REFUGE IN THE DEEP? ASSESSING THE POTENTIAL OF MESOPHOTIC CORAL

ECOSYSTEMS TO ACT AS REFUGIA FOR SHALLOW CORAL REEF FISHES IN

THE HAWAIIAN ISLANDS

By

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Abstract

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Coral reefs are increasingly under threat, necessitating an emphasis to identify coral reefs with

reduced susceptibilities to local and/or global anthropogenic impacts. Mesophotic coral reefs

(MCEs; >30m) are proposed as potential refugia and/or propagule sources, yet little information

is known about deep reefs' abilities to harbor, replenish, or conserve shallow species. In this

dissertation, I examine the plausibility of MCEs to act as refugia for shallow reef fishes in the

Hawaiian Islands. Chapter One explores reef fish community structure and habitat composition

along a 3-50m gradient in West Hawai'i. Reef fish communities change gradually with depth,

with >78% of species observed at mesophotic depths (>30m) found at shallow depths. Changes

in community structure are linked closely with feeding behavior, with shallow reefs dominated

by herbivores, while mesophotic reefs are dominated by invertivore and planktivore trophic

assemblages. Changes in fish assemblages are tied to indirect effects of depth and available coral

habitat, as deeper reefs contain more patchily-distributed habitat. Chapter Two examines

mechanisms underlying herbivorous fish distributions using a suite of observational and

experimental field and laboratory techniques. Herbivorous fishes are not limited by food

resources at MCE depths, as MCE algae had similar nutritional content, species assemblages, and appears to be highly palatable from algal choice experiments. Instead, changes with depth are likely the result of top-down, non-consumptive predation effects and behavioral choices. Chapter Three undertakes a critical analysis of the deep refugia hypothesis for coral reef fishes across the Main Hawaiian Islands. Upper MCEs (30-60m) may act as refugia for shallow reef fishes, as we found they are more thermally stable and >70% of reef fishes encountered were shallow species. Conversely, MCEs contain reduced densities of reef fishes and communities are comprised almost solely of invertivore and planktivore trophic groups. The near-absence of herbivorous fishes below 30m indicate MCEs will have a limited capacity to re-seed shallow reefs with species of ecological or economic importance. Overall, MCEs may act as refugia for biodiversity conservation but their ability to restock shallow reef fish communities will result in fundamentally different community compositions that shift towards smaller-bodied and less economically/ecologically valuable species.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

GENERAL INTRODUCTION

To date, nearly all studies of coral reefs are limited to habitat less than 30 m depth (Menza 2008; Reaka et al. 2008), yet reef-building corals frequently extend to depths greater than 150 m (Reed 1985; Maragos and Jokiel 1986; Fricke et al. 1987; Kahng and Maragos 2006). As a result, our entire understanding of coral reef ecosystems, and conservation measures intended to protect them, are biased towards information collected from the shallowest one-third of total coral reef habitat by depth. These deeper extensions of shallow coral reefs, or Mesophotic Coral Ecosystems (MCEs), have been understudied largely because of technological limitations. However, recent advances in diving and remote sensing technologies are now affording safer tools for exploration and documentation of these ecosystems (Pyle 1996; Pyle 2000; Lesser et al. 2009; Puglise et al. 2009, Waddington et al. 2010).

As reef-building corals have been documented in excess of 150 meters, it is possible that these deeper sections of reef (below 30 meters) provide considerable habitat for coral reef fishes and other organisms. Unlike shallow reefs, mesophotic reefs are often too deep to be detected on traditional navigational charts and remote sensing technologies such as aerial imagery, LIDAR (light detection and ranging), LADs (laser airborne depths sensors) and satellite-based sensors. The advent and increasing use of technical diving techniques such as TRIMIX SCUBA (use of helium and oxygen with traditional air) and rebreather technologies are now allowing divers to more safely access MCE depths. The technological advances in manned submersible, ROV (remotely operated vehicle) and AUV (autonomous underwater vehicle) operating systems can also afford greater investigation of MCE communities. These tools are highly effective but also very expensive due to the cost of each individual unit as well as ship time to accommodate and deploy them. It is for these reasons that ecological studies of MCEs are understudied and poorly

characterized, especially when compared to shallow coral reefs that can be surveyed with ease and low cost per unit effort using snorkel or SCUBA.

The few ecological studies conducted on mesophotic reefs reveal general trends that support the premise of deep reefs as potentially substantial habitat (Brokovich et al. 2007, Lesser et al. 2009). The physical environment shows declined light attenuation but increased protection from wave action and relatively constant seawater temperatures as compared to shallow reefs, indicating that mesophotic reefs may be more environmentally stable (Hoegh-Guldberg et al. 2007, Lesser et al. 2009). Studies of mesophotic communities in the Caribbean reveal that coral cover is consistent through 50-60 meters in depth (Goreau and Goreau 1973, Liddell and Ohlhorst 1988, Liddell et al. 1997, Bak et al. 2005). Recent observations in the Red Sea have discovered deep nursery grounds for juvenile angelfishes in 60-65m, and expeditions to the Northwestern Hawaiian Islands have observed similar nursery grounds for damselfishes and angelfishes in 40-60m (Brokovich et al. 2007, Kane personal observations). These findings suggest that mesophotic reefs may provide significant habitat and possible refuge for shallow coral reef fishes.

MCEs as potential coral reef refugia

Coral reefs are increasingly under threat from both local and global stressors (Hoegh-Guldberg 1999, Hughes et al. 2003, Guinotte et al. 2003, Hughes et al. 2007). While coral reefs have persisted for at least 500,000 years, the last 30 years have shown unprecedented declines in the distribution and abundance of coral reef communities (Pandolfi 2002, Bongaerts et al. 2010a). It is estimated that if coral decline continues at it's current rate approximately 15% of coral reefs will be seriously threatened within 20 years and a further 20% may be lost in the next 20-40 years (Wilkinson 2008). These rapid declines have largely been attributed to

anthropogenic impacts ranging from local factors such as overfishing, destructive fishing, reductions in water quality from land-based use and pollution, to global factors such as increased sea surface temperatures and ocean acidification as a result of increased atmospheric carbon dioxide concentrations (Hoegh-Guldberg et al. 2007, Bak et al. 2009, De'ath et al. 2009). As a result, coral reef managers and conservationists have prioritized the identification of coral reef regions that may have reduced susceptibilities to current and predicted anthropogenic stressors (Riegl and Piller 2003).

It has been proposed for some time that deep reefs may act as possible refugia for more impacted shallow coral reef ecosystems yet is not widely acknowledged or considered among coral reef management agencies (Riegl and Piller 2003, Lesser et al. 2009, Bongaerts et al. 2010a, Hinderstein et al. 2010). The 'deep reef refugia' hypothesis (DRRH) was first formulated in the late 1990s largely for zones of coral reefs that had reduced susceptibilities to certain disturbances such as cyclone damage or thermal stress (Liddell and Ohlhorst 1988, Glynn 1996, Riegl and Piller 2003). Of late, the 'deep reef refugia' hypothesis has extended beyond physical sheltering capabilities and has been proposed as possible recruitment sources for shallow coral reef ecosystems (Hughes and Tanner 2000, Lesser et al. 2009, Bongaerts et al. 2010a). The 'deep reef refugia' hypothesis currently focuses on two primary questions: 1) Are deep reefs less susceptible to disturbance than shallow water, and 2) can deep reefs provide a source of propagules to re-seed nearby shallow coral reefs experiencing decline (Hughes and Tanner 2000, Lesser et al. 2009, Slattery et al. 2011, Bridge and Guinotte 2012).

Despite a plethora of literature describing the effects of natural and anthropogenic disturbances on shallow coral reefs, very few studies incorporate disturbance effects on reefs below 20 meters (Bongaerts et al. 2010a). Although a large amount of variation exists in MCE

susceptibilities to disturbance, there is a general trend among literature indicating that MCEs may be less susceptible to disturbance events. Several studies from the Great Barrier Reef have noted that MCE regions have experienced lower coral bleaching and subsequent mortality following thermal stress events than adjacent shallow water regions (Guinotte 2007, Bongaerts et al. 2010a). The decreased coral bleaching in MCE regions is likely due to lower overall water temperatures in deeper water. Frade et al. (2008) observed a 1°C difference in average summer water temperatures between shallow (5-7m) and deep (35-40m) coral reefs in the Caribbean, with even more pronounced differences over broader depth ranges. Given that coral bleaching events have been strongly correlated with 1-2°C increases in temperatures over the long-term monthly averages, these small decreases in temperature between shallow and deep regions may be highly significant for coral bleaching and mortality events (Liu et al. 2001, Liu et al. 2003, Hoegh-Guldberg 1999, McClanahan et al. 2012).

In addition to being more thermally stable, MCE regions may be less susceptible to turbulent oceanographic conditions than coral reefs in more shallow depths. Episodic wave energy resulting from events such as tsunamis or hurricanes is negatively correlated with depth, thus coral reef ecosystems in deeper waters my be sheltered from direct storm events and other surface-related energy (Liddell and Ohlhorst 1988). While MCEs appear to be spared of direct damage resulting from storms, tsunamis and hurricanes, they are highly susceptible to secondary effects. These secondary effects are largely a result of debris avalanches, where corals and other reef organisms are dislodged from shallow waters and slide down the reef slope (Dollar 1982, Harmelin-Vivien and Laboute 1986).

Mesophotic regions may also provide refuge from direct anthropogenic impacts such as fishing pressure, recreational snorkeling and diving, and ship groundings. Reef fish collections

and fishing is generally restricted to depths of less than 20 meters. Most reef fish fishing is conducted via spearfishing while freediving or on SCUBA, and divers rarely attain depths greater than 20 meters due to physiological and gear restraints. Overfishing is common among populated coral reef regions, as documented by extreme shifts in trophic structure away from apex predator dominated reefs in pristine/uninhabited locations to primary and secondary consumer dominated reefs in populated regions (Friedlander et al. 2010, Williams et al. 2011). Friedlander (2010) notes that apex predator abundance is most pronounced at greater depths (20 meters). These findings may indicate that large predators, which are the most prized fishes for consumption, are seeking refuge at deeper depths to escape fishing pressure. While entirely plausible, refuge from fishing pressure has not been investigated to date.

Coral reef recreation has been increasing in popularity in the past two decades, largely from technological advances and increased safety for SCUBA diving. Tourism in coral reef regions is a booming business, and recent estimates of the main Hawaiian Islands' coral reefs are valued at approximately $34 billion (Bishop et al. 2011). The majority of coral reef tourism revolves around snorkeling and SCUBA diving. While tourism depends largely on healthy coral reefs, many tourist activities unintentionally cause direct damage to reefs from lack of spatial awareness while snorkeling or SCUBA diving, or anchoring boats and kayaks (Barker and Roberts 2004). The depth of MCE reefs provides protection from most recreational activities, as MCE reefs exceed recreational SCUBA diving depths (30m) and are typically too deep to effectively anchor vessels.

The concept of connectivity between reef areas has been a focus of investigation for the past decade, largely a result of increased precision in DNA and RNA studies. Connectivity studies have mainly been employed to investigate and design Marine Protected Areas (MPAs;

McCook et al. 2009, Almany et al. 2009) but recent studies in Hawaii have investigated connectivity between islands and island groups (Christie et al. 2010, Toonen et al. 2011). While all of the above studies have been conducted solely within shallow waters (<20m), there has been some recent work on determining connectivity between shallow and deep regions of coral reef habitats. Most of this work has been investigated in the Great Barrier Reef with corals as the focal study group (Bongaerts et al. 2010b, van Oppen et al. 2011, Bongaerts et al. 2011). One study found strong genetic partitioning between deep slope, upper slope and backreef habitats, indicating that corals (or at least the Pocilloporid species studied here) likely do not transport larvae from deep to shallow reef areas (Bongaerts et al. 2010b, 2011). Van Oppen (2011) found that the same coral species as noted above had less genetic structuring in a different geographic region, and analyses of individuals produced evidence of gene flow between deep and shallow coral populations. The limited number of genetic studies has been focused solely on coral species of one genus (Pocillopora) and have found varied results, indicating that more work is needed to further resolve the deep reefs as a source of propagules hypothesis. Additionally, there has been no work published to date on the connectivity of reef fish populations. Due to reef fishes' movement patterns and habitat plasticity it is feasible that typically shallow fishes may move to deeper waters either for spawning events or for additional feeding grounds and thus become a deep source of propagules for reefs of all depths.

In this dissertation I undertake a critical analysis of the "deep reefs as refugia" hypothesis as it pertains to coral reef fishes in the Hawaiian Islands. At the start of this study, no information existed on mesophotic coral reefs in the Main Hawaiian Islands, and particularly in Hawaiʻi Island. Due to the paucity of data at mesophotic depths, I first had to collect baseline data on the abundance and distribution of mesophotic coral reefs and the reef fishes that inhabit them. In

Chapter One, I detail the first descriptions of mesophotic coral reef fish communities from 3-50 meters depth in West Hawai′i. Community structure changed gradually with depth, with more than 78% of fish species observed at mesophotic depths also found in shallow reef habitats. Depth explained 17% of the variation in reef-fish community structure; live coral cover explained 10% and prevalence of sand accounted for 7% of the fitted variation indicating that depth-related factors and coral habitat play a predominant role in structuring these communities. Differences in community structure also appear to be linked closely with feeding behavior. Trophic designation accounted for 31% of the fitted variation, with changes in herbivore abundance accounting for 10% of the variation. These findings suggest that changes in reef-fish community composition from shallow to mesophotic environments are largely influenced by trophic position, coral habitat and indirect effects of depth itself.

Chapter Two explores possible mechanisms underlying the decline in herbivorous fishes from shallow to mesophotic depths. Herbivorous fishes often play an integral role in preventing macroalgae from displacing corals, yet these fishes decline with depth despite the presence of coral-dominated habitats to >80m. Chapter Two investigates the relationship between herbivorous fish species and a suite of bottom-up and top-down parameters along a depth gradient from 3-50m. Turf algae, macroalgal and coral cover, nutrient quality and palatability, temperature, light, and predator density were studied as potential mechanisms limiting the distribution of herbivorous fishes with depth. Mesophotic reefs were not limited by food resources as mesophotic algal communities had similar nutrient content, species assemblages, and appeared highly palatable from algal choice experiments. Results of in-situ and long-term temperature recordings suggest temperature does not restrict herbivorous fish distributions at upper mesophotic depths. Multivariate redundancy analyses reveal the combination of increased

habitat patchiness and reduced light levels best explain herbivorous fish distributions with depth, indicating the reduction in herbivorous fishes at upper mesophotic depths is likely the result of non-consumptive predation effects. These results suggest changes in herbivorous fish populations with depth are likely the result of top-down effects and behavioral choices rather than abiotic constraints or resource limitation.

Chapter Three comprises the culmination of data collected on coral reef fishes and their environments across the Main Hawaiian Islands. In this chapter, I undertake a critical analysis of the "deep reefs as refugia" hypothesis as it pertains to coral reef fishes. In summary, MCEs between 30-60m may offer limited refugia for shallow reef fishes. Upper MCEs offer more thermally stable environments and >70% of fishes encountered were shallow reef fishes. Below 60m, shallow reef fish occurrence is reduced to 36%. It is also probable that upper MCEs act as a source of propagules for shallow reefs as we documented the first recorded mesophotic spawning aggregations of two species of parrotfish. Conversely, MCEs overall had lower fish densities and reef fish communities are largely comprised of invertiore and planktivore functional groups that are generally not of commercial or recreational value. Significant reductions in herbivorous fishes with depth indicate that MCEs will have limited capacity to re-seed shallow reefs with fishes of economic and ecological importance. While there is support for upper mesophotic reef environments (30-60m) to act as environmental refugia and harbor shallow reef fishes, the potential to restore shallow environments is limited and likely would not result in similar community compositions that currently comprise shallow reefs.

REFERENCES

Almany GR. SR Connolly, DD Heath, JD Hogan, GP Jones, LJ McCook, M Mills, RL Pressey, DH Williamson. 2009. Connectivity, biodiversity conservation and the design of marine reserve networks for coral reefs. *Coral Reefs* 28: 339–351.

Bak RPM. G Nieuwland, EH Meesters. 2005. Coral reef crisis in deep and shallow reefs: 30 years of constancy and change in reefs of Curacao and Bonaire. *Coral Reefs* 24: 475-479.

Bak RPM. G Nieuwland, EH Meesters. 2009. Coral growth rates revisited after 31 years: what is causing lower extension rates in *Acropora palmata*? *Bull Mar Sci* 84: 287-294.

Barker NHL and CM Roberts. 2004. Scuba diver behavior and the management of diving impacts on coral reefs. *Biol Conserv* 120(4):481-489.

Bishop RC. DJ Chapman, BJ Kanninen, JA Krosnick, B Leeworthy, and NF Meade. 2011. *Total economic value for protecting and restoring Hawaiian coral reef ecosystems: final report.* Silver Spring, MD: NOAA Office of National Marine Sanctuaries, Office of Response and Restoration, and Coral Reef Conservation Program. NOAA Technical Memorandum CRCP 16. 406 pp.

Bongaerts P. T Ridgway, EM Sampayo, and O Hoegh-Guldberg. 2010a. Assessing the 'deep reef refugia' hypothesis: focus on Caribbean reefs. *Coral Reefs* 29: 309-327.

Bongaerts P. C Riginos, T Ridgway, EM Sampayo, MJH van Oppen, N Englebert, F Vermuelen, O Hoegh-Guldberg. 2010b. Genetic divergence across habitats in the widespread coral *Seriatopora hystrix* and its associated *Symbiodinium*. *PLoS ONE* 5(5): e10871. doi:10.1371/journal.pone.0010871

Bongaerts P. C Riginos, KB Hay, MJH van Oppen, O Hoegh-Guldberg, S Dove. 2011. Adaptive divergence in a scleractinian coral: physiological adaptation of *Seriatopora hystrix* to shallow and deep reef habitats. *BMC Evolutionary Biology* 11: 303. doi:10.1186/1471-2148-11-303

Bridge T and J Guinotte. 2012. *Mesophotic coral reef ecosystems in the Great Barrier Reef world heritage area: their potential distribution and possible role as refugia from disturbance.* Great Barrier Reef Marine Park Authority. Townsville.

Brokovich E. S Einbinder, S Kark, N Shashar and M Kiflawi. 2007. A deep nursery for juveniles of the zebra angelfish *Genicanthus caudovittatus*. *Environ. Biol. Fish* 80: 1-6.

Brokovich E. S Einbinder, N Shashar, M Kiflawi, and S Kark. 2008. Descending to the twilight-zone: changes in coral reef fish assemblages along a depth gradient down to 65 m. *Mar Ecol Prog Ser* 371: 253-262.

Christie MR. BN Tissot, MA Albins, JP Beets, Y Jia, DM Ortiz, SE Thompson, MA Hixon. 2010. Larval connectivity in an effective network of Marine Protected Areas. *PLoS ONE* 5(12): e15715. Doi:10.1371/journal/pone.0015715

De'ath G. JM Lough, and KE Fabricius. 2009. Declining coral calcification on the Great Barrier Reef. *Science* 323: 116-119.

Dollar SJ. 1982. Wave stress and coral community structure in Hawaii. *Coral Reefs* 1: 71-81.

Frade PR. F de Jongh, F Vermeulen, J van Bleijswijk, and RPM Bak. 2008. Variation in symbiont distribution between closely related coral species over large depth ranges. *Mol Ecol* 17:691-703.

Fricke HW, Vareschi E, and D Schlichter. 1987. Photoecology of the coral *Leptoseris fragilis* in the Red Sea twilight zone (an experimental study by submersible). *Oecologia* 73: 371-381.

Friedlander AM. SA Sandin, EE DeMartini, and E Sala. 2010. Spatial patterns of the structure of reef fish assemblages at a pristine atoll in the Central Pacific. *Mar. Ecol. Prog. Ser.* 410: 219-231.

Goreau T and N Goreau. 1973. The ecology of Jamaican coral reefs. II. Geomorphology, zonation, and sedimentary phases. *Bull. Mar. Sci.* 23: 399-464.

Glynn PW. 1996. Coral reef bleaching: facts, hypotheses and implications. *Global Change Biol* 2: 495-509.

Guinotte JM. 2007. *Understanding environmental controls and predicting climate change effects on the health and occurrence of coral communities and their constituent organisms.* PhD Thesis, School of Tropical Environment Studies and Geography, James Cook University, Townsville. 45 pp.

Harmelin-Vivien ML and P Laboute. 1986. Catastrophic impact of hurricanes on atoll outer reef slopes in the Tuamotu (French Polynesia). *Coral Reefs* 5:55-62.

Hinderstein LM, JCA Marr, FA Martinez, MJ Dowgiallo, KA Puglise, D Zawada, and R Pyle. 2010. Introduction to mesophotic coral ecosystems: Characterization, ecology, and management. *Coral Reefs* 29: 247-251.

Hoegh-Guldberg O. 1999. Climate change, coral bleaching and the future of the world's coral reefs. *Mar Freshw Res* 50:839-866.

Hoegh-Guldberg O. PJ Mumby, AJ Hooten, RS Steneck, P Greenfield, E Gomez, CD Harvell, PF Sale, AJ Edwards, K Caldeira, N Knowlton, CM Eakin, R Iglesias-Prieto, N Muthiga, RH Bradbury, A Dubi, and ME Hatziolos. 2007. Coral reefs under rapid climate change and ocean acidification. *Science* 318: 1737-1742.

Hughes TP and JE Tanner. 2000. Recruitment failure, life histories, and long-term decline of Caribbean corals. *Ecology* 81: 2241–2249.

Hughes TP. AH Baird, DR Bellwood, M Card, SR Connolly, C Folke, R Grosberg, O Hoegh-Guldberg, JBC Jackson, J Kleypas, JM Lough, P Marshall, M Nystrom, SR Palumbi, JM Pandolfi, B Rosen, J Roughgarden. 2003. Climate change, human impacts, and the resilience of Coral Reefs. *Science* 301: 929–933.

Hughes TP. RJ Rodrigues, DR Bellwood, D Ceccarelli, O Hoegh-Guldberg, L McCook, L Moltschaniwskyj, MS Pratchett, R Steneck, and B Willis. 2007. Phase shifts, herbivory, and the resilience of coral reefs to climate change. *Curr Biol* 17:360-365.

Kahng SE and JE Maragos. 2006. The deepest zooxanthellate, scleractininan corals in the world? *Coral Reefs* 25: 254

Lesser, MP. M Slattery and JJ Leichter. 2009. Ecology of mesophotic coral reefs. *J. Exp. Mar. Biol. Ecol.* 375: 1-8.

Liddell WD and SL Ohlhurst. 1988. Hard substrata community patterns, 1-120m, North Jamaica. *Palaios* 3: 413-423.

Liddell WD. WE Avery and SL Ohlhurst. 1997. Patterns of benthic community structure, 10-250m, the Bahamas. *Proc. 8th Int. Coral Reef Symp.* Vol 1 pp 437-442.

Liu, G. JE Meyer, IC Guch, and MA Toscano. 2001. NOAA's satellite coral reef bleaching early warning products aimed at local reef sites around the globe. *Reef Encounter* 30: 10-13.

Liu G. AE Strong, and W Skirving. 2003. Remote sensing of sea surface temperature during 2002 Barrier Reef coral bleaching. *EOS* 84(15): 137-144.

Maragos JE and P Jokiel. 1986. Reef corals of Johnston Atoll – one of the World's most isolated reefs. *Coral Reefs* 4: 141-150

McClanahan, TR. SD Donner, JA Maynard, MA MacNeil, NAJ Graham, JM Maina, AC Baker, JB Alemu, IM Beger, SJ Campbell, ES Darling, CM Eakin, SF Heron, SD Jupiter, CJ Lundquist, E McLeod, PJ Mumby, MJ Paddack, ER Selig, and R van Woesik (2012). Prioritizing key resilience indicators to support coral reef management in a changing climate, *PLoS ONE* 7(8): e42884.

McCook LJ. GR Almany, ML Berumen, JC Day, AL Green, GP Jones, JM Leis, S Planes, GR Russ, PF Sale, SR Thorrold. 2009. Management under uncertainty: guide–lines for incorporating connectivity into the protection of coral reefs. *Coral Reefs* 28: 353–366.

Menza, C. M. Kendall and S. Hile. 2008. The deeper we go the less we know. *Rev. Biol. Trop.* 56(1): 11-24

Pandolfi JM. 2002. Coral community dynamics at multiple scales. *Coral Reefs* 21:13-23.

Puglise KA. LM Hinderstein, JCA Marr, MJ Dowgiallo and FA Martinez. 2009. *Mesophotic Coral Ecosystems Research Strategy: International workshop to prioritize research and management needs for mesophotic coral ecosystems, Jupiter, Florida, 12-15 July 2008.* Silver Spring, MD: NOAA National Centers for Coastal Ocean Science, Center for Sponsored Coastal Ocean Research, and Office of Ocean Exploration and Research, NOAA Undersea Research Program. NOAA Technical Memorandum NOS NCCOS98 and OAR OER 2. 24 pp.

Pyle RL. 1996. How much coral reef biodiversity are we missing? *Global Biodiversity* 6(1): 3-7.

Pyle RL. 2000. Assessing undiscovered fish biodiversity on deep coral reefs using advanced self-contained diving technology. *Mar. Tech. Soc. J.* 34(4): 82-91.

Reaka ML, PJ Rodgers and AU Kudla. 2008. Patterns of biodiversity and endemism on Indo-West Pacific coral reefs. *Proc Natl Acad Sci* USA 105: 11474-11481

Reed JK. 1985. Deepest distribution of Atlantic hermatypic corals discovered in the Bahamas. In: *Proc. 5th Int. Coral Reef Symp*, Vol 6, pp 249-254.

Riegl B and WE Piller. 2003. Possible refugia for reefs in times of environmental stress. *Int J Earth Sci* 92: 520-531.

Slattery M. MP Lesser, D Brazeau, MD Stokes, and JJ Leichter. 2011. Connectivity and stability of mesophotic coral reefs. *J. Exp. Mar. Bio Ecol*. 408: 32-41.

Toonen RJ. KR Andrews, IB Baums, CE Bird, GT Concepcion, TS Daly-Engel, JA Eble, A Faucci, MR Gaither, M Iacchei, JB Puritz, JK Schultz, DJ Skillings, MA Timmers, BW Bowen. 2011. Defining boundaries for Ecosystem-Based Management: A multispecies case study of marine connectivity across the Hawaiian Archipelagao. *J Mar Bio* doi:10.1155/2011/460173

van Oppen MJH. P Bongaerts, JN Underwood, LM Peplow, TF Cooper. 2011. The role of deep reefs in shallow reef recovery: an assessment of vertical connectivity in a brooding coral from west and east Australia. *Mol Ecol* 20: 1647–1660.

Waddington KI. JJ Meeuwig, SN Evans, and LM Bellchambers. 2010. Assessment of the benthic biota of a deep coastal ecosystem by remote and in situ sampling techniques. *Mar. Fresh. Res.* 61: 1164-1170.

Wilkinson C. 2008. *Status of Coral Reefs of the World: 2008.* Global Coral Reef Monitoring Network and Reef and Rainforest Research Centre, Townsville.

Williams ID. BL Richards, SA Sandin, JK Baum, RE Schroeder, MO Nadon, B Zgliczynski, P Craig, JL McIlwain, and RE Brainard. 2011. Differences in reef fish assemblages between populated and remote reefs spanning multiple archipelagos across the Central and Western Pacific. *J Mar Bio* doi:

CHAPTER ONE

TROPHIC DESIGNATION AND LIVE CORAL COVER PREDICT CHANGES IN

REFF-FISH COMMUNITY STRUCTURE ALONG A SHALLOW TO

MESOPHOTIC GRADIENT IN HAWAII

ATTRIBUTION

C.N. Kane contributed to the conceptual development of this manuscript, collected and analyzed all data, and was the primary author of the text. B.N. Tissot contributed to manuscript review and editing.

TROPHIC DESIGNATION AND LIVE CORAL COVER PREDICT CHANGES IN REFF-FISH COMMUNITY STRUCTURE ALONG A SHALLOW TO MESOPHOTIC GRADIENT IN HAWAII

1.1 Abstract

Reef-fish community structure and habitat associations are well documented for shallow coral reefs (<20 m) but are largely unknown in deeper extensions of reefs (mesophotic reefs; >30 m). We documented the community structure of fishes and seafloor habitat composition through visual observations at depth intervals from 3 to 50 m in West Hawaii. Community structure changed gradually with depth, with more than 78% of fish species observed at mesophotic depths also found in shallow reef habitats. Depth explained 17% of the variation in reef-fish community structure; live coral cover explained 10% and prevalence of sand accounted for 7% of the fitted variation indicating that depth-related factors and coral habitat play a predominant role in structuring these communities. Differences in community structure also appear to be linked closely with feeding behavior. Trophic designation accounted for 31% of the fitted variation, with changes in herbivore abundance accounting for 10% of the variation. These findings suggest that changes in reef-fish community composition from shallow to mesophotic environments are largely influenced by trophic position, coral habitat and indirect effects of depth itself.

1.2 Introduction

Factors structuring reef-fish communities have been researched extensively in the past 50 yr, largely following the advent and widespread use of SCUBA diving (Sale 1991; Pyle 2000; Hixon 2011). Much of the spatial variation in reef-fish community composition has been linked

to habitat availability and complexity (Hixon and Beets 1993; Munday 2000; Almany 2004; Gratwicke and Speight 2005; Brokovich et al. 2006). In addition to habitat parameters, depth and associated abiotic gradients are also known to play substantial roles in the community composition and abundance of reef fishes at shallow depths (<20 m; McGehee 1994; Friedlander and Parrish 1998; Arreola-Robles and Elorduy-Garay 2002; Donaldson 2002; Brokovich et al. 2006). Due to the limitations of conventional SCUBA gear, most knowledge of reef fishes and their habitats has been limited to depths of 20 m or less, yet coral reefs commonly extend to depths in excess of 80 m (Maragos and Jokiel 1986; Kahng and Maragos 2006; Menza et al. 2007 Reaka et al. 2008; Kahng et al. 2010).

The few ecological studies conducted on mesophotic reefs (30–150 m) support the premise of deep reefs providing substantial habitat for fishes (Brokovich et al. 2007, 2008; Lesser et al. 2009; Bejarano et al. 2014). Studies of mesophotic communities in the Caribbean and Red Sea reveal that coral cover is consistent to at least 50–60 m depth (Goreau and Goreau 1973; Fricke and Schumacher 1983; Liddell and Ohlhorst 1988; Liddell et al. 1997; Bak et al. 2005; Brokovich et al. 2008). Recent work in the Au'au Channel in Hawaii has revealed a unique and extensive mesophotic coral bed, providing structure for immense mesophotic fish communities (Kahng et al. 2010). These findings suggest that the extent of coral reefs is much greater than previously thought and that mesophotic ecosystems may provide significant habitat and possible refuge for shallow coral-reef fishes (Bridge et al. 2013; Baker et al 2016).

Mechanisms underlying changes in coral-reef communities from shallow to mesophotic depths are not well understood (Slattery et al. 2011; van Oppen et al. 2011; Kahng et al. 2014). Corals, fish and other organisms have been found from shallow to upper mesophotic zones (30–50 m); in the Caribbean, nearly 80% of coral species occur to depths greater than 30 m, while

over 40% of coral species extend to 30+ m in the Indo-Pacific (Bridge et al. 2013). Few studies have focused on the ecology of fishes of mesophotic coral ecosystems (MCE, >30 m). These studies predominantly describe species composition and species–habitat relationships using observational techniques, and relate changes in community structure largely to depth and changes in coral abundance or morphology (Colin 1974; Thresher and Colin 1986; Pyle 2000; Feitoza et al. 2005; Brokovich et al. 2007, 2008, 2010a; Garcia-Sais 2010; Bryan et al. 2013; Bejarano et al. 2014; Schultz et al. 2014; Lindfield et al. 2016). To date, five studies have investigated reef fishes at mesophotic depths in Hawaii: three in the remote northwestern Hawaiian Islands (Parrish and Boland 2004; Kane et al. 2014; Fukunaga et al. 2016); one from Maui in the main Hawaiian Islands (Boland and Parrish 2005); and one encompassing both the northwestern Hawaiian Islands and Au'au Channel in Maui (Pyle et al. 2016). Thus, our understanding of reef-fish abundance and community structure in the Hawaiian Archipelago from shallow to mesophotic depths is extremely limited, and in the majority of islands, completely absent.

In this study, we aimed to test two primary hypotheses: (1) community composition of coral-reef fishes changes gradually from shallow to upper mesophotic depths; and (2) changes in reef-fish community composition with depth can be predicted by trophic position or habitat variables. Given the absence of quantitative reef-fish data below 20 m in West Hawaii, this study provides the first baseline estimates for reef fishes in shallow mesophotic depths and sheds light on depth-related changes in community structure and function in this underrepresented region.

1.3 Materials and methods

1.3.1 Study sites

Hawaii Island is geologically the youngest of all the islands in the archipelago with a steep bathymetric gradient and coral reefs that extend continuously from shore to depths of approximately 50 m. Beyond that depth, large sand flats occur to 70 m depth or more, with reef often re-emerging around 70 m and patchily continuing to depths currently undetermined. Eleven sites were selected along West Hawaii's coastline in areas where continuous coral-reef habitat occurs from shallow waters to at least 30 m (Fig. 1). Due to the steeply sloping bathymetry, the 50 m depths surveyed were often within 100–200 m of the shoreline, thus physical distance between the shallowest and deepest surveys were between 50 and 200 m from shore.

1.3.2 Sampling design

A stratified sampling design was used to explore changes in reef-fish abundance and community structure with depth. Concordant depth and bathymetric relief among sites was sought to maintain consistency and reduce potential survey bias. Visual surveys of reef fish and benthic substrates were conducted on SCUBA at approximately 10-m depth intervals during July–August in 2013–2015. Due to variability of habitat below 30 m, three sites were sampled to 50 m, seven to 40 m and one to 30 m. In each site, transects were laid parallel to shore at 3 m, 10 m and then in 10-m depth intervals until the deepest extent of the reef. A minimum of three surveys were conducted at each depth contour at each site. Transects were spaced a minimum of 5 m apart. This resulted in an incomplete survey design with 35 surveys at 3 m, 37 surveys at 10 m, 38 surveys at 20 m, 38 surveys at 30 m, 34 surveys at 40 m, and 30 surveys at 50 m (Electronic supplementary material, ESM Table S1). Visual fish surveys were conducted at each depth (3–50 m) using a 25 m x 4 m belt transect (Hill and Wilkinson 2004). Belt length, width and level of replication per depth (minimum n = 3) were dictated by technical constraints resulting from the short bottom times allowed from using SCUBA at 50 m depths (Brokovich et

al. 2008; Sandin et al. 2008). Within each fish belt transect, all fishes were identified to species (according to Randall 2007) and tallied individually. Benthic survey data was collected at two of the replicate transects within each depth by photographing a 0.25-m^2 quadrat every meter (n = 25 per transect) using an underwater camera equipped with a PVC photoquad framer and dual lighting system.

