# Effects of the 2021 Heatwave on Intertidal Clam and Oyster Populations in Washington State



Technical Report Prepared for the Jamestown S'Klallam Tribe

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COVER PHOTO Cockle clams after the 2021 heatwave at Similk Bay, WA. By James McArdle, Swinomish Tribal Community

# Executive summary

Climate change poses multiple threats to nearshore ecosystems and species that are predicted to increase in frequency and severity in the coming decades. Sessile intertidal organisms, such as clams and oysters, are particularly susceptible to one of these threats, atmospheric heatwaves. From 26 - 28 June 2021, the Pacific Northwest (including the Puget Sound region) experienced the most extreme atmospheric heatwave in recorded history with temperatures at or above  $38^{\circ}$  C ( $100^{\circ}$  F) for multiple days. The heatwave coincided with the lowest daytime low tides of the year, and some of the lowest tides in the 18.6 yr tidal cycle, leaving intertidal organisms exposed to extreme temperatures for multiple hours. This combination of events led to observations of dead and dying shellfish across the region and raised alarm of the immediate and long-term impact of the event. Building off an initial qualitative assessment of heatwave impacts, the project team collated quantitative data on clams and oysters from a suite of regional partners with the goal of comparing abundance, biomass, and size data from before and after the heatwave.

Our analysis found that within site and species comparisons ranged from negative to positive effects of the heatwave. However, many datasets displayed a high degree of variability making statistical attribution of effects challenging. Species-specific responses across sites displayed a wide range of heatwave effects, suggesting site-specific patterns that may relate to local environmental conditions and geomorphology. Overall, this work provides the best available quantitative analysis of species-specific responses to the June 2021 heatwave of Puget Sound clams and oysters. While results are variable in magnitude and confidence, nearly all species have at least one example of positive, negative, or little effect of the heatwave. Of further interest may be the juxtaposition of species that show large changes in estimated abundance, biomass, or size (percent difference > 20 %) but lack statistical evidence for those changes. Higher levels of statistical evidence may not be necessary for management actions; however, this pattern raises the question of the ability of current survey methods to detect the effects of acute events. Comanagers and scientists should examine the patterns of data and effects presented here as they consider future stewardship of intertidal clam and oyster resources.

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### Introduction

### 1.1. Background

Changing marine conditions associated with global climate change, such as increased sea surface temperatures and ocean acidification, present a considerable physiological challenge to marine organisms that can lead to changes in growth, survival, reproduction, and community structure (Harley et al. 2006, IPCC 2021). A changing climate can manifest in multiple ways including relatively slow changes, like decreasing pH and increasing average annual temperature, but can also occur as an increase in anomalous weather events such as heatwaves.

Heatwaves, defined as periods of anomalously high temperatures, occur in the atmosphere and the world's oceans and are increasing in intensity and frequency as a result of anthropogenic climate change (Coumou and Rahmstorf 2012, Perkins et al. 2012, Hobday et al. 2016). Both marine and atmospheric heatwaves pose direct and long-term threats to nearshore marine communities by affecting species distributions and ecosystem structure and function (Rosenzweig et al. 2008, Wernberg et al. 2013).

Climate change poses an immediate threat to the protection, and recovery of Puget Sound's marine environment in Washington State (WA), as well management of important natural resources (Mauger et al. 2015). From 26 - 28 June 2021, the Pacific Northwest of the United States, including the Puget Sound region, experienced the most significant heat event in recorded history that would have been nearly impossible without the effects of anthropogenic climate change (Philip et al. 2021). The event broke numerous all-time records with daytime high temperatures reaching well over  $38^{\circ}$ C ( $100^{\circ}$ F) for multiple consecutive days. Puget Sound and the Strait of Juan de Fuca in Washington State cover approximately 2492km (1549mi) of shoreline, much of which is suitable habitat for clams and oysters. Clams and oysters in the region support recreational, commercial, and cultural fisheries and are major components of softsediment nearshore ecosystems. Some species are also cultivated for aquaculture.

A preliminary examination of the effects of the June 2021 heatwave on intertidal shellfish revealed that many species, including clams and oysters, experienced unusually high mortality following the heatwave (Raymond et al. 2022). Raymond et al. (2022) utilized a 5-point rating system to evaluate the condition of intertidal shellfish following the heatwave which is referenced herein as the "qualitative assessment". A key factor in this observed mortality was that high temperatures coincided with the lowest daytime low tides of the year, exposing intertidal organisms to anomalously high air temperatures for extended periods of time. Further investigation of these preliminary observations suggests species- and location-specific responses to the heatwave, which in some cases point to resilience to extreme air temperatures. These preliminary observations provided a crucial first look at the effects of the 2021 heatwave on intertidal organisms; however, questions remained on the acute impact of the heatwave on intertidal shellfish populations.

### 1.2. Project goals and objectives

Given that such climate-driven events are predicted to increase in frequency and intensity (NOAA 2022, IPCC 2021), there is an urgent need to quantify and document the effects of such

extreme weather events on natural and enhanced intertidal bivalve populations. The purpose of this assessment was to collect and analyze available Puget Sound clam and oyster population data from before and after the June 2021 heatwave to better understand species- and/or location-specific acute impacts, and report findings to resource managers and regional stakeholders. Ideally, this type of quantitative assessment of heatwave impacts will help to increase awareness and preparedness of future extreme events, promote stewardship, inform resource management, and ultimately promote sustainability of valuable shellfish resources. The primary goal of this report was to quantify the impact of extreme atmospheric heatwave events on intertidal clam and oyster populations across Puget Sound pre- and post- the June 2021 heatwave. To accomplish this, the specific project objectives were:

- 1. Collaborate with regional partners to collate available pre- and post- heatwave survey data on clams and oyster populations in Puget Sound, WA.
- 2. Identify criteria for data selection and reformat datasets to be comparable across standard metrics: population size, density, bivalve size and biomass.
- 3. Conduct a quantitative assessment on the heatwave's acute impacts on clam and oyster populations.
- 4. Disseminate project outcomes to shellfish resource managers and regional stakeholders.

### 1.3. Report overview

This report details comparisons of clam and oyster populations, size structure, and biomass from data contributing partners before and after the 2021 heatwave. Specific comparisons will depend on the data available for a given species, location, and protocols used by contributing partners. Below we detail the contributors, data processing and analysis, display result plots and tables, and discuss general and specific observed patterns and how they relate to intertidal clam and oyster ecology and fisheries. The compiled data used in this assessment, along with R code used for data processing and analysis, can be made available upon request to the Project Manager, Elizabeth Tobin at the Jamestown S'Klallam Tribe.

# 2. Methods

### 2.1. Contributors

We leveraged the partnerships built from the initial qualitative assessment of heatwave impacts on intertidal shellfish to gather quantitative data on clam and oyster populations. We sent an email request to contributors to the qualitative assessment conducted in summer 2021 (Raymond et al. 2022) requesting quantitative population and size data on intertidal clams and/or oysters from beaches they survey. This included only quantitative data on intertidal clams and/or oysters that was collected before and after the June 2021 heatwave at the same site, following the same survey methods to ensure consistency in before and after heatwave data collection. We defined "before" data as those collected before 26 June 2021 ideally within the last few years, and "after" data as those collected between July and September 2021. Potential collaborators were targeted that routinely sample clam and oyster populations as part of their shellfish management, research or restoration activities. We received data from the following contributors: Jamestown S'Klallam Tribe, Jefferson County Marine Resource Committee, Port Gamble S'Klallam Tribe, Puget Sound Restoration Fund, Skagit County Marine Resource Committee, Swinomish Indian Tribal Community, Squaxin Island Tribe, and Washington Department of Fish and Wildlife. An overview of the data from each contributor is detailed in Appendix A and locations in Figure 1.

### 2.2. Data pre-processing

The project team received data in the rawest form possible. This meant that data was presented in a form similar to how it was originally collected and entered into each contributors' internal database. Since different contributors structure their data in different ways to accommodate their own needs, we developed a universal template to store and analyze data (Appendix K). This built consistency in the data which made it easier to check for data discrepancies among contributors and to simplify our analysis. Our template followed a 'long (= tall) data' structure where each row represents a unique observation, either from a sample quadrat (in the case of count data) or from an individual organism (in the case of size data). The exact process that converted raw data format to 'long' data format varied among contributors, but the raw data values were preserved. The project team took care to track internal versions of the data so that discrepancies discovered in the 'long' data format could be traced back to the original datasets. After datasets were converted to the standardized format, each were analyzed and sent back to the contributor as standalone reports to review for quality control, assessment, and validation. For some contributors, this was an effort that required meticulous auditing by both parties to identify and resolve any errors or discrepancies.

In some cases, only size data was provided by the contributor. If the contributor indicated that they measured all organisms that they surveyed, then we used size data to create count data for that survey site.

Many contributors included weight data along with length data; the weight data was used to compute biomass (Equation 1). In cases where length data was provided without weight data, we used contributor provided length-to-weight conversion factors (i.e., species and site-specific  $\alpha$  and  $\beta$  values) to compute the weight of individuals. If species and site-specific conversion factors were not provided, we used species and shellfish management region-specific conversion factors developed by Washington Department of Fish and Wildlife or the Swinomish Indian Tribal Community (Bradbury et al. 2005, Barber et al. 2012). Length-to-weight conversion were performed using Equation 1.

Equation 1: Weight  $(g) = \alpha L^{\beta}$ 

Where *L* is the length in mm and  $\alpha$  and  $\beta$  are species-specific conversion constants.

### 2.3. Analysis

### 2.3.1. Overview and general calculations

Analysis focused on three main metrics measured before and after the June 2021 heatwave: population size, organism size, and population biomass. These metrics were summarized at the site level for each species measured. We note that different contributors conducted surveys with slightly different methods, leading to deferring sample size, quadrat size, and total survey area

among sites. Since analyses focus on before and after comparisons at a single site, methods and sample size are largely the same for each site. Contributor, site, and/or species-specific sample size and other survey methods, and how they were accommodated are detailed in each contributor's results section (Appendices B - J).

For the purposes of this report, we created a variable 'heatwave stage' with two levels, 'before' and 'after', to differentiate data collected before and after the June 2021 heatwave. We define the 2021 heatwave as occurring between 26 - 28 June 2021. Surveys conducted before 26 June 2021, were defined as 'before' and surveys conducted after 28 June 2021 were defined as 'after'. Some sites in the dataset are not sampled annually, therefore a 'before' survey may have been conducted in years prior to 2021. All 'after' surveys were conducted between July and September 2021. Specific details on surveys can be found in contributor-specific reports (Appendices B – J).

Count data was used to produce population size estimates. We first calculated density (counts per square meter or number/ $m^2$ ) shown in Equation 2.

Equation 2:  $density = \frac{counts}{sample area}$ 

Where *counts* is the number of individuals in a unique sample area, and *sample area* is the size of the sampling unit or quadrat in square meters. Species density (i.e., counts per square meter) was then converted to a population estimate in Equation 3.

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Equation 3: population estimate = density × survey area
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Where *survey area* is the total area, the survey is meant to characterize in square meters provided by the contributor. We then computed mean population estimate (hereafter 'abundance') across all samples and the 95% confidence interval (CI) for each unique combination of site, species, and heatwave stage. Ninety-five percent confidence intervals were calculated following Equation 4.

Equation 4: 95%  $CI = mean \pm (1.96 \times standard \, error)$ 

We used size data to compute mean length (+/- 95% CI) and population biomass for each unique combination of site, species, and heatwave stage. Mean length (+/- 95% CI) was computed from all measured individuals. For estimated population biomass, we first divided mass (grams) by the sample area to calculate mass per square meter shown in Equation 5.

Equation 5: mass per square meter =  $\frac{mass}{sample area}$ 

Where *mass* is the total mass of a species at a unique sample area and heatwave stage, and *sample area* is the size of the sampling unit or quadrat in square meters. Mass per square meter was then converted to population biomass in Equation 6.

