GIANT KELP GENETIC MONITORING BEFORE AND AFTER DISTURBANCE REVEALS STABLE GENETIC DIVERISTY IN SOUTHERN CALIFORNIA

by

William H. Klingbeil III

A Thesis Submitted in
Partial Fulfillment of the
Requirements for the Degree of

Master of Science
in Biological Sciences

at
The University of Wisconsin-Milwaukee
August 2020

ABSTRACT

GIANT KELP GENETIC MONITORING BEFORE AND AFTER DISTURBANCE REVEALS STABLE GENETIC DIVERISTY IN SOUTHERN CALIFORNIA

by

William Klingbeil

The University of Wisconsin-Milwaukee, 2020
Under the Supervision of Professor Filipe Alberto

Given the impacts of climate change and other anthropogenic stressors on marine systems, there is a need to accurately predict how species respond to changing environments and disturbance regimes. The use of genetic tools to monitor temporal trends in populations gives ecologists the ability to estimate changes in genetic diversity and effective population size that may be undetectable by traditional census methods. Although multiple studies have used temporal genetic analysis, they usually involve commercially important species, and rarely sample before and after disturbance. In this study, we use newly collected samples, coupled with previously characterized microsatellite data to assess the genetic consequences of disturbance in several populations of giant kelp (*Macrocystis pyrifera*) in the Southern California Bight. We performed a pre- and post-disturbance microsatellite analysis to look at changes over a 10-year period, which included the 2015/16 El Nino Southern Oscillation event. We used canopy biomass estimated by remote sensing (Landsat) to quantify the extent of disturbance to kelp beds, and sea surface temperature data to understand how kelp was pushed towards its temperature limits during this period. Despite prolonged periods with decreased canopy at several sites, no changes in genetic structure and allelic richness was observed. We discuss how deep refugia of subsurface sporophytes and cryptic microscopic life stages could have kept genetic diversity through disturbance, with the latter being the only possible mechanism in one shallow continental site. Given the increasing effects of climate change and uncertainty in modeling impacts of species with cryptic life history stages, we suggest further investigation to reveal the role such stages play in species resilience.

Artist: Kate Klingbeil

To all who have helped engender my curiosity for the ocean

and scientific inquiry.

TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

ACKNOWLEDGMENTS

This thesis work was made possible through consultation and mentorship of a dedicated and passionate number of educators, researchers, and fellow outdoor enthusiasts. I would like to start by acknowledging my advisor, Filipe Alberto, who always expressed genuine and constructive advice throughout my degree at UWM. Thank you for supporting me in the exploration of my research interests, having an incredible amount of patience, and for providing me with a concrete foundation in experimental design and statistical analysis. Special thanks to my thesis committee: Emily Latch, Jeff Karron, Erica Young, and John Berges. Thank you all for your time and for helping me to see the bigger picture. To my lab mate, Lily Gierke, for always providing me with comic relief, quality friendship, and proper laboratory etiquette. To my lab technician and good friend, Gabriel J. Montecinos, for making the lab a productive and positive environment. Thank you for being an incredible role model and always exemplifying a real work hard play hard mentality. Special thanks to my post-doc and friend Rachael Wade for writing mentorship, and genuine professional advice. To all my lab mates who were always a pleasure to work with.

I would like to thank the Catalina Island Marine Institute at Fox Landing (CIMI), especially Heather Peterson, who always provided me with unconditional support and gave me a base of operation for field sampling. Special thanks to my fellow CIMI co-workers who engendered my passion for kelp forests and enthusiastically helped with fieldwork and sample preparation. To Dan Reed's team at the University of California Santa Barbara and Dave Siegel's team at the Earth Research Institute for additional field sampling. Special thanks to Tom Bell and Kyle Cavanaugh for support with Landsat data analysis. To Mattias Johansson and

previous Alberto lab mates for providing the baseline genetic data used in this study. To the University of Wisconsin-Biotechnology Center (Madison, WI), who always provided high-quality sequencing of samples.

I would not be where I am today if it were not my undergraduate mentors at the University of Tampa: Mark McRae, Natalia Belfiore, and Kevin Beach. Thank you for inspiring me to want to pursue a career in biology and a degree in both population genetics and phycological work. Finally, special thanks to my parents and family, who have always been incredibly supportive and allowed me to follow my childhood curiosity about the ocean and genetics.

GIANT KELP GENETIC MONITORING BEFORE AND AFTER DISTURBANCE REVEALS STABLE GENETIC DIVERISTY IN SOUTHERN CALIFORNIA

INTRODUCTION

Habitats of the world's oceans and species therein are changing and projected to continue to alter vastly due to anthropogenic stressors, such as pollution, habitat loss, invasive species, and climate change (Rahel and Olden 2008, Johnston and Roberts 2009, Claudet and Fraschetti 2010, Davidson et al. 2015, McCauley et al. 2015, Garciá Molinos et al. 2016). Predicting how species react to changing regimes has been of concern in species level and biodiversity conservation (Bellard et al. 2012, Tilman et al. 2017). Resilience, the capacity of a system to absorb disturbance and reorganize to a previous state (Walker et al. 2004), is often studied by census and area cover changes, informed by current and historical distributions, and population demography (Timpane-Padgham et al. 2017). However, other essential biodiversity layers, such as genetic diversity, go undetected when populations recover from disturbance to previous census sizes. Only when temporal genetic data is collected (i.e., genetic monitoring) can putative reductions in genetic diversity be revealed.

Genetic monitoring is a powerful tool that can provide ecologists with specific details about how populations change through time and eliminate assumptions that single time point studies confer (Schwartz et al. 2007). Important elements of populations, measured using genetics, such as genetic diversity (i.e., allelic richness and genetic structure), connectivity (i.e., gene flow), effective population size, and the potential for local adaptation have been used as predictors of population resilience (Bernhardt and Leslie 2013, Timpane-Padgham et al. 2017). It has been shown that more genetically diverse populations have greater adaptive potential and functioning (Reusch et al. 2005, Johnson et al. 2006, Hughes et al. 2008). When measured across a species range, and in conjunction with other ecological variables, ecologists can use genetic parameters (i.e., diversity, connectivity, effective population size) to target critical regions for conservation. The efficacy of using genetic measures in conservation largely depends on the knowledge of their stability through time. Only temporal studies can address this issue through empirical analysis of changes in the genetic components of populations.

Genetic monitoring of both captive and natural populations has been used, for example, to assess the stability of population structure, and its relation to both ecological (i.e., natural and anthropogenic) and evolutionary factors (i.e., Barcia et al. 2005, Athrey et al. 2012, Perrier et al. 2013, Gurgel et al. 2020). The use of historical samples in the alpine chipmunk (*Tamias alpinus*) of Yellowstone National Park revealed declines in genetic structure over a century as driven by climate-induced range contraction and habitat fragmentation. Other studies have shown stability in population structure (Tessier and Bernatchez 1999, Ruzzante et al. 2001, Heath et al. 2002, Hoffman et al. 2004, Saltonstall 2011, DeFaveri and Merilä 2015). A study by Arnaud and Laval (2004) in France revealed the stability of genetic structure in a land snail (*Helix aspersa*) over two years, despite perceived (unmeasured) disturbance from farming practices. The authors attributed stability to cryptic life-history traits (undetected by traditional census methods), such as multiple mating and sperm storage. This study hints that genetic monitoring can be of particular use for revealing cryptic life-history traits (Orsini et al. 2016) that may promote population persistence during a disturbance.

Currently, genetic monitoring studies are typically conducted on terrestrial vertebrates (i.e., mammals and birds) or commercially important species (i.e., fish) (Bellinger et al. 2003, Schwartz et al. 2007), and few studies have been done on other important marine taxa (Reynolds et al. 2017, Gurgel et al. 2020, Manent et al. 2020). Until recently, only a few studies had applied genetic monitoring techniques over periods of disturbance (Gurgel et al. 2020, Holt et al. 2020, Manent et al. 2020), This is primarily due to the stochastic nature of disturbance events, and the lack of previous genetic data. The studies previously cited, revealed alarming loss of genetic diversity as in the case of Gurgel et al. (2020), who noted cryptic genetic loss despite demographic recovery. These studies express the need to use genetic monitoring to understand the impact disturbance has on many ecologically important marine species.

The major habitat forming macroalgae is a group that has largely been neglected of genetic monitoring studies (Gurgel et al. 2020). Their ecological (Bertocci et al. 2015, Bennett et al. 2016, Blamey and Bolton 2018) and economic (Chung et al. 2011, Bennett et al. 2016, FAO 2019) importance make them a key target group for conservation. Additionally, recent work has already shown declines of key habitat-forming macroalgae in response to anthropogenic disturbance (i.e., Krumhansl et al. 2016, Smale 2020). Kelps are large brown algae that make up the order Laminariales. They are important foundation species, forming extensive forests in nutrient-rich temperate coastal regions worldwide, and provide habitat for upwards of 270 common species (Buschmann et al. 2007). Kelp forests are among some of the most productive systems on earth (Mann 1973, Vilalta-Navas et al. 2018) and are important in supporting additional coastal and open ocean pelagic systems through detrital export (Duggins et al. 1989, Hobday 2000, Krumhansl and Scheibling 2012).

As coastal ecosystems, kelp forests are exposed to a variety of anthropogenic and natural stressors that are both regional (pollution, invasive species, sedimentation, harvesting, fishing, storm surges, disease, herbivory) and global (climate change, El Niño Southern Oscillation) (Steneck et al. 2002, Schiel and Foster 2015, Bennett et al. 2016). Despite reports of declining populations, several kelp forests have shown a large degree of resilience (Reed et al. 2016, Cavanaugh et al. 2019) with fast recovery following disturbance, as they tend to colonize quickly and have high growth rates (Dayton et al. 1992, Ladah et al. 1999). All of the studies showing resilience in kelps have focused solely on changes in area cover (using remote sensing). By contrast, none have examined temporal changes in genetic diversity and genetic structure. Although there is considerable research on kelp population genetics, only two studies have incorporated a temporal context (Valero et al. 2011, Wootton and Pfister 2013), and none have been conducted following a major disturbance.

One species of particular interest for conservation is the habitat-forming kelp, *Macrocystis pyrifera* (giant kelp). In southern California, much work has been done to understand the population dynamics of giant kelp (Deysher and Dean 1986, Reed 1990, Dayton et al. 1992a, Gaylord et al. 2002, 2006b, Cavanaugh et al. 2010, 2011, 2013, Castorani et al. 2015, D. Reed et al. 2016), including genetics studies (e.g., Alberto et al. 2010b, 2011, Johansson et al. 2013, 2015, Castorani et al. 2017, Hargarten et al. 2019). The surface breaching nature of giant kelp has enabled the use of remote sensing techniques (i.e., Landsat imagery) to build long-term time series in canopy area coverage and biomass estimates (Cavanaugh et al. 2010, Bell et al. 2020). Analysis from both remote sensing and fieldwork has revealed several environmental factors (herbivory, wave-height, light, nutrients, temperature, sedimentation and substrate availability) that influence the distribution and range of giant kelp in the northeast pacific (Cavanaugh et al. 2011, Bell et al. 2015, Young et al. 2016a). Events such as El Niño Southern Oscillation (ENSO) that bring warm nutrient-depleted waters can have substantial adverse effects on canopy biomass and are increasing in frequency and intensity (Solomon et al. 2007, Oliver et al. 2018). Despite these events, giant kelp in southern California has historically been shown to have the highest level of genetic diversity across the species range (Macaya 2010, Johansson et al. 2015) and exhibits resilience of kelp area coverage (Reed et al. 2016, Cavanaugh et al. 2019). However, kelp area resilience is uninformative about the putative cryptic loss of genetic diversity.

In this study, we aim to address the lack of information on changes in genetic diversity by analyzing the temporal variation in genetic structure and allelic richness. We looked at changes that occurred over ten years, for five sites of giant kelp in the Southern California bight. We test the hypothesis that demographic bottlenecks produced by disturbances from 2014-2016 have depressed the levels of genetic diversity in giant kelp. We choose sites that differ in genetic coancestry, oceanographic conditions, and severity of disturbance to assess how impacts of disturbance may vary across the study region.

