Check for updates

RESEARCH ARTICLE

Testing the roles of local adaptation and genetic diversity to improve Giant kelp (*Macrocystis pyrifera*) restoration

Lauren N. Dykman[1,2,3] , S. Clay Steell[2], Jordan B. Bemmels[1,2], Grace K. Melchers[4],
Brian D. Timmer[1,2], Gregory L. Owens[1,2], Christopher J. Neufeld[2,5,6], Julia K. Baum[1,2]

Climate change is causing significant losses of coastal foundation species globally, heightening the need for their restoration. Despite the urgency, it remains unclear if enhancing genetic diversity by using distant source populations will improve restoration outcomes, or if local sources will perform better regardless of their diversity. We conducted a reciprocal transplant with Giant kelp (*Macrocystis pyrifera*) between a warm and cool microclimate 7 km apart in Barkley Sound, British Columbia, and tracked survivorship and growth over 6 months. We seeded kelp gametophyte cultures from the warm and cool site onto small rocks (i.e. "green gravel") in a nursery, then outplanted them into experimental plots nearby the sites where the parents were collected. The number of parents used to make the cultures was also manipulated (two vs. 10) to simulate different levels of genetic diversity (average heterozygosity). Overall, we found inconsistent evidence for local adaptation between microclimates, and possibly signs of maladaptation in kelp from the warm site. Kelp from the more genetically diverse population at the cooler site survived 16% better and grew 8% larger regardless of the outplant site. Kelp grew up to 223% larger at the cool site but had higher mortality due to either urchin grazing or gravel turnover from swell. Surprisingly, kelp produced from two-parent cultures survived 278% better than kelp from 10-parent cultures at 6 months, despite higher observed rates of selfing. Our study provides new insights into factors influencing the restoration of these important temperate coastal foundation species.

Key words: adaptation, climate change, genetic diversity, Giant kelp, green gravel, *Macrocystis pyrifera*, reciprocal transplant, restoration

Implications for Practice

- As ocean warming causes widespread loss of kelp forests, restoration practitioners must increasingly consider the genetic provenance and thermal tolerance of outplanted kelp to ensure success.
- Sourcing kelp from cooler, less-impacted sites may ensure more genetically diverse propagules and better performance even at warm sites, since kelp sourced from warm sites may be maladapted to their environment due to stress and genetic bottlenecks.
- Creating kelp cultures from a greater number of parents reduces self-fertilization rates but may not improve immediate restoration outcomes, although longer-term consequences remain unknown.

Introduction

Climate change is creating drastic, persistent shifts in historical climate patterns that are occurring more rapidly than many species can adapt (IPCC 2022). Although restoration best practices recommend preserving the genetic composition of local populations (Shearer et al. 2009; Aavik & Helm 2018), this strategy is unlikely to ensure long-term climate resilience, as local climates will quickly become unsuitable for local genotypes (Merilä & Hendry 2014; Vranken et al. 2021). Accordingly, recent attention has turned toward sourcing propagules from farther locales where the climate matches a projected future scenario (Harris et al. 2006) as a form of proactive restoration or "future-proofing" populations (Schweitzer et al. 2014; Wood et al. 2019). Specifically, using thermally tolerant genotypes from a warmer climate or microclimate as source material could increase the likelihood that populations at restored sites persist far into the future (Coleman & Wernberg 2020; Pazzaglia et al. 2021). However, the introduction

of genotypes from afar is controversial given the risks of introducing novel genetic material, and many questions remain before this concept can be applied responsibly at scale.

Sourcing propagules for restoration from nearby has been practiced for decades because it is known to enhance restoration success (in the absence of climate change) since species are often adapted to local environmental conditions (Jones 2013; Merilä & Hendry 2014). This practice further avoids the risk of outbreeding depression or loss of local genotypes that comes with sourcing material from afar (Solas et al. 2024). The general wisdom of maximizing genetic diversity and minimizing inbreeding to enhance ecological restoration is not new and is achieved by increasing the number or geographic spread of parents from which propagules are sourced (Hughes & Stachowicz 2004; Miller et al. 2017). Despite the advantages of sourcing propagules locally, no local populations may remain in an area after extreme climatic events (e.g. heatwaves and droughts), or the nearest surviving population may have suffered from the same disturbance as the restoration site, resulting in decreased genetic diversity (Jones 2013). Repopulating degraded sites with stocks of low genetic diversity may create bottlenecks, where genetic drift causes a small population to lose genetic variation that could aid in adaptation to future climate stressors (Barrett & Kohn 1991; Biebach et al. 2016). Genetic drift in bottlenecked populations may also lead to the fixation of recessive deleterious variants that contribute to inbreeding depression (Dussex et al. 2023). To avoid these outcomes, restoration projects usually introduce propagules from many parent individuals to maintain the allelic richness of the source population (Alsuwaiyan et al. 2021), yet this has limited benefit if the source population itself has low allelic richness. To enhance restoration success in the era of climate change, it is still not clear whether propagules should come from a locally adapted population, even in cases of low genetic diversity, or whether propagules should have higher genetic diversity, even if that means sourcing some individuals from further away.

Kelp (brown algae in the Order Laminariales) forms the foundation of rich, productive coastal ecosystems along one-third of the world's coastline (Jayathilake & Costello 2021) and are considered the most vulnerable temperate marine ecosystems to climate change (IPCC 2022). Kelp forests provide complex habitat, act as nurseries for commercial fish species, fuel coastal food webs, produce oxygen, contribute to nutrient cycling, and may be a carbon sink in some places (Graham 2004; Filbee-Dexter & Wernberg 2020; Eger et al. 2023a). Canopy-forming kelps are declining rapidly in many parts of the world, largely due to climate change and alterations to trophic structure caused by the loss of key predator species (Harley et al. 2012; Krumhansl et al. 2016; Rogers-Bennett & Catton 2019). Such declines have raised considerable concern and prompted an increasing number of kelp forest restoration initiatives worldwide (Eger et al. 2023b). Yet, there is still much to learn about how to successfully restore kelp forests. One emerging kelp reintroduction method is "green gravel" which involves "seeding" fragmented kelp gametophytes onto gravel or small rocks, growing the resulting sporophytes in a nursery, then deploying the seeded gravel in the field (Alsuwaiyan et al. 2022). This method has the advantage of not requiring divers to deploy outplants, as the rocks

can be scattered by hand from a boat (Wood et al. 2024). Green gravel is still a relatively new technique that has been developed for some key canopy kelp species but requires development and testing for each new species due to their unique ecologies and life cycles.

To advance understanding of the potential role of local adaptation for kelp restoration, we conducted a reciprocal transplant experiment to examine whether restoration outcomes can be enhanced by: (1) sourcing and culturing kelp from the most local microclimate, and (2) experimentally increasing the genetic diversity of cultured kelp by manipulating the number of parents used for propagation. Simultaneously, we tested the effectiveness of the "green gravel" restoration technique for outplanting Giant kelp (*Macrocystis pyrifera* sensu lato [Macaya & Zuccarello 2010] or *M. tenuifolia* [Lindstrom 2023]). We made three predictions relevant to restoration ecology, none of which are mutually exclusive: (1) kelp sourced from a warm site will survive and grow better in its source (warm) environment than in a cooler transplant site and vice versa due to local adaptation (i.e. significant site × source interaction as in Martin et al. 2021; Becheler et al. 2022; Johnson et al. 2022); (2) kelp sourced from a warm site may show lower survival and growth rates overall due to possible maladaptive inbreeding depression caused by the 2014–2016 marine heatwave, when substantial kelp die-back was observed at warm sites (significant source effect but no significant interaction) (Johnson et al. 2022); and (3) kelp grown from a greater number of mixed parents will survive and grow better than kelp grown from fewer parents due to an increased chance of introducing individuals with alleles suited to the environment and a decreased chance of self-fertilization (significant parent number effect) (Jarne & Charlesworth 1993). This study tests classical theories in restoration ecology and local adaptation to optimize restoration outcomes and examines the potential to leverage local microclimates and genetic diversity to enhance climate resilience of Giant kelp in British Columbia and the Indigenous territories therein.