Equation 6: population biomass = mass per square meter  $\times$  survey area

Where *survey area* is the total area of which the survey is meant to characterize in square meters. We then computed mean biomass across all samples (+/- 95% CI) for each unique combination of site, species, and heatwave stage.

For all data, records of 'zero' were examined carefully and were used or excluded from the above equations. Zeros were included in population estimate and population biomass calculations but were excluded from length calculations. Samples noted as 'flooded' were completely excluded from analysis as this denoted samples that would have been taken but were not due to high water. Samples noted as 'broken' were included in count analyses, but excluded from length and mass analysis unless a contributor provided value was available.

### 2.3.2. Statistical analysis

We implemented four measures of analysis: (1) percent difference, (2) Cohen's D effect size, and (3) Mann-Whitney U (MWU) tests, to compare abundance, length, and biomass between surveys before and after the June 2021 heatwave, and (4) Kolmogorov-Smirnov (KS) tests to investigate differences in length distribution before and after the heatwave by beach. All tests were applied to each unique site and species combination. Percent difference was calculated following Equation 7.

Equation 7: Percent difference = 
$$\frac{after-before}{before} \times 100$$

Where *after* is the mean population abundance, length, or biomass calculated after the heatwave, and *before* is the same measurements calculated before the heatwave.

Cohen's D effect size was calculated using the 'cohen.d' function with pooled standard deviation in the 'effsize' package in R (Torchiano 2016). Cohen's D, is a way to quantitatively measure the magnitude of the effect of an experimental treatment. Generally, Cohen's D is computed by taking the difference between groups and dividing that by the standard deviation. In this case, the groups are the measures before and after the heatwave. We used pooled (by heatwave stage) standard deviations due to differences in sample size and error between sampling periods for much of the data. See Torchiano (2016) for details on computation of pooled standard deviation. Cohen's D produces a range of values with no strict cut-off that determines significance or importance. However, general guidelines suggest Cohen's D of  $\pm 0.2$  or less are considered to be a small effect,  $\pm 0.2 - \pm 0.5$  a moderate effect,  $\pm 0.5 - \pm 0.8$  a large effect, and greater than  $\pm 0.8$  a very large effect (McLeod 2019).

Mann-Whitney U tests were used to test for a difference in abundance, length, and biomass. These tests are the non-parametric version of commonly used t-tests and were used to accommodate for the non-normal distribution many datasets. We used the 'wilcox.test' function in the 'stats' package in R (R Core Team 2022) to perform Mann-Whitney U tests.

Kolmogorov-Smirnov tests are used to test for a difference in a distribution of data compared to a reference distribution. While often used to compare a sample distribution to a known distribution, such as a normal or Poisson distribution, they can also be used to compare two sample distributions, which is what we do here. We used the 'ks.test' function with two-sided p-values in the 'dgof' package in R (Arnold and Emerson 2011) to perform Kolmogorov-Smirnov tests.

### 2.3.3. Presentation of results

Following the analytical approach above, results are reported by site and species. These results are presented in two formats: regional and contributor. Regional results (Section 3) bring together contributor specific results for a species and place them in a regional context (Figure 1). Contributor specific results, as well as detailed data and statistical analysis for each contributor dataset and are found in Appendices B-J.



Figure 1: Study area and locations of data collection and analysis. Note that each species presented in the regional results is not necessarily present at all sites. See Appendix A for specifics on what species are present at each data collection site.

## 3. Regional results

### 3.1. Analytical considerations

The goal of the regional analysis was to place site-specific results in a broader regional context for Puget Sound. We collated and plotted results on all available metrics described above for all species if data were available at two or more sites (Table 1). While this analysis uses the same metrics for all species (percent difference and Cohen's D effect size), many contributors methods differed slightly in site sample size, survey area, and sampling size (Appendix A). For example, the beach area or number of sampled quadrats may have differed slightly between pre- and postheatwave surveys. These inconsistencies, while difficult to avoid, introduce sampling error specific to each site which cannot be quantitatively normalized among sites. Therefore, regional analyses should be viewed as general patterns and not necessarily direct tests of location-specific effects of the heatwave on species. Future analysis could be done to directly address methodological inconsistencies among data contributors to build a robust regional analysis framework on heatwave effects but that was beyond the scope of this project.

Table 1: Number (N) of sites with abundance, biomass, or length data for each species included in the regional analysis. Multiple data types are present at most sites, and therefore, the N sites values are not exclusive of each other. N sites presented here reflects data availability only, and not necessarily data suitability for analysis. Sites may be omitted from regional analysis and if so, are detailed in Sections 3.2 - 3.10.

		N sites		
Species	Common name	Abundance	Biomass	Length
Clinocardium nuttallii	Cockle clam	4	2	2
Crassostrea gigas	Pacific oyster	7	0	5
Leukoma staminea	Littleneck clam	7	7	6
Mya arenaria	Softshell clam	5	4	1
Nuttallia obscurata	Purple varnish clam	3	0	2
Ostrea lurida	Olympia oyster	12	0	3
Ruditapes philippinarum	Manila clam	12	12	11
Saxidomus gigantea	Butter clam	2	3	2
Tresus capax	Horse clam	2	2	2

### 3.2. Manila clams

Twelve sites recorded Manila clam (*Ruditapes philippinarum*) abundance and biomass, and 10 sites had records of length before and after the June 2021 heatwave. Site E was removed from analyses due to its large increase (> 300%) in abundance after the heatwave. Specifically, while patterns observed at site E are presented in the contributor report (Appendix G), its inclusion in the regional assessment masked patterns observed at other sites and the site was therefore removed. Note that surveys at three sites (J, R, and S) did not collect data on clams smaller than the legal harvest size of 38mm (1.5in) (as denoted by \* in Figure 2), nor were these surveys designed to collect data on smaller clams. This difference in survey approach may affect how data from sites excluding clams smaller than 38mm compared to other sites that measured all individuals for this species.

Across the region, Manila clams display a range of patterns for all metrics following the heatwave (Figure 2). Abundance and biomass across the sites ranged from approximately 50% increase to 60% decrease. There was also a slight pattern of more negative effects of the heatwave on abundance and biomass at the more southerly sites. Length also varied from before and after the heatwave but at a much smaller magnitude. The range of heatwave effects on this species suggests that local, site-specific, factors may play a large role in how Manila clams were impacted by the heatwave. Although challenging to pinpoint these factors, it appears that southerly sites experienced greater losses than northerly sites (as demonstrated by the Cohen's D effect size). This observation aligns with the higher air temperatures and later low tides in southern reaches of the Puget Sound region during the heatwave (Raymond et al. 2022).

### 3.3. Pacific oyster

Six sites recorded Pacific oyster (*Crassostrea gigas*) abundance and four sites recorded lengths before and after the heatwave. Length data from site N was removed because lengths were only recorded after the heatwave. Biomass data from site M was removed from analyses due to its large increase (> 400%) in abundance after the heatwave which may have resulted from a significant recruitment event between surveys. Results from this site are examined and discussed in more detail in Appendix J but were removed from the regional assessment as it would mask patterns at other sites.

We observed increases in Pacific oyster abundance at all but two sites after the heatwave (Figure 3). However, there was universal decrease in length ranging from -15.0% to -36.7% compared to before heatwave surveys. This increase in observed abundance was unexpected given the generally negative observations made post-heatwave in our qualitative assessment (Raymond et al. 2022). The combination of a limited effect on population size with good evidence of a reduction in length suggests a demographic shift in Pacific oyster population across the region. One potential contributing factor is the time span between 'before' and 'after' heatwave surveys. Five of the six before-heatwave surveys were conducted in 2020, often over a year before the 'after' heatwave survey. Following the heatwave, Pacific oyster populations appear to be dominated by small individuals, possibly indicating that a recruitment event occurred before the heatwave but after the "before heatwave" surveys were complete (Figure 3). Pacific oysters reproduce naturally in parts of Puget Sound, including Hood Canal where our observations come from. Therefore, it is likely that 'before' heatwave surveys at these sites did not accurately represent the true population immediately before the heatwave. This demographic shift is further evident in the analysis presented in Appendix J. We observed a high degree of variability in population estimates, similar to other species analyzed in this report.

#### 3.4. Olympia oyster

Twelve sites recorded Olympia oyster (*Ostrea lurida*) abundance and three sites recorded lengths before and after the heatwave. We observed a range of heatwave effects on abundance, ranging from -85% to 78.6% change across sites, with most sites (8 of 12) exhibiting a decrease. While effect size analysis suggests a weak heatwave effect at most sites, sites L and O suggest a stronger negative heatwave effect. The three sites with size data ranged from -21.5% to 18.3% change in length (Figure 4). Two of these sites, H and L experienced an increase in length. This length increase, along with the variability in abundance, suggests that site-specific factors may have modulated heatwave effects across the region.

Patterns of abundance at sites G, H, and I suggest location-specific effects. Sites G and H are separated by only ~100m and exhibited a very similar decline in abundance (-36.9% and -44.4% respectively). In contrast, site I had the opposite response, exhibiting a 43.5% increase in abundance. Site I has similar substrate and aspect as sites G and H but is separated by ~10km (straight line distance). These observations are likely a result of differences in both location and duration between before and after sampling events. Sites G and H had sampling events only months apart whereas site I had a year between sampling. Site I also demonstrated evidence of a strong recruitment event in 2021 (Appendix C) that was not observed at sites G and H.

### 3.5. Littleneck clam

Seven sites recorded littleneck clam (*Leukoma staminea*) abundance, and six sites recorded length and biomass. We observed a wide range of heatwave effects on abundance and biomass from greater than 75% increase to near 100% decrease (Figure 5). Despite this large range in percent change, effect size results suggest little influence of the heatwave. This result is likely due to the large degree of variability in estimated effect. We observed a smaller degree of percent change in length, ranging from -11% to - 3% with a similarly small effect size values. Interestingly, site F stands out as showing good evidence of a decline in abundance and biomass without exhibiting a change in length, suggesting that all size classes existing before the heatwave were affected somewhat equally at this site. Note that data collection at three sites (J, R, and S) did not include measuring clams smaller than the legal harvest size of 38mm (1.5in) (as denoted by \* in Figure 5) shell width so any heatwave effect to smaller size clams would not have been detected. Furthermore, the surveys at these three sites were not designed to target <38mm individuals, therefore comparing these surveys to others should be done with caution.

### 3.6. Cockle clam

Four sites recorded cockle clam (*Clinocardium nuttallii*) abundance and two sites recorded length and biomass. We observed reductions in abundance after the heatwave in three out of four sites and in biomass at both sites (Figure 6). However, effect size analysis suggests little to weak effect of the heatwave. As with other species, the lack of evidence for strong effects is likely due to high variability in the data. The observed reduction in abundance and biomass at sites D and E but the lack of any major effect on length suggests that all size classes experienced mortality somewhat equally.

### 3.7. Softshell clam

Five sites recorded softshell clam (*Mya arenaria*) abundance and one site recorded length before and after the heatwave. Sites J, R, and S did record lengths but had limited sample sizes (N < 30). Furthermore, site E, which has a naturally small population of softshell clams, did not record this species in the 'after' heatwave survey, leaving only lengths from the 'before' heatwave survey and precluding addition in the regional analysis. We observed a range of heatwave effects on abundance ranging from 50% to -100%, although effect size analysis suggests little to weak effect of the heatwave (Figure 7).

### 3.8. Butter clam

Two sites recorded butter clam (*Saxidomus gigantea*) abundance, biomass, and length before and after the heatwave. Site S and R also reported some butter clam data but had limited sample sizes (N < 30) and/or did not record data before and after the heatwave, precluding use in this regional analysis. Opposite patterns of heatwave effects on abundance and biomass were observed for

sites D and E with -24% and 52%, and -43% to 31%, respectively (Figure 8). Similar to observations for several other species, the effect size analysis suggested little effect of the heatwave. On average, butter clams at site D were ~5mm larger than site E both before and after the heatwave. This length pattern, in conjunction with patterns of abundance and biomass, may suggest that smaller length clams were less susceptible to heat stress than larger ones, but further data and assessment would be required for validation.

