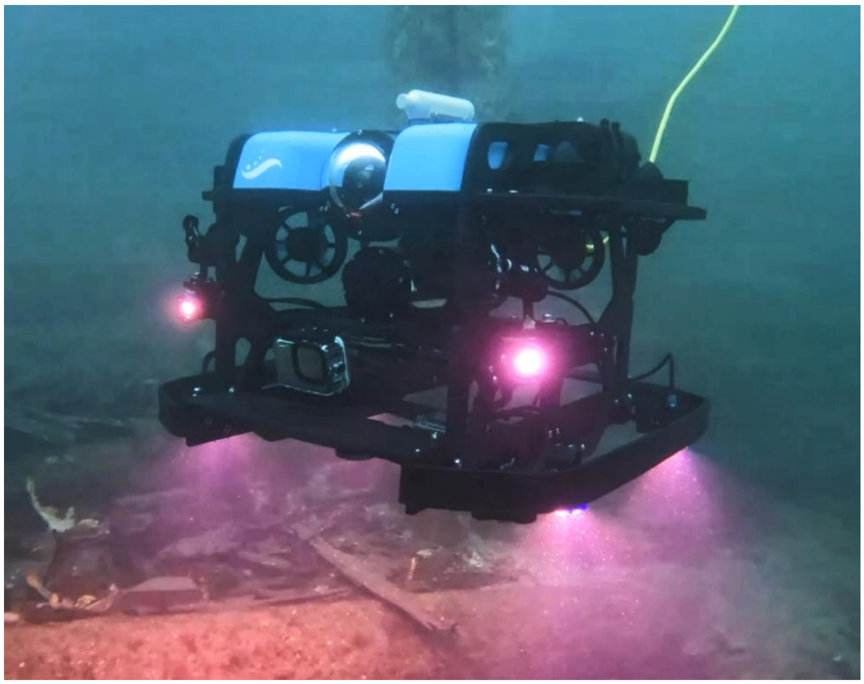
Quality Assurance Project Plan

Remotely operated vehicles (ROVs) and scientific scuba divers: A methods comparison to optimize bull kelp conservation and restoration in Puget Sound



May 2024

SEATTLE AQUARIUM

Publication Information

This project has been funded wholly or in part by the United States Environmental Protection Agency (EPA) under assistance agreement PC-01J89501 to the Washington State Department of Fish and Wildlife (WDFW). The contents of this document do not necessarily reflect the views and policies of the Environmental Protection Agency or the Washington State Department of Fish and Wildlife, nor does mention of trade names or commercial products constitute endorsement or recommendation for use.

The EPA requires an approved Quality Assurance Project Plan (QAPP) for all EPA-funded projects that generate or use environmental information, including modeling efforts, before the projects begin. The plan describes the objectives of the study and the procedures to be followed to achieve those objectives. After completing the study, the author will post the final report of the study to the internet. This QAPP describes a project selected for funding through the Habitat Strategic Initiative Lead (HSIL) Request for Proposals in Fall 2022 and is described in the February 2023 Investment List for strategic investment of Puget Sound Geographic Program funds.

This Quality Assurance Project Plan is available upon request from Zachary Randell, at z.randell@seattleaquarium.org

Data for this project are available in EPA's Water Quality Exchange (WQX) database (https://www.epa.gov/waterdata/water-quality-data-wqx). The QAPP is valid for one year from date of certification. All QAPPs for programs or projects exceeding one year in duration shall be reviewed and recertified annually.

This QAPP was approved to begin work in March 2024. It was finalized and approved in May 2024.

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COVER PHOTO: ROV *Nereo* flying beneath Pier 59 at the Seattle Aquarium. PHOTO BY: Alex Tanz, Seattle Aquarium.

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Remotely operated vehicles (ROVs) and scientific scuba divers: A methods comparison to optimize bull kelp conservation and restoration in Puget Sound.

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May 2024

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2.0 Abstract

Bull kelp (*Nereocystis luetkeana*) provides robust ecological functioning to nearshore taxa such as fishes, invertebrates, sea birds, and marine mammals throughout coastal Washington. However, over the past several decades, bull kelp has experienced substantial declines in Puget Sound, and multiple government, tribal, non-profit, and academic entities are now focused upon bull kelp conservation and restoration. Traditionally, data regarding the abundance and distribution of kelp is gathered either via scientific scuba divers or, in the case of bull kelp which forms a surface canopy, aerial imagery from airplanes or satellites. Remotely operated vehicles (ROVs) have the potential to gather data across a larger seafloor extent than scuba divers can operate, though ROVs have yet to be systematically evaluated for kelp forest survey operations.

The purpose of this project is to evaluate how ROVs can complement ongoing and future bull kelp monitoring and conservation. Our project consists of three pillars: (**1**) we have partnered with Reef Check to conduct a methods comparison between ROVs and scuba divers by flying the ROV above specific locations (transects) surveyed by divers. Our objective is to understand the pros and cons of both platforms. We have also partnered with Puget Sound Restoration Fund (PSRF) to (**2**) monitor their outplanted bull kelp with the ROV to evaluate how best the ROV can contribute to bull kelp restoration. Finally, we will leverage the data collected in this project and our work with the Port of Seattle to (**3**) expand upon an existing bull kelp habitat suitability model via incorporating our ROV-derived data to provide finer-scale predictions for locations suitability for bull kelp restoration. In summary, we envision this project will provide a comprehensive accounting for how best ROVs can complement ongoing and future bull kelp monitoring, conservation, and restoration throughout Puget Sound.

3.0 Background

3.1 Introduction and problem statement

As the predominant habitat-forming species in temperate coastal ecosystems, kelps (brown algae in the Order Laminariales) carry out critical ecological functions that provide food and shelter for other species (Dayton, 1985; Schiel & Foster, 2015), exhibit rapid growth that locally modifies water conditions (Krause-Jensen & Duarte, 2016; Wilmers et al., 2012), and provide ecosystem services essential to human well-being. Bull kelp is of particular interest as the dominant canopy forming species within Puget Sound (Calloway et al., 2020; Mumford, 2007), as its surface canopy provides extensive habitat and shelter for fish, including larval species such as rockfish that are transitioning out of the pelagic phase (Johnson, 2006; White et al., 2019). Numerous other understory kelp species also form three-dimensional habitat, though as their growth does not reach the sea surface and thus cannot be imaged from kayak or aerial platforms, much less is known about their abundance and distribution. In addition to ecological functioning, kelp forests in Washington have deep significance to indigenous and other local communities, and numerous efforts are sprouting up centered around kelp aquaculture.

Puget Sound has experienced marked bull kelp declines in recent decades, with an overall 65% loss of surface canopy since 1980, with some areas exhibiting a 96% decline (Berry et al., 2019, 2021). These declines have been most pronounced throughout south and central Puget Sound. The cause of these declines is not fully understood, though it is likely due to a combination of factors including climate-change related increases in water temperatures, as well as human related changes to sedimentation and nutrient regimes, especially related to coastal development (Khangaonkar et al., 2018). While herbivory, e.g., via sea urchin overgrazing (Filbee-Dexter & Scheibling, 2014) has been a driver of kelp loss in other temperate regions, it does not appear to be a proximate driver throughout Puget Sound.

In response to the observed declines of bull kelp canopy coverage throughout Puget Sound, state and federal agencies as well as tribal, non-profit, and academic entities marshalled to advance working groups, funding priorities, and state legislation. In 2020 the Northwest Straits Commission (NSC) and partners published the Puget Sound Kelp Conservation and Recovery Plan—the Kelp Plan, (Calloway et al., 2020)—linked here. With the Kelp Plan framework in place, January 2022 Washington Dept. of Natural Resources (DNR) developed their kelp canopy indicator (linked here) to prioritize funding for Puget Sound Partnership's Vital Signs Program (linked here). Additionally, the state has advanced numerous organizational efforts such as an accounting of all known kelp conservation and workgroup efforts (linked here), and the NSC has released kelp policy recommendations (linked here). These efforts also resulted in the signing of Senate Bill 5619 which set a goal to conserve and restore 10,000 acres of kelp and seagrass by 2040 (linked here).

Before these ambitious conservation and restoration objectives can be achieved, substantial knowledge gaps must be filled (Eger et al., 2020, 2022). For example, a recent 2023 re-convening of the Kelp Plan workgroup to assess progress thus far and identify priority next steps resulted in the publication of a Status Update (linked here). In it, the kelp community concluded that while bull kelp canopy distribution data collection is "on-track", knowledge about the abundance and distribution of other kelp species that associate with bull kelp such as understory algae (algae that do not reach the sea surface, and thus cannot be documented by kayak or aerial methods) are "off track". Specifically, for Goal 3 Distribution and trends status, one of the primary "Next Steps" items identified was "Develop and Standardize understory monitoring protocols (e.g., ROV)". Likewise, under Goal 5: Restore kelp forests, kelp forest reintroduction at recruitment limited sites was found to be "Not Started", the lowest on the scale of progress, with "Next Steps" including using tools such as ROVs to identify barriers to recruitment.

Thus far, the primary method of surveying the benthos—the seafloor and the invertebrates, algae, and fishes living in and upon it—are via scientific scuba divers. Standardized survey protocols have been in place for decades across the pacific northeast. For example, Partnership for Interdisciplinary Studies of Coastal Oceans, PISCO, linked here, have been conducting benthic surveys for decades (Menge et al., 2019). Reef Check (linked here), a world wide organization of citizen science scuba divers has very similar benthic survey protocols (see **Appendix F. Reef Check benthic survey protocols**), and have recently expanded into Washington and established a network of subtidal survey sites (see their web map of sites here).

However, limitations exist regarding how much data divers can collect. Scuba diving requires swimming with life support equipment underwater (i.e., one must carry a tank of breathing gas on their back), thus divers are limited on how long they can stay down, how deep they can operate, and the amount of information that can be written on an underwater slate. That being said, divers provide unparalleled "search" potential—they can look under cracks and crevices with a flashlight, lift blades of algae to find cryptic taxa underneath, and can manipulate and interact with the environment in indispensable ways. We do not seek to replace divers—the entire Seattle Aquarium research team on this project are themselves scientific scuba divers—but rather we seek to flesh out and further refine a complementary benthic monitoring tool that can expand upon what divers do.

ROVs have great potential to augment scientific divers by surveying kelp forests across larger areas of seafloor than divers can operate, thereby increasing the amount of information on hand to guide conservation and restoration (Boavida et al., 2016; Buscher et al., 2020; Hamel et al., 2020). Thus far however, the large size and expense of ROVs have focused them to deeper areas where they are deployed off large, seafaring vessels. Kelp forest operations require a ROV than is small and sufficiently maneuverable to get in and out of canopy forming kelp forests without the ROV's tether (a line from the ROV to the surface vessel containing communication information) becoming entangled. Fortunately, over the past few years ROV technology has advanced considerably, and the latest ROV models on the consumer market are small, maneuverable, relatively inexpensive, and are suited for deployment within kelp forests.

In partnership with the Port of Seattle, in 2022 the Seattle Aquarium launched its Coastal Climate Resilience (CCR) research program centered around using customized versions of Blue Robotics' BlueROV2, a small and highly customizable ROV, to conduct standardized video surveys within bull kelp forests (see information about our work here on GitHub). We first developed novel methods of entering and exiting bull kelp forests without getting the tether inextricably entangled (see, e.g., here and here). We also customized camera and lighting placement to illuminate the seafloor underneath the ROV, optimizing data collection of substrate type, invertebrates, algae, and bottom-fishes. Additionally, a forward-facing camera collects video of bull kelp stipes as well as schooling fish in the lower portion of the water column. As an example, you can see a synchronized side-by-side of the two video streams here.