METHODS

Giant kelp is the largest and most widely distributed benthic species in the world (Buschmann et al. 2007). It forms large surface breaching forests along the coastline from Alaska to Bahia Tortugas, Mexico, at depths of up to 30 meters. As with other kelps, *M. pyrifera* has a biphasic haplodiplontic life cycle in which a large sporophyte (2N) produces spores (1N) via meiosis that settle on hard, rocky substrate, which then differentiates into microscopic male and female gametophytes (1N). Male gametophytes release motile sperm that fertilize a non-motile egg, retained on the female thallus. Following fertilization, the zygote develops into a blade, producing the next generation of sporophytes (reviewed in North 1994). Dispersal occurs mainly through the spore stage, in which planktonic individuals can survive for at least one week (Reed et al. 1992) and may settle several kilometers from the origin. However, most spores settle within a few meters (Reed et al. 1992, Gaylord et al. 2006). There is some evidence for longer dispersal distance by kelp rafts (Hobday 2000, Hernández-Carmona et al. 2006). However, this dispersal mechanism is negligible in maintaining connectivity between local populations when compared to spores (Reed et al. 2004, Hargarten et al. 2019).

Study sites and sampling

To conduct a temporal genetic analysis, we sampled five sites between 2018 and 2019 located in three regions differing in giant kelp coancestry in the Southern California Bight, hereafter referred to as 2018 samples. These had also been sampled before, in 2007 and 2008, and genotyped by Johansson et al. (2015) using microsatellite marker analysis, hereafter referred to as 2008 samples. Two of our locations are continental, Leo Carrillo, and Carlsbad, with one site sampled in each. Our third location was Catalina Island, where we sampled three sites (Fig. 1). We sampled more sites on Catalina Island because this was the location where most anecdotal evidence suggested a disturbance to kelp forests during the 2008 to 2018 period. Catalina Island belongs to a genetic coancestry group comprised of both the northern and southern channel island archipelagos while the northernmost continental site, Leo Carrillo, showed admixture between three genetic coancestry groups in Southern California (Johansson et al. 2015). The southernmost sampling location, Carlsbad, belonged to the Southern California/Baja California genetic group. We note that the 2018 site from this location is located 30 km south of the 2018 site (San Mateo) from Johannsson et al. 2015, due to logistic constraints that precluded sampling the same site. Johansson et al. (2015), reported that kelp forests from a large swath of the coast, south of Los Angeles to Baja California, belonged to the same genetic group, and all sites had similar allelic richness.

New sample collections (Fig. 1) occurred between January 2018 and June 2019. The sampling protocol involved haphazardly collecting (~n=30 per site) sporophyte blade tissue by snorkeling and SCUBA. We collected tissue from individual holdfasts to ensure non-repetitive sampling. Blades were either dried in silica or immediately frozen and desiccated using an Eppendorf Vacufuge Plus (Hamburg, Germany).

Disturbance quantification

There are several anecdotal reports of disturbance to kelp forests in Southern California from 2008 to 2018. During this period, a series of large marine heatwaves (MHW) occurred, starting in the summer of 2014, followed by the 2015/16 El Nino-Southern Oscillation (ENSO) event. On Catalina, two large storm surges hit the Island in the summer and winter of 2014. Additionally, the invasive algal species, *Sargassum horneri*, continued to spread on the Island and spread was likely facilitated by the ENSO event (Marks et al. 2017).

To quantify the scale of disturbance to kelp forests in our study, we estimated proportional changes in surface canopy biomass from 2008 to 2018 for Leo Carrillo, Catalina Island, and Carlsbad using satellite data. We used surface canopy biomass derived from Landsat 5 and 7 Thematic mapper multispectral 30-meter resolution data taken quarterly (Cavanaugh et al. 2010, Bell et al. 2020, available from SBC-LTER). To compare across locations and determine the scale of surface canopy changes, we summed biomass within a buffer surrounding the sampling coordinates for each year and divided by the maximum biomass observed from

2008 to 2018 using the GEOSPHERE v.10 R Package (Hijmans et al. 2019). We used 5, 15, and 30 km buffers to compare the severity of local changes in biomass while controlling for neighboring kelp beds that could disperse individuals into studied sites (had they experienced different dynamics). We choose a maximum buffer of 30 km, as it well exceeds the mean dispersal capabilities of giant kelp (0.5-2.92 km) (Alberto et al. 2010). We averaged the proportions for 5 and 15 km buffers for each of the three sites on Catalina to obtain a single time-series for the Island. Given the size of the island and site spacing, the same 30 km buffer encompassed all possible habitat surrounding the three sites on the Island. For Carlsbad, we averaged the 5, 15, and 30 km proportions between the 2008 and 2018 sample sites. Differences in distances for biomass changes between the three spatial buffers were compared with Pearson's correlations.

The number of marine heatwaves and their duration were quantified for January 2008 to December 2018. We also calculated the time each site experienced above a conservative species threshold, defined as above 22°C. We choose this threshold as populations near the southern edge of giant kelps distribution appears to have reduced biomass above this threshold (Ladah et al. 1999). We used daily temperature from January 1984 to December 2019, obtained from the National Climatic Data Center Optimal Interpolation Sea Surface Temperature (OISST) dataset (Banzon et al. 2016), to produce a climatology baseline for determining temperature anomalies. We estimated the quantity and duration of marine heatwaves as in Hobday et al. (2016) and the number of events lasting five consecutive days above a species threshold of 22 °C from 2008 to 2018 using the heatwaveR v. 0.4.2 R package (Schlegel and Smit 2018).

Genotyping

DNA was extracted using the NucleoSpin 96 Plant Kit II (Macherey-Nagel, Duren, Germany) with standard protocols. Six microsatellite loci (Mpy-8, Mpy-14, BC-4, BC-18, BC-19, BC-25) were examined, following modified PCR protocols in Alberto et al. (2009). PCR product fragment sizes were determined on an ABI 3730 FVNPL (Applied Biosystems) using GeneScan-500 LIZ as a size standard (at the UW-Wisconsin Biotechnology Center). We scored alleles using STRAND v. 2.4.110 and binned them using the MsatAllele v. 1.0.4 R package (Alberto 2009).

Population genetics summary statistics

To compare changes in allelic richness between sample periods, we standardized sample size at n=15 samples for both 2008 and 2018 samples, using the StandArich R package v. 1.00 (Alberto 2005). We tested for significant temporal changes in richness across all sites using a non-parametric Wilcoxon signed-rank test (Arnaud and Laval 2004). Additionally, we tested for population bottlenecks at all sites, for both 2008 and 2018 samples, using BOTTLENECK v. 1.2.02 (Piry et al. 1999). This approach aims to detect the predicted heterozygosity-excess following a demographic bottleneck and loss of rare alleles; observed heterozygosity values are compared with those from simulated populations at mutation-drift equilibrium. We used a two-phase model (TPM) with a 30% variance and a 70% proportion stepwise mutational model (SMM). Deviations from mutation-drift equilibrium were tested with a Wilcoxon rank sign test and run for 10,000 iterations. We applied a Bonferroni correction to all p-values to control for multiple testing. We calculated general population genetics statistics such as expected and observed heterozygosity, as well as F_{IS} (inbreeding coefficient) for 2008 and 2018 data using GENETIX v 4.05.2 (Belkhir et al. 2004). We calculated F_{IS} using a permutation test consisting of 1,000 permutations. We estimated Fisher's probability of deviations from Hardy-Weinberg equilibrium using a Markov chain Monte Carlo method (Ayres and Balding 1998) with 10,000 dememorizations steps, 1,000 batches, and 5,000 iterations in the GENEPOP v. 4.7.5 (Raymond and Rousset 1995, Rousset 2008). We applied a Bonferroni correction to all Fisher's probabilities to control for multiple testing.

Genetic differentiation and spatial structure

We calculated pairwise F_{ST} (fixation index) between all paired 2008 and 2018 sites, and associated p-values, using an exact G-test with default settings in GENEPOP v.4.7.5. To visualize changes in genetic differentiation between 2008 and 2018 data, we used a factorial correspondence analysis (FCA) performed in GENETIX v. 4.05.2. We included all Southern California sites analyzed in Johannsson et al. (2015) in the FCA analysis to visualize how the 2018 sites were ordered compared to 2008 ones in the context of a large number of sites. We estimated individual genetic coancestry using STRUCTURE v. 2.3.4 for all specimen in 2018 and 2008 samples (Pritchard et al. 2000). Given that the 2008 samples in our study belonged to two of the genetic coancestry clusters identified in Johansson et al. (2015), we ran the current analysis for K=2-3 with 2008 and 2018 samples run separately. The run at K=3 revealed no additional structure, so K=2 was used in this analysis. We used parameters that included an admixture model with allele frequencies correlated across populations and an initial burn-in period of 250,000 and 750,000 reps post burn-in. To compare 2008 and 2018 samples, we produced bar plots in R v.3.6.2 (R Core Team 2019) of individual proportion assignment to each cluster (K=2) and visually compared pairwise differences for each site.

RESULTS

Disturbance quantification with Landsat and temperature analysis

Giant kelp canopy coverage varied on both intra- and interannual time scales at all locations from 2008 to 2018. With marine heatwave events from 2014 to 2016, all locations showed seasonal reductions in canopy coverage, especially Catalina Island and Carlsbad (Fig. 2). Reduction in canopy biomass was variable among locations in both magnitude and duration. Leo Carrillo had the lowest peak biomass values in the summers of 2015 and 2016, but recovered quickly in the summer of 2017, never declining below 17% of maximum biomass. Following the ENSO event, Leo Carrillo reached maximum biomass in the summer of 2018 (Fig. 2). Catalina Island and Carlsbad, however, showed substantial reductions in biomass for extended periods (Fig. 2; Table 2); Catalina experienced two and a half years with biomass below 10% of the maximum, from summer 2014 to the end of 2016 (winter) (Fig. 2; Table 2). Some canopy recovery occurred on Catalina in the spring of 2017 but remained below 32% of the maximum biomass observed in the period studied (Fig. 2). Carlsbad had reduced biomass in the fall of 2014 with minimal recovery until the end of the time-series. Peak biomass during this period occurred in the fall of 2015 and summer of 2018 (11% of max, 30 km buffer), but remained well below pre-disturbance canopy coverage (Fig. 2; Table 2). Correlations were high ($R > 0.94$) between 5, 15, and 30 km buffers for all locations (Table 2), suggesting that canopy changes were synchronous at spatial scales of at least 30 km.

Between sampling periods from 2008 to 2018, all three locations experienced multiple heatwaves (as defined by Hobday et al., 2016) and exceeded periods above the conservative temperature tolerance for giant kelp of 22 °C (Table 2). However, locations varied in the quantity and duration of both MHWs and events ≥ 22 °C. Between 2014 and 2016, Leo Carrillo had a total of 15 MHW events, in which 43% of days met the criteria for marine heatwaves (Hobday et al., 2016). The longest event in Leo Carrillo was 93 days long, from January to March of 2015, but was half as long as Catalina and Carlsbad's' longest events (Fig. 2; Table 2). From 2008 to 2018, Leo Carrillo had several events that exceeded 22 °C, the longest of which lasted 52 days in April and October of 2015. Catalina experienced similar trends in the total number of MHW events and heatwave days but had a long MHW event that well exceeded that of Leo Carrillo and Carlsbad. This MHW event lasted for 235 days from the summer of 2014 to the spring of 2015 (Table 2). Catalina also had multiple events that exceeded 22 °C, and the longest was of the same duration as Leo Carrillo (Table 2). Carlsbad also showed similar trends in the total number of MHW events, but experienced more days in marine heatwaves, with the longest lasting 188 days from the fall of 2014 to spring of 2015. The number of days exceeding species threshold well exceeded Catalina and Leo Carrillo, with the longest-lasting for 76 days from August to October of 2015 (Table 2).

Temporal analysis of genetic diversity

Despite canopy coverage loss at all sites and substantial reductions on Catalina and Carlsbad, all five sites showed little change in within and among-population genetic diversity following disturbance. The mean difference in standardized allelic richness between 2008-2018 paired sites was 0.43 ± 0.35 (mean alleles per locus ± sd, for n=15, Table 1; Fig. 3). There was no significant change in allelic richness between paired sites across sample periods (Wilcoxon sign test: $p= 0.1875$). Sites retained a high level of allelic richness (mean= 7.84 alleles per locus with n standardized to 15, sd= 0.45), maintaining a pattern of high genetic diversity towards the southern distribution of the species (Johansson et al. 2015). Additionally, BOTTLENECK did not suggest signs of heterozygosity excess in any of the 2008 or 2018 samples (Wilcoxon sign test: $p < 0.34$).