1.3.3 Data analysis

Species accumulation curves were used to verify sampling effort (MM model; Primer-E+; ESM Fig. S1). Sites were grouped into three geographic regions (North, Central, South) to ensure adequate sampling effort for analyses (Fig. 1; ESM Table S1). To ascertain general trends in reef-fish diversity and abundance with depth we calculated the mean species richness (alpha diversity) and abundance of all fishes with depth. Richness (alpha diversity) was analyzed using a generalized linear model (GLM) based on a Gaussian distribution while abundance was analyzed using a GLM based on a Poisson distribution (two factors: depth and region; JMP 12.1.0, SAS Institute Inc.) Post hoc Tukey tests were used to identify significant differences among depths.

Changes in reef-fish community composition were analyzed using principal coordinates analyses (PCO) with PERMANOVA (two factors: depth and region) to identify significant differences among species assemblages and depth using Bray–Curtis similarity matrices on square-root transformed abundance data (Primer v. 6.1.13, Primer-E+). Changes in reef-fish community composition were tested against trophic position, benthic composition and physical structure using a distance-based linear model (DistLM) and redundancy analysis (dbRDA) (Primer v. 6.1.13, Primer-E+).

DistLM partitions the variation of multivariate data according to multiple regression models to analyze relationships within a multivariate data cloud (Legendre and Anderson 1999). A dbRDA provided an ordination of fitted values using the multivariate multiple regression of the relationship between reef-fish community structure and the predictor variables (Anderson et al. 2008). Vector overlays on dbRDA ordination diagrams were used to aid in determining the strength and direction of the relationship between variables and redundancy analysis axes. The length of each vector corresponds to the size of the effect the variable had on the construction of dbRDA axes (Anderson et al. 2008). We tested for multicollinearity before DistLM and dbRDA analyses. No two variables had correlations stronger than 0.6 and were therefore considered independent.

Trophic position was examined by binning reef-fish species into six broad trophic guilds. Trophic assignment was classified according to gut content analyses published on Hawaiian fish species in Hobson (1974) and Randall (2007). Species lists and assignments were corroborated with the National Oceanic and Atmospheric Administration's Coral Reef Ecosystem Program. Comparisons of trophic guilds with depth were performed using one-way ANOVAs with post hoc Tukey tests.

Benthic composition and physical structure estimates were examined to investigate changes in the benthos with depth. Benthic photoquadrat samples were analyzed using CoralNet benthic image analysis (Beijbom et al. 2015). Fifty random points were placed within each photograph and the biota or substratum directly underneath each point was identified to the lowest taxonomic classification possible (ESM Table S2; Fenner 2005; Huisman et al. 2007). Taxonomic identifications were pooled to create descriptive habitat categories which included turf algae, macroalgae, sand, and live coral cover. These broad categories were selected as live

coral cover, turf and macroalgal cover are strongly correlated with reef-fish community composition at shallow depths (Friedlander and Parrish 1998; Arreola-Robles and Elorduy-Garay 2002; Donaldson 2002; Wilson et al. 2010; Hoey et al. 2013; Evans et al. 2014). Live coral cover was further categorized into designations based on general morphology as fishes have been shown to strongly associate with branching and plating coral morphologies. Three categories were included for analysis: branching/plating, encrusting, and lobate morphologies. Categories were summed by photoquadrat and averaged (n = 25) to obtain one cover estimate per transect to correspond with fish transects. Log-transformed benthic percentage cover data for turf algae, sand, and coral were compared across transect depths using one-way ANOVAs with post hoc Tukey tests. Macroalgal data were analyzed using the non-parametric Wilcoxon test and post hoc Wilcoxon planned comparisons as variances were not equal.

We tested whether the underlying physical substrate influenced fish community composition. Each photoquatrat sample was visually assigned a primary (>50% of photograph) structure identification using one of eight structural categories: basalt — continuous bare lava formations; compressa bed — complex interstitial matrix created by either live or dead *Porites compressa* beds; pavement — flat, hard bottom structure; pebble/cobble — small (<25 cm) rocks or pebbles; rubble — small fragments of dead coral; carbonate — outer layer of live or dead coral skeletons, predominantly mounding corals; sand/sediment — fine-grain sands or sediments; and boulder — large rocks (>25 cm). Primary benthic structure codes were then summed by transect (n = 25) and averaged to generate percentage cover of structural categories by transect for comparison.

1.4 Results

1.4.1 Community composition

We recorded 26,553 fish from 150 species and 33 families within 212 transects at 11 sites along the West Hawaii coast. Species richness significantly differed across depths and regions sampled (GLM: depth $\chi^2_{5,212} = 54.46$, $P \le 0.001$; region $\chi^2_{2,212} = 7.95$, $P = 0.019$; depth x region $\chi^2_{10,212} = 44.15$, $P \le 0.001$; Fig. 2a). Richness generally decreased from shallow (3–20 m) to mesophotic (30–50 m) depths. A significant interaction between depth and region occurred because of unexpectedly high richness at 40 and 50 m depths at one site (Kona Paradise) in the southern region. Overall fish abundance (Fig. 2b) significantly differed across depth (GLM: $\chi^2_{5,212} = 59.01$, $P \le 0.001$) and regions sampled (GLM: $\chi^2_{2,212} = 14.15$, $P = 0.001$) but there was no depth x region interaction (GLM: $\chi^2_{10,211} = 7.61$, $P = 0.67$). Overall fish abundance decreased with depth, with each region displaying similar declines with depth but slightly different overall abundances. Northern sites generally had lower abundance of fishes than Central and Southern sites. Fish abundance was significantly greater at 3–20 m than at mesophotic depths (30–50 m) (post hoc Tukey tests).

The community structure of reef fishes transitioned gradually with depth (PCO analysis; Fig. 3). Shallow reef-fish communities were more distinct (spatially separated groupings), while deeper communities were similar (groupings largely overlapping), particularly at 30–50 m. Axis 1 explained 21% of the total variation and corresponds most strongly with transect depth. PERMANOVA tests using species abundance data indicated that both region and depth were significant in structuring communities (depth: Pseudo-$F_{5,212} = 18.47$, p(perm) = 0.001; region: Pseudo-$F_{2,212} = 6.06$, p(perm) = 0.001; depth x region: Pseudo-$F_{10,212} = 3.13$, p(perm) = 0.001).

1.4.2 Patterns with depth

Species distributions varied greatly with depth (ESM Table S3), with some species found at only a single depth and others found at all six depths (Table 1). Depth-generalist species

(observed at four or more depths, n = 79) outnumbered depth-specialist species (found at one or two contiguous depths, n = 50). Depth specialists were evenly divided between shallow specialists (<30 m; n = 21) and mesophotic specialists (>30 m; n = 21), and were not concordant with genus or family (ESM Table S3).

DistLM analysis indicated significant relationships between the reef-fish community structure and 20 of the 21 variables tested (pavement not statistically significant; Table 2). Depth accounted for the largest proportion of fitted variance in fish community structure (17%) while herbivore abundance and live coral cover accounted for ~10% each (Table 2). The decline in herbivore, detritivore, and corallivore abundance with depth accounted for nearly 25% of the variation in community structure, while the decrease in live coral cover and increase in sand accounted for approximately 18% of variation observed in the model. Axis 1 of the dbRDA accounted for 46% of fitted variation (18% of the total variation) and strongly correlated with depth (Fig. 4). Vector overlays indicate that sandy habitats increase with depth, while coral-dominated habitats are associated more strongly with shallow depths. Both herbivorous and corallivorous fishes are more closely associated with shallow depths. Axis 2 explained 14% of the fitted variation (5% of total variation) and appears largely associated with site-based differences.

Habitat analyses reveal significant increases in sand and macroalgal cover with depth (ANOVA; Macroalgae: $F_{5,146} = 13.79$, $P < 0.001$; Fig. 5a; Sand: $F_{5,146} = 17.65$, $P < 0.001$; Fig. 5d). While sand increased gradually with depth, macroalgae remained at low cover from 3 to 40 m depth and significantly increased at 50 m depth. Turf algae significantly differed among depths but did not exhibit any depth-related patterns (ANOVA: $F_{5,146} = 3.7$, $P = 0.004$; Fig. 5b). Coral cover peaked at 10 m and gradually but significantly decreased with depth; mesophotic

depths had significantly less coral cover than shallower depths (ANOVA: $F_{5,146} = 25.06$, $P < 0.001$; Fig. 5c). Coral morphologies also differed significantly with depth (Fig. 5c). Branching/plating morphologies peaked in cover at 20 m and declined gradually with depth ($F_{5,137} = 6.58$, p < 0.001). Lobate morphologies also differed significantly with depth, peaking in percentage cover at 10 m and declining gradually with depth, with mesophotic depths having significantly lower lobate cover than 3–20 m depths ($F_{5,137} = 20.9$, $P < 0.001$). Encrusting morphologies remained at low but consistent cover across depth ($F_{5,137} = 1.92$, $P = 0.11$).

Trophic composition varied significantly across depth. Abundance of fishes in nearly every trophic group significantly declined with depth (Detritivores: $F_{5,211} = 13.47$, $P < 0.001$; Herbivores: $F_{5,211} = 24.25$, $P < 0.001$; Omnivores: $F_{5,211} = 4.68$, $P < 0.001$; Zooplanktivores: $F_{5,211} = 3.89$, $P < 0.002$; Corallivores: $F_{5,211} = 24.36$, $P < 0.001$; Fig 6). The only exception were the invertivores, which differed significantly among depths but did not exhibit any directional pattern ($F_{5,211} = 4.39$, $P < 0.001$) and piscivorous fishes which did not differ significantly with depth ($F_{5,211} = 1.3$, $P = 0.27$). Trophic groups differed in the rate of decline between shallow (0–20 m) and mesophotic (30–50 m) depths. Detritivore (84%), corallivore (72%) and herbivore (68%) fish abundances declined nearly twice as much as zooplanktivore (40%) fishes from shallow to mesophotic depths.

1.5 Discussion

The mesophotic zone of coral reefs has been defined as beginning at 30 m and extending to the lower distributional limit of light-dependent coral-reef communities (Hinderstein et al. 2010; Kahng et al. 2010). Our study highlights that while technically in the MCE range, the 30–50 m fish communities are largely extensions of shallow reef communities, with more than 78% of the fish species observed at MCE depths typically associated with shallow waters. Our data

corroborates the few other mesophotic fish studies in that these shallow MCE regions act as transition zones between shallow and deep fish communities (Brokovich et al. 2008; Garcia-Sais 2010; Bejarano et al. 2014; Rosa et al. 2016). In particular, Rosa et al. (2016) found nearly identical overlaps of shallow fishes in upper mesophotic depths (80%) as our study (78%), indicating that the upper mesophotic zone (30–50 m) is still largely dominated by common shallow reef-fish species. The deep specialists noted here are species found commonly on mesophotic reefs in the Hawaiian Archipelago, indicating that the 40–50 m zone in West Hawaii acts as the upper limit for common mesophotic fish species and is congruent with results from other islands within the Hawaiian Archipelago (Pyle et al. 2016).

Trophic designation accounted for 33% of the variability in our model. Abundance analyses of trophic groups with depth indicate herbivores, detritivores, and corallivores decreased significantly and accounted for 24% of the variation in community structure in this study. Comparisons of trophic assemblages indicate a shift from herbivore-dominated communities in shallow depths to zooplanktivore-dominated communities at mesophotic depths. Decreasing herbivorous fish abundances in mesophotic depths was first reported at Enewetak in the 1980s (Thresher and Colin 1986), but has received scant attention until recently; it has now been confirmed in the Red Sea, Caribbean, and northwestern Hawaiian Islands (Brokovich et al. 2008, 2010a; Garcia-Sais 2010; Bejarano et al. 2014; Fukunaga et al. 2016).

The reduction in herbivorous fishes with depth does not appear to be related to food availability as we noted consistent turf algal cover across depth and significant increases in macroalgal cover at 50 m. In addition, many previous studies have reported significant increases in turf and macroalgae at mesophotic depths. Grazing pressure is reduced in mesophotic depths compared with shallow regions although grazing is still evident (Brokovich et al. 2010a; Kahng

et al. 2010). Two main hypotheses explain the reduction in herbivorous fishes with depth; first, mesophotic temperatures may be too low for herbivores to efficiently metabolize algae (Floeter et al. 2005); and second, deep algal species may be chemically defended and thus unpalatable for herbivorous fishes (Hay 1981, 1984). While herbivore abundances have been positively correlated with temperature across large latitudinal gradients (Floeter et al. 2005), the only mesophotic study of herbivory suggests that temperatures between shallow and upper mesophotic reefs are not substantially different (Brokovich et al. 2010a). Chemical defenses of mesophotic algal species have not been studied to date, but we have observed algal grazing by parrotfish and surgeonfish species at mesophotic depths, indicating that at least some of the algal community is edible. Decreases in herbivore abundance despite adequate food supply may be a result of differences in algal quality, not quantity. Studies in shallow systems have found that herbivore patchiness is strongly correlated with differences in algal productivity (Russ 2003; Tootell and Steele 2016). Although currently untested, it is plausible that productivity of turf and macroalgal resources decreases with depth given the low light availability at mesophotic depths, which could then account for observed declines in herbivorous fishes with depth.

While all trophic groups declined in abundance between shallow and mesophotic depths, herbivorous and detritivorous fishes declined at nearly twice the rate of zooplanktivorous fish abundances (herbivores 68%, detritivores 84%, zooplanktivores 40%).The dominance of zooplanktivorous fishes at mesophotic depths is supported at many locations worldwide (Thresher and Colin 1986; Brokovich et al. 2008; Garcia-Sais 2010; Bejarano et al. 2014) and has been hypothesized to result from higher zooplankton abundances and more nutrient-rich water at depth (Kahng et al. 2010). The only study to evaluate zooplankton communities at mesophotic depths reported negligible abundances but dramatic differences among locations,

indicating that more study is needed (Rodriguez-Jerez 2004). Water clarity often increases in mesophotic depths (R. Pyle pers. comm.), but it is unclear whether zooplankton abundance is correlated with water clarity or phytoplankton abundance in tropical insular waters. We have observed increased water clarity at mesophotic depths in West Hawaii, likely a result from reduced turbidity and lower phytoplankton concentrations common in shallow waters. It is more likely, though, that zooplanktivorous fish dominance results from reductions in herbivorous fish abundance at mesophotic depths rather than increased habitat suitability or food availability in upper mesophotic depths. More investigation is needed to confirm or refute possible mechanisms underlying these observed trophic shifts.

The decline in coral cover and gradual increase in sand/sediment cover with depth indicates that habitat is more patchily distributed at mesophotic sites in West Hawaii. Nine of the 11 sites surveyed in this study had reefs that ended into large sand beds at approximately 40–70 m depth. It is thus likely that reductions in overall abundance and species richness are linked with reduced habitat availability. Live coral cover is a significant predictor of reef-fish abundance and diversity in shallow reef systems (Williams 1991) but is not typically a strong predictor of fish assemblages within mesophotic systems, as coral habitat is greatly reduced at these depths. We found significant declines in cover of lobate and branching/plating coral morphologies at mesophotic depths, likely resulting in less suitable live coral habitat for many species of fishes. Brokovich et al. (2008) found declines in branching corals and live cover were the most pronounced habitat variables correlating with fish community structure in the Red Sea, while benthic habitat metrics accounted for little variation in fish community structure in Western Australia (Fitzpatrick et al. 2012). Our studies generally support those in the Red Sea, with reduced coral cover and increased sand cover explaining the most variability (17%

combined) in fish communities with depth among the 11 benthic and structural variables tested. Overall colony size appeared smaller in mesophotic depths than in shallow waters — in shallow waters lobate colonies were often meters long while at mesophotic depths most colonies were less than 20 cm (pers. obs.). Reductions in size of colonies also likely influenced the size and species composition of fishes at mesophotic depths. While not ubiquitous across all mesophotic depths, many mesophotic reefs experience reduced coral cover and increased algal cover, thus dissociation between coral cover and fish community structure is not unexpected. It is likely that the depth variable represents indirect effects on community structure through reductions in light availability which subsequently affect benthic cover and primary productivity, as well as predation success and behavior modifications among fishes in mesophotic systems (Brokovich 2010a).

While not tested directly, habitat complexity may play a stronger role overall than benthic cover or general structure of the benthos. Variation within each depth is largely tied to differences between coral rich areas (% live coral cover) and highly complex coral matrices (*Porites compressa* beds) versus basalt regions and sand/sediment areas that lack complex interstitial spaces. While coral cover decreases significantly with depth and live coral dominance is rare in West Hawaii below depths of 20 m, it is likely that remnant reef structures and boulder habitats compensate for the complexity typically provided by branching corals in shallow regions. Numerous studies have shown that complexity, via the abundance and size of shelter holes, is important in characterizing fish assemblages at shallow depths (Hixon and Beets 1989; Friedlander and Parrish 1998; Almany 2004). The prevalence of shallow reef-fish species to depths of 50 m is influenced by similarities in structural components but it is likely that mesophotic reefs lack the degree of fine-scale complexity found in shallow reefs resulting from

complex matrices provided by branching corals, which may account for observed reductions in overall abundance and species richness at mesophotic depths in West Hawaii. Many MCE regions describe high coral cover to depths of 60 m or more but these corals are largely comprised of mounding and plating morphologies, which provide far less complex shelter space than branching morphologies (Brokovich et al. 2008; Hinderstein et al. 2010; Kahng et al. 2010). The change in coral morphologies with depth results in changes in overall complexity and thus likely contributes to the variability in reef-fish composition between shallow and upper mesophotic systems.

Accumulating baseline information on mesophotic coral reefs is imperative to enhance both general knowledge and predictive capabilities regarding coral-reef fishes. While coral cover is one of the main drivers of reef-fish community composition in shallow waters, additional forces beyond live coral habitat appear to gain importance with depth; thus, more detailed studies of habitat complexity and niche availability would be very interesting. Trophic assemblages at mesophotic depths differ from those at shallow depths, and correlations between herbivorous fishes and algal abundance observed in shallow waters are not supported in mesophotic habitats (Choat 1991; Hughes et al. 2007; Brokovich et al. 2010a; Bejarano et al. 2014). It is therefore important to consider differential drivers of community structure with depth in future modeling efforts for population and community structure of reef fishes.

1.6 Acknowledgements

1.7 References

Almany GR (2004) Differential effects of habitat complexity, predators and competitors on abundance of juvenile and adult coral reef fishes. Oecologia 141:105–113

Anderson MJ, RN Gorley and KR Clarke (2008) PERMANOVA+ for PRIMER: guide to software and statistical methods. PRIMER-E, Plymouth, UK

Arreola-Robles JL, Elorduy-Garay JF (2002) Reef fish diversity in the region of La Paz, Baja California Sur, Mexico. Bull Mar Sci 70:1–18

Bak RPM, Nieuwland G, Meesters EH (2005) Coral reef crisis in deep and shallow reefs: 30 years of constancy and change in reefs of Curacao and Bonaire. Coral Reefs 24:475–479

Baker EK, Puglise KA, Harris PT (Eds.) (2016) Mesophotic coral ecosystems — A lifeboat for coral reefs? The United Nations Environment Programme and GRID-Arendal, Nairobi and Arendal, 98 p.

Beijbom O, Edmunds PJ, Roelfsema C, Smith J, Kline DI, Neal BP, Dunlap MJ, Moriarty V, Fan TY, Tan CJ, Chan S, Treibitz T, Gamst A, Mitchell BG, Kriegman D (2015) Towards automated annotation of benthic survey images: variability of human experts and operational modes of automation. PLoS One 10:e0130312

Bejarano I, Appeldoorn RS, Nemeth M (2014) Fishes associated with mesophotic coral ecosystems in La Parguera, Puerto Rico. Coral Reefs 33:313–328

Boland RC, Parrish FA (2005) Description of fish assemblages in the black coral beds off Lahaina, Maui, Hawai'i. Pacific Science 59:411–420

Bridge TCL, Hughes TP, Guinotte JM, Bongaerts P (2013) Call to protect all coral reefs. Nat Clim Chang 3:528–530

Brokovich E, Baranes A, Goren M (2006) Habitat structure determines coral reef fish assemblages at the northern tip of the Red Sea. Ecol Indic 6:494–507

Brokovich E, Einbinder S, Kark S, Shashar N, Kiflawi M (2007) A deep nursery for juveniles of the zebra angelfish *Genicanthus caudovittatus*. Environ Biol Fishes 80:1–6

Brokovich E, Einbinder S, Shashar N, Kiflawi M, Kark S (2008) Descending to the twilight-zone: changes in coral reef fish assemblages along a depth gradient down to 65 m. Mar Ecol Prog Ser 371:253–262

Brokovich E, Ben-Ari T, Kark S, Kiflawi M, Dishon G, Iluz D, Shashar N (2010a) Functional changes of the visual system of the damselfish *Dascyllus marginatus* along its bathymetric range. Physiol Behav 101:413–421

Brokovich E, Ayalon I, Einbinder S, Segev N, Shaked Y, Genin A, Kark S, Kiflawi M (2010b) Grazing pressure on coral reefs decreases across a wide depth gradient in the Gulf of Aqaba, Red Sea. Mar Ecol Prog Ser 399:69–80

Bryan DR, Kilfoyle K, Gilmore RG, Spieler RE (2013) Characterization of the mesophotic reef fish community in south Florida, USA. J Appl Ichthyol 29:108–117

Choat JH (1991) The biology of herbivorous fishes on coral reefs. In: Sale PF (ed.) The ecology of fishes on coral reefs. Academic Press, San Diego, CA, p 120−155

Colin PL (1974) Observation and collection of deep reef fishes off the coasts of Jamaica and Honduras. Mar Biol 24:29–38

Donaldson TJ (2002) Habitat association and depth distribution of two sympatric groupers of the genus *Cephalopholis* (Serranidae: Epinephelinae). Ichthyological Research 49:191–193

Evans RD, Wilson SK, Field SN, Moore JAY (2014) Importance of macroalgal fields as coral reef fish nursery habitat in north-west Australia. Mar Biol 161:599–607

Feitoza B, Rosa R, Rocha L (2005) Ecology and zoogeography of deep-reef fishes in northeastern Brazil. Bull Mar Sci 76:725–742

Fenner D (2005) Corals of Hawaii: a field guide to the hard, black, and soft corals of Hawaii and the northwest Hawaiian Islands, including Midway. Mutual Publishing, LLC, Honolulu, HI, 144 pp

Fitzpatrick BM, Harvey ES, Heyward AJ, Twiggs EJ, Colquhoun J (2012) Habitat specialization in tropical continental shelf demersal fish assemblages. PLoS One 7:e39634

Floeter S, Behrens M, Ferreira C, Paddack M, Horn M (2005) Geographical gradients of marine herbivorous fishes: patterns and processes. Mar Biol 147:1435–1447

Fricke HW, Schumacher H (1983) The depth limit of Red Sea stony corals: an ecophysiological problem (a deep diving survey by submersible). Mar Biol 4:163–194

Friedlander AM, Parrish JD (1998) Habitat characteristics affecting fish assemblages on a Hawaiian coral reef. J Exp Mar Bio Ecol 224:1–30

Fukunaga A, Kosaki RK, Wagner D, Kane C (2016) Structure of mesophotic reef fish assemblages in the Northwestern Hawaiian Islands. PLoS One 11:e0157861

Garcia-Sais JR (2010) Reef habitats and associated sessile-benthic and fish assemblages across a euphotic–mesophotic depth gradient in Isla Desecheo, Puerto Rico. Coral Reefs 29:277–288

Goreau TF, Goreau NI (1973) The ecology of Jamaican coral reefs. II. Geomorphology, zonation, and sedimentary phases. Bull Mar Sci 23:399–464

Gratwicke B, Speight M (2005) The relationship between fish species richness, abundance and habitat complexity in a range of shallow tropical marine habitats. J Fish Biol 66:650–667

Hay M (1981) Herbivory, algal distribution, and the maintenance of between-habitat diversity on a tropical fringing reef. Am Nat 118:520–540

Hay M (1984) Patterns of fish and urchin grazing on caribbean coral reefs: are previous results typical? Ecology 65:446–454

Hill J, Wilkinson C (2004) Methods for ecological monitoring of coral reefs. Australian Institute of Marine Science, Townsville, Australia, 117pp

Hinderstein LM, Marr JCA, Martinez FA, Dowgiallo MJ, Puglise KA, Pyle RL, Zawada DG, Appeldoorn R (2010) Theme section on mesophotic coral ecosystems: characterization, ecology, and management. Coral Reefs 29:247–251

Hixon M (2011) 60 years of coral reef fish ecology: past, present, future. Bull Mar Sci 87:727–765

Hixon MA, Beets JP (1989) Shelter characteristics and Caribbean fish assemblages: experiments with artificial reefs. Bull Mar Sci 44:666–680

Hixon MA, Beets JP (1993) Predation, prey refuges, and the structure of coral-reef fish assemblages. Ecol Monogr 63:77–101

Hobson E (1974) Feeding relationships of teleostean fishes on coral reefs in Kona, Hawaii. Fishery Bulletin 72:915–1031

Hoey AS, Brandl SJ, Bellwood DR (2013) Diet and cross-shelf distribution of rabbitfishes (f. Siganidae) on the northern Great Barrier Reef: implications for ecosystem function. Coral Reefs 32:973–984

Hughes TP, Bellwood DR, Folke CS, McCook LJ, Pandolfi JM (2007) No-take areas, herbivory and coral reef resilience. Trends Ecol Evol 22:1–3

Huisman JM, Abbott IA, Smith CM (2007) Hawaiian reef plants. University of Hawaii Sea Grant College Program, Honolulu, HI, 264 pp

Kahng SE, Maragos JE (2006) The deepest, zooxanthellate scleractinian corals in the world? Coral Reefs 25:254

Kahng SE, Garcia-Sais JR, Spalding HL, Brokovich E, Wagner D, Weil E, Hinderstein L, Toonen RJ (2010) Community ecology of mesophotic coral reef ecosystems. Coral Reefs 29:255–275

Kahng SE, Copus JM, Wagner D (2014) Recent advances in the ecology of mesophotic coral ecosystems (MCEs) *Curr Opin Environ Sustain* 7: 72-81.

Kane C, Kosaki R, Wagner D (2014) High levels of mesophotic reef fish endemism in the Northwestern Hawaiian Islands. Bull Mar Sci 90:1–12

Legendre P, Anderson MJ (1999) Distance-based redundancy analysis: testing multispecies responses in multifactorial ecological experiments. Ecol Monogr 69:1–24

Lesser MP, Slattery M, Leichter JJ (2009) Ecology of mesophotic coral reefs. J Exp Mar Bio Ecol 375:1–8

Liddell WD, Ohlhorst SL (1988) Substrata community patterns, 1–120 m, North Jamaica. Palaios 3:413–423

Liddell WD, Avery WE, Ohlhurst SL (1997) Patterns of benthic community structure, 10–250 m, the Bahamas. Proc 8th Int Coral Reef Symp 1:437–442

Lindfield SJ, Harvey ES, Halford AR, McIlwain JL (2016) Mesophotic depths as refuge areas for fishery-targeted species on coral reefs. Coral Reefs 35:125–137

Maragos JE, Jokiel P (1986) Reef corals of Johnston Atoll – one of the World's most isolated reefs. Coral Reefs 4:141–150

McGehee M (1994) Correspondence between assemblages of coral reef fishes and gradients of water motion, depth, and substrate size off Puerto Rico. Mar Ecol Prog Ser 105:243–255

Menza C, Kendall M, Rogers C, Miller J (2007) A deep reef in deep trouble. Cont Shelf Res 27:2224–2230

Munday PL (2000) Interactions between habitat use and patterns of abundance in coral-dwelling fishes. Environ Biol Fishes 58:355–369

Parrish FA, Boland RC (2004) Habitat and reef-fish assemblages of banks in the Northwestern Hawaiian Islands. Mar Biol 144:1065–1073

Pyle RL (2000) Assessing undiscovered fish biodiversity on deep coral reefs using advanced self-contained diving technology. Marine Technology Society Journal 34:82–91

Pyle RL, Boland R, Bolick H, Bowen BW, Bradley CJ, Kane C, Kosaki RK, Langston R, Longenecker K, Montgomery A, Parrish FA, Popp BN, Rooney J, Smith CM, Wagner D, Spalding HL (2016) A comprehensive investigation of mesophotic coral ecosystems in the Hawaiian Archipelago. PeerJ 4:e2475

Randall J (2007) Reef and shore fishes of the Hawaiian Islands. University of Hawaii Sea Grant College Program, Honolulu, HI

Reaka M, Rodgers P, Kudla A (2008) Patterns of biodiversity and endemism on Indo-West Pacific coral reefs. Proc Natl Acad Sci U S A 105:11474–11481

Rodriguez-Jerez Z (2004) Zooplankton communities of Isla Desecheo and adjacent waters of Mona Passage. MS Thesis, University of Puerto Rico, San Juan. 85pp

Rosa MR, Alves AC, Medeiros DV, Coni EOC, Ferreira CM, Ferreira BP, Rosa R, Souza R, Amado-Filho GH, de Moura RL, Thompson FL, Sumida PYG, Francini-Filho RB (2016) Mesophotic reef fish assemblages of the remote St. Peter and St. Paul's Archipelago, Mid-Atlantic Ridge, Brazil. Coral Reefs 35:113–123

Russ GR (2003) Grazer biomass correlates more strongly with production than with biomass of algal turfs on a coral reef. Coral Reefs 22:63–67

Sale P (1991) Ecology of coral reef fishes. In: Sale PF (ed) The ecology of fishes on coral reefs. Academic Press, San Diego, CA, pp 1–11

Sandin SA, Smith JE, Demartini EE, Dinsdale EA, Donner SD, Friedlander AM, Konotchick T, Malay M, Maragos JE, Obura D, Pantos O, Paulay G, Richie M, Rohwer F, Schroeder RE, Walsh S, Jackson JBC, Knowlton N, Sala E (2008) Baselines and degradation of coral reefs in the Northern Line Islands. PLoS One 3:e1548

Schultz AL, Malcolm HA, Bucher DJ, Linklater M, Smith SD (2014) Depth and medium-scale spatial processes influence fish assemblage structure of unconsolidated habitats in a subtropical marine park. PLoS One 9:e96798

Slattery M, Lesser MP, Brazeau D, Stokes MD, Leichter JJ (2011) Connectivity and stability of mesophotic coral reefs. J Exp Mar Bio Ecol 408:32–41

Thresher R, Colin P (1986) Trophic structure, diversity and abundance of fishes of the deep reef (30–300 m) at Enewetak, Marshall Islands. Bull Mar Sci 38:253–272

Tootell JS, Steele MA (2016) Distribution, behavior, and condition of herbivorous fishes on coral reefs track algal resources. Oecologia 181:13–24

van Oppen MJH, Bongaerts P, Underwood JN, Peplow LM, Cooper TF (2011) The role of deep reefs in shallow reef recovery: an assessment of vertical connectivity in a brooding coral from west and east Australia. Mol Ecol 20:1647–1660

Williams D (1991) Patterns and processes in the distribution of coral reef fishes. In: Sale P (ed.) Ecology of coral reef fishes. Academic Press, Cambridge, MA, pp 437–474

Wilson SK, Depczynski M, Fisher R, Holmes TH, O'Leary RA, Tinkler P (2010) Habitat associations of juvenile fish at Ningaloo Reef, Western Australia: the importance of coral and algae. PLoS One 6:e15185

1.8 TABLES

Table 1.1 Number of species observed from one to six depths (3, 10, 20, 30, 40, and/or 50 m), indicating the proportion of depth specialists (found at only one or two contiguous depths) to depth generalists (found at four or more depths)

# Depths	# Species	% of Total
1	31	21%
2	19	13%
3	20	13%
4	28	19%
5	22	15%
6	29	19%

Table 1.2 Percentage of variation explained in a distance-based multivariate linear model of

reef-fish communities

Variable	P	% Variation Explained
Depth	0.001	16.89
Herbivore	0.001	10.31
Live coral	0.001	10.23
Sand/Sediment	0.001	7.67
Detritivore	0.001	7.01
Corallivore	0.001	6.92
Rubble	0.001	6.24
Basalt	0.001	5.79
Macroalgae	0.001	5.43
Compressa bed	0.001	4.45
Zooplanktivore	0.001	3.7
Site	0.002	2.28
Invertivore	0.001	2.17
Turf algae	0.001	2.14
Carbonate	0.004	1.97
Boulder	0.006	1.65
Pebble/Cobble	0.012	1.45
Omnivore	0.029	1.31
Piscivore	0.042	1.16
Pavement	0.454	0.67