The large quantity of ROV photo and videos would be challenging for a human to manually annotate (extract data) in a reasonable timeframe. Therefore, we are in the process of creating image analysis pipelines using open-source (freely available), artificial intelligence (AI) algorithms. AI has great potential to expedite the image analysis process and turn imagery into actionable data, though they still require large data sets of labeled (human annotated) data to "train" upon (Jacobs & Galluaudet, 2020). In 2023 our CCR team developed a program to train Seattle Aquarium volunteers to annotate ROV survey imagery using CoralNet (Williams et al., 2019), linked here, an open-source platform that uses convolutional neural networks (CNNs) to train an image classifier on user-uploaded images (Elawady, 2015; Villon et al., 2018). Our volunteer training program involves educating personnel on 66 categories of substrate, algae, and invertebrates. To date, we have 10 AI Teacher volunteers who have annotated over 800 ROV survey images, generating 80,000 data points from our Urban Kelp ROV surveys with the Port of

Seattle. Our CoralNet algorithm has been trained sufficiently such that it is making predictions on unlabeled data, and our AI Teachers now review and either “verify” or “correct” the algorithm’s percent-cover predictions. As an example, one of our most common categories, the understory algae sugar kelp, is being detected with an accuracy of 89%.

At its core, this project is centered around increasing the amount of information we have about the substrate, algae, invertebrates, and fishes within kelp forests, all to fill knowledge gaps identified by the state of Washington to help guide future bull kelp monitoring, conservation, and restoration. To do this, we will (**1**) conduct a ROV-scuba diver methods comparison with Reef Check to identify how data derived from both platforms differs in order to optimize each. We will also (**2**) team up with PSRF in order to evaluate how best ROVs may inform bull kelp restoration via surveys of treatment and control sites surveyed at the beginning and end of the growing season, as well as regular ROV monitoring of the outplanted kelp population. Finally, data from the first two portions of the project in addition to leveraged data from our Urban Kelp project with the Port of Seattle will guide (**3**) our expansion of an existing bull kelp habitat suitability model to increase the fine-scale resolution of the model’s predictions of locations optimal for bull kelp restoration in Puget Sound.

3.2 Study area and surroundings

This study is designed to capture as much variation in benthic substrate and community structure as possible to (1) produce a more robust ROV-scuba methods comparison, and (2) to provide our habitat suitability model a range of substrate and benthic community structure conditions. Accordingly, we will run our ROV along kelp forests growing upon soft sediment with small rocks (Centennial Park in Elliott Bay), large boulders comprised of riprap along a breakwater (the Elliott Bay Marina breakwater), along the basalt and limestone (characterizing Deadman’s Bay and Smallpox Bay along the west end of San Juan Island), and along soft-sediment and large rocky boulders (offshore of Burrows Lighthouse on Burrows Island by Anacortes) (**Figure 1**). The two sites in Elliott Bay are already part of our Urban Kelp ROV research with the Port of Seattle, and we have flown the ROV at Deadman’s Bay and Burrows Lighthouse as well.

As expected and intended, benthic community structure varies widely across these sites. From south to north, Centennial Park supports a narrow, fringing bull kelp bed along with extensive understory algae. Sugar kelp is widely abundant, as is the invasive *Sargassum muticum*. Red algae and green algae are also frequently observed, especially in the shallow portions of the bed (<4m). Just to the north, the riprap along the Elliott Bay Marina supports a similar benthic community though one with a greater diversity of invertebrates such as sea cucumbers, sea stars, and sea anemones along the hard substrate, and several additional species of brown algae such as sea colander kelp. Demersal and midwater fishes such as lingcod, cabezon, and various species of rockfish and perch are also more abundant along the riprap relative to the soft-sediment of Centennial Park.

We briefly flew the ROV at Deadman's Bay in the San Juan Islands, and we anticipate a greater diversity of invertebrates and algae, including patches of higher densities of sea urchins.

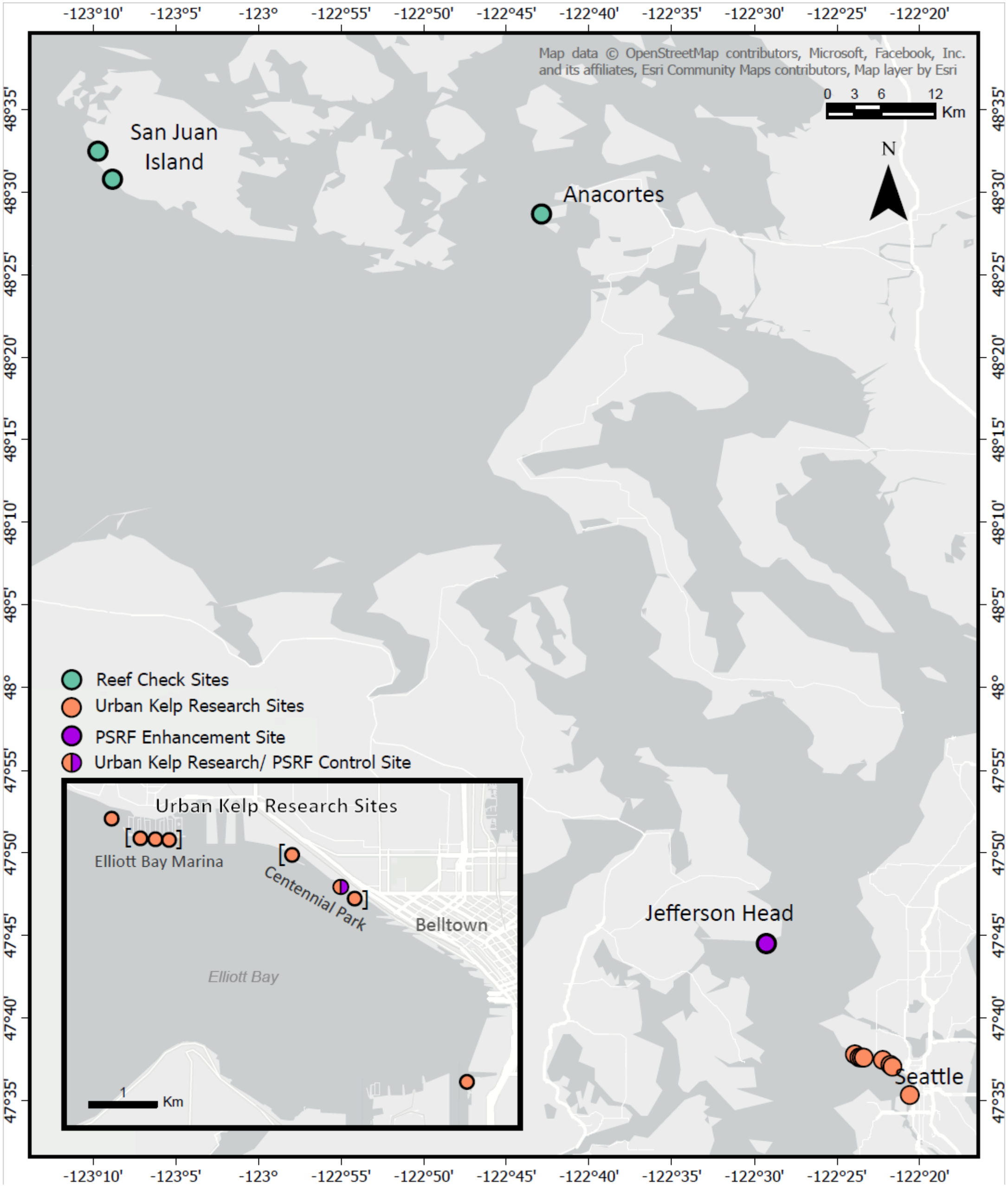

Figure 1. Map of study area with an inset of Elliott Bay and our Urban Kelp ROV sites

These sites include leveraged data from the Urban Kelp project with the Port of Seattle (inset). The Reef Check sites along with Elliott Bay Marina breakwater and Centennial Park will be surveyed as part of the methods comparison. The PSRF enhancement site is our survey location for the bull kelp restoration work. Data from all sites including the Urban Kelp sites will be utilized as part of our expanded bull kelp habitat suitability model. See **Figure 2** in **6.2.2.1 Comparability** for map of our ROV transects at the x3 Centennial Park sites in Elliott Bay.

We also flew the ROV at the Burrows Lighthouse dive site with the Samish Dive Team in 2022 and encountered a rich diversity of brown algae as well as numerous sea stars and embedded sea cucumbers. We intend for our San Juan Island ROV surveys to capture understory algae such as *Ptyerogophoa californica*, which, being 1-2m tall, may be challenging for the ROV to survey while maintaining a 1m altitude above the seafloor, and we want to capture any such difficulty in our methods comparison.

Finally, in terms of ROV surveys sites, we will also survey PSRF's bull kelp enhancement site at Jefferson Head. This will involve before and after site surveys of the "treatment," or "enhancement" site (where bull kelp seed are outplanted) as well as a control site where bull kelp grows naturally (along Centennial Park). We have not flown the ROV at Jefferson Head yet, though according to PSRF personnel it consists of unconsolidated sediments such as mud, sand, and shell-debris mixed with gravel and cobble. During the growing season red and brown understory algae form patches along suitable substrate.

3.2.1 History of study area

Puget Sound, located in the Pacific Northwest region of the United States, is a complex and intricate estuarine system. As part of the Salish Sea, it extends from the Strait of Juan De Fuca through to Olympia, Washington. In terms of geological formation, the most significant event was the Vashon Glaciation, which occurred 15,000 years ago during the last Ice Age. As the glaciers retreated, they carved out the deep basins, channels, and sills that that now make up Puget Sound (Haugerud, 2021). In doing so, these glaciers left a variety of sediments such as glacial till. Marine sediments were introduced as sea levels rose, and these are typically finer grained. Other sediments in the nearshore include more recently delivered such as via rivers and landslides, and these are more often found in the deltas and the nearshore areas of Puget Sound.

3.2.2 Summary of previous studies and existing data

Our study area was designed to capture a broad set up these varying substrate characteristics as well as varying benthic community structure. In terms of recent data collection efforts, the most similar to our study are Washington Reef Check's scuba diver surveys which operate throughout the state; their program was initiated in 2021 and surveys fully got off the ground in 2022. Furthermore, in terms of bull kelp canopy coverage, the Northwest Straits Initiative maintains a robust kayak monitoring program (linked here). Additionally, DNR conducts aerial flyovers of kelp canopies, especially in the western Strait of Juan de Fuca and along the Olympic Coast (Pfister et al., 2018), as well as camera tows and other monitoring throughout Puget Sound (Berry et al., 2021). Furthermore, while the project has yet to fully get underway with data collection, our partners at DNR recently noted that summer 2024 they will initiate a project to map via aerial drone all the canopy forming kelp in Elliott Bay—there is much potential to collaborate and combine data where possible, and we look forward to coordinating efforts.

In terms of existing data we will leverage as part of this project, especially when expanding upon the bull kelp habitat suitability model, please see here and here for high level overviews of our Urban Kelp Research Project with the Port of Seattle. In 2022, we established a network of eight ROV survey sites in Elliott Bay (see **Figure 1**, inset). At each of the eight sites we conduct four ROV transects, ranging in length between 50-100m, depending upon the local configuration of the kelp bed and surrounding area. We surveyed these sites in the summers of 2022 and 2023, and the Port of Seattle has renewed our contract and we will continue these surveys in 2024 and 2025. In particular, we will expand our surveys to include winter surveys in January and February when the annual vegetation is absent, enabling our ROV to extensively document the underlying substrate that is not typically visible in the summer, when it is covered by algae. These winter surveys in particular will be useful for our bull kelp habitat suitability model, as the ROV obtains fine-scale metrics of substrate categories such as sand, silt, cobble, shell-debris, pebbles, and hard substrate, all of which have varying associations with bull kelp's ability to grow and persist.

You can see an example of some of our Urban Kelp ROV data we will leverage for our habitat suitability model linked here. The substrate and benthic community data (columns "AE" through "BE") are the output from our manual annotations in CoralNet, and they have been appended to metadata including site information, photo ID, as well as GPS coordinates to facilitate spatial analyses (columns "A" through "AD"). Each row contains data from an individual geospatially referenced image.

3.2.3 Parameters of interest and potential sources

The parameters of interest for this project include:

- *ROV derived*: percent-coverage of benthic taxa and substrate type; n=50 percent-coverage data points per image from the downward-facing camera (x2 GoPro HERO12).
- *ROV derived*: abundances of invertebrates, certain conspicuous algae, and fishes from both the downward- and forward-facing cameras.
- *ROV derived*: spatial tracking of the ROV, i.e., 1Hz (1 per second) latitude and longitude coordinates of the ROV's position underwater to facilitate spatial analyses.
- *ROV derived*: additional ROV sensor telemetry, from, e.g., our WaterLinked Doppler Velocity Log (DVL) used to measure the altitude of the ROV above the seafloor and enable autonomous mission planning.
- *Scuba diver derived*: abundances of invertebrates and algae as well as percent-coverage data using Reef Check scuba diver protocols.
- *Habitat suitability model derived*: map layers with finer-scale predictions of specific locations where bull kelp is forecasted to persist, given the inclusion of our ROV abundance and percent-coverage data.