Pairwise comparisons of genetic differentiation between sampling periods showed that there was little change in genetic structure following the ENSO event. Pairwise F_{ST} values were significant, except for Carlsbad (Table 3). The lowest F_{ST} values occurred between the same sites temporally (mean =0.01 ± 0.006), except for the 2018 samples for both Catalina backside sites (CIB and CIR) being lower than either was to their 2008 samples (Table 3). The ordination of microsatellite alleles (FCA) showed paired sites residing adjacent to one another in the same genetic groups (Fig. 4). Catalina remained in a coancestry group containing the northern and southern Channel Islands, while Leo Carrillo and Carlsbad clustered with the Southern California/Baja California group (Fig. 4). Comparison of STRUCTURE results between sample periods revealed that individual genetic assignment remained similar over time (Fig. 5). The majority of individual proportions for Leo Carrillo and Carlsbad assigned to one cluster and Catalina individuals for all sites to a separate cluster.

DISCUSSION

In a pre- and post-disturbance analysis on three giant kelp populations in the Southern California Bight, we observed that despite profound disturbance, from marine heatwave events and temperatures above a 22 □C species threshold, there was no change in allelic richness nor evidence for genetic bottlenecks within kelp beds, and genetic differentiation among them remained unchanged. Additional anecdotal evidence from two large storm surges in 2014 and an increase in the abundance of the invasive alga, *Sargassum horneri* (Marks et al. 2015), suggested Catalina may have received greater disturbance than other locations. However, we reject this notion, as Catalina had more canopy recovery than Carlsbad following disturbance regimes from 2014 to 2016. These results support several hypotheses concerning the maintenance of genetic diversity and structure of giant kelp in southern California and provide a clear example where cryptic genetic diversity was stable despite the demographic decline of the macroscopic stage.

The stability of allelic richness and genetic structure in *M. pyrifera* between 2008 and 2018 implies that populations in this region maintained drift-gene flow equilibrium and have a large effective population size, despite major canopy reductions at Catalina and Carlsbad. The fact that F_{ST} remained lowest within sites, across time, indicates that recruitment occurred mostly from population self-replenishment instead of migration into unoccupied space. The only exception to this was the two backside sites on Catalina (CIR and CIB), where geographic and genetic distance (lower F_{ST}) between sites was minimal. Samples collected in the same year had comparably lower F_{ST} values than temporally spaced samples within the same CIR and CIB site. Given these sites are both on Catalina, and are relatively close (~10km), it is likely that we have insufficient power to detect genetic distances on such small spatial scales. Overall, the observed pattern of stability between three locations (Leo Carrillo, Catalina, and Carlsbad) supports the maintenance of historical gene flow, independent of canopy biomass reductions, suggesting that gene flow occurs intermittently between populations over long periods.

Previous analysis in both the Santa Barbara Channel and Southern California showed that genetic differentiation among giant kelp beds can be predicted mainly by both oceanographic distance and habitat continuity (Alberto et al. 2010, Alberto et al. 2011, Johansson et al. 2015). Given the large swaths of ocean between sites, it is conceivable that habitat discontinuity plays a large role in limiting dispersal probability between Island and continental populations. Additionally, there is evidence of the potential for isolation by environment between giant kelp populations in the Southern California Bight, which may further limit gene flow (Kopczak et al. 1991, Johansson et al. 2015). Given the unaltered gene flow between our populations, we can infer that to maintain drift-gene flow equilibrium populations must have maintained a high effective population size during disturbance events to counteract the effects of drift.

The effect of drift is most profound in isolated populations with small numbers of individuals and is most easily detected as a reduction in allelic richness (Frankham 2005). Our study showed that all populations maintained their historic allelic richness, with little evidence for a genetic bottleneck, despite variation in canopy biomass reductions. Our Landsat and temperature analyses showed that Leo Carrillo experienced lesser disturbance than both Carlsbad and Catalina. Leo Carrillo retained at least some canopy during disturbance regimes, while Carlsbad and Catalina showed major reductions for more than two and a half years. Recruitment and maintenance of large effective population size at Leo Carrillo can conceivably be explained by remaining adult sporophytes that survived temperature anomalies and recruited spores locally.

The prolonged absence of canopy at both Carlsbad and Catalina, however, makes the mechanism for the maintenance of a high effective population size less clear. Two possible mechanisms, elusive to Landsat census, could explain the maintenance of large effective population size. Given the nature of remote sensing in an aquatic environment, our biomass quantification might be biased as Landsat does not detect sub-surface juvenile sporophytes. It is possible that disturbance was not severe enough to affect sub-surface individuals. Another possibility is that local populations survive extended periods of disturbance through more tolerant microscopic life stages, such as embryonic sporophytes or gametophytes. These stages may resist growth (juvenile and embryonic sporophytes) or reproduction (gametophytes) until conditions return favorable for canopy-forming sporophytes (Ladah et al., 1999, Kinlan et al. 2003, Carney and Edwards 2006, Ladah and Zertuche-González 2007).

A unique aspect of the 2015/16 ENSO event is that it was not associated with the onset of large storm surges throughout the region (Reed et al. 2016), which is a major factor controlling the presence of giant kelp (Cavanaugh et al. 2011; Young et al. 2016). However, anecdotal evidence of storm surge impacts on Catalina in 2014, along with the delay of recovery despite favorable growth conditions in the spring of 2015 hints towards the absence of at least shallow subsurface individuals. Storms would have likely removed shallow sub-surface individuals, increased sedimentation, and limit recruitment around Catalina. Recruitment on the Island in shallow water could also have been limited by competition with the introduced and more temperature tolerant *Sargassum horneri* (Sullaway 2017). Together these mechanisms imply that surviving giant kelp on the Island would be limited to deeper habitat. Catalina and other island populations of *Macrocystis* can occur much deeper than their continental counterparts, as increased light penetration enables deeper colonization. It is possible that individuals survived deeper, escaping storm effects, competition, and persisted with nutrients influx from internal waves below the thermocline (Zimmerman and Kremer, 1984). Such observations of deep refugia have been previously noted in a Baja, Mexico population of *Macrocystis* during the 1997/1998 ENSO (Ladah Zertuche-González 2004), and in other kelp species (Assis et al. 2016, Graham et al. 2007). Observations of initial deep individuals on the Island following the 2015/16 ENSO, supports this hypothesis (Personal Observation). Canopy typically reforms within six months of removal (Schiel and Foster, 2015), making it less likely that subsurface adult sporophytes were soley responsible for the maintenance of large effective population size, as both Catalina and Carlsbad had minimal canopy for two and a half years. This mechanism is even less likely to have occurred at Carlsbad, where light availability would limit the growth of giant kelp in deeper depths.

Alternatively, genetic diversity may have been maintained through a bank of gametophyte or embryonic sporophyte stages, or both, that delay their development in poor conditions. Evidence of recruitment from such delayed development stages comes from both laboratory and field-based studies and has good support in other brown algae (Edwards 2000, Santelices et al. 2002, Barradas et al. 2011). However, this remains a topic of controversy for giant kelp. Laboratory studies have shown that both gametophyte and juvenile sporophytes can be induced into a state of dormancy, either by limited nutrients or altered light conditions and resume reproduction and growth when sufficient conditions are reestablished (Kinlan et al. 2003). Carney (2011), showed that laboratory-reared giant kelp gametophytes could resume sexual reproduction following a dormancy state for seven months. Although several field studies have demonstrated the role of dormant microscopic stages in sporophyte recruitment during population recovery (Barradas et al. 2011, Carney et al. 2013), it is unknown how long such stages can survive in the wild and how much they contribute to standing population recruitment. Other studies have found no evidence for giant kelp recruitment via delayed microscopic stages (Deysher and Dean 1986, Reed et al. 1997). Reed et al. (1997) compared sporophyte recruitment on substrate out planted for varying lengths of time and suggested recruitment should be higher on substrate out planted longer if dormant stages exist. Their findings suggest the majority of sporophyte recruitment occurred from recently released spores and not dormant stages. Additionally, other field studies suggest gametophyte stages can survive only for short periods in the field (Deysher and Dean 1986, Reed et al. 1994). However, these studies did not look at the contribution of dormant stages to recruitment following extreme environmental conditions, such as ENSO, or the potential for variation in dormancy capacity between different populations.

In comparison to adult sporophytes, the microscopic life-history stages of giant kelp are more tolerant of extreme environmental conditions (Schiel and Foster 2015) and may be critical in population persistence during unfavorable conditions for adult sporophytes. For example, the intertidal red algal species, *Gigartina papillate*, exhibits an alternative life-history stage, that is resistant to herbivores during periods of high grazing pressure, and alternates with a fleshy stage when grazer densities are low (Slocum 1980). Well established examples of such strategies during periods of stress exist in both terrestrial (Pake and Venable 1996) and marine systems (Maier 1990, Dahms 1995), including other kelp taxa (Edwards 2000). Following the 1997-1998 El Nino event, Ladah et al. (1999) reported giant kelp sporophyte recruitment in Baja California, after a 7-month absence of adult sporophytes, with the nearest source population of adult sporophytes over 100 km away (further than spore dispersal capability) (Ladah et al. 1999). Given this, they proposed that recruitment occurred via local gametophyte or embryonic seedbanks. In the current study, although population persistence on Catalina could be explained via a deep refugium, it is much less likely at Carlsbad, where giant kelp depth distribution is limited by high light extinction. This supports that the maintenance of genetic diversity and structure at Carlsbad was maintained by either a juvenile sporophyte or gametophyte seed bank.

Although our study demonstrates that genetic diversity within and among beds of giant kelp in the Southern California Bight was unaltered, it underlines the importance of understanding the mechanisms responsible for population persistence and how they may differ across an individual species distribution (Cavanaugh et al. 2019). For species with limited spatial dispersal, such as many marine and terrestrial plants, temporal dispersal through cryptic life cycle stages, may play an essential role in the persistence of populations. Such may be the case of giant kelp along the southern edge of its northeastern Pacific distribution where population isolation is higher, temperature tolerance limits are closer, and nutrient limitations occur, in particular during ENSO events (Ladah and Zertuche-González 2007). A push to understand the mechanisms of resilience is paramount, as we are already seeing the damaging effects of climate change in both other populations of giant kelp (Wernberg et al. 2015, Arafeh-Dalmau et al. 2019, Cavanaugh et al. 2019), and other kelp taxa (Rogers-Bennett and Catton 2019, Smale 2020). Furthermore, recent work shows the potentially damaging effects of climate change on microscopic stages (Hollarsmith, 2019). However, little is known about how future sea surface temperature models relate to the rocky benthos where these stages reside. More information is needed to understand where seedbanks play a role in population persistence, and how long they can survive without a large adult population to replenish them.

With such uncertainty of the impact of climate change on kelp systems, gametophyte banking is a valuable tool that may aid in both the restoration of future populations and further understanding of cryptic life-history stages (Wade et al. 2020). By housing kelp gametophytes and other macroalgal taxa in dormant stages, we can maintain a genetically diverse collection of individuals, that can be used to reseed threatened populations. This approach has already been implemented in terrestrial seed collections held by many botanical institutions (Dooren 2010) but is still in its infancy in their marine analogs. Our study highlights the need to establish such collections while genetic diversity remains. These collections can be used in both laboratory and field-based studies to understand variability in the niches of microscopic stages and can help target specific traits that may promote resilience for future populations (Coleman et al. 2020).

Although tools such as gametophyte seed banking are promising, genetic monitoring is still an underutilized tool in marine conservation (Reynolds et al. 2017, Manent et al. 2020). Currently, there are few studies to date have utilized temporal genetic analyses in important land plants, let alone marine macrophytes (i.e., Gurgel et al., 2020). Such data, used in conjunction with other methods, such as remote sensing and in-situ survey, can decrease the uncertainty associated with isolated studies. In doing so, wildlife managers can more accurately detect where vulnerable populations exist and what traits drive resilience.