1.9 Figures
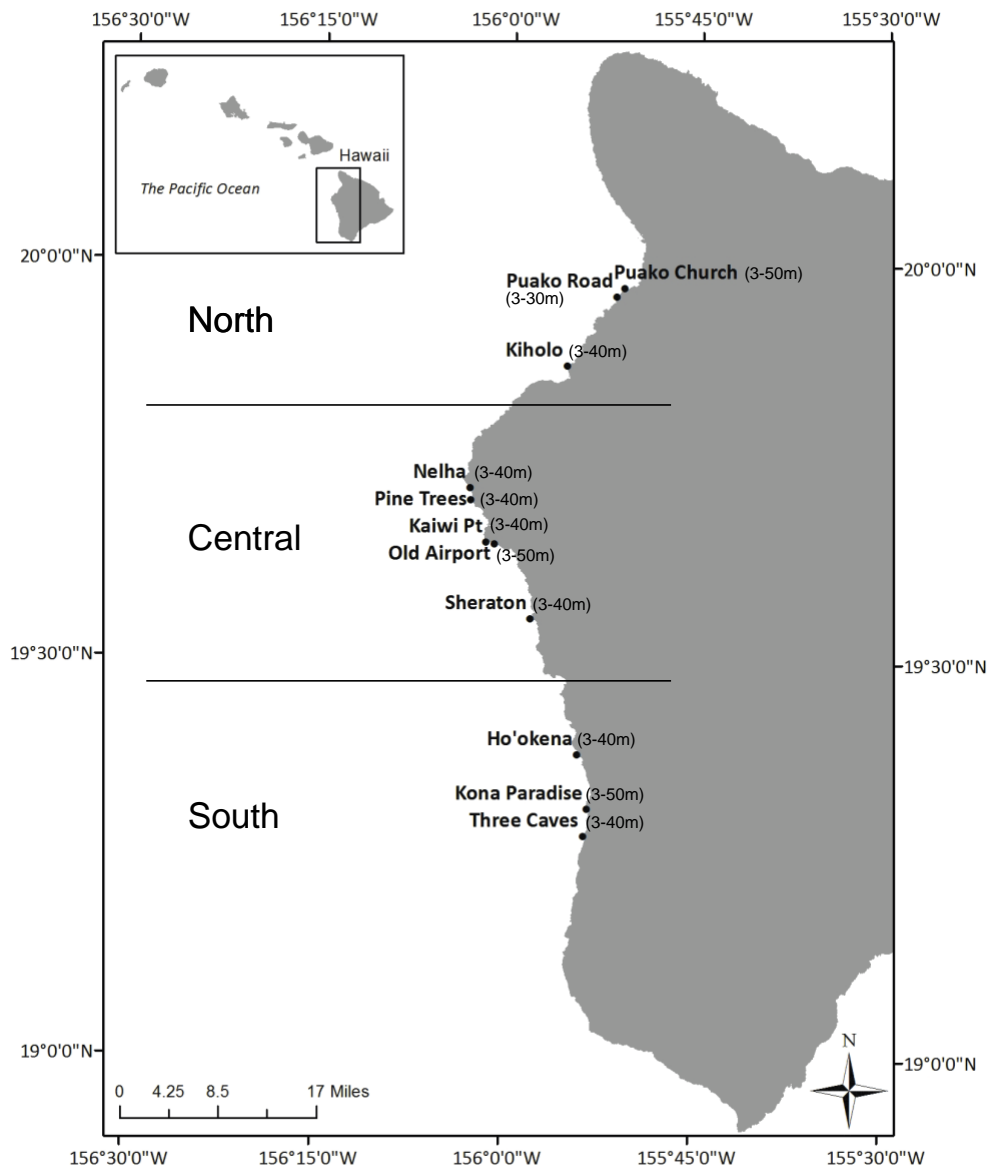
Fig. 1.1 Location of study sites in West Hawaii with depth ranges in parentheses.

a) b)
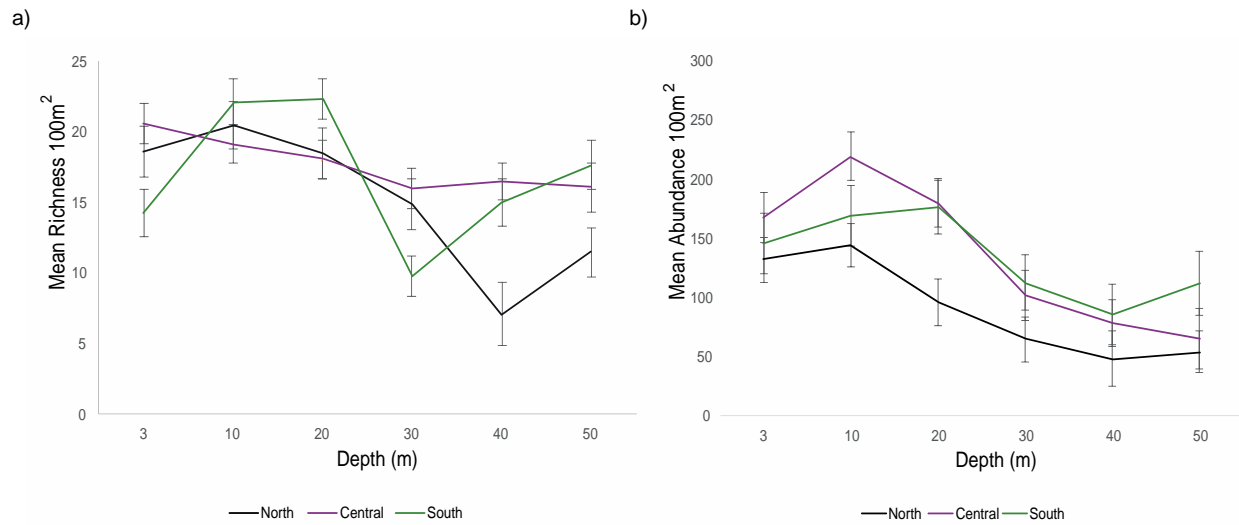
Fig. 1.2 a) Mean reef-fish species richness per $100m^2$ by depth. **b)** Mean reef-fish abundance per $100m^2$ by depth. *Error bars* are standard errors.
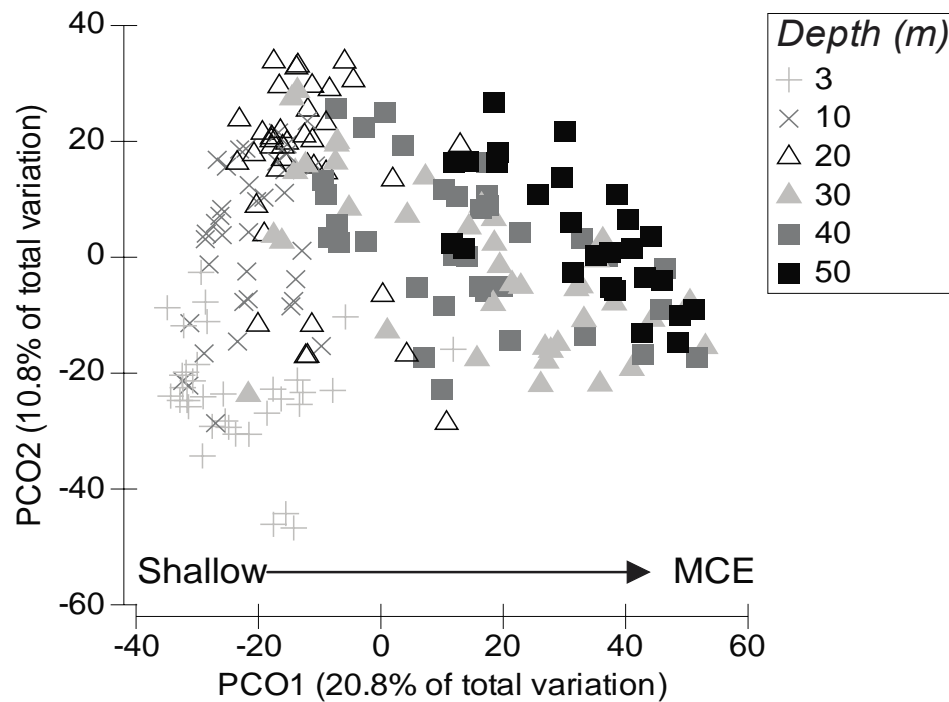
Fig. 1.3 Principal coordinates analysis ordination of square-root-transformed fish community structure at different depths using Bray–Curtis similarity resemblance matrix. *Axes* describe percentage of variation in terms of the total fish community structure.
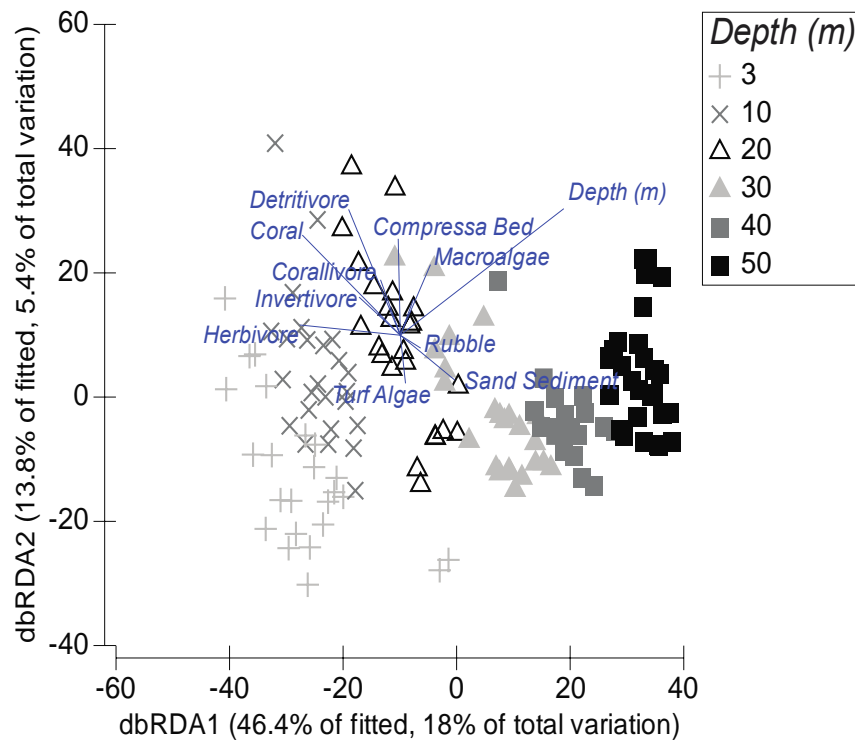
Fig. 1.4 Distance-based redundancy analysis ordination of reef-fish abundance using Bray–Curtis similarity resemblance matrix. *Axes* describe percentage of variation in terms of the total fish community structure. *Vectors* represent strength of variable in model.
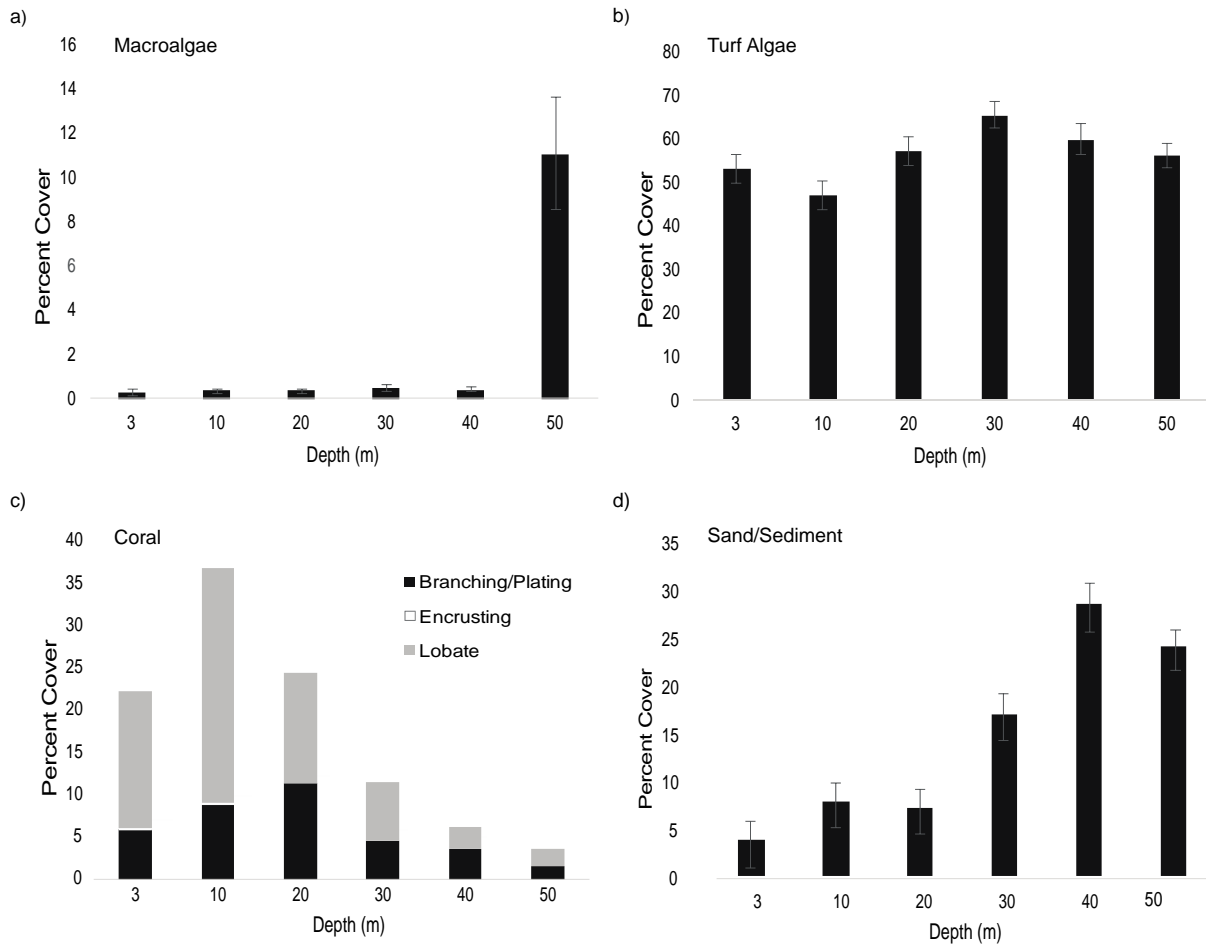
Fig. 1.5 Mean percentage cover of **a** macroalgae, **b** turf algae, **c** coral morphologies, **d** sand/sediment along a depth gradient (in m). *Error bars* are standard errors.
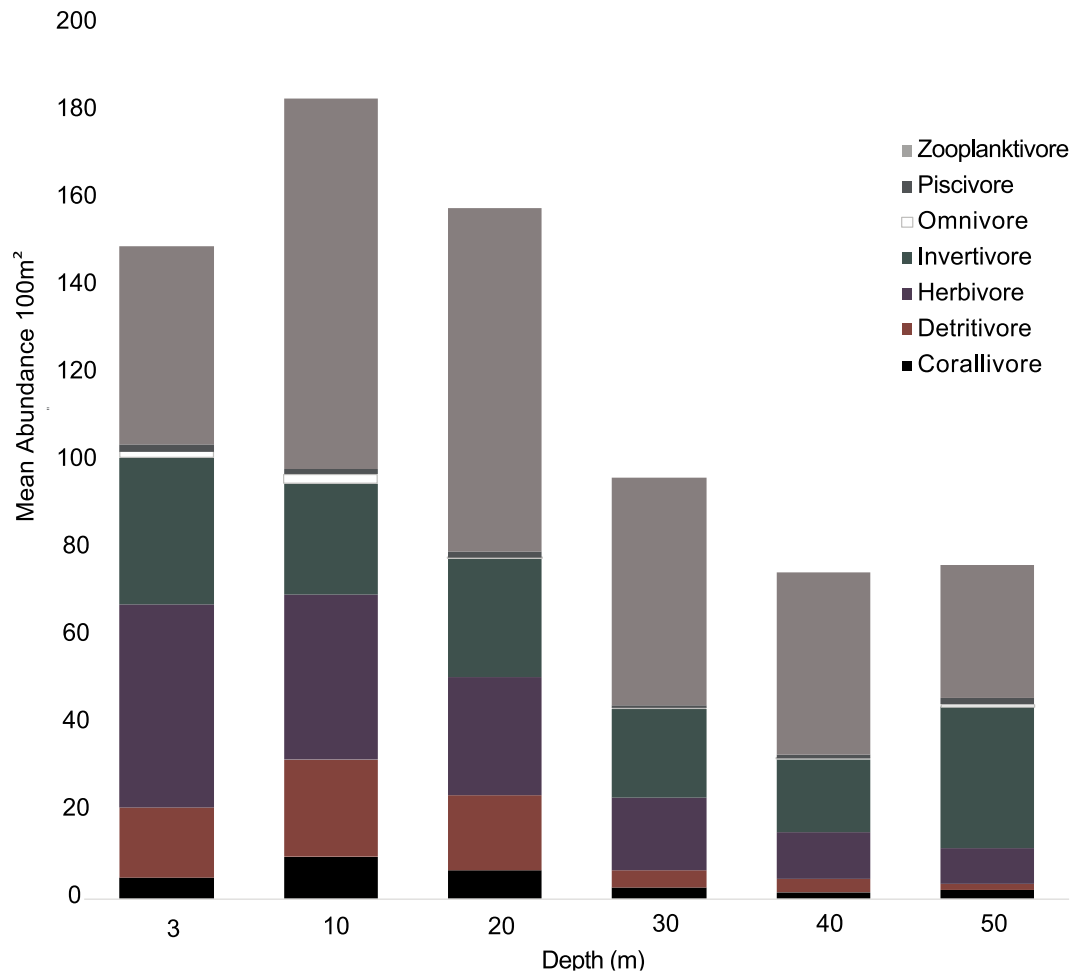
Fig. 6 Mean abundance ($100\ m^{-2}$) of reef-fish trophic groups by depth (m).
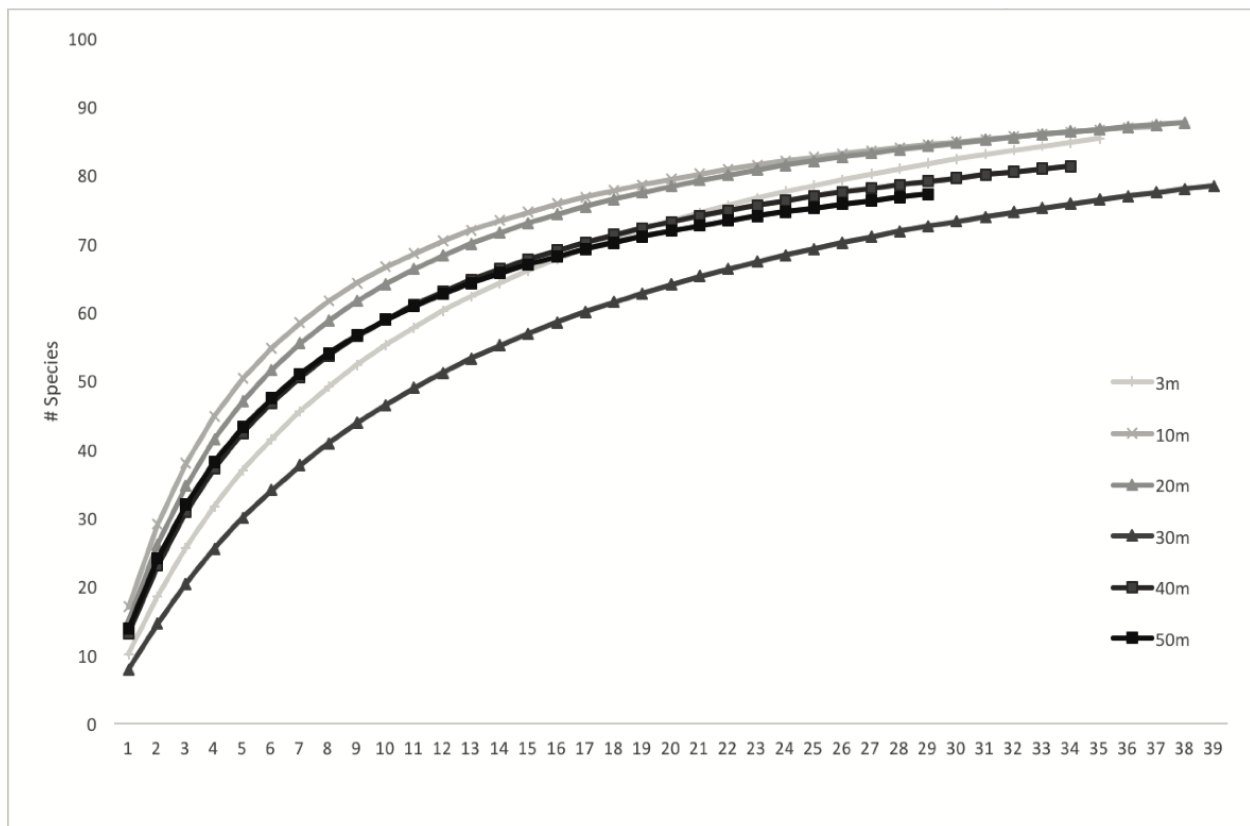
Fig. S1.1 Species accumulation curves for the six depths surveyed.

CHAPTER TWO

INDIRECT EFFECTS OF PREDATION CONSTRAIN HERBIVOROUS FISH

DISTRIBUTIONS AT MESOPHOTIC DEPTHS

INDIRECT EFFECTS OF PREDATION CONSTRAIN HERBIVOROUS FISH DISTRIBUTIONS AT MESOPHOTIC DEPTHS

2.1 Abstract

Herbivorous fishes play an integral role in preventing macroalgae from displacing corals, yet these fishes are reported to decline with depth despite the presence of coral-dominated habitats to >80m. Mechanisms underlying this decline remain largely untested due to time and safety constraints of diving at mesophotic reef depths (>30m). We investigated the relationship between herbivorous fish species and a suite of bottom-up and top-down parameters along a depth gradient from 3-50m. We examined turf, macroalgal and coral cover, nutritional quality and palatability, temperature, light, and predator density as potential mechanisms limiting the distribution of herbivorous fishes with depth. Mesophotic reefs were not limited by food resources as mesophotic algae had similar nutritional content, species assemblages, and appeared highly palatable from algal choice experiments. Results of in-situ and long-term temperature recordings suggest temperature does not restrict herbivorous fish distributions at upper mesophotic depths. Multivariate redundancy analyses reveal the combination of increased habitat patchiness and reduced light levels best explain herbivorous fish distributions with depth, indicating the reduction in herbivorous fishes at upper mesophotic depths is likely the result of non-consumptive predation effects. These results suggest changes in herbivorous fish populations with depth are likely the result of top-down effects and behavioral choices rather than abiotic constraints or resource limitation.

2.2 Introduction

Herbivorous fishes are an integral component of coral reef ecosystems and play crucial roles in maintaining coral-dominated states (Hay 1997; Fox and Bellwood 2007; Adam et al. 2015; Steneck et al. 2017). These fishes are one of the most studied trophic groups in tropical marine ecosystems, yet mechanisms underlying their spatial distributions are still unclear (Hay 1984; Russ 2003; Lapointe et al. 2004; Brokovich et al. 2008; Carlson et al. 2017). Limitations to spatial distribution of herbivorous fishes in shallow waters are largely attributed to bottom-up processes. Temperature has been shown to limit herbivorous fish distributions across latitudinal gradients within shallow tropical systems (Ferreira et al. 2004; Witman et al. 2004; Floeter et al. 2005; Smith 2008). Temperature differences as little as 8°C can inhibit metabolic efficiency in herbivorous fishes, thus constraining their distributions regardless of algal resources (Floeter et al. 2005; Smith 2008).

At finer spatial scales, herbivorous fish distribution is largely attributed to bottom-up effects resulting from resource availability (Carpenter 1988; Russ and McCook 1999; Russ 2003; Cvitanovic and Hoey 2010). Recent studies reveal herbivorous fishes cue specifically to algal quality and productivity rather than simply algal prevalence (Russ 2003; Fox and Bellwood 2007; Adam et al. 2015; Tootell and Steele 2016). Algal quality/productivity is limited by the availability of nutrients (largely nitrogen and phosphorus) and herbivorous fishes have been documented to prefer nitrogen-rich algae (Lapointe et al. 2004; Adam et al. 2015).

Resource availability and productivity may also interact with predation risk to influence the spatial distribution of herbivorous fishes. While algal resources influence centers of distributions, predation may influence the margins of distributions at smaller spatial scales. Top-down effects influencing distributions of prey organisms have been thoroughly demonstrated in

rocky intertidal systems (Paine 1980; Menge 1995) yet has received less attention in structuring coral reef fish distributions. The role of predation (direct or indirect) in structuring herbivorous fish communities appears variable. Davis et al. (2017a, 2017b) has shown predation influences behavior but not space use in parrotfishes, while others contend predation risk shapes behavior and distribution (Catano et al. 2016, 2017; Gil et al. 2017). Catano et al. (2016) further found predator effects were size-dependent, with smaller fishes less affected by predation risk than larger-bodied fishes.

One major caveat regarding spatial distributions of herbivores is that most studies have encompassed only a small portion of the total reefscape. Nearly all studies of herbivorous fishes are conducted at less than 15m depth, with a large proportion at 5m depth or less. A number of studies have explored herbivorous fish distributions across a wider depth range (Hay 1981a; Feitoza et al. 2005; Brokovich et al. 2008, 2010a; Nemeth and Appeldoorn 2009; Garcia-Sais 2010; Bejarano et al. 2014; Fukunaga et al. 2016; Kane and Tissot 2017). These studies, which span the Red Sea, the Caribbean Sea, and the Pacific Ocean, conclude that herbivorous fish abundance and biomass significantly decreases with depth. The past decade has seen a proliferation in documentation of MCEs (mesophotic coral ecosystems; >30m) and specifically MCE fish communities, but research into mechanisms underlying observed patterns is still in its infancy. Two studies to date have evaluated mechanisms underscoring reduced herbivore abundances at depth. Working in Belize, Hay (1981) found algae from deep reefs (20-30m) yielded increased chemical defenses compared to shallow algae. Brokovich et al. (2010) found reduced algal productivity and reduced grazing pressure with depth in Israel.

Mesophotic reefs are often characterized by cooler, more nutrient rich water (Hinderstein et al. 2010; Kahng et al. 2010; Smith et al. 2013) which may inhibit herbivorous fishes from

inhabiting these habitats. It is purported that nutrient levels increase at MCE depths due to the influx of colder, more nutrient rich waters (Kahng et al. 2010). Conversely, reductions in light with depth are inferred to reduce algal productivity and inhibit visual acuity (Vandenhoek et al. 1978; Nemeth and Appeldoorn 2009; Brokovich et al. 2010a, 2010b). It is currently unclear how the interplay between nutrient and light levels affect algal productivity. Additionally, studies have shown deep algae to be more highly chemically defended and thus less palatable to herbivorous fishes (Hay 1981b; Gutow et al. 2014). Seaweed palatability is determined by a combination of structural (tissue toughness, thickness) and chemical mechanisms (Gutow et al. 2014). The idea that deep algae have enhanced chemical defenses was initially proposed by Hay in the 1980s, but this hypothesis has received scant attention as a mechanism underlying herbivorous fish distributions with depth in the following decades. Recent work by Papastamatiou et al. (2015) reveal the first evidence of sharks and trevally movement and foraging between shallow and mesophotic depths, indicating that mobile predators are utilizing mesophotic reefs as foraging grounds.

Based on our understanding of shallow-water mechanisms controlling herbivorous fish abundance and possible limitations with depth, we conducted a suite of observational and experimental studies on the leeward coast of Hawaii to evaluate potential bottom-up and top-down mechanisms affecting the depth distribution of herbivorous fishes. Specifically, we sought to test how herbivorous fish distributions between shallow and mesophotic depths were influenced by 1) bottom-up effects resulting from thermal barriers, habitat, resource composition/nutritional value; and 2) predation effects resulting from direct and indirect predation pressures.

2.3 Methods

2.3.1 Study sites

Hawaii Island has a steeply sloping bathymetry and extensive coral reefs that spread continuously from shore to depths of approximately 50 m. Beyond 50m, large sand flats occur to 70 m depth or more, with reef often re-emerging and patchily continuing to depths currently undetermined. Eleven sites were selected along West Hawaii's coastline in areas where continuous coral-reef habitat occurs from shallow waters to at least 30 m (Fig. 1). Due to the steep slope, the 50 m depths surveyed were often within 100–200 m of the shoreline, thus physical distance between the shallowest and deepest surveys were between 50 and 200 m from shore.

2.3.2 Fish and benthic sampling design

A stratified sampling design was used to explore changes in fish abundance and trophic structure with depth. Concordant depth and bathymetric relief among sites was sought to maintain consistency and reduce potential survey bias. Visual surveys of reef fish and benthic substrates were conducted on SCUBA at approximately 10-m depth intervals during June–August in 2013–2015. Due to variability of habitat below 30 m, three sites were sampled to 50 m, seven to 40 m and one to 30 m. In each site, transects were laid parallel to shore at 3 m, 10 m and then in 10-m depth intervals until the deepest extent of the reef. A minimum of three transects were surveyed at each depth contour at each site. Transects at each depth contour were spaced a minimum of 5 m apart. This resulted in an unequal survey design with 35 surveys at 3 m, 37 surveys at 10 m, 38 surveys at 20 m, 38 surveys at 30 m, 34 surveys at 40 m, and 30 surveys at 50 m (for complete details see Kane and Tissot 2017). Visual fish surveys were conducted at each depth (3–50 m) using a 25 m x 4 m belt transect (Hill and Wilkinson 2004). Belt length, width and level of replication per depth (minimum n = 3) were dictated by technical

51

constraints resulting from the short bottom times allowed from using SCUBA at 50 m depths (Brokovich et al. 2008; Sandin et al. 2008). Within each fish belt transect, all fishes were identified to species (following Randall 2007) and tallied individually. Benthic survey data were collected at two of the replicate transects within each depth by photographing a 0.25-m^2 quadrat every meter (n = 25 per transect) using an underwater camera equipped with a PVC photoquadrat framer and dual lighting system.

All fishes encountered in surveys were assigned trophic designations according to gut content analyses published on Hawaiian fish species in Hobson (1974) and Randall (2007). Species lists and assignments to trophic guilds were corroborated with the database of the National Oceanic and Atmospheric Administration's Pacific Islands Fisheries Science Center for Hawaii. All herbivorous and piscivorous fish species were then extracted from the dataset for use in this study. Herbivorous fish were further assigned family/sub-family designations, and piscivorous fish species were assigned as site-attached (benthic-associated or ambush predators) or mobile (apex or wide-ranging predators) for additional analyses.

Benthic composition was examined to investigate correlations between herbivorous fish abundance and benthic taxa across depths surveyed. Benthic photoquadrat samples were analyzed using CoralNet benthic image analysis (Beijbom et al. 2015). Fifty random points were placed within each photograph and the biota or substratum directly underneath each point was identified to the lowest taxonomic classification possible (see Kane and Tissot 2017). Taxonomic identifications were pooled to create descriptive habitat categories which included turf algae, macroalgae, sand, and live coral cover. These broad categories were selected as live coral cover, turf and macroalgal cover are strongly correlated with reef-fish community composition at shallow depths (Friedlander and Parrish 1998; Arreola-Robles and Elorduy-Garay 2002;

Donaldson 2002; Wilson et al. 2010; Hoey et al. 2013; Evans et al. 2014). Categories were summed by photoquadrat and averaged (n = 25) to obtain one cover estimate per transect to correspond with fish transects.

2.3.3 Temperature and light sampling

Temperature and irradiance estimates were collected at depths from 3-40m. In 2013, divers collected temperature and irradiance levels at each survey site and survey depths from 3-40m (50m depths not recorded as sensors malfunctioned due to exceeding max depth ratings). Four identical HOBO Pendant Temperature/Light data loggers (UA-002-64; Onset Computer Corporation) were affixed to an aluminum plate with each sensor spaced linearly at 10cm intervals. Loggers were programmed to begin recording at an interval of 1 measurement every 10 seconds upon submersion.

At the beginning of surveys, a diver would position the plate on top of the substrate with the face of the sensors facing directly upward. The sensor panel would remain in place for a minimum of eight minutes before moving to the next depth interval. Divers conducted detailed recordings of the depth (3, 10, 20, 30, or 40m) and time of each placement. Data loggers were then downloaded and the last five minutes of each depth recording were extracted to ensure sufficient time to calibrate at each depth. Temperature and irradiance levels were averaged for each sensor at each depth within each site, then the four sensors were averaged together at each depth among sites.

We also deployed a series of HOBO Pendant Temperature/Light data loggers for long term data recording at one site (Kona Paradise, southern region) in 2013-2014. One data logger was placed at each of five depths: 3, 10, 20, 30, and 40m. Data loggers were set to record measurements at 25 minute intervals and were deployed for a period of 6 months. Loggers were

inspected, cleaned, and had data extracted every two weeks for the six-month period. Only temperature recordings were used for these loggers as the shallow loggers fouled too quickly to obtain accurate long-term irradiance recordings. After 3 months, a large swell dislodged the 10m sensor so we have excluded this depth from our analysis.

2.3.4 Algal identification

Initial estimates of algal composition and prevalence were compiled via photoquadrat analyses. As certain algal groups could not be easily identified via photographs (turfs, small algae) we collected voucher samples from one site to further classify algae not readily identifiable in photographs. Three representative rubble/rock matrices were collected from 10m (shallow; 7.0 x 5.5 x 4.5 cm) and 40m (deep; 7.5 x 5.0 x 4.5 cm) depths via SCUBA on 4 August 2016 at Puako. Each sample was individually frozen in a plastic bag after collection. Rocks were unthawed on 27 May 2017, photographed, and all visible algae were removed under a dissecting scope and identified to the lowest possible taxonomic level using a microscope and reference books (Abbott 1999, Abbott and Huisman 2004, Huisman et al. 2007). Algal nomenclature was updated using Algaebase (Guiry and Guiry 2017). Each specimen was processed according to Tsuda and Abbott (1985) and given a collection number (HS) from the herbarium of H. L. Spalding (HS452 – HS479).

2.3.5 Algal quality

To test nutrient contents of turf algae between shallow and mesophotic depths we investigated C:N ratios. Thirty samples of turf algae were collected by hand from benthic substrate at 10m and 40m depths at Puako in August 2016. Samples were placed in zip-lock bags and transported in sea water to the lab, rinsed, wet-weighted and then dried at 60°C for 48 hours. Samples were then ground with mortar and pestle until a fine powder was produced. C

and N contents were determined using a Flash 2000 NC soil analyzer (CE Elantech) against an

aspartic acid, $C_3H_7NO_4$ organic analytical standard (OAS: Elemental C values 36.09, Elemental

N values 10.52).

We assessed the palatability of turf algae from mesophotic depths by conducting an in-

situ algal choice experiment (cf. Hixon 1980, Hay 1981a). Three sand/rubble patches located

adjacent to large coral reef tracts were selected at 10m depth at Old Airport. In each of these

patches, ten pairs of experimental plots were set up (n=30 total). Each plot consisted of a large

cinder block with a 60cm PVC pipe (1 in diameter) attached to it. A GoPro camera was affixed

to the top of the PVC pipe to record fish behaviors without distractions by divers. In front of

each camera we created a paired design consisting of one control and one experimental plot set

1m apart from each other. The control plots consisted of a 30cm x 30cm area filled with turf

algae-covered rubble collected from the adjacent reef area at 10m depth (shallow treatment).