### 3.9. Horse clam

Two sites recorded horse clam (*Tresus capax*) abundance, biomass, and length before and after the heatwave. Site E horse clam data had limited sample size (N < 30), precluding use in the regional analysis for biomass or length. Change in abundance for site D and E was -34.4% to - 2.9%, respectfully (Figure 9). However, effect size analysis indicated weak heatwave effects due to high variability in the data. Mean clam size at site D increased slightly after the heatwave, indicating that small individuals may be more susceptible to thermal stress than large ones, but further data and assessment would be required for validation.

### 3.10. Purple varnish clam

Two sites recorded purple varnish clam (*Nuttallia obscurata*) abundance and length before and after the heatwave. Site R also reported purple varnish clam lengths but had limited sample size (N < 30), precluding use in the regional analysis. We observed little effect of the heatwave on abundance or length of purple varnish clams at both sites (Figure 10). Mean size differed by ~4mm between sites both before and after the heatwave indicating little size-specific effect of thermal stress.



Figure 2: Regional results of heatwave effects on Manila clams (Ruditapes philippinarum) including location of sites (A), mean size (B), percent difference and effect size of abundance (C, D), biomass (E, F) and length (G, H). \* denotes sites where only clams  $\geq$  38mm (1.5in) were counted. Magnitude of Cohen's D effect size is indicated as strong (red), moderate (yellow), weak (green), none (white).



Figure 3: Regional results of heatwave effects on Pacific oysters (Crassostrea gigas) including location of sites (A), mean size (B), percent difference and effect size of abundance (C, D), and length (E, F). Magnitude of Cohen's D effect size is indicated as strong (red), moderate (yellow), weak (green), none (white).



Figure 4: Regional results of heatwave effects on Olympia oysters (Ostrea lurida) including location of sites (A), mean size (B), percent difference and effect size of abundance (C, D), and length (E, F). Magnitude of Cohen's D effect size is indicated as strong (red), moderate (yellow), weak (green), none (white).



Figure 5: Regional results of heatwave effects on littleneck clams (Leukoma staminea) including location of sites (A), mean size (B), percent difference and effect size of abundance (C, D), biomass (E, F) and length (G, H). \* denotes sites where only clams  $\geq 38mm$  (1.5in) were counted. Magnitude of Cohen's D effect size is indicated as strong (red), moderate (yellow), weak (green), none (white).



Figure 6:Regional results of heatwave effects on cockle clams (Clinocardium nuttallii) including location of sites (A), mean size (B), percent difference and effect size of abundance (C, D), biomass (E, F) and length (G, H). Magnitude of Cohen's D effect size is indicated as strong (red), moderate (yellow), weak (green), none (white).



Figure 7: Regional results of heatwave effects on softshell clams (Mya arenaria) including location of sites (A), mean size (B), and percent difference and effect size of abundance (C, D). Magnitude of Cohen's D effect size is indicated as strong (red), moderate (yellow), weak (green), none (white).



Figure 8: Regional results of heatwave effects on butter clams (Saxidomus gigantea) including location of sites (A), mean size (B), percent difference and effect size of abundance (C, D), biomass (E, F) and length (G, H). Magnitude of Cohen's D effect size is indicated as strong (red), moderate (yellow), weak (green), none (white).



Figure 9: Regional results of heatwave effects on horse clams (Tresus capax) including location of sites (A), mean size (B), percent difference and effect size of abundance (C, D), biomass (E, F) and length (G, H). Magnitude of Cohen's D effect size is indicated as strong (red), moderate (yellow), weak (green), none (white).



Figure 10: Regional results of heatwave effects on purple varnish clam (Nuttallia obscurata) including location of sites (A), mean size (B), percent difference and effect size of abundance (C, D), and length (E, F). Magnitude of Cohen's D effect size is indicated as strong (red), moderate (yellow), weak (green), none (white).

### 4. Discussion

This report provides the best available quantitative analysis of species-specific responses to the June 2021 heatwave of Puget Sound intertidal clams and oysters to date. The species examined here displayed varied responses to the heatwave ranging from strongly negative to positive, with the exception of cockles that had no observed positive change. The patterns observed in this quantitative assessment reinforce observations from the qualitative assessment that suggested heatwave effects likely differ by species and location (Raymond et al. 2022). While the data were highly variable, some notable patterns did emerge. Pacific oyster (*Crassostrea gigas*) data suggest somewhat positive effects of the heatwave across the region. However, as noted above, the 'before' heatwave effects. Furthermore, all Pacific oyster data come from sites within the Hood Canal basin where they are known to reproduce naturally with large, episodic recruitment events. In contrast, Manila clams and Olympia oysters suggest somewhat negative responses to the heatwave across the region; however, results vary greatly across locations.

We did not observe any strong regional or species-specific patterns in response to the 2021 heatwave. Overall, Cohen's D effect size for the majority of species indicates little to no effect of the heatwave. Furthermore, many species show a high degree of variability of the estimated effect. This result introduces statistical uncertainty even though multiple species displayed large percent differences (> 20%) from before to after the heatwave across all metrics but particularly in abundance and biomass.

Examining data across multi-contributor scales presents unique challenges for the analysis and assessment of results. At the smaller scales, such as a site or a collection of sites monitored by the same organization over time, this may not present any issues as there is a substantial baseline for assessment or comparison. Contributor organizations and their staff tend to have an innate sense of the status of a species/location based on many years of experience and institutional knowledge (Raymond et al. 2022). Importantly, for these reasons, the lack of strong statistical evidence does not necessarily signal the lack of biologically significant changes in populations. To a third party, this report could be read as showing little to no effect of the heatwave. However, we recognize that most, if not all, contributors would disagree with that sentiment based on the findings of Raymond et al. (2022) and their own understanding of shellfish populations at beaches they monitor or manage. A stark example of this is the widespread observation in June and July 2021 of significant cockle mortality post-heatwave. While the quantitative data presented here does suggest negative effects to cockles, the magnitude and lack of strong statistical support does not align with previous observations. Herein lies perhaps the most important result from this analysis, that in many cases, variability is swamping mean effects.

Multiple unaccounted factors may have contributed to the observed variability in the data. Nearly all the sites examined in this study are open to recreational or tribal harvest of many of the species studied, especially Manila, littleneck and cockle clams, and Pacific oysters. If harvest occurred, when it occurred, at what magnitude, and on what species was beyond the scope of this analysis but may be an important area of future investigation and analysis. Finally, clams and oysters are not distributed evenly across the tidelands (e.g., Barber et al. 2012). Indeed, the patchy nature of these species can result in a transect being deployed over a dense tract of a

particular species in one survey but not the next. All these factors are potentially exacerbated by the variable time span between before and after surveys. It is a reasonable assumption that larger time spans leave more opportunity for additional external factors, include those stated above, to affect clam and oyster populations in ways that cannot be accounted for in the available survey data and this analysis.

Identifying and describing patterns of change, or lack thereof, at various scales is a central question and challenge in ecology that often does not have a straightforward solution (Levin 1992). This fact is clear in this dataset and the analysis. Standard survey methods designed to assess species abundance and biomass have been effective for managing clam and oyster populations for decades. These standardized methods are extremely valuable and continue to be sufficient for annual population estimates, setting harvest quotas and examination of long-term population trends (Barber et al. 2019). Our analysis testing for heatwave effects reduced this dataset to essentially two points per site, before and after the heatwave, thereby reducing the time scale of interest. Our analysis highlights that this reduced scale of assessment can lead to data variability overpowering the mean effects, making the detection of patterns challenging, if not impossible, at both the regional and site scale. Future analyses could consider selecting sites with longer-term datasets to determine if heatwave impacts present as a discrete anomaly when compared to decadal patterns in population metrics.

Considering these results, and the increased likelihood of extreme weather events, co-managers and other stakeholders may need to supplement their population survey techniques with the following in mind. First, is there a need at the beach and/or region-wide level to be able to assess the effect of acute events on intertidal bivalve populations? If so, are the current survey methods and the data they produce sufficient for testing these effects? We suggest that there is a strong need for contributors to be able to rapidly assess acute impacts of extreme weather events on intertidal bivalve populations. However, based on results in this report, we do not believe that the current population survey methods are sufficient for accurately assessing acute impacts on species that show a high degree of patchiness in their distribution. It is important to note that we understand that any significant change or deviation from standard population survey methodology would present a significant challenge to co-managers, as such methods have been standardized and implemented by several organizations for many years. Thus, a different solution will be needed such as incorporating consistent and/or discrete supplemental surveys to address acute needs. This may be best accomplished through the development of a rapid response plan that provides a standardized framework for baseline data collection and responding to extreme events when they occur.

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# Appendix A

				Before survey	After survey
Species	Contributor (Appendix)	Site	Data type	date	date
Clinocardium nuttallii	Swinomish Tribal Community (I)	D	abundance	6/17/2019	7/21/2021
Clinocardium nuttallii	Swinomish Tribal Community (I)	D	biomass	6/17/2019	7/21/2021
Clinocardium nuttallii	Swinomish Tribal Community (I)	D	length	6/17/2019	7/21/2021
Clinocardium nuttallii	Swinomish Tribal Community (I)	Е	abundance	5/27/2021	7/20/2021
Clinocardium nuttallii	Swinomish Tribal Community (I)	Е	biomass	5/27/2021	7/20/2021
Clinocardium nuttallii	Swinomish Tribal Community (I)	E	length	5/27/2021	7/20/2021
Clinocardium nuttallii	WA Dept. Fish and Wildlife (I)	J	abundance	6/14/2021	8/8/2021
Clinocardium nuttallii	WA Dept. Fish and Wildlife (I)	S	abundance	5/28/2021	8/22/2021
Crassostrea gigas	Port Gamble S'Klallam Tribe (D)	Q	abundance	4/30/2021	4/20/2022
Crassostrea gigas	Puget Sound Restoration Fund (E)	L	abundance	5/27/2021, 5/28/2021	8/20/2021
Crassostrea gigas	WA Dept. Fish and Wildlife (J)	J	abundance	7/8/2020	8/19/2021
Crassostrea gigas	WA Dept. Fish and Wildlife (J)	J	length	7/8/2020	8/19/2021
Crassostrea gigas	WA Dept. Fish and Wildlife (J)	K	abundance	7/8/2020	8/19/2021
Crassostrea gigas	WA Dept. Fish and Wildlife (J)	K	length	7/8/2020	8/19/2021
Crassostrea gigas	WA Dept. Fish and Wildlife (J)	M	abundance	6/9/2020	8/6/2021
Crassostrea gigas	WA Dept. Fish and Wildlife (J)	M	length	6/9/2020	8/6/2021
Crassostrea gigas	WA Dept. Fish and Wildlife (J)	0	abundance	8/17/2020	7/9/2021
Crassostrea gigas	WA Dept. Fish and Wildlife (J)	0	length	8/1//2020	7/9/2021
Crassostrea gigas	WA Dept. Fish and Wildlife (J)	S	abundance	8/1/2020	8/20/2021
Crassostrea gigas	WA Dept. Fish and Wildlife (J)	5	length	8/1/2020	8/20/2021
Cryptomya californica	Swinomish Tribal Community (H)	D	abundance	6/17/2019	9/10/2021
Leukoma staminea	Jamestown S Klallam Tribe (B)	F	abundance	6/23/2021	8/10/2021
Leukoma staminea	Jamestown S Klanam Tribe (B)	Г	longth	6/22/2021	8/10/2021
Leukoma staminea	Jamestown S Klanam Tribe (D)	Г	abundanca	6/22/2021	5/10/2021
Leukoma staminea	Port Gamble S'Klallam Tribe (D)	r D	biomass	6/22/2021	5/16/2022
Leukoma staminea	Port Gamble S'Klallam Tribe (D)	r D	length	6/22/2021	5/16/2022
Leukoma staminea	Swinomish Tribal Community (H)	D	abundance	6/17/2019	7/21/2021
Leukoma staminea	Swinomish Tribal Community (H)	D	biomass	6/17/2019	7/21/2021
Leukoma staminea	Swinomish Tribal Community (H)	D	length	6/17/2019	7/21/2021
Leukoma staminea	Swinomish Tribal Community (H)	F	abundance	5/27/2021	7/20/2021
Leukoma staminea	Swinomish Tribal Community (H)	E	biomass	5/27/2021	7/20/2021
Leukoma staminea	Swinomish Tribal Community (H)	E	length	5/27/2021	7/20/2021
Leukoma staminea	WA Dept. Fish and Wildlife (I)	J	abundance	6/14/2021	8/8/2021
Leukoma staminea	WA Dept. Fish and Wildlife (I)	J	biomass	6/14/2021	8/8/2021
Leukoma staminea	WA Dept. Fish and Wildlife (I)	J	length	6/14/2021	8/8/2021
Leukoma staminea	WA Dept. Fish and Wildlife (I)	R	abundance	4/27/2021	8/10/2021
Leukoma staminea	WA Dept. Fish and Wildlife (I)	R	biomass	4/27/2021	8/10/2021
Leukoma staminea	WA Dept. Fish and Wildlife (I)	R	length	4/27/2021	8/10/2021
Leukoma staminea	WA Dept. Fish and Wildlife (I)	S	abundance	5/28/2021	8/22/2021
Leukoma staminea	WA Dept. Fish and Wildlife (I)	S	biomass	5/28/2021	8/22/2021
Mya arenaria	Swinomish Tribal Community (H)	D	abundance	6/17/2019	7/21/2021
Mya arenaria	Swinomish Tribal Community (H)	D	biomass	6/17/2019	7/21/2021
Mya arenaria	Swinomish Tribal Community (H)	D	length	6/17/2019	7/21/2021
Mya arenaria	Swinomish Tribal Community (H)	E	abundance	5/27/2021	7/20/2021
Mya arenaria	WA Dept. Fish and Wildlife (I)	J	abundance	6/14/2021	8/8/2021
Mya arenaria	WA Dept. Fish and Wildlife (I)	J	biomass	6/14/2021	8/8/2021
Mya arenaria	WA Dept. Fish and Wildlife (I)	R	abundance	4/27/2021	8/10/2021
Mya arenaria	WA Dept. Fish and Wildlife (I)	R	biomass	4/27/2021	8/10/2021
Mya arenaria	WA Dept. Fish and Wildlife (I)	S	abundance	5/28/2021	8/22/2021
Mya arenaria	WA Dept. Fish and Wildlife (I)	S	biomass	5/28/2021	8/22/2021
Nuttallia obscurata	WA Dept. Fish and Wildlife (I)	J	abundance	6/14/2021	8/8/2021
Nuttallia obscurata	WA Dept. Fish and Wildlife (I)	J	length	6/14/2021	8/8/2021
Nuttallia obscurata	WA Dept. Fish and Wildlife (I)	S	abundance	5/28/2021	8/22/2021
Nuttallia obscurata	WA Dept. Fish and Wildlife (I)	S	length	5/28/2021	8/22/2021
Ostrea lurida	Jamestown S'Klallam Tribe (B)	G	abundance	5/25/2021	8/18/2021
Ostrea lurida	Jamestown S'Klallam Tribe (B)	Н	abundance	5/27/2021	7/26/2021
Ostrea lurida	Jamestown S'Klallam Tribe (B)	H	length	5/27/2021	7/26/2021
Ostrea lurida	Jeff. Co. Marine Resource Committee (C)	I	abundance	8/2/2020	7/26/2021