Methods

Study Sites

The fjords and sounds along the British Columbia coastline inhabited by kelps provide an ideal setting for testing the effects of local microclimate and genetic diversity on adaptation and restoration success. Barkley Sound is a fjord-like embayment on the Pacific coast of Vancouver Island, British Columbia, Canada, spanning the territories of several First Nations (Fig. 1A). Barkley Sound has a natural gradient in sea surface temperature that varies by approximately 3°C along its length during summer months (Starko et al. 2022), with colder (approximately 15°C) average temperatures to the southwest, where the sound opens to the Pacific, and warmer (approximately 18°C) average temperatures to the northeast farther into the sound. A prolonged marine heatwave that occurred in the Northeast Pacific Ocean between 2014 and 2016 ("The Blob") (Rogers-Bennett & Catton 2019), including along the coast of British Columbia, caused a loss of the dominant canopy-forming kelp, Giant kelp (*Macrocystis pyrifera*) and Bull kelp (*Nereocystis luetkeana*), from 40% of sites along a 16 km gradient of Barkley
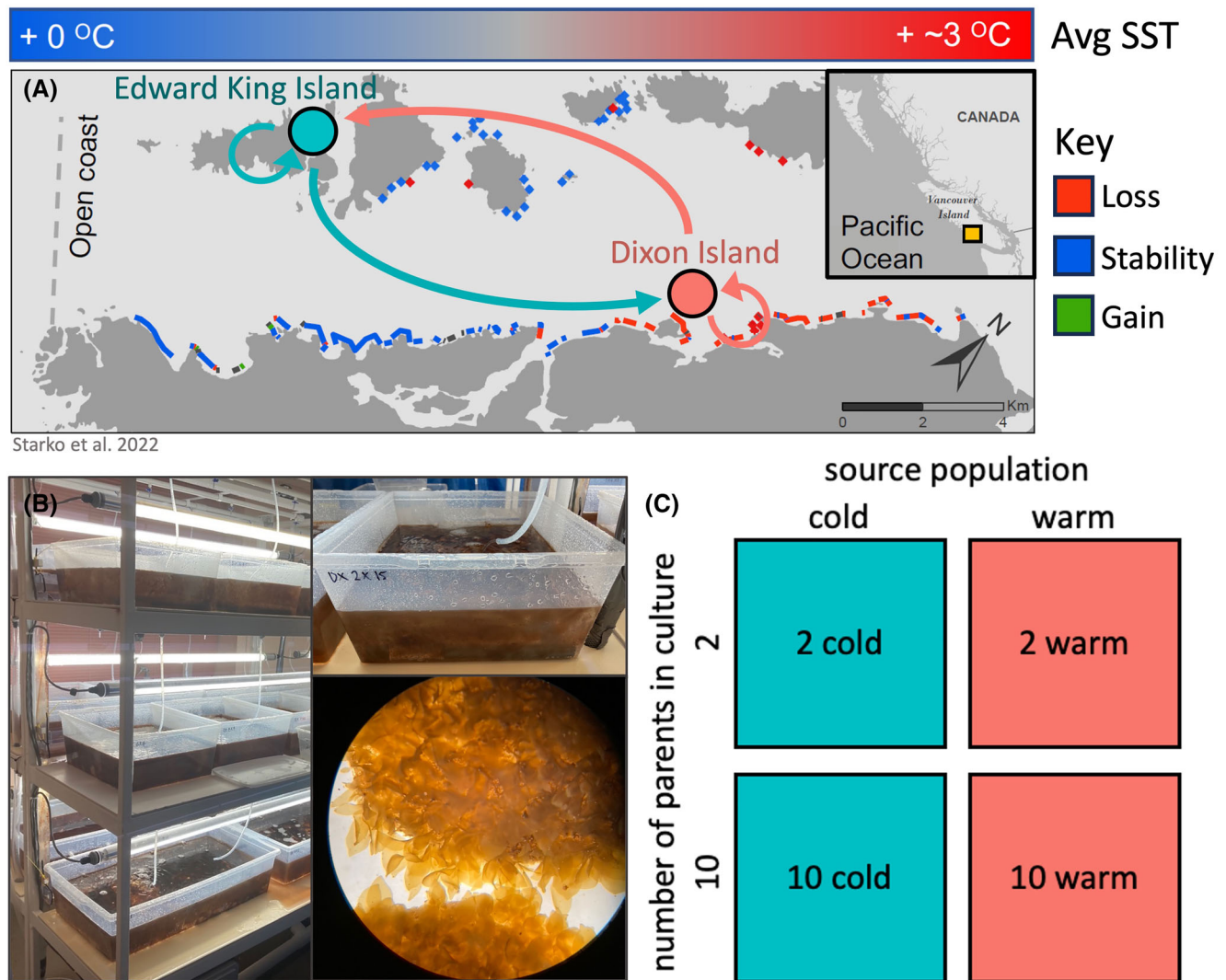
Figure 1. (A) Map of Barkley Sound showing the sea surface temperature (SST) gradient, kelp persistence after the 2014–2016 marine heatwave, and restoration sites, adapted from Starko et al. 2022; (B) nursery system and magnified image of kelp growing on gravel; (C) diagram of the 2 × 2 factorial experimental design testing two parent numbers and two population treatments, which was replicated eight times at each of two sites.

Sound (Starko et al. 2022). Microclimate was an important predictor of kelp persistence during this extreme climate event (Starko et al. 2022), indicating kelp populations in Barkley Sound may be subject to a selection gradient on a small spatial scale. We focused on two sites for this reciprocal transplant experiment: one cool site near the mouth of Barkley Sound, where kelp coverage remained relatively stable during the marine heatwave (Edward King Island, 48°50.214N, 125°12.319W), and one warmer site 7 km farther into the sound that experienced intense kelp die-off and near-extirpation during the same heatwave (Dixon Island, 48°51.271N, 125°07.096W) (Fig. 1A).

Sporophyll Collection and Spore Release

Sporophylls and somatic tissue from *M. pyrifera* parent sporophytes were collected by SCUBA divers from Dixon Island ($n = 15$ individuals) and Edward King Island ($n = 12$ individuals) on 5 December 2022, and held overnight in flow-through seawater tanks at the Bamfield Marine Sciences Centre. On 6 December 2022, reproductive tissue from each individual's sporophyll was sectioned, shaken in 7% iodine in autoclaved seawater, rinsed in autoclaved seawater, patted dry, and stored in a separate paper towel for incubation overnight at 7°C. A section of somatic blade tissue was collected from each individual and preserved in silica gel for genetic analysis.

Prepared sporophylls were soaked in individual 200 mL jars of autoclaved seawater to induce spore release on 7 December 2022. For each individual kelp, spore density was calculated with counts from a hemocytometer; inocula were diluted with autoclaved seawater to achieve a concentration of 10,000 spores/mL in a 200 mL flask for amplification. All cultures were initially incubated at 10°C under shaded white light (<3 μmol photons m^{-2} s^{-1}) for

12 hours. They were then treated with f/2-enriched autoclaved seawater under red light at 10–15 μmol photons $m^{-2} s^{-1}$ and a 16:8 light/dark cycle and incubated at $9 \pm 2°C$ for 53 days. Despite starting with the same spore density, differences in the gametophyte biomass per flask were evident. At the time of fragmentation, their relative biomasses were calculated for applying standardized concentrations to green gravel (Supplement S1; Tables S1 & S2).

Sporophyte Cultures on Green Gravel

On 28 January 2023, gametophytes were fragmented and seeded onto pre-soaked 3.5–5 cm-diameter white marble gravel held in plastic tanks with a spray bottle that was sterilized between treatments (Supplement S1). Gametophytes were diluted to apply a consistent and proportional biomass between treatments in two-parent or 10-parent mixtures. A different combination of two parents was used for each replicate (tank), while for 10-parent crosses the same 10 parents were used in each replicate. Each two-parent culture was held in one of eight 30×35 cm tanks, and each 10-parent cross was held in one of four 64×35 cm tanks. Culturing conditions were initially as follows: flow-through seawater (<0.1 L/min) passing through a 25 μm mesh filter, a 1 μm mesh filter, and an ultraviolet filter; 15–30 μmol photons $m^{-2} s^{-1}$ light on a 16:8 light/dark cycle; mean water temperature 8.8°C (range: 7.5–11.5°C), which is close to the average winter temperature at both source sites, except during two 1-hour periods when temperatures rose to 13°C. Microscopic sporophytes were visually confirmed 10 days after crossing and became macroscopic 24 days after crossing. At this time, flow was increased to 1 L/min in the two-parent tanks and 2 L/minutes in the 10-parent tanks (to equalize water turnover rates in the different tank sizes) and illuminance was increased to 50–60 μmol photons $m^{-2} s^{-1}$. These conditions were maintained for the remainder of culturing (Fig. 1B). Once juvenile sporophytes were approximately 1–4 cm, 20 per tank were individually preserved in silica gel for genetic analysis.

Experimental Design

The experimental design included four treatments based on the combination of source population (warm or cool) and number of parents (two or 10) and eight replicate plots of each treatment at each of two sites ($n = 64$ plots total; Fig. 1C). The position of the experimental plots within a replicate was randomized relative to one another and spaced 1–1.5 m apart from neighboring plots, and no more than 5 m apart from plots in the same replicate. Replicates were spaced 2–20 m from other replicates. Eight replicates of the four treatments were deployed in the warmer site, Dixon Island ($n = 32$ plots) and the same eight replicates were deployed in the cooler site, Edward King Island ($n = 32$ plots). The experimental area within each site was chosen to be: (1) along a relatively sheltered coast to avoid disturbance to the experiment, (2) at 3–7 m depth to Canadian chart datum lower low water large tide where canopy kelp grows in this region of British Columbia, (3) in areas where kelp had been present before the marine heatwave (Starko et al. 2022), and (4) on substrate of gravel and cobbles, which we targeted to minimize the likelihood of urchin grazing (urchins have been observed to cross these substrates less than boulder and bedrock; Starko et al. 2022). Bull kelp was not present at either restoration site, so there would be no interaction with this other canopy-forming species. Both restoration sites were placed immediately adjacent to the small remaining Giant kelp populations from which the parents were sourced.