3.2.4 Regulatory criteria or standards

Not applicable.

3.3 Water quality impairment studies

Not Applicable.

3.4 Effectiveness monitoring studies

Not Applicable.

4.0 Project Description

4.1 Project goals

The goals of this project are to:

- Identify the specific pros and cons of conducting ROV surveys in kelp forests via the ROV-diver methods comparison. Identify the species or taxa that ROVs are well-suited to survey, as well as the species or taxa that scuba divers are more appropriate suited to survey. Generate a scientific publication as well as technical report detailing these findings for the Washington kelp monitoring, conservation and research community.
- Identify how best to incorporate ROVs into bull kelp restoration. We will help account for any differences in local scale factors (e.g., substrate type, community structure) via before and after site surveys of the enhancement site and a "natural" kelp bed (a control site). Additionally, we will conduct routine monitoring of the outplanted bull kelp via the ROV.
- Expand upon an existing bull kelp habitat suitability model by first updating all environmental data layers from the original model. We will then incorporate simulated ROV data into the model in order to conduct a sensitivity analysis. Finally, once the model's behavior is fully understood, we will incorporate our real ROV data into the habitat suitability model in order to obtain finer-scales of predictions of bull kelp habitat suitability.

4.2 Project objectives

The objectives of this project are to:

- Conduct a methods comparison where ROVs and Reef Check divers gather data above the same 30m transects (survey locations) on the same day. These surveys will take place summer (late August/September) 2024 and 2025, as well as in the winter (January/February) 2025 and 2026. Surveys will take place at a minimum at two locations in Elliott Bay (Centennial Park and along the Elliott Bay Marina breakwater) and at two locations on the west end of San Juan Island. A fifth site at Burrows Island was not originally included in the scope of work or Statement of Work for this project, though we intend to survey it anyways to fully practice our ROV-diver choreography with Samish Reef Check divers prior to the surveys along the west end of San Juan Island.
- Conduct ROV-based bull kelp restoration site surveys and regular monitoring with PSRF. The site surveys will take place in March and June of 2024 and 2025, and the routine bull kelp outplanting monitoring will take place every other week between March and June in 2024 and 2025.
- Update an existing bull kelp habitat suitability model's environmental layers and incorporate ROV derived percent-cover and abundance information. We will first update the existing data layers and incorporate new tools such as an analysis of bathymetry layers via the Benthic Terrain Modeling tool, we will simulate and incorporate ROV data

to perform a sensitivity analysis, then we will incorporate real ROV data to obtain finer-scale predictions of optimal bull kelp habitat.

4.3 Information needed and sources

Below we first summarize the data we will collect in the field, followed by Table 1 which outlines the GIS layers to be utilized as part of Task 4.

ROV derived data will include sensor and telemetry files including .bin files and .csv files containing all geospatial information (e.g., the GPS coordinates where the ROV flew along the seafloor), as well as flight data such as ROV altitude above the seafloor, temperature, battery power remaining, etc. These data are obtained from the ROV laptop after each flight, and we download them and store them on the Seattle Aquarium DropBox.

ROV photo and video survey data will include 23 megapixel (MP) images shot on "time-lapse" mode on x2 GoPro HERO12 cameras, as well as 4K, 30 frames per second (fps) video, also shot on a GoPro HERO12. Our downward-facing cameras shoot photos, and our forward-facing cameras shoot video. The camera settings as well as a SOP for photo and video processing are described in **Appendix B. SOP for camera settings and imagery processing.**

We will extract percent-coverage information for substrate type and aggregate taxa such as brown, red, and green algae from the 23MP photos taken by the downward-facing camera using CoralNet (Williams et al., 2019), an open-source AI program. We will extract abundances of bull kelp stipes and certain schooling fishes from the forward-facing video that is to be analyzed in Video and Image Analytics for the Marine Environment—VIAME, (Dawkins et al., 2017)—linked here, and we will also use VIAME to analyze the downward-facing photos to extract abundances of individually conspicuous invertebrates (such as sea urchins, sea stars, and sea anemones), certain algae, and some demersal fish.

Scuba divers will gather invertebrate and algae "swath" data (counting abundances of individually conspicuous invertebrate and algae species) and Uniform Point Contact (UPC) to measure percent-coverage of substrate type as well as species not captured by abundance counts, such as, e.g., colonial sponges and fleshy red and green algae. These Reef Check protocols are detailed extensively in their recent QAPP signed October 12th, 2023, and the precise benthic protocols are also linked in **Appendix F. Reef Check benthic survey protocols**.

Table 1 below details the data layers that will be updated as part of our expansion of the existing bull kelp habitat suitability model.

Table 1. Data layers comprising the bull kelp habitat suitability model

Dataset	Source	Description
NCEI Bathymetric Attributed Grid (BAG) Mosaic from 1955-2018	Bathymetric Data Viewer (https://www.ncei.noaa.gov/maps/bathymetry/)	Mosaic of high-resolution quality-controlled seafloor elevation from NOAA National Ocean Service (NOS) Hydrographic Survey Bathymetric Attributed Grids (BAGs) in U.S. coastal waters.

Shoreline Infrastructure	Puget Sound Change Analysis (PSNERP) (http://www.pugetsoundnearshore.org)	Geodatabase data including armoring, breakwaters, marinas, nearshore fill, railroads, nearshore roads, tidal barriers, wetlands, land cover, shore form, and drift cells.
Hydrodynamic and Water Quality Solution Files	Salish Sea Model (SSM) 2017 Hydrodynamic Solution (Khangaonkar et al., 2018)	A diagnostic hydrodynamic and biogeochemical model (nutrients, phytoplankton, carbon, dissolved oxygen, and pH) of the Salish Sea.
Washington State ShoreZone Inventory: Floating Kelp Extent 1994-2000	Washington Department of Natural Resources, 2000 and Dethier, 2014	Line layer showing the distribution of floating kelp from the larger Washington State ShoreZone Inventory.
Marine Water and Sediment Monitoring	Washington State Department of Ecology, 2017	Long term monitoring of water and sediment in Puget Sound, Grays Harbor, and Willapa Bay.
Floating Kelp Indicator for Washington State	Washington Department of Natural Resources, 2023	Floating kelp monitoring indicator for Washington State (KelpForestWA). These data include 171 floating kelp monitoring sites in Washington State.
ROV-derived data	Urban Kelp Research Project, Seattle Aquarium, 2022-2024	Percent-coverage information for substrate type and aggregate taxa such as brown, red, and green algae

4.4 Tasks required

Task 4.1: **Field work to conduct a ROV-scuba diver methods comparison.**

- Randell, Williams, and Bogdanova will coordinate with the Seattle Aquarium Dive Program, Reef Check, and the Samish Indian Nation Reef Check divers to fly the ROV above Reef Check survey sites the same day that divers survey them.
- This methods comparison will take place at four sites in two distinct geographic locations: (1) Elliott Bay (specifically, offshore of Centennial Park and along the Elliott Bay Marina breakwater) and (2) the west side of San Juan Island (specifically, Deadman's Bay and Smallpox Bay). We will additionally perform a "shakedown" field trial with Reef Check and the Samish Dive Team at Burrows Lighthouse, just west of Anacortes, before proceeding to the main San Juan Island data collection trip.
- Seattle Aquarium staff will attend the Reef Check partner retreat to become certified in Reef Check's methodology (in early May), and upon completion of the training, will bring the methodology back to the Seattle Aquarium in order to certify additional dive personnel (in early June)

- The ROV-diver methods overlap will take place in summer 2024 (September), winter 2025 (February), summer 2025 (September), and winter 2026 (February).
- At minimum, x3 30m transects will be surveyed via Reef Check's divers as well as our ROV, matching precisely the Reef Check protocol (please see **Appendix F** for a link to the full Reef Check protocol).

Task 4.2: **ROV site surveys and outplanted bull kelp monitoring at Jefferson Head.**

- The work required includes training our HSIL Research Technician to operate the ROV (as this is our earliest field work), as well as to complete all telemetry file processing, AI image annotation, and video editing.
- We are coordinating closely with PSRF personnel. On our first PSRF ROV survey day, we will motor our vessel from Bell Harbor Marina, Seattle, to the Indianola pier, where we will pick up PSRF personnel, who will accompany us to their Jefferson head enhancement site.
- At the start of the field season (early March, 2024 and 2025), we will complete a ROV site survey (with x4 50m ROV transects) around the enhancement location. This ROV site survey will also take place at one of our Centennial Park sites which will represent a natural kelp bed "control site" (see **Figure 1**).
- Once these above site surveys have taken place, we will then monitor the bull kelp on the "off weeks", the weeks where PSRF divers are not surveying, as to increase the number of total sample points available. These surveys will involve a single ROV "pass" filming the outplanted bull kelp (as opposed to the more involved site surveys).

Task 5.1: **Process, edit, and color-correct ROV-derived photos and video**.

- As we shoot video using the "Native" white-balance setting, our imagery requires color-correction. We will use programs such as DaVinci Resolve Studio to edit videos and Adobe Lightroom to edit photos in order to process them for further analysis.
- All data will be uploaded to DropBox folders with downloadable links that we will make available to the HSIL team.

Task 5.2: **Use open-source AI programs such as CoralNet and VIAME.**

- We will use CoralNet to extract metrics of percent-coverage of aggregate taxa (e.g., substrate type, fleshy red algae, green algae, etc.) from our imagery.
- We will additionally use VIAME to generate metrics of abundance for individually conspicuous objects such as sea stars, sea urchins, bull kelp stipes, and fishes.
- All data from both CoralNet and VIAME will be extracted in the form of .csv files, and we will merge these data with our ROV's telemetry files as to provide a complete record of our benthic surveys.

- Both of these objectives will leverage our Seattle Aquarium volunteers, i.e., our "AI Teachers", who have gone through an extensive and standardized training process enabling them to help annotate our ROV imagery and train our AI algorithms.

Task 5.3: **Provide links to GIS database containing maps of our ROV survey locations.**

- We will process our telemetry files from the acoustic GPS system and the Doppler Velocity Log (DVL) to generate geospatial data for the locations where we flew the ROV.

Task 6.1: **Share our GIS database containing all model components of our updated and expanded bull kelp habitat suitability model.**

- We will iteratively update the model with new inputs (see Table 7). These will focus upon new and updated environmental data layers (e.g., from the Salish Sea Model), as well as more detailed bathymetry and substrate data using the benthic terrain modeler tool.
- We will generate simulated data for our ROV to test the behavior and output from the habitat suitability model.
- Only once the model has been extensively updated and tested will we incorporate real ROV-derived data (metrics of percent-coverage and abundance).

4.5 Systematic planning process

Not applicable.

5.0 Organization and Schedule

5.1 Key individuals and their responsibilities

Table 2 shows the responsibilities of those who will be involved in this project.

Table 2. Organization of project staff and responsibilities.