Experimental plots consisted of the same sized area with turf algae-covered rubble collected

from 40m depth and transported up to the experimental site (deep treatment). Once rubble plots

were set up, the camera was turned on and recorded for approximately 30 minutes. To control

for discovery time, we began analysis of each treatment video at 5 minutes and analyzed fish

activity for 20 minutes. Within the 20-minute time frame, we identified every fish visible to

species and recorded total visitation time by each fish, the amount of time each fish investigated

and/or grazed on algae, and the number of bites taken during each foraging bout.

2.3.6 Behavioral observations

We conducted observations of focal individuals of herbivorous fishes to examine

behavioral patterns in both shallow (<10m) and mesophotic (>30m) coral reef environments.

We selected *Zebrasoma flavescens* (Acanthuridae) as a representative species as it was the most

commonly observed fish in both shallow and mesophotic depths during transect surveys. We

conducted focal observations of adult *Z. flavescens* at two sites, Puako (northern region) and Old

Airport (central region). Within each site we selected 20 adult *Z. flavescens* at each depth

treatment; shallow observations included fishes found at 3-10m depths, while deep observations

included fishes from 30-50m depths. Focal observations were conducted by attaching a GoPro

camera to a 2m PVC pipe. At each depth, SCUBA divers selected one fish and cautiously filmed

them for a period of five minutes from a distance of at least 5m. If the focal fish was startled by

the diver or went out of view of the diver at any time during the 5-minute session, filming was

aborted and another fish was selected. Extensive communication between divers at each depth,

along with detailed notes collected on visual identities of fishes during video analysis were

employed to ensure no pseudo-replication occurred by filming the same fish multiple times.

Behaviors were binned into six categories; foraging, swimming, aggressor, victim,

courtship, or other. Foraging behaviors were classified as fish actively grazing on substrate.

Swimming was classified as fish actively moving. Agonistic behavior was divided into two

categories, an aggressive (focal fish actively chasing another fish) or submissive (focal fish

actively being chased by another fish). Courtship behaviors included spawning behaviors and

territorial patrolling (swimming >2m above substrate in a repetitive pattern). The "other"

category included any additional behaviors not previously outlined, such as time spent at

cleaning stations.

2.3.7 Data analysis

Sites were grouped into three geographic regions (North, Central, South Kohala-Kona

coast) to ensure adequate sampling effort for analyses (Fig. 1). To ascertain general trends in the

distribution of herbivorous and piscivorous fish families across depths, we calculated generalized

linear models (GLM) based on a Poisson distribution (two factors: depth and region). Focal observations of fishes were also compared using two-factor GLMs based on Gaussian distributions (factors: treatment and site). Analysis of environmental predictors for herbivorous fish distributions were conducted using a multivariate distance-based linear model with Best selection approach (distLM; Primer v. 6.1.13, Primer-E+). Prior to analysis, all variables were tested for skewness and multi-collinearity. Macroalgal and sand cover estimates were log-transformed to correct for skewed distributions. Multi-collinearity tests revealed no two variables showed correlations greater than 0.65 so all variables were retained for analysis.

Percent carbon, nitrogen, and C:N ratios were analyzed using paired t-tests. To analyze algal choice experiments we calculated two metrics: total foraging time (mean number of seconds each fish spent grazing) and grazing intensity (mean number of bites/fish/visit). Both total foraging time and grazing intensity were analyzed using GLMs based on Poisson distributions to account for non-normal distributions. All statistical analyses were conducted using JMP statistical software (JMP 12.1.0, SAS Institute Inc.).

2.4 Results

2.4.1 Herbivore distributions

A total of 5,253 fish from 33 herbivorous species were counted during transect surveys (ESM 1). The herbivore assemblage was dominated largely by acanthurids (17 species), which encompassed 80% of all fishes encountered. Labrid fishes (subfamily Scarinae) and pomacanthid fishes were the second-most common families (labrid: 5 spp., pomacanthid: 2 spp.) and each comprised approximately 7% of fish surveyed. Herbivorous fishes as a group declined with depth (Fig. 2) but this pattern was largely a result of the decline in acanthurid fishes. Acanthurid fishes differed significantly across both depth and regions sampled (GLM: depth

$\chi^2_{5,212} = 1792.28$, P ≤ 0.001; region $\chi^2_{2,212} = 35.42$, P ≤ 0.001; depth x region $\chi^2_{10,212} = 262.11$, P ≤ 0.001; Fig. 3a). While all regions declined with depth, a significant interaction between depth and region occurred as a result of higher abundances of acanthurids at one site in the southern region (Kona Paradise). Scarinae differed significantly across both depths and regions sampled (GLM: depth $\chi^2_{5,212} = 111.67$, P ≤ 0.001; region $\chi^2_{2,212} = 16.31$, P = 0.003; depth x region $\chi^2_{10,212} = 104.4$, P ≤ 0.001; Fig. 3b), but in the northern and central regions abundance peaked at 20-30m depths while in the southern region abundances were extremely low across all depths. Pomacanthid species exhibited significant patterns across depth and depth x region interaction, but no significant differences among regions sampled (GLM: depth $\chi^2_{5,212} = 127.95$, P ≤ 0.001; region $\chi^2_{2,212} = 0.0004$, P =0.99; depth x region $\chi^2_{10,212} = 54.83$, P ≤ 0.001; Fig. 3c). Pomacanthids also exhibited higher abundances at 20-30m depths. The remaining herbivorous families were pooled due to low overall abundances, and were also found to decrease significantly with depth, but did not differ significantly between regions (GLM: depth $\chi^2_{5,212} = 247.7$, P ≤ 0.001; region $\chi^2_{2,212} = 5.89$, P =0.052; depth x region $\chi^2_{10,212} = 38.64$, P ≤ 0.001; Fig. 3d). While herbivorous fishes in all regions declined with depth, a significant interaction between region and depth occurred as a result of higher abundances of herbivores at the northern sites.

Herbivorous fish densities were compared against a suite of biotic and abiotic predictor variables. DistLM analysis indicated significant relationships between herbivorous fish community structure and 5 of 7 variables tested (macroalgae and temperature not statistically significant; Table 1). The model that best explains herbivorous fish distributions (lowest AIC values) incorporates a combination of light, coral cover and sand cover variables and accounts for approximately 14% of the total variation in herbivorous fish communities (Table 2).

2.4.2 Predator distributions

Piscivorous fishes were mostly comprised of site-attached species (17 spp.) while mobile piscivores were rare and comprised of few species (6 spp.; Fig. 4a). Piscivores differed significantly between depths and between depth x region interactions, but were not significantly different between regions (Fig. 4b; GLM: depth $\chi^2_{5,212}$ = 18.96, P = 0.002; region $\chi^2_{2,212}$ = 1.81, P =0.41; depth x region $\chi^2_{10,212}$ = 37.93, P ≤ 0.001). Piscivores generally decreased from shallow to mesophotic depths with the exception of one site (Puako) at 50m in the Central region. This site contained high abundances of the introduced serranid *Cephalopholis argus* (peacock grouper) and muraenid (moray eel) species compared to sites in the northern and southern regions.

2.4.3 Temperature and Light sampling

Water temperatures averaged 28.6°C at 3m to 26.6°C at 40m. Temperature was highly variable between sites at 3m depths, but relatively stable at 10-40m depths. Light levels followed similar patterns as temperature with much more variability in irradiance at 3m depths and more stable irradiance at deeper depths. Irradiance decreased sharply with depth; 10m depths averaged 39.5% of surface light available at 3m, 20m depths 22%, 30m depths 12.8%, and 40m depths averaged 7.8% of light levels at 3m.

Long-term temperature measurements were recorded at one site (Kona Paradise) for a period of six months. During summer months, temperatures were consistently stratified between 3, 20, and 30-40m depths (Fig. 5). Stratification collapsed by late fall and from November-January temperatures were uniform from 3-40m depths.

2.4.4 Algal identification

The shallow and deep algal communities had a low abundance of macroalgae, with turf algae and brown dictyotalean blades being most visually apparent (ESM 2). Within voucher

samples, most macroalgae were diminutive (< 1 cm long) and sterile, making identifications to the species level difficult without molecular techniques.

Shallow algal communities had the highest cover of visually apparent macroalgae and species diversity (35 species/genera), while the deep communities were more depauperate with lower diversity (21 species/genera; ESM 2). *Tolypiocladia glomerata* was the most abundant turf species at shallow depths. *Padina* sp. was mostly present as the '*Vaughaniella*' stage (creeping rhizomes) in both shallow and deep samples. Overlap in species composition was high, with two-thirds of the deep species also found in shallow samples, including three abundant brown macroalgae (*Dictyota* sp., *Padina* sp., and *Stypopodium flabelliforme*; ESM 2). These species of brown algae belong to the Dictyotaceae, a family known to have antiherbivory compounds and inducible antiherbivory defenses (Hay and Steinberg 1992; Macaya and Thiel 2008). All of the deep genera have also been described from shallow water (Abbott 1999; Abbott and Huisman 2004), although species-level identifications would be necessary to evaluate possible overlap.

2.4.5 Algal quality

We analyzed carbon and nitrogen inputs of turf algae collected from 10m and 40m depths and found no significant differences between depths for carbon ($t_{1,57}=0.15$, P=0.88), nitrogen ($t_{1,57}=0.65$, P=0.52), or C:N ratios ($t_{1,57}=0.24$, P=0.81). Mean levels of %C were 13.47% (+/- 0.28 SE) at 10m vs. 13.53% (+/- 0.28 SE) at 40m. Nitrogen composition averaged 0.67% (+/- 0.03 SE) at 10m while 0.7% (+/- 0.03 SE) at 40m. Mean C:N ratios at 10m were 21.19 (+/- 1.07 SE) at 10m and 20.83 (+/- 1.05 SE) at 40m.

During the algal choice experiment, both grazing time (Fig. 6a; $\chi^2_{1,58}= 11393.99$, P $\leq$ 0.001) and grazing intensity (Fig. 6b; $\chi^2_{1,58}= 81.14$, P $\leq$ 0.001) were significantly greater on algae collected from 40m depths, indicating that algae at MCE depths is both palatable and preferred

by herbivorous fishes when translocated to shallow depths. Grazing was dominated largely by small-bodied surgeonfish (*Zebrasoma flavescens, Acanthurus nigrofuscus, Ctenochaetus strigosus*), which accounted for 87% of the grazing time in shallow treatments, and 65% of the grazing time in deep treatments. Large-bodied surgeonfish (*Naso lituratus, Acanthurus olivaceous, Naso unicornis*) accounted for 5% of shallow grazing time and 15% of deep grazing time, while parrotfish (*Calotomus carolinus, Calotomus zonarchus, Chlorurus spilosoma, Scarus psittacus*) accounted for 7% of shallow grazing time and 13% of deep grazing time.

2.4.6 Behavioral observations

Zebrasoma flavescens exhibited differences in foraging (Fig. 7; GLM: depth $t_{1,40} = 0.49$, P $=0.62$; site $t_{1,40} = 2.64$, P $=0.009$; depth x site $t_{1,40} = 1.56$, P $=0.12$), swimming (Fig. 7; GLM: depth $t_{1,40} = 0.76$, P $=0.45$; site $t_{1,40} = 2.32$, P $=0.02$; depth x site $t_{1,40} = 1.85$, P $=0.06$) and aggressive behaviors (Fig. 7; GLM: depth $t_{1,40} = 1.77$, P $=0.08$; site $t_{1,40} = 2.24$, P $=0.03$; depth x site $t_{1,40} = 1.3$, P $=0.2$) between sites but no significant differences between depths. Differences were observed among both site and depth in submissive behaviors (Fig. 7; GLM: depth $t_{1,40} = 2.8$, P $=0.006$; site $t_{1,40} = 2.21$, P $=0.03$; depth x site $t_{1,40} = 1.91$, P $=0.06$) with more fishes being chased in shallow waters than deeper waters. No significant differences were found among either sites or depths for courtship behaviors, as they were rarely observed (Fig. 7; GLM: depth $t_{1,40} = 0.58$, P $=0.57$; site $t_{1,40} = 0.1$, P $=0.92$; depth x site $t_{1,40} = 0.75$, P $=0.45$).

2.5 Discussion

Results of this study suggest that the lower limits of herbivorous fish distribution results from top-down, non-consumptive predation effects rather than resource limitation or metabolic constraints. Terrestrial research has highlighted the importance of non-consumptive predator effects, whereby prey are forced to modify foraging ranges to reduce predation risk (Peckarsky et

al. 2008; Preisser et al. 2009). The "landscape of fear" model suggests prey alter their foraging behaviors in response to spatial and temporal heterogeneity in predation risk, and has recently been evaluated in marine ecosystems (Matassa and Trussell 2011; Catano et al. 2016; Davis et al. 2017a). While predator abundance was low among all depths surveyed (Kane and Tissot 2017), the low-light environment at mesophotic depths likely inhibits the visual acuity of herbivorous fishes relative to that of piscivores, whose eyes are more well-adapted to low-light conditions (McFarland 1991). Our models of herbivore distribution indicate the combination of more patchily distributed habitat (increased sand cover and lower coral cover at mesophotic depths) as well as reduced light levels best explain the reduction in herbivorous fishes at mesophotic depths, supporting landscape of fear models.

Many key predator species have been overfished both in Hawaii and reefs worldwide – while abundance may currently appear low and even decreased at mesophotic depths, the historic threat of these species and their preferences for deeper waters may instinctually inhibit larger herbivorous fishes from spending significant time at mesophotic depths (Jackson et al. 2001; Papastamatiou et al. 2015). Studies of predators on mesophotic reefs are limited, but initial studies by Papastamatiou et al. (2015) in the Northwestern Hawaiian Islands reveal apex predators undergo regular diel movements between shallow and mesophotic depths and obtain approximately 35% of their diet from mesophotic reefs. The observed movement patterns of predators, in conjunction with decreased visual acuity from low light levels and reduced sheltering space likely enhance predation threat in herbivorous fishes at mesophotic depths (Brokovich et al. 2010b; Papastamatiou et al. 2015; Catano et al. 2016, 2017).

Light levels have been widely documented to generate strong bottom-up effects in ecological systems (Wootton and Power 1993; Stoepler and Lill 2013; Bennie et al. 2015). From

savannahs to streams, light strongly affects upper trophic levels through regulation of terrestrial and aquatic plant productivity. In marine systems, algal quality and productivity is driven by both nutrient availability (specifically nitrogen and phosphorus) and light (Lapointe et al. 2004; Gutow et al. 2014; Adam et al. 2015). Conversely, we found that light levels likely facilitate top-down regulation of herbivorous fishes at mesophotic depths. Unlike studies in other systems, we found that decreased light levels do not constrain the quantity or quality of algal resources at upper mesophotic depths. Nitrogen composition was virtually identical between 10m and 40m turf algal samples. C:N ratios between shallow and mesophotic depths were also nearly identical, indicating the nutritional value of turf algae is not reduced at MCE depths. Instead, reductions in light levels at mesophotic depths likely affect the visual acuity of shallow reef fish species, thereby enhancing perceived predation risk. Brokovich et al. (2010b) studied eye and retina anatomy of a common damselfish from 5, 20, and 40m depths in the Red Sea and found no discernable changes in eye anatomy, suggesting that fishes do not readily adapt to lower light levels - hence overall visual acuity may be reduced in fishes that encompass both shallow and mesophotic depths.

Acanthurids, the most abundant of the herbivorous fishes, decreased significantly and universally with depth. Conversely, scarines displayed variable and site-dependent patterns with depth, with increased abundances at 20-30m in the northern and central regions of Hawaii. The increase in abundance at 20-30m was a result of spawning behavior observed during surveys. At sites without spawning activity, parrotfish were rarely observed at depth. Additional differences among species' distributions with depth may be attributed to body size. The most common herbivores at depth were *Zebrasoma flavescens*, a small-bodied surgeonfish, while large-bodied fishes such as surgeonfish and parrotfish spp. were rarely observed. In contrast with most

terrestrial studies, vulnerability to predation in reef fishes has been positively correlated with body size, suggesting smaller bodied fishes are less wary in response to predators (Sinclair et al. 2003; Januchowski-Hartley et al. 2011; Burkepile et al. 2013; Catano et al. 2016). Our work supports this observation, with smaller-bodied herbivores more commonly observed at mesophotic depths and larger-bodied herbivores rarely observed.

We were subsequently interested in parsing out changes in algal quality between shallow and mesophotic depths as a potential mechanism for reductions in herbivorous fish abundances. While deep algae appear to be just as nutritious as shallow algae, it is possible that deep algae contain antiherbivory compounds and inducible antiherbivory defenses (Hay and Steinberg 1992, Macaya and Thiel 2008). We identified three abundant brown macroalgae in deep collections (*Dictyota* sp., *Padina* sp., and *Stypopodium flabelliforme*) that belong to the Dictyotaceae, a family known to have antiherbivory compounds. Previous work in the Caribbean has shown reduced herbivory below 30m was largely a result of increased chemical defenses in algal communities (Hay 1981b, 1997). Choice experiments reveal that unlike Caribbean reefs, deep algal communities in Hawaii appear to be highly palatable. When algal communities from 40m were transplanted to 10m depth, fish spent significantly more time grazing and grazed more intensely on deep algal communities than control (10m) algal communities. Behavioral observations of the acanthurid *Zebrasoma flavescens* further indicate that when these fishes are present in mesophotic depths, they behave the same way as when they are in shallow waters. When averaged between sites, shallow *Zebrasoma* spent 49% of the observed time foraging, while deep *Zebrasoma* spent 52% of time foraging; percentage time swimming was also within one percentage point between shallow and deep fishes observed. We commonly observed turf algal patches with turfs in excess of 3cm length, indicating that turf algae at these depths are not

commonly or heavily grazed. Brokovich et al (2010) directly measured grazing along a depth continuum in the Red Sea and similarly found decreased grazing intensity by herbivorous fishes at mesophotic depths. Yet, evidence of feeding at MCE depths and preferential feeding on deep algae when transplanted to shallow depths do not translate to high grazer abundance or high grazing pressure at mesophotic depths, further supporting non-consumptive predation risk as the most probable mechanism limiting herbivore distribution with depth.

Our work demonstrates that herbivorous fish distributions across a shallow to mesophotic gradient are largely the result of indirect, top-down processes. We still have much to learn regarding algal-herbivore interactions at mesophotic depths. Differences in abundance trajectories among herbivorous families further support the need for additional species-specific insight. Additionally, very few studies investigate mesophotic algal communities, and the roles they may (or may not) play in influencing overall reef communities at these depths. Many of the predictive models for coral-algal-herbivore interactions do not include the role depth may play in limiting herbivore abundance (Mumby and Hastings 2007; Brokovich et al. 2010a; Adam et al. 2015). Hence future attempts to address herbivore effects on coral reef resilience would benefit by addressing the lower extent of herbivorous fish depth ranges. Multiple studies have documented extensive coral-dominated reefs below 30m, yet herbivorous fishes are rare at these depths (Kahng and Maragos 2006; Brokovich et al. 2008; Kahng et al. 2014). These findings highlight opportunities for exploring alternative mechanisms influencing coral reef resilience, and the overall need to continue and expand mesophotic coral reef studies.

2.6 Acknowledgments

2.7 References

Abbott IA. (1999) *Marine red algae of the Hawaiian Islands*. Bishop Museum Press, Honolulu, HI

Abbott IA, Huisman JM (2004) *Marine green and brown algae of the Hawaiian Islands*. Bishop Museum Press, Honolulu, HI

Adam TC, Burkepile DE, Ruttenberg BI, Paddack MJ (2015) Herbivory and the resilience of Caribbean coral reefs : knowledge gaps and implications for management. Mar. Ecol. Prog. Ser. 520:1–20

Bejarano I, Appeldoorn RS, Nemeth M (2014) Fishes associated with mesophotic coral ecosystems in La Parguera, Puerto Rico. Coral Reefs 33:313–328

Bennie J, Davies TW, Cruse D, Inger R, Gaston KJ (2015) Cascading effects of artificial light at night: resource-mediated control of herbivores in a grassland ecosystem. Philos. Trans. R. Soc. Lond. B. Biol. Sci. 370:20140131

Brokovich E, Ayalon I, Einbinder S, Segev N, Shaked Y, Genin A, Kark S, Kiflawi M (2010a) Grazing pressure on coral reefs decreases across a wide depth gradient in the Gulf of Aqaba, Red Sea. Mar. Ecol. Prog. Ser. 399:69–80

Brokovich E, Ben-Ari T, Kark S, Kiflawi M, Dishon G, Iluz D, Shashar N (2010b) Functional changes of the visual system of the damselfish Dascyllus marginatus along its bathymetric range. Physiol. Behav. 101:413–21

Brokovich E, Einbinder S, Shashar N, Kiflawi M, Kark S (2008) Descending to the twilight-zone: changes in coral reef fish assemblages along a depth gradient down to 65 m. Mar. Ecol. Prog. Ser. 371:253–262

Burkepile DE, Burns CE, Tambling CJ, Amendola E, Buis GM, Govender N, Nelson V, Thompson DI, Zinn AD, Smith MD (2013) Habitat selection by large herbivores in a southern African savanna: the relative roles of bottom-up and top-down forces. Ecosphere 4:art139

Carpenter RC (1988) Mass mortality of a Caribbean sea urchin: Immediate effects on community metabolism and other herbivores. Proc. Natl. Acad. Sci. U. S. A. 85:511–4

Catano LB, Barton MB, Boswell KM, Burkepile DE (2017) Predator identity and time of day interact to shape the risk–reward trade-off for herbivorous coral reef fishes. Oecologia 183:763–773

Catano LB, Rojas MC, Malossi RJ, Peters JR, Heithaus MR, Fourqurean JW, Burkepile DE (2016) Reefscapes of fear: Predation risk and reef hetero-geneity interact to shape herbivore foraging behaviour. J. Anim. Ecol. 85:146–156

Cvitanovic C, Hoey AS (2010) Benthic community composition influences within-habitat variation in macroalgal browsing on the Great Barrier Reef. Mar. Freshw. Res. 61:999–1005

Davis K, Carlson PM, Bradley D, Warner RR, Caselle JE (2017a) Predation risk influences feeding rates but competition structures space use for a common Pacific parrotfish. Oecologia 1–11

Davis K, Carlson PM, Lowe CG, Warner RR, Caselle JE (2017b) Parrotfish movement patterns vary with spatiotemporal scale. Mar. Ecol. Prog. Ser. 577:149–164

Feitoza B, Rosa R, Rocha L (2005) Ecology and zoogeogrpahy of deep-reef fishes in northeastern Brazil. Bull. Mar. Sci. 76:725–742

Ferreira CEL, Floeter SR, Gasparini JL, Ferreira BP, Joyeux JC (2004) Trophic structure patterns of Brazilian reef fishes: a latitudinal comparison. J. Biogeogr. 31:1093–1106

Floeter S, Behrens M, Ferreira C (2005) Geographical gradients of marine herbivorous fishes: patterns and processes. Mar. Biol. 1–19

Fox R, Bellwood D (2007) Quantifying herbivory across a coral reef depth gradient. Mar. Ecol. Prog. Ser. 339:49–59

Fukunaga A, Kosaki RK, Wagner D, Kane C (2016) Structure of mesophotic reef fish assemblages in the Northwestern Hawaiian Islands. PLoS One 11:e0157861

Garcia-Sais JR (2010) Reef habitats and associated sessile-benthic and fish assemblages across a euphotic–mesophotic depth gradient in Isla Desecheo, Puerto Rico. Coral Reefs 29:277–288

Gil MA, Zill J, Ponciano JM (2017) Context-dependent landscape of fear: Algal density elicits risky herbivory in a coral reef. Ecology 98:534–544

Gutow L, Rahman MM, Bartl K, Saborowski R, Bartsch I, Wiencke C (2014) Ocean acidification affects growth but not nutritional quality of the seaweed Fucus vesiculosus (Phaeophyceae, Fucales). J. Exp. Mar. Bio. Ecol. 453:84–90

Hay M (1981a) Spatial patterns of agrazing intensity on a Caribbean barrier reef: herbivory and algal distribution. Aquat. Bot. 11:97–109

Hay M (1981b) Herbivory, Algal distribution, and the maintenance of between-habitat diversity on a tropical fringing reef. Am. Nat. 118:520–540

Hay M (1984) Patterns of fish and urchin grazing on caribbean coral reefs: are previous results typical? Ecology 65:446–454

Hay ME (1997) The ecology and evolution of seaweed-herbivore interactions on coral reefs. Coral Reefs 16:S67–S76

Hinderstein LM, Marr JC a., Martinez F a., Dowgiallo MJ, Puglise K a., Pyle RL, Zawada DG, Appeldoorn R (2010) Theme section on "Mesophotic Coral Ecosystems: Characterization, Ecology, and Management." Coral Reefs 29:247–251

Jackson JB, Kirby MX, Berger WH, Bjorndal K a, Botsford LW, Bourque BJ, Bradbury RH, Cooke R, Erlandson J, Estes J a, Hughes TP, Kidwell S, Lange CB, Lenihan HS, Pandolfi JM, Peterson CH, Steneck RS, Tegner MJ, Warner RR (2001) Historical overfishing and the recent collapse of coastal ecosystems. Science 293:629–37

Januchowski-Hartley FA, Graham NAJ, Feary DA, Morove T, Cinner JE (2011) Fear of fishers: human predation explains behavioral changes in coral reef fishes. PLoS One 6:e22761

Kahng S, Copus J, Wagner D (2014) Recent advances in the ecology of mesophotic coral ecosystems (MCEs). Curr. Opin. Environ. Sustain. 7:72–81

Kahng SE, Garcia-Sais JR, Spalding HL, Brokovich E, Wagner D, Weil E, Hinderstein L, Toonen RJ (2010) Community ecology of mesophotic coral reef ecosystems. Coral Reefs 29:255–275

Kahng SE, Maragos JE (2006) The deepest, zooxanthellate scleractinian corals in the world? Coral Reefs 25:254–254

Kane CN, Tissot BN (2017) Trophic designation and live coral cover predict changes in reef-fish community structure along a shallow to mesophotic gradient in Hawaii. Coral Reefs 1–11

Lapointe BE, Barile PJ, Yentsch CS, Littler MM, Littler DS, Kakuk B (2004) The relative importance of nutrient enrichment and herbivory on macroalgal communities near Norman's Pond Cay, Exumas Cays, Bahamas: A "natural" enrichment experiment. J. Exp. Mar. Bio. Ecol. 298:275–301

Matassa CM, Trussell GC (2011) Landscape of fear influences the relative importance of consumptive and nonconsumptive predator effects. Ecology 92:2258–2266

Menge B (1995) Indirect effects in marine rocky intertidal interaction webs: patterns and importance. Ecol. Monogr. 65:21–74

Mumby PJ, Hastings A (2007) The impact of ecosystem connectivity on coral reef resilience. J. Appl. Ecol. 45:854–862

Nemeth M, Appeldoorn R (2009) The distribution of herbivorous coral reef fishes within fore-reef habitats: the role of depth, light and rugosity. Caribb J Sci 45:247–253

Paine RT (1980) Food Webs: Linkage, Interaction Strength and Community Infrastructure. J. Anim. Ecol. 49:666–685

Papastamatiou YP, Meyer CG, Kosaki RK, Wallsgrove NJ, Popp BN (2015) Movements and foraging of predators associated with mesophotic coral reefs and their potential for linking ecological habitats. Mar. Ecol. Prog. Ser. 521:155–170

Peckarsky B, Abrams P, Bolnick D, Dill L, Grabowski J, Luttbeg B, Orrock J, Peacor S, Preisser E, Schmitz O, Trussell G (2008) Revisiting the classics : considering nonconsumptive effects in textbook examples of predator – prey interactions. Ecology 89:2416–2425

Preisser EL, Bolnick DI, Grabowski JH (2009) Resource dynamics influence the strength of non-consumptive predator effects on prey. Ecol. Lett. 12:315–323

Russ GR (2003) Grazer biomass correlates more strongly with production than with biomass of algal turfs on a coral reef. Coral Reefs 22:63–67

Russ GR, McCook LJ (1999) Potential effects of a cyclone on benthic algal production and yield to grazers on coral reefs across the central Great Barrier Reef. J. Exp. Mar. Bio. Ecol. 235:237–254

Sinclair ARE, Mduma S, Brashares JS (2003) Patterns of predation in a diverse predator–prey system. Nature 425:288–290

Smith T, Glynn P, Maté J, Toth L, Gyory J (2013) A depth refugium from catastrophic coral bleaching prevents regional extinction. Ecology ???

Smith TB (2008) Temperature effects on herbivory for an Indo-Pacific parrotfish in Panam{á}: implications for coral--algal competition. Coral Reefs 27:397–405

Steneck RS, Bellwood DR, Hay ME (2017) Herbivory in the marine realm: shaping ecosystems and colliding with the Anthropocene. Curr. Biol. 27:in press

Stoepler TM, Lill JT (2013) Direct and indirect effects of light environment generate ecological trade-offs in herbivore performance and parasitism. Ecology 94:2299–310

Tootell JS, Steele MA (2016) Distribution, behavior, and condition of herbivorous fishes on coral reefs track algal resources. Oecologia 181:13–24

Vandenhoek C, Breeman AM, Bak RPM, Vanbuurt G (1978) Distribution of algae, corals and gorgonians in relation to depth, light attenuation, water-movement and grazing pressure in fringing coral-reef of Curacao, Netherlands Antilles. Aquat. Bot. 5:1–46

Witman J, Etter R, Smith F (2004) The relationship between regional and local species diversity in marine benthic communities: a global perspective. … Acad. Sci. … 101:15664–15669

Wootton JT, Power ME (1993) Productivity, consumers, and the structure of a river food chain. Proc. Natl. Acad. Sci. U. S. A. 90:1384–7

2.8 Tables

Table 2.1: Percentage of variation explained by each variable in DistLM marginal test.

Variable	P	% Variation Explained
Macroalgae	0.307	0.97
Turf Algae	0.001	4.91
Corallivore	0.001	8.78
Sand	0.001	5.62
Temperature	0.563	0.68
Light	0.001	3.79
Piscivore	0.032	1.72

Table 2.2: Highest ranking DistLM models using BEST selection procedure.

AIC	R2	Selections
926.53	0.138	Coral, Sand, Light
926.93	0.150	Turf Algae, Coral, Sand, Light
927.24	0.118	Coral, Light
927.28	0.147	Coral, Sand, Light, Piscivore
927.31	0.117	Coral, Sand

Table S2.1: Mean density (MD) and +/- 1 SEM of herbivorous fish species across survey depths. Shaded columns indicate mesophotic depths.

Family	Species	3m		10m		20m		30m		40m		50m	
		MD	SE	MD	SE	MD	SE	MD	SE	MD	SE	MD	SE
Acanthuridae	*Acanthurus achilles*	0.26	0.13	0.16	0.07	0.05	0.05	0	0	0	0	0	0
	Acanthurus blochii	0.11	0.09	0.05	0.04	0.03	0.03	0	0	0	0	0.27	0.00
	Acanthurus dussimieri	0.2	0.15	0.3	0.17	0.08	0.04	0.08	0.06	0.26	0.11	0.37	0.20
	Acanthurus guttatus	0.03	0.03	0	0	0	0	0	0	0	0	0	0
	Acanthurus leucopareius	3.83	2.08	0.05	0.04	0.11	0.11	0.08	0.08	0	0	0.13	0.11
	Acanthurus nigricans	0.2	0.13	0.11	0.05	0.16	0.09	0.13	0.11	0	0	0	0
	Acanthurus nigrofuscus	22.11	3.53	13.62	2.42	4.37	0.91	3	0.66	3.29	0.71	2.40	0.53
	Acanthurus nigroris	0.74	0.64	0.22	0.14	0.42	0.24	0.74	0.64	0	0	0.10	0.13
	Acanthurus olivaceous	0.97	0.61	0.73	0.23	0.92	0.29	0.61	0.17	0.65	0.17	0.03	0.00
	Acanthurus triosteges	0.31	0.17	0	0	0	0	0	0	0	0	0	0
	Acanthurus xanthopterus	0	0	0	0	0	0	0.05	0.05	0.09	0.06	0	0
	Ctenochaetus hawaiiensis	0.43	0.2	1.05	0.41	0.74	0.16	0.26	0.11	0.62	0.38	0.30	0.24
	Ctenochaetus strigosis	15.54	4.46	21.32	2.17	16.18	2.15	3.87	1.48	2.56	0.73	1.13	0.36
	Naso lituratus	0.8	0.28	0.95	0.23	1.11	0.23	0.71	0.2	0.32	0.09	1.50	3.39
	Naso unicornis	0.17	0.1	0.05	0.05	0	0	0	0	0.29	0.24	0.07	0.00
	Zebrasoma flavescens	8.57	1.23	17.51	1.69	13.89	2.61	3.92	1.02	2.03	0.52	0.20	0.08
	Zebrasoma veliferum	0.09	0.06	0.08	0.08	0	0	0	0	0	0	1.27	0.26
Balistidae	*Melichthys vidua*	0.11	0.09	0.19	0.09	0.29	0.13	0.21	0.09	0.18	0.08	0.03	0.00
Blennidae	*Cirripectes vanderbilti*	0.14	0.12	0	0	0	0	0	0	0	0	0	0
Kyphosidae	*Kyphosus sandwicensis*	0.06	0.04	0	0	0	0	0	0	0	0	0	0
Labridae	*Calotomus carolinus*	0.03	0.03	0.05	0.04	0.11	0.05	0.11	0.08	0	0	0.07	0.00
	Chlorurus perspicillatus	0	0	0	0	0.03	0.03	0.03	0.03	0	0	0	0
	Chlorurus sordidus	1.54	0.72	0.81	0.38	1.89	0.47	1.5	0.8	0.62	0.32	0	0
	Scarus psittacus	1	0.55	0.03	0.03	0.32	0.17	0.61	0.36	0	0	0.10	0.00

	Scarus rubroviolaceus	0.06	0.06	0.14	0.1	0.05	0.04	0.16	0.11	0.03	0.03	0.40	0.48
Monocanthidae	*Cantherhines verecundus*	0.03	0.03	0	0	0	0	0	0	0	0	0	0
Pomacanthidae	*Centropyge fisheri*	0	0	0.03	0.03	1.03	0.38	2.79	0.83	0.79	0.3	0.40	0.37
	Centropyge potteri	0	0	1.05	0.29	1.76	0.34	0.68	0.23	0.82	0.24	0.47	0.09
Pomacentridae	*Stegastes fasciolatus*	3.6	0.82	0.54	0.34	0.03	0.03	0	0	0	0	0	0
Tetradontidae	*Canthigaster amboinensis*	0.23	0.09	0	0	0	0	0	0	0.03	0.03	0	0
	Canthigaster coronata	0	0	0	0	0	0	0.13	0.08	0.24	0.11	0.13	0.00
	Canthigaster epilampra	0	0	0	0	0	0	0.08	0.06	0.29	0.13	0.13	0.11
	Canthigaster jactator	1.14	0.26	0.84	0.18	0.32	0.1	0.45	0.13	0.79	0.17	0.13	0.26

Table S2.2: Macroalgal species identified from photoquadrat analysis and voucher samples. Algae were noted as rare (1) or common (2). "*" were epiphytes.