Green Gravel Outplanting and Monitoring

At each site, between 7 and 9 March 2023, green gravel from each of the four treatments was outplanted into 1 m^2 experimental plots by SCUBA divers, who carefully transferred the gravel by hand from a small bin into the designated treatment plots. Twenty gravel pieces from a single treatment were placed haphazardly but relatively evenly spaced per plot. If plots naturally had mostly gravel substrate, larger nearby cobbles were transferred into the plot by hand to provide material for holdfasts to grow onto.

The experimental plots were monitored at 4 weeks (April), 9 weeks (May), 15 weeks (June), and 25 weeks (August) after outplanting. At each time point, the number of gravel left in the plot, the number of gravel with live kelp, and the total length of five randomly selected individuals on separate gravel was recorded for each plot. It was easy to distinguish our kelp from wild-set kelp, of which there was a small amount in the area, since our white marble gravel was distinct from the natural substrate. During the June monitoring, we tagged all surviving individuals with flagging tape so we could track their condition and pair them to their plot in case their increasing size and buoyancy caused them to be swept outside. At each monitoring time point, temperature and salinity were measured at every meter depth with a YSI Pro DSS and seawater was collected for nutrient analysis 1 m from the surface and 1 m from the bottom in triple-rinsed ethanol-sterilized bottles at both restoration sites. Seawater samples were frozen at $-20°C$ for 6-9 months and were sent to the Institute of Ocean Sciences, Sidney, British Columbia, for analysis of dissolved nitrate + nitrite. Tissue from all survivors in June and August were preserved in silica gel for genetic analysis.

Genetic Diversity

DNA was extracted from preserved somatic tissue from all parent kelp, 20 offspring individuals in each culture treatment, and a subset of surviving outplanted individuals from the field sites following Bringloe et al. (2021) with minor modifications (described in Supplement S2). A library of 96 individuals was sequenced using reduced-representation genotyping by sequencing (GBS) at Plateforme d'Analyze Génomique at Université Laval, Quebec. Filtered sequencing reads were aligned to a *M. pyrifera* reference genome (Diesel et al. 2023) using *bwa-mem* ver. 0.7.17-r1188 (Li 2013) and single nucleotide polymorphism (SNP) genotypes were called using *BCFtools* ver. 1.11–1.19 (Danacek et al. 2021). To demonstrate differences in genetic diversity between experimental treatments, we calculated the average observed heterozygosity (H_o) of each individual using *BCFtools*, and the statistic nucleotide

diversity (π) of each population using *pixy* ver. 1.2.7.beta1 (Korunes & Samuk 2021). To verify the genetic distinctiveness of the two parental source populations, we calculated population genetic differentiation (F_{ST}) using *hierfstat* ver. 0.5-11 (Goudet 2005) and performed a principal component analysis (PCA) of genetic variation using *SNPRelate* ver. 1.6.4 (Zheng et al. 2012, 2017). Details of genetic methods are provided in Supplement S2.

Statistical Analysis

Analyses were conducted in R ver. 4.3.1 (2023-06-16) (R Core Team 2022) using the function *glmer* in the package *lme4* Ver1.1-32 (Bates et al. 2015).

We tested whether kelp survivorship was influenced by the source population, number of parents, outplant site, and their interactions (all fixed effects) using generalized linear mixed effects models with a binomial distribution (Table S3). Additionally, the replicate was included as a random effect to account for the non-independence of kelp growing in similar areas within the site. Initially, all interactions between fixed effects were included in the model, and non-significant interactions were removed stepwise to find the simplest model that included main significant effects following the protocol of Crawley (2013). The response variable for survivorship was the number of gravel with live kelp out of the number of gravel remaining in the plot at each survey time, since mortality due to gravel loss was not related to the population $\times$ site effect we aimed to test. Statistical results from alternative models with survivorship as the number of live gravels out of the total number initially deployed, as well as a Cox proportional hazards model, are presented in Supplement S3 (Tables S5 & S6) for comparison. These different methods of analysis did not change the key findings of this study.

We ran a similar statistical test for kelp total length, but used a gamma distribution (suitable for continuous, positive data) and plot nested in replicate as a random effect to account for the non-independence of kelp individuals measured within the same plot (Table S4). In some cases, the generalized linear mixed effects model was too complex to include a random effect when many interactions between the fixed effects were included, so the models were reduced to generalized linear models (function *glm*; package *lme4*; Bates et al. 2015) by removing the random effect. Model iterations that were run as a glm are noted in Table S4; however, all final models were mixed effects models. Additional details on statistical analysis are provided in Supplement S3, and scripts for running analysis are available at https://doi.org/10.5281/zenodo.15707791.

Results

Effects of Source Population and Outplant Site on Kelp Survivorship and Growth

Kelp survivorship was influenced by different factors at different time points, with some evidence of local adaptation (Tables 1, 2, & S3; Fig. 2A). At 55 days, source population (warm vs. cool) and outplant site were both significant ($z = -3.071$, $p = 0.00213$; $z = 1.975$, $p = 0.04822$), with a significant interaction between the two ($z = 4.995$, $p = 5.89e-07$) (Fig. 2A). Consistent with local adaptation, kelp sourced from the cool site survived 28% better at its home site than at its away site. Kelp from the warm site also survived 85% better at the cool site, which is suggestive of maladaptation. At 100 days, source population had a significant effect on survivorship ($z = -3.117$, $p = 0.001825$), with kelp sourced from the cool site surviving 16% better than kelp sourced from the warm site. However, by 140 and 200 days, no significant effects of source population or outplant site on kelp survivorship were detected (Supplement S5, Fig. S1a). All survivorship, growth, and environmental data are available at https://doi.org/10.5683/SP3/GEXCSU.

Similarly, kelp growth was influenced by different factors at different time points, and we found no strong evidence of local adaptation (Fig. 3A, Tables 2 & S4). At 55 days, the source population had a significant effect on kelp total length ($t = 2.472$, $p = 0.0134$) and a significant interaction with outplant site ($t = -2.040$, $p = 0.0413$) (Fig. 3A). Kelp from the cool site grew 12% better at the cool site, suggesting local adaptation, but kelp from the warm site grew 47% better at the cool site, suggesting maladaptation. At 100 days, source population had a significant effect on total length ($t = 2.269$, $p = 0.0232$), with kelp sourced from the cool site

Table 1. Summary of the number of plots with surviving kelp and the number of gravels with kelp remaining in the different sites and treatments throughout the experiment. Originally, 32 plots and 640 gravels were outplanted at each site, and 16 plots and 320 gravels were outplanted in each treatment. The bottom lines show the total number and percentage of plots and gravels surviving across the whole experiment. A description of overall restoration results from the experiment over time can be found in Supplement S4.

Days old	55		100		140		200	
Category	# Plots	# Kelp	# Plots	# Kelp	# Plots	# Kelp	# Plots	# Kelp
Dixon	32	324	21	99	13	39	7	12
Edward King	32	461	9	34	4	8	2	3
10C	16	190	9	44	5	10	1	1
10W	16	200	7	22	3	9	2	2
2C	16	201	7	30	5	13	4	8
2W	16	194	7	37	4	15	2	4
Total # surviving	**64**	**785**	**30**	**133**	**17**	**47**	**9**	**15**
Total % surviving	**100%**	**61%**	**47%**	**10%**	**27%**	**3.7%**	**14%**	**1.2%**

Table 2. Summary table showing which kelp cultures performed best (survival and growth) at each survey time under the conditions tested. The top two rows show whether kelp sourced from the warm (W) or cool (C) population performed better in the warm or cool environment. The bottom row shows whether kelp cultures produced from 10 or two parents performed better. Within each survey date, survival results are shown on the left and growth results are shown on the right.

Days old	55	100	140	200
Warm environment	C | C	C | W	W | W	C | W
Cool environment	W | W	W | C	C | C	C | W
Which parent # is better?	2 | 2	2 | 2	2 | 10	2 | 10

growing 8% larger than kelp sourced from the warm site. By 140 days, outplant site was the only significant driver of kelp total length ($t = -3.817$, $p = 0.000135$), with kelp outplanted at the cool site reaching larger sizes (140 ± 29 cm) than kelp outplanted at the warm site (47.6 ± 5.9 cm). At 200 days, outplant site was again significant ($t = -2.485$, $p = 0.0130$), with kelp outplanted at the cool site growing larger (285 ± 164 cm) than kelp outplanted at the warm site (88.3 ± 25.5 cm),

although sample size was very low (Fig. S1b). Patterns suggesting local adaptation (i.e. greater length of kelp at its home site than its away site) were visually apparent at both 100 and 140 days, but sample size was too low to detect significance.

The Effect of Parent Number on Kelp Survivorship and Growth

At 100 days, parent number had a significant effect on survivorship ($z = -2.143$, $p = 0.032140$) and a significant interaction with source population ($z = 3.562$, $p = 0.000368$) (Tables 1, 2, & S3; Fig. 2B). The greatest survivorship was among kelp cultures with 10 parents from the cool site ($20.4 \pm 6.3\%$) and the lowest survivorship was in kelp cultures with 10 parents from the warm site ($10.6 \pm 4.2\%$). At 200 days, the number of parents was the only significant driver of kelp survivorship ($z = 2.030$, $p = 0.0423$), with cultures made from two parents surviving better ($3.7 \pm 2.1\%$) than cultures made from 10 parents ($0.99 \pm 0.79\%$) (Fig. S1c).