Staff	Title	Responsibilities
Dr. Zachary Randell Seattle Aquarium Phone: (408) 660-7842	Project Manager and Principal Investigator; Research Scientist	Writes the QAPP. Oversees project design and implementation. Coordinates field work with partner organizations. Participates in field and oversees imagery processing, AI training, and data analysis. Writes the draft report and final report.
Megan Williams Seattle Aquarium Phone: (206) 356-9850	Research Technician	Conducts field work, photo/video processing, AI analyses of photos/stills, and data analyses including spatial analyses.
Elena Bogdanova Seattle Aquarium Phone: (973) 652-2527	Research Technician	Conducts field work, photo/video processing, AI analyses of photos/stills, and data analyses including spatial analyses.
Dr. Shawn Larson Seattle Aquarium Phone: (206) 618-3762	Senior Conservation Research Manager	Dr. Randell's supervisor at the Seattle Aquarium. Provides internal review of the QAPP, approves the budget, and approves the final QAPP.
Joel Hollander Seattle Aquarium Phone: (206) 714-8968	Dive Program Manager	Oversees all Seattle Aquarium field work to ensure the safe conduct of scuba, ROV, and vessel operations.
Ken Nelson Department of Ecology Phone: (360) 522-2722	National Estuary Program Quality Coordinator	Reviews and approves the draft QAPP and the final QAPP.
Dr. Jodie Toft Puget Sound Restoration Fund Phone: (831) 251-5824	Deputy Director	We coordinate with Jodie and PSRF on several projects, and she will facilitate communication and collaboration as we conduct joint field operations as part of **Task 4.2**.
Jackie Selbitschka Reef Check Phone: (651) 492-8827	Washington Regional Manager	We coordinate with Jackie to ensure the Seattle Aquarium scientific divers are also certified Reef Check divers, and we have coordinated with Jackie to plan field operations associated with **Task 4.1**

5.2 Special training and certifications

The core personnel at the Seattle Aquarium who will be responsible for advancing this project on a daily basis are as follows:

- Dr. Zachary Randell: Primary Boat Operator (PBO) for the Seattle Aquarium vessel. Scientific Diver at the Seattle Aquarium. Trained ROV operator. Experience conducting subtidal and surface operations in temperate waters since 2010. Ph.D. in Integrative Biology with a Minor in Statistics from Oregon State University (defended 2021; his public dissertation defense can be found here on YouTube), with research focused on kelp forest monitoring, analysis, and dynamical modeling (dissertation linked here).

- Megan Williams: PBO for the Seattle Aquarium vessel. Scientific Diver at the Seattle Aquarium. Trained ROV operator. Trained in video/photo processing and machine learning methods of image analysis. In-progress three quarter Geographic Information Systems (GIS) credential course (linked here) at the University of Washington. Master of Science degree in Marine Biology from James Cook University, 2022. Her graduate research was centered around the effects of "sea-weeding" (removing sargassum from coral reefs) on non-coral benthic communities.
- Elena Bogdanova: Trained ROV operator. Certified AAUS Scientific diver. Trained in video/photo processing and machine learning methods of image analysis. Masters of Arts in Remote Sensing and GIS, University of Haifa, Israel, 2019. Master of Science in Marine Biology, Leon Charney School of Marine Sciences, University of Haifa, 2023. Thesis: Investigating links between fish community and structural complexity using 3D mapping in protected and non-protected Mediterranean shallow rocky reefs.

5.3 Organization chart

Not applicable – See Table 2.

5.4 Proposed project schedule

Tables 3—5 list key activities, due dates, and lead staff, and a budget breakdown for this project.

Table 3. Schedule for completing field and laboratory work

Task	Due date	Lead staff
4.1 – ROV/scuba methods comparison with Reef Check.	**October 2024, April 2025, October 2025, and April 2026**	**Randell, Williams, Bogdanova**
4.2 – ROV surveys of bull kelp restoration with PSRF.	**August 2024, August 2025**	**Randell, Williams, Bogdanova**
5.1 – Process video imagery.	**Continually via quarterly reports starting July 10 2024 through October 2026**	**Randell, Williams, Bogdanova**
5.2 – AI analyses of ROV imagery.	**Continually via quarterly reports starting July 10 2024 through October 2026**	**Randell, Williams, Bogdanova**
5.3 – links to maps of ROV survey locations.	**January 2025, January 2026**	**Randell, Williams, Bogdanova**

Task	Due date	Lead staff
6.1 – link to GIS database containing habitat suitability model.	**January 2025, January 2026**	**Randell, Williams, Bogdanova**

Table 4. Schedule for final report

Task	Due date	Lead staff
Draft to supervisor	**June 2026**	**Randell**
Draft to client/peer reviewer	**July 2026**	**Randell**
Draft to external reviewers	**July 2026**	**Randell**
Final draft to Strategic Initiative	**September 2026**	**Randell**
Final report due on web	**September 2026**	**Randell**

5.5 Budget and funding

PI Randell and Research Technician Williams are both covered by the Seattle Aquarium's General Operations budget, though a small portion of Randell's time is covered on this grant to supervise the completion of the project. Research Technician Bogdanova's entire salary and benefits are covered by this HSIL project, and completing the ROV field work, photo/video processing, AI analyses, data analysis, and GIS analyses are her primary responsibilities. Total personnel salary costs are $189,150.02 for the project, including $66,202.51 for fringe benefits (**Table 5**). The Seattle Aquarium applies a 10% indirect rate on personnel and fringe costs, totaling $22,300.00.

We also budgeted $22,300.00 for field work and travel. This largely will go towards fuel and moorage costs for the Seattle Aquarium vessel in order to complete **Tasks 4.1** and **4.2.** The Seattle Aquarium makes priority use of its vessel for this research. These travel funds also include support to send Seattle Aquarium personnel to the Salish Sea Ecosystem Conference (SSEC) as the HSIL Virtual Subrecipient Summit.

In terms of equipment, the Seattle Aquarium has funded (matched) over $35,000 worth of ROV and related equipment to initiate the CCR program. We have also budgeted approximately $38,000 worth of equipment funds into this HSIL project to upgrade our ROVs, purchase additional sensors, and purchase equipment necessary to power the ROV from the Seattle Aquarium's vessel. We also included $13,408 in supplies, largely consisting of relatively small spare and replacement ROV components or camera parts.

Table 5. Budget breakdown by category

Budget category	Total funds allocated
Personnel	**$189,150.02**
Fringe benefits	**$66,202.51**
Travel	**$22,300.00**
Equipment	**$38,000.00**
Supplies	**$13,408.00**
Indirect at 10% on personnel and fringe	**$25,535.25**
Total Grant Amount	**$354,605.00**

6.0 Quality Objectives

6.1 Data quality objectives

The overall data quality objectives for this project are to (**1**) gather usable (expanded upon below) photo and video imagery of the benthos via ROV surveys, along with (**2**) usable (also expanded upon below) ROV telemetry data including latitude/longitude ROV tracking information, during the course of ROV surveys within bull kelp forests in Washington. These imagery and telemetry will be processed to generate data about substrate type and benthic community structure, and these spatial data will contribute to an expansion of an existing bull kelp habitat suitability model, the predictions from which will (**3**) provide finer-scale spatial predictions about where bull kelp may thrive in Puget Sound.

The data quality objectives are met when:

- ROV surveys are completed when annual water clarity conditions are such that image quality (sharpness, clarity) is maximized. That is to say, we avoid surveying in June, July, and early August as much as possible as the annual algae blooms render the water brown and turbid, thus challenging our ability to obtain clear imagery of the seafloor. We also survey during a flooding (rising) or slack high tides as much as possible, again to obtain clearer, sharper, imagery. See here for an example of clear, sharp, benthic imagery.
- We conduct surveys during the same time each year as to obtain a consistent time series (for the 2024, 2025, and 2026 surveys).
- We conduct at-minimum x3 and hopefully (depending upon topside conditions) x6 30m transects (individual surveys) for the ROV-diver methods comparison.
- Our ROV navigational system provides consistent and precise geospatial information enabling us to use those spatial data as part of our bull kelp habitat suitability model.
- Our scuba diver data adheres to Reef Check’s standardized monitoring protocols.

6.2 Measurement quality objectives

Our ROV surveys within kelp forests are relatively new—we have developed the protocols ourselves since PI Randell starting working at the Seattle Aquarium in March of 2022. However, our ROV surveys are fundamentally derived from scientific scuba diving benthic survey procedures, such as PISCO's methods as well as Reef Check's methods (see **Appendix F. Reef Check benthic survey protocols**). Therefore, our primary Measurement Quality Objective is to conduct ROV surveys in a manner that expands upon the rigor, statistical independence, and repeatability of standardized benthic scuba based surveys to capture abundances of mobile and conspicuous invertebrates and algae, and percent-coverage data regarding substrate type and aggregate (not individually discernable) taxa.

Central to the standardization of benthic scuba diver surveys is the ability to link abundances and percent-coverage estimates to precise areas of seafloor—this allows one to generate density estimates. For divers, this involves laying down a meter tape (often a 30m tape) and counting conspicuous and individually identifiable invertebrates, algae, and fish species. Likewise, divers also conduct Uniform Point Contact (UPC) surveys to record the percent-category of aggregate taxa as well as seafloor type along each meter mark down the tape.

As an expansion of these benthic diver surveys, our ROV methodology also captures abundances and percent-coverage information. However, instead of laying down a meter tape to track the area of seafloor surveyed, we precisely track the spatial position (i.e., latitude/longitude coordinates) of the ROV along the seafloor. And a step further, because we know the altitude of the ROV above the seafloor (via a Doppler Velocity Log), and because we know the angle of the camera lens that is facing downwards, via a simple trigonometric function in *R* we can calculate the area captured by the downward-facing camera at any moment in time (see, e.g., [here]). We can combine these area and geospatial tracking data streams to analyze a variety of ROV surveys parameters, as exampled by **Figure 2**, which display our real-world survey parameters for our summer 2022 ROV surveys with the Port of Seattle.

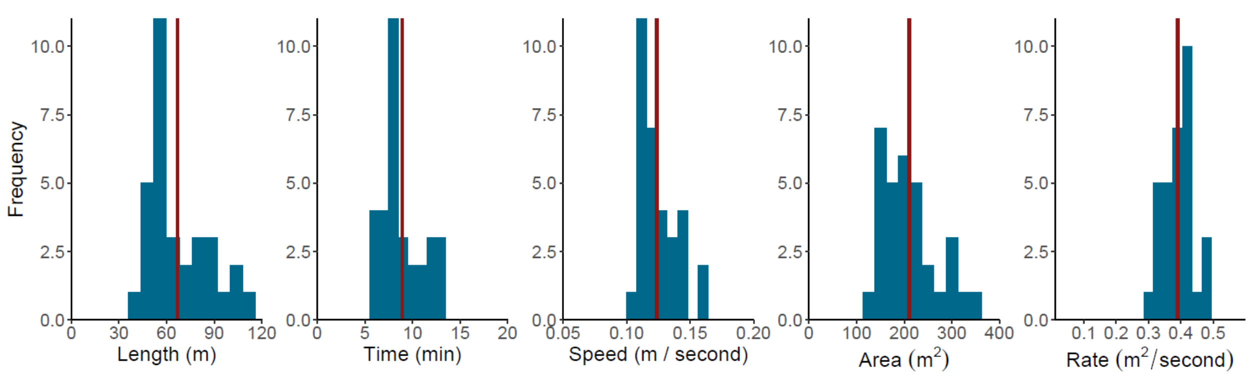

Figure 2. ROV survey parameters

Frequency histograms of ROV survey parameters from n=32 ROV transects (surveys) completed summer 2022 for the Port of Seattle. The red line denotes the mean value of each parameter. Survey parameters include transect length in meters, survey time in minutes, ROV speed in meters per second, seafloor area filmed in units of meters squared, and rate (or ROV survey effort) in units of meters squared filmed by the downward-facing camera per second.

This is all to say, precise spatial tracking is an essential component of our ROV surveys, thus it is a key Measurement quality objective. Please see **Appendix A: SOP for ROV Navigation and survey flights** for details about how we achieve this objective. We have x2 separate systems that provide geospatial information, providing redundancy. The first is WaterLinked Underwater GPS (UGPS) system, and the second is WaterLinked's Doppler Velocity Log DVL-A50. In particular, the latter sensor provides highly precise spatial tracking across the seafloor via x4 acoustic beams. The "performance" model of the DVL-A50 (which we have) has a long term accuracy of 0.1%.

Additionally, regarding our AI analyses, they will take extended periods of time to train before they can generate output that does not need to be "reviewed" by a human (where the human either "verifies" or "corrects" the algorithm's predictions). Our target is for our algorithms to perform at 90% accuracy. For some categories of taxa that are commonly observed, we are near this goal—our CoralNet algorithm can detect sugar kelp with 89% accuracy—but we have over 10,000 data points of sugar kelp. Most other categories in CoralNet do not yet have as many data points and thus are not at that high of an accuracy, and until they are, we will maintain our "human review" of all project data.