Family	Species	3-20m	30-50m
Cyanobacteria	*Leptolyngbya* sp.	2	
	Oscillatoria sp.	1	
Chlorophyta	*Calothrix* sp.		2
	Caulerpa sp.	1	1
	Cladophora sp.	1	1
	Dictyosphaeria sp.		1
	Halimeda sp.	2	2
	Microdictyon sp.	1	1
	Neomeris sp.	1	2
	Phyllodictyon sp.	1	
	Rhipidosiphon sp.	2	
	Siphonogramen sp.	2	
	Ulothrix sp.*	1	
	Ulvaceae	1	
	Unidentifiable green sp.	2	2
	Ventricaria sp.	1	
Phaeophyceae	*Cutleria irregularis*	2	
	Dictyota sp.	2	2
	Feldmannia indica		2
	Padina sp.	2	2
	Sargassum sp.	1	
	Sphacelaria novae-hollandiae	2	
	Stypopodium flabelliforme	2	2
	Turbinaria sp.	1	
	Unidentifiable brown sp.	2	1
Rhodophyta	*Amphiroa* sp.	2	1
	Asparagopsis sp.	2	1
	Ceramium sp.*	1	
	Chondria sp.	2	
	Dasysiphonia japonica	2	
	Dichotomaria sp.		1
	Gibsmithia sp.	1	2
	Herposiphonia sp.	2	
	Neosiphonia sp.		2
	Nongeniculate coraline algae (CCA)	2	
	Peyssonnelia sp.	2	
	Polysiphonia sp.	2	

	Stenopeltis sp.		1
	Tolypiocladia glomerata	2	
	Unidentifiable red sp.	2	2
	Unidentifiable calcareous		1
Sarcinochrysidaceae	*Chrysocystis* sp.	1	

2.9 Figures
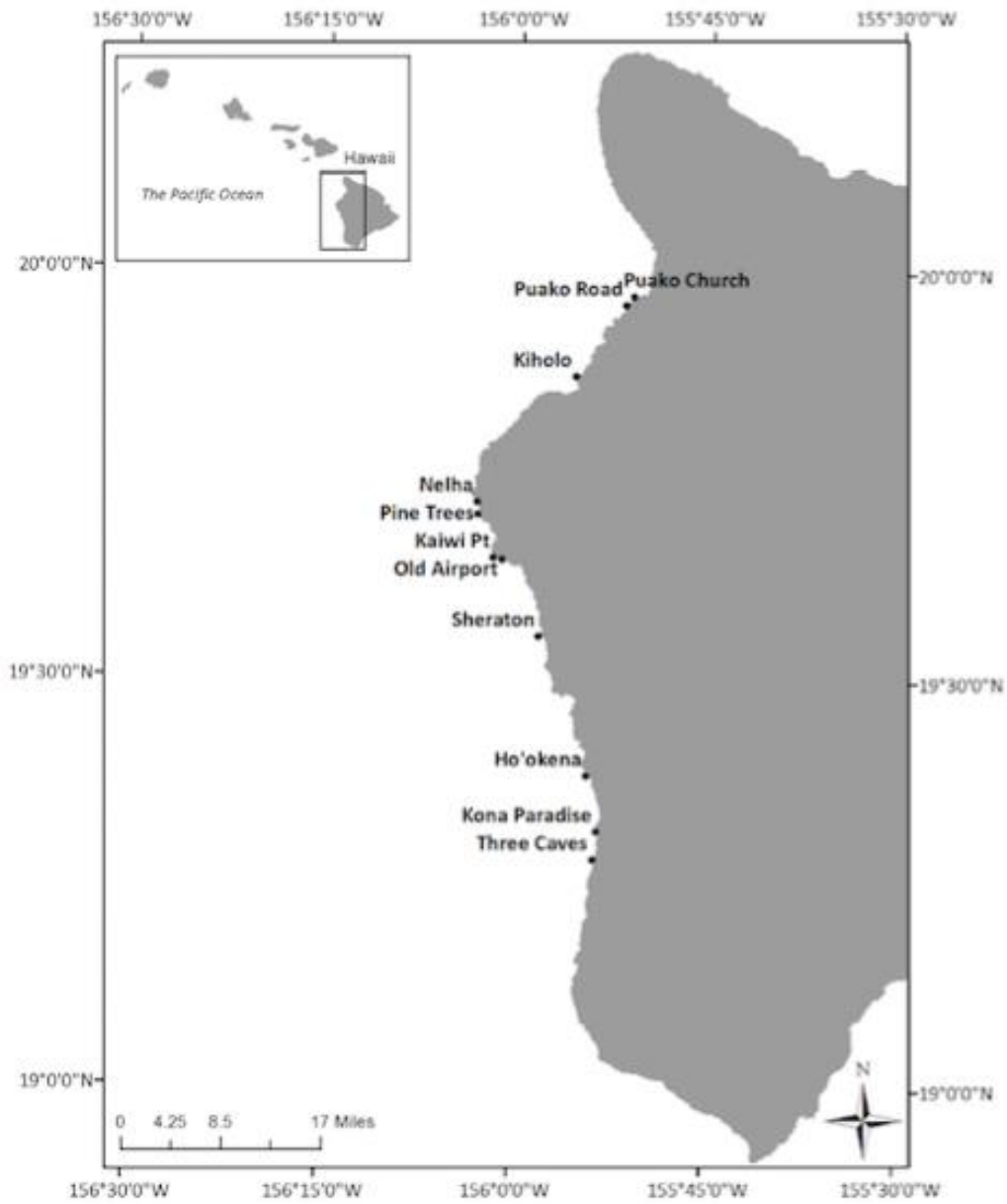
Figure 2.1: Locations of surveys sites along West Hawaii. Starred sites indicate locations of behavioral studies.
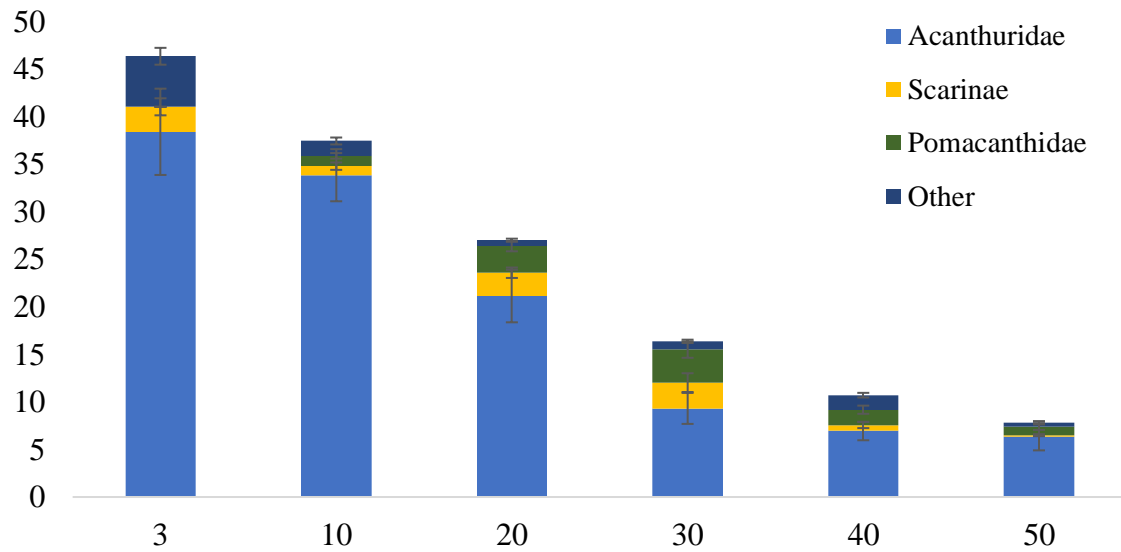
Figure 2.2: Mean (+/- SEM) abundance of herbivorous fishes across survey depths (m).
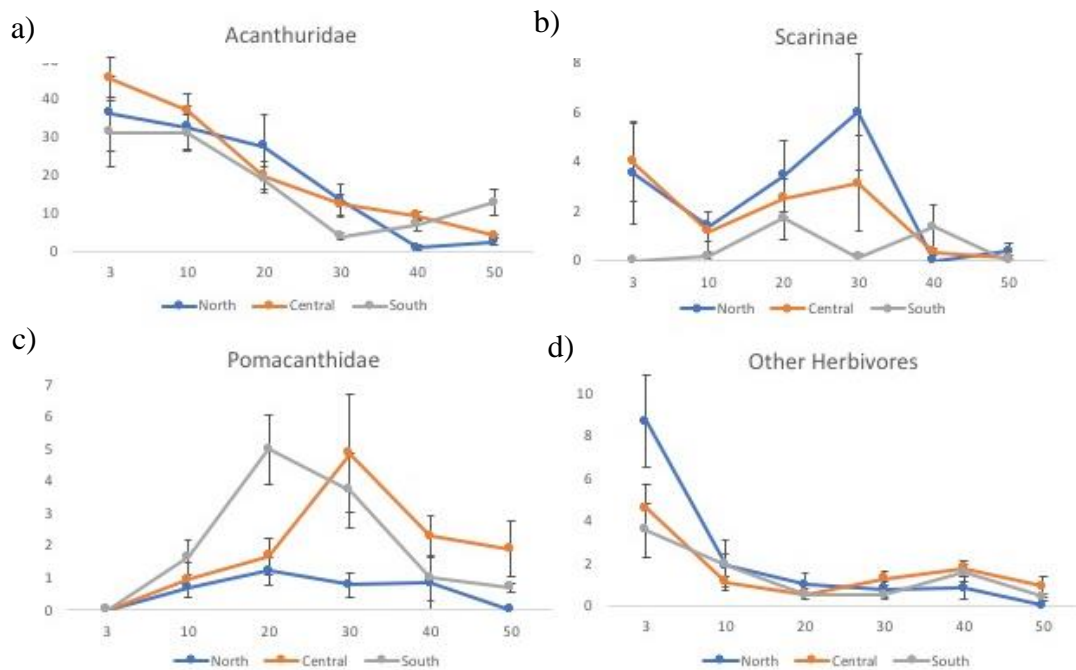
Figure 2.3: Mean (+/- SEM) abundance of a) Acanthuridae, b) Scarinae, c) Pomacanthidae, and d) other Herbivorous fishes across survey depths (m).
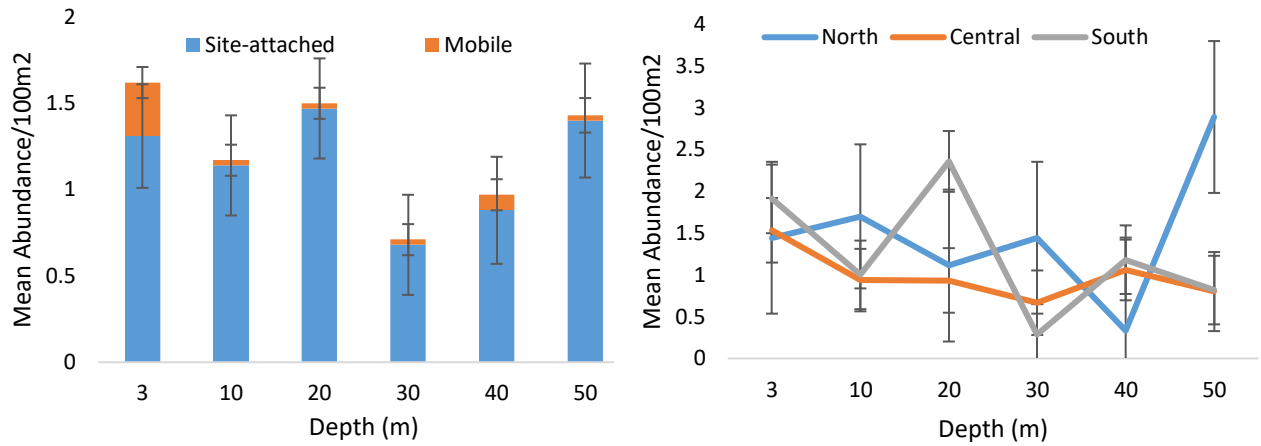
Figure 2.4: Mean (+/- SEM) abundance of a) mobile and site-attached piscivores by depth, and b) piscivorous fishes with each region across depths.
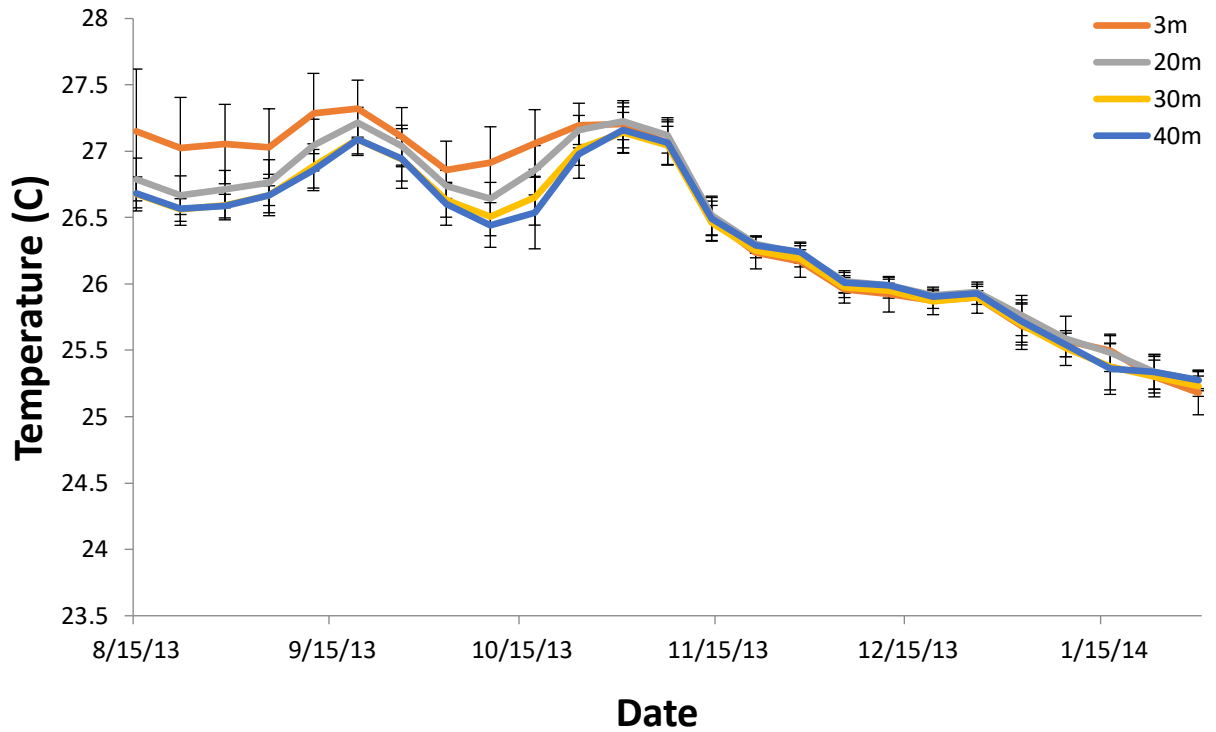
Figure 2.5: Mean weekly temperature (C) at Kona Paradise for a six-month period. Error bars indicate +/- 1 Standard Deviation.
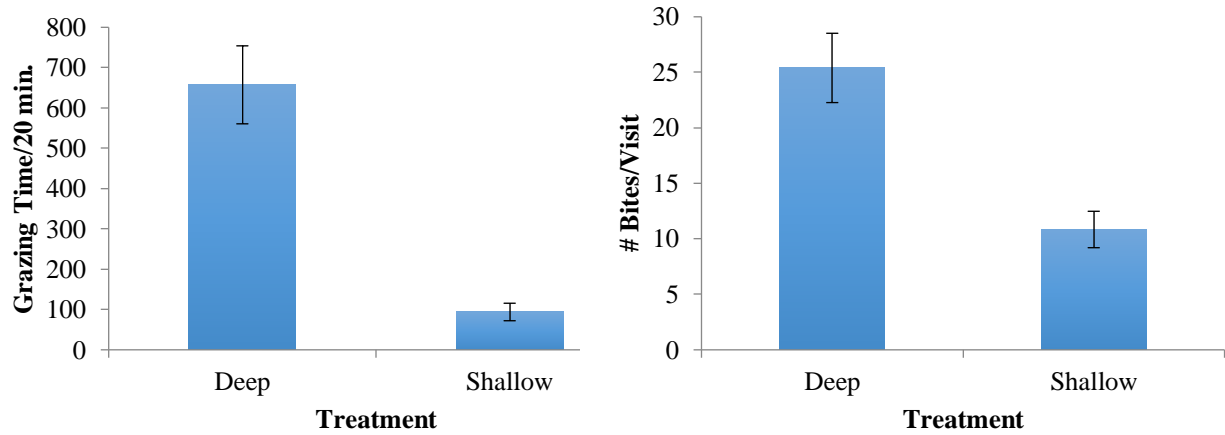
Figure 2.6: Mean proportion (+/- SEM) of a) mean total grazing time and b) bites per visit of herbivorous fish during algal choice experiment.
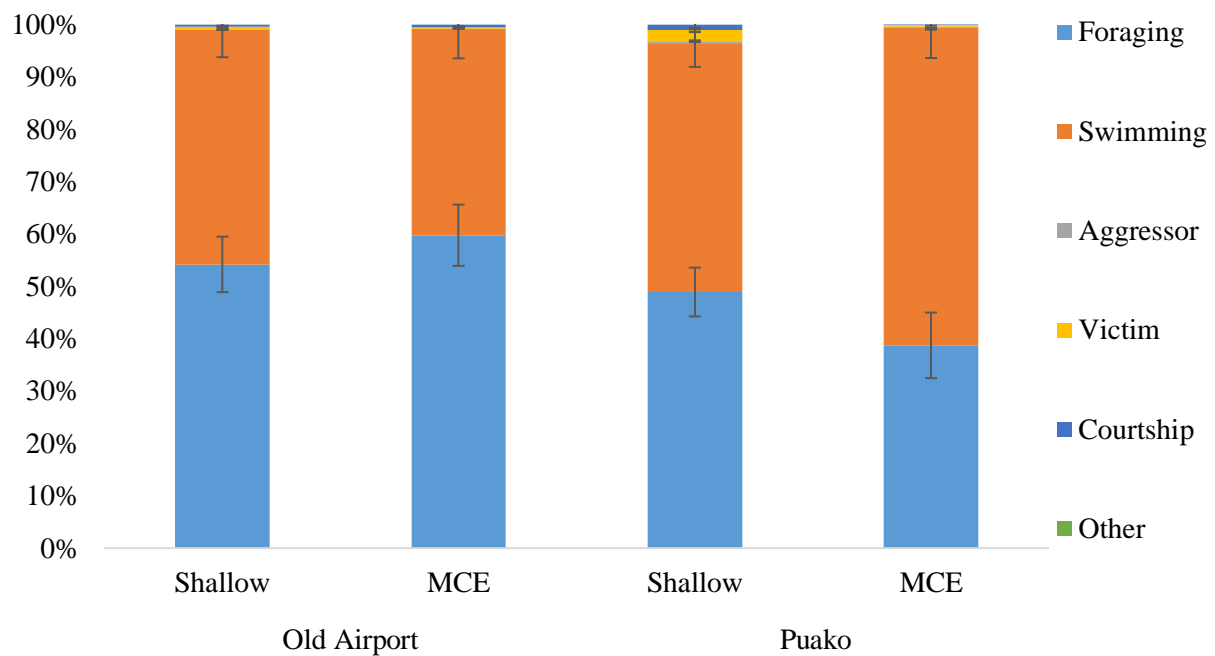
Figure 2.7: Mean (+/- SEM) proportion of time *Zebrasoma flavescens* displayed each behavior.

CHAPTER THREE

HAWAIIAN MESOPHOTIC CORAL REEFS HAVE LIMITED CAPACITY TO ACT AS

REFUGIA FOR SHALLOW REEF FISHES

HAWAIIAN MESOPHOTIC CORAL REEFS HAVE LIMITED CAPACITY TO ACT AS REFUGIA FOR SHALLOW REEF FISHES

3.1 Abstract

The effects of local and global stressors have increased precipitously in coral reef environments over the past few decades. Increasingly, scientists, managers, and conservationists have been seeking coral reef regions that may act as potential refugia from climate change or increasing localized stressors. The vast increase in knowledge of mesophotic coral reef ecosystems (MCEs; 30-150+m) in the past decade has led to hypotheses regarding whether MCEs may act as climate refugia for coral reef species from degraded shallow reef environments. Here we review this hypothesis with regards to the potential of MCEs to act as refugia for shallow coral reef fishes in the Main Hawaiian Islands. We found that MCEs between 30-60m offer limited refugia for shallow reef fishes. Upper MCEs offer more thermally stable environments and >70% of fishes encountered were shallow reef fishes. Below 60m, shallow reef fish occurrence was reduced to 36%. It is possible that shallow MCEs act as a source of propagules for shallow reefs as we documented the first recorded mesophotic spawning aggregations of two species of parrotfish. Yet MCEs overall had lower fish densities and reef fish communities were largely comprised of invertivore and planktivore functional groups that are generally not of commercial or recreational fisheries value. Significant reductions in herbivorous fishes with depth indicate that MCEs will have limited capacity to re-seed shallow reefs with similar fish assemblages currently occupying shallow reefs. While we found evidence that upper mesophotic reef environments (30-60m) act as environmental refugia and harbor shallow reef fishes, the potential to restore shallow

environments is limited and likely would not result in similar community compositions that currently inhabit shallow reefs.

3.2 Introduction

It has been proposed that mesophotic reefs (MCEs; 30-150+m) may act as refugia for more impacted shallow coral reef ecosystems, and this hypothesis has gained interest among coral reef management agencies (Riegl and Piller 2003; Lesser et al. 2009; Hinderstein et al. 2010; Bridge et al. 2013). The 'deep reef refugia' hypothesis (DRRH) was first formulated in the late 1990s, largely for zones of coral reefs that had reduced susceptibilities to certain disturbances such as cyclone damage or thermal stress (Liddell and Ohlhorst 1988; Glynn 1996; Riegl and Piller 2003). Of late, DRRH has extended beyond physical sheltering capabilities and has been proposed as possible recruitment sources for shallow coral reef ecosystems (Bongaerts et al. 2010; Slattery et al. 2011; Laverick et al. 2016; Turner et al. 2017). The DRRH currently focuses on two primary questions: 1) Are deep reefs less susceptible to disturbance than shallow reefs, and 2) can deep reefs provide a source of propagules to re-seed nearby shallow coral reefs experiencing decline (Hughes and Tanner 2000; Bridge and Guinotte 2012; Kahng et al. 2014; Loya et al. 2016).

Refugia for coral reef organisms is gaining priority among managers and conservationists as coral reefs are increasingly under threat from both local and global stressors (Hughes et al. 2003; Pandolfi et al. 2005; Spalding and Brown 2015)It is estimated that if coral decline continues at the current rate approximately 50% may be lost in the next 20-40 years (Carpenter et al. 2008; van Hooidonk et al. 2013; Hughes et al. 2017). These rapid declines have largely been attributed to anthropogenic impacts ranging from local factors such as overfishing, destructive fishing, reductions in water quality from land-based runoff and pollution, to global factors such

as increased sea surface temperatures and ocean acidification as a result of increased atmospheric carbon dioxide concentrations (Hoegh-Guldberg and Bruno 2010; Spalding and Brown 2015; Albright et al. 2016; Hoey et al. 2016; Eyre et al. 2018).

The widespread degradation of coral reefs has cascading effects on organisms dependent on reefs for food and shelter. It is estimated that 60-70% of reef fishes have been negatively impacted by the recent loss of coral communities (Jones et al. 2004; Munday et al. 2008; Hoey et al. 2016). Degradation of coral composition and structural complexity resulting from local and global pressures will likely result in declines in the abundance of a wide range of coral reef fishes (McClanahan et al. 2014; Pratchett et al. 2015). Ocean warming and acidification will also directly affect reef fishes' physiology and behavior, ultimately affecting the abundance, distribution, and species composition of reef fishes (Munday et al. 2008; Wilson et al. 2010; Hoey et al. 2016; Leis 2018). As a result, coral reef managers and conservationists have prioritized the identification of coral reef regions that may have reduced susceptibilities to current and predicted anthropogenic stressors (Riegl and Piller 2003; Bridge et al. 2013; Chollett and Mumby 2013).

3.2.1 MCEs as environmental refugia

Despite a plethora of literature describing the effects of natural and anthropogenic disturbances on shallow coral reefs, very few studies report disturbance effects on reefs below 20 meters. Although a large amount of variation exists in MCE susceptibilities to disturbance, there is a general trend in the literature indicating that MCEs may be less susceptible to disturbance events. Several studies have noted that MCE regions have experienced lower coral bleaching and subsequent mortality following thermal stress events than adjacent shallow water regions (Bridge and Guinotte 2012; Sinniger et al. 2012; Muir et al. 2015). Decreased coral bleaching in

84

MCE regions is likely due to lower overall water temperatures in deeper water. Frade et al. (2008) observed a 1°C difference in average summer water temperatures between shallow (5-7m) and deep (35-40m) coral reefs in the Caribbean, with even more pronounced differences over broader depth ranges. Given that coral bleaching events have been strongly correlated with 1-2°C increases in temperatures over the long-term annual maximum averages, these small decreases in temperature between shallow and deep regions may be highly important for preventing coral bleaching and mortality events (Liu et al. 2001, Liu et al. 2003, Hoegh-Guldberg 1999, McClanahan et al. 2012). Recent studies of MCEs in the Caribbean, though, reported more significant coral bleaching in mesophotic corals than their shallower counterparts, indicating that MCEs may not be thermal refugia for corals after all (Chollett and Mumby 2013; Neal et al. 2013; Smith et al. 2016d).

MCE regions may be less susceptible to turbulent oceanographic conditions than coral reefs in shallow depths. Episodic peaks in wave energy resulting from hurricanes is negatively correlated with depth, thus coral reef ecosystems in deeper waters may be sheltered from surface-related wave energy, yet this effect is dependent on storm characteristics (Liddell and Ohlhorst 1998; White et al. 2013). Tidal forcing and turbulence common in shallow systems are also likely to have muted effects on reef organisms at mesophotic depths, although this has yet to be empirically tested. The occurrence of internal waves and tides within deep reefs systems can bring cooler, more nutrient rich waters to mesophotic depths, but again, few studies have consdered the effects of these events on mesophotic ecosystems (Leichter et al. 2005; Leichter and Genovese 2006; Bridge et al. 2010; Smith et al. 2016c).

Mesophotic regions may also provide refuge from direct anthropogenic impacts such as fishing pressure, recreational snorkeling and diving, and ship groundings. Reef fish collections and fishing are generally restricted to depths of less than 20 meters. Most reef fish fishing is conducted via gillnets, traps, and spearfishing while freediving or on SCUBA, and divers rarely attain depths greater than 20 meters due to physiological and gear restraints. Overfishing is common among shallow coral reefs in populated regions, as documented by extreme shifts in trophic structure away from apex predator dominated reefs in pristine/uninhabited locations to primary and secondary consumer dominated reefs in populated regions (Friedlander et al. 2010; Williams et al. 2011). Mesophotic reefs have shown promise as refugia for fishery-targeted species with fished species found to be both larger and more abundant at mesophotic depths (Lindfield et al. 2014a, 2016; Asher et al. 2017a).

3.2.2 MCEs as a source for propagules

The concept of connectivity between reef areas has been a focus of investigation for the past decade, largely a result of increased precision in DNA and RNA studies. Connectivity studies have mainly been employed to investigate and design marine protected areas (Mumby and Hastings 2007; Rocha et al. 2007; Christie et al. 2010; Sale et al. 2010; Toonen et al. 2013), and recent studies in Hawaii have investigated connectivity between islands and island groups (Toonen et al. 2011; Selkoe et al. 2014; Tenggardjaja et al. 2016). A number of studies have employed genetic sequencing to determine connectivity between shallow and mesophotic organisms. These studies have predominantly centered around coral connectivity and have yielded mixed results, with some studies showing strong genetic partitioning (Bongaerts et al. 2011, 2013, 2015) and others showing mixed gene flow between shallow and mesophotic corals (van Oppen et al. 2011; Holstein et al. 2015, 2016). Studies of reef fish connectivity between

shallow and mesophotic depths has been extremely limited, yet show weak genetic structuring between depth zones (Tenggardjaja et al. 2014, 2016; Vaz et al. 2016). Due to the movement patterns of reef fishes, habitat plasticity and pelagic larval stages, it is likely that typically shallow fishes may move to deeper waters when spawning and thus become a source of propagules for reefs of all depths.

Here, we investigate the possibility that mesophotic reefs serve as refugia for shallow reef fishes in the Main Hawaiian Islands. We explore whether Hawaii's mesophotic reefs act as propagule sources by examining 1) reef fish composition between shallow and mesophotic depths, 2) deep spawning potential, 3) fish size structure, and 4) long-term temperature recordings.

3.3 Methods

3.3.1 Location of Studies

This study encompasses reef fish survey data from six islands in the Main Hawaiian Island region of the Hawaiian Archipelago (Fig. 1). Diver-based surveys were conducted from 3-100m depths at Niʻihau, Oʻahu, Lanaʻi, Molokaʻi, Maui and Hawaiʻi islands (Table 1). Islands were selected to span the extent of the Main Hawaiian Islands, and sampling locations were chosen to represent a range of depths and reef types among the islands. Despite the range of reef types among the islands (fringing reef, fore-reef, lagoon) all surveys were conducted in areas of sufficient hard bottom reef or structure to maintain consistency in sampling habitats from depths of 3-100m. Water temperature monitoring was conducted at Hawaiʻi Island from August 2013 to February 2014 to ascertain long-term temperature trends from 3-40m depth.

3.3.2 Sampling Design

Reef fishes were sampled using diver-based observational surveys. Mesophotic surveys were conducted via a belt transect method using both open-circuit and closed-circuit technologies. We combined surveys from open and closed circuit technologies as recent research suggests that there are no discernable differences in surveys between the two methodologies for moderate and low fishing pressure regions in the main Hawaiian Islands (Gray et al. 2016). As MCE depths are seldom accessed and/or fished in the main Hawaiian Islands, we have inherently categorized them as moderate to low fishing pressure and included both methodologies for analysis. Mesophotic surveys at Niʻihau, Oʻahu, Lanaʻi, Molokaʻi and Maui were conducted using closed-circuit rebreathers in the spring of 2015. At each of these survey sites, all conspicuous, diurnally active fishes were counted and sized to the nearest centimeter total length along a 25x2m belt transect (Kane et al. 2014). Additionally, 6 belt transect surveys were conducted at Hawaiʻi island from 70 to 91m depth using closed-circuit rebreathers to enhance diver safety. All conspicuous, diurnally active fishes were counted and sized to the nearest centimeter for each survey method described.

Shallow coral reef fish surveys (3-28m) were conducted using open-circuit SCUBA between 2010 and 2016. Stationary point count surveys were conducted at Niʻihau, Oʻahu, Lanaʻi, Molokaʻi, and Maui islands using a circular plot 15m in diameter during the NOAA Reef Assessment and Monitoring Program (RAMP) cruises to the main Hawaiian Islands (Heenan et al. 2017). At each survey site, all conspicuous, diurnally active fishes were counted and sized to the nearest centimeter. At Hawaii Island, a combination of stationary point count surveys (n=257) and belt transect (n=110) surveys were conducted. Stationary point count surveys followed the above survey protocol conducted as part of NOAA's RAMP expeditions (Heenan et

al. 2017) from 3 to 28m depths. Belt transect surveys were conducted between 2013-2015 using a 25x4m belt method and spanned depths from 3 to 50m (Kane and Tissot 2017).

All fishes encountered in surveys were additionally assigned trophic designations according to gut content analyses published on Hawaiian fish species in Hobson (1974) and Randall (2007). Species lists and assignments were corroborated with the NOAA's Pacific Islands Fisheries Science Center. Fishes were binned into one of six broad trophic designations: corallivore, herbivore, invertivore, omnivore, piscivore, and planktivore.

Four representative reef fish species were selected to understand species-specific trends in fish distribution with depth. Two of these species, *Scarus psittacus* and *Chlorurus spilurus*, are highly valued for their ecological roles as herbivores and are also highly prized recreational and commercial fishing targets, especially larger individuals (Gray et al. 2016; Kelly et al. 2016). One species, *Zebrasoma flavescens,* was selected as juveniles of this species are a heavily targeted aquarium fishery species (Williams et al. 2009; Stevenson et al. 2011; Stevenson and Tissot 2013; Gray et al. 2016). The final species selected, *Thalassoma duperrey*, is one of the most abundant fishes on Hawaiian reefs, but is not fished commercially or recreationally.

