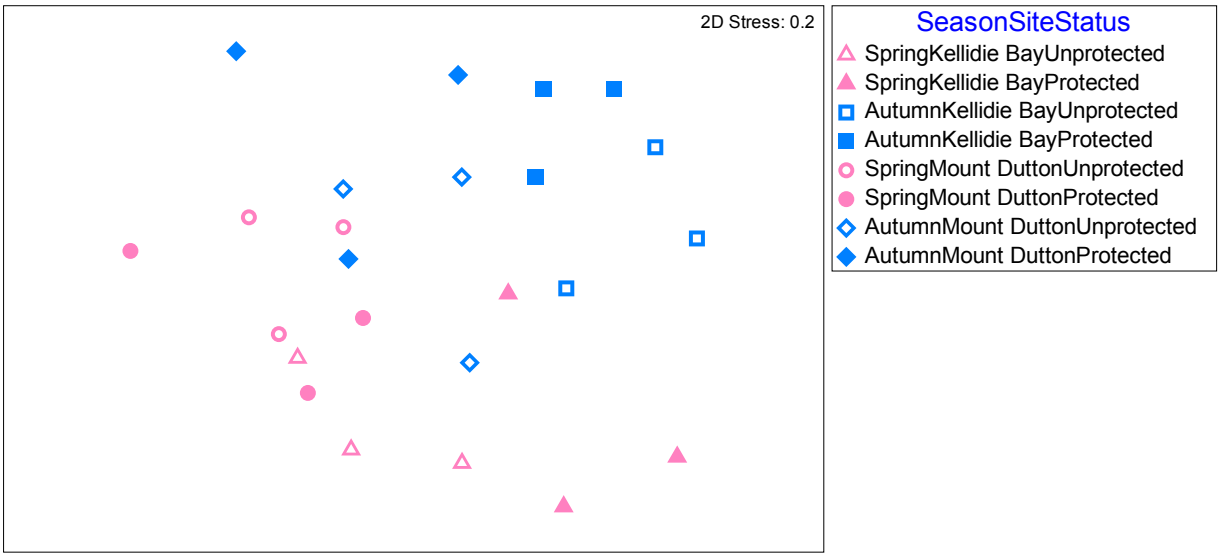

Figure 4.16: nMDS ordination plot showing the influence of protection on fish assemblages observed for the two sites where protected (filled symbols) and unprotected (open symbols) areas were sampled. Autumn deployments are shown in blue, while spring are shown in pink.

Table 4.10: PERMANOVA results for the reduced dataset including only those two sites where protected and unprotected areas could be sampled (Kellidie Bay and Mount Dutton) as the nested factor Status. Significant values are shown in bold. Unique permutation ranged from 981 – 999.

Main test	**df**	**MS**	**Pseudo-*F***	***p*(perm)**
Season [Se]	1	7793.3	4.64	**0.001**
Site [Si]	1	7802.3	4.64	**0.001**
Status [St] within (Si)	1	2185.1	1.30	0.18
Se x Si	1	2592.2	1.54	0.113
Se x St (Si)	1	2529.7	1.51	0.065
Res	16	1679.6		

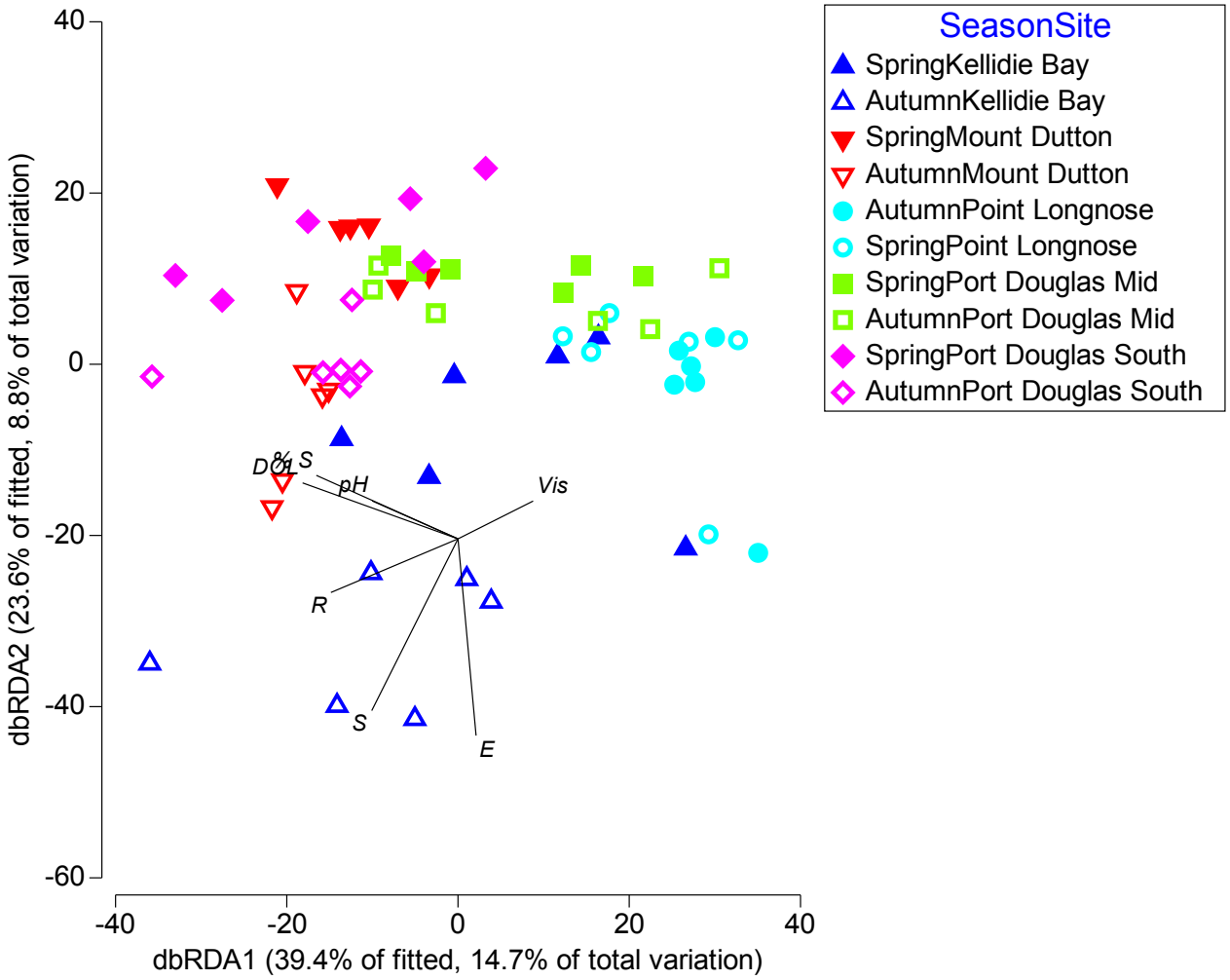

Figure 4.17: Unconstrained ordination plot constructed using distance-based redundancy analysis (dbRDA) on the results of a DistLM on multivariate fish assemblage data. The first two axes out of eight are shown, with the length and angle vectors representing the strength and direction, respectively, of the correlations of the chosen (based on DistLM) environmental variables. DOL = distance to oyster lease, Vis = visibility, % S = % seagrass habitat cover, E = effluent, R = relief, S = salinity, pH; see Table 4.8 for a summary of each environmental variable.

Table 4.11: Pearson correlation values for each of the seven chosen variables from DistLM with the first two dbRDA axes. The strongest correlation for each axis is shown in bold.

Variable	dbRDA1	dbRDA2
Effluent	0.063	**0.691**
Distance to oyster lease	**-0.545**	-0.196
Visibility	0.264	-0.133
Relief	-0.446	0.188
% Seagrass	-0.497	-0.223
Salinity	-0.303	0.603
pH	-0.302	-0.133

Synthesis

The shallow waters of Coffin Bay were sampled across two seasons, showing distinct assemblages for each of the different bays for each season. The taxa characteristic to each sampling site often differed but generally comprised temperate demersal teleosts, although decapods were commonly seen and also had influential contributions to similarities within sites. Along with seasonal impacts, the distance to oyster lease and the effluent input were also related to fish assemblages.

The small openings to the inner bays of Coffin Bay influence water currents and circulation patterns (Strutton et al. 1996; Kämpf and Ellis 2014), which contribute to differences in flushing times, water quality and turbidity between bays. Such differences may explain why the inner, more enclosed sites (KB, MD, PDS) had differences in habitat types compared to the more open, outer sites (PDM, PL; see Appendix 2D, Table 7.10), which drove the differences in fish assemblages among sites. Two variables (relief and % seagrass) relating to habitat type were included in the best model to explain variation in fish assemblages. Species preferring soft sediments (i.e. the absence of seagrass) include those characterising Point Longnose and Port Douglas Mid: bluespotted-flathead *Platycephalus speculator*, and the sand crab *Ovalipes australiensis*. While those species more commonly associated with habitats dominated by seagrass or algae, e.g. weedy whiting *Neoodax balteatus,* were observed in Kellidie Bay, Mount Dutton and Port Douglas South. Previous studies have found similar patterns in fish assemblages with significant differences based on habitat types, particularly between seagrass and unconsolidated, soft-sediment habitats (Jenkins and Wheatley 1998; Guidetti 2000). These studies also found a difference in abundance between habitats, with soft sediment areas having generally lower abundances than seagrass habitats, a result not seen in my study. Furthermore, relief is usually influenced by habitat type with sandier, soft-sediment areas tending to be flatter and less structured leading to lower relief profiles. Seagrass and algae areas are more vertically structured than soft-sediments, with more microhabitats and higher relief profiles. Previous studies have also found assemblages to be strongly influenced by relief (Heck and Orth 1980; Hyndes et al. 2003) with areas of high relief (and structural complexity) generally having greater abundance and diversity. Within the current study, I found that the assemblages changed with differing relief but found no correlation with abundances. The reason for

this lack of correlation between relief and total abundance is unknown but could be related to the generally low numbers of individuals observed across all deployments.

Proximity to effluent input was one of the strongest explanatory variables included in the model. Natural freshwater input, run-off, and stormwater input have the ability to change water quality and affect fish through changes in abundances or assemblage structure (Islam and Tanaka 2004; McKinley and Johnston 2010). However, there was no strong correlation in my study with any water quality variables, such as salinity, pH, and visibility which are commonly affected by such effluent inputs (Islam and Tanaka 2004; Brooks et al. 2006). Thus, the reason for proximity to effluent input to be strongly correlated with variation in assemblages could be related to changes in water quality not measured in the present study (e.g. dissolved oxygen, heavy metals, and viral or bacterial pathogens). Excess nutrients from anthropogenic effluents can also increase macroalgae cover through eutrophication (Ryther and Dunstan 1971; Smith 2003), leading to a subsequent increase in sand cover through smothering (Short and Wyllie-Echeverria 1996). However, there was similarly minimal evidence of such occurrences based on the correlations between measured variables (i.e. proximity to effluent input was not strongly correlated with habitat variables). Thus, the direct impacts of the various effluent inputs are unknown but did seem to affect fish assemblages. Further research on the exact sources of effluent input into the system and the composition of such inputs may help our understanding of their influence on fish assemblages.

Distance to oyster lease was also included in the best model. This variable has a small influence on assemblages which may be a result from increased boat traffic within the area, the artificial structure providing additional habitat, or changes to the benthic substrate in the vicinity (Everett et al. 1995; Forrest and Creese 2006; Forrest et al. 2009). Oyster leases can affect their surrounding ecosystem (Forrest et al. 2009) with studies on fish showing generally neutral or positive impacts in terms of increased abundance and diversity (Leguerrier et al. 2004; Erbland and Ozbay 2008; Lin et al. 2009). Lin et al. (2009) found a decrease in biomass of the fish communities of between 40 to 100 % after the removal of oyster leases within Tapong Bay, Taiwan, and a declining abundance of reef-associated fishes but an increase in pelagic and soft-sediment-associated fish species. No reef-associated species were commonly observed

at any sites in my study regardless of proximity to oyster lease, the general lack of submerged reef habitat within Coffin Bay likely contributed to these differences as such fish are not naturally present within the bays.

My fishing intensity index was not found to be correlated with the variation in fish assemblages and was thus not included in the model. Similarly, no significant difference was found between some protected and unprotected areas in Coffin Bay. Therefore, fishing activities may either not be having a significant impact on the communities observed within Coffin Bay, or the relatively young protected areas (only enforced since 2014) may not have been protected long enough for effects to manifest (Claudet et al. 2008; Edgar et al. 2014). Increasing replicate numbers in any future study may also serve to increase the power to detect differences, as assemblages within each site were quite varied as evidenced by the low within-site similarity values (Figure 4.13).

The environmental variables measured are not the only drivers influencing assemblages. Cumulatively, the variables in the best model explained approximately 40 % of the total variation observed in the fish assemblages, leaving a large portion of the variation unexplained. Furthermore, none of the variables significantly explained any variation for the total abundance of individuals observed. This lack of correlation in relation to the total abundance of fish could indicate that the anthropogenic stressors investigated do not have an effect (e.g. causing a loss of overall abundance) even at sites or areas exposed to multiple stressors. Or it could be that any strong effects occur at the individual species level but they do not add up to any net change in the sum of all individuals.

Past studies on cumulative impacts have highlighted the complex and variable nature of the effects that stressors may have upon fish and other assemblages (Crain et al. 2008; Shears and Ross 2010). Previous studies assessing fish assemblages using similar statistical techniques (i.e. DistLM) often correlated assemblages to habitat and anthropogenic-related environmental variables with similar explanatory power for their models (e.g. Moore et al. 2010, 37 %; Olsson et al. 2012, 20–50 %; Teixeira-Neves et al. 2015, 39 %). Moore et al. (2010) assessed fish assemblages in relation to habitat-related environmental variables only and found depth was a key explanatory variable,

but depths in that study ranged over a much greater distance (11–100 m) than in my study (0.5–3.5 m). Olsson et al. (2012) assessed the effect of a range of environmental variables and found temperature and salinity to be important variables in the structuring of fish assemblages. Salinity was a variable also included in the best model for my study but may not have been so strongly correlated due to the smaller overall range, i.e. 34–42 (present study) vs. 3–20 (Olsson et al. 2012). Teixeira-Neves et al. (2015) assessed anthropogenic and environmental variables in relation to reef fish communities and, similar to Moore et al. (2010), found depth to be an important explanatory variable. They also found distance to coast (a proxy for general anthropogenic influence potential) to be an important variable. Overall, these studies and the present study show the highly variable nature of fish assemblages and that the explanatory variables best explaining such variation are not always consistent across studies.

Conclusion

In summary, I found a diverse fish community within Coffin Bay that was variable and had distinct communities for each site studied. These assemblages were influenced by environmental (e.g. habitat type and relief) and anthropogenic stressors (e.g. effluent and distance to oyster lease) that served to alter the composition of species rather than affect the overall total abundance. The best model explained 37 % of the total variation and included seven out of the 11 variables measured. My results show that anthropogenic stressors are influencing the fish communities in Coffin Bay and continued monitoring should be undertaken to ensure detrimental effects do not develop over time. Further research investigating the exact effluent inputs and their composition (in terms of water quality parameters) would also be beneficial for understanding why effluent input is influencing fish assemblages.

Chapter discussion

Across the four case-studies, I have found mixed influences from different anthropogenic stressors. Fishing and subsequently protection from fishing appeared to have varying results with some areas showing significant differences in assemblages between protected and unprotected sites (Case study #1), while others showed no difference (Case study #4). These observations, however, were taken only months to years after the enforcement of zoning and as such the influence of protection may not yet have been observable on all assemblages (Edgar et al. 2014). The species that were so far benefiting from protection were not always the expected species targeted by commercial or recreational fisheries but were often those considered as by-products. Effluent from desalination was observed to be having no impact on fish assemblages but it is likely that the artificial structure built to house the diffuser was serving to create reef-like habitat that was attracting some fish to the area. Tourism activities in the form of bait and berley use in the shark-cage diving industry showed limited association with overall fish assemblages but attracted *Meuschenia hippocrepis* and *Pseudocaranx s*pp. When assessing the impacts of cumulative stressors within Coffin Bay, effluent input and proximity to oyster leases had the biggest influences on shallow-water fish communities, more so than fishing intensity or natural environmental variables.

Temporal change was a common occurrence in all case studies, including both seasonal (Case studies #2 and #4) and interannual (Case studies #1 and #3) variations. Such variations over different timescales continues (along with Chapter #3) to reinforce the need to plan and account for changes in fish assemblages through time when designing studies, and it also shows the highly variable nature of marine ecosystems. The response of fish assemblages to anthropogenic stressors was shown to varying over time for some case studies. For example, the influence of protection changed between seasons (Case study #2) and years (Case study # 1 and # 3). However, the temporal changes did not always influence the fish assemblages response to potential stressors with ADP (as the most impacted site by effluent input) being the only site that did not vary between seasons (Case study #2). Therefore, the influence of stressors may not be constant, and will vary depending on how the assemblages within an area inherently change over time. Previous research has shown that timing of stressors may influence outcomes significantly and that negative effects can be alleviated by planning when

stressors occur (Wu et al. 2017). For example, Wu et al. (2017) found that dredging during autumn had the least detrimental impact on seagrass habitats. Such methods of reducing the effects of stressors can, however, only be achieved with stressors that are pulse-type disturbances (e.g. dredging or effluent release) rather than press disturbances (e.g. climate change or increased nutrient loads; Crowe et al. 2000; Wu et al. 2017). Future research could sample at more frequent intervals to better assess whether the influences of anthropogenic stressors on fish assemblages might be reduced at some times (e.g. when fish are not spawning or identifying times there are naturally low abundances within an area).

Sampling within some protected areas was also common across the four case-studies and incorporated sites sampled from protected areas of varying ages, e.g. Port Noarlunga Reef was protected ~40 years ago vs. the upper Gulf St Vincent sanctuary zone protected for <2 years at the time of sampling. The influence of protection on fish assemblages varied across case studies, with some clear effects seen in the recently established protected areas (Case study #1), while no effect was seen in other areas that have been protected for a similar period (Case study #4). In addition to age of protected area, inherent fish communities, previous exposure to varying levels of fishing pressure, enforcement strategies, and size of the area can all contribute to the success of marine protected area (Halpern 2003; Claudet et al. 2008; Edgar et al. 2014; Kelaher et al. 2015). There has been growing research for the past few years to use protected areas to alleviate some on the effects of anthropogenic stressors (Lester et al. 2009; Lubchenco and Grorud-Colvert 2015; Roberts et al. 2017). Marine protected areas elsewhere are commonly used to mitigate the impacts of fishing (e.g. Roberts et al. 2005; Babcock et al. 2010; Kelaher et al. 2014; Coleman et al. 2015), but continued research shows they may be useful for a range of stressors (Halpern et al. 2008a; McLeod et al. 2009; Marta et al. 2012). There is potential for the protected areas sampled in my case studies to compensate for some of the influences of the stressors studied, perhaps through increased resilience (Game et al. 2009; Mellin et al. 2016). Additionally, influences from stressors may have been stronger if sampling had not incorporated protected areas (i.e. Case study #2 and #3).

Habitat-modifying stressors such as artificial structures also appeared to be one of the main influences with structures changing assemblages in Case study #2 and to a lesser extent Case study #4. A previous study (Wakefield et al. 2013) assessing the effects of anthropogenic habitat modification (in the form of rock-walls and dredge channels) found reduced diversity and abundances compared to similar natural habitats, which indicates such areas may not be providing all necessary habitat components for assemblages. Further research into the potential influences of anthropogenic structures and whether they are synergistic, antagonistic or help alleviate some of the potential influences is needed. For example a study on seawalls within harbours has shown they can be modified to increase biodiversity within the area (Browne and Chapman 2014).Understanding such interactions between stressors may be key to better protecting and managing our marine ecosystems, particularly for coastal areas which are under increasing modification from urbanisation (Lotze et al. 2006; Halpern et al. 2008b; Bulleri and Chapman 2010).

Stressors that added components to the ecosystem (such as bait and berley or effluent) also showed effects on fish assemblages in some circumstances. Such stressors may be either considered as a pulse or press depending on how they are added to the system (i.e. bait and berley added for only a limited number of days per year vs. an open license for tourism activities to be conducted every day). This length of presence could have a significant effect on its overall influence (Underwood 1994), as long-term or recurrent stressors have been shown to have various effects of over time which may be different to more isolated incidents (Hughes and Connell 1999). However, sampling stressors, particularly for multiple, cumulative stressors, over the long term is rarely done (Crain et al. 2008) and would be beneficial in many scenarios to assess the response of fish assemblages to recurrent or press stressors.

Generally, the stressors I have studied had mixed effects on the fish assemblages with no clear species or group of species being primarily affected across all studies, and with no net loss of species diversity or total abundance. Previous research has shown different anthropogenic stressors can affect multiple species and ecosystems (e.g. Crowe et al. 2000; Shears and Ross 2010; O'Gorman et al. 2012; Maxwell et al. 2013). While studies such as these have investigated several species together, other studies are

more targeted (e.g. Lusseau 2003; Melcón et al. 2012; Huveneers et al. 2013), but usually focus on charismatic megafauna. Recent research has highlighted the potential for informative analysis to be conducted using traits, where ecologically relevant traits of organisms can be used to assess the influence of anthropogenic stressors, (e.g. Barausse et al. 2011; Palkovacs et al. 2012; Coleman et al. 2015). These analyses can be useful as they provide a broader look at concepts like ecosystem services and functions that species provide rather than effects on individuals themselves or the assemblage. Trait-based analysis on the data I have collected, or for future studies on such stressors, may reveal further ways in which the ecosystems are influenced by anthropogenic stressors and how this may affect ecosystem function.

No particular habitat type or geographic location seemed to be more prone to being influenced by a stressor. Shallow areas, close to significant population centres are typically considered at increased risk of impacts from anthropogenic stressors (Halpern et al. 2008b; Gelcich et al. 2014), and I did see significant influences in my shallowest sites from multiple anthropogenic stressors (Case study #4). Many studies are focussed on reef areas (e.g. Crowe et al. 2000; Wilson et al. 2006), but there is also much literature stating the vulnerability of seagrass to a range of anthropogenic stressors (Duarte 2002; Orth et al. 2006; Grech et al. 2011). Thus, further research into these less-studied habitats (i.e. seagrass and soft sediment) may prove valuable in our overall understanding on the effects of anthropogenic stressors.

Across my case studies, I have assessed the influence of single (protection [Case study #1]), multiple (effluent, artificial structures, and protection [Case study #2]; bait and berley, and protection [Case study #3]), and cumulative (Case study #4) stressors across a range of temporal time scales and locations. The outcomes from these studies show that it can be easier to attribute variation within the assemblage to particular stressors when looking at them in isolation, but this can also limit our understanding of the total ecosystem such as when the variable assessed does not account for the differences observed. Assessing multiple and cumulative stressors is challenging and less frequently conducted (Crain et al. 2008; Stelzenmuller et al. 2010), but such studies can enable researchers to have a more complete picture of what is truly occurring within the ecosystem (e.g. Case studies #2–4).

Overall, I have found that anthropogenic stressors can have significant influences on fish assemblages and be associated with significantly changed species composition and their abundances within an area. Improving our understanding of the fish assemblages studied and their responses to different anthropogenic stressors can be used to inform management decisions. Indeed, results from these case studies will be directly used by the relevant management agencies (e.g. SA Water for Case study #2). Without knowledge of how fish assemblages respond to anthropogenic stressors we are unable to make informed management decisions, leading to potential harm for the ecosystems involved.

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Case-study #1

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Case-study #2

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Case-study #3

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Case study #4

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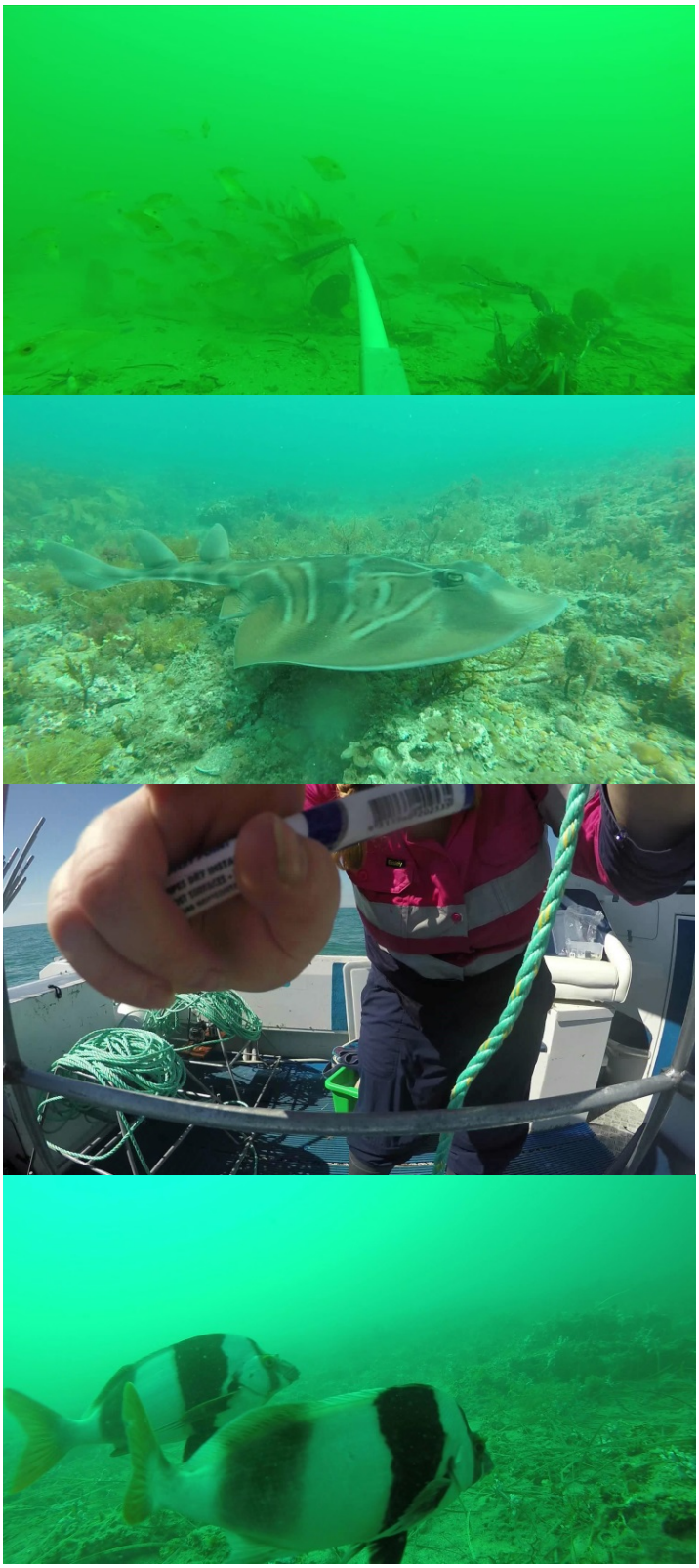

Chapter 5

What are we missing? Advantages of more than one viewpoint to estimate fish assemblages using baited video

Sasha K. Whitmarsh
Charlie Huveneers
Peter G. Fairweather

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Abstract

Counting errors can bias assessments of species abundance and richness, which can affect assessments of stock structure, population structure, and monitoring programs. Many methods for studying ecology use fixed viewpoints (e.g. camera traps, underwater video), but there is little known about how this biases the data obtained. In the marine realm, most studies using baited underwater video, a common method for monitoring fish and nekton, have previously only assessed fishes using a single bait-facing viewpoint. To investigate the biases stemming from using fixed viewpoints, we added cameras to cover 360° views around the units. We found similar species richness for all observed viewpoints but the bait-facing viewpoint recorded the highest fish abundance. Sightings of infrequently-seen and shy species increased with the additional cameras and the extra viewpoints allowed the abundance estimates of highly-abundant schooling species to be up to 60 % higher. We specifically recommend the use of additional cameras for studies focusing on shyer species or those particularly interested in increasing the sensitivity of the method by avoiding saturation in highly-abundant species. Studies may also benefit from using additional cameras to focus observation on the downstream viewpoint.