The number of parents (two vs. 10) did not have a significant effect on kelp total length at any time point, but did have a
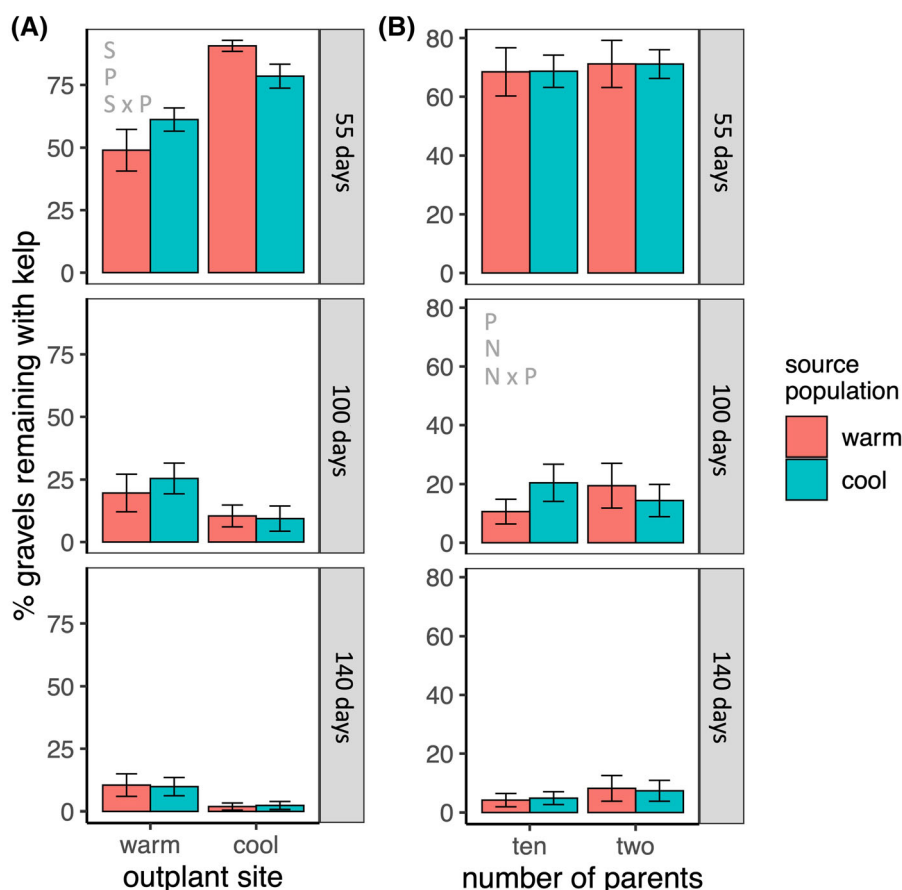

Figure 2. Mean ($\pm$ SE) survivorship for kelp from the two source populations broken down by (A) outplant site and (B) number of parents used to make the culture. Survivorship at each time point was calculated as the number of gravels with live kelp in a plot divided by the number of gravels remaining in the plot. Days since sporophytes were confirmed are shown in gray boxes to the right of the figure panels (55 days = April, 100 days = May, and 140 days = June). Significant effects and interactions from Table S3 are noted by gray letters in the top left of each panel: P = population, S = site, n = number of parents. Significant interactions are denoted by an "×" between the letters, for example, P × S indicates a significant population by site interaction. The panel for 200 days (August) was not shown since the sample size was very low (see Fig. S1).
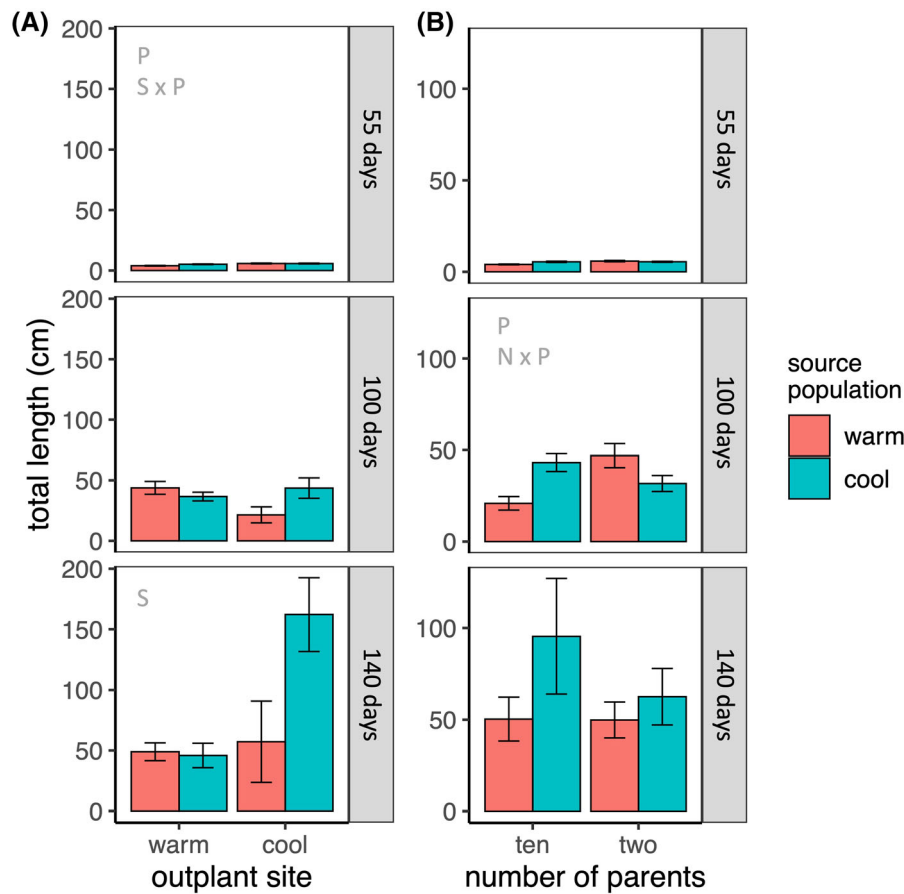
Figure 3. Mean ($\pm$ SE) total length for kelp from the two source populations broken down by (A) outplant site and (B) number of parents used to make the culture. Days since sporophytes were confirmed are shown in gray boxes to the right of the figure panels (55 days = April, 100 days = May, and 140 days = June). Significant effects and interactions from Table S4 are noted by gray letters in the top left of each panel: P = population, S = site, n = number of parents. Significant interactions are denoted by an "$\times$" between the letters, for example, S $\times$ P for a significant site by population interaction. The panel for 200 days (August) was not shown since the sample size was very low (see Fig. S1).

significant interaction with the source population at 100 days ($t = -2.021$, $p = 0.0433$) (Tables 1, 2 & S4), when kelp cultures made with two parents sourced from the warm site had the greatest total length (46.9 ± 6.6 cm) and kelp cultures made with 10 parents sourced from the warm site had the lowest total length (20.9 ± 3.7 cm).

Genetic Analyses

Our filtered genetic dataset consisted of 8871 SNPs and 75 individuals. The cool and warm parent populations were genetically distinguishable, as evidenced by the non-overlapping clusters of individuals from each population on the first two PC axes of genetic variation (Supplement S6, Fig. S2) and a moderate F_{ST} value of 0.126 between parental populations. As expected, population- and individual-level genetic variation was higher in parent kelp from the cool population ($\pi = 0.278$; mean $H_o = 0.281$) than from the warm population ($\pi = 0.230$; mean $H_o = 0.226$) (Figs. 4 & S3). Individuals were on average more heterozygous in