6.2.1 Targets for precision, bias, and sensitivity

6.2.1.1 Precision

When we conduct our ROV surveys, we use an advanced navigation system that enables us to be within 2m of a GPS point or trackline. We have precise flight control over the ROV thus our target for resurveying locations is to survey within 2m of the previous survey location.

Regarding our AI analyses, all AI Teachers go through an extensive training program before they annotate any of our real data. As part of this, they are standardized on a set of 10 images with 100 data points each, and we evaluate and correct any misunderstandings in percent-cover categories. Furthermore, we regularly go back and review annotated images, and we aim for 90% accuracy. Anything below that cutoff triggers a conversation and potentially additionally training. Certain categories of algae look very similar underwater, thus the 90% allows for a degree of subjectivity in percent-cover cases that are especially difficult to differentiate.

Regarding the Reef Check data, please refer to **Appendix F**, which links to the full Reef Check training procedure, survey protocols, and QAQC standards. In short, Reef Check personnel receive extensive training and are expected to be within 10% of the lead diver's data. Calibration training is required every year, and our Seattle Aquarium Scientific Divers will go through this training.

6.2.1.2 Bias

Not applicable.

6.2.1.3 Sensitivity

Not applicable.

6.2.2 Targets for comparability, representativeness, and completeness

6.2.2.1 Comparability

As detailed above in **6.2 Measurement quality objectives**, maintaining comparability of our ROV methodology with similar scuba diver surveys is a core goal our work. The ROV methods used in this project will expand upon the protocols developed for the Urban Kelp Research Project with the Port of Seattle (and please see **Appendices A—E** for a complete list our Standardized Operating Procedures (SOPs) associated with our ROV surveys).

To ensure comparability across our surveys, as part of the Urban Kelp study we complete x4 replicate transects at each of the x8 ROV survey sites in Elliott Bay. The x8 sites are displayed in **Figure 1**, and **Figure 3** below shows a map of x4 ROV transects at each of the x3 ROV sites in Centennial Park. These x4 transects are akin to the classic 30m scuba diver transects that are part of PISCO and Reef Check protocols, enabling comparison to the numerous Reef Check diver sites throughout coastal Washington.

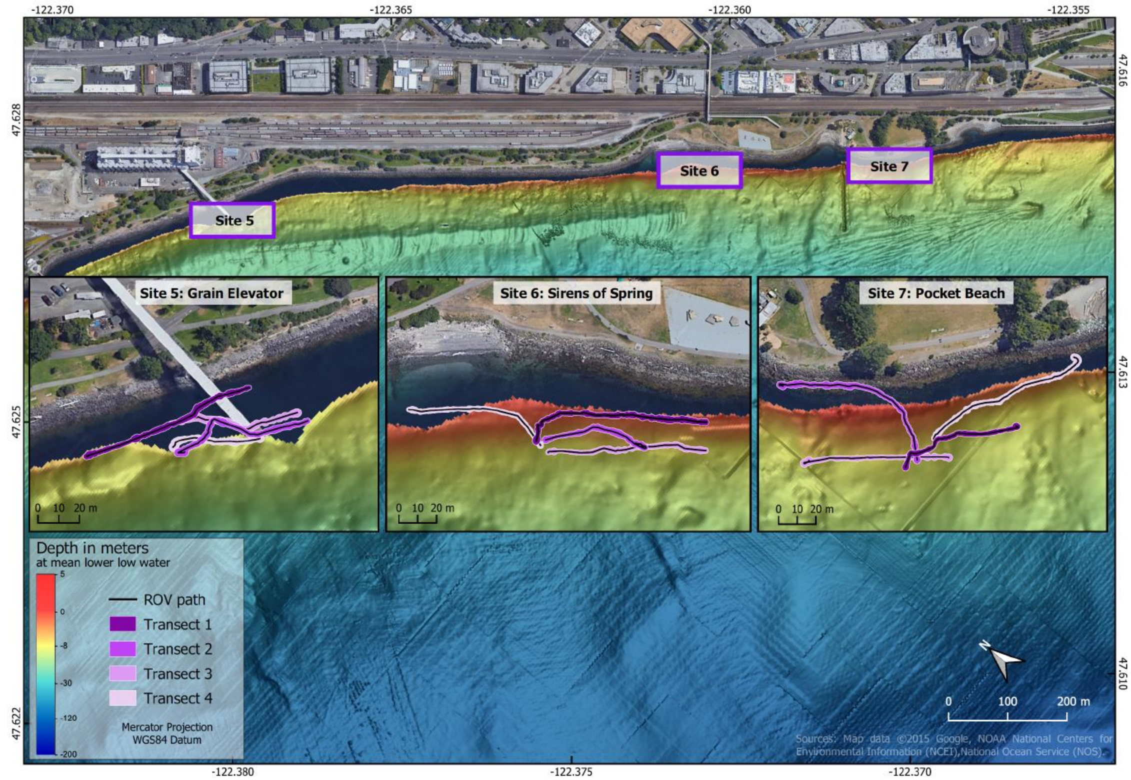

Figure 3. Site and transect maps from our Urban Kelp research

The x3 replicate sites along Seattle waterfront at Centennial Park—Grain Elevator, Sirens of Spring, and Pocket Beach—enabling statistical comparison and differentiation of these sites with the x3 replicate sites along the Elliott Bay Marina breakwater as part of the Urban Kelp Research Project with the Port of Seattle (not displayed here). The transects depicted here were from our summer 2022 ROV surveys.

Furthermore, in terms of comparability, six of the eight Urban Kelp ROV survey sites are further grouped into "super sites" (**Figure 1** inset), specifically the x3 Centennial Park (**Figure 3**) and x3 Elliott Bay Marina breakwater sites. These groupings were established to provide replication at the site level, thereby enabling statistical differentiation of the two broader locations: Centennial Park versus the Elliott Bay Marina breakwater. Thus, our ROV methodology—while relatively new—is fundamentally based on tried and tested subtidal field methods such as standardized survey areas, as well as and replication at the proper spatial scale given the desired spatial scale of inference.

Finally regarding comparability, note in **Figure 3** that the transects have a slight "wiggle" to them—this is a reflection of our seafloor area measurements—and the slight oscillation in area surveyed is a reflection of the small changes in ROV depth as we manually maneuvered the ROV and attempted to maintain a consistent 1m altitude above the seafloor. Since then, we now use *surftrak* (linked here)—open-source software developed by Clyde McQueen, a Seattle Aquarium Special Projects Volunteer (SPV), that enables our ROV to autonomously adjust its depth across varying terrain to maintain our desired 1m altitude above the seafloor. The successful development and testing of *surftrak* on ROV *Lutris*, one of our x2 ROVs, was a major milestone that is but one of several custom software efforts underway to ensure that our ROV imagery is as consistent and comparable as possible. By keeping the ROV at a consistent 1m depth above the seafloor, we minimize variation in lighting and keep the scale of our imagery consistent.

6.2.2.2 Representativeness

Our ROV survey methodology will be as—or more—representative of the underlying seafloor than scuba diver benthic surveys transects. For example, the standard diver based Uniform Point Contact (UPC) survey records x3 data points along each meter of a 30m transect: (1) substrate type, (2) relief, a measure of vertical rugosity, and (3) the primary substrate holder, e.g., red algae growing upon a rock. These tally to 90 data points per 30m transect. With our ROV surveys, our downward-facing camera takes a 23MP image every meter, and we use CoralNet to analyze any desired number of randomly placed points across that image (**Figure 4,** Row 2). After revising our analytical approach (discussed below) we switched from annotating 100 data points to now annotating 50 data points per image. For a 30m ROV transect, that equates to 1,500 data points, versus the diver UPC survey's 90 data points. In terms of representativeness, our ROV surveys provide a comprehensive accounting of the seafloor. How these 1,500 data points compare to the diver data, however, still needs to be evaluated (hence our present study), as the diver will be able to interact with the seafloor in ways the ROV cannot.

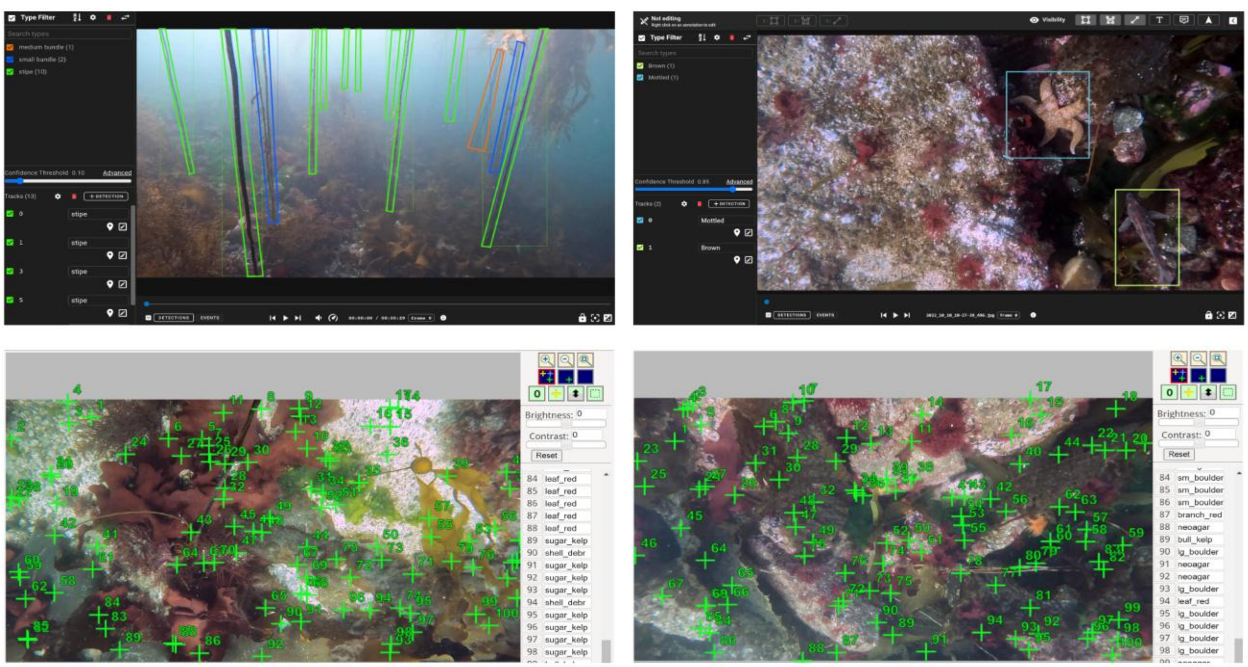

Figure 4. Examples of AI annotations from ROV surveys in Elliott Bay

Row 1: (Left) manually annotations of bull kelp stipe abundances from the forward-facing camera using VIAME. (Right) object detection in VIAME using the downward-facing imagery to capture abundances of invertebrates and fishes. Row 2: (Left and Right) percent-coverage from the downward-facing camera to generate metrics for our 66 categories of substrate type and aggregate taxa such as brown and red algae.

6.2.2.3 Completeness

As noted above, we are able to control how many data points per image we analyze. And now that we have annotated over 800 images from our ROV surveys—each with 100 data points—we can evaluate the degree to which our sampling design is "complete" relative to, e.g., annotating fewer images per survey and/or fewer data points per image. Specifically, we can evaluate species accumulation curves (also known as species richness curves) for some of our preliminary Urban Kelp ROV data (**Figure 5**). These curves allowed us to evaluate the tradeoff between "completeness" of data extracted from the photos, i.e., extracting large numbers of data points from every image, versus the efficacy of our image clearance rate (the rate at which we manually annotate our images).