3.3.3 Behavioral Observations

We conducted focal observations of two herbivorous fish species to examine behavioral patterns in both shallow (<10m) and mesophotic (>30m) coral reef environments on Hawaiʻi Island. The first species was the surgeonfish *Zebrasoma flavescens*, an important aquarium fish species and widely abundant throughout the Main Hawaiian Islands, and 2) the parrotfish *Scarus psittacus*, an important herbivore and widely fished for recreational spearfishing. Additionally, *S. psittacus* was selected for focal observations after survey analyses indicated unique spikes in abundance at 20-40m depths at two sites, Puako and Old Airport, within Hawaiʻi Island. We

conducted focal observations of *Z. flavescens* at these two sites, whereas we conducted

observations of terminal male *S. psittacus* at only one of the anomalous sites due to insufficient

abundances at MCE depths at the other site during the course of our behavioral studies. We

restricted our observations to terminal male *S. psittacus* as females of this species repeatedly

swam up and down the reef slope from 10-50m, making it impossible to maintain diver safety

while following these fish. *Zebrasoma flavescens* observations were conducted on adult fishes

(>10cm length). We selected 20 individuals at each depth treatment; shallow observations

included fishes found at 3-10m depths, while deep observations included fishes from 30 to 50m

depths. Focal observations were conducted by attaching a GoPro camera to a 2m PVC pipe. At

each depth, SCUBA divers selected one fish and filmed them for a period of five minutes from a

distance of at least 5m. If the focal fish was startled by the diver or went out of view of the diver

at any time during the 5-minute session, filming was aborted and another fish was selected.

Extensive communication between divers at each depth, along with detailed notes collected on

visual identities of fishes during video analysis were employed to ensure no pseudo-replication

occurred by filming the same fish multiple times.

Behavioral observations were categorized into six behavioral traits; foraging, swimming,

aggressor, victim, courtship, or other. Foraging behaviors were classified as fish actively grazing

on substrate. Swimming was classified as fish actively moving. Aggression was divided into

two categories, an aggressor (focal fish actively chasing another fish) or victim (focal fish

actively being chased by another fish). Courtship behaviors included spawning behaviors and

territorial patrolling (swimming >2m above substrate in a repetitive pattern). The "other"

category included any additional behaviors not previously outlined, such as time spent at

cleaning stations.

3.3.4 Environmental Parameters

We deployed five HOBO Pendant Temperature/Light data loggers (UA-002-64) for long term data recording at one site (Kona Paradise, southern region, leeward Hawaiʻi Island) in 2013-2014. One data logger was placed on the benthos at each of five depths: 3, 10, 20, 30, and 40m. Data loggers recorded temperature (with errors of about 0.1°C) every 25 minutes from August 2013 to February 2014. Loggers were inspected, cleaned, and had data extracted every three weeks for the six-month period. Owing to biofouling, long-term irradiance measurements were unreliable, and were discarded. After 3 months, a large swell dislodged and removed the 10m sensor, so we have excluded this depth from our analysis.

Surface climate data were accessed via the National Oceanographic and Atmospheric Administration's (NOAA) National Climate Data Center (NCDC; www.ncdc.noaa.gov). We utilized NCDC's Kona, HI weather station dataset as they were the closest in proximity to our research site in Kaohe Bay, HI. Daily weather information was downloaded from NCDC's monthly climate data portal from October 2013 through January 2014. Offshore wind speed (OWS) estimates were generated for each day from October 2013 to January 2014 using the equation OWS=average daily wind speed*cos((direction of wind-260)*π/180). Alongshore wind speed (AWS) estimates were generated for the same time frame using the equation AWS=average daily wind speed*cos((direction of wind-170)*π/180).

3.3.5 Statistical Analyses

Analyses of reef fish densities were calculated using generalized linear models (GLMs) based on a Poisson distribution (two factors: depth zone and island). During these analyses, we removed two transects from Niʻihau MCE depths as these were observed to be extreme outliers resulting from one species, *Luzonichthys earlei*, having anomalously high abundances (>2000

fish/transect). This species was not found on any other surveys or any other islands (ESM 1). Focal observations of *Z. Flavescens* and *S. psittacus* were also compared using GLMs based on Gaussian distributions. GLM statistical analyses were conducted using JMP statistical software (JMP 12.1.0, SAS Institute Inc.).

Analyses of fish trophic densities with depth were conducted using nonparametric quantile regression spline of 0.90 quantile using the function rq() from the *quantreg* package for R statistical software (v. 3.2.2). This method is useful for understanding relationships between depth and fish densities when relationships show heterogeneous and non-linear scatter without making assumptions of fish density shape. The models were constructed using the bs() function in the R package *splines* to find the appropriate degree and fit for the polynomial as well as determining maximum density values and 90% confidence intervals for each trophic group. See Fukunaga et al. (2016) for specific details on statistical methodology and R code.

Species-specific analyses were conducted using kernel density estimates (KDEs) to compare fish length-frequency distributions between shallow (0-30m) and mesophotic (>30m) depths (Salgado-Ugarte et al. 2000; Langlois et al. 2012). Kernel density estimates and appropriate bandwidths for each of the five selected fish species were generated using methodology outlined in Langlois et al. (2012). Additionally, we created reference bands for equality for each species using the *sm.density.compare* function in R. Assuming a null model, the reference bands give the mean KDE +/- one standard error. Areas where distribution curves lie outside the reference bands indicate differences in sizes between the two depth zones (shallow vs. MCE). Sample sizes and Interquartile Ranges are provided to describe sample size and spread of the distribution (Table 2).

3.4 Results

3.4.1 Propagule Refugia

Across all islands and depths, we counted a total of 291,615 fish from 228 species within 2,272 fish surveys (ESM 1). Sampling effort varied by island and depth (Table 1). Most fish species were found to be depth generalists and ranged from shallow to upper mesophotic reef depths. In total, 17.8% of species encountered were found only in <30m depth while 70.1% of species encountered were found from 0-60m depth. Approximately 36% of species observed were found along the entire MCE range from 0-100m. Only 8.6% of fish species were deep reef specialists and found only below 30m. Fish density was found to be significantly different between depth zones (Shallow vs. MCE) and marginally significant between islands or island x depth interaction (Fig 2: GLM: depth zone $\chi^2_{1,1286} = 6.86$, $P= 0.009$; island $\chi^2_{5,1286} = 1.74$, $P =0.057$; depth zone x island $\chi^2_{5,1286} = 1.06$, $P =0.051$). Marginal significance between islands likely results from differences in overall fish abundance between islands, with Hawaiʻi Island having the highest densities of shallow reef fishes and Niʻihau having high densities of fish in both shallow and MCE depths. Marginal significance in depth x island interaction resulted from significant differences between Niʻihau and all other islands, as Niʻihau had higher densities of fishes at MCE depths as compared to shallow depths. With the exception of Niʻihau, all other islands exhibited the same trend of higher densities of fish at shallow depths and lower densities at MCE depths.

Reef fish were subdivided by trophic group for analysis. Densities with each trophic designation f ollowed in accordance with overall fish densities with the majority of trophic groups attaining maximum densities in shallow (<30m) depths (Fig 3). Corallivorous fishes attained maximum density at 18.3m (90% CI 16.3-20.9m) and extended only into upper mesophotic depths (Fig. 3a). Herbivorous fishes reached a maximum at 22.2m (90% CI 16.7-

24.9m) and declined continuously into mesophotic depths (Fig. 3b). Invertivores were consistently observed among all depths (Fig. 3c) but attained a maximum density within upper mesophotic habitats at 41.1m (90% CI 33.7-49.1). Piscivorous fishes displayed no reliable maximum density (Fig. 3d) and were the least abundant trophic group. Planktivorous fishes also peaked within shallow depths (Fig. 3e) with a maximum at 26.3m (90% CI 24.8-28.3). Although planktivores peaked within shallow waters, they were some of the most common fishes at mesophotic depths along with invertivores. Omnivorous fishes were rare at all depths observed and 90% CI could not be determined due the low observation records of this trophic group.

Given declines in the density of herbivorous fishes with depth, we were interested in determining whether there were behavioral shifts associated with these declines. We noted two sites on Hawaii Island that contained higher abundances of herbivorous fishes at MCE depths than shallow depths. Closer inspections revealed the increased abundances of herbivores at depth resulted from high densities of one species of parrotfish, *Scarus psittacus* (Fig. 4). Focal observational surveys of two species of herbivorous fishes at these sites yielded distinct differences in behavior between species. *Zebrasoma flavescens* exhibited similar behaviors overall between shallow and deep sites, with fishes spending an average of 51% of time foraging and 46% of time swimming at shallow depths, versus 49% of time foraging and 50% swimming at mesophotic depths. Site-based differences in foraging included higher foraging at the Old Airport site (Fig. 5a; GLM: depth $t_{1,40} = 0.49$, P =0.62; site $t_{1,40} = 2.64$, P =0.009; depth x site $t_{1,40} = 1.56$, P =0.12) Swimming behaviors were found to be significantly higher at the Puako site (Fig. 5a; GLM: depth $t_{1,40} = 0.76$, P =0.45; site $t_{1,40} = 2.32$, P =0.02; depth x site $t_{1,40} = 1.85$, P =0.06). Differences in aggressor behaviors were also found between sites, with slightly higher aggressive acts at the Puako site(Fig. 5a; GLM: depth $t_{1,40} = 1.77$, P =0.08; site $t_{1,40} = 2.24$, P

=0.03; depth x site $t_{1,40}$ = 1.3, P =0.2) No significant differences were found between depths for

foraging, swimming, or aggressor behaviors. Differences were observed among both site and

depth in victim behaviors (Fig. 5a; GLM: depth $t_{1,40}$ = 2.8, P =0.006; site $t_{1,40}$ = 2.21, P =0.03;

depth x site $t_{1,40}$ = 1.91, P =0.06) with more fishes being chased in shallow waters than deeper

waters. No significant differences were found among either sites or depths for courtship

behaviors, as they were rarely observed (Fig. 5a; GLM: depth $t_{1,40}$ = 0.58, P =0.57; site $t_{1,40}$ = 0.1,

P =0.92; depth x site $t_{1,40}$ = 0.75, P =0.45).

Observations of focal individuals of *S. psittacus* revealed behaviors that strongly deviated

from those observed in *Z. flavescens*. Mesophotic depths were used almost exclusively for

courtship, comprising 93% of observed time dislaying courtship behaviors, and less than 1%

foraging or swimming. Conversely, *S. psittacus* were largely found swimming at shallow depths,

spending approximately 60% of their time moving around shallow reefs, while spending

approximately 15% of time foraging (Fig. 5b; GLM: courtship $\chi^2_{1,40}$= 3084.09, P ≤ 0.001;

swimming $\chi^2_{1,40}$ = 5159.83, P ≤ 0.001). *Scarus psittacus* also maintained significantly higher

foraging rates in shallow waters depths (Fig. 5b; GLM: $\chi^2_{1,40}$= 1074.45, P ≤ 0.001). Aggressive

interactions were not significantly different between depths and were not commonly observed

(Fig. 5b; GLM: aggressor $\chi^2_{1,40}$= 1.67, P =0.2; aggresse $\chi^2_{1,40}$= 0.21, P =0.65). These

observations indicate *S. psittacus* uses certain MCE areas almost exclusively as courtship and

spawning grounds.

We selected four fish species to more closely examine size-frequency relationships

between shallow and mesophotic depths (Table 2). *Chlorurus spilurus* and *Scarus psittacus*

displayed similar trajectories between shallow and mesophotic depths (Fig.6a, b). Both parrotfish

species showed higher densities of small sized fishes than expected in the null model (grey

bands) while fishes in mesophotic depths displayed lower densities than expected. Conversely, a shift occurred at larger size ranges in both parrotfish species, where larger individuals had higher densities in mesophotic depths than expected in the null model, while large-bodied fishes in shallow depths had lower densities than expected. *Zebrasoma flavescens*, a highly prized aquarium fish species, followed similar KDE trajectories as the parrotfish (Fig. 6c). *Zebrasoma flavescens* occurred in higher densities at smaller size ranges in shallow waters, while in mesophotic depths densities peaked at larger sizes. Lastly, we investigated *Thalassoma duperrey*, an extremely common but unfished species in Hawaiian waters and found similar patterns as the above fished species (Fig. 6d). KDE analyses indicate higher densities of small sized fishes than expected of *T. duperrey* at shallow depths and lower than expected densities at mesophotic depths, while fishes were found to have higher densities than expected in the null model at mesophotic depths for the upper size ranges, with shallow fishes displaying lower densities than expected at the larger size ranges.

3.4.2 Environmental Refugia

Water temperature was recorded at five depths (3, ~~10~~, 20, 30, 40m) at Kahoe Bay, Hawaiʻi, from August 2013 to February 2014. Temperatures were stratified by depth during summer and fall seasons, with shallow temperatures consistently higher and more variable (Fig. 7a). By November 2013, stratification was lost and temperatures remained uniform from 3-40m depth through February 2014. In general, temperatures at 3m were predominantly driven by daily heating/cooling near the surface, with one peak in temperature per day. Conversely, temperature recordings at 20-40m depths sometimes revealed roughly two temperature peaks per day, indicating that temperature fluctuations at these depths sometimes resulted from internal semidiurnal tides, rather than daytime warming (Fig.7b).

Two anomalous temperature events occurred during this study. One event occurred during mid-October, and another event occurred in late January 2014. During both of these events, temperatures decreased dramatically at 30-40m depths. Temperature anomalies during the first event were likely the result of periods of sustained offshore winds that resulted in upwelling (Fig. 8a,b). The trend of shallow (3m) depths maintaining one temperature maximum per day while 20-40m depths maintained two temperature maxima per day was consistent for the entire length of our study (6 months). The upwelling event that initiated on October 20, 2013, revealed that 20-40m depths decreased more than 1℃ during the event, and 30-40m temperatures were suppressed for approximately 3 days before reverting to normal temperatures. Shallow temperatures (3m) started dropping below underlying temperatures, suggesting that some salinity stratification may have developed during the event (Fig. 8).

The second event of interest occurred from January 15 to 18, 2014 (Fig. 9a,b). This event shows evidence of internal tides propagating up the reef slope, all the way to the shallowest temperature sensor (3m). Upslope propagation is observed with earliest records of cold water at deep sensors, and successive movement of cold water with time upslope to shallow sensors. Strong internal tides resulted in cold water moving farther up the slope than normally occurs. During this time, temperature fluctuations begin syncing along the entire 3-40m gradient and the effect of daily heating on shallow reefs is replaced by effects of internal tide propagation.

3.5 Discussion

In order to consider mesophotic reefs as climate change or fishing refugia for shallow reef fishes, MCEs need to both harbor sufficient populations of species adversely impacted in shallow reefs as well as provide sources for recolonization of shallow reef areas (Bridge and Guinotte 2012; Pyle et al. 2016). The high amount of species overlap found in the main

Hawaiian Islands between shallow and upper mesophotic reefs (30-60m) supports the notion of MCEs as potential refugia. This follows in accordance with previous work both in Hawaiʻi as well as the Western Pacific, Caribbean, Brazil, and Red Sea where high species overlap existed between 0 and 50m (Brokovich et al. 2008; Bejarano et al. 2014; Andradi-Brown et al. 2016; Fukunaga et al. 2016; Rosa et al. 2016; Kane and Tissot 2017).

We also documented the first evidence of parrotfishes utilizing mesophotic depths as courtship and spawning regions. Previous studies have noted the usage of MCEs as spawning grounds for groupers in the Caribbean (Starr et al.; Rowell et al. 2015; Pickard et al. 2016). Sedberry et al. (2006) noted that shelf edge reefs (40-60m) off South Carolina were particularly important as spawning grounds for several species of porgies, sea bass, grouper, gag, scamp, snapper, and triggerfish. We observed spawning of two common parrotfish species at two of twelve sites on Hawaiʻi Island. These spawning assemblages were temporally brief but consistent as they occurred within the same month for each of the three years visited (C. Kane pers. obs.). Combined observations and behavioral studies indicate that these parrotfishes utilize deep reefs for short periods of time for spawning aggregations, then return to shallow waters. These deep spawning events support the Deep Reefs as Refugia Hypothesis (DRRH) as potential deep sources of propagules. These observations provide initial evidence that MCEs in the Hawaiian Islands may act as spawning grounds for shallow reef fishes and provide an opportunity to further explore potential spawning grounds on other islands. Further support of DRRH for connectivity between shallow and mesophotic depths lies in our inability to study female parrotfishes during behavioral observations as females were found to swim rapidly and consistently up and down the reef slope between shallow and mesophotic depths. While we were unable to collect data on female behavior due to diver safety (limitations on moving quickly

from deep to shallow, or repeatedly moving up and down the reef), this limitation in itself lends evidence to support active movement of fishes between shallow and mesophotic depths. We were able to collect data on terminal males as they formed small territorial ranges at mesophotic depths and hence did not move quickly between depths.

Genetic connectivity studies of fishes in Hawaiian waters further support observational evidence from this and other studies. Although connectivity work in reef fishes between shallow and mesophotic depths is limited, the few studies to date indicate fishes found on shallow and mesophotic reefs are not genetically distinct. Connectivity studies of 13 reef fish species collected across the Hawaiian Islands reveal little to no genetic structuring between shallow and mesophotic reefs (J. Copus et al., in review). Tenggardjaja et al. (2014) found no genetic structure in damselfish at Johnston Atoll from 30-150m. Additionally, Tenggardjaja et al. (2016) found population structuring between the Northwestern and Main Hawaiian Islands, but no strong structuring within the Main Hawaiian Islands in three endemic damselfishes. Therefore, connectivity of reef fishes between shallow and mesophotic environments yields strong evidence for DRRH as uniform population structure indicates that fishes found at MCEs are genetically connected with shallow populations and hence potential sources of propagules to re-seed shallow reef environments. The mobility of reef fishes to actively or passively move between shallow and mesophotic environments leads to much greater probability of these fishes re-seeding shallow reefs as compared to more sessile or dispersal-limited organisms (Kahng et al. 2014; Loya et al. 2016).

Our study also supports MCEs as being probable environmental refugia. There is temperature stratification at Hawaiʻi Island between 3-40m depths during summer seasons, with shallow waters consistently warmer than mesophotic waters. However, the magnitude of

99

difference in temperatures is small (~1°C) and may not be sufficient to preclude bleaching during thermal stress events. Yet it is during this time (late summer, early fall) that bleaching is most likely to occur in the Hawaiian Islands, so it is possible even small temperature differences between shallow and mesophotic depths may alleviate bleaching potential for deeper corals (Smith et al. 2013; Couch et al. 2017). In late fall, stratification collapsed and we observed near uniform temperatures from 3-40m which lasted for the remaining duration of our study (3 months). Several anomalous events occurred during the study that are also consistent with the environmental refugia hypothesis. Strong offshore winds provided upwelling that reduced mesophotic and shallow water temperatures for up to a week following the upwelling event. Additionally, the 20-40m temperature stations recorded twice-daily peaks in temperature, indicating that water temperature at these depths were primarily driven by internal tide fluctuations and not daily heating/cooling as was demonstrated at 3m depths where temperature peaked once per day. Periods of strong internal tides also accounted for upslope propagation of cooler waters into shallow depths. Additional studies off Hawaiʻi Island reveal similar trends in temperature reductions with depth (Brill et al. 1999). Further evidence that internal waves and tides cause temperature fluctuations between shallow and mesophotic depths have been documented at Maui (Kahng and Kelley 2007), Oʻahu (Smith et al. 2016a), and at the offshore Station ALOHA (Campbell and Vaulot 1993). Previous studies have indicated effects of internal waves and internal tides reducing water temperatures at shallow depths across Pacific and Caribbean waters (Leichter et al. 2005; Leichter and Genovese 2006; Jantzen et al. 2013; Smith et al. 2016b). Evidence of internal tides and waves in the Hawaiian Islands, coupled with lower overall observed temperatures at mesophotic depths provide are consistent with the DRRH for mesophotic reefs acting as an environmental or thermal refugia.

Benefits from thermal stability in MCEs to date have not been explored for coral reef fishes yet there is anecdotal evidence that fishes may retreat to deeper waters during thermal stress events in shallow reefs. Such studies would yield important evidence for conservation and management under future climate change scenarios. Conversely, coral colonies may not benefit from thermal stability at mesophotic depths as studies show greater bleaching and stress of mesophotic corals and an inability of MCEs to adequately buffer extreme or prolonged shallow thermal stress events (Neal et al. 2013; Smith et al. 2016d).

The support of DRRH in reference to reef fishes outlined above is not universal. While there is evidence of large overlap in species composition, substantial overlap only occurs in the upper 20-30m of the mesophotic zone (upper mesophotic = 30-60m). Below 50-60m, species overlap with shallow reefs is much lower – in our study, shallow reef fish overlap was reduced from >70% in upper MCEs to 36% at lower MCE depths (60-100m). This pattern is supported by studies which found distinct species compositions that were either endemic species or deep-specialists at depths below 60m (Kosaki et al. 2016; Loya et al. 2016; Pinheiro et al. 2016). Additionally, both density and diversity declines with depth, leaving more limited potential for replenishment to shallow reefs (Brokovich et al. 2008; Fukunaga et al. 2016; Asher et al. 2017b; Kane and Tissot 2017).

Perhaps the most prominent evidence against DRRH for reef fishes is the shift in trophic assemblages and trophic dominance with depth. Corallivorous and herbivorous fishes were found to decline significantly with depth across the Main Hawaiian Islands. This is supported by further work in the Hawaiian Islands with precipitous declines in herbivorous fishes with depth across the Hawaiian Archipelago (Bridge et al. 2016; Fukunaga et al. 2016; Pyle et al. 2016; Fukunaga and Kosaki 2017; Kane and Tissot 2017; Kane et al., in review). Mesophotic

environments are dominated largely by invertivore and planktivore species, a pattern consistent in mesophotic environments in most major ocean basins (Brokovich et al. 2010; Garcia-Sais 2010; Bejarano et al. 2014; Andradi-Brown et al. 2016; Bridge et al. 2016; Kane and Tissot 2017). Invertivore and planktivore species are largely small-bodied and not of high commercial or recreational fishing importance, indicating that replenishment of these species from deep reefs may promote biodiversity but likely will not have positive effects for commercial or recreational reef fish fisheries.

The near absence of herbivorous fishes at depth greatly limits both the ecological and economic potential of mesophotic reefs as refugia for these coral reef fishes. As ecologically important species for shallow coral reefs, herbivorous fishes play an integral role in coral maintenance and recovery (Ogden and Lobel 1978; Heenan and Williams 2013; Adam et al. 2015; Hixon 2015). These fishes are also heavily fished for commercial and recreational reef fisheries and are some of the most prized reef fishes for consumption in the Hawaiian Islands (Weijerman et al. 2013; Gray et al. 2016). The disparity in behavioral patterns between two herbivorous fish species in Hawaiʻi (*Z. flavescens* and *S. psittacus*) indicate that mesophotic reefs are not preferential habitat for these species. *Zebrasoma flavescens* abundances declined significantly with depth (Bogeberg et al., in review), yet those few that were found at MCE depths exhibited similar behaviors as shallow congeners. Foraging observations indicate that MCEs are potential foraging grounds yet are just not utilized often by *Z. flavescens* (Kane et al., in review). Although we observed deep spawning grounds for two species of parrotfish in Hawaiʻi Island, these are likely temporary migrations to deep waters while they predominantly live in shallow waters. It is likely that herbivorous fishes decline with depth as a result of predation threats and are not limited by resource availability or quality (Kane et al., in review).

Thus, while mesophotic reefs may provide critical habitat for spawning aggregations, they will not act as refugia from fishing pressure for herbivorous fishes.

When assessing the size structure of four shallow reef fish species, we found that fish at mesophotic depths are generally at the larger ends of their size ranges and lack smaller sized individuals. During our mesophotic surveys, we rarely encountered recruits or juveniles of shallow fish species and typically observed only fish that were sub-adult or adult stages. The higher than expected density probabilities of small-sized fish in shallow waters further indicates that these fish are recruiting to shallow reefs, not deep reefs. This trend was consistent across all four species of fish investigated individually (2 species heavily targeted for consumption, 1 species targeted for aquarium collection, and 1 species not fished), indicating that greater density probabilities of fish in mesophotic depths is not necessarily a result of fishing pressure. There has been some evidence of mesophotic fishing refugia in the Marianas Islands (Lindfield et al. 2014b, 2016), but our data do not support this trend as herbivorous fishes were rare at MCE depths and piscivorous fishes had low but consistent densities across depths.

3.6 Conclusions

These findings provide evidence that mesophotic reefs may act as refugia for coral reef fishes, but in a limited capacity. Evidence supports deep reefs as critical habitat for spawning aggregations of some shallow fishes and biodiversity reservoirs for shallow species within upper mesophotic depths (30-60m). Previously published genetic analyses indicate population connectivity between shallow and mesophotic reefs thus supporting predictions of deep reefs as potential sources for propagules. Unfortunately, the reduction in the density of herbivorous fishes with depth indicates MCEs are not likely to facilitate the recovery of coral-dominated reefs via herbivorous fish re-seeding following disturbances or restock valuable fisheries species upon

depletion in shallow reefs. Replenishment of shallow reef fishes from deep reefs is plausible, although the replenished reef fish communities are not likely to look like current fish communities. Dominance of invertivore and planktivore fish species at MCE depths may be beneficial for restoring biodiversity in depauperate shallow reef systems, but these species largely lack economic value for shallow reef systems. While MCEs may not serve as refugia for certain taxa from climate or anthropogenic impacts, they still deserve consideration as essential habitat for spawning migrations and as biodiversity conservation. Additionally, knowledge of these systems, especially with respect to reef fishes, is still in its infancy. Directed studies further investigating ecological, physiological and genetic connectivity of reef fishes between shallow and mesophotic depths is essential to further evaluate refugia hypotheses and manage fish communities for future climate change conditions. We have provided initial evidence evaluating DRRH from which future studies can build and shed more definitive light on the interplay between shallow and mesophotic reefs with regards to coral reef fishes.

3.7 Acknowledgments

3.8 References

Adam TC, Burkepile DE, Ruttenberg BI, Paddack MJ (2015) Herbivory and the resilience of Caribbean coral reefs : knowledge gaps and implications for management. Mar. Ecol. Prog. Ser. 520:1–20

Albright R, Caldeira L, Hosfelt J, Kwiatkowski L, Maclaren JK, Mason BM, Nebuchina Y, Ninokawa A, Pongratz J, Ricke KL, Rivlin T, Schneider K, Sesboüé M, Shamberger K, Silverman J, Wolfe K, Zhu K, Caldeira K (2016) Reversal of ocean acidification enhances net coral reef calcification. Nature 531:362–365

Andradi-Brown DA, Gress E, Wright G, Exton DA, Rogers AD (2016) Reef fish community biomass and trophic structure changes across shallow to upper- Mesophotic reefs in the mesoamerican barrier reef, Caribbean. PLoS One 11:1–19

Asher J, Williams ID, Harvey ES (2017a) An Assessment of Mobile Predator Populations along Shallow and Mesophotic Depth Gradients in the Hawaiian Archipelago. Sci. Rep. 7:3905

Asher J, Williams ID, Harvey ES (2017b) Mesophotic Depth Gradients Impact Reef Fish Assemblage Composition and Functional Group Partitioning in the Main Hawaiian Islands. Front. Mar. Sci. 4:

Bejarano I, Appeldoorn RS, Nemeth M (2014) Fishes associated with mesophotic coral ecosystems in La Parguera, Puerto Rico. Coral Reefs 33:313–328

Bongaerts P, Carmichael M, Hay KB, Tonk L, Frade PR, Hoegh-guldberg O (2015) Prevalent endosymbiont zonation shapes the depth distributions of scleractinian coral species Subject Category : Subject Areas : R. Soc. Open Sci.

Bongaerts P, Frade PR, Ogier JJ, Hay KB, Van Bleijswijk J, Englebert N, Vermeij MJ, Bak RP, Visser PM, Hoegh-Guldberg O (2013) Sharing the slope: Depth partitioning of agariciid corals and associated Symbiodinium across shallow and mesophotic habitats (2-60 m) on a Caribbean reef. BMC Evol. Biol. 13:

Bongaerts P, Ridgway T, Sampayo EM, Hoegh-Guldberg O (2010) Assessing the "deep reef refugia" hypothesis: focus on Caribbean reefs. Coral Reefs 29:309–327

Bongaerts P, Riginos C, Hay K, van Oppen M, Hoegh-Guldberg O, Dove S (2011) Adaptive divergence in a scleractinian coral : physiological adaptation of Seriatopora hystrix to shallow and deep reef habitats. BMC Evol. Biol. 11:1–14

Bridge T, Guinotte J (2012) Mesophotic Coral Reef Ecosystems in the Great Barrier Reef World Heritage Area in the Great Barrier Reef World.

Bridge TCL, Done TJ, Beaman RJ, Friedman a., Williams SB, Pizarro O, Webster JM (2010) Topography, substratum and benthic macrofaunal relationships on a tropical mesophotic

shelf margin, central Great Barrier Reef, Australia. Coral Reefs 30:143–153

Bridge TCL, Hughes TP, Guinotte JM, Bongaerts P (2013) Call to protect all coral reefs. Nat. Clim. Chang. 3:528–530

Bridge TCL, Luiz OJ, Coleman RR, Kane CN, Kosaki RK (2016) Ecological and morphological traits predict depth-generalist fishes on coral reefs. Proc. R. Soc. B 283:

Brill RW, Block BA, Boggs CH, Bigelow KA, Freund E V., Marcinek DJ (1999) Horizontal movements and depth distribution of large adult yellowfin tuna (Thunnus albacares) near the Hawaiian Islands, recorded using ultrasonic telemetry: implications for the physiological ecology of pelagic fishes. Mar. Biol. 133:395–408

Brokovich E, Ayalon I, Einbinder S, Segev N, Shaked Y, Genin A, Kark S, Kiflawi M (2010) Grazing pressure on coral reefs decreases across a wide depth gradient in the Gulf of Aqaba, Red Sea. Mar. Ecol. Prog. Ser. 399:69–80

Brokovich E, Einbinder S, Shashar N, Kiflawi M, Kark S (2008) Descending to the twilight-zone: changes in coral reef fish assemblages along a depth gradient down to 65 m. Mar. Ecol. Prog. Ser. 371:253–262

Campbell L, Vaulot D (1993) Photosynthetic picoplankton community structure in the subtropical North Pacific Ocean near Hawaii (station ALOHA). Deep Sea Res. Part I Oceanogr. Res. Pap. 40:2043–2060

Carpenter KE, Abrar M, Aeby G, Aronson RB, Banks S, Bruckner A, Chiriboga A, Cortés J, Delbeek JC, Devantier L, Edgar GJ, Edwards AJ, Fenner D, Guzmán HM, Hoeksema BW, Hodgson G, Johan O, Licuanan WY, Livingstone SR, Lovell ER, Moore J a, Obura DO, Ochavillo D, Polidoro B a, Precht WF, Quibilan MC, Reboton C, Richards ZT, Rogers AD, Sanciangco J, Sheppard A, Sheppard C, Smith J, Stuart S, Turak E, Veron JEN, Wallace C, Weil E, Wood E (2008) One-third of reef-building corals face elevated extinction risk from climate change and local impacts. Science 321:560–3

Chollett I, Mumby PJ (2013) Reefs of last resort: Locating and assessing thermal refugia in the wider Caribbean. Biol. Conserv. 167:179–186

Christie MR, Tissot BN, Albins M a, Beets JP, Jia Y, Ortiz DM, Thompson SE, Hixon M a (2010) Larval connectivity in an effective network of marine protected areas. PLoS One 5:e15715

Couch CS, Burns JHR, Liu G, Steward K, Gutlay TN, Kenyon J, Eakin CM, Kosaki RK (2017) Mass coral bleaching due to unprecedented marine heatwave in Papahānaumokuākea Marine National Monument (Northwestern Hawaiian Islands). PLoS One 12:1–27

Eyre BD, Cyronak T, Drupp P, De Carlo EH, Sachs JP, Andersson AJ (2018) Coral reefs will transition to net dissolving before end of century. Science 359:908–911

Friedlander A, Sandin S, DeMartini E, Sala E (2010) Spatial patterns of the structure of reef fish assemblages at a pristine atoll in the central Pacific. Mar. Ecol. Prog. Ser. 410:219–231

Fukunaga A, Kosaki RK (2017) Use of multivariate control charts to assess the status of reef fish assemblages in the Northwestern Hawaiian Islands. PeerJ 5:e3651

Fukunaga A, Kosaki RK, Wagner D, Kane C (2016) Structure of mesophotic reef fish assemblages in the Northwestern Hawaiian Islands. PLoS One 11:1–15

Garcia-Sais JR (2010) Reef habitats and associated sessile-benthic and fish assemblages across a euphotic–mesophotic depth gradient in Isla Desecheo, Puerto Rico. Coral Reefs 29:277–288

Glynn P (1996) Coral reef bleaching: facts, hypotheses and implications. Glob. Chang. Biol. 2:495–509

Gray AE, Williams ID, Stamoulis KA, Boland RC, Lino KC, Hauk BB, Leonard JC, Rooney JJ, Asher JM, Lopes KH, Kosaki RK (2016) Comparison of reef fish survey data gathered by open and closed circuit SCUBA divers reveals differences in areas with higher fishing pressure. PLoS One 11:1–18

Heenan A, Williams I, DesRochers A, Kanemura T, Kosaki R, Nadon M, Brainard R (2017) Long-term monitoring of coral reef fish assemblages in the Western Central Pacific. Sci. Data 4:1–12

Heenan A, Williams ID (2013) Monitoring herbivorous fishes as indicators of coral reef resilience in American Samoa. PLoS One 8:e79604

Hinderstein LM, Marr JC a., Martinez F a., Dowgiallo MJ, Puglise K a., Pyle RL, Zawada DG, Appeldoorn R (2010) Theme section on "Mesophotic Coral Ecosystems: Characterization, Ecology, and Management." Coral Reefs 29:247–251

Hoegh-Guldberg O, Bruno JF (2010) The impact of climate change on the world's marine ecosystems. Science 328:1523–8

Hoey A, Howells E, Johansen J, Hobbs J-P, Messmer V, McCowan D, Wilson S, Pratchett M (2016) Recent Advances in Understanding the Effects of Climate Change on Coral Reefs. Diversity 8:12

Holstein DM, Paris CB, Vaz AC, Smith TB (2016) Modeling vertical coral connectivity and mesophotic refugia. Coral Reefs 35:23–37

Holstein DM, Smith TB, Gyory J, Paris CB (2015) Fertile fathoms: Deep reproductive refugia for threatened shallow corals. Sci. Rep. 5:1–12

van Hooidonk R, Maynard JA, Planes S (2013) Temporary refugia for coral reefs in a warming world. Nat. Clim. Chang. 3:508–511

Hughes TP, Baird AH, Bellwood DR, Card M, Connolly SR, Folke C, Grosberg R, Hoegh-Guldberg O, Jackson JBC, Kleypas J, Lough JM, Marshall P, Nyström M, Palumbi SR, Pandolfi JM, Rosen B, Roughgarden J (2003) Climate Change, Human Impacts, and the Resilience of Coral Reefs. Science 301:929 LP-933

Hughes TP, Kerry JT, Álvarez-Noriega M, Álvarez-Romero JG, Anderson KD, Baird AH, Babcock RC, Beger M, Bellwood DR, Berkelmans R, Bridge TC, Butler IR, Byrne M, Cantin NE, Comeau S, Connolly SR, Cumming GS, Dalton SJ, Diaz-Pulido G, Eakin CM, Figueira WF, Gilmour JP, Harrison HB, Heron SF, Hoey AS, Hobbs J-PA, Hoogenboom MO, Kennedy E V., Kuo C, Lough JM, Lowe RJ, Liu G, McCulloch MT, Malcolm HA, McWilliam MJ, Pandolfi JM, Pears RJ, Pratchett MS, Schoepf V, Simpson T, Skirving WJ, Sommer B, Torda G, Wachenfeld DR, Willis BL, Wilson SK (2017) Global warming and recurrent mass bleaching of corals. Nature 543:373–377

Hughes TP, Tanner JE (2000) RECRUITMENT FAILURE, LIFE HISTORIES, AND LONG-TERM DECLINE OF CARIBBEAN CORALS. Ecology 81:2250–2263

Jantzen C, Schmidt GM, Wild C, Roder C, Khokiattiwong S, Richter C (2013) Benthic reef primary production in response to large amplitude internal waves at the Similan Islands (Andaman Sea, Thailand). PLoS One 8:e81834

Jones GP, McCormick MI, Srinivasan M, Eagle J V (2004) Coral decline threatens fish biodiversity in marine reserves. Proc. Natl. Acad. Sci. U. S. A. 101:8251–3

Kahng S, Copus J, Wagner D (2014) Recent advances in the ecology of mesophotic coral ecosystems (MCEs). Curr. Opin. Environ. Sustain. 7:72–81

Kahng SE, Kelley CD (2007) Vertical zonation of megabenthic taxa on a deep photosynthetic reef (50–140 m) in the Au'au Channel, Hawaii. Coral Reefs 26:679–687

Kane C, Kosaki R, Wagner D (2014) High levels of mesophotic reef fish endemism in the Northwestern Hawaiian Islands. Bull. Mar. Sci. 90:1–12

Kane CN, Tissot BN (2017) Trophic designation and live coral cover predict changes in reef-fish community structure along a shallow to mesophotic gradient in Hawaii. Coral Reefs 1–11

Kelly ELA, Eynaud Y, Clements SM, Gleason M, Sparks RT, Williams ID, Smith JE (2016) Investigating functional redundancy versus complementarity in Hawaiian herbivorous coral reef fishes. Oecologia 182:1151–1163

Kosaki RK, Pyle RL, Leonard JC, Hauk BB, Whitton RK, Wagner D (2016) 100% endemism in mesophotic reef fish assemblages at Kure Atoll, Hawaiian Islands. Mar. Biodivers.