the 10-parent cultures (mean H_o cool = 0.247, warm = 0.223) than in two-parent cultures (mean H_o cool = 0.212, warm = 0.175) (Figs. 4 & S3). The reduced heterozygosity in two-parent cultures was driven largely by an increased likelihood of self-fertilization. The presence of selfed individuals was inferred from a bimodal distribution of H_o values in the offspring, with some offspring exhibiting H_o that was approximately half that of parents (Fig. 4), as expected for selfed individuals. For two-parent cultures, the rate of selfing was 50% (15/30), and for 10-parent cultures it was 18.75% (3/16) (Fig. 4). Some selfed individuals survived until the end of the season (August) which is consistent with the detection of selfed individuals in wild populations (Bemmels et al. 2025). The proportion of selfed individuals in the survivors at the end of the season was lower than in the cultures before outplanting (survivors: 1/10 = 10%; cultures: 18/46 = 39%), but this difference was not statistically significant (χ^2 test, $p = 0.14$). Due to time and budgetary constraints, we were only able to sequence one culture of each treatment, and the sample size and statistical power to detect differences were low. Genetic data are available in NCBI, BioProject ID: PRJNA1279248.
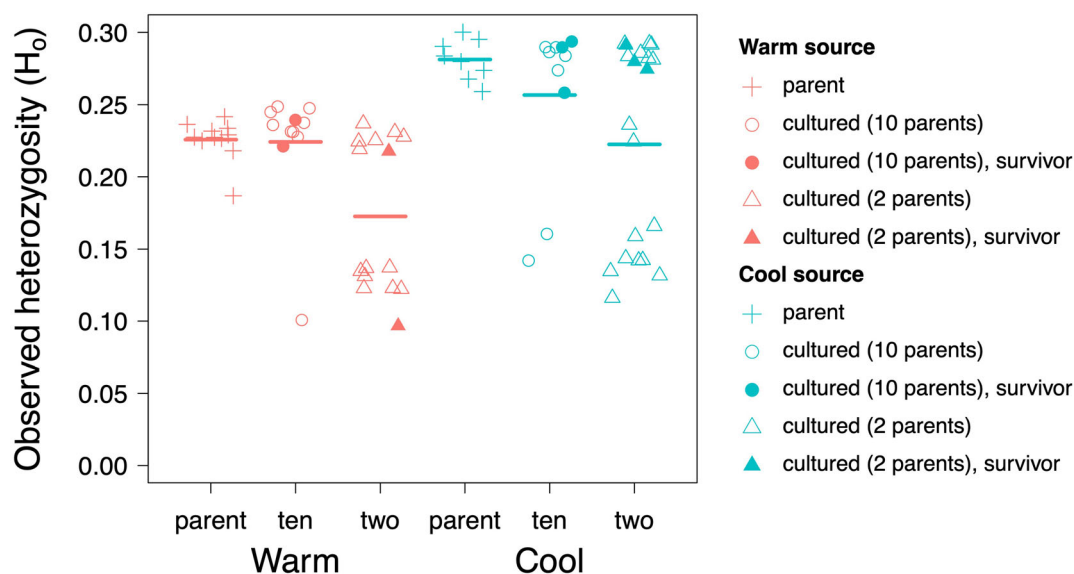
Figure 4. Individual average heterozygosity (H_o) measurements for each individual sequenced, with horizontal bars representing the mean. The legend shows whether the individual was a parent, a child taken from a culture before outplanting, or a child survivor at the end of the experiment in August. Red samples are sourced from the warm population (Dixon Island), and blue samples are sourced from the cool population (Edward King Island).

Biotic and Abiotic Site Conditions

The sites differed in key biotic and abiotic features that may have driven differences in restoration success. Overall, temperatures increased from April to June, especially in the shallowest 3 m, and declined slightly by August (Fig. 5A). A strong thermal difference of 2–3°C between the two sites was established above the thermocline, but these differences were not as pronounced at depth. Note that this experiment did not take place during an El Niño event or marine heatwave. The warmer site had a greater number of plots outplanted in areas with a higher percentage of sand (Fig. 5B). Urchins were more abundant at the cool site, with roughly similar numbers on both cobbles and boulders. At the warm site, urchins were more abundant on boulders than cobbles (Fig. 5C). Nutrient (nitrate + nitrite) concentrations were generally higher at the cooler site, particularly at the bottom (Fig. 5D). Nutrients at the surface were generally low until August when they showed a distinctive peak. Nutrients at the bottom were high in March and April, lower in May and June, then high again in August.

Discussion

Our reciprocal transplant experiment provides support for green gravel as an effective kelp restoration technique, but found inconsistent evidence of local adaptation in the kelp populations being restored (Leimu & Fischer 2008) and variable importance of number of parents on kelp growth and survival at different time points after outplanting (Hughes et al. 2008). We demonstrated that cultures produced with two parents have a higher likelihood of self-fertilization, which may reduce survivorship (Raimondi et al. 2004). Given the complexity of conducting manipulative experiments underwater, we could not control for all factors and

site differences were at times the most influential, for example, enhanced kelp growth at the cooler, nutrient-rich site.

Inconsistent Evidence for Local Adaptation

Our study found inconsistent evidence for local adaptation, with some populations at some time points surviving and growing best at their home site. However, at some time points, kelp sourced from the warm site survived and grew better away from its home site, which may indicate maladaptation (Leimu & Fischer 2008). These results could arise from several mechanisms. First, the warmer average temperatures combined with the stress of the 2014–2016 marine heatwave may not have imposed significant selection for heat-tolerant alleles at the warm site. If the warm population had a small initial effective population size—as supported by the lower observed average heterozygosity among parents at the warm site—it may have had a limited ability to adapt and been subject to genetic drift (Robertson 1960; Vranken et al. 2021). Indeed, Leimu and Fischer (2008) found that larger populations more frequently exhibited local adaptation than smaller ones. Second, if Barkley Sound is a well-mixed water body (Stronach et al. 1993) with effective dispersal, the genetic signature of kelp loss from the heat wave at the warm site may have been diluted by later immigration, given that Giant kelp can disperse up to 10 km in one generation, though most spores disperse only a few meters (Raimondi et al. 2004; Alberto et al. 2010). The moderate F_{ST} (0.126) and non-overlapping clusters in the genetic PCA, however, suggest that our study sites do constitute genetically distinguishable populations (Bemmels et al. 2025). Third, the thermal environment at the sites during the experimental period may not have been distinct enough for differences in performance to become apparent, since our kelp remained below the thermocline for much of the experiment. Finally, the lack of
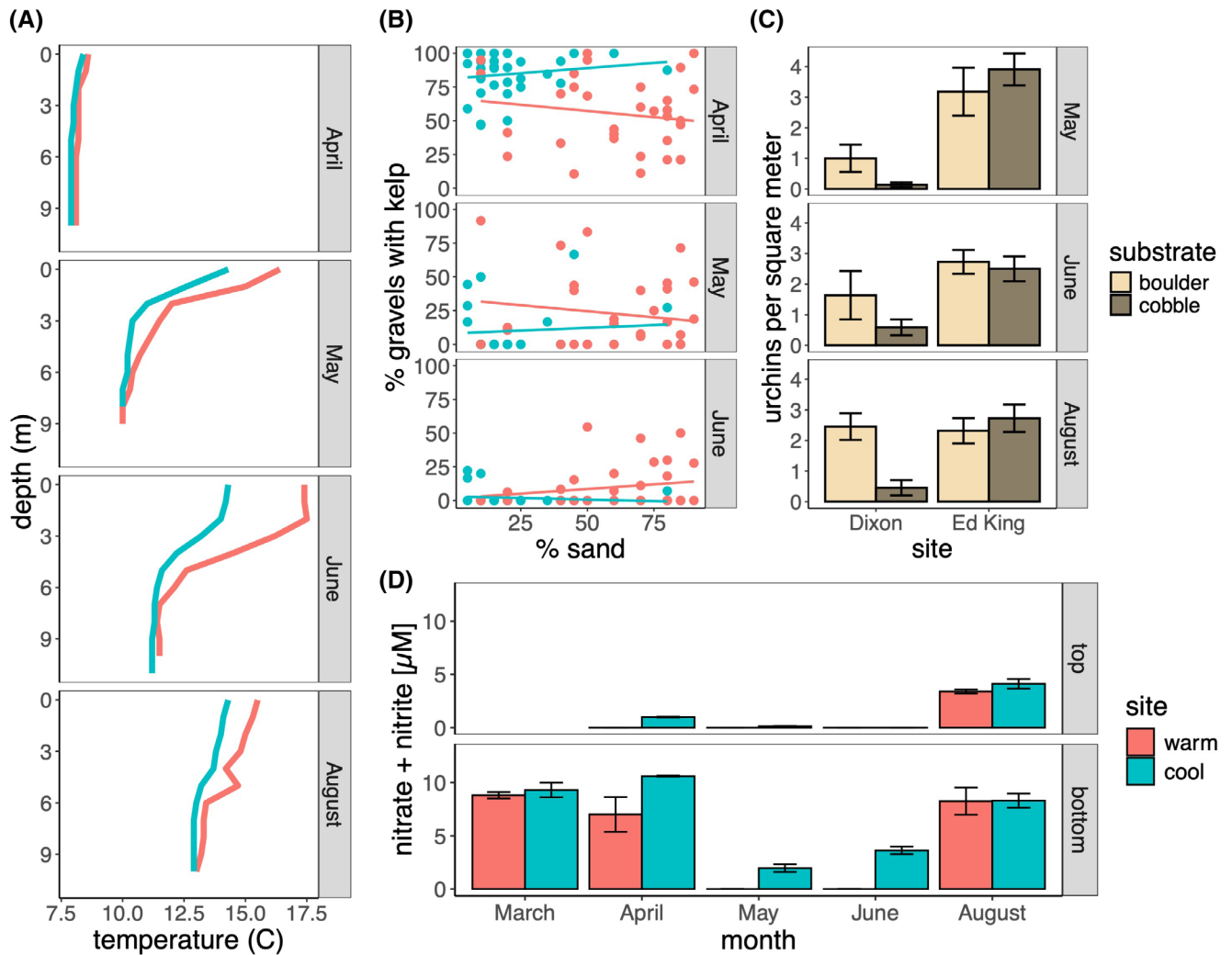
Figure 5. Biotic and abiotic factors at the two outplant sites. (A) Temperature profiles for each survey time point at the warm (red) and cool (blue) site from April to August. (B) The effect of proportion of sand on the survivorship of Giant kelp on green gravel at both sites from April to June. (C) Mean (± SE) urchin density at the warm site (Dixon Island) and the cool site (Edward King Island) from transect surveys on boulder (tan) and cobble (brown) around the restoration area from May to August. (D) Mean (± SE) nitrate plus nitrite concentrations at the top (1 m from surface) and bottom (1 m from bottom) of the water column at both sites during the study period from March to August.

significant results could indicate that phenotypic plasticity compensated for differences in underlying genotypes. More work is required to test this hypothesis (Merilä & Hendry 2014), as in some systems plasticity is not enough to overcome genetic adaptation (e.g. Barott et al. 2021).