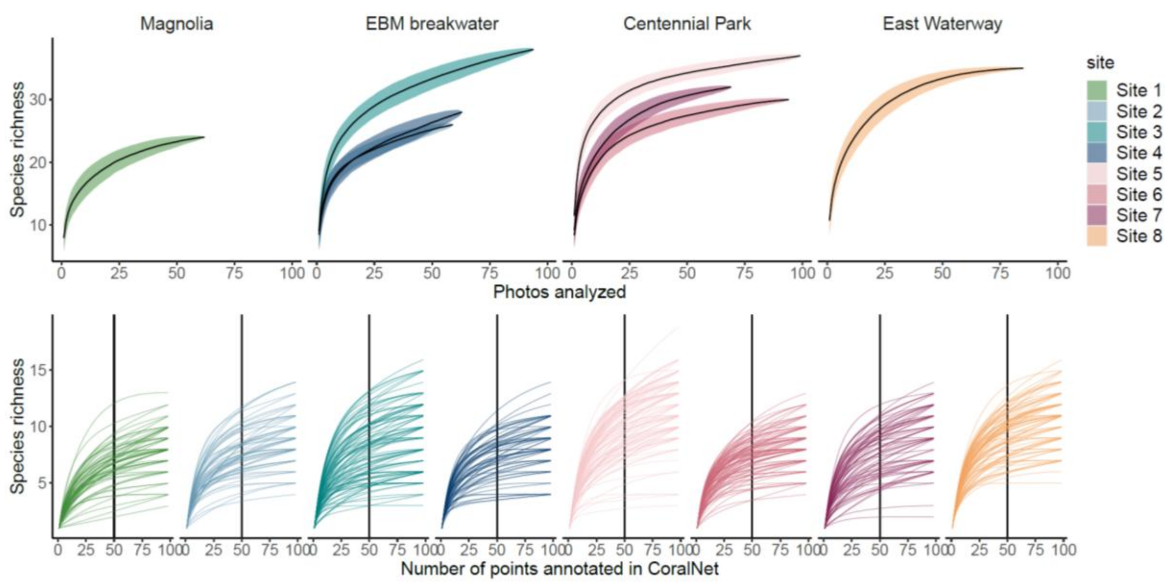

Figure 5. Diagnostic species richness curves

Row 1: The relationship between the number of photos per transect on the x-axis and the resulting species richness on the y-axis. The asymptotic leveling out of these figures depicts the point (the number of photos) at which additional photos do not yield further returns on additional species captured by the survey. Curves were calculated using the *specaccum()* function in *R*. Row 2: The relationship between the number of data points per image and the associated patterns of species richness captured. The black vertical bars and their intersection with the y-axis curves denote the hypothetical implication in terms of species richness of dropping from 100 to 50 data points per image. Curves were calculated using the *rarecurve()* function in *R*. The *R* code for these analyses can be found [here](#).

As visible in **Figure 5** Row 1, there would be very little loss of species richness if we switched from annotating one image every meter, to instead one image every other meter. Likewise, **Figure 5** Row 2 indicates that very little loss of species richness captured would be incurred if we switched from annotating 100 data points per image to instead annotating 50 data points. Following these results, we have adjusted our AI training regime, switching from annotating 10,000 data points per a 100m transect to 2,500 data points per a 100m transect–this yielded a x4 increase in the rate at which we clear (or annotate) our ROV survey imagery, while still extracting large amounts of information from the benthos.

Note that as the ROV "flies above" the seafloor taking downward- and forward-facing imagery, we are not able to see underneath, e.g., understory algae, as to infer seafloor type, or to see cryptic taxa. For example, the ROV (and therefore our AI analyses) cannot see a kelp crab underneath a blade of sugar kelp, which a diver, searching through the understory algae, would come across. This is a known limitation of our ROV surveys, and this is part of our motivation for comparing ROV and diver data.

However, regarding the inability to see substrate underneath understory algae—given that a better understanding of fine-scale substrate variation is a key question of interest, we have implemented a two-phase survey approach for our Urban Kelp Research Project with the Port of Seattle. Specifically, we first survey in the winter (January and February) when the water is clear and the annual vegetation (algae/kelp) is absent. This allows us to comprehensively film the underlying substrate. Then, in the summer (late August and September), when the annual algae is present and when the summer plankton blooms have subsided, we conduct our summer surveys. Given our ROV's navigation system, we can return to the same precise locations surveyed previously, thus allowing us to combine data afterwards as to overlay our summer community structure data atop the underlying substrate survey in the winter.

For more information pertaining to our field and analysis protocols, the following SOPs can be found in **Appendices**:

- **Appendix A. SOP for ROV navigation and survey flights.**
- **Appendix B. SOP for camera settings and imagery processing.**
- **Appendix C. SOP to train CoralNet and extract data.**
- **Appendix D. SOP to conduct ROV-scuba diver methods overlap.**
- **Appendix E. SOP for PSRF ROV monitoring**

6.3 Acceptance criteria for quality of existing data

As part of expansion of a bull kelp habitat suitability model, we will utilize some of our existing ROV data from the Urban Kelp Research Project with the Port of Seattle. Specifically, we will use some of the data from the x8 ROV survey sites (with x4 transects at each site) that we have gathered in the course of our annual surveys summer 2022 and 2023. Please see **10.0 Quality**

Control Procedures for our protocol to maintain integrity of the imagery and derived data, including Quality Assurance / Quality Control QAQC procedures. For example, we only conduct our ROV surveys if the water visibility is more than 3m, and once the imagery is gathered, we review it immediately; if it is found to contain excessive particulates, or excessive shimmer or hazing due to, e.g., halocline or thermocline layers, we discard the imagery and repeat the survey until the acceptance criteria are achieved.

6.4 Model quality objectives

Model quality objectives from our expansion of the bull kelp habitat suitability model consist of the following:

- The environmental data layers we incorporate should be as up to date or newer than the layers that comprised the original model.
- Environmental model layers can be toggled on and off in a real-time map viewer in order to evaluate their spatial extent and underlying patterns.
- The effects upon the overarching model i.e. the suitability of specific locations for bull kelp ought to also refresh and change as individual environmental layers are toggled on and off. The current model we are expanding upon only displays a single, static output that is the result of the Weighted Overlay sum of all environmental layers—the relative contribution of the various input layers cannot be visually assessed in the current model formulation.
- The Benthic Terrain Modeling (BTM) tool will enable us to calculate seafloor characteristics from existing bathymetry data such as slope, aspect, and rugosity.
- Use of the BTM tool will further enable us to look at the relationships between our fine-scale substrate type data from the ROV surveys and the output from the BTM analysis of bathymetry data as to create a new layer containing extrapolated percent-cover values for the broader Elliott Bay region.
- Our primary response variable—bull kelp canopy extent—will rely upon the most up to date information possible, including bull kelp canopy data to be gathered by DNR summer 2024 throughout Elliott Bay via drone. We are collaborating with DNR personnel led by Helen Berry to coordinate our respective efforts.

7.0 Study Design

7.1 Study boundaries

The ROV surveys and ROV-diver method comparisons conducted for this project will encompass three sites within Elliott Bay, two sites on San Juan Island, and one site near Anacortes, Washington. **Figure 1** in **3.2 Study area and surroundings** details of overarching spatial extent of our study, and **Figure 3** in **6.2.2.1 Comparability** presents a finer spatial scale of ROV transects by displaying our ROV surveys along Centennial Park for the Port of Seattle from the summer 2022 field season.

Regarding our expansion of the bull kelp habitat suitability model, please see **7.3.1 Analytical framework** for a complete description of our approach and the layers.

7.2 Field data collection

7.2.1 Sampling locations and frequency

In the context of sampling design, our ROV-scuba methods comparison can best be thought of as stratified random, with the known differences in substrate type providing the stratified parameter that varies locally (e.g., soft-sediment at Centennial Park vs riprap along the Elliott Bay Marina breakwater vs basalt and limestone along the west end of San Juan Island). However, we will differ slightly from the traditional structure of stratified random in that the number of replicate transects (surveys) will be held fixed across all strata (substrate types). The transects will be randomly placed along the seafloor at a midpoint along the fringing bull kelp beds (approximately 7m depth). We will sample a minimum of x3 transects per site, with each transect spaced at minimum 5m from the others as to provide statistical independence.

Note that the spatial scale of inference for the ROV-scuba methods comparison is the individual 30m transect: we seek to differentiate whether the data derived by the ROV from one 30m transect differs from the data gathered by scuba divers *along that same 30m transect* (and see **14.5 Data analysis and presentation** for details about the specific statistical analyses we will use given this sampling design). Therefore, individual transects themselves represent the scale of replication, and x3 transects is the minimum that we seek to complete. Given the conditions encountered in the field and the constraints of subtidal field operations, we will attempt to complete additional transects, e.g., x6, if possible. With transect as our scale our replication, we do not seek to differentiate one area from another (e.g., we are not comparing the whole of Elliott Bay Marina breakwater to Centennial Park, which would require replication at, e.g., the site level). Rather, our individual transect-level comparison at one location can instead then be compared and contrasted with the transect-level comparison at other locations.

In terms of timing, the ROV-diver methods comparison will take place at all sites first in August-September 2024 (summer), then again January/February 2025 (winter). We will then repeat these surveys summer 2025 and winter 2026. We are coordinating with Reef Check personnel to train our Seattle Aquarium Scientific Divers, as well as line up the vessel, diver, and ROV logistics necessary to execute these field operations in Elliott Bay as well as along the west end of San Juan Island. We are presently scheduled to first accompany Reef Check divers on their trip to the west end of San Juan Island this September. Please see **Appendix D. SOP to conduct ROV-diver methods overlap** for the complete sequencing of how the ROV-diver methods comparison will take place.

Regarding the ROV site surveys alongside PSRF as part of their bull kelp restoration work, those surveys can be split into two components: a restored vs native site comparison, and the routine monitoring of the outplanted bull kelp. We address the former below, and the latter in the next paragraph. For the restored vs native site comparison, our objective is to differentiate local scale factors (differences in substrate type and benthic community structure) between the two sites. These site surveys will employ the two-phase winter and summer survey modality described above as to ensure we thoroughly capture the underlying seafloor. That is to say, we will first survey mid-March 2024 when the underlying vegetation is still absent and the seafloor is exposed. We will again survey in June at the conclusion of PSRF's routine monitoring of the site.

In terms of study design, we will utilize the same approach we have for the Urban Kelp Research Project. If we seek to differentiate two locations from one another statistically, we require replication at the site level. Therefore, we will sample x3 sites (with 50m minimum spacing between the sites) from one another at Jefferson Head, as well as x3 sites along Centennial Park. Each site will consist of x4 50m transects.

For the routine monitoring, we will use the ROV to supplement the already planned PSRF diver monitoring of their outplanted bull kelp. We will specifically survey on the "off weeks" when PSRF does not conduct their diver surveys. These ROV surveys will consist of us approaching the outplanted bull kelp, and flying the ROV laterally such that we film the entirety of the outplanted bull kelp forest.

7.2.2 Field parameters and laboratory analyses to be measured

The ROV surveys will gather photo/video imagery from the downward- and forward-facing cameras. From those imagery (the divers will also gather abundance and percent coverage counts by manually counting those metrics – see Appendix F) we will gather the following types of community and ROV sensor and telemetry data:

- Abundances of mobile and individually conspicuous invertebrates such as sea stars, solitary anemones, sea urchins, and sea cucumbers, as well as individually conspicuous algae such as bull kelp stipes.
- Percent-coverage estimates for substrate type (sand, shell debris, hard substrate) as well as colonial/aggregate taxa such as colonial tunicates and sponges and fleshy red, green, and some brown algae.
- In the course of our ROV operations we will also gather a suite of data electronically and automatically (at 1Hz, i.e., one data point per second) such as the ROV's altitude above

the seafloor, latitude and longitude coordinates for the ROV at all times underwater, depth, power levels, etc.

7.3 Modeling and analysis design

7.3.1 Analytical framework

Habitat suitability models (HSMs) provide a framework to predict species distribution given observed relationships with environmental variables (Havron et al., 2017; Rowden et al., 2017). Our initiative builds upon a HSM developed during the University of Washington GIS Certification Program in partnership with Puget Sound Restoration Fund by Gray McKenna, Victoria Fox, Avery Wolf, and Charlotte Ainsworth. First published June 1st (linked here), the *Bull Kelp Restoration Site Selection Tool* involved defining suitability ranges for input variables crucial to bull kelp growth, survival, and reproduction (**Tables 1** and **6**). These ranges facilitated the reclassification of input rasters onto a standardized scale (0-9). Each input received relative weights informed by the scientific literature, and a weighted overlay method was employed to generate a suitability model output, assigning a suitability "score" to each cell based on the inputs (**Figure 6**). At a high level, our expanded model will incorporate additional input variables (**Table 7**) absent in the previous version, encompassing environmental factors (e.g., sediment, chlorophyll levels, future sea temperature predictions) and taxonomic considerations, including understory algal species, benthic fauna, fish, and substrate type (e.g., sand/silt, pebble, cobble, boulder) at a finer-scale resolution (< 10m).