FIGURES

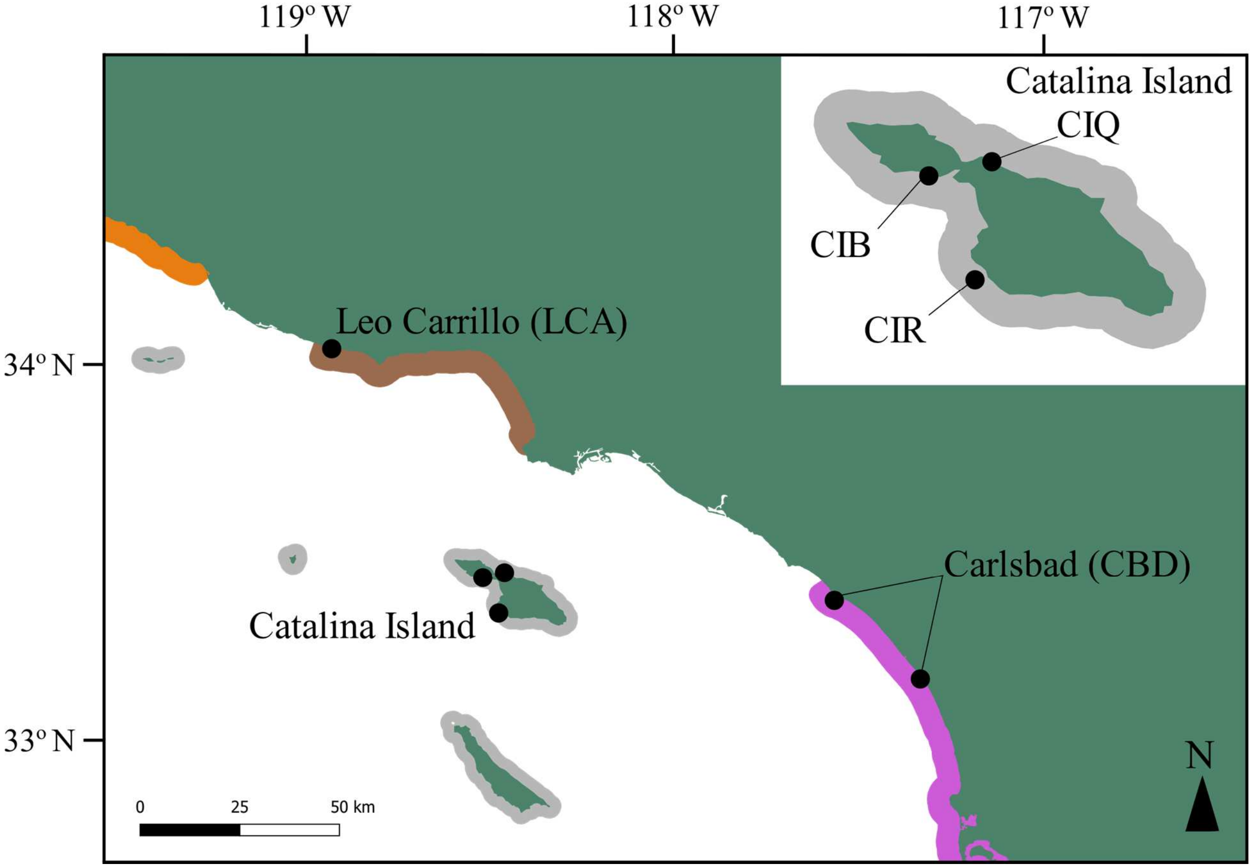

Figure 1. Map showing *M. pyrifera* resampled locations in the Southern California Bight. Colors represent three genetic coancestry groups described by Johansson et al. (2015), including one region of admixture (brown): Continental Santa Barbara (orange), Northern and Southern Channel Islands (gray) and Southern California/Baja California (purple) groups. The 2018 sampl location for Carlsbad is located 30 km SE of the 2008 location. Note that giant kelp beds do not occur continuously along the colored regions.

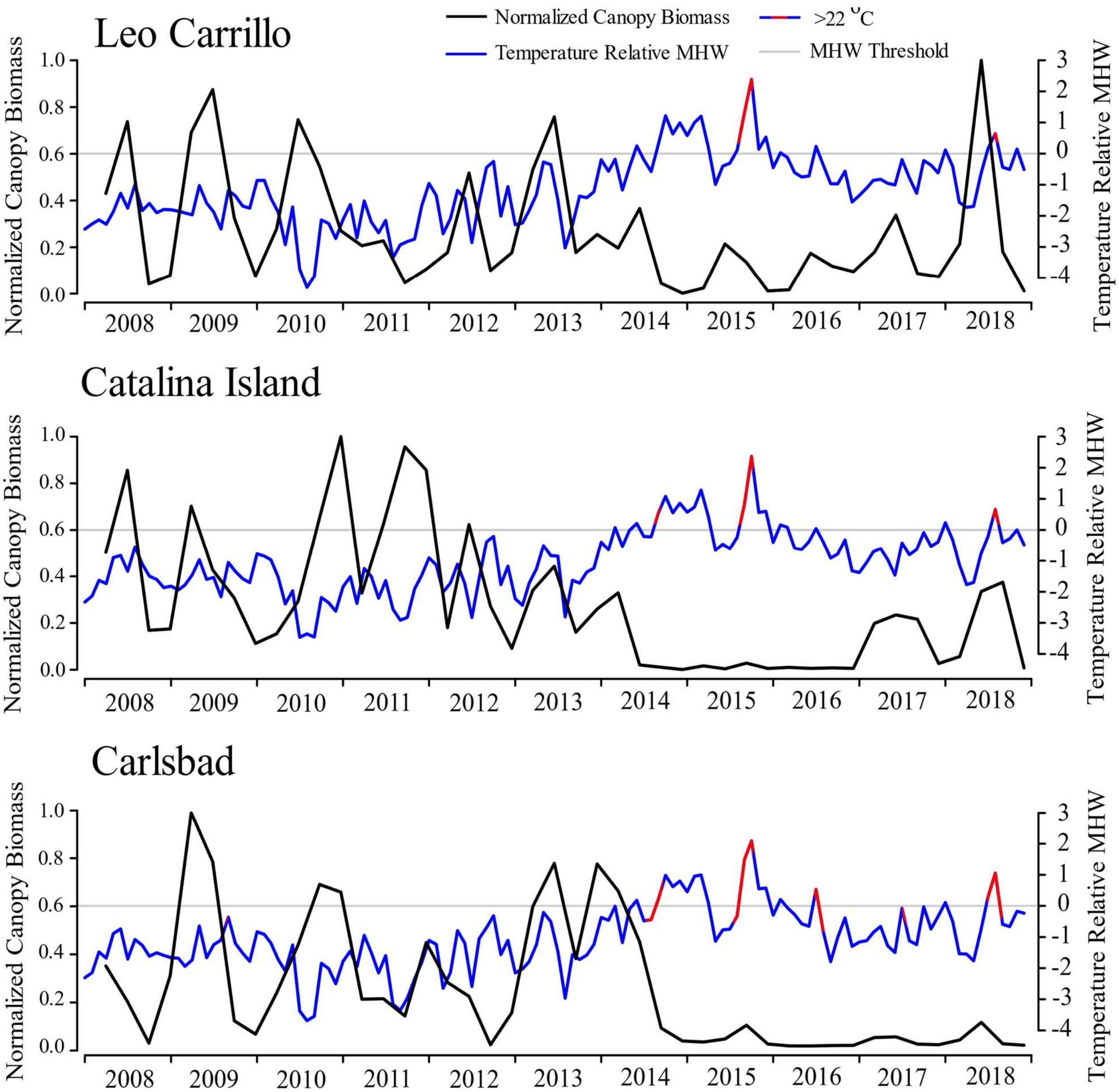

Figure 2. Normalized *M. pyrifera* canopy Biomass (30 km) and Marine Heatwave (MHW) events from 2008 to 2018/19 for Leo Carrillo, Catalina Island (averaged over different locations), and Carlsbad (averaged over 2008 and 2018/19 locations). Canopy biomass is shown quarterly and normalized by the quarter of maximum biomass from 2008 to 2018/19 (black). Temperatures (Blue) are shown as monthly means relative to the MHW threshold (gray). Shown in red are periods when the mean monthly temperature exceeded a conservative species tolerance threshold of 22 □C.

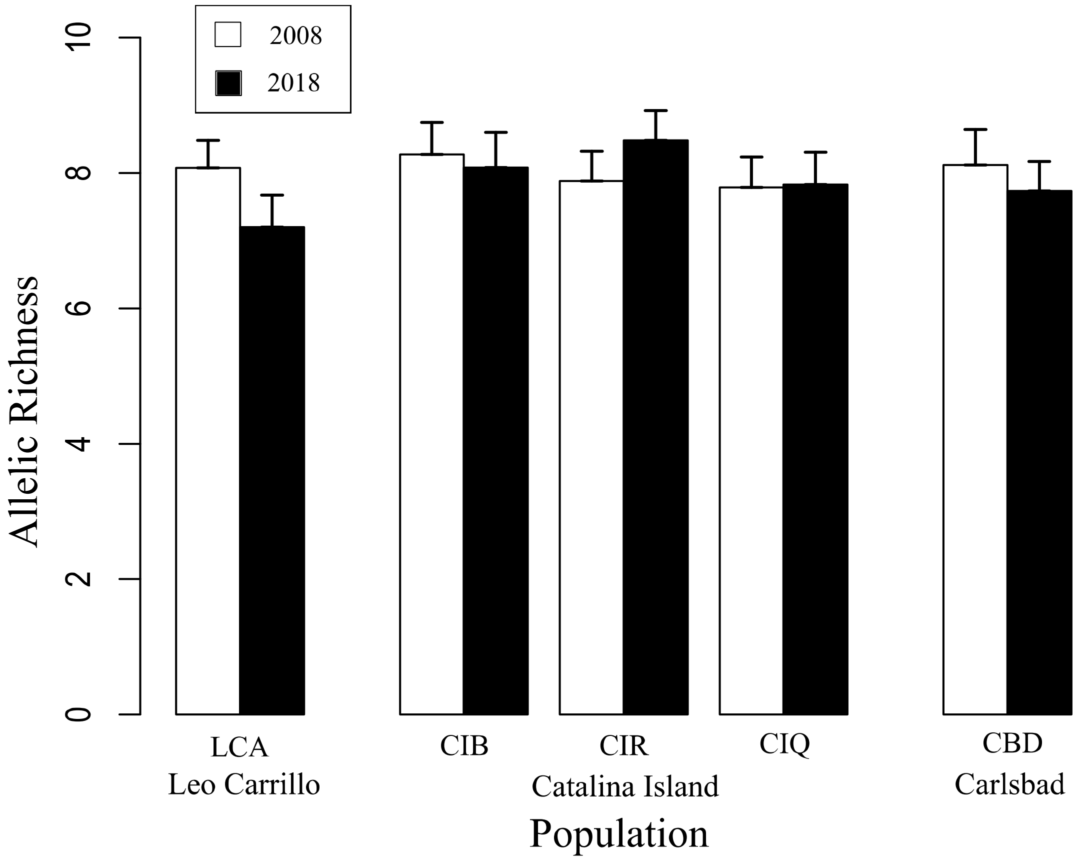

Figure 3. Pairwise comparisons of standardized microsatellite allelic richness (n=15) between 2008 and 2018/19 *M. pyrifera* samples at each of the three locations sampled in Southern California. No significant change in allelic richness was found between sampling periods (Wilcoxon: p= 0.1875)