Langlois TJ, Fitzpatrick BR, Fairclough D V., Wakefield CB, Hesp SA, McLean DL, Harvey ES, Meeuwig JJ (2012) Similarities between Line Fishing and Baited Stereo-Video Estimations of Length-Frequency: Novel Application of Kernel Density Estimates. PLoS

One 7:1–9

Laverick JH, Andradi-Brown DA, Exton DA, Bongaerts P, Bridge TCL, Lesser MP, Pyle RL, Slattery M, Wagner D, Rogers AD (2016) To what extent do mesophotic coral ecosystems and shallow reefs share species of conservation interest? Environ. Evid. 5:16

Leichter J, Deane G, Stokes M (2005) Spatial and temporal variability of internal wave forcing on a coral reef. J. Phys. Oceanogr. 35:1945–1962

Leichter J, Genovese S (2006) Intermittent upwelling and subsidized growth of the scleractinian coral Madracis mirabilis on the deep fore-reef slope of Discovery Bay, Jamaica. Mar. Ecol. Prog. Ser. 316:95–103

Leis JM (2018) Paradigm Lost: Ocean Acidification Will Overturn the Concept of Larval-Fish Biophysical Dispersal. Front. Mar. Sci. 5:47

Lesser MP, Slattery M, Leichter JJ (2009) Ecology of mesophotic coral reefs. J. Exp. Mar. Bio. Ecol. 375:1–8

Liddell WD, Ohlhorst SL (1988) Substrata Community Patterns , 1-120 M, North Jamaica. Palaios 3:413–423

Liddell WD, Ohlhorst SL (1998) Hard substrata community patterns, 1-120m, North Jamaica. Palaios 3:413–423

Lindfield SJ, Harvey ES, Halford AR, McIlwain JL (2016) Mesophotic depths as refuge areas for fishery-targeted species on coral reefs. Coral Reefs 35:125–137

Lindfield SJ, McIlwain JL, Harvey ES (2014a) Depth refuge and the impacts of SCUBA spearfishing on coral reef fishes. PLoS One 9:e92628

Lindfield SJ, McIlwain JL, Harvey ES (2014b) Depth Refuge and the Impacts of SCUBA Spearfishing on Coral Reef Fishes. PLoS One 9:e92628

Loya Y, Eyal G, Treibitz T, Lesser MP, Appeldoorn R (2016) Theme section on mesophotic coral ecosystems: advances in knowledge and future perspectives. Coral Reefs 35:1–9

McClanahan TR, Graham NA, Darling ES (2014) Coral reefs in a crystal ball: predicting the future from the vulnerability of corals and reef fishes to multiple stressors. Curr. Opin. Environ. Sustain. 7:59–64

Muir P, Wallace C, Bridge TCL, Bongaerts P (2015) Diverse staghorn coral fauna on the mesophotic reefs of north-east Australia. PLoS One 10:1–17

Mumby PJ, Hastings A (2007) The impact of ecosystem connectivity on coral reef resilience. J. Appl. Ecol. 45:854–862

Munday PL, Jones GP, Pratchett MS, Williams AJ (2008) Climate change and the future for coral reef fishes. Fish Fish. 9:261–285

Neal BP, Condit C, Liu G, dos Santos S, Kahru M, Mitchell BG, Kline DI (2013) When depth is no refuge: cumulative thermal stress increases with depth in Bocas del Toro, Panama. Coral Reefs 33:193–205

Ogden JC, Lobel PS (1978) The role of herbivorous fishes and urchins in coral reef communities. Environ. Biol. Fishes 3:49–63

van Oppen MJH, Bongaerts P, Underwood JN, Peplow LM, Cooper TF (2011) The role of deep reefs in shallow reef recovery: an assessment of vertical connectivity in a brooding coral from west and east Australia. Mol. Ecol. 20:1647–60

Pandolfi JM, Jackson JBC, Baron N, Bradbury RH, Guzman HM, Hughes TP, Kappel C V, Micheli F, Ogden JC, Possingham HP, Sala E (2005) Are U.S. Coral Reefs on the Slippery Slope to Slime? Science 307:1725 LP-1726

Pickard AE, Vaudo JJ, Wetherbee BM, Nemeth RS, Blondeau JB, Kadison EA, Shivji MS (2016) Comparative Use of a Caribbean Mesophotic Coral Ecosystem and Association with Fish Spawning Aggregations by Three Species of Shark. PLoS One 11:e0151221

Pinheiro HT, Goodbody-Gringley G, Jessup ME, Shepherd B, Chequer AD, Rocha LA (2016) Upper and lower mesophotic coral reef fish communities evaluated by underwater visual censuses in two Caribbean locations. Coral Reefs 35:139–151

Pratchett MS, Wilson SK, Munday PL (2015) Effects of climate change on coral reef fishes. In: Mora C. (eds) Ecology of fishes on coral reefs. Cambridge University Press, pp 127–134

Pyle RL, Boland R, Bolick H, Bowen BW, Bradley CJ, Kane C, Kosaki RK, Langston R, Longenecker K, Montgomery A, Parrish FA, Popp BN, Rooney J, Smith CM, Wagner D, Spalding HL (2016) A comprehensive investigation of mesophotic coral ecosystems in the Hawaiian Archipelago. PeerJ 4:e2475

Riegl B, Piller WE (2003) Possible refugia for reefs in times of environmental stress. Int. J. Earth Sci. 92:520–531

Rocha L a., Craig MT, Bowen BW (2007) Phylogeography and the conservation of coral reef fishes. Coral Reefs 26:501–512

Rosa MR, Alves AC, Medeiros DV, Coni EOC, Ferreira CM, Ferreira BP, de Souza Rosa R, Amado-Filho GM, Pereira-Filho GH, de Moura RL, Thompson FL, Sumida PYG, Francini-Filho RB (2016) Mesophotic reef fish assemblages of the remote St. Peter and St. Paul's Archipelago, Mid-Atlantic Ridge, Brazil. Coral Reefs 35:113–123

Rowell TJ, Nemeth RS, Schärer MT, Appeldoorn RS (2015) Fish sound production and acoustic

telemetry reveal behaviors and spatial patterns associated with spawning aggregations of two Caribbean groupers. 518:239–254

Sale PF, Lavieren H Van, Lagman M, Butler M, Atema J, Fauvelot C, Hogan J, Jones GP, Lindeman K, Paris C, Steneck R, Stewart H (2010) Preserving Reef Connectivity A Handbook for Marine Protected Area Managers.

Salgado-Ugarte IH, Shimizu M, Taniuchi T, Matsushita K (2000) Size frequency analysis by averaged shifted histograms and kernel density estimators. Asian Fish. Sci. 13:1–12

Selkoe KA, Gaggiotti OE, Bowen BW, Toonen RJ (2014) Emergent Patterns of Population Genetic Structure for a Coral Reef Community. Mol. Ecol. n/a-n/a

Sinniger F, Morita M, Harii S (2012) "Locally extinct" coral species Seriatopora hystrix found at upper mesophotic depths in Okinawa. Coral Reefs 32:153–153

Slattery M, Lesser MP, Brazeau D, Stokes MD, Leichter JJ (2011) Connectivity and stability of mesophotic coral reefs. J. Exp. Mar. Bio. Ecol. 408:32–41

Smith KA, Rocheleau G, Merrifield MA, Jaramillo S, Pawlak G (2016a) Temperature variability caused by internal tides in the coral reef ecosystem of Hanauma bay, Hawai'i. Cont. Shelf Res. 116:1–12

Smith KA, Rocheleau G, Merrifield MA, Jaramillo S, Pawlak G (2016b) Temperature variability caused by internal tides in the coral reef ecosystem of Hanauma bay, Hawai'i. Cont. Shelf Res. 116:1–12

Smith T, Glynn P, Maté J, Toth L, Gyory J (2013) A depth refugium from catastrophic coral bleaching prevents regional extinction. Ecology

Smith TB, Brandtneris VW, Canals M, Brandt ME, Martens J, Brewer RS, Kadison E, Kammann M, Keller J, Holstein DM (2016c) Potential Structuring Forces on a Shelf Edge Upper Mesophotic Coral Ecosystem in the US Virgin Islands. Front. Mar. Sci. 3:115

Smith TB, Gyory J, Brandt ME, Miller WJ, Jossart J, Nemeth RS (2016d) Caribbean mesophotic coral ecosystems are unlikely climate change refugia. Glob. Chang. Biol. 22:2756–2765

Spalding MD, Brown BE (2015) Warm-water coral reefs and climate change. Science 350:769–771

Starr RM, Sala E, Ballesteros E, Zabala M Spatial dynamics of the Nassau grouper Epinephelus striatus in a Caribbean atoll. Mar. Ecol. Prog. Ser. 343:239–249

Stevenson TC, Tissot BN (2013) Evaluating marine protected areas for managing marine resource conflict in Hawaii. Mar. Policy 39:215–223

Stevenson TC, Tissot BN, Dierking J (2011) Fisher behaviour influences catch productivity and selectivity in West Hawaii's aquarium fishery. ICES J. Mar. Sci. 68:813–822

Tenggardjaja KA, Bowen BW, Bernardi G (2014) Vertical and Horizontal Genetic Connectivity in Chromis verater, an Endemic Damselfish Found on Shallow and Mesophotic Reefs in the Hawaiian Archipelago and Adjacent Johnston Atoll. PLoS One 9:e115493

Tenggardjaja KA, Bowen BW, Bernardi G (2016) Reef Fish Dispersal in the Hawaiian Archipelago : Comparative Phylogeography of Three Endemic Damselfishes. J. Mar. Biol. 2016:in press

Toonen RJ, Andrews KR, Baums IB, Bird CE, Concepcion GT, Daly-Engel TS, Eble J a., Faucci A, Gaither MR, Iacchei M, Puritz JB, Schultz JK, Skillings DJ, Timmers M a., Bowen BW (2011) Defining Boundaries for Ecosystem-Based Management: A Multispecies Case Study of Marine Connectivity across the Hawaiian Archipelago. J. Mar. Biol. 2011:1–13

Toonen RJ, Wilhelm TA, Maxwell SM, Wagner D, Bowen BW, Sheppard CRC, Taei SM, Teroroko T, Moffitt R, Gaymer CF, Morgan L, Sheppard ALS, Parks J, Friedlander AM (2013) One size does not fit all : The emerging frontier in large-scale marine conservation. Mar. Pollut. Bull. 2010–2013

Turner JA, Babcock RC, Hovey R, Kendrick GA (2017) Review Article Deep thinking : a systematic review of mesophotic coral ecosystems.

Vaz AC, Paris CB, Olascoaga MJ, Kourafalou VH, Kang H, Reed JK (2016) The perfect storm: Match-mismatch of bio-physical events drives larval reef fish connectivity between Pulley Ridge mesophotic reef and the Florida Keys. Cont. Shelf Res. 125:136–146

Weijerman M, Fulton E a, Parrish F a (2013) Comparison of coral reef ecosystems along a fishing pressure gradient. PLoS One 8:e63797

White KN, Ohara T, Fujii T, Kawamura I, Mizuyama M, Montenegro J, Shikiba H, Naruse T, McClelland T, Denis V, Reimer JD (2013) Typhoon damage on a shallow mesophotic reef in Okinawa, Japan. PeerJ 1:e151

Williams ID, Richards BL, Sandin S a., Baum JK, Schroeder RE, Nadon MO, Zgliczynski B, Craig P, McIlwain JL, Brainard RE (2011) Differences in Reef Fish Assemblages between Populated and Remote Reefs Spanning Multiple Archipelagos Across the Central and Western Pacific. J. Mar. Biol. 2011:1–14

Williams ID, Walsh WJ, Claisse JT, Tissot BN, Stamoulis K a. (2009) Impacts of a Hawaiian marine protected area network on the abundance and fishery sustainability of the yellow tang, Zebrasoma flavescens. Biol. Conserv. 142:1066–1073

Wilson SK, Adjeroud M, Bellwood DR, Berumen ML, Booth D, Bozec Y-M, Chabanet P, Cheal a, Cinner J, Depczynski M, Feary D a, Gagliano M, Graham N a J, Halford a R, Halpern BS, Harborne a R, Hoey a S, Holbrook SJ, Jones GP, Kulbiki M, Letourneur Y, De Loma TL, McClanahan T, McCormick MI, Meekan MG, Mumby PJ, Munday PL, Ohman MC, Pratchett MS, Riegl B, Sano M, Schmitt RJ, Syms C (2010) Crucial knowledge gaps in current understanding of climate change impacts on coral reef fishes. J. Exp. Biol. 213:894–900

3.9 Tables

Table 3.1: Number of reef fish surveys conducted for each island/depth combination.

Island	Shallow (0-30m)	Upper MCE (30-60m)	Lower MCE (60-100m)	TOTAL MCE (30-100m)
Niihau	**103**	1	4	**5**
Oahu	**228**	1	1	**2**
Lanai	**115**	0	4	**4**
Molokai	**170**	0	2	**2**
Maui	**174**	1	9	**10**
Hawaii	**367**	102	6	**108**

Table 3.2: Kernel density estimate (KDE) sample size and interquartile ranges for each focal species.

Species	Sample Size		Interquartile Range	
	Shallow	MCE	Shallow	MCE
Chlorurus spilurus	2885	33	8	20
Ctenochaetus strigosus	20963	296	4	3
Scarus psittacus	2230	67	7	8
Thalassoma duperrey	20947	297	4	4
Zebrasoma flavescens	13241	312	5	3

Table S3.1: Total abundance of fishes observed within each island/depth combination

Species	Niihau 0-30	Niihau 30-60	Niihau 60-100	Oahu 0-30	Oahu 30-60	Oahu 60-100	Molokai 0-30	Molokai 60-100	Lanai 0-30	Lanai 60-100	Maui 0-30	Maui 30-60	Maui 60-100	Hawaii 0-30	Hawaii 30-60	Hawaii 60-100
Abudefduf abdominalis	39			72			73		99		120			188		
Abudefduf sordidus	24			10			25		15		19			41		
Abudefduf vaigiensis	10			47			59		43		91			65		
Acanthrusu xanthopterus															5	
Acanthurus achilles	45			2			7	2	37		14			110		
Acanthurus blochii	88			73			111		74		39		15	250	8	3
Acanthurus dussumieri	93	1		67		2	123	2	57	2	120		1	264	23	1
Acanthurus guttatus	13						21		3		13			31		
Acanthurus leucopareius	649			507			841		274		1861			1999	7	
Acanthurus nigricans				1			4		11		9			54	5	
Acanthurus nigrofuscus	1621			3462			3231		2871		4561		2	8714	298	
Acanthurus nigroris	137	1	3	93			57		82		78		9	117	31	14
Acanthurus olivaceus	496	9		572			280		152	14	318	3	19	701	46	
Acanthurus thompsoni	42			29			90		596		48			376	53	
Acanthurus triostegus	393			450			887		257		478			185		
Acanthurus xanthopterus	11			5			16		10		8			30		
Amblycirrhitus bimacula														1		
Anampses chrysocephalus	127			77			64		31		30			16	31	
Anampses cuvier	13			18			62		5		76			56		
Aphareus furca	55	2		1			12		24		12		3	140	1	
Apogon sp				41			3				13		4	6		
Apolemichthys arcuatus	11		2	4			15	3	1	1	10			34	7	

116

Aprion virescens	93		15	43	3	20		20	7	22	2	2
Arothron hispidus	1		3							1	1	
Arothron meleagris			1							3		
Aulostomus chinensis	7		18	19		18		23		71	7	
Balistes polylepis			1					3		3		
Bodianus albotaeniatus	217	1	150	265		103		270	5	319	11	2
Bodianus sanguineus		2										
Bothus mancus										3		
Bryaninops yongei											1	
Calotomus carolinus	41		39	20		8		37		21	6	
Calotomus zonarchus	5		2	1				2		1		
Cantherhines dumerilii	6		13	4		6		18		32	2	
Cantherhines sandwichiensis	6		59	28		8		40		74	2	
Cantherhines verecundus			1	1		2		3	1	0		
Canthigaster amboinensis	21		35	21		14		28		44	1	
Canthigaster coronata	13	1	146	36		5	1	24	1	9	17	
Canthigaster epilampra	1		2			3				0	17	
Canthigaster jactator	98		760	376		271		533		390	48	
Canthigaster solandri	2									1		
Caracanthus typicus	2		7	2		5		12		15		
Carangoides ferdau	9			3		1				0		
Carangoides orthogrammus	3			6		6		1		0		
Caranx ignobilis	1									0		
Caranx melampygus	87		52	127		45		30	2	63	1	
Carcharhinus amblyrhynchos	2									0		
Centropyge fisheri	32		81	17		31		36		376	145	3

117

Species																		
Centropyge loricula				2			3									23		
Centropyge potteri	94	1	1	139			164			140			212	1	12	746	68	3
Cephalopholis argus	146	4		37			195			178			96			299	19	
Chaetodon auriga	33			8			19			13	2		23		2	64	2	
Chaetodon citrinellus							1									0		
Chaetodon ephippium	4						3			4						7		
Chaetodon fremblii	28			28			40			7			51			26		
Chaetodon kleinii	73		2	644			111	1		129	6		228	2	8	81	87	
Chaetodon lineolatus				1									3			5		
Chaetodon lunula	37			18			41			51			40			186	18	
Chaetodon lunulatus			2				19			18			19			17		
Chaetodon miliaris	122		2	305			151	16		43	6		65		35	55	20	
Chaetodon multicinctus	111	1		346			436			315			357			1348	85	
Chaetodon ornatissimus	16			56			140			111			179			385	14	
Chaetodon quadrimaculatus	70			130			100			61			80			455		
Chaetodon reticulatus										5			2			3		
Chaetodon tinkeri			3			2										9	17	1
Chaetodon unimaculatus				39			79			28			61			55	3	
Cheilio inermis	3			2			6						8			0		
Chlorurus perspicillatus	8			5			23			25			10		2	9	1	
Chlorurus spilurus	8			107			958			515			302			368	78	
Chromis acares	78			30						63			10			156		
Chromis agilis	62	14		105			538			1910			408			16780	1228	
Chromis hanui	430			341			484			327			563		12	1764	93	7
Chromis leucura			12				3								3	242	2037	3
Chromis ovalis	600			113			266			23			535		20	197	56	12

	S1a	S1b	S1c	S2a	S2b	S3a	S3b	S4a	S4b	S5a	S5b	S6a	S6b	S6c
Chromis vanderbilti	7780	25		6518		6615		5430		7854		26370	6	
Chromis verater	198	5	3	42		104	18	80		239	71	285	302	8
Cirrhilabrus jordani				2		3		6		7		0	178	
Cirrhitops fasciatus	56			205		68		4		98		108		
Cirrhitus pinnulatus	23			20		25		10		16		43		
Cirripectes obscurus						1		1				1		
Cirripectes vanderbilti	25			78		37		29		50		75		
Coris ballieui	30			6		12				10		1		
Coris flavovittata	17	1	2	3		2				3		3		
Coris gaimard	83			163		104		65		81		233	23	
Coris venusta	1190			813		237		91		182		129		
Ctenochaetus hawaiiensis	1			3		5		2		6	8	469	40	
Ctenochaetus strigosus	502	5	3	386		2355		1793		2626	2	8672	268	7
Cymolutes lecluse				3		7				1		0		
Dascyllus albisella	227			313	1	291		199		575		200	45	
Decapterus macarellus	270			137		1443		303		600		384	6	
Dendrochirus barberi				1				1				0		
Diodon holocanthus				1								0		
Diodon hystrix	1									3		4		
Echidna nebulosa				1						1		0		
Elagatis bipinnulata						2						3	1	
Enchelynassa canina						1						1	1	
Exallias brevis	4			4		8		6		16		32		
Fistularia commersonii				34		6		9		18		14		
Forcipiger flavissimus	43	2		81		87		58	3	98	14	324	120	15
Forcipiger longirostris	8			3		24		6		17		79	48	
Gnatholepis anjerensis													1	
Gomphosus varius	38			119		385		225		246		597	18	

Goniistius vittatus			1							0		
Gorgasia hawaiiensis											8	
Gunnellichthys curiosus	16		18	5	2		8			25		
Gymnomuraena zebra			1	1						2		
Gymnothorax eurostus	1	1	5	2			3			3		
Gymnothorax flavimarginatus	6	1	11	10	7		10			18	4	1
Gymnothorax javanicus			2	2	2		1			0	1	
Gymnothorax melatremus	1		1	1			1			6		
Gymnothorax meleagris	9		24	10	4		20			42	1	
Gymnothorax nudivomer		4										
Gymnothorax rueppellii										1	5	
Gymnothorax steindachneri							1			0		
Gymnothorax undulatus	1		4				4			1		
Halichoeres ornatissimus	297	1	229	268	220		363			852	24	
Hemitaurichthys polylepis	16		1	5	55		20	2		22	35	
Hemitaurichthys thompsoni					1		15			74	11	
Heniochus acuminatus						10						
Heniochus diphreutes	145	29	387	82	60	2	16	35	6	11	2	
Heteropriacanthus cruentatus				1						11		
Iniistius aneitensis			1							0		
Iniistius pavo				2			1			1	1	
Kuhlia sandvicensis	1			1			3			0		
Kyphosus sp	323		34	571	8		131			210		
Labroides phthirophagus	42		81	143	131		79			242	14	
Lactoria fornasini			1	1			1			0		
Liopropoma aurora		1										

Species												
Lutjanus fulvus	194	1	6		63	68	60	8		112	1	
Lutjanus kasmira	1268		1484		685	252	97	40		3109	32	21
Luzonichthys earlei		3607										
Macropharyngodon geoffroy	431		264		166	66	176			135	252	
Malacanthus brevirostris	21		37		44	2	15			3		
Melichthys niger	48		92		141	101	167			847		
Melichthys vidua	80		102		75	91	107			232	15	
Monotaxis grandoculis	103		77		113	61	54	2		218	10	
Mulloidichthys flavolineatus	90		7		155	111	656			200	33	
Mulloidichthys mimicus										1		
Mulloidichthys pfluegeri		25			4	3	2			3		
Mulloidichthys vanicolensis	61		69		444	59	262			323	48	
Myrichthys magnificus										1		
Myripristis amaena	1	16			19	4	1			50		
Myripristis berndti	233		29		58	70	53			269	3	
Myripristis chryseres											42	
Myripristis kuntee	13		505		14	42	5			527	4	
Myripristis vittata					1	3				0		
Naso annulatus					2	1	1			6		
Naso brevirostris	17	1	153		233	214	278	2	1	72		
Naso caesius		2										
Naso hexacanthus	281	56	209	6	386	427	382	40	25	169	15	
Naso lituratus	239		348		263	199	373		5	632	83	
Naso unicornis	188		187	4	91	25	71	2		70	12	
Nemateleotris magnifica						1				1		
Neoniphon aurolineatus									4			1
Neoniphon sammara					2	26	7			40	1	

Species	1a	1b	1c	2a	2b	2c	3a	3b	3c	4a	4b	4c	5a	5b	5c	6a	6b	6c
Novaculichthys taeniourus	15			35			7			1			10			7	1	
Oplegnathus punctatus							2									0		
Ostracion meleagris	7			17			22			14			41			46	5	
Ostracion whitleyi	1												1			2	2	
Oxycheilinus bimaculatus	113			840	1		354	1		68			179			6	11	
Oxycheilinus unifasciatus	7			3			101			74			87			158	29	
Oxycirrhites typus				1								1	1		1	0		
Paracirrhites arcatus	1067			1171	1		937			883			899			3179	23	
Paracirrhites forsteri	55			40			75			57			55			151	1	
Parapercis schauinslandii	42			134	1		5					3	15		1	0	1	
Parupeneus chrysonemus												3						
Parupeneus cyclostomus	52			52			14			26			41		1	68	13	2
Parupeneus insularis	63			31			28			16			23			165	6	
Parupeneus multifasciatus	329		3	1014			387	1		302	10		460		16	720	65	6
Parupeneus pleurostigma	88			212			88			24	1		79			53	46	
Parupeneus porphyreus	14			14			40			7			22		9	21	14	
Pervagor aspricaudus	3			64			49			2			50			46	4	
Pervagor spilosoma				43			7					1	6			5		
Plagiotremus ewaensis	39			37			30			11			26			64	2	
Plagiotremus goslinei	84			28			150			34			47			74	1	
Plectroglyphidodon imparipennis	1021			855			540			303			294			485		
Plectroglyphidodon johnstonianus	275			635			749			641			977			1123	9	
Plectroglyphidodon sindonis				1			40			18			27			25		
Priacanthus meeki													1			2		
Pristiapogon kallopterus	84	19	6	141			104			22			24			131	13	
Pseudanthias bicolor	13		8	127			47		6	1			13			7	36	

Species																
Pseudanthias hawaiiensis			115	1			9		3						16	
Pseudanthias thompsoni			3														
Pseudocheilinus evanidus	117	4	4	304	1		311		352		163	6	2	1129	607	4
Pseudocheilinus octotaenia	9			75			153		110		145			518	147	
Pseudocheilinus tetrataenia	34			69			108		111		77			769	4	
Pseudojuloides cerasinus	164			542	1		154		88		174			97	54	
Ptereleotris heteroptera	34			241			11		13		81			5		
Rhinecanthus aculeatus	5			4			4				4			4		
Rhinecanthus rectangulus	109			334			88		70		155			74		
Sarda orientalis											36			0		
Sargocentron caudimaculatum														1		
Sargocentron diadema	10			156			61		2		5			39	16	
Sargocentron ensifer							2							0		
Sargocentron spiniferum	1	1		1			4		5		2			11	5	
Sargocentron tiere	3			4			1		9					41		
Sargocentron xantherythrum				23			31		3		24		2	1	3	
Saurida flamma														1		
Scarus dubius	27			6			45		30		39			15		
Scarus psittacus	56			165			429		444		363			180	44	
Scarus rubroviolaceus	134			29	7		87	3	73	3	103			181	19	2
Scomberoides lysan	1			7					1		4			0		
Scorpaenopsis cacopsis	1										1			2		
Scorpaenopsis diabolus				3							4			5	3	
Scuticaria tigrina				1										0		
Sebastapistes ballieui	2										1			3		
Sebastapistes coniorta	5			3			2				3			2		

Species																		
Selar crumenophthalmus																10		
Seriola dumerili	3	1													1	2	1	
Sphyraena barracuda	1															2		
Sphyraena helleri										1						0		
Stegastes fasciolatus	210			470			711			455			1390			1212		
Stethojulis balteata	338			881			682			523			931			600		
Sufflamen bursa	262	1		837			285	1		153			389	1	3	813	81	2
Sufflamen fraenatum	108	1		297			120			34	1		66	1		70	50	1
Synodontidae	1			7			10			9			10	1		18	12	
Thalassoma ballieui	16			1			34			5			18			20	6	
Thalassoma duperrey	2637	4		3596			2572			2295			2618			3451	181	4
Thalassoma lutescens				1			1			9						0		
Thalassoma purpureum	21			17			7			5			17			13		
Thalassoma quinquevittatum				4			3			2						0		
Thalassoma trilobatum	92			14			61			57			27			31	72	
Xanthichthys auromarginatus			5	83						14	4		14	2	5	100	99	14
Xanthichthys caeruleolineatus																1		
Xanthichthys mento										10				1	1			7
Zanclus cornutus	47			88			73	2		74	3		87		12	294	192	7
Zebrasoma flavescens	40	6		100			923			1248			663		10	6493	224	11
Zebrasoma veliferum				3			20			15			7			40	38	

3.10 Figures
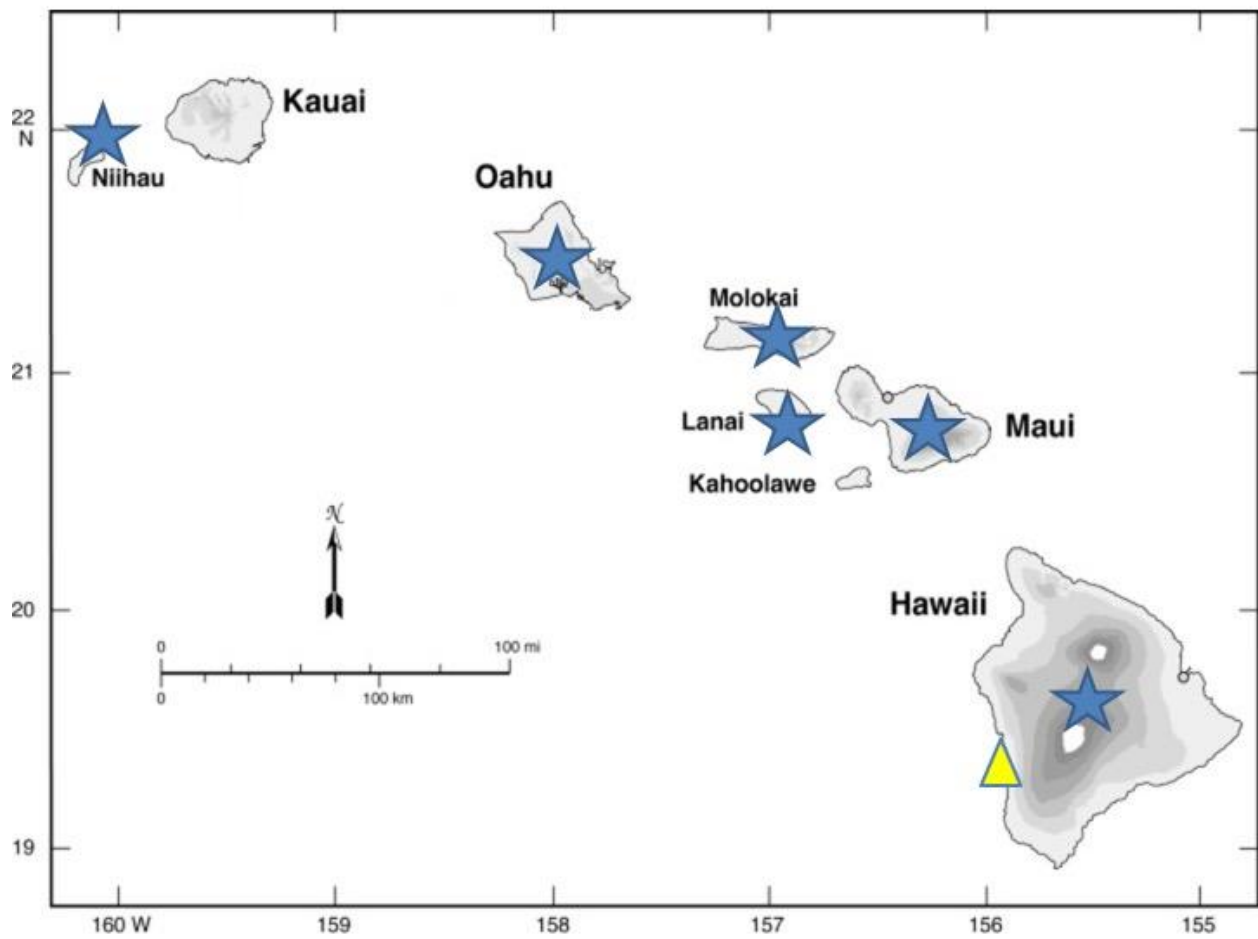
Figure 3.1: Map of the Main Hawaiian Islands. Starred islands indicate regions with survey data.