Figure 6. Predicted habitat suitability from current HSM formulation

(Left): Bull kelp habitat suitability throughout Puget Sound from the current instantiation of the model, with green shoreline representing good habitat, and progressively yellow, orange, and red shoreline representing poor habitat. (Right): Elliott Bay, virtually uniformly predicted to be favorable for bull kelp restoration. Bull kelp restoration failed to thrive in Smith Cove (red dot), despite favorable habitat predicted by the HSM. Our ROV surveys throughout Elliott Bay have documented substantial fine-scale variation in substrate type and benthic community structure that may contribute to variable bull kelp population outcomes.

However, subsequent to the model's development, a bull kelp outplanting endeavor in Smith Cove, Elliott Bay, an area deemed suitable by the model, proved unsuccessful. *In situ* observations at the time suggested that bull kelp herbivory by kelp crabs, as well as competition from other algae such as understory red and brown algae may have contributed. However, bull kelp thrives in other locations throughout Elliott Bay, including less than 1km away along the Elliott Bay Marina breakwater and offshore of Magnolia, as our ROV surveys have documented. These variable outcomes in bull kelp population success throughout Elliott Bay indicate that our understanding about the local scale drivers of bull kelp dynamics (e.g., substrate preferences, herbivory, competition) are incomplete, and accordingly, the current instantiation of the bull kelp habitat suitability model is likewise incomplete. Our core objective therefore in updating and expanding the existing HSM is to improve fine-scale predicted habitat suitability in order to improve accuracy and further guide bull kelp conservation and restoration.

To expand upon the HSM, our initial phase involves acquiring project package files from the developers of the bull kelp HSM and recreating the model ourselves. The original model's creators conducted a literature review, pinpointing environmental factors affecting bull kelp distribution, persistence, growth, and reproduction (**Tables 1** and **6**). An initial review by us found more current and/or higher resolution data that may be able to increase the effectiveness of the model (**Table 7**).

The new input variables for the HSM will be selected based on environmental factors influencing bull kelp distribution, persistence, growth, and reproduction. An extensive literature review and data search will be conducted to identify more recent and higher-resolution data sources (e.g., the 2017 Salish Sea Model and ~1m bathymetric resolution in Elliott Bay) relative to those used in the 2022 HSM. Additionally, any data omitted from the 2022 model due to time constraints and data availability during the class will be incorporated, particularly those that may enable relationships or correlations with our ROV-derived data (e.g., information about substrate slope, aspect, and rugosity).

The model will be constructed utilizing the Suitability Modeler (linked here) within ArcGIS Pro. Developing habitat suitability models is an iterative process, and the Suitability Modeler provides analytical feedback via interactive maps, plots, and panes. This empowers us to make informed decisions regarding specified parameters and visualize their impact on the final output. Additionally, we will use the Benthic Terrain Modeler (BTM) ArcGIS extension (linked here) to classify the benthic environment. This involves extracting bathymetric layers from NCEI's Bathymetric Data Viewer. Grids resulting from the BTM will represent bathymetric position index (BPI) and standardized BPIs, and slope and terrain ruggedness. Once ROV-derived data regarding substrate type (e.g., sand, shell-debris, cobble, hard substrate) have been incorporated, we can calculate the correlations between our fine-scale substrate characteristics and those derived from the BTM, thereby enabling our fine-scale characteristics to be extrapolated and forecasted across the larger study area.

Various raster interpolation techniques, such as Spline and Kriging, will be generated from the input variable layers. These interpolations will then be compared to choose the best interpolation method for the model. Input rasters will then be reclassified to a common scale, 1-9, assigning lower values to variable ranges that are less suitable for kelp and higher values to variable ranges more suitable for kelp. As with the original HSM, these assignments will be based upon the most up-to-date published scientific literature. Finally, reclassified raster layers will be combined using the Weighted Overlay (linked here) tool which multiplies each cell in the input layer by its assigned relative value. Each input raster will be weighted according to its importance on bull kelp bull growth, survival, and reproduction.

For the calibration of our HSM, we will assess the values of the output cells derived from the reclassified raster against the bull kelp presence data from the 1994-2000 DNR ShoreZone dataset. Specifically, we will compare the mean cell value for regions within the bull kelp distribution in the dataset to the mean cell value obtained from our model output. In the event that a more recent or complete data set of bull kelp canopy coverage becomes available—which in collaboration with DNR we suspect may be the case—we will compare our model output to those data.

Table 6. Original layers comprising the bull kelp HSM

Variable	Input Layer	Relationship with Suitability	Data Source
Temperature	Mean Daily Maximum Summer (Jun - Sept) Surface Temperature (°C)	Negative	Salish Sea Model 2014 Hydrodynamic Solution (Khangaonkar et al., 2018; Premathilake & Khangaonkar, 2022)
Current Speed	Mean Daily Maximum Current Speed (m/s)	Positive	
Salinity	Mean Daily Minimum Winter (Nov - April) Salinity (ppt)	Positive	
Substrate	Substrate Classification	Categorical	ShoreZone Inventory (Washington Department of Natural Resources 2000, Dethier 2014)
Light	Mean Daily Maximum Summer Light Transmission (%) within 1m of Surface	Positive	Washington State Department of Ecology Marine Water Column Monitoring, 2018

	Mean Daily Minimum Winter Light Transmission (%) at 10m	Positive	
Nutrients	Mean Summer Surface DIN (uM)	Positive	Washington State Department of Ecology Marine Nutrient Monitoring, 2018
Historic Kelp Presence	Qualified Kelp Presence 1911-1912	Positive	Cameron F. Potash from kelp. Washington, DC: US Govt. Print. Off.; 1915. Data digitized by Washington State Department of Natural Resources; described in Pfister et al. 2017
Depth	0 to -10m MLLW	Bounding	NOAA, 2014. Puget Sound 1/3 arc-second NAVD 88 Coastal Digital Elevation Model and NOAA, 2011. Port Townsend, Washington 1/3 Arc-second NAVD 88 Coastal Digital Elevation Model.

Table 7. Additional input layers for the expanded bull kelp habitat suitability model

Variable	Input Layer	Relationship with Suitability	Data Source
Temperature	Mean Daily Maximum Summer (Jun - Sept) Sea Surface Temperature (°C)	Negative	Salish Sea Model 2017 Hydrodynamic Solution, ~100m resolution (Unpublished, provided by T. Khangaonkar and N. Wenfei, January 2024)
	Mean Daily Maximum Summer (Jun - Sept) Seafloor Temperature (°C)		
Current Speed	Mean Daily Maximum Current Speed Sea Surface (m/s)	Positive	Salish Sea Model 2017 Hydrodynamic Solution, ~100m resolution (Unpublished, provided by T. Khangaonkar and N. Wenfei, January 2024)
	Mean Daily Maximum Current Speed Seafloor (m/s)		

Salinity	Mean Daily Minimum Winter (Nov - April) Salinity Sea Surface (ppt) Mean Daily Minimum Winter (Nov - April) Salinity Seafloor (ppt)	Positive	Salish Sea Model 2017 Hydrodynamic Solution, ~100m resolution (Unpublished, provided by T. Khangaonkar and N. Wenfei, January 2024)
Substrate	Substrate Classification	Categorical	NOAA National Ocean Service (NOS) Hydrographic Survey Bathymetric Attributed Grids (BAGs) in U.S. coastal waters. ~1m resolution in Elliott Bay, 2013.
Substrate complexity	Rugosity	Positive	Benthic Terrain Modeler output with NOAA National Ocean Service (NOS) Hydrographic Survey Bathymetric Attributed Grids (BAGs) in U.S. coastal waters. ~1m resolution in Elliott Bay, 2013.
Turbidity	Sedimentation	Negative	Washington State Department of Ecology's Environmental Information Management system.
Aggregate taxa	Percent cover	Correlations will be calculated from our ROV data	Image classification on CoralNet. Urban Kelp Research Project, Seattle Aquarium, 2022-2024

7.3.2 Model setup and data needs

Our expansion of the bull kelp HSM will involve both refreshing the older, existing environmental variables (**Table 6**) and incorporating new layers (**Table 7**). Please see the respective layers for information regarding year and grain size, where applicable.

Please see the text in **7.3.1 Analytical framework** for details about our modeling approach.

Furthermore, please see **13.4 Model quality assessment** and **13.4.1 Calibration and validation** for details about how we will simulate ROV data as well as the layer weightings to iteratively explore the model and its output, as well as revise and update as necessary, especially via through conversations with subject-matter experts with DFW, DNR, and HSIL.

7.4 Assumptions of study design

Our ROV-scuba methods comparison assumes that the underlying substrate and community structure varies across the geographical extent of our sites (e.g., comparing Elliott Bay to the San Juan Islands) such that we are able to gain a broader suite of inference as opposed to conducting the entire methods comparison in one location.

The suitability ranges to reclassify input rasters for input variables will be established according to insights obtained from the existing scientific literature. It is assumed that clear suitability ranges can be identified for each input variable used in the model, and that the ranges we ascribe to these environmental layers are reasonably reflective of how bull kelp responds to these specific environmental variables. However, given the ongoing nature of research and the continuous emergence of new findings, there is no guarantee that a definitive range will be available for every variable—this precise situation was encountered in the original formulation of the model. To mitigate this as much as possible, we will closely follow the scientific literature and we will maintain communication with key kelp personnel at DNR in order to maintain an iterative dialogue regarding our modeling decisions and assumptions.

The weighted overlay, employed to generate the habitat suitability model output, operates on the assumption that the ranges in the reclassified raster layers accurately depict the suitability for bull kelp.

The calibration method, relying on bull kelp presence data from 1994-2000, presupposes that areas suitable for bull kelp during that period remain equally suitable today. However, given the alterations in environmental conditions over the last two decades, areas considered suitable for bull kelp back then may not necessarily be suitable now. Therefore, our efforts will include obtaining more recent bull kelp presence data from DNR for improved accuracy, especially the upcoming effort in summer 2024 to obtain aerial imagery of all bull kelp throughout Elliott Bay via Unmanned Aerial Vehicles (UAVs, i.e., drones/quadcopters).

7.5 Possible challenges and contingencies

7.5.1 Logistical problems

Conducting subtidal operations is inherently challenging. Field days often have to be postponed due to inclement conditions such as wind (especially when conducting our winter ROV surveys in Puget Sound). Furthermore, visibility can be heavily impacted by summer algal blooms, though we target our ROV surveys during specific times of the year when the water is relatively clear (most anytime *other* than May, June, July, and early August). We also target flooding tides or slack high tides, as these are associated with the clearest water. However, even when the marine and topside conditions are favorable, there is inherent risk associated with subtidal operations, especially when operating electronics such as ROVs in salt water.

To mitigate this risk, we have x2 ROVs from Blue Robotics—ROV *Nereo* and ROV *Lutris*. They are configured identically, such that if one goes down for whatever reason, we have a backup that can be seamlessly rotated into the field. Furthermore, part of the appeal of the Blue Robotics ROV system is that any given component can be easily replaced. If, e.g., the electronics enclosure floods (which we have not experienced before), we can simply obtain a new ROV electronics enclosure for $1,400 (linked here). This modular approach provides an added degree of redundancy for our operations. Finally, in order to further mitigate risk, we regularly conduct "pressure tests" of the ROV's enclosed and sealed spaces. These pressure tests (via a vacuum pump) are a key early warning of any issues with the ROV's o-rings, and conducting this test proactively enables us to correct any problems.

Please also note that both ROV *Nereo* and ROV *Lutris*—and all constituent components and sensors—are listed on the Seattle Aquarium's insurance policy. So, in the (unlikely) event of a complete vehicle loss, we will be able to get back up and running relatively quickly.

7.5.2 Practical constraints

One constraint to our ROV surveys has been the duration that the ROV can stay underwater. For example, when the ROV runs off a 15V battery, it can stay underwater for about a hour when operating in less than 1kt of current. When operating in 3+ kts of current, we have to crank the thrusters up to 100%, which reduces our runtime to around 20min. In an effort to increase the ROV's survey duration we are reconfiguring the ROV to run off of "shore power", where the ROV is powered from the Seattle Aquarium's vessel. These modifications are scheduled to take place February 2024, and once complete, the Aquarium vessel will have a 200Ah lithium-ion battery, which translates to virtually an unlimited run time for our ROV during the course of a survey day.