Introduction

All scientific research methods have inherent biases, and so understanding and mitigating against such biases is essential to the correct interpretation of results (Tyre et al. 2003; Elphick 2008). In many cases, biases are either ignored or merely acknowledged and, at best, their effects on the findings discussed rather than actively addressed or mitigated against. In particular, many traditional sampling methods such as using fishing gear in the marine realm have limited ways to address potential biases compared to more modern camera-based methods which can provide additional information such as behaviour, habitat and oceanographic conditions (e.g. water clarity and current). Taking action to diminish known biases may enable a better use of scarce resources, including research funds, and improve the accuracy of the data collected (Moore et al. 2014). The most likely way in which detection biases are incorporated into ecological data occurs when conducting counts of organisms (Elphick 2008; Dénes et al. 2015). Such errors can be classified into two types: 1) false negatives (leading to under-estimations of abundance) occur when some individuals cannot be detected and

counted or when the whole population of interest is not included within the sampled location; and 2) false positives (leading to over-estimations of abundance) occur through individuals erroneously being counted as being present (Tyre et al. 2003; Elphick 2008; Dénes et al. 2015). Some studies have previously attempted to address such biases. For example, biases related to species richness estimates within avian communities have been assessed using detection probabilities to determine the number of visits and grid size required to detect rare and common species (Sliwinski et al. 2015). In the marine realm, studies have addressed biases associated with fish behaviour seen while scuba-diving during underwater surveys by using bubble-free equipment, i.e. rebreathers (Lindfield et al. 2014a). Point-independence analysis has been used to assess the detectability of penguins during ship-based surveys using mark-recapture methods compared to standard strip transects (Southwell and Low 2009). However, while some methods are suitably studied to address biases, other more novel methods have not had the same level of scrutiny to potential biases inherent to the equipment or experimental design.

With increased use due to availability of low-cost technology, Baited Remote Underwater Video Stations (BRUVS), comprised of a camera mounted within a frame with bait attached, have become a popular method to assess fish assemblages over the last two decades (Whitmarsh et al. 2017). They are currently used for a wide variety of purposes (Whitmarsh et al. 2017), ranging from species-specific behavioural information (e.g. Santana-Garcon et al. 2014) to community analyses, particularly within marine protected areas (e.g. Whitmarsh et al. 2014). BRUVS are often chosen due to their non-destructive and non-extractive nature along with their ease of use, archivable footage, and replicability (Colton and Swearer 2010; Murphy and Jenkins 2010). BRUVS are suitable to assess a wide range of fish species, predominantly the larger, more mobile species and those often targeted by fishers (Colton and Swearer 2010). However, BRUVS are known to have several biases that are usually only acknowledged rather than being addressed explicitly. For example, BRUVS can be biased towards carnivorous species while being biased against smaller, more cryptic species (Colton and Swearer 2010).

Fish behaviour can affect assemblage data obtained using BRUVS if it affects differences in detection probability among species. For example, shy species might exhibit avoidance behaviour (e.g. due to increased predation risk) as a result of the fish activity surrounding the bait (Lowry et al. 2012). Rare or uncommon species might not be observed on the BRUVS, particularly if they are not piscivorous (Lowry et al. 2012), as non-piscivorous (e.g. herbivorous or planktivorous) species are generally less likely to be attracted to the usually fish-based bait. Species that are territorial or have small home ranges are also less likely to be observed on BRUVS than those that are schooling or highly mobile (Watson et al. 2010).

Along with detection biases, the standard metric used in BRUVS studies can also bias the relative count obtained. Most BRUVS studies (81 % in Whitmarsh et al. (2017)) use *MaxN* as an abundance measure. *MaxN* is the maximum number of individuals seen within a single frame (for each species) either across the entirety of the sampling period or some time within a video (e.g. each 15 min), and is considered a conservative estimate of relative abundance (Cappo et al. 2007; Farnsworth et al. 2007). *MaxN* has been shown under some circumstances to be non-linearly related to true abundance, such as when abundances at the bait are great but more fish cannot physically fit within the video frame, referred to as screen saturation (Stobart et al. 2015). Other metrics have been suggested as alternatives to *MaxN,* e.g. *MeanCount* (Schobernd et al. 2014), but they also have their own associated biases (e.g. decreased detection probability, Campbell et al. 2015) and have been shown to be similarly non-linearly related to true abundance and cause underestimations for highly-abundant species (Kilfoil et al. 2017).

Currents may also affect fish assemblages observed using BRUVS but there has been little published work investigating their influence on the assemblages observed, despite recommendations for such work to be carried out (Taylor et al. 2013). Of the research that has been conducted, studies have shown the bait plume will travel downstream and act as an attractant, so that fish will then travel upstream towards the source of the plume (Trenkel and Lorance 2011; Bacheler et al. 2014). Trenkel and Lorance (2011) also found that individuals had differing reactions when encountering a bait plume which may further complicate the ability to understand the effect of plume dispersal on fish assemblages. Thus, the direction that the BRUVS faces may influence the number of

species observed and so bias abundance estimates through missing individuals or species.

The issues surrounding the potential for counting errors and biases with BRUVS may hinder the uptake of this method by some researchers or cause widespread biases within monitoring datasets. The use of additional cameras, facing in directions other than toward the bait, could increase species richness from observations of shy individuals typically reluctant to approach the bait or through increased abundance estimates due to a greater field of view than with one camera only. This increase in field of view also allows the downstream current direction to be observed, which may lead to sighting more individuals and species as they are likely to swim upstream towards the bait. By increasing the chances of sighting more individuals and species, this modification to BRUVS may help to address some inherent biases and assist in informing the scientific community of changing technological advancements. Thus, the objective of this study is to test whether additional cameras can increase abundance and species richness estimates compared to using a single viewpoint. We aim to 1) study how communities observed on the additional viewpoints differ from the front (bait-facing) viewpoint, and determine whether additional cameras can increase 2) the observed species richness, 3) the sightings of shy or infrequently-seen species, and 4) the ability to detect differences in abundance by maximising *MaxN* estimates, e.g. when screen saturation occurs or by counting individuals not sighted on the bait-facing camera. We also investigate whether our findings are consistent across current directions and different locations.

Methods

This study was conducted within a large temperate gulf, Gulf St Vincent, located in South Australia, Australia. Gulf St Vincent is a relatively shallow (depth <30 m) coastal waterbody that acts as a large inverse estuary (Bye and Kampf 2008). A variety of habitats are contained within this gulf including high and low profile rocky reefs, large seagrass meadows, and extensive sandy or finer soft-sediment areas (Tanner 2005). Interspersed among these are numerous ship wrecks, some of which were purposefully sunk and others the result of historical accidents.

Sampling was conducted at five sites within four habitat types of Gulf St Vincent (Appendix 3, Figure 7.3). Aldinga Reef is a high-profile reef system with depths ranging from 4 to 20 m. Long Spit is a shallow (~7 m) sand bank with abundant seagrass meadows (*Posidonia* spp.). The Zanoni (a historically-significant shipwreck, sunk in 1865, protected from fishing) and The Barge (vessel purposefully sunk 1.85 km south of the Zanoni to provide an artificial reef where fishing is allowed, alleviating illegal fishing pressure on the Zanoni) are wrecks in deep (18–20 m) soft-sediment habitats, and a site called Near Zanoni (15 m) is situated in soft-sediment habitat 2 km away from the Zanoni, outside the influence of these wrecks and open to fishing. These sites were chosen due to their accessibility and distinct habitat types.

Custom-built BRUVS units were used, which consisted of a trapezoid metal frame upon which four GoPro Hero 3+ or Hero 4 cameras (set to equivalent settings and tested for field of view differences) were mounted. Each camera faced one of four directions, to the Front (facing the bait bag), each side (Left and Right), and to the Back (Figure 5.1). The opening angle of the camera underwater was calculated to be approximately 94° resulting in practicable 360° views around the BRUVS without overlapping when considering camera spacing. Each BRUVS was baited with 800 g of crushed sardines (*Sardinops sagax*) affixed to the end of the single bait arm within a mesh bag. BRUVS were deployed during daylight hours at each site over a one-month period in the austral summer of 2016. BRUVS were left on the seafloor for 60 min before retrieval. Four replicates were deployed per site. BRUVS were spaced at least 250 m apart. However, some of the BRUVS deployed around the Zanoni and the Barge were slightly closer, to a minimum of 150 m apart (similar to other studies, e.g. Gladstone et al. 2012; Lindfield et al. 2014b) because the wrecks were too small to space BRUVS units further apart without being too far from the wreck. Previous studies conducted at the same locations have shown little evidence for identifiable individuals swimming between replicates (S. K. Whitmarsh, unpublished data).

Videos were viewed using the specialised software EventMeasure (www.seagis.com.au). Fish species were identified and then counted using *MaxN*. Species richness, *MaxN* estimates and time of arrival data were calculated for each camera viewpoint. 'Infrequently seen' species were classified as species observed fewer

than three times across all replicate videos and viewpoints, while 'shy' species were classified as those that were observed to be reluctant to approach the bait, did not interact with other species, or seemed reluctant to enter open spaces. Four common species were also chosen to assess how *MaxN* estimates changed when comparing the front and additional cameras: the leatherjacket, *Thamnaconus degeni*; trevally, *Pseudocaranx* spp.; snapper, *Chrysophrys auratus*; and Port Jackson sharks, *Heterodontus portusjacksoni*. These species were selected as they were either abundant schooling species that aggregated in large numbers to feed on the bait (*T. degeni* and *Pseudocaranx* spp.), commonly observed to feed and mill around the BRUVS (*C. auratus*), or in the case of *H. portusjacksoni* have low *MaxN*, but present in higher number than *MaxN* based on the ability to identify individuals using colour patterns and size.

The video from each viewpoint was processed separately, with *MaxN* estimates recorded for each species. To estimate whether *MaxN* increases when including all four viewpoints, time at *MaxN* on the bait-facing camera was identified, with the *MaxN* observed on the other three viewpoints at that time added to calculate the maximum number of individuals sighted across all four viewpoints while avoiding double-counting. Times between cameras were synched to either the time the BRUVS entered the water or reached the seafloor.

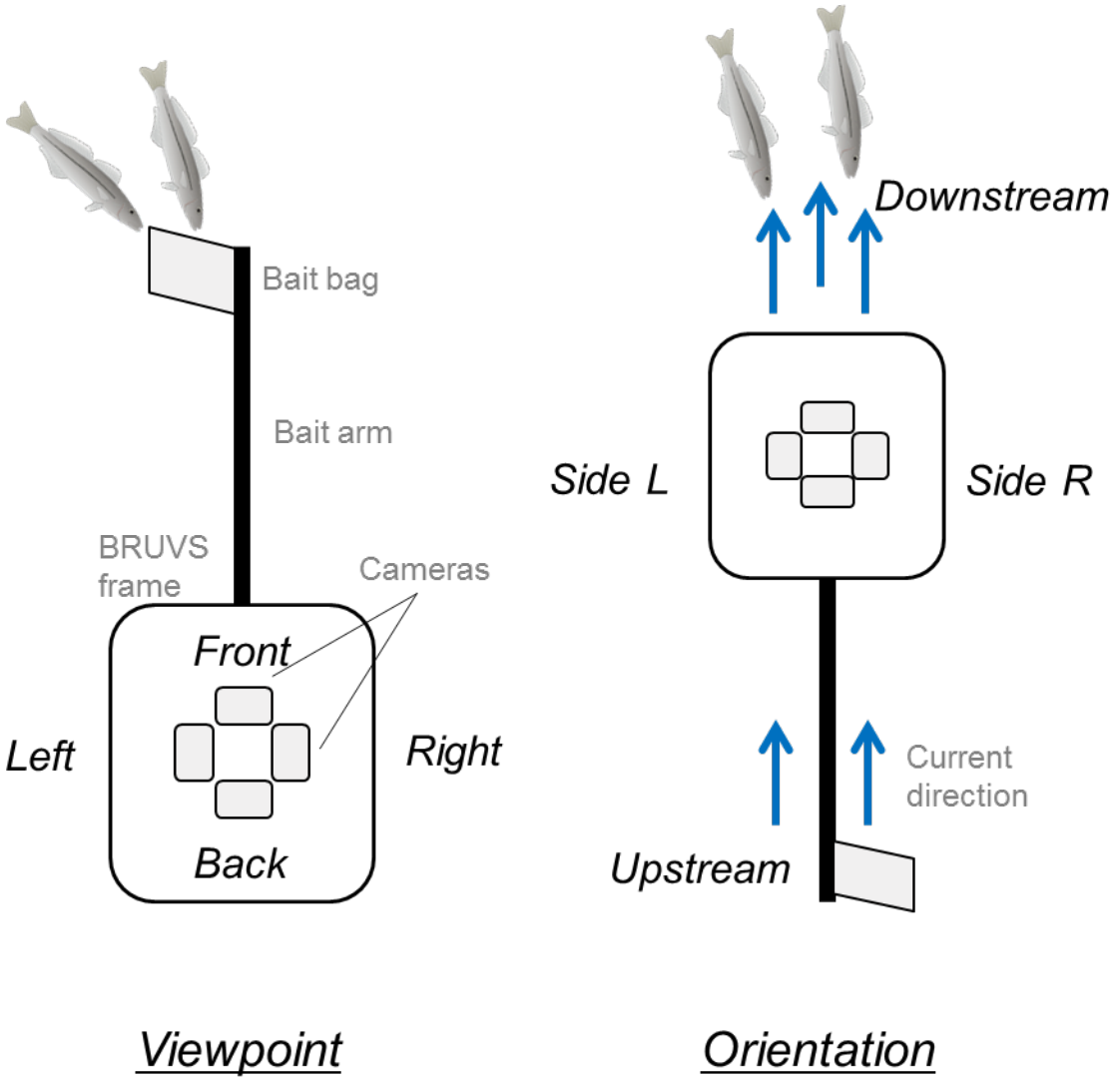

Figure 5.1: Plan view of the BRUVS set-up used, showing both the Viewpoint and Orientation (in relation to current direction) factors. Each BRUVS unit has one bait bag on a bait arm in front of one camera; the other three cameras do not face any bait. Fish (*Sillaginodes punctatus*) image source: Dieter Tracey, Integration and Application Network, University of Maryland Center for Environmental Science (ian.umces.edu/imagelibrary/)

Data analysis

Due to issues with battery longevity, two replicates had viewpoints which did not record for the whole 60 min soak time and thus were excluded from analysis (one from Aldinga Reef and one from Near Zanoni), leaving a final *N* of 18. Most statistical analysis was conducted in PRIMER v7 (Clarke and Gorley 2015) with the PERMANOVA+ add-on (Anderson et al. 2008). Multi-species assemblage data were transformed using dispersion weighting (by Site; Clarke et al. 2006) and then a square-root transform to account for the variable schooling nature of particular fish species and to down-weight the influence of highly abundant species. Raw and transformed data contributions were visualised using shade plots (Clarke et al. 2014).

To assess whether assemblages differed between viewpoints, a two-factor multivariate PERMANOVA test based on Bray-Curtis similarities was conducted on the factors Site (random factor with 5 levels) and Viewpoint (fixed factor with 4 levels). Canonical Analysis of Principal co-ordinates (CAP) analysis was also used to test for the influence of Viewpoint alone on assemblages observed and to provide a visual representation via constrained ordination of the similarities within the data. We also used a presence/absence transformation on the multivariate community data to assess whether the species composition alone was affected by Viewpoint and Site using PERMANOVA. Univariate total abundance and species richness data were used to construct Euclidean distance matrices and to subsequently test for differences among viewpoints using the same PERMANOVA model as above. Significant effects were explored further using pair-wise tests on the factor Viewpoint or the Site x Viewpoint interaction.

To assess the influence of current on the assemblages observed, current direction was determined based on the flow of particles and marine plants in front of the cameras. Each viewpoint within a BRUVS deployment was classified in relation to the observed current direction (Figure 5.1). We ran a Pearson Chi-square goodness-of-fit analysis to test whether the distribution of viewpoints across current directions differed from the expected random allocation of 25 % (i.e. by chance the Front Viewpoint would face the Downstream direction 25 % of the time). CAP analyses were

then used to assess the effect of current direction on fish assemblage (multivariate) and abundance (univariate).

To assess whether fish activity around the bait served to attract infrequently-seen or shy species we used Pearson correlations with Bonferroni probability tests for total fish abundance per replicate against the number infrequently-seen or shy species per replicate.

Results

Overall, this study observed 3,601 individuals based on *MaxN* estimates from 46 species, 38 of those being teleost fishes along with four chondrichthyans and four invertebrate species (two decapods, one cephalopod and one echinoderm; Appendix 3, Table 7.12).

Assemblages and abundances of fish

Accounting for both species abundances and composition, there was a significant difference among viewpoints determined from multivariate PERMANOVA (Viewpoint *Pseudo-F* = 2.2571, *p(perm)* = 0.035) but not from CAP analysis (p = 0.999, Figure 5.2). The CAP analysis also had a very low allocation-success rate of 11 %, implying no distinct differences across viewpoints. Pairwise PERMANOVA tests on the factor Viewpoint were unable to differentiate which pairs had a significant difference (p > 0.064, Appendix 3, Table 7.12); however, the smallest p values were recorded for the pairs involving the Front viewpoint. For presence/absence-transformed data, no significant differences were detected for taxonomic composition alone (multivariate PERMANOVA *Pseudo-F* = 0.27009, *p(perm)* = 0.974), with these trends also being consistent across the locations studied (i.e. NS Site x Viewpoint interaction *p(perm)* >0.05).

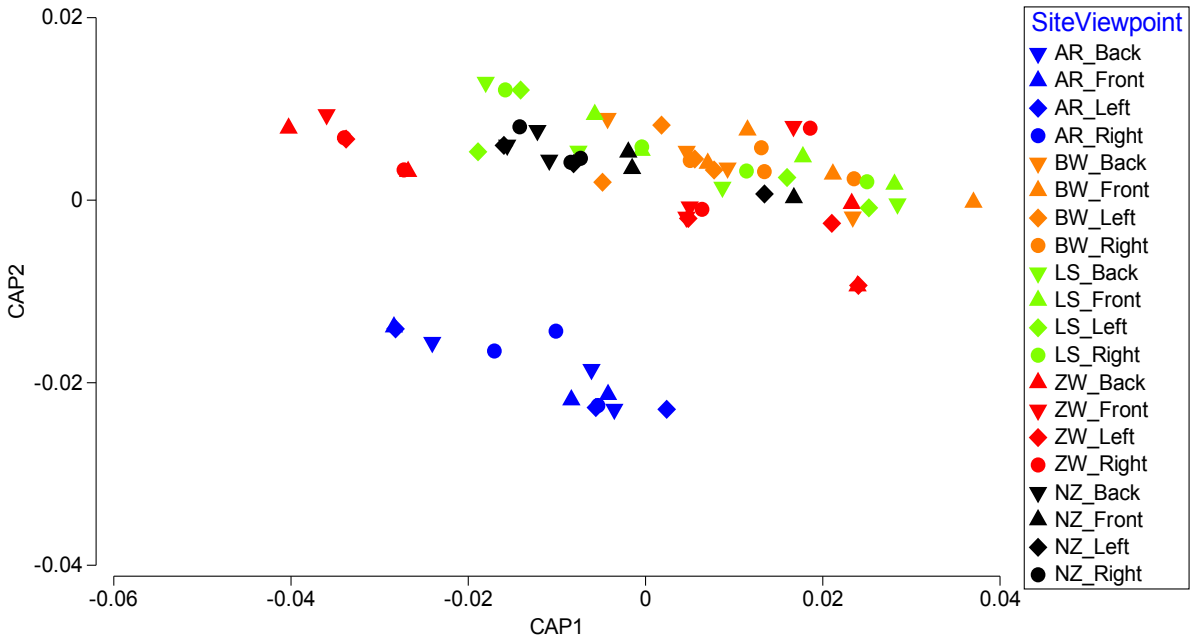

Figure 5.2: CAP constrained-ordination plot based on multivariate assemblage data showing the separation due to the factor Viewpoint alone (for *m* = 4 axes, allocation success rate = 11.1 %). Post-analysis symbols have been modified to also show the influence of sites, where AR = Aldinga Reef, BW = Barge wreck, LS = Long Spit, ZW = Zanoni wreck, NZ = Near Zanoni. Colour version available online

For total abundance (of pooled species), the Front viewpoint had the highest mean count (Figure 5.3a), and univariate PERMANOVA analysis showed similar results as above, with significant differences observed for the factor Viewpoint (*Pseudo-F* = 3.4916, *p(perm)* = 0.045) but pairwise tests were unable to differentiate which pairs were different ($p > 0.058$, Appendix 3, Table 7.13). When considering differences among Viewpoint by Site combinations, some significant differences were able to be identified (Table 5.1), with significant differences being observed between Viewpoints at Near Zanoni and the Barge, where assemblages were dominated by the highly-abundant schooling species *T. degeni* (Appendix 3, Table 7.14). The Front view was also the most likely to have the highest abundance of any viewpoint within a replicate deployment (Figure 5.4a).

Because we were unable to direct each BRUVS to face a particular current direction, we had an uneven distribution of viewpoint to current orientations, with the Front view facing the Downstream direction on three occasions, the Back on four, the Left on six, and the Right on five occasions (total $N = 18$). However, this distribution was not significantly different from the expected random placement of 25 % in each direction (Pearson Chi-square = 1.111, $p = 0.774$, df = 3). CAP detected no significant difference for the species abundance and composition observed from the different viewpoints in relation to the Current Direction ($p = 0.981$, allocation success rate = 12.5 %). Similarly, there was also no significant difference for total abundance of individuals in relation to current (CAP $p = 0.602$, allocation success rate = 4.2 %; Figure 5.3c).

Table 5.1: Pairwise PERMANOVA tests of the Viewpoint factor listed by site for univariate PERMANOVA analysis of total individuals per viewpoint, where bold values are significant (α = 0.05) Monte-Carlo *p* values whereas NS are non-significant (*p* > 0.05). Comparisons between pairs not involving the Front viewpoint (i.e. Back vs Left, Left vs Right and Back vs Right) are not shown here because they were non-significant (all *p* > 0.12).