				Before survey	After survey
Species	Contributor (Appendix)	Site	Data type	date	date
Ostrea lurida	Jeff. Co. Marine Resource Committee (C)	Ι	length	8/2/2020	7/26/2021
Ostrea lurida	Puget Sound Restoration Fund (E)	L	abundance	5/27/2021, 5/28/2021	8/20/2021
Ostrea lurida	Puget Sound Restoration Fund (E)	L	length	5/27/2021, 5/28/2021	8/20/2021
Ostrea lurida	Skagit Co. Marine Resource Committee (G)	Α	abundance	6/12/2018	7/10/2021
Ostrea lurida	Skagit Co. Marine Resource Committee (G)	В	abundance	6/15/2018	7/8/2021
Ostrea lurida	Swinomish Tribal Community (J)	С	abundance	5/25/2021	7/15/2021
Ostrea lurida	Swinomish Tribal Community (J)	D	abundance	5/24/2021	7/26/2021
Ostrea lurida	WA Dept. Fish and Wildlife (J)	J	abundance	7/8/2020	8/19/2021
Ostrea lurida	WA Dept. Fish and Wildlife (J)	K	abundance	7/8/2020	8/19/2021
Ostrea lurida	WA Dept. Fish and Wildlife (J)	М	abundance	6/9/2020	8/6/2021
Ostrea lurida	WA Dept, Fish and Wildlife (J)	0	abundance	8/17/2020	7/9/2021
Ruditapes philippinarum	Jamestown S'Klallam Tribe (B)	F	abundance	6/23/2021	8/10/2021
Ruditapes philippinarum	Jamestown S'Klallam Tribe (B)	F	biomass	6/23/2021	8/10/2021
Ruditapes philippinarum	Jamestown S'Klallam Tribe (B)	F	length	6/23/2021	8/10/2021
Ruditapes philippinarum	Port Gamble S'Klallam Tribe (D)	Р	abundance	6/22/2021	5/16/2022
Ruditapes philippinarum	Port Gamble S'Klallam Tribe (D)	P	biomass	6/22/2021	5/16/2022
Ruditanes philippinarum	Port Gamble S'Klallam Tribe (D)	P	length	6/22/2021	5/16/2022
Ruditapes philippinarum	Squaxin Island Tribe (G)	T	abundance	4/8/2020	7/27/2021
Ruditapes philippinarum	Squaxin Island Tribe (G)	T	biomass	4/8/2020	7/27/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	T	length	4/8/2020	7/27/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	Ū	abundance	6/7/2021	7/8/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	U	biomass	6/7/2021	7/8/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	U	length	6/7/2021	7/8/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	W	abundance	6/26/2020	7/13/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	W	biomass	6/26/2020	7/13/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	W	length	6/26/2020	7/13/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	x	abundance	6/24/2021	7/19/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	X	biomass	6/24/2021	7/19/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	X	length	6/24/2021	7/19/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	X V	abundance	3/31/2021	7/7/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	v	biomass	3/31/2021	7/7/2021
Ruditanes philippinarum	Squaxin Island Tribe (G)	Y	length	3/31/2021	7/7/2021
Ruditanes philippinarum	Swinomish Tribal Community (H)	D	abundance	6/17/2019	7/21/2021
Ruditanes philippinarum	Swinomish Tribal Community (H)	D	biomass	6/17/2019	7/21/2021
Ruditanes philippinarum	Swinomish Tribal Community (H)	D	length	6/17/2019	7/21/2021
Ruditanes philippinarum	Swinomish Tribal Community (H)	F	abundance	5/27/2021	7/20/2021
Ruditanes philippinarum	Swinomish Tribal Community (H)	E	biomass	5/27/2021	7/20/2021
Ruditanes philippinarum	Swinomish Tribal Community (H)	E	length	5/27/2021	7/20/2021
Ruditanes philippinarum	WA Dept. Fish and Wildlife (I)	I	abundance	6/14/2021	8/8/2021
Ruditanes philippinarum	WA Dept. Fish and Wildlife (I)	J	biomass	6/14/2021	8/8/2021
Ruditanes philippinarum	WA Dept. Fish and Wildlife (I)	J	length	6/14/2021	8/8/2021
Ruditanes philippinarum	WA Dept. Fish and Wildlife (I)	R	abundance	4/27/2021	8/10/2021
Ruditanes philippinarum	WA Dept. Fish and Wildlife (I)	R	biomass	4/27/2021	8/10/2021
Ruditanes philippinarum	WA Dept. Fish and Wildlife (I)	S	abundance	5/28/2021	8/22/2021
Ruditanes philippinarum	WA Dept. Fish and Wildlife (I)	S	biomass	5/28/2021	8/22/2021
Ruditanes philippinarum	WA Dept. Fish and Wildlife (I)	S	length	5/28/2021	8/22/2021
Saxidomus gigantea	Swinomish Tribal Community (H)	D	abundance	6/17/2019	7/21/2021
Saxidomus gigantea	Swinomish Tribal Community (H)	D	biomass	6/17/2019	7/21/2021
Saxidomus gigantea	Swinomish Tribal Community (H)	D	length	6/17/2019	7/21/2021
Saxidomus gigantea	Swinomish Tribal Community (H)	F	abundance	5/27/2021	7/20/2021
Saxidomus gigantea	Swinomish Tribal Community (H)	F	hiomass	5/27/2021	7/20/2021
Saxidomus gigantea	Swinomish Tribal Community (H)	F	length	5/27/2021	7/20/2021
Saxidomus gigantea	WA Dept Fish and Wildlife (I)	I	hiomass	6/14/2021	8/8/2021
Tresus capar	Swinomish Tribal Community (H)	J D	abundance	6/17/2021	7/21/2021
Tresus capax	Swinomish Tribal Community (H)	D	hiomass	6/17/2019	7/21/2021
Tresus capax	Swinomish Tribal Community (H)	<u>д</u>	length	6/17/2019	7/21/2021
Tresus capax	Swinomish Tribal Community (H)	F	abundance	5/27/2021	7/20/2021
Tresus capar	Swinomish Tribal Community (H)	F	hiomass	5/27/2021	7/20/2021
Tresus capar	Swinomish Tribal Community (H)	F	length	5/27/2021	7/20/2021
2. como cupun				0, 1, 2021	

# Appendix B: Jamestown S'Klallam Tribe

### 1. Data overview

The Jamestown S'Klallam Tribe (JST) measured abundance and length of *Leukoma staminea* (Littleneck clam), *Ruditapes philippinarum* (Manila clam), and *Ostrea lurida* (Olympia oyster) at Sequim Bay Calms (SBC – site F), and Sequim 1.5 acre (site H) before and at the June 2021 heatwave. They only measured abundance of *O. lurida* at Sequim Subsistence (site G). Sample sizes and number of sampled individuals vary between surveys.

Table 1: Overview of data from Jamestown S'Klallam Tribe. Total survey area, size of samples, number of samples, and number of individual lengths are displayed for each species and site as the value before/after the heatwave.

		Survey area	Sample	Ν	N indv.		
Species	Site	(m <sup>2</sup> )	area (m <sup>2</sup> )	samples	lengths	Before date	After date
L. staminea	SBC	1918/1918	0.25	37/43	504/281	23-June-21	10-Aug-21
R. philippinarum	SBC	1918/1918	0.25	38/43	170/180	23-June-21	10-Aug-21
	Sequim 1.5 acre	6146/6146	13.25/16.25	53/65	51/44	27-May-21	26-July-21
O. lurida	Sequim	1195/1195	0.25	53/51	_	25-May-21	18-Aug-21
	Subsistence	1195/1195	0.25	55/51	-	23-1v1ay-21	10-Aug-21

### 2. Results

2.1. Abundance



Figure 1: Population estimate of L. staminea, R. philippinarum, and O. lurida at sites sampled by Jamestown S'Klallam Tribe before and after the June 2021 heatwave.

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Table 2: Results of the difference in population size of L. staminea, R. philippinarum, and O. lurida at sites sampled by Jamestown S'Klallam Tribe before and after the June 2021 heatwave.

		Effect size - Cohen's D				
Species	Site	Percent difference	(95% CI)	MWU p-value		
L. staminea	SBC	-39.1	0.43 (-0.09 - 0.96)	0.122		
R. philippinarum	SBC	-10.4	0.09 (-0.58 - 0.76)	0.913		
O lumida	Sequim 1.5 acre	-44.4	0.28 (-0.09 - 0.65)	0.682		
0. iuriaa	Sequim Subsistence	-36.9	0.21 (-0.21 – 0.64)	0.971		

### 2.2. Size



Figure 2: Size-frequency histograms (A) and box plots (B) of L. staminea and R. philippinarum lengths measured at Jamestown S'Klallam Tribe site SBC before and after the June 2021 heatwave. Vertical bars in box plots represent median.