The lack of strong evidence for local adaptation between the relatively close sites (7 km) in this experiment suggests that environmental factors, not genetics, largely controlled growth and survivorship. For practice, this indicates that kelp do not necessarily need to be sourced from the most local sites for restoration. In fact, kelp from the cool site survived and grew moderately better at some time points regardless of outplant site and had higher genetic diversity. Thus, farther, undisturbed sites may provide healthier propagules than sites that have experienced more intense heat stress. The findings that kelp from cool regions appeared to be more locally adapted, whereas kelp from warm regions may be maladapted at some time points, are both consistent with findings of Solas et al. (2024), although their study tested for local adaptation in a metapopulation structure sensu Blanquart et al. (2013). We unfortunately did not identify a site with genotypes that perform markedly better in warm conditions, and effect sizes of the main studied factors (e.g. outplant site, source population, and genetic diversity) were generally quite small. Future studies should target more populations with greater geographic distance and be informed by population genetic data, which were not available prior to this study. The experimental matching of genotypes to environment in canopy-forming kelp remains critical for the conservation and restoration of these cold-water species, as climate change is projected to intensify (Schwalm et al. 2020).

Surprising Patterns in Survivorship with Genetic Diversity

The significantly higher survivorship of the two-parent cultures at 6 months (200 days) after outplant was surprising given a vast literature of theoretical and experimental work demonstrating the benefits of enhanced genetic diversity and larger population size (Hughes et al. 2008), including decreased inbreeding depression and an enhanced ability to adapt to future stressors (Wootton & Pfister 2013; Alsuwaiyan et al. 2021). Our two-parent cultures had substantially more individuals with reduced heterozygosity (H_o), which resulted from the greater likelihood of selfing. Self-fertilization has been shown to result in lower fitness and competitive ability in Giant kelp (Raimondi et al. 2004; Johansson et al. 2013) as well as many plant and algae species (Wright et al. 2013), because recessive deleterious mutations are more likely to be expressed (Charlesworth & Willis 2009). The few known benefits of self-fertilization (increased reproductive assurance, maintaining "good" local genotypes, allaying the genetic costs of male function or meiosis) (Jarne & Charlesworth 1993) do not explain our results, as these are relevant to evolutionary processes or population demographics, not the performance of offspring in a single generation. In addition, the (non-significant; $p = 0.08$) lower proportion of selfed individuals among survivors at the end of the season (6%) than in cultures prior to outplanting (35%) hints that inbreeding depression may have occurred, despite the overall greater survival of individuals from the two-parent culture.

The success of the two-parent cultures could have arisen from slight differences in culturing and nursery conditions in early life, although differences in growth were not observed between these treatments at the time of outplanting. Another possible explanation might be that the two-parent offspring are genetically more homogenous. Relative fitness differences between non-selfed individuals (all full siblings) may be less pronounced than among non-selfed individuals in 10-parent cultures. In this case, we might expect more of these similarly fit individuals to survive but grow to smaller size in two-parent plots, whereas in 10-parent plots a smaller number of individuals with superior genotypes might grow to larger size and outcompete other individuals, resulting in lower survivorship. However, this potential phenomenon cannot fully explain our results, as the surviving 10-parent individuals were not substantially larger than the two-parent individuals, and in the case of the cool source population were in fact (non-significantly) smaller.

Increasing numbers in culture might be particularly crucial in kelps, which can self-fertilize, leading to even more drastic reductions in individual fitness (Raimondi et al. 2004). Although we did not see a significant difference in survival between selfed and non-selfed individuals, the low sample size and overall low survival mean that we are underpowered for detecting differences, and other work has found large fitness penalties (Raimondi et al. 2004). This propensity of Giant kelp to self-fertilize means that the number of parents has a more immediate effect on restoration efforts than in species without selfing. For a species without selfing, the first generation released will be relatively unaffected by the number of parents, and issues of inbreeding will only arise in subsequent generations. For Giant kelp, our results suggest that a small number of parents likely causes fitness effects to a large proportion of the offspring immediately. Although we do not see this effect in overall survival, previous measures of the effect of selfing on Giant kelp found that the costs were more pronounced at late life stages (Raimondi et al. 2004), specifically on reproductive output, which was not measured in this experiment.

Future studies could include more than 10 parents in the "high genetic diversity" treatment, which may enhance differences in observed effects. We chose 10 parents to be more genetically tractable, and this was sufficient to capture the heterogeneity of the parents for the cool, but not the warm population. More parents, for example 30 (Shearer et al. 2009), would further decrease the probability of selfing. Studies should also investigate admixture of two or more populations, which has potential to enhance ecosystem function or stress tolerance (Miller et al. 2017).

Green Gravel as a Tool for Giant Kelp Restoration

Our test of the emergent green gravel technique for Giant kelp in British Columbia showed promise for a first trial but requires further enhancement of efficiency. Although survivorship was initially high, by 6 months just over 1 % of kelp were still alive, for a total of 15 individuals. Four were observed to grow holdfasts onto native rock, some holdfasts were 8 cm in diameter, and the tallest kelp was 5.3 m. This success is important because Giant kelp is more buoyant than most other species for which this method has been developed (Fredriksen et al. 2020; Wood et al. 2024), making it less likely to stay in place. This study is among the first to report survivorship from a field experiment of Giant kelp grown with green gravel. Unfortunately, our outplanted kelp did not survive to reproductive age—which is a prerequisite in establishing self-sustaining populations that are resistant and resilient to perturbations (Aavik & Helm 2018)—because they disappeared from plots over the subsequent winter.

Influence of Site on Kelp Restoration Success

On the influence of the environment, we noted three general trends which largely agreed with our expectations. First, kelp survived (non-significantly) better at the warm site where grazer density was low. The warm site with 1–2 urchins/m^2 had fourfold improved survivorship compared to the cool site with 3–4 urchins/m^2. The impacts of overgrazing on kelp are well-demonstrated (Miller et al. 2022), and our study further supports the importance of conducting synergistic restoration activities such as grazer harvesting in concert with outplanting or selection of sites with naturally fewer grazers. However, a confounding factor that could have caused high mortality is that the cool site is more wave-exposed, and high swell can cause gravel to roll (Earp et al. 2024).

While grazers and swell likely drove patterns of survivorship, nutrient concentration likely played a role in kelp growth. Outplanted kelp grew best at the cool site, which had higher concentrations of dissolved nitrate and nitrite. Sufficient dissolved inorganic nitrogen can increase the resistance of

Giant kelp to heat stress (Wheeler & North 1980; North & Zimmerman 1984). In fact, some studies have suggested that nutrient limitation, rather than thermal limits, are reasons for Giant kelp tissue deterioration in summer during strong stratification (North & Zimmerman 1984). These results suggest that kelp productivity could be maximized by restoring in cooler, higher-nutrient sites via urchin removal, since these are the conditions in which urchins will be the most abundant and populations most genetically diverse. However, because the difference in performance between our source populations was rather low, restoring at key warm sites is still possible and encouraged. In this case, sourcing kelp from cooler areas with higher genetic diversity could yield positive results.

Caveats and Limitations

Controlled manipulative experiments are challenging in the ocean environment for many reasons, not least of which is that some aspects of the environment cannot be controlled. Primarily, temperatures between sites at the depths where our kelp grew were not substantially different for much of the year, which likely partially explains the lack of a significant population × site interaction at later time points. Further, kelp mortality made our main hypotheses untestable at later time points due to low sample size. Sample size in general was a limiting factor due to time and budgetary constraints, and future studies should include greater replication, urchin control, more populations nested within different environments (e.g. Blanquart et al. 2013), and more individuals genetically sequenced. Finally, co-varying environmental factors (e.g. greater nutrients with cooler temperatures) make it hard to untangle specific drivers of kelp growth and survivorship. Regardless, the environmental information collected and distinct conditions between the sites illustrated some likely drivers of restoration success that will be useful to restoration practitioners.

Conclusions

We successfully cultured and outplanted Giant kelp in British Columbia, performing an important first test of the green gravel technique that resulted in kelps growing holdfasts onto other rocks and some growing up to 5.3 m in length. Despite modest effects of source population, genetic diversity, and outplanting site on restoration outcomes, overall survivorship was low in all cases. The selection of kelp propagules from the most local site did not appear to enhance restoration success. In fact, kelp from the warm site sometimes showed signs of maladaptation. Although this study found only slight evidence for local adaptation between two geographically close sites, it revealed complex environmental drivers of kelp growth and survivorship. Kelp that originated from the cool site had moderately greater survivorship and growth overall, possibly because the source population had higher genetic diversity and may not have experienced a recent bottleneck. Kelp grew larger and faster at the exposed, colder site likely because of greater water movement and nutrient supply. Kelp survived best at the warmer, more sheltered site likely because there were fewer urchin grazers and less intense swell. Finally, although cultures resulting from only two parents survived better than cultures from 10 parents after 6 months in the field, we saw evidence of frequent self-fertilization in two-parent cultures, indicating negative consequences might be seen with greater statistical power or at later stages of the life cycle. This work represents an early test of the role of genetic diversity and provenance in the success of kelp forest restoration, and continued investigation will be important to further improve these emerging techniques.