Pairwise comparison of viewpoints	**Site**				
	Aldinga	**Barge**	**Long Spit**	**Zanoni**	**Near Zanoni**
Front vs Back	NS	NS	NS	NS	**0.035**
Front vs Left	NS	**0.008**	NS	NS	**0.003**
Front vs Right	NS	NS	NS	NS	**0.008**

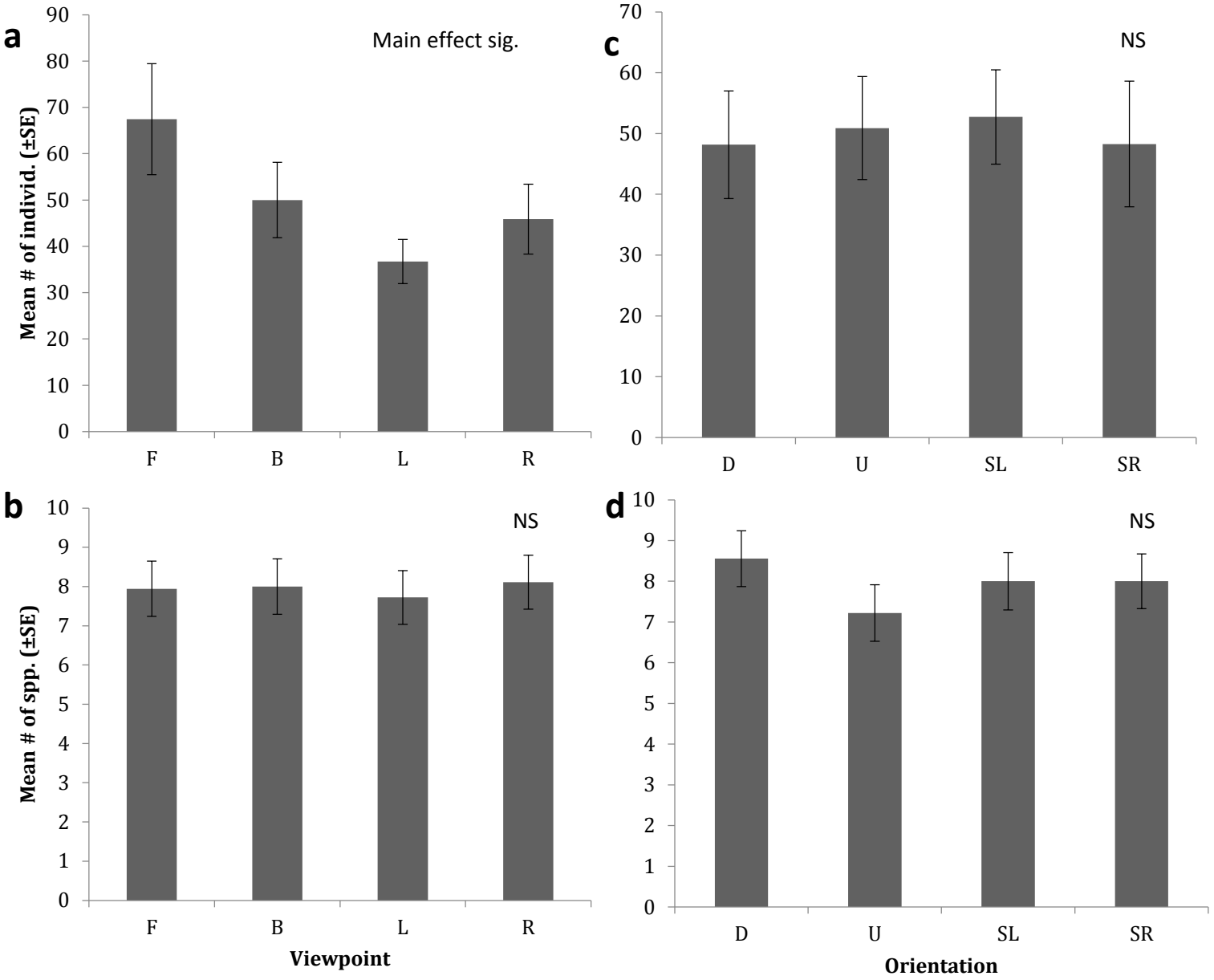

Figure 5.3: a) Mean (± SE) number of total individuals and b) mean (± SE) number of species per camera viewpoint for all sites; c) Mean (± SE) number of total individuals and d) mean (± SE) number of species observed based on the unit's relation to the direction of current for all sites. *N* = 18; F = front, B = back, L = left, R = right, D = downstream, U = upstream, SL = side left, SR = side right

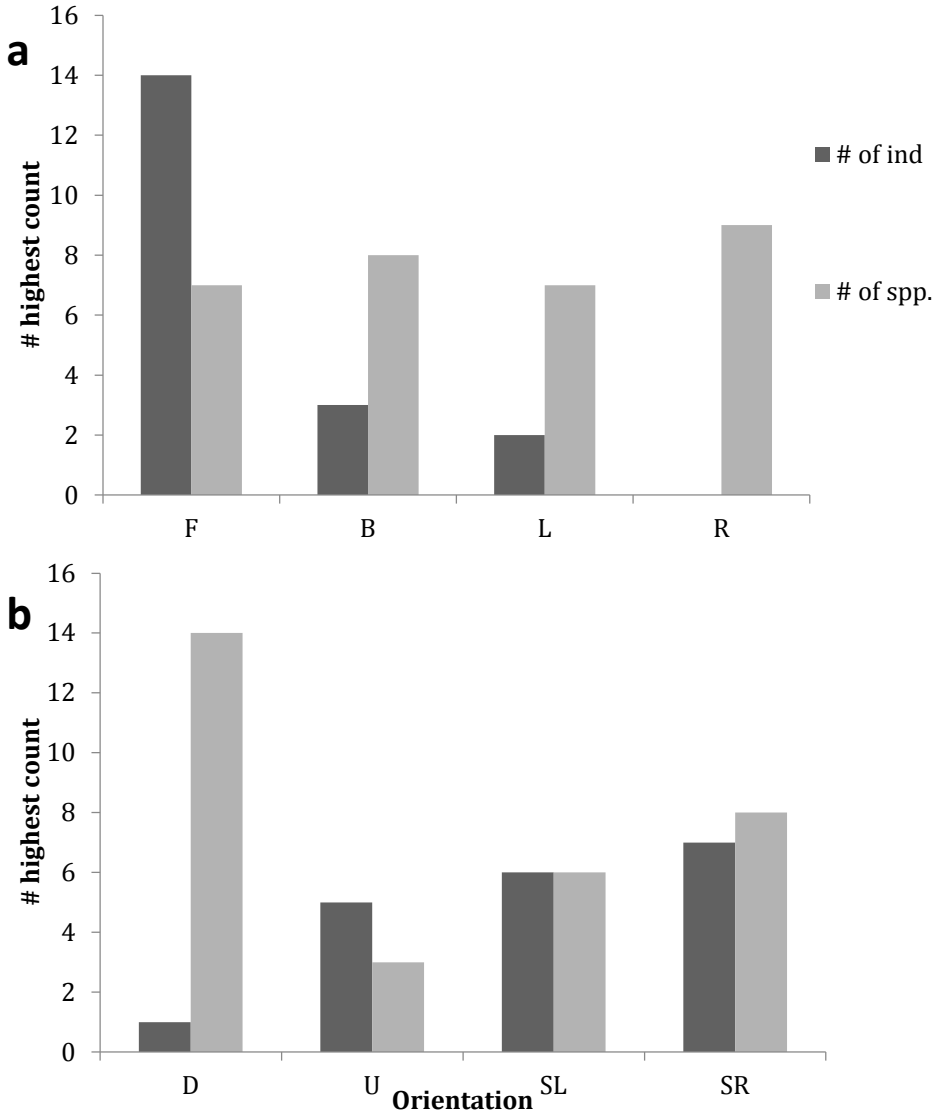

Figure 5.4: Count of the times (out of 18) that each a) viewpoint or b) current orientation within each replicate had the highest value of either number of species or number of individuals. Draws were counted for all equal Viewpoints, thus the sum of N may be greater than 18. F = front, B = back, L = left, R = right, D = downstream, U = upstream, SL = side left, SR = side right

Species richness

There were no significant differences among viewpoints in the number of species seen per Viewpoint (Figure 5.3b; univariate PERMANOVA *Pseudo-F* = 0.27368, *p(perm)* = 0.84) but cumulatively there was a moderate increase in the observed number of species when more cameras were used (mean ± SE = 7.9 ± 3.0 for one camera compared to 9.8 ± 3.5 for four cameras). When comparing the viewpoints within each replicate, there was also no clear trend for any particular viewpoint to show the highest number of species (Figure 5.4a). There was no significant difference in the number of species observed in relation to the Current Direction factor (p >0.05, allocation success rate = 27.8 %; Figure 5.3d). It was, however, more common for the Downstream direction to observe the most species compared to the other orientations within each replicate (Figure 5.4b). There was also a trend for the Downstream direction to observe a species for the first time within a replicate deployment with 48 % of first sightings occurring in that orientation compared to 9 % for the Upstream and 21 % for each side. The Downstream orientation also had the most occurrences of a species first appearing compared to other orientations with 12 out of 18 replicates (67 %) observing the most first-sightings Downstream of any orientation, while Upstream never had the most and the Side Right and Side Left had the most on three and four replicates, respectively.

Infrequently-seen or shy species

Ten species out of the total 46 were classified as infrequently seen (Table 5.2) and similar numbers of sightings of infrequently-seen species were observed across viewpoints, with slightly more being observed on the right side than other viewpoints (Figure 5.5a). There was, however, a trend for more of these species to be observed in the Downstream direction (Figure 5.5b), with approximately 4.5 % of total species sightings that were infrequently seen for the Downstream compared to 1.5 % for the Upstream, 2.1 % and 2.8 % for the Side Right and Side Left, respectively.

Table 5.2: A list of the infrequently-seen and/or shy species and the number of times each was observed from the 72 videos

Species	Infrequently seen	Shy	# of times observed
Acanthaluteres brownii	X		2
Chelmonops curiosus	X		2
Leptomithrax gaimardii	X		1
Olisthops cyanomelas	X		1
Omegophora armilla	X	X	2
Parma victoriae	X		2
Platycephalus speculator	X		3
Sepioteuthis australis	X		3
Siphonognathus radiates	X	X	1
Coscinasterias muricata	X		1
Aracana ornata		X	5
Haletta semifasciata		X	9
Neoodax balteatus		X	8
Parapercis haackei		X	35
Siphamia cephalotes		X	10
Siphonognathus sp.		X	4

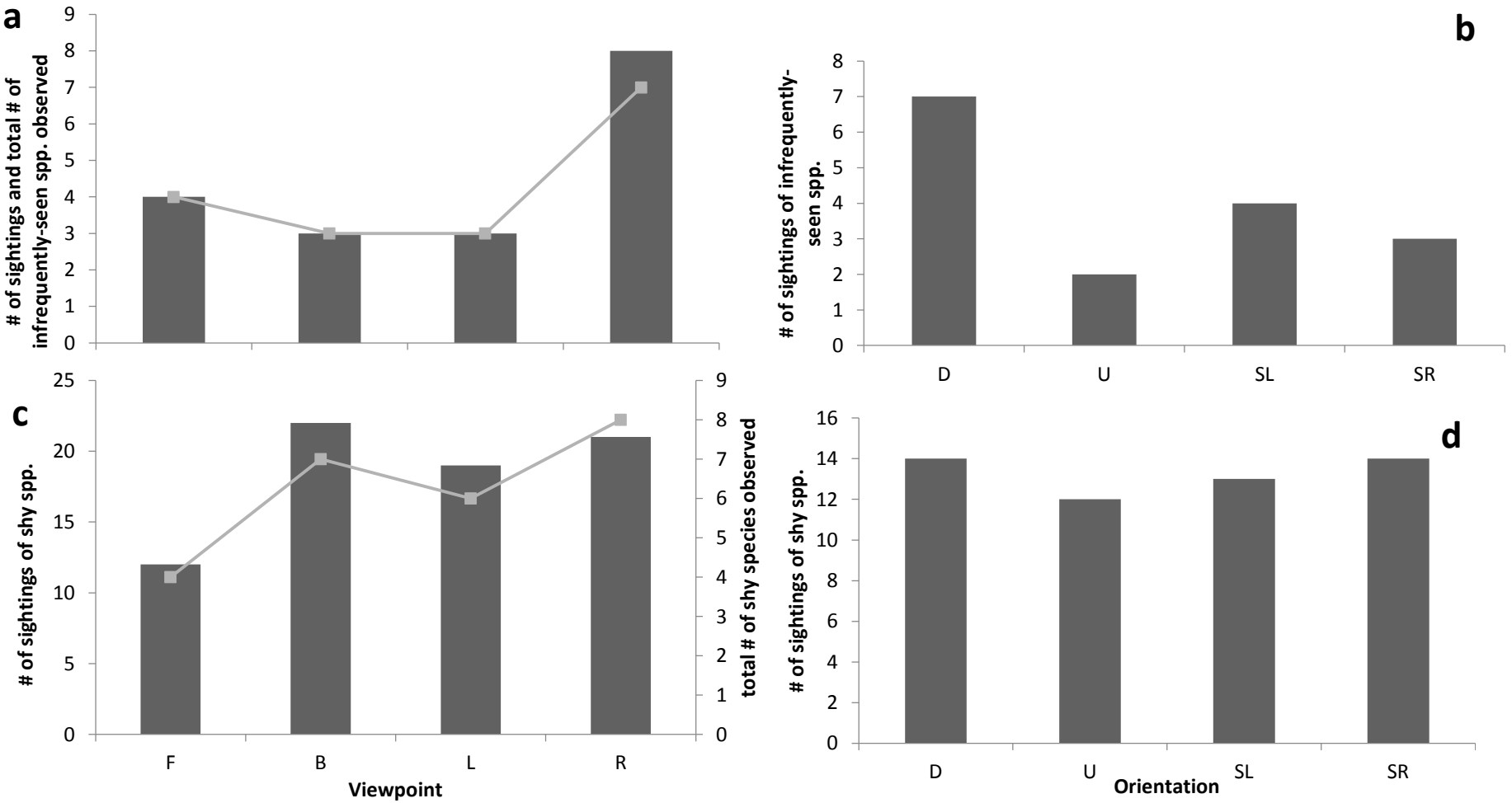

Figure 5.5: The total number of sightings of infrequently-seen species for a) Viewpoint (shown as bars), with the total number of infrequently-seen species observed from each viewpoint (shown as a line) and b) current Orientation. The total number of sightings of shy species for c) Viewpoint (shown as bars), with the total number of shy species observed from each viewpoint (shown as a line) and d) current Orientation. F = front, B = back, L = left, R = right, D = downstream, U = upstream, SL = side left, SR = side right

Eight species were considered to be shy (Table 5.2). There was a consistent trend for both lower numbers of sightings of shy species on the front camera and fewer overall shy species observed from the front viewpoint compared to all other viewpoints (Figure 5.5c). There appeared to be no influence of current direction on the number of sightings of shy species (Figure 5.5d). Pearson correlations and Bonferroni probabilities showed that neither shy species (r = 0.005 and p = 0.983) nor infrequently-seen species (r = -0.463 and p = 0.053) were significantly correlated with total fish abundance around the BRUVS.

MaxN estimates

Similar *MaxN* estimates were gained from front-only estimates compared to the cumulative total for all cameras for both *Pseudocaranx* spp. and *H. portusjacksoni* (Figure 5.6b & d). For 4 out of 6 replicates that observed *C. auratus*, the combined estimate was slightly higher than the front-only estimate (Figure 5.6c). *Thamnaconus degeni* showed the greatest difference between front and combined estimates for *MaxN* (Figure 5.6a), with some replicates having up to 60 % more individuals counted using the combined estimates than the front-only estimate but this only occurred at the highest densities.

Discussion

The use of three additional viewpoints with BRUVS allowed us to better understand and mitigate potential biases associated with the single-camera method. The additional viewpoints were able to reduce the likelihood of false negatives by increasing the chance of observing shy and infrequently-seen species, thus sampling a larger proportion of the total population in the vicinity of the BRUVS unit. The additional viewpoints also provided an increase in sensitivity for abundance estimates when assemblages became saturated on the front camera.

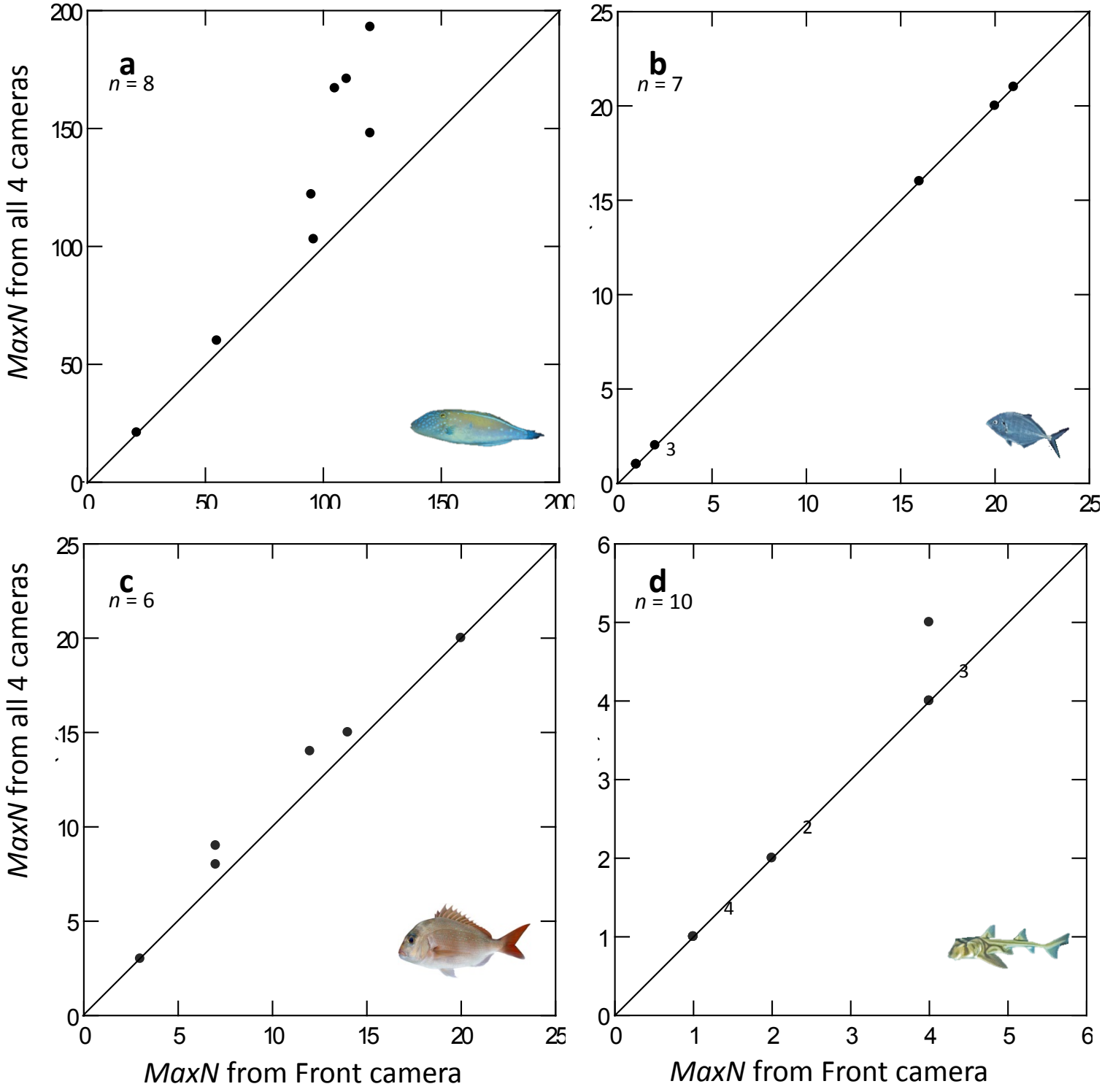

Figure 5.6: MaxN estimates for four species for the front camera only compared to all cameras combined for the species: a) leatherjacket *Thamnaconus degeni*; b) trevally *Pseudocaranx* spp.; c) snapper *Chrysophrys auratus;* d) Port Jackson shark *Heterodontus portusjacksoni*. Only replicates that had a sighting of the target species on the front camera were included in the analysis. Each point indicates a single replicate deployment with numbers above the points indicating the number of any overlapping points. Photo source: Rudie H. Kuiter, except for *C. auratus* (© State of New South Wales)

There were detectable differences among the front and additional viewpoints with most of these differences being driven by the higher abundance of some species on the front viewpoint. Attracted individuals are likely to be enticed to the bait bag and congregate within this vicinity to feed, increasing the abundance seen from the front viewpoint compared to those without bait (Taylor et al. 2013; Harasti et al. 2015). This pattern seems particularly prevalent for schooling species, especially those that commonly feed upon the bait, such as *T. degeni* (see Figure 5.6a).

The additional viewpoints had little impact on overall observed species richness, with a trend for additional cameras to only slightly increase the number of species observed. While the increase in species numbers is small, there are situations where it could be necessary for scientists to observe the maximum diversity within an area (e.g. to assess marine protected areas). Monitoring programs focussed on protected areas desire the maximal biodiversity of these areas to be recorded (Murphy and Jenkins 2010), thus enabling researchers to know which species may be benefiting from protection and conservation efforts, and having an ability to track assemblages over time.

Currents may also play an important role in determining the numbers of species observed, with the downstream direction the most likely to observe the highest species richness within a replicate deployment. The attraction of individuals towards the bait along the bait plume is likely to be the reason that more species were observed in the downstream direction. Fish (and other animals) attracted to BRUVS by the olfactory stimulus of the bait plume travel upstream towards the source (Taylor et al. 2013) and are, therefore, more likely to be observed in the downstream direction. Corroborating this, time-of-first-arrival data implied that most fish are following the bait plume to the BRUVS because the downstream direction is usually the first direction to detect a species and thus has more first sightings of species compared to other orientations and viewpoints. Therefore, facing the BRUVS downstream using divers, a modified frame design, or 360° BRUVS may also lead to a reduction in false negatives. Similar to current, proximity to structures (e.g. reef) may also affect the assemblages observed (Trenkel and Lorance 2011; Bacheler et al. 2014), by using additional viewpoints researchers can ensure any viewpoints facing structures can also be adequately sampled.

While there was no preference for any viewpoint shown by infrequently-seen species, these species were more often observed in the downstream current direction. Infrequently-seen species may be less common within an area or behaviourally disinclined to approach the BRUVS unit and thus less likely to be observed by the camera. Regardless of the reason for species being seen infrequently, there may be times when a comprehensive species list is particularly important, such as when researchers are interested in documenting the biodiversity of an area. Such is the case when assessing marine protected area performance or when targeting selected species such as in the Global FinPrint project (https://globalfinprint.org/), a large-scale study with the aim of assessing populations of sharks and rays in coral reef areas. By using sampling approaches such as additional viewpoints or deliberately facing a BRUVS downstream, researchers would be better able to use BRUVS to sample certain assemblages (such as those species that are seen infrequently) and thus reduce bias.

In contrast, shy species had a tendency to prefer viewpoints that were not the front. The high levels of activity surrounding the bait might have affected the numbers of shy species observed by the front viewpoint. Species identified as shy are often smaller and may avoid the activity around bait or are disinclined to approach due to the bait type not comprising their typical diet. Researchers interested in shyer species may benefit from extra cameras at additional viewpoints to increase the likelihood of observing these shy species and also similarly reduce the prevalence of false negatives.

A major benefit of the additional viewpoints is the ability to maximise the *MaxN* estimates for abundance and increase the sensitivity of *MaxN*, allowing for detecting differences due to factors that are not detectable using a single viewpoint. *MaxN* has been reported to have a non-linear relationship with true abundance, particularly for highly abundant species that can reach a saturation point, where more individuals cannot fit within the field of view (Schobernd et al. 2014; Stobart et al. 2015). We have found similar results from our study as by having the extra viewpoints we were able to count more individuals within the areas surrounding the BRUVS. This enabled a better representation of the abundance of a given species, especially in cases where a species was highly abundant and thus more likely to have an abundance non-linearly related to *MaxN* estimates. Therefore, additional viewpoints were able to reduce biases such as

false negatives. This may be useful in studies requiring more sensitive abundance estimates such as in marine protected areas, where researchers may wish to detect any abundance differences between closed and open areas, or to conduct stock assessments. Maximising *MaxN* estimates was most effective for highly-abundant species, such as *T. degeni*. For species with lower abundances or without a tendency to mill around the BRUVS, there are fewer benefits from using additional viewpoints to maximise *MaxN*. For example, often there were no individuals occurring on any of the other viewpoints when *MaxN* occurred at the front for *Pseudocaranx* spp. or *H. portusjacksoni* (see Figure 5.6). These results are similar to others (Campbell et al. 2015), and further emphasise that those species which have lower abundances are less affected by the non-linearity issues with *MaxN*.

A different method of maximising relative abundance estimates could also be inferred from using multiple viewpoints. The additional viewpoints can allow researchers to identify times when the highest number of individuals can be observed across all viewpoints, rather than when *MaxN* is reached on the front viewpoint. It would, however, be very time consuming with the present technology to identify when such a hypothetical maximum occurred. It could nevertheless be calculated by either watching the four viewpoint videos simultaneously to find the highest *MaxN* across all viewpoints (similar to what is done by those using stitched-together 360° videos e.g. Kilfoil et al. 2017) or to record fish numbers continuously as they enter and leave the field of view and subsequently analyse the data to find the highest *MaxN*. If advances were made in the automation of software for *MaxN* analysis, better estimates could be calculated using additional viewpoints. Advancements also in the use of commercially-available 360° video units have made it possible to use stitched-together 360° viewpoints to calculate abundances (Kilfoil et al. 2017). One of the benefits of using the method of the present study to maximise *MaxN* is that it is relatively quick and only requires approximately 15 minutes to calculate for the first species within a replicate with subsequent species taking a shorter amount of time due to less file opening and calculating synchronisation times for the videos. Comparatively, to fully analyse the videos using standard analysis techniques with non-automated software, each additional camera increases video processing time. The additional cameras also add some extra capital cost to the project for equipment and may be logistically more

challenging to organise in the field, but newly commercially-available 360° units are becoming increasingly more affordable.

Findings from this study are applicable to other methods using fixed viewpoints such as terrestrially-based camera traps. For example, O'Connor et al. (2017) investigated the use of additional cameras to survey wildlife and mesopredator activity in North America and found similar results to our study on marine fishes. Common species were equally detected using one or several cameras but infrequently-seen species were more likely to be observed through the addition of extra cameras (O'Connor et al. 2017). Recent studies have also promoted the use of remote cameras to form a global network for monitoring biodiversity (Steenweg et al. 2017). It is likely that as technology improves and costs continue to fall for photography and videography equipment, then more studies and monitoring programs will use such methods.

In conclusion, the additional cameras enabled us to better understand the biases associated with counting abundance and richness from a single viewpoint. We were able to reduce the effect of one type of counting error, false negatives, by observing additional species and individuals not present on the front viewpoint. In particular, the extra viewpoints are useful for observing infrequently-seen and shy species through the ability to view the downstream current direction and viewpoints not containing the bait and associated activity. While all optical-based surveys are likely to benefit, studies incorporating additional viewpoints may particularly benefit from use of automated video analyses that dramatically cut down video processing time (e.g. through machine learning). Additionally, 360° views are useful for those studies which are likely to have a high saturation of individuals on the front camera, and studies requiring species list and diversity estimates as comprehensive as possible.

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Chapter 6

General discussion

In the Anthropocene, concern grows about the potential influence that anthropogenic stressors may exert on marine ecosystems (Worm et al. 2006; Halpern et al. 2008b; Dirzo et al. 2014). Despite some stressors, such as climate change, receiving large amounts of research attention, other stressors are typically less well-studied. Alongside this, many marine species and ecosystems are understudied, e.g. non-commercially targeted species or in temperate regions, resulting in a paucity of information for many species and regions. The cumulative effects of multiple stressors are similarly understudied even though the combination of such stressors may be worse than each one individually (Crain et al. 2008; Halpern et al. 2008a). Throughout this PhD project, I have deployed 472 BRUVS across eight separate studies. In doing so, I have sampled at 20 different sites ranging in latitude from the South Neptune Island group (S 35.34) to the Clinton Wetlands Sanctuary Zone (S 34.16) and in longitude from Port Douglas, Coffin Bay (E 135.30) to Aldinga Reef (E 134.46). These deployments were conducted across multiple habitat types, including rocky reef, shipwrecks and other artificial reef types, sand (or other soft sediment), and seagrass, and ranged in deployment depth from very shallow (~1 m) to much deeper (~30 m). Furthermore, I have also sampled from nearshore coastal habitats (e.g. Coffin Bay) to remote offshore islands (e.g. Neptune Islands group). Deployments were conducted across multiple years (2013 – 2017) and in all seasons, along with both the daytime and nighttime being sampled. Demersal and pelagic fish assemblages were studied through the use of both conventional benthic BRUVS and more novel pelagic set-ups.