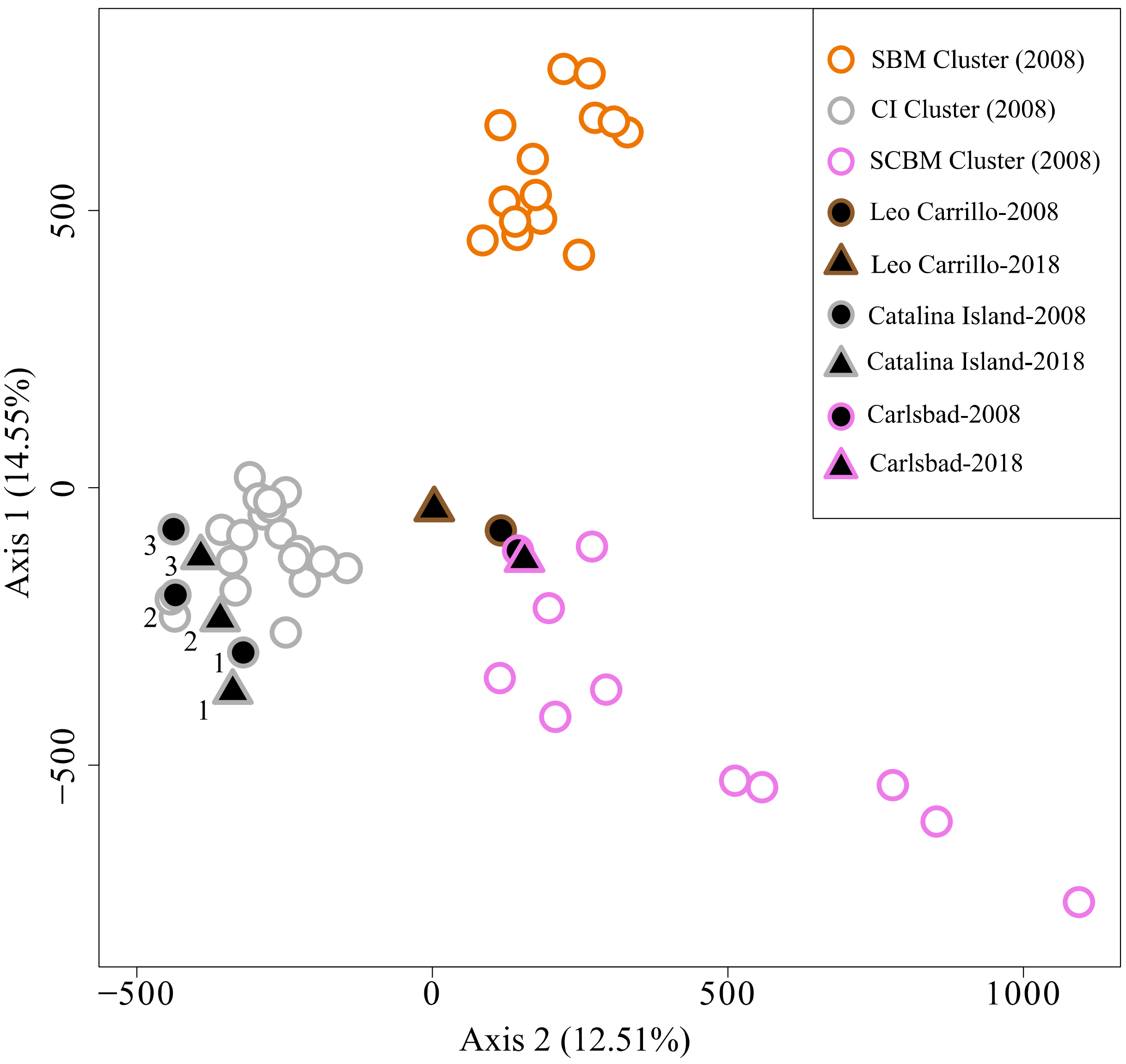

Figure 4. Factorial Correspondence Analysis (FCA) of *M. pyrifera* microsatellite alleles using GENETIX v. 4.05.2. 2008 and 2018/19 samples represented with circles and triangles, respectively. Black filled symbols are for 2008-2018/19 specific comparisons made in this study while open symbols are for additional 2008 samples from Johansson et al. (2015) used here to represent the spatial genetic structure of sites compared here in the overall regional context. The outline color in symbols represents the genetic coancestry clusters identified in Johansson et al. (2015) using STRUCTURE. 2008 and 2018/19 samples clustered together, reflecting minor temporal changes in genetic structure.

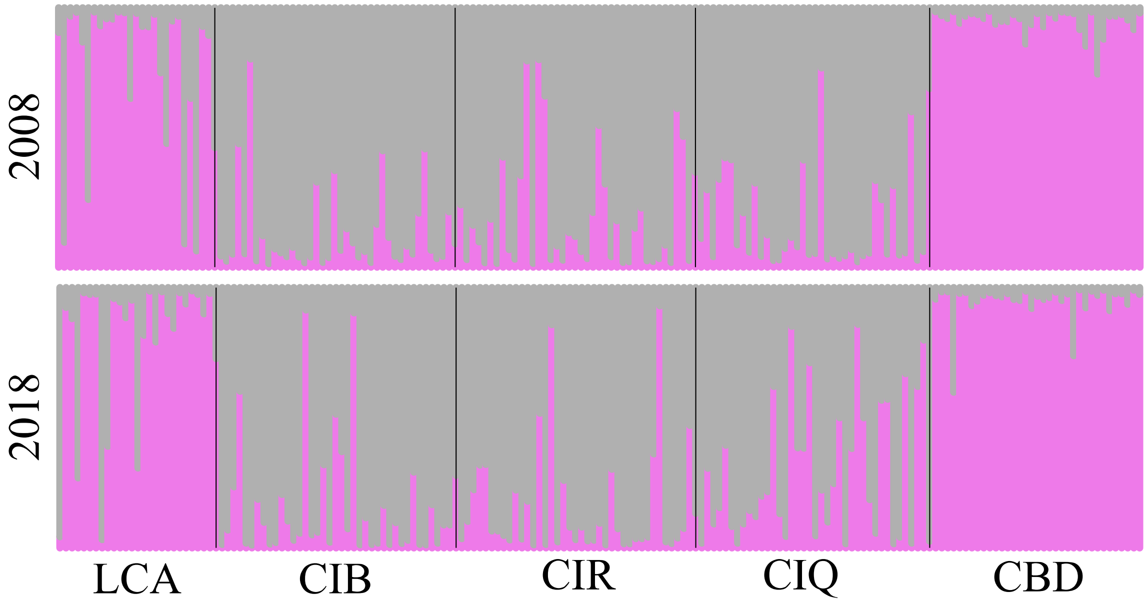

Figure 5. Structure analysis (STRUCTURE v. 2.3.4) comparison of *M. pyrifera* for K=2 clusters of 2008 and 2018/19 sites. Bars represent proportions of individual genetic assignment to two different clusters of genetic coancestry. Individual genetic assignments for all sites did not change temporally during the period studied.

Table 1. Population genetics summary statistics for *M. pyrifera* temporal genetic diversity for five sites in Southern California. The table includes: population with code, sample year, latitude and longitude, sample size (n), standardized allelic richness (A_R), Wilcoxon rank test for bottleneck (P_B), non-biased expected heterozygosity (H_{nb}), observed heterozygosity (H_{obs}), inbreeding coefficient (F_{IS}), Fisher's exact test for Hardy Weinberg equilibrium *p* value (P).

Population (Code)	Sample Year	Latitude	Longitude	n	A_R	P_B	H_{nb}	H_{obs}	F_{IS}	P
Leo Carrillo (LCA-08)	2008	34°2'34.56"N	118°56'4.20"W	27	8.07	0.4218	0.7745	0.6232	0.1991	0.0004*
Leo Carrillo (LCA-18)	2018	34°2'34.56"N	118°56'4.20"W	27	7.12	0.9453	0.7246	0.6211	0.1462	0.0017*
Catalina Island Backside 1 (CIB-08)	2008	33°20'2.76"N	118°29'16.68"W	40	8.28	0.7188	0.7742	0.723	0.0669	0.007
Catalina Island Backside 1 (CIB-19)	2019	33°20'2.76"N	118°29'16.68"W	40	8.08	0.7188	0.7218	0.6553	0.0933	0.0617
Catalina Island Backside 2 (CIR-08)	2008	33°25'45.72"N	118°31'49.32"W	40	7.89	0.7188	0.7308	0.6869	0.0608	0.4604
Catalina Island Backside 2 (CIR-19)	2019	33°25'45.72"N	118°31'49.32"W	40	8.48	0.3438	0.7606	0.6611	0.1325	0.0006*
Catalina Island Quarry (CIQ-08)	2008	33°26'32.51"N	118°28'20.88"W	39	7.79	0.5781	0.7372	0.7415	-0.0059	0.9863
Catalina Island Quarry (CIQ-19)	2019	33°26' 26.16"N	118°27' 45.00"W	39	7.83	0.7188	0.7471	0.6704	0.1042	0.0025*
Carlsbad (CBD-08)	2008	33°22'4.00"N	117°35'24.40"W	35	8.12	0.7188	0.7756	0.7353	0.0527	0.0044*
Carlsbad (CBD-18)	2018	33°9' 27.86"N	117°21' 36.14"W	35	7.73	0.7813	0.7689	0.6868	0.1083	0.0028*

**Bonferonni Correction P_B and P: $P < 0.005$*

Table 2. Summary statistics for *M. pyrifera* canopy biomass (Landsat) and marine heatwave (MHW) analysis. All canopy biomass proportions are based off 30 km buffers, with proportions averaged across all Catalina Island sites, and the 2018/19 and 2008 Carlsbad locations. Note: AMB = annual max biomass

Location (Site)	**Leo Carrillo (LCA)**	**Catalina Is. (CIB, CIQ,CIR)**	**Carlsbad (CBD)**
Buffer Correlations			
5 and 15 km	0.98*	0.98*	0.98*
5 and 30 km	0.94*	0.95*	0.96*
15 and 30 km	0.97*	0.99*	0.99*
Biomass Summary (30 km)			
Min Annual Max Biomass	0.17, 2016	0.01, 2016	0.02, 2016
Mean AMB Pre 2014	0.64 ±0.23	0.76 ± 0.21	0.59 ± 0.27
Mean AMB Post 2014	0.42± 0.34	0.2 ± 0.17	0.19 ± 0.27
Mean AMB 2014-2016	0.25 ± 0.25	0.12 ± 0.18	0.26 ± 0.35
Mean Biomass Post 2014	1.0 Summer 18	0.375, Fall 18	0.12, Summer 18
Duration <10% Max (years)	0	2.5, Summer 14-Winter 16	2.5, Winter 15-Spring 18
Marine Heatwave Summary			
Total MHW Events	22	27	26
Total Duration (days)	599	595	654
MHW Events 2014-2016	15	16	13
Days of MHW 2014-2016	472	481	473
Largest Marine Heatwave			
Date of Max MHW	Jan 5 - Mar 14, 2015	Aug 30, 2014 - Mar 26, 2015	Oct 3, 2014 - Apr 8, 2015
Duration of Max MHW (days)	93	235	188
Mean intensity MHW	2.2	2.08	2.05
Max intensity MHW	3.7	3.39	3.4
Species Threshold Summary			
Total Events	6	3	15
Total Duration (days)	120	117	392
Largest Event >22			
Date	Sep 6 - Oct 10, 2015	Sep 8 - Oct 29, 2015	Aug 14 - Sep 12, 2015
Duration (days)	52	52	76
Mean Temperature (°C)	22.9	23.0	23.4
Max Temperature	24.3	24.3	24.8

** Pearson's Correlation probability <0.05.*

Table 3. Pairwise F_{ST} with G-test of significance computed in GENEPOP v. 4.7.5. All F_{ST} values are for 2008 (-08) and 2018/19 (-18; -19) sites of *M. pyrifera* in Southern California sampled in 2008 and 2018/19, respectively. The lowest F_{ST} values occurred between 2008 and 2018/19 samples for the same site, apart from the two backside sites on Catalina (CIB and CIR).

	LCA-08	LCA-18	CIB-08	CIB-19	CIR-08	CIR-19	CIQ-08	CIQ-19	CBD-08
LCA-18	0.0117*								
CIB-08	0.0535**	0.0530**							
CIB-19	0.0849**	0.0616 **	0.0090*						
CIR-08	0.0603**	0.0385**	0.0169**	0.0133**					
CIR-19	0.0646**	0.0515**	0.0210**	0.0089*	0.0193**				
CIQ-08	0.0849**	0.0774**	0.0325**	0.0472**	0.0422**	0.0428**			
CIQ-19	0.0748**	0.0710**	0.0288**	0.0351**	0.0388**	0.0342**	0.0059*		
CBD-08	0.0617**	0.0689**	0.0692**	0.0868**	0.0772**	0.0754**	0.0668**	0.0623**	
CBD-18	0.0628**	0.0783**	0.0666**	0.0890**	0.0790**	0.0814**	0.0807**	0.0694**	0.0009†

**Significance at the 0.05 probability level. **Significance at probability level <0.001. †Non-significant.*

REFERENCES

Alberto, F. 2009. MsatAllele-1.0: An R package to visualize the binning of microsatellite alleles. *J. Hered.* 100:394–7.

Alberto, F., Raimondi, P.T., Reed, D.C., Coelho, N.C., Leblois, R., Whitmer, A. & Serrão, E.A. 2010. Habitat continuity and geographic distance predict population genetic differentiation in giant kelp. *Ecology*. 91:49–56.