Acknowledgments

LITERATURE CITED

Aavik T, Helm A (2018) Restoration of plant species and genetic diversity depends on landscape-scale dispersal. Restoration Ecology 26:S92–S102. https://doi.org/10.1111/rec.12634

Alberto F, Raimondi PT, Reed DC, Coelho NC, Leblois R, Whitmer A, Serrão EA (2010) Habitat continuity and geographic distance predict population genetic differentiation in giant kelp. Ecology 91:49–56. https://doi.org/10.1890/09-0050.1

Alsuwaiyan NA, Filbee-Dexter K, Vranken S, Burkholz C, Cambridge M, Coleman MA, Wernberg T (2022) Green gravel as a vector of dispersal for kelp restoration. Frontiers in Marine Science 9:910417. https://doi.org/10.3389/fmars.2022.910417

Alsuwaiyan NA, Vranken S, Filbee-Dexter K, Cambridge M, Coleman MA, Wernberg T (2021) Genotypic variation in response to extreme events may facilitate kelp adaptation under future climates. Marine Ecology Progress Series 672:111–121. https://doi.org/10.3354/meps13802

Barott KL, Huffmyer AS, Davidson JM, Lenz EA, Matsuda SB, Hancock JR, Innis T, Drury C, Putnam HM, Gates RD (2021) Coral bleaching response is unaltered following acclimatization to reefs with distinct environmental conditions. Proceedings of the National Academy of Sciences of the United States of America 118:e2025435118. https://doi.org/10.1073/pnas.2025435118

Barrett SC, Kohn JR (1991) Genetic and evolutionary consequences of small population size in plants: implications for conservation. Pages 3–30. In: Genetics and conservation of rare plants. Oxford University Press, New York.

Bates D, Mächler M, Bolker B, Walker S (2015) Fitting linear mixed-effects models using lme4. Journal of Statistical Software 67:1–48. https://doi.org/10.18637/jss.v067.i01

Becheler R, Haverbeck D, Clerc C, Montecinos G, Valero M, Mansilla A, Faugeron S (2022) Variation in thermal tolerance of the giant kelp's gametophytes: suitability of habitat, population quality or local adaptation? Frontiers in Marine Science 9:802535. https://doi.org/10.3389/fmars.2022.802535

Bemmels JB, Starko S, Weigel BL, Hirabayashi K, Pinch A, Elphinstone C, et al. (2025) Population genomics reveals strong impacts of genetic drift without purging and guides conservation of bull and giant kelp. Current Biology 3:688–698. https://doi.org/10.1016/j.cub.2024.12.025

Biebach I, Leigh DM, Sluzek K, Keller LF (2016) Genetic issues in reintroduction. Pages 149–183. In: Reintroduction of fish and wildlife populations. University of California Press, Berkeley.

Blanquart F, Katz O, Nuismer SI, Gandon S (2013) A practical guide to measuring local adaptation. Ecology Letters 16:1195–1205. https://doi.org/10.1111/ele.12150

Bringloe TT, Zaparenkov D, Starko S, Grant WS, Vieira C, Kawai H, et al. (2021) Whole-genome sequencing reveals forgotten lineages and recurrent hybridizations within the kelp genus *Alaria* (Phaeophyceae). Journal of Phycology 57:1721–1738. https://doi.org/10.1111/jpy.13212

Charlesworth D, Willis JH (2009) The genetics of inbreeding depression. Nature Reviews Genetics 10:783–796. https://doi.org/10.1038/nrg2664

Coleman MA, Wernberg T (2020) The silver lining of extreme events. Trends in Ecology & Evolution 35:1065–1067. https://doi.org/10.1016/j.tree.2020.08.013

Crawley M (2013) The R book. 2nd ed. Wiley, Chichester, United Kingdom

Danacek P, Bonfield JK, Liddle J, Marshall J, Ohan V, Pollard MO, et al. (2021) Twelve years of SAMtools and BCFtools. GigaScience 10:giab008. https://doi.org/10.1093/gigascience/giab008

Diesel J, Molano G, Montecinos GJ, DeWeese K, Calhoun S, Kuo A, et al. (2023) A scaffolded and annotated reference genome of giant kelp (*Macrocystis pyrifera*). BMC Genomics 24:543. https://doi.org/10.1186/s12864-023-09658-x

Dussex N, Morales HE, Grossen C, Dalén L, van Oosterhout C (2023) Purging and accumulation of genetic load in conservation. Trends in Ecology & Evolution 38:961–969. https://doi.org/10.1016/j.tree.2023.05.008

Earp HS, Smale DA, Catherall HJN, Moore PJ (2024) An assessment of the utility of green gravel as a kelp restoration tool in wave-exposed intertidal habitats. Journal of the Marine Biological Association of the United Kingdom 104:1–11. https://doi.org/10.1017/S0025315424000225

Eger AM, Aguirre JD, Altamirano M, Arafeh-Dalmau N, Arroyo NL, Bauer-Civiello AM, et al. (2023b) The kelp forest challenge: a collaborative global movement to protect and restore 4 million hectares of kelp forests. Journal of Applied Phycology 36:951. https://doi.org/10.1007/s10811-023-03103-y

Eger AM, Marzinelli EM, Beas-Luna R, Blain CO, Blamey LK, Byrnes JE, et al. (2023a) The value of ecosystem services in global marine kelp forests. Nature Communications 14:1894. https://doi.org/10.1038/s41467-023-37385-0

Filbee-Dexter K, Wernberg T (2020) Substantial blue carbon in overlooked Australian kelp forests. Scientific Reports 10:1–6. https://doi.org/10.1038/s41598-020-69258-7

Fredriksen S, Filbee-Dexter K, Norderhaug KM, Steen H, Bodvin T, Coleman MA, Moy F, Wernberg T (2020) Green gravel: a novel restoration tool to combat kelp forest decline. Scientific Reports 10:3983. https://doi.org/10.1038/s41598-020-60553-x

Goudet J (2005) HIERFSTAT, a package for R to compute and test hierarchical *F*-statistics. Molecular Ecology Notes 5:184–186. https://doi.org/10.1111/j.1471-8286.2004.00828.x

Graham MH (2004) Effects of local deforestation on the diversity and structure of southern California giant kelp forest food webs. Ecosystems 7:341–357. https://doi.org/10.1007/s10021-003-0245-6

Harley CDG, Anderson KM, Demes KW, Jorve JP, Kordas RL, Coyle TA, Graham MH (2012) Effects of climate change on global seaweed communities. Journal of Phycology 48:1064–1078. https://doi.org/10.1111/j.1529-8817.2012.01224.x

Harris JA, Hobbs RJ, Higgs E, Aronson J (2006) Ecological restoration and global climate change. Restoration Ecology 14:170–176. https://doi.org/10.1111/j.1526-100X.2006.00136.x

Hughes AR, Inouye BD, Johnson MT, Underwood N, Vellend M (2008) Ecological consequences of genetic diversity. Ecology Letters 11:609–623. https://doi.org/10.1111/j.1461-0248.2008.01179.x

Hughes AR, Stachowicz JJ (2004) Genetic diversity enhances the resistance of a seagrass ecosystem to disturbance. Proceedings of the National Academy of Sciences of the United States of America 101:8998–9002. https://doi.org/10.1073/pnas.0402642101

IPCC (Intergovernmental Panel on Climate Change) (2022) Climate change 2022: impacts, adaptation, and vulnerability. In: Pörtner H-O, Roberts DC, Tignor M, Poloczanska ES, Mintenbeck K, Alegría A, Craig M, Langsdorf S, Löschke S, Möller V, Okem A, Rama B (eds) Contribution of Working Group II to the Sixth Assessment Report of the Intergovernmental Panel on Climate Change. Cambridge University Press, Cambridge, United Kingdom; New York

Jarne P, Charlesworth D (1993) The evolution of the selfing rate in functionally hermaphrodite plants and animals. Annual Review of Ecology and Systematics 24:441–466. https://doi.org/10.1146/annurev.es.24.110193.002301

Jayathilake D, Costello MJ (2021) Version 2 of the world map of Laminarian kelp benefits from more Arctic data and makes it the largest marine biome. Biological Conservation 257:109099. https://doi.org/10.1016/j.biocon.2021.109099

Johansson MT, Raimondi PT, Reed DC, Coelho NC, Serrão EA, Alberto FA (2013) Looking into the black box: simulating the role of self-fertilization and mortality in the genetic structure of *Macrocystis pyrifera*. Molecular Ecology 22:4842–4854. https://doi.org/10.1111/mec.12444