BRUVS as a method for assessing fish assemblages

As a contemporary method that is growing in popularity, BRUVS has been widely used for a variety of studies and in a variety of ways (Chapter 2). The overall suitability for it to capture a large portion of the fish assemblage (Colton and Swearer 2010; Murphy and Jenkins 2010) and the falling price of electronics has helped increase the use of this method globally. The relatively rapid uptake of this apparatus has led to many elements within the method being highly variable (Chapter 2). The under-reporting of such elements leads to issues when trying to compare results between studies and for new researchers to choose appropriate sampling designs. Thus, I developed a checklist for researchers to ensure relevant variables are described in their method sections (Chapter 2). Recent developments with BRUVS technology has led to

an expansion of studies into new areas such as the deep sea, pelagic zone (Chapter 4 – Case study #3), nighttime (Chapter 3), and 360° viewpoints (Chapter 5). However, developments within these areas are still ongoing, with studies investigating different methodological elements such as light colour at night. Contrary to other studies in Western Australia with clearer waters that showed distinct differences between light colours (Harvey et al. 2012a; Fitzpatrick et al. 2013), I found no general difference between red and blue lights for fish observed at night (Chapter 5). Such results may indicate that fish assemblages respond differently depending on the region studied. This is also hinted at by the variability in the methods used, where some researchers have found different soak times or bait types appropriate for different areas or types of assemblages studied. These discrepancies reiterate the need for pilot studies to determine and document most suitable methods. Recently, a national protocol was developed for the deployment of BRUVS within Australian Commonwealth marine reserves designed to aid in the standardisation of studies conducted in those areas (Langlois et al. 2018). This is an example of researchers acknowledging the variability amongst past studies (Chapter 2) and the need to minimise protocol differences from impacting upon the adequacy of results and our ability to compare studies.

The development of stereo-BRUVS has allowed researchers to accurately measure fish length (Murphy and Jenkins 2010). This method can be particularly useful for studies assessing protected areas, stock biomass, and identification of nursery areas (Chapter 2). Before the advent of this technology, studies aiming to investigate the length of fishes had to rely on trained experts conducting underwater visual census (Colton and Swearer 2010) or through extractive and destructive methods such as fishing and trawling, thus the development of stereo-BRUVS has been key in providing a safe, reliable method to assess the length of fish species. My thesis, however, focused on the fish community and ecological changes at the community level, which did not require information about fish length or use of stereo-BRUVS. Thus, I mostly used single-BRUVS instead of stereo set-ups. More information about the use of stereo-BRUVS is provided in Chapter 2.

While BRUVS has the ability to observe a wide range of species, gaining a better understanding of how to capture the unobserved portion of the fish community (e.g. shy

and cryptic species) is still of concern to many researchers (Langlois et al. 2010; Watson et al. 2010). Many studies have also attempted to address the inherent biases of using *MaxN* as a metric for abundance, e.g. underestimation of abundance, Sherman et al. 2018, and screen saturation, Schobernd et al. 2014; Stobart et al. 2015. I have addressed some of these issues using additional cameras to cover 360° views around the BRUVS unit (Chapter 5), which captured a greater proportion of the total fish assemblage than traditional single viewpoint BRUVS and improved counts taken of highly abundant species. Two recent studies have also begun addressing these issues but focussed on only sharks and rays. Kilfoil et al. (2017) used 360° cameras to improve *MaxN* estimates of shark abundance, while Sherman et al. (2018) used individual identifications to improve counts of rays. New technology enabling us to cover wider views is likely to be a growing area of BRUVS. The reducing costs of such technology and the advancement of machine learning to assist with video analysis will further promote its use among researchers.

The careful analysis of previous BRUVS research resulted in recommendations for more thorough reporting and potential standardised method variables (Chapter 2), further study of contentious issues with developing uses (i.e. light colour at night; Chapter 3), and novel uses of BRUVS (encompassing 360° views). Such analyses have led to substantial increase in the available knowledge of how BRUVS is being used and ways it may be improved, leading to an increase in the standard of studies using this method. By expanding the baseline of knowledge, I have provided for other users a better foundation of expectation for their own research.

Fish assemblages of coastal South Australia

Despite high rates of endemism (purportedly up to 85 %; Government of South Australia 2004) for marine species within South Australia, there still exists a lack of research for many areas, habitat types, and taxa. By selecting sites within these understudied areas and by taking a community-level approach (where all species were identified and counted that could be viewed in the field of view of the BRUVS), I have gathered a broad baseline of knowledge (Chapter 3 and 4). I counted over 25,000 individuals from at least 145 different species (Appendix 4, Table 7.15) across all my deployments (N = 472). An average *MaxN* estimate of 47 individuals of all species were

seen per field of view (taking into account individuals observed on any additional viewpoints as separate counts). Twenty-seven species were observed on only one occasion across all studies, while 60 species were observed five times or fewer. In contrast, the nine most common species were observed on 100 or more deployments and included species such as Port Jackson shark *Heterodontus portusjacksoni* (observed on 235 deployments), blue swimmer crab *Portunus armatus* (191), red mullet *Upeneichthys vlamingii* (185), trumpeter *Pelates octolineatus* (171), and trevally *Pseudocaranx* spp. (154). My sampling conducted at offshore islands showed the greatest number of rare species and those potentially undergoing range extensions (Chapter 4 – Case study #3). These results are consistent with other studies at offshore islands that noticed some species undergoing range extensions (Duffy et al. 2017), which could mean offshore islands are particularly susceptible to such incursions or the less-frequently studied nature of such places makes these sightings more novel.

Across all studies within this project, the assemblages observed were strongly associated with habitat types, with each broad habitat classification showing quite distinct communities (e.g. reef, sand, seagrass; Chapter 3–5). The influence of habitat type on the structuring of fish assemblages is well documented (Connolly 1994; Bloomfield and Gillanders 2005; Cappo et al. 2011; Saunders et al. 2014; Borland et al. 2017) but despite the likelihood of encountering distinct assemblages which react in a variety of ways, studies infrequently assess their research aim across habitats types. For example, only 23 % (out of 161) of studies assessed in Chapter 2 sampled in multiple habitat types. By assessing multiple habitat types in many of my studies (e.g. Chapter 3 and 5), I was able to see how different factors affected the different assemblages within each habitat type. For example, in Chapter 5, I was able to investigate how 360° BRUVS were able to improve the *MaxN* estimate of different species that were only found in different habitats.

Only six out of the 161 studies included in the methodological review in Chapter 2 sampled fish assemblages at night (Svane and Barnett 2008; Svane et al. 2008; Bassett and Montgomery 2011; Harvey et al. 2012b; Fitzpatrick et al. 2013; Peters et al. 2014), with an additional relevant study using BRUVS (Barker and Cowan 2018) being published after the publication of Chapter 2. Three of these studies, Bassett and

Montgomery (2011), Fitzpatrick et al. (2013), and Peters et al. (2014), assessed fish assemblages only at night and made no comparisons to daytime assemblages. Of the four remaining studies, only one, Harvey et al. (2012b), assessed assemblages in a range of habitats, similar to my Chapter 3. There were similarities between my results and these studies assessing differences between day and night assemblages, with distinct differences between the assemblages types found for all studies (Chapter 3; Svane and Barnett 2008; Svane et al. 2008; Harvey et al. 2012b; Barker and Cowan 2018). Other studies showed higher diversity for daytime assemblages compared to nighttime (Harvey et al. 2012b; Barker and Cowan 2018) but this was the case for only two out of my six sites. The ability to distinguish among habitat types was also easier for my sites, compared to those sampled in Harvey et al. (2012a), in that I was able to distinguish between assemblages in all habitats regardless of light colour or time of day. Such differences between studies highlight the variable nature of fish assemblages and their differing reactions to sampling regimes which may be based on subtle differences in behaviour. Furthering our understanding of nighttime assemblages can be key to ensuring the conservation and appropriate management of fishes. Reliable and safe ways to sample fish at night are important to quantify species that can be missed during daytime sampling and to better understand the effects of nighttime anthropogenic stressors (e.g. artificial light). Increasing our ability to sample fish at night can help better identify changes in fish community which might not be detectable in diurnal fish which help us identify our anthropocene influence.

Previous studies have assessed the effects of time of day (e.g. Chapter 3; Willis et al. 2006; Myers et al. 2016), or tides and currents (Taylor et al. 2013) on fish assemblages, but fewer studies have investigated visibility or species interactions (but see Klages et al. 2014). Visibility was an issue for some of my studies (e.g. Chapter 4 – Case study #1) and likely stems from sampling within Gulf St Vincent, which is an inverse estuary with slow flushing times (Bye and Kampf 2008), prone to turbidity and hence reduced visibility. This is potentially an issue with the use of any video technology for temperate areas and some habitat types (e.g. seagrass), which are likely to have lower visibility than areas such as coral reefs. However, counting and identifying fish present near the bait bag was still achievable and shows that such methods remain usable in areas of lowered visibility.

Our increased understanding of fish assemblages in temperate waters through the assessment of seasonal changes (Chapters 3 and 4), diurnal and yearly variation (Chapter 3), and understudied places and habitats (Chapters 3 and 4) will lead to better management of such areas. Providing baseline data for remote marine protected areas such as in the upper Gulf St Vincent, Neptune Islands, and Coffin Bay will contribute to the ongoing management of such areas, particularly as the state government does not have the capacity to monitor these areas as frequently as others, especially those closer to metropolitan Adelaide. This baseline knowledge can be used to compare with future studies to assess how assemblages are changing over time, leading to an understanding of whether management goals are being met or if changes are required (e.g. better enforcement, or increasing the size of areas).

The effect of anthropogenic stressors on fish assemblages

During the Anthropocene, concern about the potential influences that human-induced stressors may be having on the marine environment has been growing over decades (Ban et al. 2010; Gelcich et al. 2014; Robbins et al. 2017). Understanding these effects and potential mitigation techniques are key research areas (e.g. Webster and Harris 2004; Shears and Ross 2010; Wu et al. 2017). I found that stressors in South Australia may have a range of different influences on fish assemblages, with outcomes such as higher abundances of fisheries species in protected areas (Chapter 4 – Case study #1 and #3), associations of assemblages with proximity to effluent input and oyster leases for sites within Coffin Bay (Chapter 4 – Case study #4), minimal impacts from desalination brine input (Chapter 4 – Case study #2) and limited effects from bait and berley input at the Neptune Islands group (Chapter 4 – Case study #3). Stressors are likely to influence assemblages differently depending on a variety of coincidental factors including region, habitat type, and background environmental conditions (e.g. tides, currents, water temperature, salinity). These site-specific differences thus make it hard to predict effects and, along with the general difficulty of untangling and predicting effects from cumulative stressors (Crain et al. 2008; Ban et al. 2010; Stelzenmuller et al. 2010), highlight the need for continued research and monitoring. With knowledge of how stressors impact fish assemblages, management plans can then be put into place to mitigate such influences.

The use of BRUVS to assess the influence of anthropogenic stressors (apart from assessing protected areas or fishing impacts) on fish assemblages is not so common. Previous studies have, however, assessed the impact of artificial light (Barker and Cowan 2018), water quality (Gilby et al. 2016), urbanisation (Vargas-Fonseca et al. 2016), and anthropogenic habitats (rockwall and dredge channel, Wakefield et al. 2013; windfarms, Griffin et al. 2016; marinas, Bosch et al. 2017). These studies are all recent (published within the last 5 years), which shows that researchers are beginning to expand the kinds of studies in which BRUVS are being used. None of these studies considered multiple or cumulative stressors; however, Gilby et al. (2016) and Vargas-Fonseca et al. (2016), considered the influence of multiple habitat variables with an anthropogenic stressor. In contrast to my results from Case study #4, Gilby et al. (2016) found little influence from proximity to effluent input on the fish assemblages observed, with the authors considering the best explanatory variable for their study to be piscivore abundance. Considerations of variables not measured by both studies (e.g. habitat connectivity for my study and other anthropogenic stressors (such as fishing pressure) in their study) and the fact that Coffin Bay is a more enclosed system than the region that Gilby et al. (2016) studied, may have helped generate these differences. The range of anthropogenic stressors that I have assessed in Chapter 4 showcases possible ways to use BRUVS to detect influences on fish assemblages. I expect more studies in the future to use BRUVS to assess the influence of anthropogenic stressors as the need to assess impacts in a safe and repeatable way, and thus the use of BRUVS, grows.

The results of my case studies will be useful to management agencies and private industry (e.g. Department of Environment & Water, natural resource management boards, SA Water, Adelaide Aqua desalination company, white shark cage-diving industry), with some results already being used to educate and train staff from the cage-diving industry. The knowledge of which species are affected by stressors enables stakeholders to monitor these fishes, note changes in abundance, and identify flow-on impacts. The recent change of government within South Australia has prompted a review of the existing marine protected areas (sanctuary zones). Key research locations from my project will be targeted by this review (e.g. Upper Gulf St Vincent Marine Park; Chapter 4 – Case study #1), and findings from my thesis will contribute information

towards this review. The results from my desalination case study (Chapter 4 – Case study #2) are used by the regulatory agency for the management of the site and will contribute to ongoing monitoring within the area.

Sampling design and analysis

Although the number of replicate may appear to be low in some instances, I was able to detect statistical differences throughout the chapters, indicating that I had sufficient power to avoid type II errors. The standard number of deployments was assessed as part of Chapter 2, and showed that six deployments per sites was sufficient to assess fish assemblages and detect differences between sites. I therefore planned for using 6 replicates throughout my PhD.

The sampling area of a BRUVS is a contentious issue with users having different opinions about what can be considered independent (see Chapter 2). The biggest issue with independence among replicates during my PhD was in Chapter 5 where each unit was mounted with four cameras. These four cameras could obviously not be considered independent, leading to potential issues; however, I found it was necessary to determine exactly which extra viewpoints could be considered beneficial. Without mounting the four cameras to a single unit we could not have identified the improved MaxN estimates and would have had to account for replicate to replicate variability obscuring true viewpoint differences.

Count data were used in different chapters within my PhD and ranged from 0 to 349 individuals for any one species per replicate and 0 to 23 species per replicate. These counts were analysed in different ways in different chapters of my PhD according to the aims of that chapter. The range of these counts may have been modified by transforming the data (often using dispersion weighting to account for the schooling nature of some fish species) and then analysed used included Bray-Curtis similarity matrices which are appropriate for datasets with large numbers of zeroes and permutational analyses which are more robust to data not normally distributed and which can be used for unbalanced designs. Overall, this statistical approach using count data is suitable for community-based analyses such as those in this thesis and allowed me to draw appropriate conclusions in each chapter.

Directions for future studies

It is likely that the expanding use of video technology will continue as prices decrease and developments in machine learning or other approaches to rapidly identify fishes increase (Herrnstein and de Villiers 1980). While my studies have enhanced our knowledge of how BRUVS are used, the composition and abundance of temperate fish assemblages, how such assemblages respond to changes over time and different stressors, and developed novel uses of BRUVS to reduce potential biases, gaps in the research still exist. To follow on from the research conducted within this thesis, the following studies would be beneficial and are required to further our understanding of fish assemblages:

Assessing the effects of distance between deployments on the fish assemblages observed

Spacing between BRUVS deployments varies greatly between studies, with many studies failing to report the distance used (Chapter 2). Assessing how far BRUVS must be spaced to ensure independence of the resulting data would alleviate issues related to pseudoreplication. Independence between replicates can be crucial for applying appropriate statistical analyses but to what extent a particular study may suffer from this issue can be debated (Colegrave and Ruxton 2018). By assessing the overlap or lack thereof of assemblages between closely- and sparsely-spaced BRUVS, researchers can have a more accurate understanding of the potential effect of BRUVS spacing on observed fish assemblages. Such knowledge may enable better-targeted sampling of small or patchy habitat types such as isolated reefs and shipwrecks.

Optimising soak time for assemblages in southern temperate Australia

Discrepancies exist in the soak times used across studies and between the times when *MaxN* is reached (Chapter 2). Previous research has suggested soak times of 60 minutes (Unsworth et al. 2014) or 30 minutes (Harasti et al. 2015) with longer soak times recommended for pelagic BRUVS (Santana-Garcon et al. 2014). For many species in my studies, *MaxN* occurred after 30 minutes, suggesting a 60-minute soak time may be more appropriate. These discrepancies exist even when previous studies were conducted within similar (temperate, rocky reef; Harasti et al. 2015) habitats, thus conducting further research in this area will allow researchers to make better informed decisions. Optimising soak times in terms of informational output to cost is beneficial

for many researchers and can ensure that limited research funds are used effectively and not wasted on extraneous or underperforming sampling regimes.

Investigate metrics other than MaxN and maximise data output from video analyses

BRUVS deployments and data processing can be logistically difficult and expensive, thus the need to maximise the informational output from the data collected. The use of video allows for many observations and comparisons which are not routinely made or published when analysing BRUVS footage. For example, inter- and intra-specific species interactions can be recorded allowing for improved knowledge of how species interact, which species may influence the overall assemblage observed, and a better understanding of fish behaviour. Similarly, recording aggregations across species and timing of 'events' seen on the video footage (e.g. fish scattering, large schools arriving, or predators passing) may also be informative. Further analyses on the time of arrival of species, whether they feed on the bait, how long they spend at the bait, how much bait is consumed, and observations of general species behaviour are also achievable. These additional variables can be used to further our understanding of fish behaviour and how the assemblage observed on each deployment could have been affected by the species composition. Some have potential to be turned into new metrics for repeatable and routine use.

Further research on cumulative influences

Continued research on cumulative influences would be beneficial as not many studies attempt to tease apart different stressors. This can be done through careful planning of which sites to monitor and comparing between control and impact sites that differ in the intensity of influence or stressor types. Incorporating studies on the influence of timing on different stressors (e.g. Wu et al. 2017) and also how the effects vary over time may similarly prove beneficial. Examining the role that protected areas could have in alleviating the influence of potential stressors could also be useful as studies have shown protection can make assemblages more resilient to climate change (Game et al. 2009; Roberts et al. 2017). Taking into account the traits of fishes (e.g. life history characteristics and morphology) could allow for the influence of stressors to be better understood in terms of ecosystem functioning (Coleman et al. 2015; Stuart-Smith et al. 2015).

Conclusion

This thesis had the overall objective to describe under-studied fish assemblages and assess the effects of increasing anthropogenic activities in South Australia through a contemporary non-extractive monitoring method. Results for each chapter contributed to specific aims that led to outcomes and a thesis conclusion as seen in Figure 6.1. Generally, results from my studies will contribute to the sustainable management of fish assemblages, with outcomes that are applicable to many temperate fish assemblages across Australia and worldwide. My thesis contributes to our understanding of temperate fish assemblages, including rare or shy species, and has improved our ability to effectively monitor such assemblages. My project has extended and optimised the protocols of BRUVS for assessing fish assemblages in a non-extractive way. I have furthered use of BRUVS in novel ways through 360° views to assess fish assemblages. My thesis contributes data that can assist in the ongoing monitoring and future management of assemblages by assessing differences between night and day, seasonal, and yearly sampling periods, expanding understanding of habitat usage and providing baseline data for many under-studied areas. My research has focused on a wide range of species, rather than just those which are commercially targeted or observable during the day, giving empirical data about multiple marine species across a range of ecosystems and habitats. The use of BRUVS to detect the influence of anthropogenic stressors is a developing area and within this thesis I have shown its viability and versatility to be used in a range of situations, habitats, and differing stressors. The increased knowledge of how temperate fish assemblages are affected by different anthropogenic stressors will also directly lead into management of the studied areas due to close collaboration with the governing authorities.

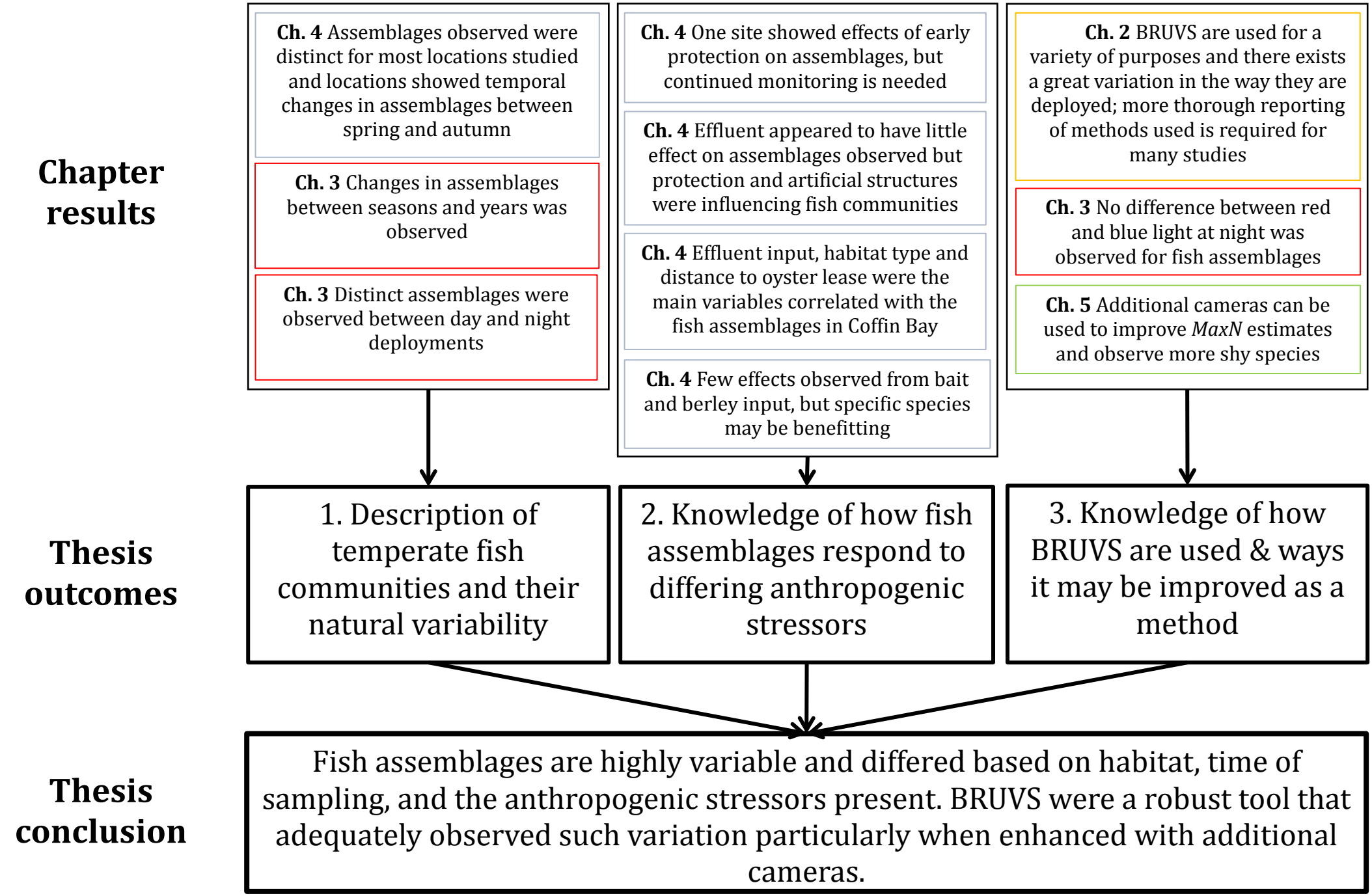

Figure 6.1: Overall thesis conclusion and main aims as outcomes highlighting the results of each chapter and how they contributed to those outcomes. Colours represent the different thesis chapters.