Alberto, F., Raimondi, P.T., Reed, D.C., Watson, J.R., Siegel, D.A., Mitarai, S., Coelho, N. et al. 2011. Isolation by oceanographic distance explains genetic structure for *Macrocystis pyrifera* in the Santa Barbara Channel. *Mol. Ecol.* 20:2543–54.

Alberto, F., Whitmer, A., Coelho, N.C., Zippay, M., Varela-Alvarez, E., Raimondi, P.T., Reed, D.C. et al. 2009. Microsatellite markers for the giant kelp *Macrocystis pyrifera*. *Conserv. Genet.* 10:1915–7.

Arafeh-Dalmau, N., Montaño-Moctezuma, G., Martinez, J.A., Beas-Luna, R., Schoeman, D.S. & Torres-Moye, G. 2019. Extreme Marine Heatwaves alter kelp forest community near its equatorward distribution limit. *Front. Mar. Sci.* 6:1–18.

Arai, K. 2003. Genetics of the Loach, *Misgurnus anguillicaudatus*: Recent Progress and Perspective. *Folia Biol. (Praha).* 51:107–17.

Arnaud, J.F. & Laval, G. 2004. Stability of genetic structure and effective population size inferred from temporal changes of microsatellite DNA polymorphisms in the land snail *Helix aspersa* (Gastropoda: Helicidae). *Biol. J. Linn. Soc.* 82:89–102.

Assis, J., Coelho, N.C., Lamy, T., Valero, M., Alberto, F. & Serrão, E.Á. 2016. Deep reefs are climatic refugia for genetic diversity of marine forests. *J. Biogeogr.* 43:833–44.

Athrey, G., Barr, K.R., Lance, R.F. & Leberg, P.L. 2012. Birds in space and time: Genetic changes accompanying anthropogenic habitat fragmentation in the endangered black-capped vireo (*Vireo atricapilla*). *Evol. Appl.* 5:540–52.

Ayres, K.L. & Balding, D.J. 1998. Measuring departures from Hardy-Weinberg: A Markov chain Monte Carlo method for estimating the inbreeding coefficient. *Heredity (Edinb).* 80:769–77.

Banzon, V., Smith, T.M., Mike Chin, T., Liu, C. & Hankins, W. 2016. A long-term record of blended satellite and in situ sea-surface temperature for climate monitoring, modeling and environmental studies. *Earth Syst. Sci. Data*. 8:165–76.

Barcia, A.R., López, G.E., Hernández, D. & García-Machado, E. 2005. Temporal variation of the population structure and genetic diversity of *Farfantepenaeus notialis* assessed by allozyme loci. *Mol. Ecol.* 14:2933–42.

Barradas, A., Alberto, F., Engelen, A.H. & Serrão, E.A. 2011. Fast sporophyte replacement after removal suggests banks of latent microscopic stages of *Laminaria Ochroleuca* (phaeophyceae) in tide pools in northern Portugal. *Cah. Biol. Mar.* 52:435–9.

Belkhir, K., Borsa P.,Chikhi L., Raufaste N., B.F. 2004. GENETIX logiciel sous Windows TM pour la genetique des populations. Laboratoire Génome Populations Interactions CNRS UMR 5000. Universite de Montpellier II, Montpellier, France.

Bell, T.W., Allen, J.G., Cavanaugh, K.C. & Siegel, D.A. 2020. Three decades of variability in California's giant kelp forests from the Landsat satellites. *Remote Sens. Environ.* 238:0–1.

Bell, T.W., Cavanaugh, K.C., Reed, D.C. & Siegel, D.A. 2015. Geographical variability in the controls of giant kelp biomass dynamics. *J. Biogeogr.* 42:2010–21.

Bellard, C., Bertelsmeier, C., Leadley, P., Thuiller, W. & Courchamp, F. 2012. Impacts of climate change on the future of biodiversity. *Ecol. Lett.* 15:365–77.

Bellinger, M.R., Johnson, J.A., Toepfer, J. & Dunn, P. 2003. Loss of genetic variation in Greater Prairie Chickens following a population bottleneck in Wisconsin, U.S.A. *Conserv. Biol.* 17:717–24.

Bennett, S., Wernberg, T., Connell, S.D., Hobday, A.J., Johnson, C.R. & Poloczanska, E.S. 2016. The "Great Southern Reef": Social, ecological and economic value of Australia's neglected kelp forests. *Marine and Freshwater Research*. pp. 47–56.

Bernhardt, J.R. & Leslie, H.M. 2013. Resilience to Climate Change in Coastal Marine Ecosystems. *Ann. Rev. Mar. Sci.* 5:371–92.

Bertocci, I., Araújo, R., Oliveira, P. & Sousa-Pinto, I. 2015. Potential effects of kelp species on local fisheries.

Blamey, L.K. & Bolton, J.J. 2018. The economic value of South African kelp forests and temperate reefs: Past, present and future. *J. Mar. Syst.* 188:172–81.

Buschmann, A., Graham, M. & Vasquez, J. 2007. Global Ecology of the Giant Kelp Macrocystis. *Oceanography and Marine Biology*. pp. 39–88.

Carney, L.T. 2011. A multispecies laboratory assessment of rapid sporophyte recruitment from delayed kelp gametophytes. *J. Phycol.* 47:244–51.

Carney, L.T., Bohonak, A.J., Edwards, M.S. & Alberto, F. 2013. Genetic and experimental evidence for a mixed-age, mixed-origin bank of kelp microscopic stages in southern California. *Ecology*. 94:1955–65.

Carney, L.T. & Edwards, M.S. 2006. Cryptic Processes in the Sea : A Review of Delayed Development in the Microscopic Life Stages of Marine Macroalgae. *Algae*. 21:161–8.

Castorani, M.C.N., Reed, D.C., Alberto, F., Bell, T.W., Simons, R.D., Cavanaugh, K.C., Siegel, D.A. et al. 2015. Connectivity structures local population dynamics: a long□term empirical test in a large metapopulation system. *Ecology*. 96:3141–52.

Castorani, M.C.N., Reed, D.C., Raimondi, P.T., Alberto, F., Bell, T.W., Cavanaugh, K.C., Siegel, D.A. et al. 2017. Fluctuations in population fecundity drive variation in demographic connectivity and metapopulation dynamics. *Proc. R. Soc. B Biol. Sci.* 284:20162086.

Cavanaugh, K.C., Kendall, B.E., Siegel, D.A., Reed, D.C., Alberto, F. & Assis, J. 2013.

Synchrony in dynamics of giant kelp forests is driven by both local recruitment and regional environmental controls. *Ecology*. 94:499–509.

Cavanaugh, K.C., Reed, D.C., Bell, T.W., Castorani, M.C.N. & Beas-Luna, R. 2019. Spatial variability in the resistance and resilience of giant kelp in southern and Baja California to a multiyear heatwave. *Front. Mar. Sci.* 6:1–14.

Cavanaugh, K.C., Siegel, D.A., Kinlan, B.P. & Reed, D.C. 2010. Scaling giant kelp field measurements to regional scales using satellite observations. *Mar. Ecol. Prog. Ser.* 403:13–27.

Cavanaugh, K.C., Siegel, D.A., Reed, D.C. & Dennison, P.E. 2011. Environmental controls of giant-kelp biomass in the Santa Barbara Channel, California. *Mar. Ecol. Prog. Ser.* 429:1–17.

Chung, I.K., Beardall, J., Mehta, S., Sahoo, D. & Stojkovic, S. 2011. Using marine macroalgae for carbon sequestration: A critical appraisal. *J. Appl. Phycol.* 23:877–86.

Claudet, J. & Fraschetti, S. 2010. Human-driven impacts on marine habitats: A regional meta-analysis in the Mediterranean Sea. *Biol. Conserv.* 143:2195–206.

Coleman, M.A., Wood, G., Filbee-dexter, K., Minne, A.J.P., Goold, H.D., Vergés, A., Marzinelli, E.M. et al. 2020. Restore or Redefine: Future Trajectories for Restoration. *Front. Mar. Sci.* 7:1–12.

Dahms, H.U. 1995. Dormancy in the Copepoda - an overview. *Hydrobiologia*. 306:199–211.

Davidson, A.D., Campbell, M.L., Hewitt, C.L. & Schaffelke, B. 2015. Assessing the impacts of nonindigenous marine macroalgae: An update of current knowledge. *Bot. Mar.* 58:55–79.

Dayton, P.K., Tegner, M.J., Parnell, P.E. & Edwards, P.B. 1992a. Temporal and spatial patterns of disturbance and recovery in a kelp forest community. *Ecol. Monogr.* 62:421–45.

Dayton, P.K., Tegner, M.J., Parnell, P.E., Edwards, P.B., Monographs, E., Sep, N. & Edwards, B. 1992b. Temporal and spatial patterns of disturbance and recovery in a kelp forest community. *Ecol. Monogr.* 62:421–45.

DeFaveri, J. & Merilä, J. 2015. Temporal stability of genetic variability and differentiation in the three-spined stickleback (*Gasterosteus aculeatus*). *PLoS One*. 10.

Deysher, L.E. & Dean, T.A. 1986. In situ recruitment of sporophytes of the giant kelp, *Macrocystis pyrifera* (L.) C.A. Agardh: Effects of physical factors. *J. Exp. Mar. Bio. Ecol.* 103:41–63.

Dooren, T. Van 2010. Banking Seed : Use and Value in the Conservation of Agricultural Diversity. *Sci. Cult. (Lond).* 18:4:373–95.

Duggins, D.O., Simenstad, C.A. & Estes, J.A. 1989. Magnification of secondary production by kelp detritus in coastal marine ecosystems. *Science (80-.).* 245:170–3.

Edwards, M.S. 2000. The role of alternate life-history stages of a marine macroalga: A seed bank analogue? *Ecology*. 81:2404–15.

FAO 2018. The State of the World Fisheries and Aquaculture 2018 - Meeting the sustainable development goals. Rome. Licence: CC BY:NC:SA 3.0 IGO. 227 pp.

Frankham, R. 2005. Genetics and extinction. *Biol. Conserv.* 126:131–40.

Garciá Molinos, J., Halpern, B.S., Schoeman, D.S., Brown, C.J., Kiessling, W., Moore, P.J., Pandolfi, J.M. 2016. Climate velocity and the future global redistribution of marine biodiversity. *Nat. Clim. Chang.* 6:83–8.

Gaylord, B., Reed, D.C., Raimondi, P.T. & Washburn, L. 2006. Macroalgal spore dispersal in coastal environments: Mechanistic insights revealed by theory and experiment. *Ecol. Monogr.* 76:481–502.

Gaylord, B., Reed, D.C., Raimondi, P.T., Washburn, L. & McLean, S.R. 2002. A physically based model of macroalgal spore dispersal in the wave and current-dominated nearshore. *Ecology*. 83:1239–51.

Graham, M.H., Kinlan, B.P., Druehl, L.D., Garske, L.E. & Banks, S. 2007. Deep-water kelp refugia as potential hotspots of tropical marine diversity and productivity. *Proc. Natl. Acad. Sci.* 104:16576–80.

Gurgel, C.F.D., Camacho, O., Minne, A.J.P., Wernberg, T. & Coleman, M.A. 2020. Marine Heatwave Drives Cryptic Loss of Genetic Diversity in Underwater Forests. *Curr. Biol.* 0:1–8.

Hargarten, H.L., Johansson, M.L., Reed, D.C., Coelho, N.C., Siegel, D.A. & Alberto, F. 2019. Seascape genetics of the stalked kelp *Pterygophora californica* and comparative population genetics in the Santa Barbara Channel. *J. Phycol.* 56:110–20.

Heath, D.D., Busch, C., Kelly, J. & Atagi, D.Y. 2002. Temporal change in genetic structure and effective population size in steelhead trout (*Oncorhynchus mykiss*). *Mol. Ecol.* 11:197–214.

Hernández-Carmona, G., Hughes, B. & Graham, M.H. 2006. Reproductive longevity of drifting kelp *Macrocystis pyrifera* (Phaeophyceae) in Monterey Bay, USA. *J. Phycol.* 42:1199–207.

Hijmans, R.J., Williams, E. & Vennes, C. 2019. geosphere: Spherical Trigonometry. R package version 1.5-10.