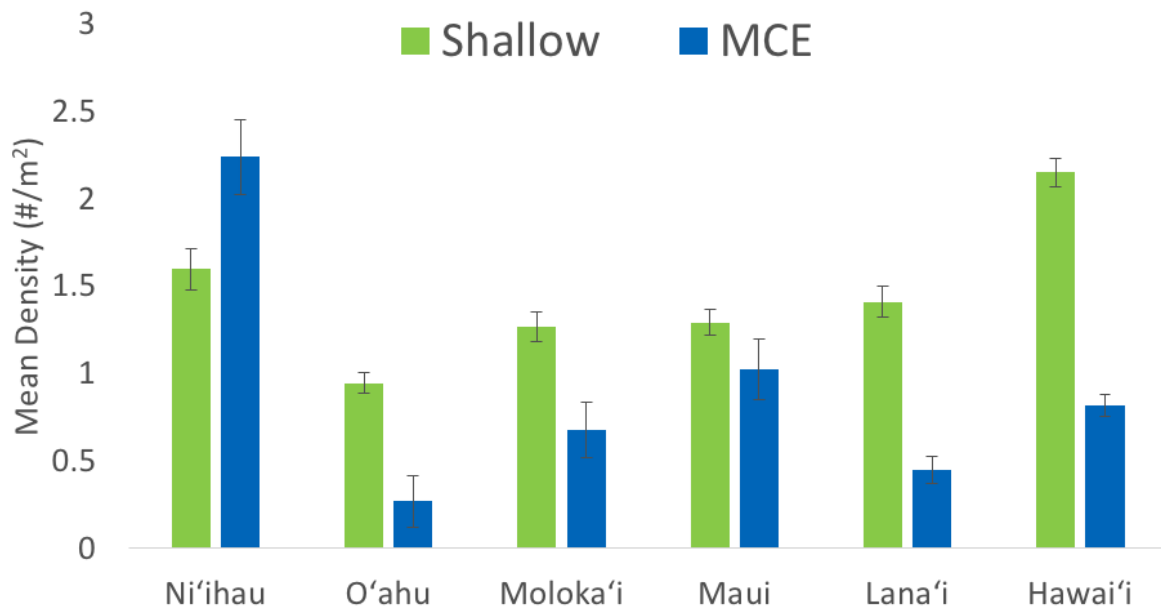Triangle indicates location of temperature recordings.

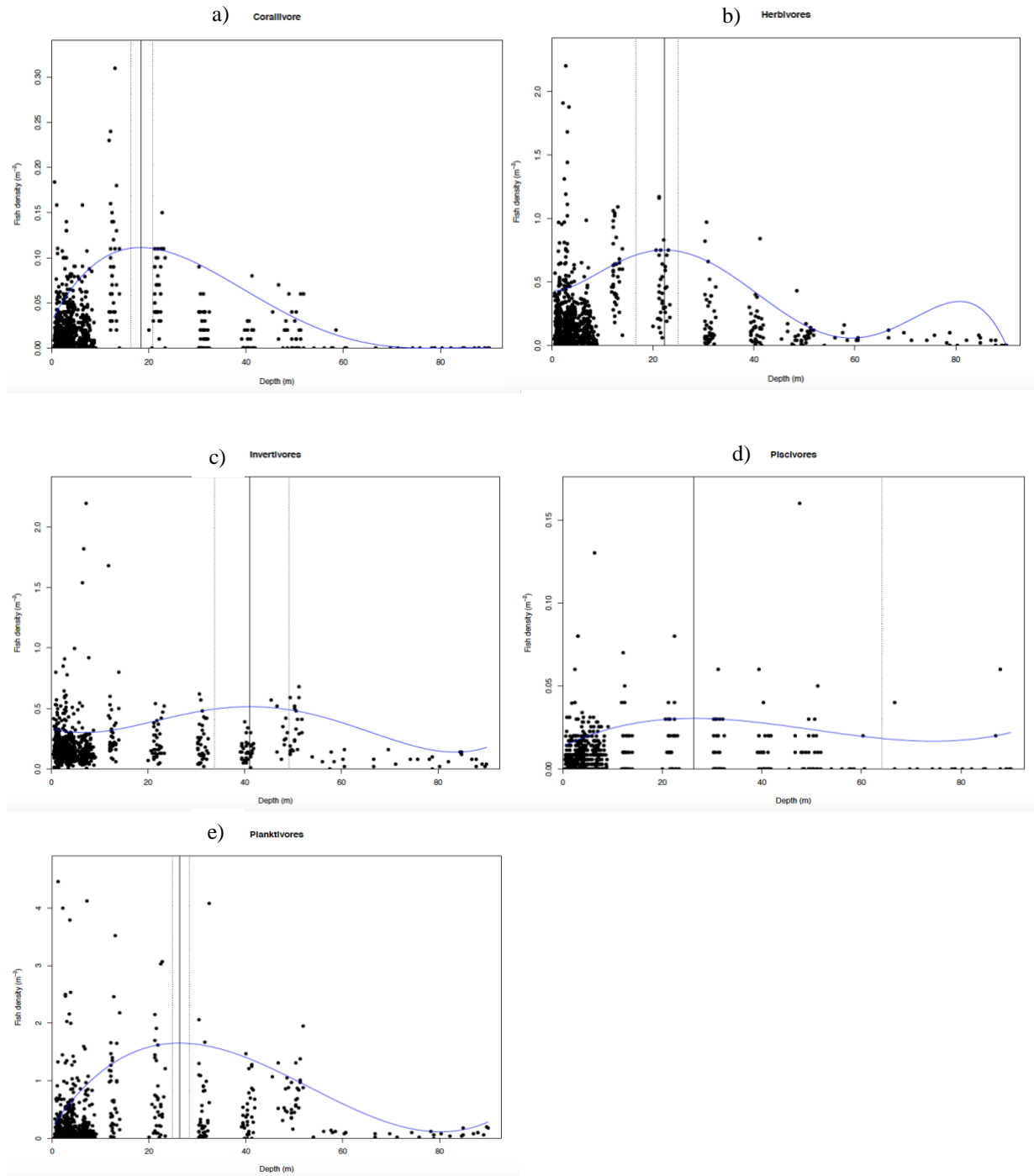Figure 3.2: Mean density (+/- SE) of reef fish at shallow and mesophotic depths within each island surveyed.

Figure 3.3: Relationship between fish trophic category and depth. Regression spline model for the 90th percentile indicated with maximum as a vertical solid line and 90% bootstrapped confidence intervals as dotted lines.
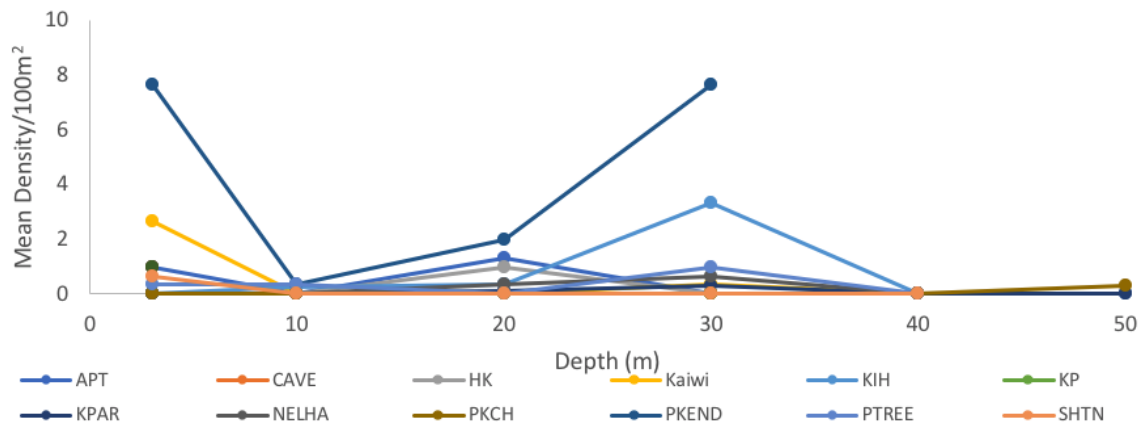
Figure 3.4: *Scarus psittacus* density by site.

a)
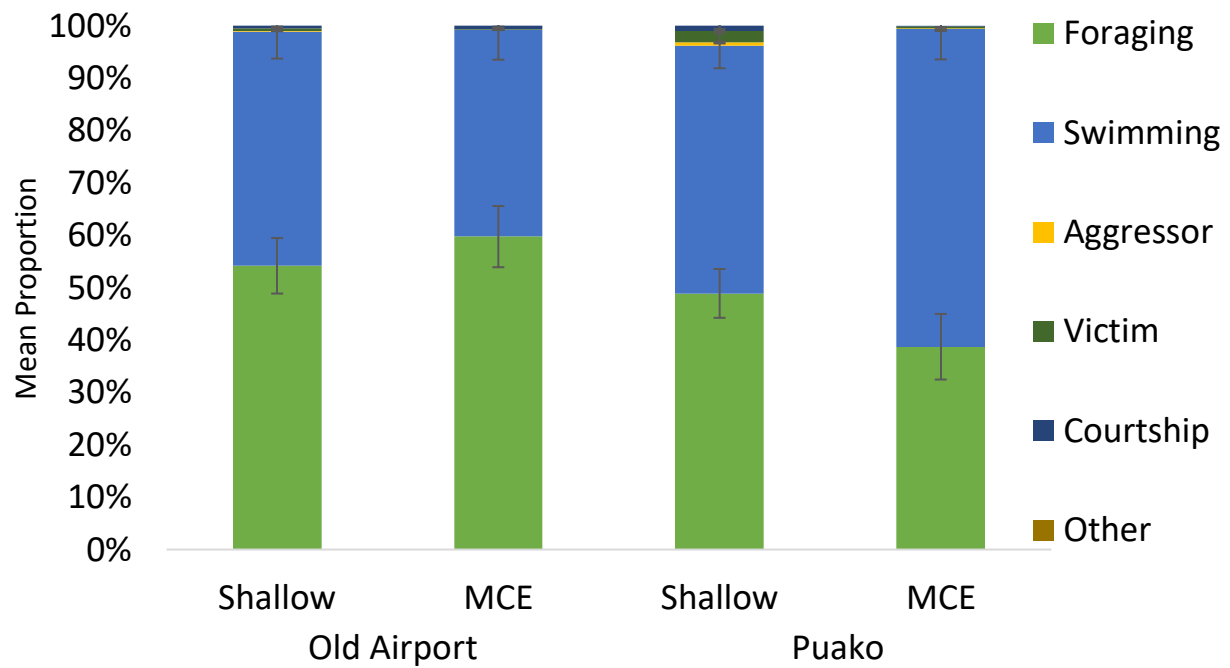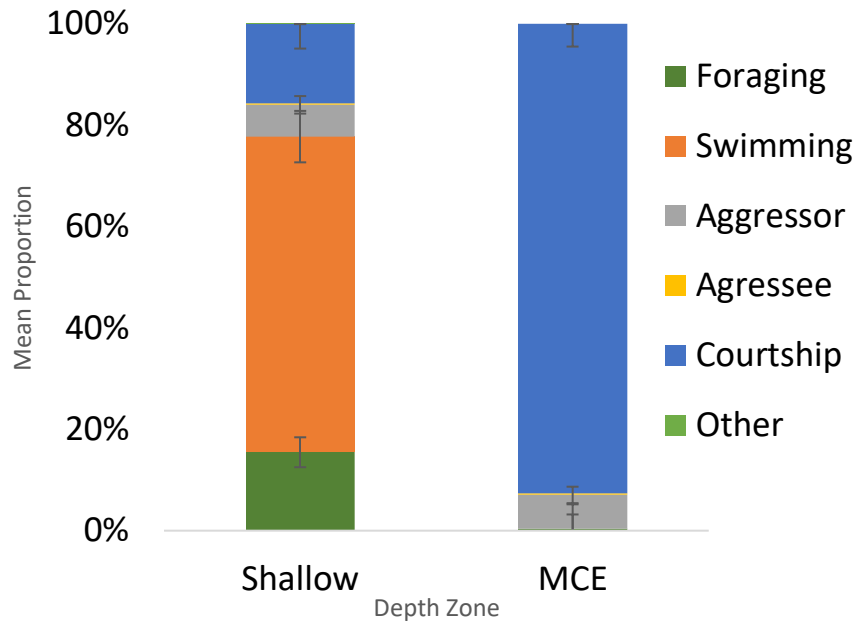
Figure 3.5: a) *Zebrasoma flavescens* and b) *Scarus psittacus* terminal male behavior between

shallow (<10m) and MCE (>30m) depths in Hawaiʻi Island.
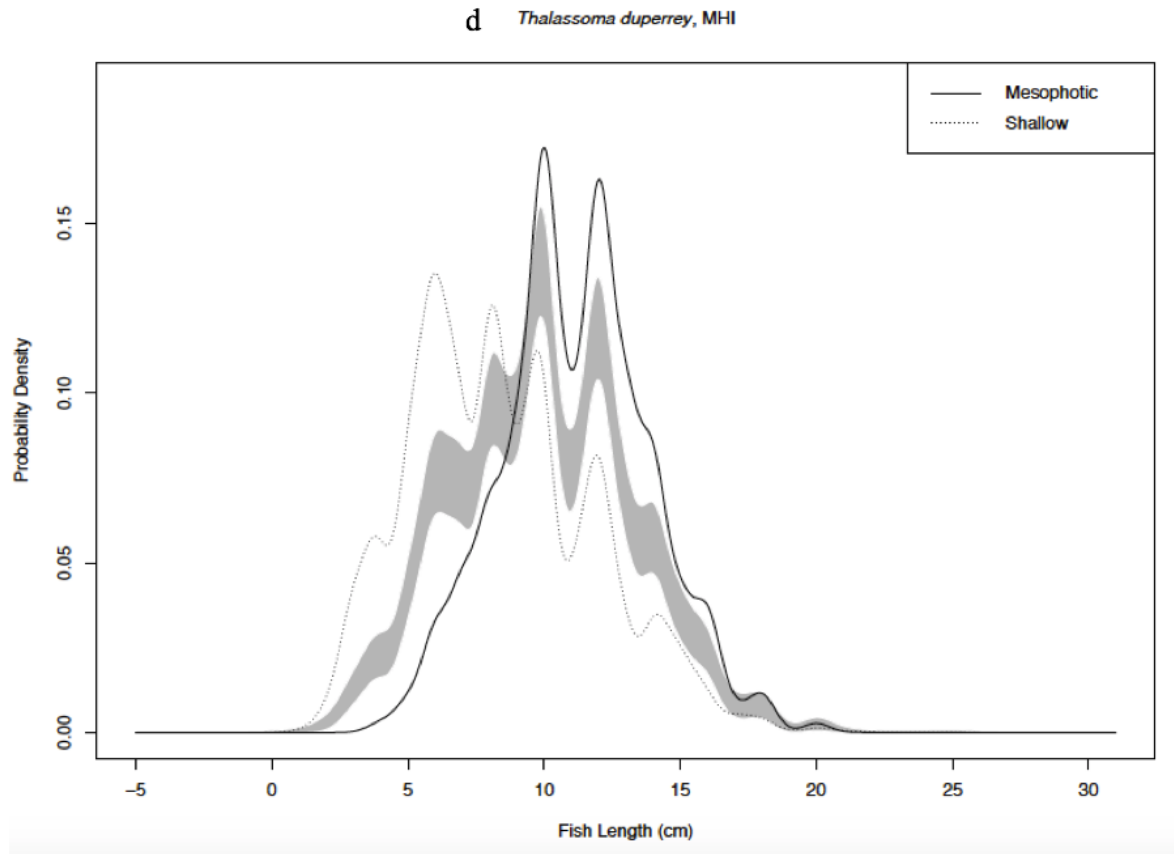
a *Chlorurus spilurus*, MHI
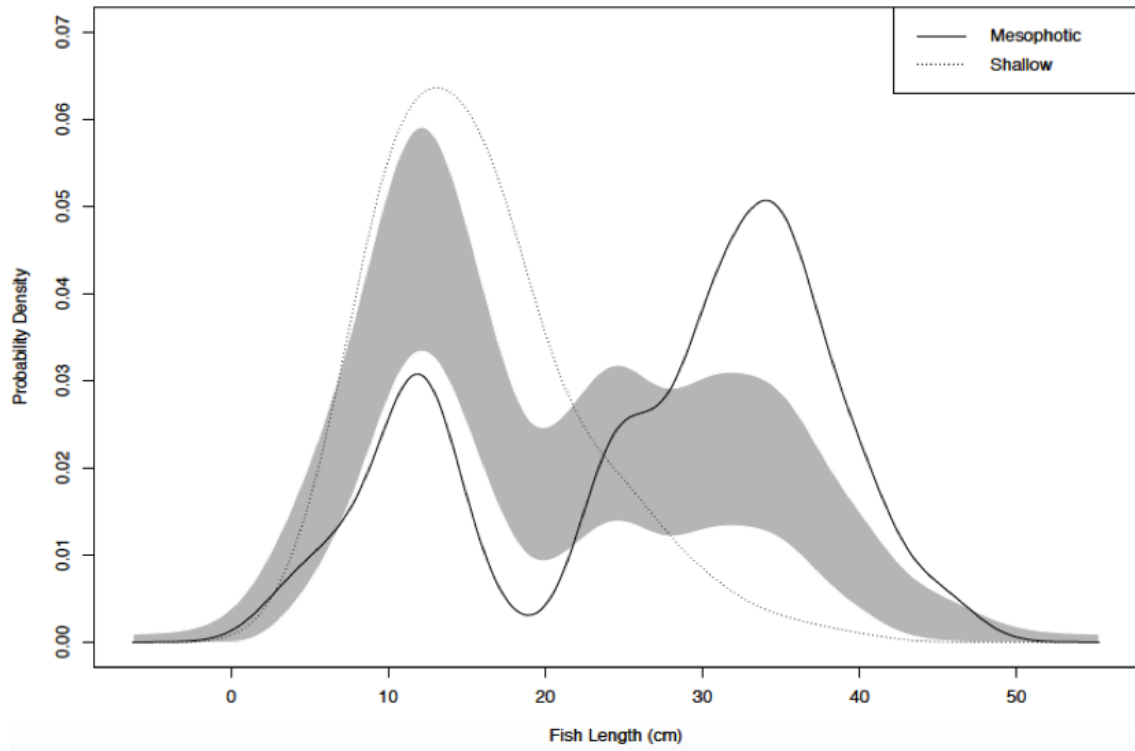
b *Scarus psittacus*, MHI
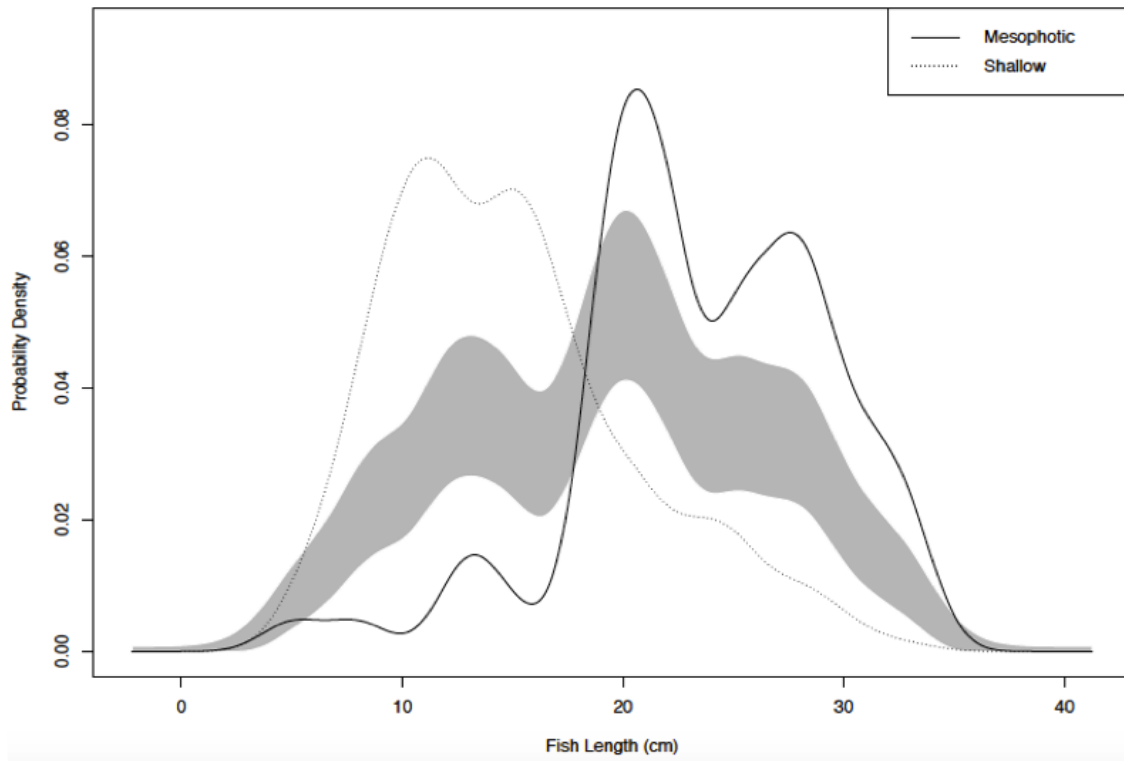
c   *Zebrasoma flavescens*, MHI
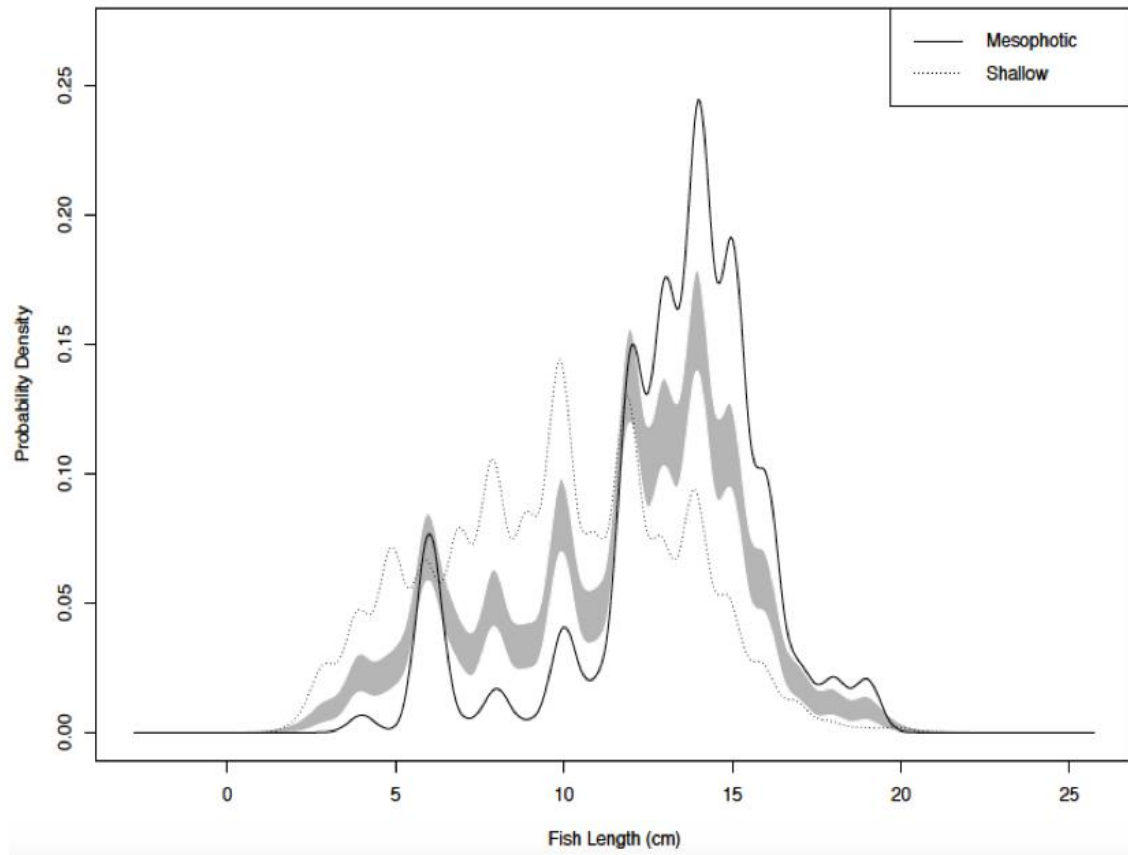
Figure 3.6: Comparison of kernel density estimates (KDEs) using mean bandwidths for a) *Chlorurus spilurus*, b) *Scarus psittacus*, c) *Zebrasoma flavescens*, and d) *Thalassoma duperrey*.
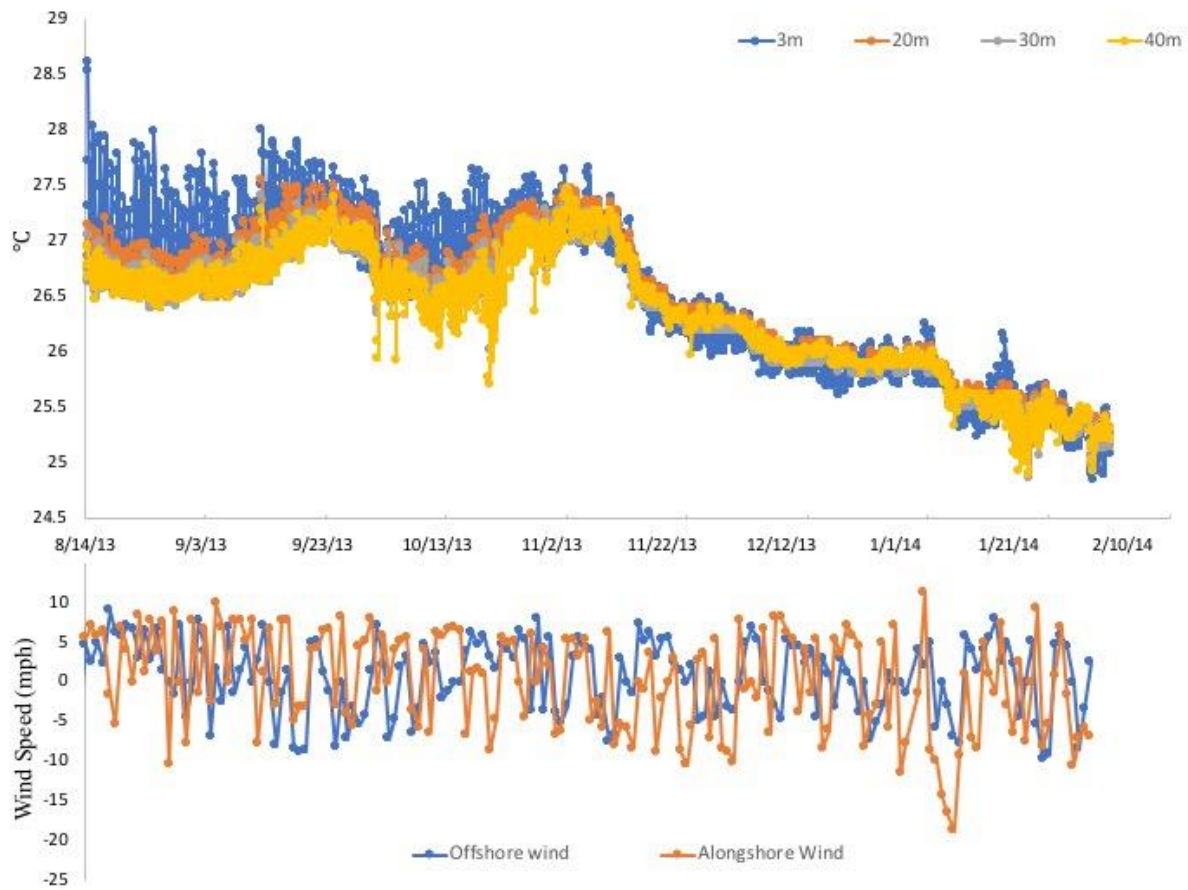
Figure 3.7: a) Temperature records for 3-40m depths at Kaohe Bay, Hawaii from August 2013-

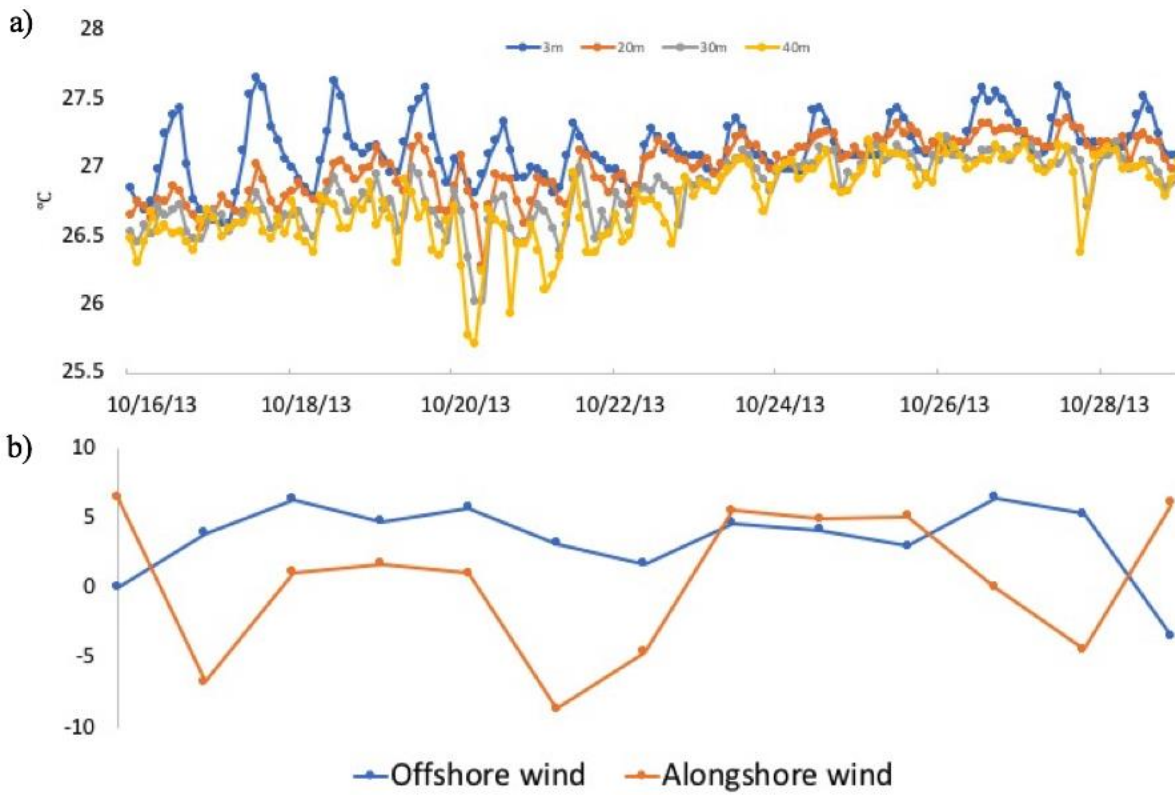February 2014; b) Corresponding offshore and alongshore wind speed for Kona, Hawaii.

Figure 3.8: a) Temperature records for 3-40m depths at Ka′ohe Bay, Hawaii for October 2013; b) Corresponding offshore and alongshore wind speed for Kona, Hawaii.
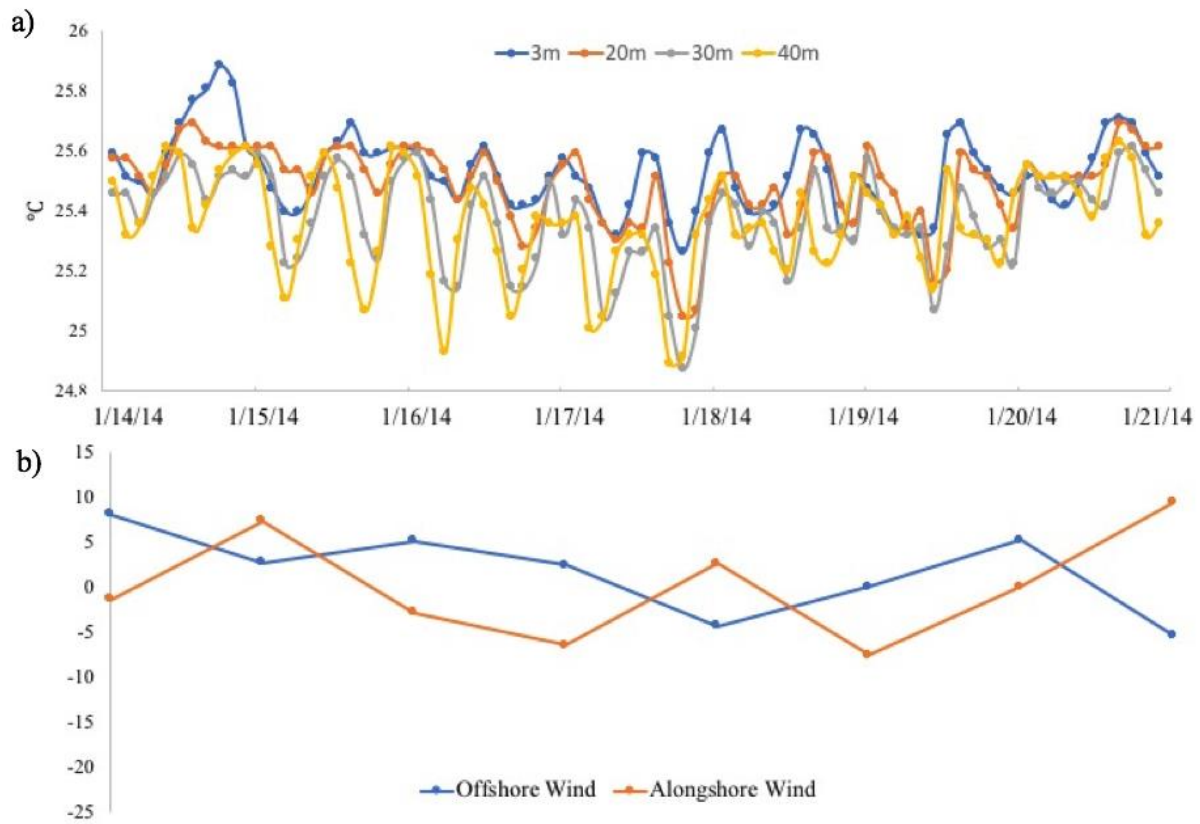
Figure 3.9: a) Temperature records for 3-40m depths at Ka′ohe Bay, Hawaii for January 2014; b)

Corresponding offshore and alongshore wind speed for Kona, Hawaii.