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Chapter 7

Appendices

Appendix 1 – Chapter 2

Table 7.1: The 161 studies used in Chapter 2 showing the method used for the 24 variables investigated. Papers were included in the analysis if published in peer-reviewed literature, bait was used in one or more replicates and if video footage was used rather than still images. The last search was conducted on the 18/07/16 using the keywords 'baited' and 'video' or 'BRUVS', on Google Scholar, Scopus, Proquest (Aquatic Sciences and Fisheries Abstracts), Biological Abstracts. Full references for the studies can be found in either the main article or at the end of this appendix

Study	Year	Country	Geographical Zone	Aquatic realm	Habitat	Name of unit	BRUVS set-up	Single or stereo	Stereo or single length data used?	Type of length measures	Max. visible range (m)	Soak time (min)	Minimum distance between replicates (m)	Bait type	Bait amount (g)	Preparation method	Bait deployment method	Depth lower limit	Depth upper limit	Depth range (m)	# of replicates	Metric	Software used	Taxa included	Standard or novel use	% identified to spp.	# of unspecifieds
Gilby et al. In press	In press	Australia	Sub-tropical	Marine	Multiple	BRUVS	Horizontal	Single	No	N/A	Unspecified	30	≤250	Sardines	Other	Chopped	Bait bag	≤5	≤5	0-10	7-9	MaxN, MaxN per trophic group, # of feeding forays on algae	None specified	Fishes	Standard	Unspecified	2
Lavaleye et al. In press	In press	Ireland	Temperate	Marine	Deepwater	Baited lander	Vertical	Single	No	Unspecified	Unspecified	>90	N/A	Sardines	Other	Whole	Bait canisters	≥100.1	≥100.1	0-10	Zero	Total counts	None specified	Crustacea, fishes, echinodermata fish + chondrichtyans + lobster	Novel	Unspecified	3
Walsh et al. In press	In press	Australia	Temperate	Marine	Rocky reef	BRUVs	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	60	≤250	Other	801-1000	Unspecified	Bait bag	≤5	20.1-50	21-50	Zero	MaxN	EventMeasure		Standard	0	3
Colefax et al. 2016	2016	Australia	Sub-tropical	Marine	Rocky reef	RUVs	Horizontal	Stereo	Yes - stereo	Fork length via stereo	2 - 4	0-29	≤150	Sardines	Unspecified	Chopped	No container	5.1-10	5.1-10	0-10	Unspecified	MaxN, T 1st, Time first fed, habitat Percent cover for habitat	EventMeasure, PhotoMeasure	Fishes, Chondrichthyans, Cephalopoda	Standard	Unspecified	3
Ghazilou et al. 2016b	2016	Iran	Sub-tropical	Marine	Coral Reef	BRUVS	Horizontal	Single	no	N/A	Unspecified	60	≤250	Unspecified	Unspecified	Unspecified	Bait bag	≤5	5.1-10	0-10	10-20	Coral point Count	N/A	Standard	N/A	4	
Ghazilou et al. 2016a	2016	Iran	Sub-tropical	Marine	Coral Reef	BRUVs	Horizontal	Single	No	N/A	2 - 4	30	Unspecified	Other	101-300	Other	Bait bag	Unspecified	Unspecified	Unspecified	4-6	T1st, MaxN	GoPro software	Fishes	Standard	<20	4
Gilby et al. 2016a	2016	Australia	Sub-tropical	Marine	Multiple	BRUVS	Horizontal	Single	No	N/A	Unspecified	30	≤250	Other	Other	Other	Bait bag	≤5	≤5	0-10	7-9	MaxN	None specified	Fishes	Standard	Unspecified	2
Griffin et al. 2016	2016	Ireland	Temperate	Marine	Multiple	BRUVs	Horizontal	Stereo	Yes - stereo	stereo lengths	Unspecified	60	≤150	Other fish	≤100	Unspecified	Unspecified	10.1-20	20.1-50	0-10	7-9	MaxN	EventMeasure	All mobile taxa	Standard	<30	3
Hesse et al. 2016	2016	New Zealand	Temperate	Marine	Rocky Reef	BUV	Horizontal	Single	No	N/A	Unspecified	30	≤150	Sardines	101-300	Chopped	Bait container	≤5	10.1-20	0-10	2-3	MaxN	VLC	All mobile taxa	Novel	Unspecified	2
Heyns-Veale et al. 2016	2016	South Africa	Temperate	Marine	Rocky Reef	BRUVs	Horizontal	Stereo	Yes - stereo	stereo lengths	Unspecified	60	≤350	Sardines	801-1000	Crushed	Bait container	10.1-20	50.1-100	51-100	Stratified	MaxN	EventMeasure	Fishes	Standard	Unspecified	2
McLean et al. 2016	2016	Australia	Tropical	Marine	Multiple	BRUVs	Horizontal	Stereo	Yes - stereo	stereo lengths	8 - 10	60	≤450	Sardines	801-1000	Crushed	Bait bag	5.1-10	20.1-50	11-20	Unspecified	MaxN	EventMeasure	Fishes, Chondrichthyans	Novel	<5	1
Misa et al. 2016	2016	USA (Hawaii)	Tropical	Marine	Deepwater	BotCam	Horizontal	Stereo	Yes - stereo	Fork length via stereo	Unspecified	31-59	Unspecified	Mix	501-800	Crushed	Unspecified	50.1-100	≥100.1	>100	Unspecified	T1st, MaxN, Time to MaxN	PhotoMeasure, EventMeasure, Vision Measurement System	Fishes	Novel	Targeted	4
Parker et al. 2016	2016	South Afrioa	Temperate	Marine	Rocky Reef	BRUVs	Horizontal	Stereo	Yes - stereo	stereo lengths	Unspecified	60	≤350	Sardines	801-1000	Crushed	Unspecified	Unspecified	Unspecified	Unspecified	Stratified	MaxN	EventMeasure	Fishes	Standard	Unspecified	6
Pejdo et al. 2016	2016	Croatia	Temperate	Marine	Unspecified	BRUV	Unspecified	Unspecified	No	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	4-6	Unspecified	None specified	Unspecified	Standard	Unspecified	17
Schmid et al. 2016	2016	Brazil	Tropical	Freshwater	Other	BRUV	Horizontal	Single	No	N/A	Unspecified	>90	≤250	Other	501-800	Crushed	Bait bag	≤5	≤5	Unspecified	7-9	MaxN, T1st	VLC	Fishes, Chondrichthyans	Standard	Unspecified	3
Spaet et al. 2016	2016	Saudi Arabia	Sub-tropical	Marine	Coral Reef	BRUVs	Horizontal	Single	No	N/A	Unspecified	>90	≤550	Other fish	801-1000	Crushed	Bait bag	≤5	≥100.1	>100	4-6	T1st, MaxN	Unspecified	Chondrichthyans	Novel	<30	2
Anderson and Santana-Garcon 2015	2015	Australia	Tropical	Marine	Pelagic	BRUV	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	>90	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Pelagic	Pelagic	Pelagic	10-20	MaxN	EventMeasure	Fishes, Chondrichthyans	Novel	Unspecified	8
Bacheler & Schertzer 2015	2015	USA	Sub-tropical	Marine	Unspecified	Video array	Horizontal	Single	No	N/A	Unspecified	0-29	Unspecified	Other	301-500	Unspecified	Bait bag	Unspecified	Unspecified	Unspecified	Stratified	MeanCount	None specified	Fishes, Chondrichthyans	Standard	Unspecified	8
Barley et al. 2015	2015	Australia	Tropical	Marine	Coral Reef	BRUVS	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	60	Unspecified	Sardines	801-1000	Crushed	Bait bag	Unspecified	Unspecified	Unspecified	Unspecified	Species specific - moray eel behaviour	Unspecified	One species - fish	Standard	0	8
Bornt et al. 2015	2015	Australia	Sub-tropical	Marine	Coral Reef	BRUV	Horizontal	Stereo	Yes - stereo	stereo lengths	6 - 8	60	≤250	Sardines	501-800	Crushed	Bait bag	5.1-10	20.1-50	11-20	4-6	MaxN	AIMS BRUVS, EventMeasure	Fishes, Chondrichthyans	Standard	Unspecified	1
Bouchet and Meeuwig 2015	2015	Australia	Temperate	Marine	Pelagic	Unspecified	Horizontal	Stereo	No - stereo	stereo lengths	Unspecified	>90	≤250	Sardines	>1000	Crushed	Bait container	Pelagic	Pelagic	Pelagic	4-6	MaxN	MATLAB, EventMeasure	All mobile taxa	Novel	<40	2
Campbell et al. 2015	2015	USA	Sub-tropical	Marine	Coral reef	Unspecified	Horizontal	Single	No	N/A	Unspecified	31-59	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	5.1-10	≥100.1	>100	Stratified	MeanCount, MaxN	None specified	Fishes, Chondrichthyans	Standard	Unspecified	8

Coleman et al. 2015	2015	Australia	Temperate, Sub-tropical	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	2 - 4	30	≤250	Sardines	301-500	Chopped	Bait bag	20.1-50	20.1-50	0-10	10-20	MaxN	None specified	Fishes, Chondrichthyans	Standard	Unspecified	1
De Vos et al. 2015	2015	South Africa	Temperate	Marine	Multiple	BRUVs	Horizontal	Single	No	N/A	Unspecified	60	≤550	Sardines	801-1000	Chopped	Bait container	≤5	20.1-50	21-50	Stratified	MaxN	VLC	Chondrichthyans	Standard	0	1
D'Onghia et al. 2015a	2015	International waters	Sub-tropical	Marine	Deepwater	Baited lander	Horizontal	Single	No	N/A	Unspecified	>90	N/A	Other fish	501-800	Whole	Unspecified	≥100.1	≥100.1	>100	Zero	MaxN	None specified	Fishes, Chondrichthyans	Novel	0	2
D'Onghia et al. 2015b	2015	International waters	Sub-tropical	Marine	Deepwater	Baited lander	Horizontal	Single	No	N/A	Unspecified	>90	N/A	Other fish	501-800	Whole	Unspecified	≥100.1	≥100.1	>100	Zero	MaxN	None specified	Fishes, Chondrichthyans	Novel	0	2
Ebner et al. 2015	2015	Australia	Tropical	Freshwater	Other	BRUVS	Horizontal	Single	No	N/A	Unspecified	60	≤150	Mix	Unspecified	whole	Bait bag	≤5	≤5	0-10	2-3	MaxN, T 1st	WMP or VLC	All mobile taxa	Standard	<20	2
Fitzpatrick et al. 2015	2015	Australia	Tropical	Marine	Coral reef	BRUV	Horizontal	Stereo	Yes - stereo	stereo lengths	4 - 6	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	4-6	MaxN	PhotoMeasure	Fishes, Chondrichthyans	Standard	Unspecified	10
Goetze et al. 2015	2015	Fiji	Tropical	Marine	Coral reef	BRUV	Horizontal	Stereo	No - stereo	Unspecified	6 - 8	60	≤250	Sardines	501-800	Unspecified	Bait bag	Unspecified	Unspecified	Unspecified	4-6	MaxN	EventMeasure	Fishes, Chondrichthyans	Standard	Unspecified	6
Harasti et al. 2015	2015	Australia	Temperate, Sub-tropical	Marine	Rocky reef	BRUVs	Horizontal	Single	No	N/A	2 - 4	Other	≤250	Sardines	801-1000	Crushed	Bait bag	10.1-20	20.1-50	11-20	4-6	MaxN	EventMeasure	Fishes, Chondrichthyans	Standard	Unspecified	1
Howarth et al. 2015	2015	United Kingdom	Temperate	Marine	Other	BRUV	Horizontal	Single	Yes - single	Single with reference marks for length estimates	Unspecified	60	N/A	Other fish	101-300	Chopped	Bait container	Unspecified	Unspecified	Unspecified	Zero	T1st, MaxN	None specified	Fishes, Chondrichthyans	Standard	Unspecified	5
Kelaher et al. 2015a	2015	Australia	Sub-tropical	Marine	Rocky Reef	BRUV	Horizontal	Single	No	N/A	2 - 4	30	Unspecified	Sardines	301-500	Unspecified	Bait bag	20.1-50	20.1-50	11-20	10-20	MaxN	None specified	Fishes, Chondrichthyans	Standard	Unspecified	3
Kelaher et al. 2015b	2015	Australia	Temperate	Marine	Rocky Reef	BRUV	Horizontal	Single	No	N/A	2 - 4	30	≤150	Sardines	301-500	Chopped	Bait container	5.1-10	20.1-50	21-50	4-6	MaxN	None specified	Unspecified	Standard	Unspecified	2
Langlois et al. 2015	2015	Australia	Tropical	Marine	Multiple	BRUV	Horizontal	Stereo	Yes - stereo	Stereo lengths	4 - 6	>90	Unspecified	Sardines	801-1000	Crushed	Bait bag	50.1-100	≥100.1	21-50	Unspecified	MaxN	CAL, BRUVS2.1.mdb, EventMeasure	Fishes	Novel	Targeted	2
Letessier et al. 2015	2015	New Caledonia	Tropical	Marine	Coral reef	BRUVS	Horizontal	Stereo	Yes - stereo	stereo lengths	6 - 8	60	Unspecified	Sardines	801-1000	Crushed	Bait bag	Unspecified	Unspecified	Unspecified	2-3	Lengths only	EventMeasure	Fishes, Chondrichthyans	Standard	<20	4
Malcolm et al. 2015	2015	Australia	Sub-tropical	Marine	Rocky reef	BRUV	Horizontal	Both	Yes - stereo	stereo lengths	Unspecified	30	≤250	Sardines	801-1000	Crushed	Bait bag	10.1-20	20.1-50	21-50	4-6	MaxN	EventMeasure	Snapper	Standard	Targeted	1
McLaren et al. 2015	2015	Australia	Temperate	Marine	Rocky reef	BRUVs	Horizontal	Stereo	Yes - stereo	stereo lengths	4 - 6	60	≤250	Sardines	501-800	Crushed	Unspecified	Unspecified	Unspecified	Unspecified	4-6	MaxN	EventMeasure, PhotoMeasure	Fishes, Chondrichthyans	Standard	Unspecified	5
McLean et al. 2015	2015	Australia	Temperate, Sub-tropical	Marine	Deepwater	BRUVs	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	>90	≤350	Other	801-1000	Unspecified	Bait bag	≥100.1	≥100.1	>100	7-9	MaxN	EventMeasure	Fishes, Chondrichthyans	Novel	<20	3
Pearson and Stevens 2015	2015	Australia	Sub-tropical	Marine	Rocky reef	BRUVs	Horizontal	Single	No	Single with reference marks for length estimates	2 - 4	30	≤550	Sardines	≤100	Crushed	Bait bag	20.1-50	50.1-100	51-100	4-6	MaxN	None specified	Fishes, Chondrichthyans	Standard	<10	0
Rees et al. 2015	2015	Australia	Temperate	Marine	Pelagic	RUVS	Horizontal	Single	No	N/A	Unspecified	31-59	≤450	Other	301-500	Other	Bait container	Pelagic	Pelagic	Pelagic	10-20	T1st, MaxN	VLC	Fishes, Chondrichthyans	Novel	<20	1
Roberson et al. 2015	2015	South Africa	Temperate	Marine	Multiple	BRUVs	Horizontal	Single	No	N/A	Unspecified	60	≤550	Sardines	801-1000	Crushed	Bait container	≤5	20.1-50	21-50	Unspecified	MaxN	Mac Media Player	Fishes, Chondrichthyans	Standard	0	2
Ryan et al. 2015	2015	Australia	Tropical, International waters	Marine	Multiple	BRUVS	Horizontal	Stereo	Yes - stereo	stereo lengths	4 - 6	Other	Unspecified	Sardines	801-1000	Crushed	Bait bag	≤5	50.1-100	51-100	>20	Behaviour of specific species	EventMeasure	Chondrichthyans	Standard	Unspecified	2
Schultz et al. 2015	2015	Australia	Sub-tropical	Marine	Multiple	BRUV	Horizontal	Single	No	N/A	Unspecified	30	≤250	Sardines	801-1000	Crushed	Bait bag	Unspecified	Unspecified	Unspecified	2-3	MaxN	EventMeasure	Fishes, Chondrichthyans	Standard	Unspecified	5
Scott et al. 2015	2015	Australia	Sub-tropical	Marine	Pelagic	PBRUV	Horizontal	Single	No - Single	N/A	Unspecified	31-59	Unspecified	Vegetable mix	≤100	Crushed	Bait container	Pelagic	Pelagic	Pelagic	Unspecified	MaxN	None specified	Fishes, Chondrichthyans	Novel	0	3
Stobart et al. 2015	2015	Spain	Sub-tropical	Marine	Multiple	BUV	Vertical	Single	No	N/A	Unspecified	>90	Unspecified	Sardines	301-500	Crushed	Bait bag	50.1-100	50.1-100	11-20	>20	MaxN per time	None specified	Lobsters	Novel	Targeted	2
Tanner and Williams 2015	2015	Australia	Temperate	Marine	Other	BRUV	Vertical	Single	No	N/A	Unspecified	30	≤250	Other	301-500	Crushed	Bait bag	5.1-10	20.1-50	11-20	4-6	MaxN	None specified	All mobile taxa	Standard	<30	1
Terres et al. 2015	2015	Australia	Sub-tropical	Marine	Multiple	BUV	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	31-59	≤250	Sardines	501-800	Crushed	Bait bag	10.1-20	20.1-50	11-20	>20	Species specific	None specified	Snapper	Standard	Targeted	2
Trobbiani and Venerus 2015	2015	Argentina	Temperate	Marine	Multiple	MBUV	Vertical	Single	No	Single using mirrors	>10	0-29	≤150	Other	101-300	Unspecified	Bait container	≤5	10.1-20	11-20	2-3	MaxN variant	None specified	Fishes	Novel	Unspecified	2
Anderson and Bell 2014	2014	Canada	Temperate	Marine	Other	observatory	Vertical	Single	No	N/A	Unspecified	Other	N/A	Other	>1000	Whole	No container	50.1-100	50.1-100	0-10	Zero	MaxN over time	None specified	All mobile taxa	Novel	Unspecified	2
Barord et al. 2014	2014	Oceania	Tropical	Marine	Unspecified	BRUVS	Horizontal	Single	No	N/A	Unspecified	>90	Unspecified	Mix	Unspecified	Chopped	Bait bag	≥100.1	≥100.1	51-100	4-6	Individuals by colour pattern (Hotspotter)	Hotspotter	Nautilus	Novel	Targeted	4
De Vos et al. 2014	2014	South Africa	Temperate	Marine	Rocky reef	BRUV s	Horizontal	Single	No	N/A	Unspecified	60	≤250	Sardines	801-1000	Crushed	Bait container	≤5	20.1-50	21-50	10-20	*MaxN*	Apple Quicktime	Fishes, Chondrichthyans	Standard	0	1
Dunlop et al. 2015	2015	NZ	Temperate	Marine	Rocky reef	BUV	Vertical	Single	No	N/A	Unspecified	30	Unspecified	Sardines	Whole fish	Whole	Bait container	≤5	20.1-50	21-50	10-20	MaxN for snapper and behaviours	None specified	Snapper	Standard	Targeted	2
Espinoza et al. 2014	2014	Australia	Tropical	Marine	Multiple	BRUVS	Horizontal	Single	No	N/A	>10	Other	≤350	Sardines	801-1000	Crushed	Bait bag	5.1-10	≥100.1	>100	Stratified	MaxN	BRUVS1.5.mdb	Chondrichthyans	Standard	<10	0
Hannah & Blume 2014	2014	USA	Temperate	Marine	Multiple	Baited lander	Horizontal	Stereo	Yes - stereo	Stereo lengths	Unspecified	0-29	≤450	Other fish	801-1000	Chopped	Bait bag	20.1-50	≥100.1	51-100	10-20	MaxN	MATLAB, Adobe premier Pro	Fishes	Standard	Unspecified	2
Harasti et al. 2014	2014	Australia	Sub-tropical	Marine	Other	Mini-BRUV	Horizontal	Single	No	Single with reference marks for	Unspecified	Other	Unspecified	Sardines	Whole fish	Crushed	Bait bag	≤5	≤5	0-10	4-6	MaxN, T1st, Time first fed	EventMeasure	Fishes	Novel	Targeted	2

Hill et al. 2014	2014	Australia	Temperate	Marine	Multiple	BRUV	Horizontal	Stereo	No - stereo	length estimates Unspecified	Unspecified	60	Unspecified	Sardines	801-1000	Crushed	Unspecified	20.1-50	Unspecified	Unspecified	4-6	MaxN	EventMeasure	All mobile taxa	Standard	<10	6
Kelaher et al. 2014	2014	Australia	Sub-tropical	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	2 - 4	30	≤250	Sardines	301-500	Crushed	Bait bag	10.1-20	20.1-50	21-50	4-6	*MaxN*	Unspecified	Fishes, Chondrichthyans	Standard	Unspecified	2
Klages et al. 2014	2014	Australia	Sub-tropical	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	2 - 4	30	Unspecified	Sardines	301-500	Crushed	Bait bag	10.1-20	20.1-50	21-50	10-20	*MaxN*, behaviour	Unspecified	Fishes, Chondrichthyans	Standard	Unspecified	3
Letessier et al. 2013	2014	Australia	Tropical	Marine	Pelagic	Mid-water rig	Horizontal	Stereo	Yes - stereo	Stereo lengths	Unspecified	>90	Unspecified	Mix	Unspecified	Crushed	Unspecified	≤5	20.1-50	21-50	>20	T1st, MaxN	EventMeasure	Fishes, Chondrichthyans	Novel	<20	4
Lindfield et al. 2014	2014	Guam/CNMI	Tropical	Marine	Coral reef	BRUV	Horizontal	Stereo	Yes - stereo	Fork length via stereo	8 - 10	60	≤150	Other fish	801-1000	Crushed	Unspecified	5.1-10	20.1-50	11-20	4-6	*MaxN*	EventMeasure	Fishes, Chondrichthyans	Standard	Unspecified	2
Lowry et al. 2014	2014	Australia	Sub-tropical	Estuarine	Multiple	BRUV	Horizontal	Single	No	N/A	>1	30	Unspecified	Vegetable mix	Unspecified	Crushed	Bait container	≤5	10.1-20	0-10	4-6	*MaxN*	None specified	Unspecified	Standard	Unspecified	4
Morton & Gladstone 2014	2014	Australia	Sub-tropical	Estuarine	Rocky reef	BRUVS	Horizontal	Single	No	N/A	Unspecified	31-59	≤150	Sardines	Unspecified	Crushed	Bait bag	≤5	5.1-10	0-10	4-6	*MaxN*	None specified	Fishes, Chondrichthyans	Standard	<10	2
Peters et al. 2014	2014	United Kingdom	Temperate	Marine	Seagrass	BRUV s	Horizontal	Stereo	Yes - stereo	Stereo lengths	Unspecified	>90	Unspecified	Other	Unspecified	Crushed	Bait bag	≤5	5.1-10	0-10	7-9	MaxN	EventMeasure	All mobile taxa	Novel	Unspecified	4
Rizzari et al. 2014	2014	Australia	Tropical	Marine	Coral reef	BRUV	Horizontal	Single	No	N/A	Unspecified	60	Unspecified	Sardines	801-1000	Crushed	Bait bag	Unspecified	Unspecified	Unspecified	10-20	Total numbers of sharks	None specified	Chondrichthyans	Standard	Unspecified	6
Santana-Garcon et al. 2014a	2014	Australia	Sub-tropical	Marine	Pelagic	BRUV s	Horizontal	Stereo	Yes - stereo	Stereo lengths	6 - 8	>90	Unspecified	Other fish	801-1000	Chopped	Bait bag	Pelagic	Pelagic	Pelagic	2-3	MaxN per hr	EventMeasure	Fishes, Chondrichthyans	Novel	Unspecified	2
Santana-Garcon et al. 2014b	2014	Australia	Tropical	Marine	Pelagic	BRUV s	Horizontal	Stereo	Yes - stereo	Stereo lengths	2 - 4	>90	Unspecified	Sardines	501-800	Unspecified	Bait bag	Pelagic	Pelagic	Pelagic	2-3	Behaviour and length of 1 species	EventMeasure	big eyes	Novel	Targeted	2
Santana-Garcon et al. 2014c	2014	Australia	Tropical	Marine	Pelagic	BRUV s	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	>90	≤550	Sardines	501-800	Crushed	Bait bag	Pelagic	Pelagic	Pelagic	4-6	MaxN and MaxN per 15min	EventMeasure	Fishes, Chondrichthyans	Novel	<30	2
Santana-Garcon et al. 2014d	2014	Australia	Sub-tropical	Marine	Pelagic	BRUV s	Horizontal	Stereo	Yes - stereo	Fork length via stereo	6 - 8	>90	≤550	Sardines	501-800	Crushed	Bait bag	Pelagic	Pelagic	Pelagic	10-20	MaxN, range (distance of the fish to the camera)	EventMeasure	Fishes, Chondrichthyans	Novel	<20	0
Schultz et al. 2014	2014	Australia	Sub-tropical	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	2 - 4	30	≤250	Sardines	801-1000	Crushed	Bait bag	5.1-10	20.1-50	21-50	>20	*MaxN*	EventMeasure	Fishes, Chondrichthyans	Standard	<10	0
Stevens et al. 2014	2014	United Kingdom	Temperate	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	Unspecified	0-29	Unspecified	Other fish	≤100	Chopped	Bait bag	Unspecified	Unspecified	Unspecified	2-3	Modified *MaxN*	None specified	All mobile taxa	Standard	Unspecified	6
Udyawer et al. 2014	2014	Australia	Tropical	Marine	Coral Reef	BRUVS	Horizontal	Single	No	N/A	Unspecified	60	≤350	Sardines	801-1000	Crushed	Bait bag	≤5	≥100.1	>100	Stratified	*MaxN*	BRUVS2.1.mdb	Sea snakes	Standard	Targeted	1
Unsworth et al. 2014	2014	United Kingdom	Temperate	Marine	Seagrass	BRUV s	Horizontal	Stereo	Yes - stereo	Stereo lengths	>1	>90	≤150	Mix	Unspecified	Crushed	Bait bag	Unspecified	Unspecified	Unspecified	7-9	MaxN	EventMeasure	Fishes, Chondrichthyans, Cephalopoda	Novel	<40	4
Whitmarsh et al. 2014	2014	Australia	Temperate	Marine	Seagrass	BRUVS	Horizontal	Single	No	N/A	Unspecified	60	≤250	Sardines	301-500	Crushed	Bait bag	≤5	≤5	0-10	4-6	*MaxN*	EventMeasure	All mobile taxa	Standard	<20	1
Ebner & Morgan 2013	2013	Australia	Tropical	Freshwater	Other	BRUVS	Horizontal	Single	No	N/A	8 - 10	60	≤150	Mix	Unspecified	Whole	Bait bag	≤5	5.1-10	0-10	2-3	MaxN, T1st, MaxN per 10 min	None specified	Fishes	Novel	Unspecified	2
Fitzpatrick et al. 2013	2013	Australia	Sub-tropical	Marine	Coral reef	BRUV	Horizontal	Stereo	No - stereo	Stereo lengths	2 - 4	60	≤250	Sardines	801-1000	Crushed	Bait bag	10.1-20	20.1-50	0-10	4-6	MaxN	EventMeasure	Fishes, Chondrichthyans	Novel	Unspecified	1
Folpp et al. 2013	2013	Australia	Sub-tropical	Marine	Multiple	BRUV	Horizontal	Single	No	N/A	>1	30	Unspecified	Vegetable mix	Unspecified	Crushed	Bait container	≤5	10.1-20	0-10	4-6	MaxN, T1st	None specified	Fishes, Chondrichthyans	Standard	<5	2
Gardner & Struthers 2013	2013	NZ	Temperate	Marine	Rocky Reef	BUV	Vertical	Single	No	Single with reference marks for length estimates	Unspecified	30	Unspecified	Mix	101-300	Whole	Bait container	5.1-10	10.1-20	0-10	4-6	Bcmax	Sigmascan Pro 4.0	Blue cod	Standard	Targeted	2
Hardinge et al. 2013	2013	Australia	Temperate	Marine	Rocky Reef	BRUV s	Horizontal	Stereo	Yes - stereo	Stereo lengths	Unspecified	60	≤550	Sardines	Other	Crushed	Bait bag	10.1-20	10.1-20	0-10	10-20	*MaxN*	EventMeasure	Fishes, Chondrichthyans	Standard	<5	1
Harvey et al. 2013	2013	Australia	Temperate	Marine	Multiple	BRUV s	Horizontal	Both	No - stereo	Unspecified	4 - 6	60	≤550	Sardines	801-1000	Crushed	Unspecified	Unspecified	Unspecified	Unspecified	>20	*MaxN*	None specified	Fishes, Chondrichthyans	Standard	Unspecified	6
Misa et al. 2013	2013	USA (Hawaii)	Tropical	Marine	Multiple	BotCam	Horizontal	Stereo	Yes - stereo	Stereo lengths	Unspecified	31-59	≤450	Mix	501-800	Crushed	Bait container	50.1-100	≥100.1	>100	10-20	MaxN	VF Deep Portal, Adobe Premiere Pro CS4, Visual Measurement System, PhotoMeasure	Fishes, Chondrichthyans	Novel	Unspecified	2
Moore et al. 2013	2013	USA (Hawaii)	Tropical	Marine	Deepwater	BotCam	Horizontal	Stereo	Yes - stereo	Stereo lengths	8 - 10	31-59	≤450	Mix	501-800	Chopped	Bait bag	50.1-100	≥100.1	>100	>20	MaxN	Visual Measurement System, PhotoMeasure	Fishes, Chondrichthyans	Novel	Targeted	0
Poulos et al. 2013	2013	Australia	Sub-tropical	Marine	Multiple	BRUVS	Horizontal	Single	No	N/A	Unspecified	30	≤250	Sardines	801-1000	Crushed	Unspecified	Unspecified	Unspecified	Unspecified	7-9	MaxN	EventMeasure	Fishes, Chondrichthyans	Standard	<5	5
Rees et al. 2013	2013	Australia	Sub-tropical	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	Unspecified	30	Unspecified	Sardines	301-500	Crushed	Unspecified	10.1-20	20.1-50	21-50	4-6	*MaxN*	None specified	Fishes, Chondrichthyans	Standard	Unspecified	4
Ruppert et al. 2013	2013	Australia	Tropical	Marine	Coral reef	BRUVS	Unspecified	Single	No	N/A	Unspecified	60	≤550	Unspecified	Unspecified	Unspecified	Unspecified	5.1-10	50.1-100	21-50	4-6	MaxN	None specified	Chondrichthyans	Standard	Unspecified	7
Sackett et al.	2013	USA (Hawaii)	Tropical	Marine	Deepwate	BotCam	Horizontal	Stereo	Yes -	Fork	Unspecifie	31-59	≤450	Mix	501-800	Chopped	Bait bag	50.1-100	≥100.1	>100	Unspecifie	MaxN	Visual	Fishes,	Novel	Unspecifie	3