Hobday, A.J. 2000a. Persistence and transport of fauna on drifting kelp (*Macrocystis pyrifera* (L.) C. Agardh) rafts in the Southern California Bight. *J. Exp. Mar. Bio. Ecol.* 253:75–96.

Hobday, A.J. 2000b. Abundance and dispersal of drifting kelp Macrocystis pyrifera rafts in the Southern California Bight. *Mar. Ecol. Prog. Ser.* 195:101–16.

Hobday, A.J., Alexander, L. V., Perkins, S.E., Smale, D.A., Straub, S.C., Oliver, E.C.J., Benthuysen, J.A. et al. 2016. A hierarchical approach to defining marine heatwaves. *Prog. Oceanogr.* 141:227–38.

Hoffman, E.A., Schueler, F.W. & Blouin, M.S. 2004. Effective population sizes and temporal stability of genetic structure in *Rana pipiens*, the northern leopard frog. *Evolution (N. Y).* 58:2536–45.

Holt, R., Kwok, A. & Dorken, M.E. 2020. Increased spatial-genetic structure in a population of the clonal aquatic plant *Sagittaria latifolia* (Alismataceae) following disturbance. *Heredity (Edinb).* 124:514–23.

Hughes, A.R., Inouye, B.D., Johnson, M.T.J., Underwood, N. & Vellend, M. 2008. Ecological consequences of genetic diversity. *Ecol. Lett.* 11:609–23.

Johansson, M.L., Alberto, F., Reed, D.C., Raimondi, P.T., Coelho, N.C., Young, M.A., Drake, P.T. et al. 2015. Seascape drivers of *Macrocystis pyrifera* population genetic structure in the northeast Pacific. *Mol. Ecol.* 24:4866–85.

Johansson, M.L., Raimondi, P.T., Reed, D.C., Coelho, N.C., Serrão, E.A. & Alberto, F.A. 2013. Looking into the black box: Simulating the role of self-fertilization and mortality in the genetic structure of *Macrocystis pyrifera*. *Mol. Ecol.* 22:4842–54.

Johnson, M.T.J., Lajeunesse, M.J. & Agrawal, A.A. 2006. Additive and interactive effects of plant genotypic diversity on arthropod communities and plant fitness. *Ecol. Lett.* 9:24–34.

Johnston, E.L. & Roberts, D.A. 2009. Contaminants reduce the richness and evenness of marine communities: A review and meta-analysis. *Environ. Pollut.* 157:1745–52.

Kinlan, B.P., Graham, M.H., Sala, E. & Dayton, P.K. 2003. Arrested development of giant kelp (*Macrocystis pyrifera*, Phaeophyceae) embryonic sporophytes: A mechanism for delayed recruitment in perennial kelps? *J. Phycol.* 39:47–57.

Kopczak, C.D., Zimmerman, R.C. & Kremer, J.N. 1991. Variation in Nitrogen Physiology and Growth Among Geographically Isolated Populations of the Giant Kelp, *Macrocystis Pyrifera* (Phaeophyta). *J. Phycol.* 27:149–58.

Krumhansl, K.A., Okamoto, D.K., Rassweiler, A., Novak, M., Bolton, J.J., Cavanaugh, K.C., Connell, S.D. et al. 2016. Global patterns of kelp forest change over the past half-century. *Proc. Natl. Acad. Sci. U. S. A.* 113:13785–90.

Krumhansl, K.A. & Scheibling, R.E. 2012. Production and fate of kelp detritus. *Mar. Ecol. Prog. Ser.* 467:281–302.

Ladah, L.B. & Zertuche-González, J.A. 2004. Giant kelp (*Macrocystis pyrifera*) survival in deep water (25-40 m) during El Niño of 1997-1998 in Baja California, Mexico. *Bot. Mar.* 47:367–72.

Ladah, L.B. & Zertuche-González, J.A. 2007. Survival of microscopic stages of a perennial kelp (*Macrocystis pyrifera*) from the center and the southern extreme of its range in the Northern Hemisphere after exposure to simulated El Niño stress. *Mar. Biol.* 152:677–86.

Ladah, L.B., Zertuche-González, J.A. & Hernandez-Carmona, G. 1999. Giant kelp (*Macrocystis pyrifera*, Phaeophyceae) recruitment near its southern limit in Baja California after mass disappearance during ENSO 1997-1998. *J. Phycol.* 35:1106–12.

Macaya, E.C. 2010. Phylogeny, connectivity and dispersal patterns of the giant kelp *Macrocystis pyrifera* (Phaeophyceae). Victoria University of Wellington, 175 pp.

Maier, G. 1990. Spatial distribution of resting stages, rate of emergence from diapause and times to adulthood and to the appearance of the first clutch in 3 species of cyclopoid copepods.

Hydrobiologia. 206:11–8.

Manent, P., Bañolas, G., Alberto, F., Curbelo, L., Espino, F. & Tuya, F. 2020. Long-term seagrass degradation: Integrating landscape, demographic, and genetic responses. *Aquat. Conserv. Mar. Freshw. Ecosyst.* 30:1111–20.

Mann, K.H. 1973. Seaweeds: Their productivity and strategy for growth. *Science.* 182:975–81.

Marks, L.M., Reed, D.C. & Obaza, A.K. 2017. Assessment of control methods for the invasive seaweed *Sargassum horneri* in California, USA. *Manag. Biol. Invasions*. 8:205–13.

Marks, L.M., Salinas-Ruiz, P., Reed, D.C., Holbrook, S.J., Culver, C.S., Engle, J.M., Kushner, D.J. 2015. Range expansion of a non-native, invasive macroalga *Sargassum horneri* (Turner) C. Agardh, 1820 in the eastern Pacific. *BioInvasions Rec.* 4:243–8.

McCauley, D.J., Pinsky, M.L., Palumbi, S.R., Estes, J.A., Joyce, F.H. & Warner, R.R. 2015. Marine defaunation: Animal loss in the global ocean. *Science.* 347:247-254.

North, W.J. 1994. Review of Macrocystis Biology. *Biol. Econ. algae*. 447–527.

Oliver, E.C.J., Donat, M.G., Burrows, M.T., Moore, P.J., Smale, D.A., Alexander, L. V., Benthuysen, J.A. et al. 2018. Longer and more frequent marine heatwaves over the past century. *Nat. Commun.* 9:1324.

Orsini, L., Marshall, H., Cuenca Cambronero, M., Chaturvedi, A., Thomas, K.W., Pfrender, M.E., Spanier, K.I. et al. 2016. Temporal genetic stability in natural populations of the waterflea *Daphnia magna* in response to strong selection pressure. *Mol. Ecol.* 25:6024–38.

Pake, C.E. & Venable, D.L. 1996. Seed banks in desert annuals: Implications for persistence and coexistence in variable environments. *Ecology*. 77:1427–35.

Perrier, C., Guyomard, R., Bagliniere, J.L., Nikolic, N. & Evanno, G. 2013. Changes in the genetic structure of Atlantic salmon populations over four decades reveal substantial impacts of stocking and potential resiliency. *Ecol. Evol.* 3:2334–49.

Piry, S., Luikart, G. & Cornuet, J.M. 1999. BOTTLENECK: A computer program for detecting recent reductions in the effective population size using allele frequency data. *J. Hered.* 90:502–3.

Pritchard, J.K., Stephens, M. & Donnelly, P. 2000. Inference of population structure using multilocus genotype data. *Genetics*. 155:945–59.

Rahel, F.J. & Olden, J.D. 2008. Assessing the effects of climate change on aquatic invasive species. *Conserv. Biol.* 22:521–33.

Reed, D.C., Washburn, L., Rassweiler, A., Miller, R., Bell, T. & Harrer, S. 2016. Extreme warming challenges sentinel status of kelp forests as indicators of climate change. *Nat. Commun.* 7:13757.

Reed, D.C. 1990. The effects of variable settlement and early competition on patterns of kelp recruitment. *Ecology*. 71:776–87.

Reed, D.C., Amsler, C.D. & Ebeling, A.W. 1992. Dispersal in kelps: factors affecting spore swimming and competency. *Ecology*. 73:1577-1585

Reed, D.C., Anderson, T.W., Ebeling, A.W. & Anghera, M. 1997. The role of reproductive synchrony in the colonization potential of kelp. *Ecology*. 78:2443–57.

Reed, D.C., Lewis, R.J. & Anghera, M. 1994. Effects of an open-coast oil-production outfall on patterns of giant kelp (*Macrocystis pyrifera*) recruitment. *Mar. Biol.* 120:25–31.

Reed, D.C., Schroeter, S.C. & Raimondi, P.T. 2004. Spore supply and habitat availability as sources of recruitment limitation in the giant kelp *Macrocystis pyrifera* (Phaeophyceae). *J. Phycol.* 40:275-284

Reusch, T.B.H., Ehlers, A., Hämmerli, A. & Worm, B. 2005. Ecosystem recovery after climatic extremes enhanced by genotypic diversity. *Proc. Natl. Acad. Sci. U. S. A.* 102:2826–31.

Reynolds, L.K., Stachowicz, J.J., Hughes, A.R., Kamel, S.J., Ort, B.S. & Grosberg, R.K. 2017. Temporal stability in patterns of genetic diversity and structure of a marine foundation species (*Zostera marina*). *Heredity (Edinb).* 118:404–12.

Rogers-Bennett, L. & Catton, C.A. 2019. Marine heat wave and multiple stressors tip bull kelp forest to sea urchin barrens. *Sci. Rep.* 9:1-9

Rousset, F. 2008. GENEPOP'007: A complete re-implementation of the GENEPOP software for Windows and Linux. *Mol. Ecol. Resour.* 8:103–6.

Ruzzante, D.E., Taggart, C.T., Doyle, R.W. & Cook, D. 2001. Stability in the historical pattern of genetic structure of Newfoundland cod (*Gadus morhua*). *Conserv. Genet.* 2:257–69.

Saltonstall, K. 2011. Remnant native *Phragmites australis* maintains genetic diversity despite multiple threats. *Conservation Genetics*. 12:1027-1033

Santelices, B., Aedo, D. & Hoffmann, A. 2002. Banks of microscopic forms and survival to darkness of propagules and microscopic stages of macroalgae. *Rev. Chil. Hist. Nat.* 75:547–55.

Schiel, D.R. & Foster, M.S. 2015. The biology and ecology of giant kelp forests. Univ of California Press. 1–395 pp.

Schwartz, M.K., Luikart, G. & Waples, R.S. 2007. Genetic monitoring as a promising tool for conservation and management. *Trends Ecol. Evol.* 22:25–33.

Slocum, C.J. 1980. Differential susceptibility to grazers in two phases of an intertidal alga: Advantages of heteromorphic generations. *J. Exp. Mar. Bio. Ecol.* 46:99–110.

Smale, D.A. 2020. Impacts of ocean warming on kelp forest ecosystems. *New Phytol.* 225:1447–54.

Solomon, S., D., Qin, M., Manning, Z., Chen, M., Marquis, K.B., Averyt, M.T., Miller HL et al. 2007. Summary for Policymakers. In: Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. *New York Cambridge Univ. Press*.

Steneck, R., Graham B J Bourque D Corbett J M Erlandson, M.H., Graham, M.H., Bourque, B.J., Corbett, D., Erlandson, J.M., Estes, J.A. et al. 2002. Kelp Forest Ecosystems: Biodiversity, Stability, Resilience and Future. *Environ. Conserv.* 29:436–59.

Sullaway, G. 2017. Assessing the impact of the invasive species, Sargassum horneri, on kelp forest net community production. San Diego State.

Tessier, N. & Bernatchez, L. 1999. Stability of population structure and genetic diversity across generations assessed by microsatellites among sympatric populations of landlocked Atlantic salmon (*Salmo salar*). *Mol. Ecol.* 8:169–79.

Tilman, D., Clark, M., Williams, D.R., Kimmel, K., Polasky, S. & Packer, C. 2017. Future threats to biodiversity and pathways to their prevention. *Nature*. 546:73–81.

Timpane-Padgham, B.L., Beechie, T. & Klinger, T. 2017. A systematic review of ecological attributes that confer resilience to climate change in environmental restoration. *PLoS One*. 12:1–23.

Valero, M., Destombe, C., Mauger, S., Ribout, C., Engel, C.R., Daguin-Thiebaut, C. & Tellier, F. 2011. Using genetic tools for sustainable management of kelps: A literature review and the example of *Laminaria Digitata*. *Cah. Biol. Mar.* 52:467–83.