Johnson LC, Galliart MB, Alsdurf JD, Maricle BR, Baer SG, Bello NM, Gibson DJ, Smith AB (2022) Reciprocal transplant gardens as gold standard to detect local adaptation in grassland species: new opportunities moving into the 21st century. Journal of Ecology 110:1054–1071. https://doi.org/10.1111/1365-2745.13695

Jones TA (2013) When local isn't best. Evolutionary Applications 6:1109–1118. https://doi.org/10.1111/eva.12090

Korunes KL, Samuk K (2021) PIXY: unbiased estimation of nucleotide diversity and divergence in the presence of missing data. Molecular Ecology Resources 21:1359–1368. https://doi.org/10.1111/1755-0998.13326

Krumhansl KA, Okamoto DK, Rassweiler A, Novak M, Bolton JJ, Cavanaugh KC, et al. (2016) Global patterns of kelp forest change over the past half-century. Proceedings of the National Academy of Sciences of the United States of America 113:13785–13790. https://doi.org/10.1073/pnas.1606102113

Leimu R, Fischer M (2008) A meta-analysis of local adaptation in plants. PLoS One 3:e4010. https://doi.org/10.1371/journal.pone.0004010

Li H (2013) Aligning sequence reads, clone sequences and assembly contigs with BWA-MEM. arXiv: 1303.3997v2

Lindstrom SC (2023) A reinstated species name for north-eastern Pacific *Macrocystis* (Laminariaceae, Phaeophyceae). Notulae Algarum 290:1-2.

Macaya EC, Zuccarello GC (2010) DNA barcoding and genetic divergence in the Giant kelp *Macrocystis* (Laminariales). Journal of Phycology 46:736–742. https://doi.org/10.1111/j.1529-8817.2010.00845.x

Martin RA, Chick LD, Garvin ML, Diamond SE (2021) In a nutshell, a reciprocal transplant experiment reveals local adaptation and fitness trade-offs in response to urban evolution in an acorn-dwelling ant. Evolution 75:876–887. https://doi.org/10.1111/evo.14191

Merilä J, Hendry AP (2014) Climate change, adaptation, and phenotypic plasticity: the problem and the evidence. Evolutionary Applications 7:1–14. https://doi.org/10.1111/eva.12137

Miller KI, Blain CO, Shears NT (2022) Sea urchin removal as a tool for macroalgal restoration: a review on removing "the spiny enemies". Frontiers in Marine Science 9:831001. https://doi.org/10.3389/fmars.2022.831001

Miller BP, Sinclair EA, Menz MH, Elliott CP, Bunn E, Commander LE, et al. (2017) A framework for the practical science necessary to restore sustainable, resilient, and biodiverse ecosystems. Restoration Ecology 25:605–617. https://doi.org/10.1111/rec.12475

North WJ, Zimmerman RC (1984) Influences of macronutrients and water temperatures on summertime survival of *Macrocystis* canopies. Pages 419–424. In: Eleventh International Seaweed Symposium: Proceedings of the Eleventh International Seaweed Symposium, held in Qingdao, People's Republic of China, June 19–25, 1983. Springer, Dordrecht.

Pazzaglia J, Nguyen HM, Santillán-Sarmiento A, Ruocco M, Dattolo E, Marín-Guirao L, Procaccini G (2021) The genetic component of seagrass restoration: what we know and the way forwards. Water 13:1–24. https://doi.org/10.3390/w13060829

R Core Team (2022) R: a language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria

Raimondi PT, Reed DC, Gaylord B, Washburn L (2004) Effects of self-fertilization in the giant kelp, *Macrocystis pyrifera*. Ecology 85:3267–3276. https://doi.org/10.1890/03-0559

Robertson A (1960) A theory of limits in artificial selection. Proceedings of the Royal Society of London. Series B: Biological Sciences 153:234–249. https://doi.org/10.1098/rspb.1960.0099

Rogers-Bennett L, Catton CA (2019) Marine heat wave and multiple stressors tip bull kelp forest to sea urchin barrens. Scientific Reports 9:15050. https://doi.org/10.1038/s41598-019-51114-y

Schwalm CR, Glendon S, Duffy PB (2020) RCP8.5 tracks cumulative CO_2 emissions. Proceedings of the National Academy of Sciences of the United States of America 117:19656–19657. https://doi.org/10.1073/pnas.2007117117

Schweitzer C, Clark SL, Gottschalk KW, Stringer J, Sitzlar R (2014) Proactive restoration: planning, implementation, and early results of silvicultural strategies for increasing resilience against gypsy moth infestation in upland oak forests on the Daniel Boone National Forest, Kentucky. Journal of Forestry 112:401–411. https://doi.org/10.5849/jof.13-085

Shearer TL, Porto I, Zubillaga AL (2009) Restoration of coral populations in light of genetic diversity estimates. Coral Reefs 28:727–733. https://doi.org/10.1007/s00338-009-0520-x

Solas M, Correa RA, Barría F, Garcés C, Camus C, Faugeron S (2024) Assessment of local adaptation and outbreeding risks in contrasting thermal environments of the giant kelp, *Macrocystis pyrifera*. Journal of Applied Phycology 36:471–483. https://doi.org/10.1007/s10811-023-03119-4

Starko S, Neufeld CJ, Gendall L, Timmer B, Campbell L, Yakimishyn J, Druehl L, Baum JK (2022) Microclimate predicts kelp forest extinction in the face of direct and indirect marine heatwave effects. Ecological Applications 32:e2673. https://doi.org/10.1002/eap.2673

Stronach JA, Ng MK, Foreman MG, Murty TS (1993) Tides and currents in Barkley sound and Alberni inlet. Marine Geodesy 16:1–41. https://doi.org/10.1080/15210609309379675

Vranken S, Wernberg T, Scheben A, Severn-Ellis AA, Batley J, Bayer PE, Edwards D, Wheeler D, Coleman MA (2021) Genotype-environment mismatch of kelp forests under climate change. Molecular Ecology 30:3730–3746. https://doi.org/10.1111/mec.15993

Wheeler PA, North WJ (1980) Effect of nitrogen supply on nitrogen content and growth rate of juvenile *Macrocystis pyrifera* (Phaeophyta) sporophytes. Journal of Phycology 16:577–582. https://doi.org/10.1111/j.1529-8817.1980.tb03076.x

Wood GV, Filbee-Dexter K, Coleman MA, Valckenaere J, Aguirre JD, Bentley PM, et al. (2024) Upscaling marine forest restoration: challenges, solutions and recommendations from the green gravel action group. Frontiers in Marine Science 11:1364263. https://doi.org/10.3389/fmars.2024.1364263

Wood G, Marzinelli EM, Coleman MA, Campbell AH, Santini NS, Kajlich L, Verdura J, Wodak J, Steinberg PD, Vergés A (2019) Restoring subtidal marine macrophytes in the Anthropocene: trajectories and future-proofing. Marine and Freshwater Research 70:936–951. https://doi.org/10.1071/MF18226

Wootton JT, Pfister CA (2013) Experimental separation of genetic and demographic factors on extinction risk in wild populations. Ecology 94:2117–2123. https://doi.org/10.1890/12-1828.1

Wright SI, Kalisz S, Slotte T (2013) Evolutionary consequences of self-fertilization in plants. Proceedings of the Royal Society B: Biological Sciences 280:20130133. https://doi.org/10.1098/rspb.2013.0133

Zheng X, Gogarten S, Lawrence M, Stilp A, Conomos M, Weir BS, Laurie C, Levine D (2017) SeqArray–a storage-efficient high-performance data format for WGS variant calls. Bioinformatics 33:2251–2257. https://doi.org/10.1093/bioinformatics/btx145

Zheng X, Levine D, Shen J, Gogarten SM, Laurie C, Weir BS (2012) A high-performance computing toolset for relatedness and principal component analysis of SNP data. Bioinformatics 28:3326–3328. https://doi.org/10.1093/bioinformatics/bts606

Supporting Information

The following information may be found in the online version of this article:

Supplement S1. Detailed culturing and outplanting methods.

Supplement S2. Genetic details.

Supplement S3. Details for statistics presented in the manuscript.

Supplement S4. Overall kelp growth and survivorship trends relevant to restoration.

Supplement S5. Survivorship and growth data from August.

Supplement S6. Additional genetic results.

Table S1. Fragment densities after blending, flask volumes, and estimated fragment numbers by gametophyte flask culture derived from each wild sporophyte "parent."

Table S2. Volumes from the single-parent gametophyte flasks mixed for each experimental group.

Table S3. Outputs of generalized linear mixed effects models for survivorship using a binomial distribution.

Table S4. Outputs of generalized linear mixed effects models with kelp total length as the response variable using a gamma distribution.

Table S5. Alternative statistics not presented in the main manuscript.

Table S6. Statistical results of Cox proportional hazards model, not counting lost gravel as mortality events.

Figure S1. For August (200 days), mean ($\pm$SE) (a) survivorship and (b) growth for kelp from the two source populations broken down by the outplant site.

Figure S2. Principal component analysis (PCA) of genetic variation, showing the placement of individuals along the first two PC axes.

Figure S3. Mean ($\pm$SD) average heterozygosity (H_o) of parents, cultures, and end of season survivors from the warm site (Dixon) and cool site (Edward King).