2013					r				stereo	length via stereo	d									d		Measurement System, EventMeasure	Chondrichthyans		d	
Taylor et al. 2013	2013	Australia	Sub-tropical	Estuarine	Other	BRUV	Horizontal	Single	No	N/A	Unspecified	31-59	Unspecified	Vegetable mix	≤100	Crushed	Unspecified	≤5	5.1-10	0-10	2-3	*MaxN*	None specified	Fishes, Chondrichthyans	Standard	0	3
Wakefield et al. 2013	2013	Australia	Temperate	Marine	Multiple	Baited video	Horizontal	Single	No	N/A	Unspecified	31-59	≤150	Sardines	101-300	Crushed	Bait bag	≤5	10.1-20	11-20	2-3	*MaxN*	BRUVS version 2.1	Unspecified	Standard	Unspecified	3
White et al. 2013	2013	Australia	Tropical	Marine	Multiple	BRUVS	Horizontal	Single	No	N/A	Unspecified	Other	Unspecified	Sardines	801-1000	Crushed	Bait bag	5.1-10	≥100.1	>100	Stratified	*MaxN*	BRUVS 1.5.mdb	Chondrichthyans	Standard	Targeted	2
Wraith et al. 2013	2013	Australia	Temperate	Marine	Rocky reef	BRUVS	Horizontal	Single	No	N/A	2 - 4	30	≤250	Mix	101-300	Chopped	Bait bag	≤5	5.1-10	0-10	4-6	T1st, MaxN, other	None used	Fishes, Chondrichthyans	Standard	0	0
Aguzzi et al. 2012	2012	International waters	Temperate	Marine	Deepwater	Baited lander	Vertical	Single	No	N/A	Unspecified	>90	N/A	Other fish	801-1000	Chopped	Unspecified	≥100.1	≥100.1	>100	Zero	Individual counts	None specified	Fishes, Chondrichthyans	Novel	<20	2
Bernard & Götz 2012	2012	South Africa	Temperate	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	Unspecified	60	≤350	Sardines	801-1000	Crushed	Bait container	≤5	20.1-50	21-50	Stratified	MaxN	Adobe Premiere	Fishes, Chondrichthyans	Standard	<10	1
Birt et al. 2012	2012	Australia	Temperate	Marine	Rocky Reef	BRUVS	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	30	≤250	Sardines	801-1000	Crushed	Bait bag	5.1-10	10.1-20	0-10	Unspecified	MaxN	EventMeasure	Fishes, Chondrichthyans	Standard	Unspecified	4
Bloomfield et al. 2012	2012	United Kingdom	Temperate	Marine	Multiple	Baited Video	Horizontal	Single	No	N/A	Unspecified	61-90	Unspecified	Mix	Unspecified	Chopped	Bait bag	5.1-10	20.1-50	Unspecified	7-9	MaxN	None specified	Fishes	Standard	<30	4
Bond et al. 2012	2012	Belize	Tropical	Marine	Coral reef	BRUV s	Horizontal	Single	No	N/A	Unspecified	61-90	≥550	Other fish	801-1000	Crushed	Bait bag	5.1-10	20.1-50	11-20	Unspecified	# of sharks	None specified	Chondrichthyans	Standard	Targeted	2
Colton & Swearer 2012	2012	Australia	Temperate	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	Unspecified	60	Unspecified	Sardines	301-500	Crushed	Unspecified	≤5	20.1-50	11-20	10-20	MaxN	None specified	Fishes, Chondrichthyans	Standard	Unspecified	4
Dorman et al. 2012	2012	Australia	Sub-tropical	Marine	Coral reef	BRUV s	Horizontal	Stereo	Yes - stereo	Fork length via stereo	Unspecified	60	≤250	Mix	501-800	Crushed	Bait bag	10.1-20	20.1-50	11-20	4-6	MaxN	EventMeasure	Fishes, Chondrichthyans	Standard	<10	1
Fitzpatrick et al. 2012	2012	Australia	Tropical	Marine	Multiple	BRUVS	Horizontal	Stereo	Yes - stereo	Stereo lengths	4 - 6	60	≤250	Sardines	501-800	Crushed	Bait bag	≤5	≥100.1	>100	4-6	MaxN	BRUVS1.5.mdb	Fishes, Chondrichthyans	Standard	Unspecified	1
Gladstone et al. 2012	2012	Australia	Sub-tropical	Estuarine	Seagrass	BRUVS	Horizontal	Single	No	N/A	<2	Other	≤150	Sardines	801-1000	Chopped	Unspecified	≤5	10.1-20	0-10	4-6	MaxN, total MaxN	None specified	Fishes, Chondrichthyans	Standard	<30	1
Harvey et al. 2012	2012	Australia	Temperate	Marine	Multiple	BRUV s	Horizontal	Stereo	No - stereo	Unspecified	2 - 4	60	≤550	Sardines	801-1000	Crushed	Bait bag	≤5	20.1-50	21-50	4-6	MaxN	None specified	Fishes, Chondrichthyans	Novel	<5	1
Harvey et al. 2012 (traps)	2012	Australia	Tropical	Marine	Other	BRUVs	Horizontal	Stereo	Yes - stereo	Fork length via stereo	Unspecified	60	≤550	Sardines	801-1000	Crushed	Bait bag	20.1-50	50.1-100	11-20	Unspecified	MaxN, T1st, Time first fed, behaviour Length data for individual species	BRUVS1.5.mdb © PhotoMeasure	Fishes, Chondrichthyans	Standard	<20	2
Langlois et al. 2012a	2012	Australia	Temperate	Marine	Rocky Reef	BRUVS	Horizontal	Stereo	Yes - stereo	Fork length via stereo	4 - 6	60	≤250	Sardines	501-800	Crushed	Bait bag	≤5	50.1-100	51-100	>20		PhotoMeasure	Fishes	Standard	Targeted	0
Langlois et al. 2012c	2012	Australia	Temperate	Marine	Rocky Reef	BRUVS	Horizontal	Stereo	Yes - stereo	Fork length via stereo	4 - 6	60	≤550	Sardines	501-800	Crushed	Bait bag	5.1-10	50.1-100	51-100	10-20	MaxN	EventMeasure	Fishes, Chondrichthyans	Standard	Unspecified	1
Langlois et al. 2012b	2012	Australia	Temperate	Marine	Rocky Reef	BRUV	Horizontal	Stereo	No - stereo	Unspecified	4 - 6	60	≤250	Sardines	501-800	Crushed	Bait bag	20.1-50	20.1-50	21-50	4-6	MaxN	BRUVS1.5.mdb, PhotoMeasure	Fishes, Chondrichthyans	Standard	Unspecified	2
Lowry et al. 2012	2012	Australia	Sub-tropical	Estuarine	Multiple	BRUV	Horizontal	Single	No	N/A	>1	30	Unspecified	Vegetable mix	Unspecified	Crushed	Bait container	5.1-10	5.1-10	0-10	4-6	MaxN	BRUV tape reading interface	Fishes, Chondrichthyans, Cephalopoda	Standard	<5	2
Schultz et al. 2012	2012	Australia	Sub-tropical	Marine	Rocky Reef	BRUV	Horizontal	Single	No	N/A	Unspecified	30	≤250	Sardines	801-1000	Crushed	Bait bag	20.1-50	20.1-50	0-10	2-3	MaxN	EventMeasure	Fishes, Chondrichthyans	Standard	Unspecified	2
Zintzen et al. 2012	2012	NZ	Temperate	Marine	Deepwater	BRUV s	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	>90	≤550	Sardines	>1000	Chopped	Bait bag	20.1-50	≥100.1	>100	4-6	MaxN, T1st	EventMeasure	Fishes, Chondrichthyans	Novel	<30	2
Bassett & Montgomery 2011	2011	NZ	Temperate	Marine	Rocky reef	BUV	Horizontal	Single	No	N/A	Unspecified	30	Unspecified	Sardines	101-300	Unspecified	Bait container	5.1-10	10.1-20	0-10	>20	T1st, Other	None specified	Fishes, Chondrichthyans	Novel	Unspecified	4
Brooks et al. 2011	2011	Bahamas	Tropical	Marine	Unspecified	BRUVS	Horizontal	Single	No	Single with reference marks for length estimates	Unspecified	61-90	Unspecified	Other fish	101-300	Crushed	Bait bag	10.1-20	20.1-50	0-10	4-6	All sharks counted	Screen Calipers, Iconico Software	Chondrichthyans	Standard	0	3
Cappo et al. 2011	2011	Australia	Tropical	Marine	Multiple	BRUVS	Horizontal	Single	No	N/A	Unspecified	60	≤450	Sardines	801-1000	Crushed	Bait bag	≤5	20.1-50	11-20	Stratified	MaxN	BRUVS2.5.mdb	Fishes + Chondrichthyans, Sea snakes	Standard	Unspecified	2
Craig et al. 2011	2011	International waters	Temperate	Marine	Deepwater	Baited lander	Vertical	Single	No	Single with reference marks for length estimates	<2	60	N/A	Other fish	301-500	Unspecified	No container	≥100.1	≥100.1	>100	Zero	Total species and abundance	None specified	All mobile taxa	Novel	Unspecified	2
Dunstan et al. 2011	2011	Australia	Tropical	Marine	Rocky Reef	BRUVS	Horizontal	Single	No	N/A	Unspecified	>90	Unspecified	Other	Unspecified	Chopped	Unspecified	50.1-100	≥100.1	>100	10-20	Presence of juvenile nautilis	None specified	Nautilus	Novel	Targeted	4
Goetze et al. 2011	2011	Fiji	Tropical	Marine	Coral reef	BRUV s	Horizontal	Stereo	No - stereo	Stereo lengths	6 - 8	60	≤250	Sardines	501-800	Crushed	Bait bag	≤5	20.1-50	21-50	Unspecified	MaxN	EventMeasure	Unspecified	Standard	Unspecified	3
Gutteridge et al. 2011	2011	Australia	Sub-tropical	Marine	Multiple	BRUV	Horizontal	Single	No	N/A	Unspecified	0-29	≤150	Sardines	301-500	Unspecified	Bait container	≤5	5.1-10	0-10	10-20	MaxN	VLC	All mobile taxa	Standard	Unspecified	3
Jeffreys et al. 2011	2011	International waters	Sub-tropical	Marine	Deepwater	Baited lander	Horizontal	Single	No	N/A	Unspecified	>90	N/A	Other	>1000	Crushed	Bait canisters	≥100.1	≥100.1	>100	Zero	MaxN per time period	None specified	All mobile taxa	Novel	Unspecified	2
Lowry et al. 2011a	2011	Australia	Sub-tropical	Estuarine	Rocky Reef	BRUV	Horizontal	Single	No	N/A	>1	30	Unspecified	Vegetable mix	Unspecified	Crushed	Bait container	5.1-10	5.1-10	0-10	10-20	MaxN, T1st	Quicktime Pro®	Fishes + Chondrichthyans, cephalopods	Standard	<5	2

Lowry et al. 2011b	2011	Australia	Sub-tropical	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	Unspecified	30	Unspecified	Unspecified	Unspecified	Unspecified	Bait container	≤5	≤5	0-10	10-20	MaxN, T1st	BRUVS tape reading interface 2.1	Fishes, Chondrichthyans	Standard	0	5
Malcolm et al. 2011	2011	Australia	Sub-tropical	Marine	Multiple	BRUV	Horizontal	Single	No	N/A	2 - 4	30	≤250	Sardines	Unspecified	Crushed	Bait bag	≤5	50.1-100	Unspecified	2-3	MaxN	None specified	Fishes, Chondrichthyans	Standard	<20	2
Marouchos et al. 2011	2011	Australia	Temperate	Marine	Deepwater	DeepBRUVS	Horizontal	Single	No	Unspecified	Unspecified	>90	N/A	Vegetable mix	>1000	Crushed	Bait canisters	Unspecified	≥100.1	Unspecified	Unspecified	Gulper shark assessment	None specified	All mobile taxa	Novel	Unspecified	6
McIlwain et al. 2011	2011	Oman	Tropical	Marine	Multiple	BRUVS	Horizontal	Stereo	Yes - stereo	Stereo lengths	4 - 6	60	Unspecified	Sardines	801-1000	Unspecified	Unspecified	50.1-100	≥100.1	51-100	7-9	MaxN	PhotoMeasure, BRUVS1.5.mdb	Fishes, Chondrichthyans	Novel	<30	3
McLean et al. 2011	2011	Australia	Sub-tropical	Marine	Coral reef	BRUV s	Horizontal	Stereo	Yes - stereo	Fork length via stereo	6 - 8	60	≤250	Sardines	501-800	Crushed	Bait bag	5.1-10	20.1-50	11-20	4-6	MaxN	BRUVS1.5.mdb, EventMeasure	coral trout	Standard	Targeted	0
Merritt et al. 2011	2011	USA (Hawaii)	Tropical	Marine	Deepwater	BotCam	Horizontal	Stereo	Yes - stereo	Fork length via stereo	Unspecified	31-59	≤250	Mix	801-1000	Chopped	Bait bag	50.1-100	≥100.1	>100	10-20	MaxN, t1st	Visual Measurement System	Unspecified	Novel	Unspecified	3
Moore et al. 2011	2011	Australia	Temperate	Marine	Multiple	BRUVS	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	60	≤350	Sardines	501-800	Unspecified	Unspecified	5.1-10	50.1-100	51-100	Stratified	Other	None specified	Unspecified	Standard	Unspecified	6
Robbins et al. 2011	2011	Australia	Sub-tropical	Marine	Pelagic	Unspecified	Vertical	Single	No	N/A	Unspecified	Unspecified	Unspecified	Sardines	Whole fish	Whole	Bait container	Pelagic	Pelagic	Pelagic	>20	Shark behaviour	Unspecified	Chondrichthyans	Novel	Targeted	5
Zintzen et al. 2011	2011	NZ	Temperate	Marine	Unspecified	BRUV	Horizontal	Stereo	Yes - stereo	Unspecified	Unspecified	>90	Unspecified	Sardines	>1000	Crushed	Bait bag	20.1-50	≥100.1	>100	Unspecified	Hagfish behaviour	None specified	Eels	Novel	Targeted	5
Broad et al. 2010	2010	Australia	Temperate	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	Unspecified	30	≤150	Sardines	301-500	Crushed	Bait bag	≤5	5.1-10	0-10	10-20	MaxN	None specified	Fishes, Chondrichthyans	Standard	Unspecified	2
Chatfield et al. 2010	2010	Australia	Temperate	Marine	Multiple	BRUVS	Horizontal	Single	No	N/A	Unspecified	60	≤550	Sardines	501-800	Unspecified	Unspecified	Unspecified	Unspecified	Unspecified	>20	MaxN	None specified	Fishes, Chondrichthyans	Standard	Unspecified	7
Colton & Swearer 2010	2010	Australia	Temperate	Marine	Rocky reef	BRUV	Horizontal	Single	No	N/A	Unspecified	60	≤550	Sardines	301-500	Crushed	Bait bag	≤5	5.1-10	0-10	>20	MaxN	None specified	Fishes, Chondrichthyans	Standard	Unspecified	2
Fujii et al. 2010	2010	Japan/International waters	Sub-tropical	Marine	Deepwater	Baited lander	Vertical	Single	No	Single with reference marks for length estimates	<2	>90	N/A	Other fish	301-500	Unspecified	Unspecified	≥100.1	≥100.1	0-10	Zero	MaxN per time period	None specified	Fishes	Novel	Targeted	2
Langlois et al. 2010	2010	Australia	All	Marine	Coral reef	BRUV	Horizontal	Stereo	Yes - stereo	Fork length via stereo	4 - 6	60	≤250	Sardines	501-800	Crushed	Bait bag	5.1-10	5.1-10	0-10	7-9	MaxN	BRUVS1.5.mdb, PhotoMeasure	Fishes, Chondrichthyans	Standard	Unspecified	1
McLean et al. 2010	2010	Australia	Sub-tropical	Marine	Coral reef	BRUV	Horizontal	Stereo	Yes - stereo	Fork length via stereo	6 - 8	60	≤250	Sardines	501-800	Crushed	Bait bag	5.1-10	20.1-50	11-20	4-6	MaxN, time first fed	BRUVS1.5.mdb, EventMeasure	Redthroar emperor	Standard	Targeted	0
Moore et al. 2010	2010	Australia	Temperate	Marine	Multiple	BRUVS	Horizontal	Stereo	No - stereo	Stereo lengths	Unspecified	60	≤550	Sardines	501-800	Crushed	Bait bag	5.1-10	50.1-100	51-100	Stratified	MaxN	BRUVS1.5.mdb, PhotoMeasure	Fishes, Chondrichthyans	Standard	Unspecified	2
Ryer et al. 2010	2010	USA (Alaska)	Temperate	Marine	Other	Baited camera	Vertical	Single	No	N/A	Unspecified	0-29	Unspecified	Sardines	101-300	Whole	Bait bag	≤5	10.1-20	11-20	4-6	Total abundance	None specified	Fishes	Standard	Targeted	2
Watson et al. 2010	2010	Australia	Sub-tropical, tropical	Marine	Coral reef	BRUV	Horizontal	Stereo	Yes - stereo	Fork length via stereo	6 - 8	60	≤250	Sardines	501-800	Crushed	Bait bag	≤5	5.1-10	0-10	4-6	MaxN	AIMS BRUVS, PhotoMeasure	Fishes, Chondrichthyans	Standard	Unspecified	1
Gomelyuk 2009	2009	Australia	Tropical	Marine	Multiple	BRUVS	Horizontal	Single	No	N/A	2 - 4	60	Unspecified	Sardines	801-1000	Crushed	Bait bag	≤5	10.1-20	11-20	10-20	MaxN	None specified	Fishes, Chondrichthyans	Standard	<10	1
Jamieson et al. 2009	2009	Japan/International waters	Sub-tropical, temperate	Marine	Deepwater	Baited lander	Vertical	Single	No	Single with reference marks for length estimates	Unspecified	>90	Unspecified	Other fish	801-1000	Chopped	No container	≥100.1	≥100.1	>100	2-3	Individual decapods	None specified	Crustacea	Novel	Unspecified	3
Watson & Harvey 2009	2009	Australia	Sub-tropical	Marine	Coral reef	BRUVS	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	60	≤250	Sardines	501-800	Crushed	Bait bag	5.1-10	10.1-20	0-10	4-6	MaxN, habitat	EventMeasure	Fishes, Chondrichthyans	Standard	Unspecified	3
Watson et al. 2009	2009	Australia	Sub-tropical	Marine	Coral reef	BRUV s	Horizontal	Stereo	Yes - stereo	Fork length via stereo	6 - 8	60	≤250	Sardines	501-800	Crushed	Bait bag	5.1-10	20.1-50	11-20	4-6	MaxN	PhotoMeasure	Fishes	Standard	Targeted	0
Westera et al. 2009	2009	Australia	Temperate	Marine	Rocky reef	BRUV	Horizontal	Stereo	No - stereo	Unspecified	Unspecified	31-59	Unspecified	Unspecified	Unspecified	Unspecified	Bait bag	5.1-10	10.1-20	0-10	Unspecified	MaxN	None specified	Fishes, Chondrichthyans	Standard	0	7
Kleczkowski et al. 2008	2008	Australia	Temperate	Marine	Rocky reef	BUV	Vertical	Single	No	Single with reference marks for length estimates	Unspecified	30	≤250	Sardines	Whole fish	Whole	Bait container	Unspecified	Unspecified	Unspecified	10-20	MaxN	Sonic Foundry, Sigma Scan Pro	Fishes, Chondrichthyans	Standard	<5	4
Stoner et al. 2008	2008	USA (Alaska)	Temperate	Marine	Multiple	Baited camera	Horizontal	Single	No	N/A	Unspecified	0-29	Unspecified	Sardines	101-300	Chopped	Bait bag	≤5	≤5	0-10	4-6	T1st, Number of fish arriving, MaxN	None specified	Cod	Standard	Targeted	2
Svane & Barnett 2008	2008	Australia	Temperate	Marine	Other	Unspecified	Vertical	Single	No	N/A	Unspecified	30	Unspecified	Sardines	301-500	Whole	No container	20.1-50	10.1-20	0-10	2-3	MaxN per time period	None specified	All mobile taxa	Standard	Unspecified	4
Svane et al. 2008	2008	Australia	Temperate	Marine	Other	Unspecified	Vertical	Single	No	N/A	Unspecified	30	Unspecified	Mix	Whole fish	Whole	No container	20.1-50	20.1-50	11-20	4-6	Bait loss, MaxN per time	None specified	All mobile taxa	Standard	Unspecified	4
Bailey et al. 2007	2007	International waters	Temperate	Marine	Deepwater	Baited lander	Vertical	Single	No	Single with reference marks for length	2 - 4	>90	N/A	Other fish	Whole fish	Whole	Unspecified	≥100.1	≥100.1	0-10	Zero	Individual fish counts, swimming speed	None specified	Fishes	Novel	Targeted	1

Cappo et al. 2007a	2007	Australia	Tropical	Marine	Multiple	BRUVS	Horizontal	Both	No - stereo	Single with reference marks for length estimates Unspecified	Unspecified	31-59	≤350	Sardines	801-1000	Crushed	Bait bag	Unspecified	50.1-100	Unspecified	Stratified	MaxN	BRUVS1.5.mdb	Fishes + Chondrichthyans, Sea snakes	Standard	Unspecified	5
Harvey et al. 2007	2007	Australia	Temperate, tropical	Marine	Multiple	BRUVS	Horizontal	Both	No - stereo	Unspecified	6 - 8	60	≤550	Mix	801-1000	Crushed	Bait container	≤5	20.1-50	21-50	4-6	MaxN	None specified	Fishes, Chondrichthyans	Standard	Unspecified	2
Heagney et al. 2007	2007	Australia	Sub-tropical	Marine	Pelagic	BRUV	Horizontal	Single	No	Unspecified	>10	31-59	≥550	Vegetable mix	≤100	Crushed	Bait container	Pelagic	Pelagic	Pelagic	10-20	MaxN	None specified	Fishes, Chondrichthyans	Novel	<10	1
Malcolm et al. 2007	2007	Australia	Sub-tropical	Marine	Rocky reef	BRUVS	Horizontal	Single	No	N/A	2 - 4	30	≤250	Sardines	Unspecified	Crushed	Bait bag	10.1-20	20.1-50	11-20	7-9	MaxN	None specified	Fishes, Chondrichthyans	Standard	<5	1
Smale et al. 2007	2007	Antarctica	Polar	Marine	Other	Unspecified	Vertical	Single	No	N/A	Unspecified	>90	Unspecified	Other fish	101-300	Chopped	Unspecified	10.1-20	10.1-20	0-10	4-6	T1st, residence time, # feeding on bait, bait mass lost	None specified	All mobile taxa	Novel	<10	4
Stobart et al. 2007	2007	France/Spain	Sub-tropical	Marine	Rocky reef	BUV	Horizontal	Single	No	Single with reference marks for length estimates	2 - 4	30	Unspecified	Mix	Other	Crushed	Bait bag	5.1-10	10.1-20	0-10	2-3	MaxN	ScreenGet v1.0	Fishes, Chondrichthyans	Novel	<10	1
Watson et al. 2007	2007	Australia	Sub-tropical	Marine	Rocky reef	BRUV s	Horizontal	Stereo	No - stereo	Unspecified	8 - 10	60	≤250	Sardines	501-800	Crushed	Bait bag	5.1-10	20.1-50	11-20	4-6	MaxN	None specified	Fishes, Chondrichthyans	Standard	Unspecified	2
Jamieson et al. 2006	2006	International waters	Temperate	Marine	Deepwater	Baited lander	Vertical	Single	No	N/A	Unspecified	Unspecified	N/A	Other fish	>1000	Whole	No container	≥100.1	≥100.1	>100	Zero	Species specific for grenadier	None specified	Fishes	Novel	Targeted	2
Watson et al. 2005	2005	Australia	Temperate	Marine	Rocky reef	Baited video	Horizontal	Stereo	No - stereo	Unspecified	8 - 10	0-29	≤550	Mix	501-800	Crushed	Bait bag	Unspecified	Unspecified	Unspecified	4-6	MaxN	None specified	Fishes, Chondrichthyans	Standard	0	4
Cappo et al. 2004	2004	Australia	Tropical	Marine	Other	BRUVS	Horizontal	Single	No	Single with reference marks for length estimates	Unspecified	60	≤450	Sardines	801-1000	Crushed	Bait bag	10.1-20	20.1-50	11-20	4-6	MaxN, T1st	None specified	Fishes, Chondrichthyans	Standard	Unspecified	2
Denny & Babcock 2004	2004	NZ	Temperate	Marine	Rocky reef	Baited video	Horizontal	Single	No	Single with reference marks for length estimates	Unspecified	30	Unspecified	Sardines	101-300	Unspecified	Bait container	Unspecified	20.1-50	Unspecified	4-6	MaxN per 1 min	Sigmascan	Fishes, Chondrichthyans	Standard	0	5
Denny et al. 2004	2004	NZ	Temperate	Marine	Rocky Reef	BUV	Horizontal	Single	No	Single with reference marks for length estimates	Unspecified	30	Unspecified	Sardines	101-300	Unspecified	Bait container	Unspecified	20.1-50	Unspecified	>20	MaxN per 1 min for snapper only	Sigmascan	Snapper	Standard	Targeted	5
Westera et al. 2003	2003	Australia	Tropical	Marine	Coral reef	BRUV	Horizontal	Single	No	N/A	Unspecified	30	Unspecified	Unspecified	Unspecified	Unspecified	Bait bag	≤5	≤5	0-10	10-20	MaxN	None specified	Fishes, Chondrichthyans	Standard	0	5
Collins et al. 2002	2002	South Georgia	Polar	Marine	Deepwater	Baited lander	Horizontal	Single	No	Single with reference marks for length estimates	Unspecified	Other	N/A	Mix	501-800	Whole	No container	≥100.1	≥100.1	>100	Zero	MaxN per sequence	None specified	Crustaceans	Novel	Unspecified	2
Yau et al. 2002	2002	South Georgia	Polar	Marine	Deepwater	Baited lander	Vertical	Single	No	Single with reference marks for length estimates	Unspecified	Other	N/A	Mix	501-800	Whole	No container	≥100.1	≥100.1	>100	Zero	Total species and abundance	None specified	All mobile taxa	Novel	<30	1
Willis & Babcock 2000	2000	NZ	Temperate	Marine	Rocky reef	BUV	Vertical	Single	No	Single with reference marks for length estimates	Unspecified	31-59	Unspecified	Sardines	101-300	Chopped	Bait container	Unspecified	Unspecified	Unspecified	4-6	MaxN	Mocha image analysis	Blue cod, snapper	Standard	Targeted	5
Willis et al. 2000	2000	NZ	Temperate	Marine	Rocky reef	BUV	Vertical	Single	No	Single with reference marks for length estimates	Unspecified	30	Unspecified	Mix	Whole fish	Whole	Bait container	Unspecified	Unspecified	Unspecified	4-6	MaxN	Mocha image analysis	Fishes, Chondrichthyans	Standard	Unspecified	6
Ellis and DeMartini 1995	1995	USA (Hawaii)	Tropical	Marine	Other	Unspecified	Horizontal	Single	Yes - single	Single with reference marks for length estimates	Unspecified	0-29	≤150	Mix	Unspecified	Whole	Bait container	50.1-100	50.1-100	21-50	2-3	MaxN, T1st, time within view	None specified	Fishes, Chondrichthyans	Standard	<40	3