Table 3: Results of the difference of L. staminea, R. philippinarum lengths measured at Jamestown S'Klallam Tribe at SBC before and after the June 2021 heatwave.

		Percent	Effect size - Cohen's		
Species	Site	difference	D (95% CI)	MWU p-value	KS test p-value
L. staminea	SBC	1.0	-0.05 (-0.19 – 0.10)	0.570	0.846
O. lurida	Sequim 1.5 acre	10.9	-0.30 (-0.71 – 0.11)	0.200	0.345
R. philippinarum	SBC	-1.0	-0.06 (-0.17 – 0.28)	0.475	0.586

### 2.3. Biomass



Figure 3: Biomass estimate of L. staminea and, R. philippinarum at SBC sampled by Jamestown S'Klallam Tribe before and after the June 2021 heatwave.

Table 4: Results of the difference of L. staminea, R. philippinarum biomass measured at Jamestown S'Klallam Tribe site SBC before and after the June 2021 heatwave.

			Effect size - Cohen's D	
Species	Site	Percent difference	(95% CI)	MWU p-value
L. staminea	SBC	-52.5	0.54 (0.09 - 1.00)	0.005
R. philippinarum	SBC	-10.2	0.06 (-0.38 - 0.50)	0.979

### 3. Discussion

There is little evidence that the estimated population size of *L. staminea*, *R. philippinarum*, and *O. lurida* was affected by the June 2021 heatwave from a statistical perspective, despite reductions in estimated mean biomass (Figure 1; Table 2). Similarly, *L. staminea*, *R. philippinarum* mean size and size distribution and biomass of *R. philippinarum* was also not affected by the heatwave. However, there is strong evidence for a decrease in biomass of *L. staminea* at SBC after the heatwave, representing a 52.5% reduction in biomass (p = 0.005; Figure 3; Table 4). As with many data sets analyzed in this project, the degree of observed variability may be limiting the power to detect statistical differences. However, abundance and biomass data presented here suggest large decreases post heatwave.

These data provide a good opportunity to compare non-native *R. philippinarum* (Manila clams) to their native counterpart, *L. staminea* (littleneck clams). In general, the data suggest that *L. staminea* experienced intense negative effects from the heatwave compared to *R. philippinarum* (Figure 2, 3; Table 2, 4). As these species are similar in their habitat and ecology, it suggests that the native *L. staminea* may be more susceptible to extreme heat stress. In the face of climate change and the likelihood of more frequent and intense heatwaves, this pattern suggests that *L. staminea* may be in danger of widespread population losses.

# Appendix C: Jefferson County MRC

### 1. Data overview

The Jefferson County Marine Resource Committee (MRC) measured abundance and length of *Ostrea lurida* at Discovery Bay (Disco – site I) before and at the June 2021 heatwave. Sample sizes vary somewhat between surveys but are scale with the total area sampled.

Table 1: Overview of data from Jefferson Count MRC. Total survey area, size of samples, number of samples, and number of individual lengths are displayed for each species and site as the value before/after the heatwave.

		Survey area	Sample	Ν	N indv.		
Species	Site	(m <sup>2</sup> )	area (m <sup>2</sup> )	samples	lengths	Before date	After date
O. lurida	Disco	2023/1826	0.25	83/60	420/460	2-Aug-20	26-July-21

### 2. Results

2.1. Abundance



Figure 1: Population estimate of O. lurida sampled by Jefferson MRC at Discovery Bay before and after the June 2021 heatwave.

Table 2: Results of the difference in population size of O. lurida sampled by Jefferson MRC at Discovery Bay before and after the June 2021 heatwave.

			Effect size - Cohen's D	
Species	Site	Percent difference	(95% CI)	MWU p-value
O. lurida	Disco	43.5	-0.17 (-0.51 – 0.16)	0.549

2.2. Size



Figure 2: Size-frequency histograms (A) and box plots (B) of O. lurida lengths measured at Jefferson MRC site Discovery Bay before and after the June 2021 heatwave. Vertical bars in box plots represent median.

Table 3: Results of the difference in O. lurida lengths measured at Jefferson MRC site Discovery Bay before and after the June 2021 heatwave.

		Percent	Effect size - Cohen's		
Species	Site	difference	D (95% CI)	MWU p-value	KS test p-value
O. lurida	Disco	-21.8	0.59 (0.45 - 0.73)	< 0.001	< 0.001

### 3. Discussion

There is little evidence that the estimated population size of *O. lurida* was affected by the June 2021 heatwave from a statistical perspective. However, there is strong evidence for a decrease in *O. lurida* size and overall size distribution (p < 0.001, p < 0.001; Figure 2; Table 3). *O. lurida* decreased from 40.7mm before the heatwave to 31.9mm after the heatwave. Visual examination of size distributions suggests a decrease in abundance of 40mm to 60mm individuals and an increase in 10mm to 30mm individuals.

Much of the *O. lurida* data presented in this report does not include size data leaving a knowledge gap on the effects of the June 2021 heatwave. Here, we see a lack of effect at the abundance scale but a clear reduction in size and change in size distribution. This suggests that the effects of the heatwave may have a range of effects on species and a single measure may not capture the full effect or lack thereof.

# Appendix D: Port Gamble S'Klallam Tribe

### 1. Data overview

The Port Gamble S'Klallam Tribe (PGST) measured abundance and size of *Ruditapes philippinarum* (Manila clam) and *Leukoma staminea* (Littleneck clam) at Guillemot Cove – Clam (GC – Clam, site P) and abundance of *Crassostrea gigas* (Pacific oyster) at Guillemot Cove – Oyster (GC – Oyster, site Q). The number of sample quadrats and individuals measured were consistent between surveys. Of note, after surveys occurred close to one year after the heatwave.

Table 1: Overview of data from the Port Gamble S'Klallam Tribe. Total survey area, size of samples, number of samples, and number of individual lengths are displayed for each species and site as the value before/after the heatwave.

		Survey	Sample	Ν	N indv.		
Species	Site	area (m <sup>2</sup> )	area (m <sup>2</sup> )	samples	lengths	Before date	After date
C. gigas	GC – Oyster	9827/16,663	0.19	85/103	-	30 Apr 21	20 Apr 22
L. staminea	GC – Clam	8782	0.19	61/52	67/69	22 June 21	16 May 22
R. philippinarum	GC - Clam	8782	0.19	61/52	2375/2139	22 June 21	16 May 22

### 2. Results



2.1. Abundance

Figure 1: Population estimate of Crassostrea and Ostrea at sites sampled by Port Gamble S'Klallam Tribe before and after the June 2021 heatwave.

Species	Site	Percent difference	Effect size - Cohen's D	MWU p-value
-			(95% CI)	-
C. gigas	GC – Oyster	118.7	-0.65 (-0.940.35)	0.089
L. staminea	GC – Clam	64.2	-0.21 (-0.59 – 0.16)	0.921
R. philippinarum	GC – Clam	5.8	-0.08 (-0.45 - 0.30)	0.645

Table 2: Results of the difference in population size of Ruditapes, Leukoma, and Crassostrea at Port Gamble S'Klallam Tribe sites from before and after the June 2021 heatwave.

### 2.2. Size



*Figure 2: Size-frequency histograms (A) and box plots (B) of Leukoma and Ruditapes lengths measured at Port Gamble S'Klallam site GC-Clam before and after the June 2021 heatwave. Vertical bars in box plots represent median.* 

Table 3: Results of the difference in Leukoma and Ruditapes length at Port Gamble S'Klallam site GC-Clam from before and after the June 2021 heatwave.

		Percent	Effect size - Cohen's		
Species	Site	difference	D (95% CI)	MWU p-value	KS test p-value
L. staminea	GC-Clam	-4.2	0.27 (-0.31 – 0.87)	0.378	0.816
R. philippinarum	GC-Clam	-5.8	0.37 (0.31 – 0.42)	< 0.001	< 0.001

### 2.3. Biomass



Figure 3: Estimated biomass of Leukoma and Ruditapes at Port Gamble S'Klallam site GC-Clam from before and after the June 2021 heatwave.

Table 4: Results of the difference of estimated biomass of Leukoma and Ruditapes at Port Gamble S'Klallam site GC-Clam from before and after the June 2021 heatwave.

			Effect size - Cohen's D	
Species	Site	Percent difference	(95% CI)	MWU p-value
L. staminea	GC – Clam	88.1	-0.25 (-0.62 – 0.13)	0.962
R. philippinarum	GC – Clam	13.0	-0.14 (-0.52 – 0.23)	0.551

### 3. Discussion

There is little evidence that the estimated population size, of *L. staminea*, *R. philippinarum*, and *C. gigas* and biomass of *R. philippinarum* and *L. staminea* was affected by the June 2021 heatwave form a statistical perspective. However, there is evidence for an effect of heatwave on *R. philippinarum* size. Mean size of *R. philippinarum* decreased following the heatwave (p < 0.001) and experienced and overall change in size distribution (p < 0.001; Figure 2; Table 3). It should be noted that size data are treated as means computed from all measured individuals, leading to a considerably large sample size (Table 1). This leads to a small error estimates, and therefore statistically significant differences. Whether an approximate 3mm reduction in mean size is biologically significant is up for debate.

In contrast to many other sites in this report, *R. philippinarum*, *L. staminea*, and *C. gigas* all increased mean abundance and biomass after the heatwave. This may suggest that the increased temperature increased metabolism causing increased growth. This assumes that there were sufficient food resources during the period of metabolic increase. However, what this

explanation does not account for the large increase in observed individuals, especially *L. staminea* and *C. gigas*. Alternatively, the variability of the data indicates a large range of plausible abundances and biomass estimates, leading to insignificant p-values, and general uncertainty of a heatwave effect.

# Appendix E: Puget Sound Restoration Fund

### 1. Data overview

Puget Sound Restoration Fund (PSRF) sampled *Crassostrea gigas* (Pacific oyster) and *Ostrea lurida* (Olympia oyster) at Port Gamble Bay (PGB – site L). They measured abundance of both species and length of *Ostrea*. While surveys were conducted in the same general area and methods, PSRF noted that 22 total records that were collected during the before survey were not collected in the after survey. This leads to lower sample size in the sample area.

Table 1: Overview of data from Puget Sound Restoration Fund. Total survey area, size of samples, number of samples, and number of individual lengths are displayed for each species and site as the value before/after the heatwave.

		Survey	Sample	Ν	N indv.		
Species	Site	area (m <sup>2</sup> )	area (m <sup>2</sup> )	samples	lengths	Before date	After date
C. gigas	PGB	4590	0.25	45/20	-	27-28 May 21	20 Aug 21
O. lurida	PGB	4590	0.25	45/20	72/12	27-28 May 21	20 Aug 21

### 2. Results

2.1. Abundance



Figure 1: Population estimate of native and Pacific oysters in Port Gamble Bay before and after the June 2021 heatwave. Data are displayed in thousands and with 95% confidence intervals.

Table 2: Comparison of native and Pacific oyster abundance at Port Gamble Bay between surveys conducted before and after the June 2021 heatwave.

		Effect size - Cohen's D	
Species	Percent difference	(95% CI)	MWU p- value
C. gigas	-32.4%	0.21 (-0.33 – 0.75)	0.659
O. lurida	-85.0%	0.76 (-0.20 - 1.31)	0.002





Figure 2: Size frequency and distribution of native oysters at Port Gamble Bay before and after the June 2021 heatwave. Solid lines in boxplots represent median size.