Vilalta-Navas, A., Beas-Luna, R., Calderon-Aguilera, L.E., Ladah, L., Micheli, F., Christensen, V. & Torre, J. 2018. A mass-balanced food web model for a kelp forest ecosystem near its southern distributional limit in the northern hemisphere. *Food Webs*. 17:e00091.

Schlegel W., R. & J. Smit, A. 2018. heatwaveR: A central algorithm for the detection of heatwaves and cold-spells. *J. Open Source Softw.* 3:821.

Wade, R., Augyte, S., Harden, M., Nuzhdin, S., Yarish, C. & Alberto, F. 2020. Macroalgal germplasm banking for conservation, food security, and industry. *PLoS Biol.* 18:e3000641.

Walker, B., Holling, C.S., Carpenter, S.R. & Kinzig, A. 2004. Resilience, adaptability and transformability in social-ecological systems. *Ecol. Soc.* 9.

Wernberg, T., Bennett, S., Babcock, R.C., Bettignies, T. De, Cure, K., Depczynski, M., Dufois, F. et al. 2015. Climate-driven regime shift of a temperate marine ecosystem. *Science.* 353:169–72.

Wootton, J.T. & Pfister, C.A. 2013. Experimental separation of genetic and demographic factors on extinction risk in wild populations. *Ecology*. 94:2117–23.

Young, M., Cavanaugh, K., Bell, T., Raimondi, P., Edwards, C.A., Drake, P.T., Erikson, L. et al. 2016. Environmental controls on spatial patterns in the long-term persistence of giant kelp in central California. *Ecol. Monogr.* 86:45–60.

Zimmerman, R.C. & Kremer, J.N. 1984. Episodic Nutrient Supply To a Kelp Forest Ecosystem in Southern California. *J. Mar. Res.* 42:591–604.

APPENDICES

Appendix A: R Code for determining normalized canopy biomass from 2008-2018

```
#The following script uses Quarterly Landsat Canopy Biomass data obtained from (Bell et
#al.,2020) to achieve a time series
#of summed canopy biomass proportional to the quarter with the largest biomass between
2008 and 2018. Three buffers of 5, 15 and 30 km are used to select
#cells to sum within a site. See page 12 for more details

#Load R package geosphere v. 10 R Package (Hijmans et al.,2019)
library(geosphere)

options(digits=10)   #Makes sure R grabs all decimals for coordinates

#Reads in Appropriate Data set of Quarterly Landsat Canopy Wet Biomass (kg) from 2008 to
2018
#buffered at 30 km around each sample location (Bell et al., 2020).

#Leo Carrillo data
data<-read.delim("Leo_Carrillo_2008_2018_RevisedHeader.txt",header=T,na.strings = "NaN")

#Catalina Island data
data<-read.delim("Catalina_2008_2018_RevisedHeader.txt",header=T,na.strings = "NaN")

#Carlsbad data
data<-read.delim("CBD_2008_2018_RevisedHeader.txt",header=T,na.strings = "NaN")

#San Mateo data
data<-read.delim("San_Mateo_2008_2018_RevisedHeader.txt",header=T,na.strings = "NaN")

#Number of quarters
nquarts<-ncol(data)-2

#Sample site Coordinates in decimal degrees of Longitude and Latitude respectively. Note
Calculations are done for
#each individual site separately for all three buffers (5,15 and 30 km) using the same
code. See p.12 for rational of
#calculations for each sample location.

longlat<-c(-118.934500, 34.042933) #Leo Carrillo
longlat<-c(-118.450127,33.397824) #Catalina All for 30km buffer
longlat<-c(-118.483167,33.343600) #Catalina Island Backside 1 (CIB)
longlat<-c(-118.530367,33.429367) #Catalina Island Backside 2 (CIR)
longlat<-c(-118.472467,33.442364) #Catalina Island Quarry (CIQ)
longlat<-c(-117.590111,33.367778) #Carlsbad Old (CBD-OLD) (Originally San Mateo (SMA) in
#Johansson et al. (2015))
longlat<-c(-117.360039,33.157740) #Carlsbad New (CBD-NEW)

#Starting here each calculation for the above sample site coordinates is done
#seperatly using their respective data set from above.

#Measuring different distances from central cell
cellCoords<-matrix(c(data$Lon,data$Lat),ncol=2)

spatdis<-distVincentyEllipsoid(longlat,cellCoords)
```

```
#Indexing Only Data within the specified distance (buffer)
data5<-data[spatdis<=5000,] #5 km buffer

data15<-data[spatdis<=15000,] #15 km buffer

data30<-data[spatdis<=30000,] #30 km buffer

#Summing all cells for each buffer seperatly and Normalizing (diving) all values by the
#maximum biomass year from 2008-2018

#5 km buffer
sum5<-apply(data5[,-c(1,2)],2,sum,na.rm=T) #Sum canopy biomass per quarter

PropmaxBiomass5<-sum5/max(sum5) #Normalizing by maximum canopy biomass year

#Repeating for 15 km Buffer
sum15<-apply(data15[,-c(1,2)],2,sum,na.rm=T)

PropmaxBiomass15<-sum15/max(sum15)

#Repeating for 30 km Buffer
sum30<-apply(data30[,-c(1,2)],2,sum,na.rm=T)

PropmaxBiomass30<-sum30/max(sum30)

#Renaming biomass objects and producing a single data frame of total canopy biomass and
normalized biomass for one site
Biomass_5<-sum5
PropBiomass_5<-PropmaxBiomass5

Biomass_15<-sum15
PropBiomass_15<-PropmaxBiomass15

Biomass_30<-sum30
PropBiomass_30<-PropmaxBiomass30

Alldata<-cbind(Biomass_5,PropBiomass_5,PropBiomass_15,Biomass_30,PropBiomass_30)

#Exporting data frame of total canopy biomass and normalized canopy biomass for 5,15 and
#30 km buffers from 2008-2018.
write.csv(Alldata,"All_2008_2018_CBD_Processed.csv") #Using CBD-NEW as an example

#####The script above was repeated for each sample site coordinate with their respective
datasets.#####

#Note: Post processing occurred in excel to achieve mean canopy biomass proportions for
#Catalina and Carlsbad Sites.
#A mean proportion across all sites (CIB,CIR,CIQ) was taken for 5 and 15 km buffers on
#Catalina and 5,15 and 30 km buffers for Carlsbad (CBD-NEW and CBD-OLD). See
#page 12 for more information.
```

Appendix B: R code for sea surface temperature analysis and Figure 3 plot

```
#The following code uses daily temperature from January 1984 to December 2019 obtained
#from the National Climatic Data Center Optimal Interpolation Sea Surface Temperature
#(OISST) dataset (Banzon et al.,2016) and
#the heatwaveR v. 0.4.2 R package (Schlegel and Smit, 2018) to detect Marine Heatwave
#events (MHW) based on Hobday et al. (2016) definition
#and detects events occurring above a conservative species threshold of 22 degrees
#celcius. This code is also used to plot Figure 3.

#Load necessary packages
library(heatwaveR)
library(lubridate)
library(dplyr)
options(stringsAsFactors = FALSE)

#Read appropriate daily SST data set
data<-read.delim("dataLCA.txt") #Leo Carrillo

data<-read.delim("dataCI_avg.txt") #Catalina Island

data<-read.delim("dataCBD_avg.txt") #Carlsbad (CBD-OLD and CBD-NEW average)

#Setting time column as a date object
data$t<-as.Date(data$t)

#Produce climatology data Baseline from 1984-2019
dclimdata<-ts2clm(data,x=t,y=temp,climatologyPeriod = c("1984-01-01","2019-12-31"))

#Detect Marine heatwave events
detecteventdata<-detect_event(dclimdata)

#Detect events exceeding a species threshold of 22 degrees Celcius
data22<-exceedance(data,threshold=22)

#Writing a dataframe of all detected MHW events and Species threshold events respectivly
#from 1983-2019
write.csv(detecteventdata$event,"CBD_events.csv") #example location CBD
write.csv(data22$exceedance,"CBD_exceedance.csv") #example location CBD

#Note all descriptive statistics for MHW events and Species Threshold events in Table 2
#were calculated in Microsoft excel using the data frames exported above.

######The folloing code was used to prepare data for Figure 3######

#Indexing detecteventdata object (MHW events) to only include values occuring from 2008-
2018
detecteventdata_08<-droplevels(detecteventdata$climatology[8767:12784,])

############Calculating monthly averages for SST, MHW threshold and Species threshold
#from 2008-2018##########

#creating month column
detecteventdata_08<-detecteventdata_08 %>%
  mutate(month=month(t))

#creating year column
```

```
detecteventdata_08<-detecteventdata_08 %>%
  mutate(year=year(t))

#calculating mean monthy SST
data_temp_bymonth <-detecteventdata_08 %>%
  group_by(year, month) %>%
  summarise(avg_temp = mean(temp))

#calculating mean monthly MHW threshold
data_thresh_bymonth <-detecteventdata_08 %>%
  group_by(year, month) %>%
  summarise(avg_thresh = mean(thresh))

#Combining all above vectors in a single dataframe
data_bymonth<-
cbind(data_temp_bymonth$year,data_temp_bymonth$month,data_temp_bymonth$avg_temp,data_thre
sh_bymonth$avg_thresh)
colnames(data_bymonth)<-c("year","month","temp","thresh") #Renaming Columns

data_bymonth<-as.data.frame(data_bymonth) #setting as class dataframe

#########################################################################

#Adding a new column representing date
data_month<-data_bymonth %>%
  mutate(month2 = as.Date(paste0(year,"-", month,"-15"),"%Y-%m-%d"))

#Adding 22 degrees threshold column
data_month<-data_month %>%
  mutate(sp_thresh=rep(22,132))

#Calculating the difference between mean monthly SST and the MHW threshold
data_month<-data_month %>%
  mutate(d_thresh=temp-thresh)

#Calculating the difference between mean monthly SST and the Species threshold
data_month<-data_month %>%
  mutate(dsp_thresh=temp-sp_thresh)

#Calculating the difference between MHW threshold and species threshold
data_month<-data_month %>%
  mutate(sp_rel_thresh=sp_thresh-thresh)

###################PLOT#########################

#The following code uses the data month data frame created above to create Figure 3.

#Read in proportional canopy biomass data at 30 km buffer. Note Read in one at a time
#when producing plot.

CIdata30km<-read.csv("All_2008_2018_Leo_Carrillo_Processed.csv") #Leo Carrillo

CIdata30km<-read.csv("All_2008_2018_Catalina30kmBuffer_Processed.csv") #Catalina

CIdata30km<-read.csv("All_2008_2018_AVG_SMA_CBD_Processed.csv") #Carlsbad


par(mfrow=c(3,1)) #To produce multiple plots in one figure (run once)
```

```
################################PLOT##########################

#To plot temperature relative to MHW threshold along with time axis and relative
#temperature axis
plot(seq(from=as.Date("2008-01-01"),to=as.Date("2018-12-
31"),by="month"),data_month$d_thresh,type="l",ylim=c(-
4.5,3),lwd=3,col="blue",cex=.25,yaxt="n",bty="n",xaxt="n",ylab="Normalized Canopy
Biomass",xlab="Year")
axis(1, at=seq(from=as.Date("2008-01-01"),to=as.Date("2019-01-
01"),by="year"),labels=seq(from=as.Date("2008-01-01"),to=as.Date("2019-01-
01"),by="year"),lwd=2)
axis(4,at=-4:3,pos=as.Date("2019-01-31"),lwd=2)

abline(a=0,b=1,h=0,lwd=2,col="gray") #Creates gray MHW threshold line

#This line is used to tell when the temperature relative to the MHW threshold was above
#the species threshold
par(new=T)
lines(seq(from=as.Date("2008-01-01"),to=as.Date("2018-12-
31"),by="month"),data_month$sp_rel_thresh)

#Adds proportional biomass data with axis
par(new=T)
plot(seq(from=as.Date("2008-03-31"),to=as.Date("2018-12-
31"),by="quarter"),CIdata30km$PropBiomass_30,lwd=3,bty="n",type="l",axes=F,xlab="",ylab="
",xlim=c(as.Date("2008-01-01"),as.Date("2018-12-31")))
axis(2,pos=as.Date("2008-01-01"),lwd=2)

#Rerun all of the above code minus par() for each site independently until all sites have
#been plotted.
```