Table 7.2: The 161 studies used Chapter 2 showing the purpose of the study. Full references for the studies can be found in either the main article or at the end of this appendix

Study	Species specific/ behavioural information	Marine protected areas	Other fishing related	Changes along a gradient or between habitats	Method comparison within BRUVS	Method comparison with another method	Night and/or day	Other
Gilby et al. In press		X						
Lavaleye et al. In press				X				
Walsh et al. In press		X		X	X			
Colefax et al. 2016			X					
Ghazilou et al. 2016b					X			
Ghazilou et al. 2016a								X
Gilby et al. 2016				X				
Griffin et al. 2016								X
Hesse et al. 2016				X				
Heyns-Veale et al. 2016		X		X				
McLean et al. 2016				X				
Misa et al. 2016					X			
Parker et al. 2016		X				X		
Pejdo et al. 2016						X		
Schmid et al. 2016				X	X			
Spaet et al. 2016	X					X		
Anderson and Santana-Garcon 2015					X			X
Bacheler & Schertzer 2015				X				
Barley et al. 2015	X							
Bornt et al. 2015		X						
Bouchet and Meeuwig 2015	X	X						
Campbell et al. 2015		X						
Coleman et al. 2015					X			
De Vos et al. 2015	X							
D'Onghia et al.		X						

2015a								
D'Onghia et al. 2015b	X							
Ebner et al. 2015							X	
Fitzpatrick et al. 2015							X	
Goetze et al. 2015			X				X	
Harasti et al. 2015		X			X			
Howarth et al. 2015		X						
Kelaher et al. 2015a		X						X
Kelaher et al. 2015b		X						X
Langlois et al. 2015					X			
Letessier et al. 2015	X	X						
Malcolm et al. 2015		X					X	
McLaren et al. 2015							X	
McLean et al. 2015					X			
Pearson and Stevens 2015				X				
Rees et al. 2015	X							
Roberson et al. 2015		X		X				
Ryan et al. 2015				X				X
Schultz et al. 2015	X	X			X	X		
Scott et al. 2015			X	X	X			
Stobart et al. 2015	X	X						
Tanner and Williams 2015					X			
Terres et al. 2015							X	
Trobbiani and Venerus 2015		X		X				
Anderson and Bell 2014								X
Barord et al. 2014				X				
De Vos et al. 2014		X	X					
Dunlop et al. 2014								X
Espinoza et al. 2014				X				

Hannah & Blume 2014								X
Harasti et al. 2014				X				X
Hill et al. 2014	X	X		X				
Kelaher et al. 2014	X	X						
Klages et al. 2014				X				
Lindfield et al. 2014				X				
Morton & Gladstone 2014		X				X		
Lowry et al. 2014			X				X	
Peters et al. 2014						X		
Rizzari et al. 2014	X							
Santana-Garcon et al. 2014a					X			
Santana-Garcon et al. 2014b		X						
Santana-Garcon et al. 2014c				X				
Santana-Garcon et al. 2014d		X						
Schultz et al. 2014	X							
Stevens et al. 2014					X		X	
Udyawer et al. 2014		X						
Unsworth et al. 2014	X	X				X		
Whitmarsh et al. 2014	X							
Ebner & Morgan 2013							X	
Fitzpatrick et al. 2013		X			X			
Folpp et al. 2013	X	X						
Gardner & Struthers 2013	X	X		X				
Hardinge et al. 2013	X							
Harvey et al. 2013					X			
Letessier et al. 2013				X				
Misa et al. 2013	X							

Moore et al. 2013		X								
Poulos et al. 2013	X									
Rees et al. 2013							X			
Ruppert et al. 2013				X						
Sackett et al. 2013			X					X		
Taylor et al. 2013							X			
Wakefield et al. 2013	X	X								
White et al. 2013		X			X					
Wraith et al. 2013	X						X			
Aguzzi et al. 2012				X						
Bernard & Götz 2012	X	X								
Birt et al. 2012		X	X							
Bloomfield et al. 2012				X						
Bond et al. 2012					X					
Colton & Swearer 2012	X									
Dorman et al. 2012		X								
Fitzpatrick et al. 2012		X		X						
Gladstone et al. 2012		X		X						X
Harvey et al. 2012a							X			
Harvey et al. 2012b										
Langlois et al. 2012a		X								
Langlois et al. 2012b			X					X		
Langlois et al. 2012c					X					
Lowry et al. 2012a		X								X
Lowry et al. 2011b					X					
Schultz et al. 2012										
Zintzen et al. 2012	X									
Bassett & Montgomery 2011		X					X			
Brooks et al.							X			

2011												
Cappo et al. 2011	X											
Craig et al. 2011							X		X			X
Dunstan et al. 2011			X				X				X	
Goetze et al. 2011												
Gutteridge et al. 2011	X											
Jeffreys et al. 2011			X						X			
Lowry et al. 2011					X							
Malcolm et al. 2011					X							
Marouchos et al. 2011			X									
McIlwain et al. 2011					X							
McLean et al. 2011	X											
Merritt et al. 2011									X			
Moore et al. 2011	X		X						X			
Robbins et al. 2011					X							
Zintzen et al. 2011									X			
Broad et al. 2010									X			
Chatfield et al. 2010				X								
Colton & Swearer 2010					X							X
Fujii et al. 2010									X			
Langlois et al. 2010												
McLean et al. 2010							X		X			
Moore et al. 2010					X							
Ryer et al. 2010			X									
Watson et al. 2010	X											
Gomelyuk 2009							X					
Jamieson et al. 2009			X		X							
Watson & Harvey 2009			X		X							
Watson et al. 2009	X				X							
Westera et al.			X		X							

2009										
Kleczkowski et al. 2008				X	X			X		
Stoner et al. 2008	X									
Svane & Barnett 2008				X						
Svane et al. 2008	X									
Bailey et al. 2007	X	X								
Cappo et al. 2007a				X			X			
Harvey et al. 2007		X			X					
Heagney et al. 2007				X						
Malcolm et al. 2007					X					
Smale et al. 2007					X					
Stobart et al. 2007		X								
Watson et al. 2007	X									X
Jamieson et al. 2006		X	X							
Watson et al. 2005										
Cappo et al. 2004		X			X					
Denny & Babcock 2004								X	X	
Denny et al. 2004					X		X			
Westera et al. 2003										
Collins et al. 2002		X								
Yau et al. 2002	X									
Willis & Babcock 2000	X									
Willis et al. 2000		X								
Ellis and DeMartini 1995	X						X			

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Appendix 2 – Chapter 4

Appendix A – Case-study #1

Table 7.3: List of species, their fisheries classification, their taxonomic authority and the pictogram (Appendix 4, Table 7.15) of the distinctive as used to represent them in Figure 4.2. In addition, there was an unidentified teleost species allotted as 'not-sought'.

Taxa	Fisheries group	Taxonomic authority	Fish pictogram
Arripis georgianus	Targeted	(Valenciennes 1831)	
Mustelus antarcticus	Targeted	Günther 1870	
Chrysophrys auratus	Targeted	(Forster 1801)	
Portunus armatus	Targeted	(A. Milne-Edwards, 1861)	
Pseudocaranx spp.	Targeted		
Pseudorhombus arsius	Targeted	(Hamilton-Buchanan 1822)	
Sepioteuthis australis	Targeted	Quoy & Gaimard, 1832	
Sillaginodes punctatus	Targeted	(Cuvier 1829)	
Sphyraena novaehollandiae	Targeted	Günther 1860	
Acanthaluteres brownii	By-product	(Richardson 1846)	
Acanthaluteres spilomelanurus	By-product	(Quoy & Gaimard 1824)	
Acanthaluteres vittiger	By-product	(Castelnau 1873)	
Meuschenia scaber	By-product	(Forster 1801)	
Nelusetta ayraud	By-product	(Quoy & Gaimard 1824)	
Ovalipes australiensis	By-product	Stephenson & Rees, 1968	
Scobinichthys granulatus	By-product	(White 1790)	
Thamnaconus degeni	By-product	(Regan 1903)	
Bathytoshia brevicaudata	By-product	(Hutton 1875)	
Dinolestes lewini	By-product	(Griffith 1834)	
Haletta semifasciata	By-product	(Valenciennes 1840)	
Myliobatis tenuicaudatus	By-product	Hector 1877	
Pelates octolineatus	By-product	(Jenyns 1840)	
Sphyrna zygaena	By-product	(Linnaeus 1758)	
Trachurus novaezelandiae	By-product	Richardson 1843	
Trygonorrhina dumerilii	By-product	(Castelnau 1873)	
Upeneichthys vlamingii	By-product	(Cuvier 1829)	
Aracana aurita	Not-sought	(Shaw 1798)	
Aracana ornata	Not-sought	(Gray 1838)	
Atherinidae sp.	Not-sought		
Brachaluteres jacksonianus	Not-sought	(Quoy & Gaimard 1824)	

Heterodontus portusjacksoni	Not-sought	(Meyer 1793)	
Nectocarcinus integrifrons	Not-sought	(Latreille, 1825)	
Neoodax balteatus	Not-sought	(Valenciennes 1840)	
Parequula melbournensis	Not-sought	(Castelnau 1872)	
Siphamia cephalotes	Not-sought	(Castelnau 1875)	
Torquigener pleurogramma	Not-sought	(Regan 1903)	
Vincentia conspersa	Not-sought	(Klunzinger 1872)	

Table 7.4: Species contributing most to the similarity within a group for each zone by Year and Area from SIMPER analysis in case study #1. Av. Sim is the average similarity of a species within a group, Sim/SD is the similarity divided by the standard deviation and gives a measure whether that species is a consistent indicator for that group (values >1 in bold show indicator species). Contrib% is the percent contribution that a species gives to the similarity within a group and Cum.% shows the cumulative percent of this contribution to the total with the cut-off at 70 %.

Group	Average group similarity	Species	Average abundance per replicate	Av.Sim	Sim/SD	Contrib%	Cum.%
20151Unprotected	66.64	*Pelates octolineatus*	5.21	29.35	**3.03**	44.05	44.05
		Torquigener pleurogramma	4.22	19.83	**2.24**	29.76	73.80
20152Unprotected	38.25	*Heterodontus portusjacksoni*	1.83	11.20	**2.62**	29.28	29.28
		Neoodax balteatus	1.67	8.49	**1.71**	22.20	51.48
		Portunus armatus	1.58	5.54	0.71	14.48	65.97
		Scobinichthys granulatus	1.25	2.77	0.60	7.24	73.21
20151Protected	63.53	*Pelates octolineatus*	7.29	38.60	**2.32**	60.76	60.76
		Torquigener pleurogramma	3.86	14.21	**1.20**	22.36	83.12
20152Protected	44.17	*Scobinichthys granulatus*	2.18	10.42	**1.55**	23.59	23.59
		Heterodontus portusjacksoni	2.09	9.52	**1.08**	21.55	45.15
		Neoodax balteatus	1.64	6.67	**1.02**	15.11	60.25
		Pelates octolineatus	1.14	4.15	0.94	9.39	69.64
		Portunus armatus	0.82	3.81	0.99	8.63	78.27
20162Unprotected	47.78	*Portunus armatus*	3.60	21.81	**3.55**	45.64	45.64
		Heterodontus portusjacksoni	1.70	11.00	**3.51**	23.02	68.67
		Torquigener pleurogramma	1.23	3.59	0.74	7.52	76.19
20162Protected	56.26	*Portunus armatus*	3.17	18.95	**2.91**	33.68	33.68
		Heterodontus portusjacksoni	2.42	14.34	**3.40**	25.50	59.17
		Pelates octolineatus	2.24	8.44	**1.42**	15.00	74.17

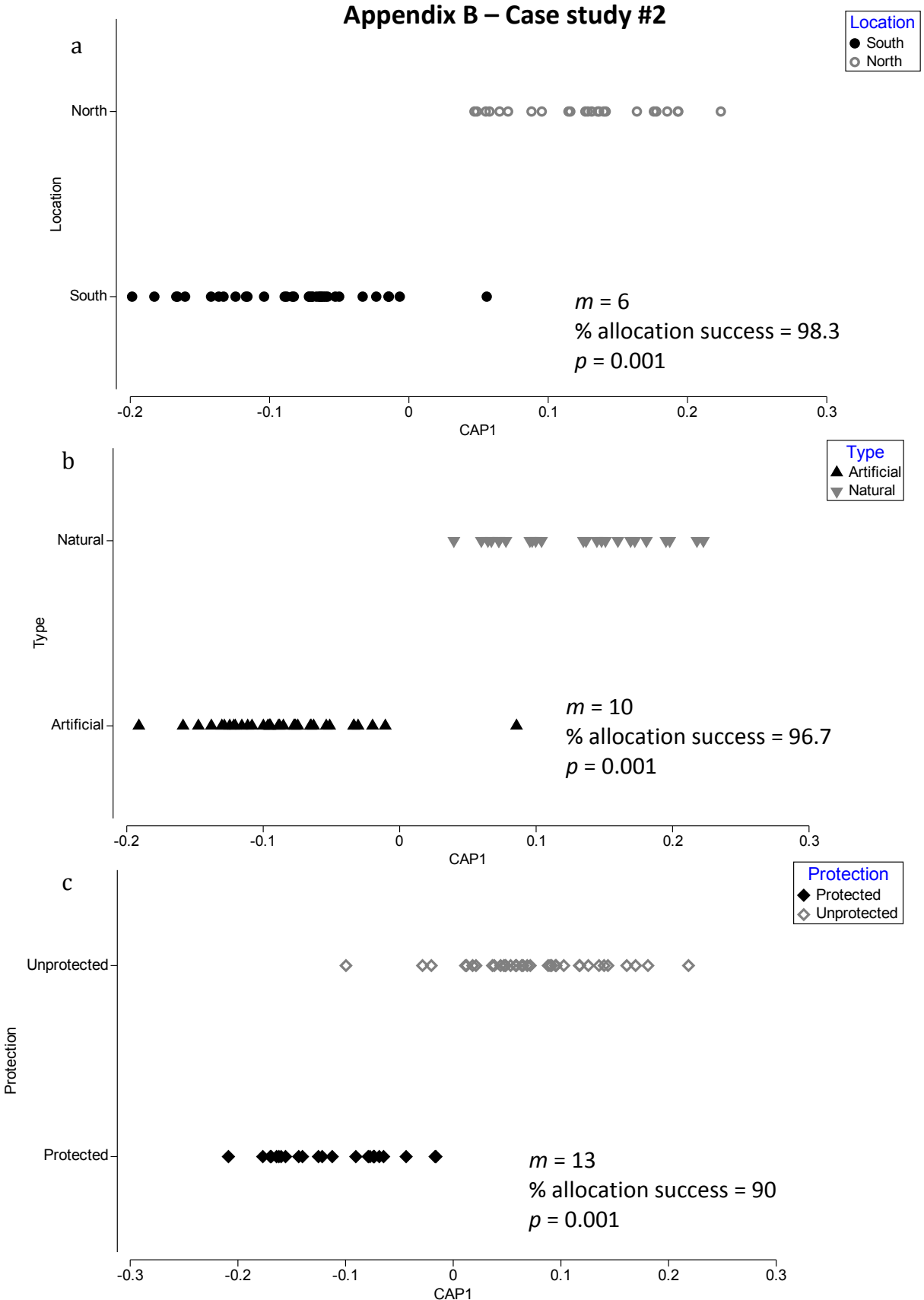


Figure 7.1: CAP ordination plots from Case study #2 showing discriminant factors for the 1st PCO axis (out of *m* axes) for the factor of a) Geographic location, b) Type, and c) Protection.

Appendix C – Case study #3

Table 7.5: Summary of replicate numbers available for analysis from Case study #3and the habitat type observed from the video footage. Unusable replicates were either too obstructed by habitat to view or had technical issues with their recording.

	2016				2017			
Location	**Proposed replicates**	**Usable replicates**	**Habitat classification**		**Proposed replicates**	**Usable replicates**	**Habitat classification**	
North Neptunes	6	13 deep 6 shallow	Reef Sand Seagrass Pelagic	15 2 1 0	18	17	Reef Sand Seagrass Pelagic	9 2 0 6
South Neptunes	6	6	Reef Sand Seagrass Pelagic	1 3 2 0	18	17	Reef Sand Seagrass Pelagic	5 6 0 6
Dangerous Reef	6	6	Reef Sand Seagrass Pelagic	2 4 0 0	18	18	Reef Sand Seagrass Pelagic	1 7 4 6
Liguanea Island	6	5	Reef Sand Seagrass Pelagic	5 0 0 0	Not sampled			
Sub- Total	24	35			54	52	**Total**	**87**

Table 7.6: Species contributing to the dissimilarity between Impact levels for 2016 fish assemblage data from Case study #3. Colours indicate species used to characterise each Impact level as shown in Figure 4. Only the top three species were used for each factor level. Diss/SD is the average dissimilarity divided by the standard deviation with values above 1 (shown in bold) being considered species which are consistent indicators for that factor level. Note: *Mustelus antarcticus* was not used to represent Low impact areas as it was not included in the species contributing most to the similarity of that level.

Pairs	Species	Average abundance (A)	Average abundance (B)	Average dissimilarity	Diss/SD	% contribution
Control (A) vs. High impact (B) Average dissimilarity = 71.76	*Pseudocaranx* spp.	4.03	0.78	10.87	**1.02**	15.15
	Meuschenia hippocrepis	2.45	2.70	7.78	**1.26**	10.84
	Notolabrus tetricus	0.94	1.45	3.52	**1.05**	4.90
	Pictilabrus laticlavius	0.73	1.22	3.28	0.98	4.57
Control (A) vs. Low impact (B) Average dissimilarity = 86.77	*Pseudocaranx* spp.	4.03	0.26	13.43	0.96	15.47
	Meuschenia hippocrepis	2.45	0.36	10.14	0.89	11.69
	Myliobatis tenuicaudatus	0.18	2.00	7.64	**1.08**	8.81
	Mustelus antarcticus	0.25	0.74	3.48	0.62	4.01
	Upeneichthys vlamingii	0.36	0.83	3.40	**1.00**	3.92
High impact (A) vs. Low impact (B) Average dissimilarity = 80.56	*Meuschenia hippocrepis*	2.70	0.36	10.18	**1.60**	12.11
	Myliobatis tenuicaudatus	0.72	2.00	7.48	**1.05**	8.89
	Notolabrus tetricus	1.45	0.48	5.35	**1.08**	6.36
	Pictilabrus laticlavius	1.22	0.17	4.63	**1.01**	5.51

Table 7.7: Species contributing to the dissimilarity between Impact levels for 2017 fish assemblage data from case study #3. Colours indicate species used to characterise each Impact level for Figure 4. Only the top three species were used for each factor level. Diss/SD is the average dissimilarity divided by the standard deviation with values above 1 (shown in bold) being considered species which are consistent indicators for that factor level.

Pairs	Species	Average abundance (A)	Average abundance (B)	Average dissimilarity	Diss/SD	% contribution
Control (A) vs. High impact (B)	*Meuschenia hippocrepis*	0.16	2.05	9.14	**1.54**	10.11
Average dissimilarity = 90.41	*Pseudocaranx* spp.	0.29	1.06	6.10	0.74	6.75
	Sphyraena novaehollandiae	1.06	0.12	5.79	0.63	6.40
	Sillaginodes punctatus	1.02	0.11	4.52	0.77	5.00
	Notolabrus parilus	0.83	0.29	3.74	0.77	4.14
Control (A) vs. Low impact (B)	*Sphyraena novaehollandiae*	1.06	0.00	7.20	0.69	7.76
Average dissimilarity = 92.74	*Pseudocaranx* spp.	0.29	0.57	5.99	0.52	6.46
	Sillaginodes punctatus	1.02	0.11	5.76	0.78	6.22
	Notolabrus parilus	0.83	0.18	4.29	0.71	4.63
	Myliobatis tenuicaudatus	0.17	0.55	4.00	0.58	4.31
	Notolabrus tetricus	0.35	0.67	3.80	0.99	4.09
High impact (A) vs. Low impact (B)	*Meuschenia hippocrepis*	2.05	0.70	9.34	**1.34**	11.59
Average dissimilarity = 80.56	*Pseudocaranx* spp.	1.06	0.57	5.95	0.67	7.39
	Meuschenia freycineti	0.78	0.37	4.09	0.91	5.08
	Cheilodactylus nigripes	0.82	0.53	3.81	**1.06**	4.73
	Notolabrus tetricus	0.57	0.67	3.57	**1.22**	4.44
	Myliobatis tenuicaudatus	0.29	0.55	3.57	0.64	4.43

Table 7.8: The top 10 species contributing to the dissimilarity between Protection levels for each year for Case study #3. Diss/SD is the average dissimilarity divided by the standard deviation with values above 1 (shown in bold) being considered species which are consistent indicators for that factor level. Dissimilarity between factor levels was 77 % for 2016 and 87 % for 2017.

Species	Fisheries targeted?	Average abundance Protected	Average abundance Unprotected	Average dissimilarity	Diss/SD	% contribution
2016						
Meuschenia hippocrepis	No	2.18	2.31	9.01	**1.27**	11.77
Pseudocaranx spp.	Yes	2.38	0.24	7.70	0.76	10.05
Myliobatis tenuicaudatus	No	0.63	1.09	4.84	0.79	6.32
Notolabrus tetricus	No	1.30	0.73	3.95	0.98	5.15
Pictilabrus laticlavius	No	1.04	0.55	3.55	0.92	4.64
Cheilodactylus nigripes	No	0.75	0.55	3.09	**1.09**	4.04
Girella zebra	No	0.75	0.06	2.92	0.67	3.81
Achoerodus gouldii	No	0.83	0.27	2.85	**1.00**	3.73
Mustelus antarcticus	Yes	0.07	0.56	2.30	0.49	3.01
Upeneichthys vlamingii	No	0.29	0.45	2.30	0.75	3.01
2017						
Meuschenia hippocrepis	No	1.08	0.70	6.24	0.98	7.19
Pseudocaranx spp.	Yes	0.66	0.57	5.97	0.58	6.88
Sphyraena novaehollandiae	Yes	0.60	0.00	4.93	0.47	5.68
Myliobatis tenuicaudatus	No	0.23	0.55	3.79	0.60	4.36
Notolabrus tetricus	No	0.45	0.67	3.69	**1.08**	4.25
Sillaginodes punctatus	Yes	0.58	0.11	3.42	0.58	3.94
Meuschenia freycineti	No	0.48	0.37	3.27	0.74	3.77
Cheilodactylus nigripes	No	0.51	0.53	3.19	0.93	3.68
Notolabrus parilus	No	0.57	0.18	3.02	0.62	3.48
Achoerodus gouldii	No	0.26	0.59	2.96	0.81	3.41

Table 7.9: Results from case study #3 pairwise PERMANOVA analyses conducted on two species individually, with each year analysed separately for the factor Location which was significant as a main effect for all analyses conducted. Bold values indicate significant differences. Blanks involve Liguanea Island, which was not visited in 2017.