		Effect size - Cohen's		
Species	Percent difference	D (95% CI)	MWU p-value	KS test p-value
O. lurida	18.3 %	-0.56 (-1.18 – 0.07)	0.092	0.292

#### 3. Discussion

The June 2021 heatwave appears to have effected *O. lurida* in both abundance and size, while *C. gigas* showed little change in abundance and size. There is strong evidence of a decline in *O. lurida* abundance (d = 0.78, p < 0.001). *O. lurida* experienced an 85% decline in abundance from 24,480 (14,979 – 33,980 95% CI) before the heatwave to 7,534 (370 – 6,974 95% CI) after the heatwave. There is also strong evidence of an increase in mean size of *O. lurida* after the heatwave (p = 0.009). Mean size increased from 31.2mm (27.7mm – 33.7mm 95% CI) before the heatwave to 36.9 (33.8mm – 40.0mm 95% CI) after the heatwave. Size-frequency data indicates that the heatwave resulted in a loss of small individuals (< 25 mm), leading to the increase in mean size. However, this change in size-frequency distribution was not significant (p = 0.292).

However, there is somewhat limited sample size to preform this test. In contrast, *C. gigas* appears to have experienced little effect from the heatwave. However, it should be noted that the population experience an estimated 32.4% decline in abundance. This difference is likely not statistically significant due to the large degree of estimated error.

Available data suggest that there are species dependent heatwave effects on population abundance at Port Gamble Bay. While size data is not available for *C. gigas*, the species is often larger than *O. lurida* which may have provided resilience to extreme temperatures. The sizefrequency patterns of *O. lurida* suggest that smaller individuals may be more susceptible to thermal stress at this location. Therefore, the presumed larger *C. gigas* may have been at an advantage compared to *O. lurida*. Both species also display a large degree of variability in abundance. This feature may make detection of short-term disturbances difficult to detect. Longterm monitoring of these populations may provide a more complete picture of the effects of the heatwave on these species' abundance and size distribution.

# Appendix F: Skagit County MRC

### 1. Data overview

The Skagit County Marine Resources Committee (MRC) measured abundance of *Ostrea lurida* (Olympia oyster) at Fidalgo Bay Causeway (FCB – site B) and Shell RV Park – North (SRV-N – site A) before and at the June 2021 heatwave. Before surveys were conducted in 2018, nearly three years before the heatwave. Sample sizes vary between sites but are the same within sites between surveys.

Table 1: Overview of data from Skagit County MRC. Total survey area, size of samples, number of samples, and number of individual lengths are displayed for each species and site as the value before/after the heatwave.

Species	Site	Survey area $(m^2)$	Sample	N samples	N indv.	Before date	After date
species	bite	(111)	area (III )	samples	ienguis	Delore dute	The dute
O lumida	FCB	1515	0.1	31	-	15-June-18	8-July-21
O. iuriaa	SRV-N	2080	0.1	10	-	12-June-18	10-July-21

### 2. Results





Figure 1: Population estimate of O. lurida by Skagit MRC at FCB and SRV-N before and after the June 2021 heatwave.

Table 2: Results of the difference in population size of O. lurida sampled by Skagit MRC at FCB and SRV-N before and after the June 2021 heatwave.

			Effect size - Cohen's D	
Species	Site	Percent difference	(95% CI)	MWU p-value
O lumi da	FCB	11.1	-0.08 (-0.59 - 0.43)	0.658
<i>0. iuriaa</i>	SRV-N	-1.5	0.03 (-0.91 – 0.97)	0.520

### 3. Discussion

There is no evidence that the June 2021 heatwave had an effect on the abundance of *O. lurida* at Skagit County MRC sites FCB and SRV-N. While there were measurable differences in abundance, these changes were not statistically significant (Figure 1; Table 2). At FCB, the estimated 11.1% increase in abundance is contrary to predicted heatwave effects and suggests that the *O. lurida* population is growing at the site.

These results suggest sites of resilience to extreme heat events for *O. lurida* and may provide clues to site level features that contribute to this resilience. These sites are located in Northern Puget Sound which generally experienced lower air temperatures, and importantly low tide earlier in the day than South Puget Sound sites.

# Appendix G: Swinomish Indian Tribal Community

### 1. Data overview

The Swinomish Indian Tribal Community (SITC) measured abundance, length, and biomass, of a suite of bivalve species at Lone Tree (LT – site D), Kiket (KI – site C), and Twin Lagoons (TW – site E). While multiple *Macoma* species were identified, we pooled to genus *Macoma* spp. Furthermore, *Mya arenaria* (Softshell clam) were excluded from statistical testing due to low sample size and *Tresus capax* (Horse clam) (TW) and *Ruditapes philippinarum* (Manila clam) (TW) were removed from size analysis due to low sample size.

Table 1: Overview of data by Swinomish Indian Tribal Community. Total survey area, size of samples, number of samples, and
number of individual lengths are displayed for each species and site as the value before/after the heatwave.

			Sample		N indv.		
Species	Site	Survey area (m <sup>2</sup> )	area (m <sup>2</sup> )	N samples	lengths	Before date	After date
C muttallii	LT	70925.8/74042.5	0.09	197/209	87/40	17-June-19	21-July-21
C. nutiatiti	TW	22246.8	0.09	67/69	10/7	27-May-21	20-July-21
C. californica	LT	70925.8/74042.5	0.09	197/209	-	17-June-19	21-July-21
I ataminaa	LT	52871.4	0.09	197/209	66/62	17-June-19	21-July-21
L. staminea	TW	22246.8	0.09	67/69	49/59	27-May-21	20-July-21
Macomason	LT	70925.8/74042.5	0.09	197/209	-	17-June-19	21-July-21
<i>Macoma</i> spp.	TW	22246.8	0.09	67/69	-	27-May-21	20-July-21
M. anonania	LT	70925.8/74042.5	0.09	197/209	53/50	17-June-19	21-July-21
M. arenaria	TW	22246.8	0.09	67/69	3/0	27-May-21	20-July-21
O lumida	KI	222.8	0.06	14/14	-	25-May-21	17-July-21
O. iuriaa	LT	138.0	0.06	40/22	-	24-May-21	26-July-21
D. mhilimmin amun	LT	70925.8/74042.5	0.09	197/209	53/41	17-June-19	21-July-21
K. philippinarum	TW	22246.8	0.09	67/69	3/14	27-May-21	20-July-21
S. gigantag	LT	70925.8/74042.5	0.09	197/209	415/318	17-June-19	21-July-21
S. giganiea	TW	22246.8	0.09	67/69	97/151	27-May-21	20-July-21
T agnar	LT	70925.8/74042.5	0.09	197/209	50/34	17-June-19	21-July-21
т. сарах	TW	22246.8	0.09	67/69	2/2	27-May-21	20-July-21

### 2. Results

### 2.1. Abundance



Figure 1: Population estimate of bivalve species at LT, TW, and KI sites sampled by Swinomish Indian Tribal Community before and after the June 2021 heatwave.

Table 2: Results of the difference in population size of bivalve species at LT, TW, and KI sites sampled by Swinomish India	an
Tribal Community before and after the June 2021 heatwave.	

		Effect size - Cohen's D			
Species	Site	Percent difference	(95% CI)	MWU p-value	
C muttallii	LT	-55.3	0.31 (0.11 - 0.50)	0.028	
C. nunann	TW	-32.0	0.09 (-0.25 - 0.43)	0.931	
C. californica	LT	-88.4	0.17 (-0.03 – 0.37)	0.027	
I staminog	LT	-7.5	0.03 (-0.16 – 0.23)	0.584	
L. staminea	TW	24	-0.09 (-0.43 – 0.25)	0.643	
Macomacon	LT	-4.5	0.01 (-0.18 – 0.21)	0.043	
<i>Macoma</i> spp.	TW	4.7	-0.02 (-0.36 - 0.32)	0.188	
0 lunida	KI	-30.8	0.25 (-0.53 - 1.03)	0.867	
0. iuriaa	LT	78.6	-0.51 (-1.05 - 0.03)	0.036	
B. shilippin anum	LT	23.9	0.07 (-0.13 – 0.26)	0.198	
к. ратрриатит	TW	353.1	-0.26 (-0.60 - 0.08)	0.251	
C oigantag	LT	-24.1	0.14 (-0.05 - 0.34)	0.404	
5. giganiea	TW	52.2	-0.17 (-0.51 – 0.17)	0.264	
Tagnar	LT	-34.4	0.11 (-0.09 - 0.30)	0.361	
1. сарах	TW	-2.9	0.01 (-0.33 – 0.34)	0.982	





Figure 2: Size-frequency histograms (A) and box plots (B) of bivalve lengths measured at site LT by Swinomish Indian Tribal Community before and after the June 2021 heatwave. Vertical bars in box plots represent median.



Figure 3: Size-frequency histograms (A) and box plots (B) of bivalve lengths measured at site TW by Swinomish Indian Tribal Community before and after the June 2021 heatwave. Vertical bars in box plots represent median.

Table 3: Results of the difference in size and size distribution of bivalve species at LT and TW sites sampled by Swinomish Indian Tribal Community before and after the June 2021 heatwave.

		Percent	Effect size - Cohen's D	MWU p-	KS test p-
Species	Site	difference	(95% CI)	value	value
C muttallii	LT	-1.3	0.03 (-0.34 – 0.41)	0.493	0.163
<i>C. nullalli</i>	TW	-12.3	0.42 (-0.63 – 1.45)	0.591	0.922
T / ·	LT	-5.8	0.26 (-0.10 – 0.61)	0.295	0.666
L. siuminea	TW	-0.4	0.02 (-0.37 – 0.41)	0.915	0.415
R. philippinarum	LT	-3.3	0.15 (-0.26 - 0.56)	0.439	0.310
<u> </u>	LT	-0.1	0.00 (-0.14 - 0.15)	0.986	0.942
S. giganiea	TW	-2.6	0.12 (-0.14 – 0.37)	0.207	0.082
T. capax	LT	4.4	-0.23 (-0.67 – 0.22)	0.122	0.181

#### 2.3. Biomass



Figure 4: Biomass estimate of bivalve species at LT, TW, and KI sites sampled by Swinomish Indian Tribal Community before and after the June 2021 heatwave.

		Percent	Effect size - Cohen's D	
Species	Site	difference	(95% CI)	MWU p-value
C muttallii	LT	-61.0	0.29 (0.09 - 0.48)	0.015
C. nutiatiti	TW	-48.5	0.15 (-0.19 – 0.49)	0.919
I staminog	LT	-19.2	0.08 (-0.12 - 0.27)	0.361
L. staminea	TW	16.6	-0.06 (-0.40 – 0.28)	0.611
M. arenaria	LT	-6.8	0.02 (-0.17 – 0.21)	0.879
D. mhilimnin amun	LT	-15.9	0.04 (-0.16 - 0.24)	0.285
<b>K</b> . philippinarum	TW	1478.2	-0.27 (-0.61 – 0.07)	0.102
C gigantag	LT	-23.5	0.13 (-0.06 - 0.33)	0.229
S. giganiea	TW	31.1	-0.12 (-0.46 – 0.22)	0.342
T canar	LT	-19.6	0.05 (-0.14 - 0.25)	0.325
T. capax	TW	-53.6	0.10 (-0.24 - 0.44)	0.910

Table 4: Results of the difference biomass of bivalve species at LT and TW sites sampled by Swinomish Indian Tribal Community before and after the June 2021 heatwave.

#### 3. Discussion

There is little evidence that the heatwave had an effect on the abundance, size, and biomass of most species across all sites from a statistical perspective. However, there are a few exceptions. There is strong evidence of a reduction in abundance and biomass of *C. nuttallii* (cockles) at LT, and reduction of abundance of *C. cryptomya* and *Macoma* spp. at LT. Notably, there was no evidence for a difference in mean size or size distribution before and after the heatwave for any species and any site.

As with many of the datasets analyzed in this project, there is a high degree of variability in the data. This makes identification of statistical differences challenging. However, it should be noted that many species experienced reductions in mean abundance, size, and/or biomass of greater than 20 %. While not found to be statistically significant, it is conceivable that this is biologically significant. At a minimum these data suggest that many bivalve species experienced a disturbance with negative consequences. However, the exact effect of the disturbance may be hard to elucidate with available data.