	***Pseudocaranx* spp.**						***Meuschenia hippocrepis***					
	2016			**2017**			**2016**			**2017**		
Groups	**t**	**perms**	***p*(MC)**	**t**	**perms**	***p*(perm)**	**t**	**perms**	***p*(MC)**	**t**	**perms**	***p*(MC)**
Dangerous Reef, Liguanea Island	4.32	98	**0.001**				3.70	76	**0.004**			
Dangerous Reef, North Neptunes	7.20	129	**0.001**	2.83	246	**0.007**	3.23	88	**0.007**	5.99	138	**0.001**
Dangerous Reef, South Neptunes	4.73	62	**0.001**	1.31	179	0.187	0.56	8	0.572	2.17	81	**0.034**
Liguanea Island, North Neptunes	1.32	35	0.179				2.24	90	**0.037**			
Liguanea Island, South Neptunes	0.30	9	0.772				4.13	73	**0.003**			
North Neptunes, South Neptunes	1.30	35	0.193	1.94	128	0.067	3.74	51	**0.002**	3.41	72	**0.003**

Appendix D – Case study #4

Table 7.10: Case study #4 SIMPER results comparing within-site similarities of the assemblage for each site within each season sampled. Sim/SD is a measure of the similarity divided by the standard deviation and is an indication of consistency within a site with species considered as consistent having values above 1 and are shown in bold.

Site	Season	Species	Average abundance	Average similarity	Sim/SD	% contribution
Kellidie Bay	Autumn	*Arripis* spp.	1.62	20.25	**1.88**	56.74
		Aurelia aurita	1.00	9.30	**0.76**	26.07
	Spring	*Leptomithrax gaimardii*	0.49	5.27	**1.12**	38.61
		Parapercis ramsayi	0.88	4.31	**0.37**	31.56
Mount Dutton	*Autumn*	*Nectocarcinus integrifrons*	0.55	7.59	**1.04**	28.18
		Myliobatis tenuicaudatus	0.83	5.17	**0.76**	19.18
		Sillaginodes punctatus	0.66	4.88	**1.09**	18.12
		Neoodax balteatus	0.92	4.07	**0.50**	15.12
	Spring	*Nectocarcinus integrifrons*	1.10	13.33	**2.37**	33.11
		Leptomithrax gaimardii	1.17	12.96	**1.60**	32.20
		Neoodax balteatus	1.77	7.19	**1.35**	17.86
Point Longnose	*Autumn*	*Ovalipes australiensis*	2.44	30.85	**3.63**	58.09
		Platycephalus speculator	1.98	15.22	**1.11**	28.66
	Spring	*Ovalipes australiensis*	1.22	12.98	**1.39**	45.15
		Nectocarcinus integrifrons	1.56	7.55	**0.69**	26.27
Port Douglas Mid	*Autumn*	*Ovalipes australiensis*	1.59	8.75	**0.53**	35.25
		Nectocarcinus integrifrons	1.19	5.22	**0.76**	21.03
		Platycephalus speculator	1.39	3.70	**0.40**	14.93
	Spring	*Nectocarcinus integrifrons*	1.56	10.34	**0.99**	27.84
		Sillaginodes punctatus	0.93	8.45	**0.72**	22.76
		Leptomithrax gaimardii	1.03	7.27	**0.77**	19.56
Port Douglas South	*Autumn*	*Neoodax balteatus*	2.76	19.12	**1.93**	58.05
		Nectocarcinus integrifrons	1.19	7.08	**1.17**	21.50
	Spring	*Neoodax balteatus*	1.70	13.09	**0.85**	44.74
		Nectocarcinus integrifrons	0.92	5.71	**0.60**	19.52
		Leptomithrax gaimardii	0.59	3.79	**0.61**	12.96

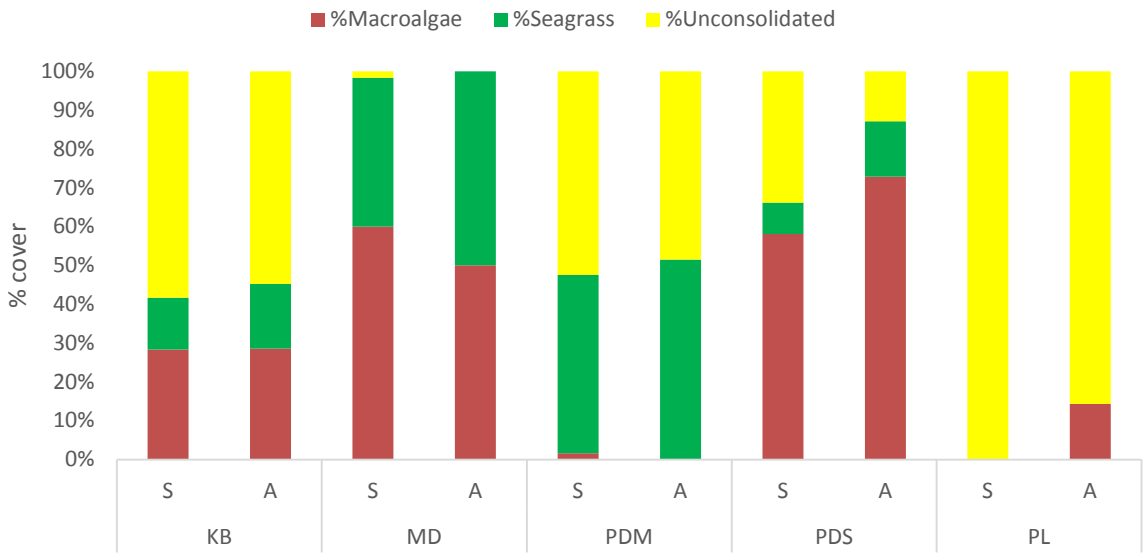


Figure 7.2: The % cover for each habitat type (excluding % open water) within the field of view of each BRUVS deployment in case study #4, averaged across site by season. S = spring, A = autumn. Sites are represented by their initials.

Table 7.11: Case study #4 results from distance-based redundancy analysis (dbRDA) showing the variation explained by the seven axes used in the DistLM model.

Axis	% explained variation out of fitted model		% explained variation out of total variation	
	Individual variable	Cumulative	Individual variable	Cumulative
1	39.41	39.41	14.7	14.7
2	23.62	63.03	8.81	23.52
3	15	78.04	5.6	29.11
4	10.53	88.57	3.93	33.04
5	6.02	94.59	2.25	35.29
6	4.1	98.68	1.53	36.82
7	1.32	100	0.49	37.31

Appendix 3 – Chapter 5

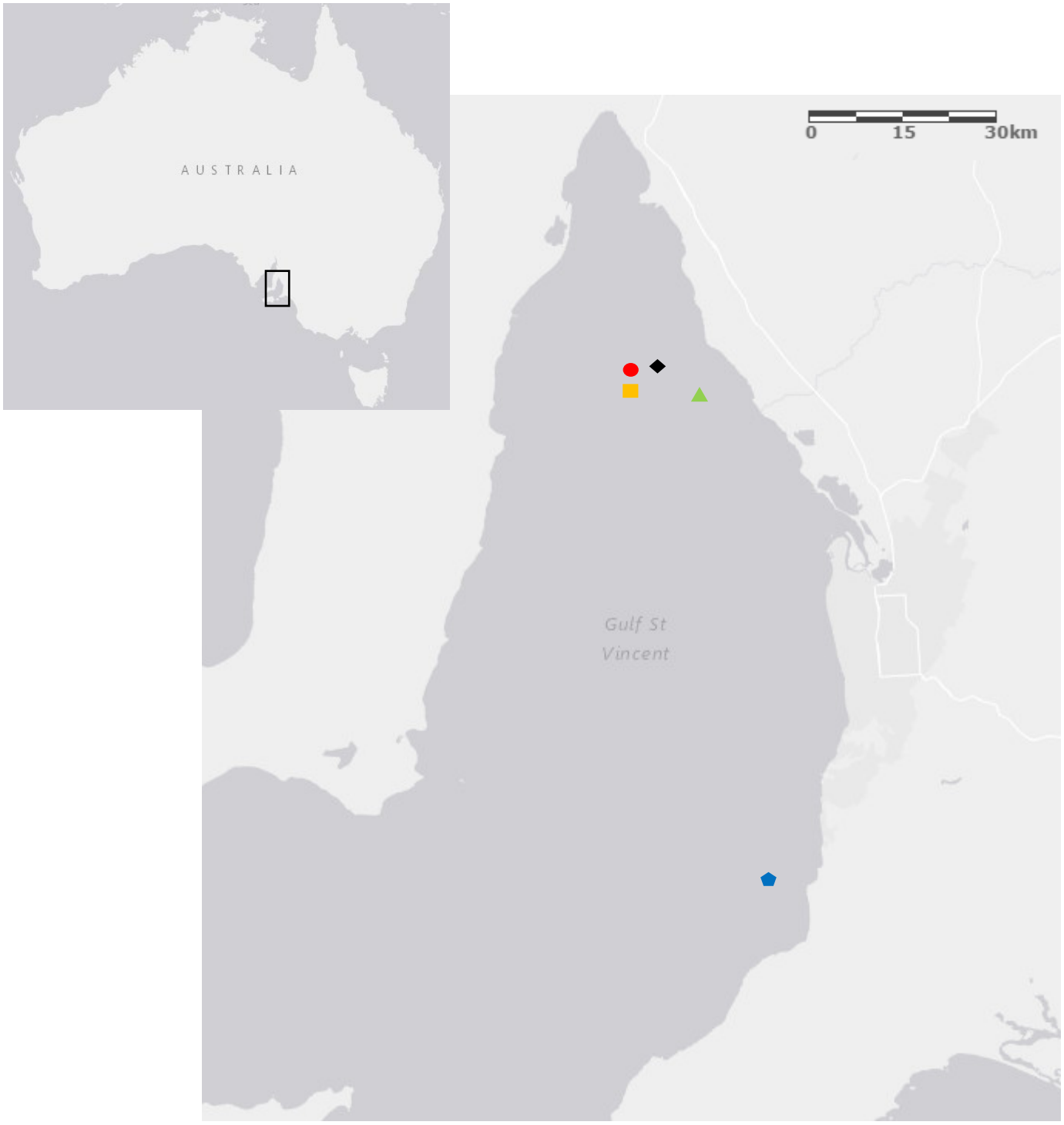


Figure 7.3: Map showing the sites where BRUVS were deployed. The blue pentagon indicates Aldinga Reef (S35.27360 E138.43265), the green triangle Long Spit (S34.56461 E138.22672), the orange square the Barge wreck (S34.52841 E138.06356), the red circle the Zanoni wreck (S34.51163 E138.06368) and the black diamond Near Zanoni (S34.51496 E138.08525).

Table 7.12: Presence of taxa at each site, with an X indicating at least one sighting, for species observed using 360° view BRUVS. *Pseudocaranx* spp. consisted of two similar co-occurring species, *Pseudocaranx wrighti* and *Pseudocaranx georgianus*.

Taxa and Taxonomic authority	Aldinga Reef	Barge	Long Spit	Near Zanoni	Zanoni
Teleosts					
Acanthaluteres brownii (Richardson 1846)	X				
Acanthaluteres vittiger (Castelnau 1873)	X		X		
Aracana ornata (Gray 1838)				X	
Arripis georgianus (Valenciennes 1831)	X		X		
Austrolabrus maculatus (Macleay 1881)	X				
Cheilodactylus nigripes Richardson 1850	X				
Chelmonops curiosus Kuiter 1986					X
Dactylophora nigricans (Richardson 1850)	X				X
Haletta semifasciata (Valenciennes 1840)			X		
Kyphosus sydneyanus (Günther 1886)	X				
Meuschenia freycineti (Quoy & Gaimard 1824)			X		X
Meuschenia hippocrepis (Quoy & Gaimard 1824)	X				
Neoodax balteatus (Valenciennes 1840)			X		
Notolabrus parilus (Richardson 1850)	X				
Notolabrus tetricus (Richardson 1840)	X				
Olisthops cyanomelas (Richardson 1850)	X				
Omegophora armilla (McCulloch & Waite 1915)	X				
Chrysophrys auratus (Forster 1801)	X	X			X
Parapercis haackei (Steindachner 1884)	X	X		X	X
Parequula melbournensis (Castelnau 1872)	X	X			
Parma victoriae (Günther 1863)	X				
Pelates octolineatus (Jenyns 1840)		X	X	X	X
Pentaceropsis recurvirostris (Richardson 1845)	X				
Pictilabrus laticlavius (Richardson 1839)	X				
Platycephalus speculator Klunzinger 1872		X			
Pseudocaranx spp.	X	X			X
Scobinichthys granulatus (White 1790)	X		X	X	
Scorpis aequipinnis Richardson 1848	X				
Sillaginodes punctatus (Cuvier 1829)	X		X	X	
Siphamia cephalotes (Castelnau 1875)			X		
Siphonognathus sp.	X				
Siphonognathus radiatus (Quoy & Gaimard 1834)			X		
Sphyraena novaehollandiae Günther 1860			X		
Thamnaconus degeni (Regan 1903)		X		X	X
Tilodon sexfasciatus (Richardson 1842)	X				
Torquigener pleurogramma (Regan 1903)			X		
Trachurus novaezelandiae Richardson 1843			X	X	X
Upeneichthys vlamingii (Cuvier 1829)	X	X	X	X	
Chondrichthyans					

Bathytoshia brevicaudata (Hutton 1875)			X		
Heterodontus portusjacksoni (Meyer 1793)		X	X	X	X
Notorynchus cepedianus (Péron 1807)		X			
Trygonorrhina dumerilii (Castelnau 1873)		X	X	X	X
Invertebrates					
Coscinasterias muricata Verrill, 1867	X				
Leptomithrax gaimardii (H. Milne Edwards, 1834)				X	
Portunus armatus (A. Milne-Edwards, 1861)		X	X	X	X
Sepioteuthis australis Quoy & Gaimard, 1832			X		
Total	**25**	**11**	**17**	**10**	**11**

Table 7.13: Pairwise PERMANOVA tests of the Viewpoint factor for multivariate analysis of assemblages and univariate analysis of total individuals per viewpoint. Unique permutations ranged from 982-996.

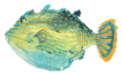
	Multivariate		**Univariate**	
Groups	***t***	**p(perm)**	***t***	***p*(perm)**
Back, Front	1.646	0.092	1.238	0.277
Back, Left	0.031	0.846	3.260	0.058
Back, Right	0.973	0.524	0.882	0.444
Front, Left	1.836	0.076	2.140	0.104
Front, Right	2.042	0.064	2.021	0.104
Left, Right	1.171	0.326	2.271	0.091

Table 7.14: SIMPER results showing the average similarity of assemblages from each site and the dominant taxa (in descending order) with their contribution to that overall site similarity.

Site	Average similarity (%)	Dominant taxa	Average abundance	% contribution to similarity
Aldinga	38	*Upeneichthys vlamingii*	2.75	15.2
		Pseudocaranx spp.	4.75	12.3
		Austrolabrus maculatus	1.92	11.9
		Notolabrus parilus	1.08	8.0
		Chrysophrys auratus	2.42	7.9
		Parequula melbournensis	1.17	7.9
		Tilodon sexfasciatus	0.83	6.9
Barge	53	*Thamnaconus degeni*	49.31	70.9
Long Spit	39	*Siphamia cephalotes*	17.75	27.7
		Heterodontus portusjacksoni	2.19	14.7
		Pelates octolineatus	2.13	11.6
		Sillaginodes punctatus	1.25	8.2
		Portunus armatus	1.63	7.8
Near Zanoni	69	*Thamnaconus degeni*	59.00	60.6
		Pelates octolineatus	27.58	28.2
Zanoni	33	*Chrysophrys auratus*	6.19	46.4
		Pseudocaranx spp.	6.63	20.0
		Trachurus novaezelandiae	2.19	11.2

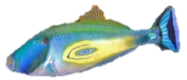
Appendix 4 - Thesis

Table 7.15: List of taxa observed across all BRUVS deployments with an X indicating that taxon was present within that study. Reference pictograms are included for when this visual representation was used in other figures (e.g. 2.1). Ch. # refers to the study chapter, with 4-# indicating the specific case study within Chapter 4 (which included four case studies). Unidentified species were grouped into spp. categories. Taxonomic authorities were taken from www.fishesofaustralia.net.au and www.ala.org.au.

Taxa	**Taxonomic authority**	**Present in study**						**Pictogram**	**Pictogram source**
		Ch. 3	**Ch. 4-1**	**Ch. 4-2**	**Ch. 4-3**	**Ch. 4-4**	**Ch. 5**		
Teleosts									
Acanthaluteres brownii	(Richardson 1846)	X	X	X	X		X		
Acanthaluteres spilomelanurus	(Quoy & Gaimard 1824)		X	X		X			
Acanthaluteres vittiger	(Castelnau 1873)	X	X	X	X	X	X		
Achoerodus gouldii	(Richardson 1843)	X			X				
Aldrichetta forsteri	(Valenciennes 1836)					X			
Ammotretis elongatus	McCulloch 1914		X						
Aracana aurita	(Shaw 1798)	X	X	X					Rudie H. Kuiter
Aracana ornata	(Gray 1838)	X	X	X		X	X		
Argyrosomus japonicus	(Temminck & Schlegel 1844)	X							
Arnoglossus sp.		X							
Arripis georgianus	(Valenciennes 1831)	X	X	X	X	X	X	 (Arripis spp.)	CSIRO

Arripis truttaceus	(Cuvier 1829)	X	X		X	X	 (Arripis spp.)	CSIRO
Atherinidae sp.		X	X					
Atherinosoma microstoma	(Günther 1861)					X		
Austrolabrus maculatus	(Macleay 1881)	X		X	X		X	 Ian Shaw
Brachaluteres jacksonianus	(Quoy & Gaimard 1824)	X	X	X			X	
Caesioperca rasor	(Richardson 1839)				X			
Centroberyx gerrardi	(Günther 1887)				X			
Centroberyx lineatus	(Cuvier 1829)				X			
Cheilodactylus nigripes	Richardson 1850	X		X	X		X	 Rudie H. Kuiter
Cheilodactylus spectabilis	Hutton 1872				X			
Chelmonops curiosus	Kuiter 1986	X		X	X		X	 Graham Short
Cnidoglanis macrocephalus	(Valenciennes 1840)	X						
Dactylophora nigricans	(Richardson 1850)	X		X	X		X	
Dinolestes lewini	(Griffith 1834)	X	X	X	X			
Diodon nicthemerus	Cuvier 1818	X						
Dotalabrus aurantiacus	(Castelnau 1872)	X						
Engraulis australis	(White 1790)	X				X		

Enoplosus armatus	(White 1790)	X		X	X				
Eubalichthys mosaicus	(Ramsay & Ogilby 1886)			X					
Eupetrichthys angustipes	Ramsay & Ogilby 1888				X				
Favonigobius lateralis	(Macleay 1881)					X			
Girella zebra	(Richardson 1846)			X	X				Rudie H. Kuiter
Gnathophis sp.		X							
Haletta semifasciata	(Valenciennes 1840)	X	X	X		X	X		
Heteroscarus acroptilus	(Richardson 1846)	X		X	X				
Hypoplectrodes nigroruber	(Cuvier 1828)	X		X	X		X		
Hyporhamphus melanochir	(Valenciennes 1847)					X			
Kyphosus sydneyanus	(Günther 1886)	X		X	X		X		Rick Stuart-Smith
Latropiscis purpurissatus	(Richardson 1843)				X				
Meuschenia flavolineata	Hutchins 1977	X		X	X				
Meuschenia freycineti	(Quoy & Gaimard 1824)	X		X	X		X		John Turnbull
Meuschenia galii	(Waite 1905)	X			X		X		

Meuschenia hippocrepis	(Quoy & Gaimard 1824)	X		X	X		X		Andrew J. Green
Meuschenia scaber	(Forster 1801)	X	X						
Meuschenia venusta	Hutchins 1977				X				
Neatypus obliquus	Waite, 1905				X				
Nelusetta ayraud	(Quoy & Gaimard 1824)		X		X	X			
Nemadactylus valenciennesi	(Whitley 1937)				X				
Neoodax balteatus	Valenciennes 1840)	X	X	X		X	X		B. Hutchins
Neosebastes bougainvillii	(Cuvier 1829)			X					
Neosebastes scorpaenoides	Guichenot 1867			X	X				
Notolabrus fucicola	(Richardson 1840)	X							
Notolabrus parilus	(Richardson 1850)	X		X	X		X		Rudie H. Kuiter
Notolabrus tetricus	(Richardson 1840)	X		X	X		X		Rudie H. Kuiter
Odax acroptilus	(Richardson 1846)	X							
Odax cyanomelas	(Richardson 1850)			X					
Olisthops cyanomelas	(Richardson 1850)				X		X		
Omegophora armilla	(McCulloch & Waite 1915)	X		X			X		
Omegophora	Hardy & Hutchins			X					

cyanopunctata	1981								
Ophichthidae sp.		X							
Ophthalmolepis lineolatus	(Valenciennes 1839)	X			X				
Chrysophrys auratus	(Forster 1801)	X	X	X			X		DPI, NSW
Parapercis haackei	(Steindachner 1884)	X		X	X	X	X		Rudie H. Kuiter
Parapercis ramsayi	(Steindachner 1883)				X	X			Dave Harasti
Parapriacanthus elongatus	(McCulloch 1911)	X							
Parequula melbournensis	(Castelnau 1872)	X	X	X	X	X	X		Erik Schlogl
Parma victoriae	(Günther 1863)	X		X	X		X		David Muirhead
Pelates octolineatus	(Jenyns 1840)	X	X	X			X		Barry Hutchins
Pempheris multiradiata	Klunzinger 1879	X		X	X				Erik Schlögl
Pentaceropsis recurvirostris	(Richardson 1845)	X		X			X		
Phyllopteryx taeniolatus	(Lacépède 1804)			X					

Pictilabrus laticlavius	(Richardson 1839)	X		X	X		X		Rudie H. Kuiter
Platycephalus bassensis	Cuvier 1829					X			
Platycephalus speculator	Klunzinger 1872	X		X	X	X	X		Rudie H. Kuiter
Pseudocaranx spp.		X	X	X	X	X	X		Rudie H. Kuiter
Pseudophycis barbata	Günther 1863	X							Rudie H. Kuiter
Pseudorhombus arsius	(Hamilton-Buchanan 1822)		X						
Rhombosolea tapirina	Günther 1862					X			
Sardinops sagax	(Jenyns 1842)	X							
Scobinichthys granulatus	(White 1790)	X	X	X	X		X		Rudie H. Kuiter
Scomber australasicus	Cuvier 1832				X				
Scorpis aequipinnis	Richardson 1848	X		X	X		X		Julian Finn
Scorpis georgiana	Valenciennes 1832			X					

Seriola hippos	Günther 1876				X				
Seriola lalandi	Valenciennes 1833				X				
Sillaginodes punctatus	(Cuvier 1829)	X	X		X	X	X		Rudie H. Kuiter
Sillago sp.		X		X		X			
Siphamia cephalotes	(Castelnau 1875)	X	X			X	X		
Siphonognathus attenuatus	(Ogilby 1897)	X			X				
Siphonognathus radiatus	(Quoy & Gaimard 1834)					X			
Siphonognathus sp.		X		X	X		X		
Siphonognathus tanyourus	Gomon & Paxton 1986						X		
Sphyraena novaehollandiae	Günther 1860	X	X		X	X	X		Rudie H. Kuiter
Stigmatopora argus	(Richardson 1840)					X			
Thamnaconus degeni	(Regan 1903)	X	X	X	X		X		Rudie H. Kuiter
Threpterius maculosus	Richardson 1850				X				
Thunnus maccoyii	(Castelnau 1872)				X				
Tilodon sexfasciatus	(Richardson 1842)	X		X	X		X		
Torquigener pleurogramma	(Regan 1903)	X	X				X		Klaus Steifel

Trachinops noarlungae	Glover 1974	X		X			X		
Trachurus declivis	(Jenyns 1841)			X		X			
Trachurus novaezelandiae	Richardson 1843	X	X	X	X		X		Richard Ling
Upeneichthys vlamingii	(Cuvier 1829)	X	X	X	X	X	X		Sarah Spieght
Vincentia conspersa	(Klunzinger 1872)	X	X			X			Rudie H. Kuiter
Unidentified fish spp.		X	X			X	X		
Chondrichthyans									
Aptychotrema vincentiana	(Haacke 1885)				X				
Asymbolus vincenti	(Zietz 1908)	X							CSIRO
Carcharhinus brachyurus	(Günther 1870)	X			X				
Carcharodon carcharias	(Linnaeus 1758)	X			X				
Bathytoshia brevicaudata	(Hutton 1875)	X	X	X	X		X		
Heterodontus portusjacksoni	(Meyer 1793)	X	X	X	X		X		Rudie H. Kuiter
Mustelus antarcticus	Günther 1870		X		X	X			

Myliobatis tenuicaudatus	Hector 1877	X	X		X	X			Malcolm Francis
Orectolobus halei	Whitley 1940	X							
Parascyllium variolatum	(Duméril 1853)				X				
Notorynchus cepedianus	(Péron 1807)	X					X		
Sphyrna zygaena	(Linnaeus 1758)		X						
Trygonorrhina dumerilii	(Castelnau 1873)	X	X	X		X	X		
Urolophus gigas	Scott 1954				X				
Urolophus paucimaculatus	Dixon 1969				X				
Nektonic Invertebrates									
Aurelia aurita	(Linnaeus, 1758)					X			
Cyanea rosella	Gershwin in Gowlett-Holmes, 2008					X			
Unidentified cnidarian spp.		X			X	X		 (ctenophore in chapter 2)	Marco Faasse
Coscinasterias muricata	Verrill, 1867	X							
Uniophora granifera	(Lamarck, 1816)					X			
Unidentified seastar					X				

spp.									
Unidentified hermit crab spp.					X				
Ibacus peronii	Leach, 1815	X							
Leptomithrax gaimardii	(H. Milne Edwards, 1834)	X		X	X	X	X		John Lewis
Melicertus latisulcatus	(Kishinouye, 1896)	X							
Naxia aurita	(Latreille, 1825)			X	X				
Nectocarcinus integrifrons	(Latreille, 1825)	X	X	X		X			Joan Hales
Ovalipes australiensis	Stephenson & Rees, 1968	X	X	X	X	X			Sarah Speight,
Ozius truncatus	H. Milne Edwards, 1834			X					
Portunus armatus	(A. Milne-Edwards, 1861)	X	X				X		R. Swainston
Euprymna tasmanica	(Pfeffer, 1884)	X							Doug Perrine
Octopus spp.		X			X	X			
Sepia apama	Gray, 1849	X		X					
Sepioteuthis australis	Quoy & Gaimard, 1832	X	X	X	X	X	X		
Other									
Arctocephalus	(Lesson, 1828)				X				

forsteri			
Tursiops sp.		X	X
Microcarbo melanoleucos	(Vieillot, 1817)		X