# Appendix H: Squaxin Island Tribe

### 1. Data overview

The Squaxin Island Tribe (SQXN) measured abundance and length of *Ruditapes philippinarum* (Manila clam) at Senior Beach (SNB, site U), PTL- Bergh (PTL-B, site X), PTL- Krishnamorti (PTL-K, site Y), PTL- Morrison (PTL-M, site W) and PTL- Orser (PTL-O, site T) and abundance of *Crassostrea gigas* (Pacific oyster) at Pickering Passage on Harstine Island (PPHI, site V). *C. gigas* was only surveyed after the heatwave. The number of quadrats sampled were relatively consistent within in a sampling location between before and after heatwave surveys. Sample area was also consistent except for PTL-O, where clam harvest occurred between the before and after survey. <u>Samples from the area of harvest were removed from this analysis.</u> Since *C. gigas* was only sampled after the heatwave, results from those data are not displayed here. Note change that some sample dates are separated by more approximately a year.

Table 1: Overview of data from the Squaxin Island Tribe. Total survey area, size of samples, number of samples, and number of individual lengths are displayed for each species and site as the value before/after the heatwave.

		Survey area	Sample area	Ν	N indv.		
Species	Site	(m <sup>2</sup> )	(m <sup>2</sup> )	samples	lengths	Before date	After date
	PTL-B	627	0.09	21/28	518/630	24-June-21	19-July-21
	PTL-K	1942	0.09	41/43	620/288	31-Mar-21	07- July -21
R. philippinarum	PTL-M	312.6	0.09	21/30	383/373	26-June-20	13- July -21
	PTL-O	1124/942	0.09	32/31	760/725	08-Apr-20	27- July -21
	SNB	6809.6	0.09	32/31	639/416	07-June-21	08- July -21
C. gigas	PPHI	-	0.37	-/20	-	-	08- July -21

### 2. Results

2.1. Abundance



Figure 1: Population estimate of R. philippinarum at sites sampled by Squaxin Island Tribe before and after the June 2021 heatwave.

			Effect size - Cohen's D	
Species	Site	Percent difference	(95% CI)	MWU p-value
	PTL-B	-10.8	0.14 (-0.43 – 0.73)	0.879
	PTL-K	-55.7	0.60 (0.16 - 1.04)	0.093
R. philippinarum	PTL-M	-31.8	0.41 (-0.17 – 0.98)	0.150
	PTL-O	-17.5	0.25 (-0.26 – 0.76)	0.390
	SNB	-18.8	0.22 (-0.28 – 0.73)	0.536

Table 2: Results of the difference in population size of R. philippinarum at Squaxin Island Tribe sites from before and after the June 2021 heatwave.

### 2.2. Size



Figure 2: Size-frequency histograms (A) and box plots (B) of R. philippinarum lengths measure before and after the June 2021 heatwave.

Table 3: Results of the difference in R. philippinarum length at Squaxin Island Tribe sites from before and after the June 2021 heatwave.

Species	Site	Percent difference	Effect size - Cohen's D (95% CI)	MWU p-value	KS test p-value
	PTL-B	0.6	-0.04 (-0.15 - 0.08)	0.638	0.574
	PTL-K	-4.1	0.24 (0.10 - 0.38)	0.048	0.009
R. philippinarum	PTL-M	0.9	-0.07 (-0.21 – 0.08)	0.331	0.227
	PTL-O	4.3	-0.26 (-0.370.16)	< 0.001	< 0.001
	SNB	-1.8	0.17(0.02 - 0.31)	0.016	0.043

### 2.3. Biomass



Figure 3: Biomass estimates R. philippinarum at sites sampled by Squaxin Island Tribe before and after the June 2021 heatwave. Table 4: Results of the difference in R. philippinarum biomass at Squaxin Island Tribe sites from before and after the June 2021 heatwave.

			Effect size - Cohen's D	
Species	Site	Percent difference	(95% CI)	MWU p-value
	PTL-B	-14.5	0.20 (-0.40 - 0.78)	0.692
	PTL-K	-55.3	0.63 (0.17 - 1.08)	0.133
R. philippinarum	PTL-M	-26.6	0.31 (-0.26 – 0.89)	0.247
* **	PTL-O	-4.7	0.06(-0.46-0.58)	0.909
	SNB	-21.6	0.27 (-0.23 – 0.78)	0.437

### 3. Discussion

There is little evidence that the estimated population size, and biomass of *R. philippinarum* was affected by the June 2021 heatwave form a statistical perspective. However, there is evidence for an effect of heatwave on *R. philippinarum* size. The exceptions are an apparent reduction in size at PTL-K and SNB (p = 0.048 p < 0.016) and an increase in size at PTL-O (p < 0.001). These represent -4.1%, -2.6%, and 4.3% differences respectively (Figure 2; Table 3). There is also evidence that the distribution of sizes changed after the heatwave at PLT-K (p = 0.009), PTL-O (p < 0.001) and SNB (p = 0.002). It is notable that at population estimate and biomass declined by approximately 50% at PTL-K but that this is not considered a statistically significant difference. This is almost certainly due to the large degree of variability in these data. Alternatively, examining the effect size (Cohen's D) suggests effects of the heatwave that may be more in line with one's intuitive sense. In general, effect sizes near 0.5 are considered to be moderate. Using this criteria, both PTL-K and PTL-M appear to have reduced population size and biomass as an effect of the heatwave.

These data offer an excellent opportunity to compare the effects of the heatwave on one species across different locations. Results suggest that PTL-K experienced a greater negative effect from the heatwave than other sites. Further analysis could formally test for an effect of site and heatwave and the interaction between the two. As with many of the dataset examined for this project there is considerable variability in the data which may make statistical detection of heatwave effects impossible.

# Appendix I: WDFW - Clam

### 1. Data overview

The Washington Department of Fish and Wildlife (WDFW) measured abundance and length of multiple clam species at three sites: North Bay (NB – site R), Twanoh State Park (TSP – site S) and Wolfe State Park (WPSP – site J). The number of quadrat samples (N samples) in which counts were collected were fairly consistent between before and after heatwave surveys, but the number of individuals measured (N indv. lengths) varied greatly (Table 1). *Saxidomus gigantea* (Butter clam), *Tresus capax* (Horse clam), and *Macoma* spp. were sampled for but many zeros were excluded from abundance and biomass analyses. Counts were only collected on legal size (=> 38 mm) *Leukoma staminea* (Littleneck clam) and *Ruditapes philippinarum* (Manila clam) so population and biomass estimates reported for these two species only represent legal size clams. Size measurements were collected on all subsampled individuals. Species where the number of individuals measured was low (< 30) in both the before and after surveys were excluded from the size analysis. WDFW also surveyed oysters (*Crassostrea gigas* and *Ostrea lurida*) at these and other sites; these data are presented in a separate document.

		Survey	Sample area	Ν	N indv.		
Species	Site	area (m <sup>2</sup> )	(m <sup>2</sup> )	samples	lengths	Before date	After date
	NB	17,678		92/97	-	27-Apr-21	10-Aug-21
C. nuttallii	TSP	33,541	0.093	121/110	1/-	28-May-21	20-Aug-21
	WPSP	31,565		143/133	-	14-June-21	8-Aug-21
	NB	17,678		92/97	56/46	27-Apr-21	10-Aug-21
L. staminea	TSP	33,541	0.093	121/110	1/-	28-May-21	20-Aug-21
	WPSP	31,565		143/133	13/11	14-June-21	8-Aug-21
	NB	17,678		92/97	21/9	27-Apr-21	10-Aug-21
M. arenaria	TSP	33,541	0.093	121/110	4/3	28-May-21	20-Aug-21
	WPSP	31,565		143/133	8/12	14-June-21	8-Aug-21
	NB	17,678	0.093	92/97	-/5	27-Apr-21	10-Aug-21
N. obscurata	TSP	33,541		121/110	392/191	28-May-21	20-Aug-21
	WPSP	31,565		143/133	162/125	14-June-21	8-Aug-21
	NB	17,678		92/97	898/602	27-Apr-21	10-Aug-21
R. philippinarum	TSP	33,541	0.093	121/110	559/452	28-May-21	20-Aug-21
	WPSP	31,565		143/133	280/273	14-June-21	8-Aug-21
	NB	17,678		92/97	-	27-Apr-21	10-Aug-21
S. gigantea	TSP	33,541	0.093	121/110	-	28-May-21	20-Aug-21
	WPSP	31,565		143/133	27/13	14-June-21	8-Aug-21
	NB	17,678		92/97	-	27-Apr-21	10-Aug-21
T. capax	TSP	33,541	0.093	121/110	-	28-May-21	20-Aug-21
	WPSP	31,565		143/133	-	14-June-21	8-Aug-21

Table 1: Overview of data by WDFW. Total survey area, size of samples, number of samples, and number of individual lengths are displayed for each species and site as the value before/after the heatwave.

### 2. Results

### 2.1. Abundance



Figure 1: Population estimate of bivalve species at NB, TSP, and WPSP sites sampled by WDFW before and after the June 2021 heatwave.

Table 2 : Results of the difference in population size of bivalve species at NB, TSP, and WPSP sites sampled by WDFW before and after the June 2021 heatwave.

			Effect size - Cohen's D	
Species	Site	Percent difference	(95% CI)	MWU p-value
C. nuttallii	TSP	10.0	0.00 (-0.27 – 0.25)	0.951
I at amin a a	NB	55.2	-0.19 (-0.47 – 0.10)	0.750
L. siaminea	WPSP	15.2	-0.05 (-0.28 - 0.19)	0.448
	NB	3.2	-0.01 (-0.30 – 0.27)	0.736
M. arenaria	TSP	-32.3	0.08 (-0.17 – 0.35)	0.550
	WPSP	46.6	-0.13 ( -0.37 – 0.10)	0.329
N abaaunata	TSP	-8.1	0.04 (-0.22 - 0.30)	0.480
N. ODSCUrata	WPSP	6.3	-0.03 (-0.27 - 0.20)	0.781
	NB	-8.8	0.07 (-0.22 – 0.36)	0.570
R. philippinarum	TSP	0.5	0.00 (-0.26 - 0.25)	0.466
	WPSP	48.7	-0.20 (-0.44 - 0.04)	0.057



Figure 2: Size-frequency histograms (A) and box plots (B) of bivalve lengths measured at site NB by WDFW before and after the June 2021 heatwave. Vertical bars in box plots represent median.



Figure 3: Size-frequency histograms (A) and box plots (B) of bivalve lengths measured at site TSP by WDFW before and after the June 2021 heatwave. Vertical bars in box plots represent median.



Figure 4: Size-frequency histograms (A) and box plots (B) of bivalve lengths measured at site WPSP by WDFW before and after the June 2021 heatwave. Vertical bars in box plots represent median.

Table 3: Results of the difference in size of	and size distribution of bivalve species at NB, TSP, and WPSP sites sampled by WI	DFW
before and after the June 2021 heatwave.	<i>Tests of N. obscutata were not preformed at NB due to low sample size.</i>	

		Percent	Effect size - Cohen's D	MWU p-	KS test p-
Species	Site	difference	(95% CI)	value	value
N. obscurata	TSP	2.8	-0.17 (-0.34 - 0.00)	0.028	0.011
	WPSP	0.8	-0.04 (-0.28 – 0.19)	0.717	0.526
	NB	2.2	-0.13 (-0.230.03)	0.026	0.015
R. philippinarum	TSP	1.2	-0.07 (-0.19 – 0.06)	0.214	0.454
	WPSP	-3.3	0.19 (0.02 - 0.36)	0.109	0.388

### 2.3. Biomass



Figure 5: Biomass estimate of bivalve species NB, TSP, and WPSP sites sampled by WDFW before and after the June 2021 heatwave. Biomass estimates were not preformed at NB for N. obscurata due to low sample size.

Table 4: Results of the difference biomass of bivalve species at NB	TSP, and WPSP sites sampled by WDFW before and after the
June 2021 heatwave.	

		Percent	Effect size - Cohen's D	
Species	Site	difference	(95% CI)	MWU p-value
I atamin aa	NB	19.9	-0.27 (-1.31 – 0.77)	0.647
L. staminea	WPSP	-24.2	0.21 (-0.69 - 1.10)	0.614
	NB	-26.9	0.32 (-0.77 – 1.41)	0.515
M. arenaria	TSP	44.6	-0.15 (0.96 - 0.67)	0.708
	WPSP	18.4	-0.14 (-1.09 - 0.80)	0.754
N abaaunata	TSP	-5.3	0.03 (-0.43 – 0.49)	0.879
IN. ODSCUPAIA	WPSP	-8.8	0.08 (-0.46 - 0.61)	0.772
	NB	11.1	-0.11 (-0.63 – 0.41)	0.696
R. philippinarum	TSP	27.0	-0.17 (-0.60 – 0.26)	0.447
	WPSP	-22.0	0.18(-0.29-0.65)	0.460

#### 3. Discussion

There is little evidence from a statistical perspective that the heatwave had an effect on the abundance, size, and biomass for most species sampled across the three surveyed sites. However, there are a few exceptions in which an effect is supported. There is good evidence of an increase in mean size after the June 2021 heatwave for *N. obscurata* at TSP and *R. philippinarum* at NB of 2.8% and 2.2%, respectively, (Table 4; Fig 4). There is also good evidence of a change in

overall size distribution of these two species at these sites (Table 4; Fig 4). Although, while significant, the effect size is notably small (d < 0.2). In some cases, large differences in mean abundance and/or biomass were observed, but not found to be statistically significant. For example, *L. staminea* at NB featured at 55% increase in biomass but Mann-Whitney U test found this to be non-significant (p = 0.75). This is likely due to the high degree of variability within and between surveys for a given species at a given site which has been seen in many other contributors' data.

As with many of the datasets analyzed in this project, there is a high degree of variability in the data. This makes identification of statistical differences challenging. However, it should be noted that three species experienced reductions in biomass of greater than 20%. While note found to be statistically significant, it is conceivable that this is biologically significant. At a minimum these data suggest that many bivalve species experienced a disturbance with negative consequences. However, the exact effect of the disturbance may be hard to elucidate with available data.

# Appendix J: WDFW - Oyster

### 1. Data overview

The Washington Department of Fish and Wildlife (WDFW) measured abundance and length of osysters *Crassostera gigas* (Pacific oyster) and *Ostrea lurida* (Olympia oyster) at Dosewallpis State Park (DSP – site N), Kitsap Memorial State Park (KMSP – site M), Triton Cove (TC – site O), Twanoh State Park (TSP – site S), Wolfe State Park (WPSP – site J) and Wolfe State Park-Enhanced (WPSP-E – site K). The number of quadrats samples was fairly consistent between surveys, but the number of individuals measured varied greatly. Note that *O. lurida* lengths were not taken at any site, and only after heatwave data is available for DSP.

Table 1: Overview of data from Washington Department of Fish and Wildlife. Total survey area, size of samples, number of samples, and number of individual lengths are displayed for each species and site as the value before/after the heatwave.

		Survey area	Sample	Ν	N indv.		
Species	Site	$(m^2)$	area (m <sup>2</sup> )	samples	lengths	Before date	After date
C. gigas	DSP	72,003	0.19	-/79	-/382	-	8-July-21
	KMSP	5,483/5,546	0.19	37/39	140/123	9-June-20	6-Aug-21
	TC		0.19	67/65	252/195	17-Aug-20	9-July-21
	TSP	16,987	0.19	58/70	265/584	1-Aug-20	9-July-21
	WPSP	3,992/3,903	0.19	49/61	118/122	8-July-20	19-Aug-21
	WPSP-E	5,785/6,314	0.19	54/58	299/248	8-Aug-20	19-Aug-21
O. lurida	DSP	72,003	0.19	-/79	-	-	8-July-21
	KMSP	5,483/5,546	0.19	37/39	-	9-June-20	6-Aug-21
	TC	13,446/12,808	0.19	67/55	-	17-Aug-20	9-July-21
	WPSP		0.19	49/61	-	8-Jul-20	19-Aug-21
	WPSP-E	5,785/6,314	0.19	54/58	-	8-Jul-20	19-Aug-21

2. Results

2.1. Abundance



Figure 1: Population estimate of oysters at KMSP, TC, TSP, WPSP, and WPSP-E sites sampled by WDFW before and after the June 2021 heatwave.

		Percent	Effect size - Cohen's D	MWU p-
Species	Site	difference	(95% CI)	value
	KMSP	478.2	-0.83 (-1.310.35)	0.002
	TC	160.2	-0.46 (-0.810.11)	0.463
C. gigas	TSP	-2.5	0.01 (-0.34 – 0.36)	< 0.001
	WPSP	33.1	-0.18 (-0.56 - 0.20)	0.402
	WPSP-E	80.3	-0.22 (-0.60 - 0.15)	0.760
	KMSP	-36.0	0.12 (-0.34 – 0.57)	0.663
O lumida	TC	-81.8	0.68 (0.31 - 1.05)	< 0.001
0. iuriaa	WPSP	-21.5	0.04 (-0.34 - 0.42)	0.868
	WPSP-E	49.2	0.11 (-0.27 – 0.48)	0.539

Table 2: Results of the difference in population size of oysters at KMSP, TC, TSP, WPSP, and WPSP-E sites sampled by WDFW before and after the June 2021 heatwave.

#### 2.2. Size



Figure 2: Size-frequency histograms (A) and boxplot (B) of Crassostrea gigas lengths measured at sites KMSP, TSP, WPSP, WPSP-E by WDFW before and after the June 2021 heatwave. Vertical bars in box plots represent median.

Table 3: Results of the difference in size and size distribution of Crassostrea gigas at KMSP, TSP, WPSP, and WPSP-E sites sampled by WDFW before and after the June 2021 heatwave.

		Percent	Effect size - Cohen's D	MWU p-	KS test p-
Species	Site	difference	(95% CI)	value	value
	KMSP	-36.8	0.96 (0.71 – 1.22)	< 0.001	< 0.001
Calibra	TSP	-34.8	0.97 (0.81 - 1.12)	< 0.001	< 0.001
C. gigas	WPSP	-38.8	1.59 (1.30 – 1.88)	< 0.001	< 0.001
	WPSP-E	-18.2	0.31 (0.14 - 0.48)	< 0.001	< 0.001

### 3. Discussion

There is little evidence for that the heatwave had an effect on the abundance, of *C. gigas* and *O. lurida* across all sites from a statistical perspective. However, there are a few exceptions. There is strong evidence for increase in *C. gigas* abundance at KMSP and decrease at TSP, and a decrease in abundance of *O. lurida* at TC (Table 2, Fig. 1). In contrast to patterns of abundance, there is strong evidence for change in size distribution and reduction in mean size of *C. gigas* at all sites (Table 3, Fig. 2). Mean size decreased by 18.2% to 38.8% representing a reduction of mean size of 17 – 63 mm. One of the challenges of comparing data from 2020 to 2021 is the fact that natural recruitment events may have occurred. Thus, the decline seen in size at some locations, such as KSMP and TSP, may be due to large recruitment events that occurred in the year between these surveys (Figure 2A). It is also plausible that our results reflect two impacts, one of a natural recruitment event and the other of a mortality event related to the heat dome. As these surveys were not designed to tease apart these confounding factors.

As with many of the datasets analyzed in this project, there is a high degree of variability in the data. This makes identification of statistical differences challenging. However, it should be noted that many species experiences reductions in mean abundance, size, and/or biomass of greater than 20%. While note found to be statistically significant, it is conceivable that this is biologically significant. At a minimum these data suggest that many bivalve species experienced a disturbance with negative consequences. However, the exact effect of the disturbance may be hard to elucidate with available data.

Survey Metadata:									
Contributor:	Jamestown S'Klallam Tribe (JST)								
Data Contact:	Annie Raymond, Liz Tobin								
Site Name (Code):	Sequim_1.5acre								
Site Location:	Sequim Bay (1.5-acre Olympia restoration site)								
Survey Type:	Ovster Population								
Pre-survey Method:	randomized transects and o	quadrats within a pre-set restoration polygon.							
Post-survey Method	randomized transects and o	quadrats within a pre-set restoration polygon.							
Pre Total Survey Area (sq m):	6,146								
Post Total Survey Area (sq m):	6,146								
Sample frame (sqs m):	0.25								
Enhanced Beach:	Yes								
Sample Measurements:	All individuals that were cou	unted were also measured for size.							
Data Tarakina									
Data Inacking:	Raw Data Eilo pathway	Spreadsheet name	Shoot						
Soquim 1 Eacro Count	https://docs.google.com/cr	spreadsheet hame	Besteration Site, Count Data						
Sequim 1 Sacre Size	https://docs.google.com/spr	Before2021_Oly_Subsistence_beach_Survey_Results	size distribution						
bequini_ribusic_oize									
Data Dictionary:									
sheet name	<u>column</u>	column_name_	description	format <u>notes</u>					
Counts	A	Site	code for site name (see Survey Metadata above)	character					
Counts	В	Date	observation date	YYYY-MM-DD					
Counts	С	Sample_ID	unique ID for quadrat/sample	character					
Counts	D	GPS_ID	number used to collect sample GPS point	character					
Counts	E	Sample Lat	sample latitude (degree decimal)	numeric					
Counts	F	Sample Long	sample longitude (degree decimal)	numeric					
Counts	G	Species	species sampled	character					
Counts	н	Total Area	size of total surveyed area - squared meters	numeric					
Counts	I	Sample Area	size of area sampled (e.g., sample frame) in square meters	numeric					
Counts	1	Live Count	number of live oysters observed in sample	numeric					
Counts	к	All Dead Count	number of dead oysters observed in sample (recently dead + old dead)	numeric					
Counts	L	Fresh Dead Count	number of recently dead oysters observed in sample (articulated w/o flesh)	numeric					
Counts	Μ	Data ID	ID to track original data source (see Data Tracking above)	string					
Counts	Ν	Data Concern?	concern with using data point in pre/post analysis: "y" or "n" (see Notes)	character					
Counts	0	Notes	data notes and/or qualitative observations	string					
Circa.	•	Cit -							
Sizes	A	Site	code for site name (see Survey Metadata above)						
Sizes	В	Date	observation date	YYYY-MM-DD					
Sizes	2	Sample_ID	unique ID for quadrat/sample	character					
Sizes	D	Species	target species of assessment: Genus_species	character					
Sizes	E	Total Area	size of total surveyed area - squared meters	numeric					
Sizes	F	Sample Area	size of area sampled (e.g., sample frame) in square meters	numeric					
Sizes	G	Size_mm	individual length in millimeters	numeric					
Sizes	Н	Weight_g	individual weight in grams	numeric					
Sizes	I	Data ID	ID to track original data source (see Data Tracking above)	string					
Sizes	J	Data Concern?	concern with using data point in pre/post analysis: "y" or "n" (see Notes)	character					
Sizes	К	Notes	data notes and/orqualitative observations	string					

#### Count Data

		Sample		Sample	Sample	•	Total	Sample	Live	All Dead	Fresh Dead		Data	
Site	Date	ID	GPS ID	Lat	Long	Species	Area	Area	Count	Count	Count	Data ID	Concern?	Notes
Sequim_1.5acre	2021-05-13	55	NA	NA	NA	Ostrea lurida	6146	0.25	0	0	NA	Sequim_1.5acre_Count	N	
Sequim_1.5acre	2021-05-13	58	NA	NA	NA	Ostrea lurida	6146	0.25	0	0	NA	Sequim_1.5acre_Count	Ŷ	Outside restoration boundary - not included in post survey

#### Size Data

Site	Date	Sample_ID	Species	Total Area	Sample Area	Size_mm	Weight_g	Data ID	Data Concern?	Notes
Sequim_1.5acre	2021-05-13	Sequim_1.5acre_before	Ostrea lurida	6,146	0.25	5	NA	Sequim_1.5acre_Size	N	
Sequim_1.5acre	2021-05-13	Sequim_1.5acre_before	Ostrea lurida	6,146	0.25	11	NA	Sequim_1.5acre_Size	Ŷ	broken shell