Vessel availability is another potentially limiting factor regarding field operations. The Seattle Aquarium has a 24' aluminum Almar vessel, and PI Randell and Williams are now Primary Boat Operators (PBO) after taking a Motorboat Operator Training Course (MOTC). This project's research is a priority for our vessel, and we are on-track to complete all our objectives, especially for our weeklong survey efforts in Elliott Bay and in the San Juan Islands. We are also scheduled to regularly monitor PSRF bull kelp outplanting locations, and for those, there may be occasional instances where some of the other Seattle Aquarium research and conservation efforts require the vessel for a week (e.g., our Pinto Abalone outplanting efforts with WDFW and PSRF may require the vessel for work in the San Juan Islands). That all being said, PI Randell coordinates very closely with Joel Hollander, the Seattle Aquarium Dive Program Manager, to ensure vessel availability for this HSIL research.

7.5.3 Schedule limitations

There may occasionally be competing interests for use of the Seattle Aquarium vessel as discussed in the previous section, but we do not anticipate these will negatively impact our project.

8.0 Field Procedures

8.1 Invasive species evaluation

The ROV does not come in contact with the seafloor thus we anticipate there is very little to no chance that any invasive species could be spread in the course of our surveys. This is especially the case because we give our ROV an extensive freshwater rinse on the vessel at the conclusion of surveys for the day. No samples are gathered as part of this study.

8.2 Measurement and sampling procedures

Please see **Appendices A—E** for SOPs regarding our ROV tracking and survey protocols, photo/video processing, AI analyses, and the sequencing of our ROV-diver methods comparison.

8.3 Containers, preservation methods, holding times

Not Applicable: no water samples or any other samples will be collected as part of this project.

8.4 Equipment decontamination

Not Applicable: no laboratory equipment will be utilized as part of this project.

8.5 Sample ID

While we are not collecting biological samples, we have stringent protocols in place for the naming conventions of our photo/video data files. These conventions ensure that as soon as the imagery are brought off of a camera, they are labeled correctly with the following format: YYYY_MM_DD_hh-mm-ss, such as 2024_01_24_14-49-25, corresponds to 2:49pm on January 24th, 2024.

8.6 Chain of custody

We do not take any physical samples, but do gather extensive photo and video imagery as well as sensor telemetry. In terms of imagery chain of custody, we immediately upload our photos and videos from our GoPro cameras to DropBox cloud storage. All content on DropBox is also stored on a hard drive at the Seattle Aquarium. Likewise, all ROV telemetry is uploaded to the DropBox at the conclusion of each day in the field. Please see **11 Data Management Procedures** for more information about data handling and storage.

8.7 Field log requirements

We have multiple tracking documents associated with our ROV field operations. These include:

- A flight/float plan filed ahead of the field day's operations with the Seattle Aquarium's Dive Program Manager. The flight/float plan outlines the plan for the day, weather forecasts, personnel involved, their affiliation and contact information, estimated departure and return times, etc.
- During the course of ROV operations we also record any anomalies that are experienced regarding the ROV software and hardware. These are recorded on "write in the rain" notebooks in the field, and then are logged electronically in a DropBox excel file once we are out of the field.
- Once we are out of the field, we additionally log the high-level metadata of the day: date, site, # of transects completed, personnel present, water conditions, surface conditions, anomalies with the vessel, and anomalies with the ROV.

- We log vessel use each day including date, location, PBO, engine hours, and any issues.
- We also have a separate log for all video imagery that requires processing and we track the status of each video: need to extract stills, color-correct, etc.
- We also contain a log of every image that is ready for analysis in CoralNet. We track whether the image has been started, is in progress, or is complete.

8.8 Other activities

We have routine (every six months) maintenance activities that we complete for our ROV including o-ring inspection, lubrication, and replacement (if necessary). This also involves checking over numerous bolts and screws to make sure they have not become loose.

9.0 Laboratory Procedures

9.1 Lab procedures table

Not Applicable: there are no laboratory procedures associated with this project.

9.2 Sample preparation method(s)

Not Applicable.

9.3 Special method requirements

Not Applicable.

9.4 Laboratories accredited for methods

Not Applicable.

10.0 Quality Control Procedures

Quality Assurance / Quality Control (QAQC) procedures are vital to ensure data collection, entry, processing, analysis, and reporting follow a predetermined plan, with steps in place to capture and correct errors. Below we outline the QAQC steps in place for our ROV surveys and habitat suitability modeling. We do not detail Reef Check's QAQC for their diver surveys, as Reef Check maintains thorough QAQC processes that are detailed in their QAPP, signed October 12th, 2023. That being said, Seattle Aquarium Scientific Divers that collect Reef Check data will participate in the Reef Check Partner Retreat, as well as internal Seattle Aquarium Reef Check training, followed by certification by the Reef Check Washington Regional Manager, Selbitschka.

Regarding our ROV surveys, there are multiple steps throughout the process by which we evaluate imagery or data quality and modify our actions accordingly.

- Upon the day we survey, ROV surveys will not be undertaken if the water visibility is less than 3m, or if the particulate count is sufficiently high as to introduce untoward backscatter in our downward- or forward-facing ROV imagery.
- Immediately following the ROV surveys, we review the imagery, and if there is excessive blurring, particulates that interfere with our lighting or the camera's focus, or stratification in the water that introduces excessive "shimmer" (e.g., from halocline or thermocline, both of which are relatively common in Elliott Bay), we will opt to discard the imagery and redo the ROV survey another day.
- Furthermore, we likewise review the geospatial tracklines of the ROV's survey path. If they are found to be slightly erroneous, smoothing procedures in *R* (e.g., by applying a spline) will be implemented. If the tracklines produced by the Underwater GPS (UGPS) system are more than slightly erroneous (e.g., if they are in an entirely incorrect location), we revert to our fall-back system for spatial tracking, the Doppler Velocity Log (DVL), which exhibits highly precise spatial tracking. Please see **Appendix A. ROV navigation and survey flights** for the SOP detailing ROV navigation and flight operations.
- Once the ROV imagery has been collected, it is processed (e.g., color-corrected) using existing color-correction templates that have been reviewed for accuracy by subject-matter experts at the Seattle Aquarium that have extensive experience conducting subtidal operations.
- Once the imagery is processed, it is ready for manual annotation in our AI programs. Our AI teachers go through an extensive training process to identify substrate type and benthic organisms. Once they are familiar with the necessary taxa and substrate types, they undergo a standardization training process, where all AI Teachers annotate the same 10 images, with 100 uniformly distributed data points across each image (1000 points total training points). These uniformly distributed data points enable our CCR team to compare our annotations (our "ground truth") with the AI Teachers'. Deviations from the ground truth annotations are noted, and we have conversations with individual AI Teachers to resolve any confusion or questions.
- Once an AI Teacher is fully trained, we review every data point from every 10th image that they annotate. If we find more than 10% deviation from CCR personnel's annotations, we once more have a conversation to talk through problems and confusion.
- Zooming out, also note that as soon as photos/videos are offload from the ROV, they are renamed according to a specific naming convention—detailed in **8.5 Sample ID**—and these files retain that naming convention throughout the entirety of our analytical pipeline, such that the .csv file resulting from our AI analyses contains a column that lists the individual file name for each still image that has been extracted from the videos. For example, see column "AD" in this .csv file—each row of data in our final output can be traced to the precise day—even to the precise second—of imagery that generated the resulting data. Therefore, if any questions arise, we can trace the entirety of the imagery/data pathway and correct the error.

- Once the .csv file from CoralNet is generated, CCR personnel review each data file to ensure that the information aligns with expectations. This can involve, for example, pulling up imagery from that day's survey to ensure that the date and time stamps align properly, and that substrate type and benthic community structure in the data are reflective of the imagery captured by the ROV.

For our habitat suitability model, we will maintain strict version control, such that changes in ArcGIS, *R*, and Python can be rolled back, if necessary. Each individual environmental data layer will be scrutinized to ensure it is accurate and appropriate. As described in **13.4 Model quality assessment**, we will add layers one at a time to evaluate model sensitivity to new environmental variables. Likewise, we will extensively simulate our ROV benthic data to understand model behavior in response to these new data. Adding layers individually along with simulating ROV data will enable us to evaluate any errors in model formulation or performance.

Furthermore, we will maintain communication with state and federal collaborators that are experienced with similar spatial methods such as Bob Pacunski with WDFW and Blake Feist with NOAA National Marine Fisheries Service. By sharing methods and results with them, as well as with our kelp collaborators at DNR, we will adjust our approach in an iterative fashion if and when any issues arise.

10.1 Table of field and laboratory quality control

Not Applicable.

10.2 Corrective action processes

Not Applicable – please see our responses above in **10.0 Quality Control Procedures.**

11.0 Data Management Procedures

11.1 Data recording and reporting requirements

All ROV data (ROV sensor and telemetry files as well photo/video imagery) are retrieved immediately following each ROV field day. The data are directly transferred to the Seattle Aquarium's DropBox account (described below in **11.3 Electronic transfer requirements**). The Reef Check diver obtained data will be treated according to Reef Check standards, which are documented in the recent Reef Check QAPP signed October 12th, 2023.

11.2 Laboratory data package requirements

Not applicable.

11.3 Electronic transfer requirements

All electronic information (such as photo/video imagery and derived benthic data) will be stored on a secure DropBox account maintained by the Seattle Aquarium. Randell, Williams, and Bogdanova all have licenses to this account and thus have read/write/delete privileges. Any of the content on DropBox can be shared, viewed, and downloaded via links like this, and anyone can download the content at such links. However, the content at the link cannot be edited by anyone except the three Seattle Aquarium staff listed above.

11.4 Data upload procedures

It is unclear to us at this time whether or not our benthic community structure data need to be uploaded to the EPA's WQX portal. However, in reviewing Reef Check's QAPP (signed October 12th, 2023), they put "Not Applicable", and they gather similar benthic community data. All compatible data collected for this project will be uploaded to EPA's Water Quality Exchange (WQX). Otherwise, until a definitive verdict has been returned, we will upload and store the data as described above in **11.3 Electronic transfer requirements**.

11.5 Model information management

All modeling information will be managed just as all our other data, i.e., on the Seattle Aquarium's DropBox. The volume of certain data layers is anticipated to be large. For example, the Salish Sea Model output file size is ~3GB for 1 day, hence the need to focus the geographic area of that model to our specific area of interest. That being said, the subscription we have with DropBox is for unlimited storage.

In terms of version control, we use GitHub, which is the standard for programmers to maintain tight and transparent version control. All our code and much project information can be found at our two GitHub repositories that detail our CCR work in the public domain, linked here and here. GitHub is also now our standard for making data publicly available, as you can see here, a GitHub repository for a recent Seattle Aquarium scientific publication.

12.0 Audits and Reports

12.1 Audits

Audits are not anticipated for this project. We will regularly update HSIL via quarterly progress reports, and we are in routine contact with key kelp personnel such as Elizabeth Spaulding and Helen Berry at DNR.

12.2 Responsible personnel

PI Randell will be responsible for all Quarterly Reports and deliverables to HSIL.

12.3 Frequency and distribution of reports

The Quarterly Reports will be completed and submitted in the SmartSimple system based on the HSIL reporting requirements. That is to say, the reports will be submitted on January 10th, April 10th, June 10th, and October 10th.

12.4 Responsibility for reports

PI Randell will be responsible for the final report and he will be assisted by Research Technician's Williams and Bogdanova.

13.0 Data Verification

13.1 Field data verification, requirements, and responsibilities

Research Technician Williams has developed a training and standardization program for our AI analyses of ROV survey imagery (**Figure 7**). Specifically, she developed a training pipeline where our Seattle Aquarium "AI Teacher" volunteers are taught to identify the 66 percent-cover categories of substrate, algae, invertebrates we identify from the downward-facing camera in CoralNet. This involves classroom sessions as well as at-home work with a species identification guide. Williams has recorded some of these training sessions so that they can be viewed repeatedly, if required.

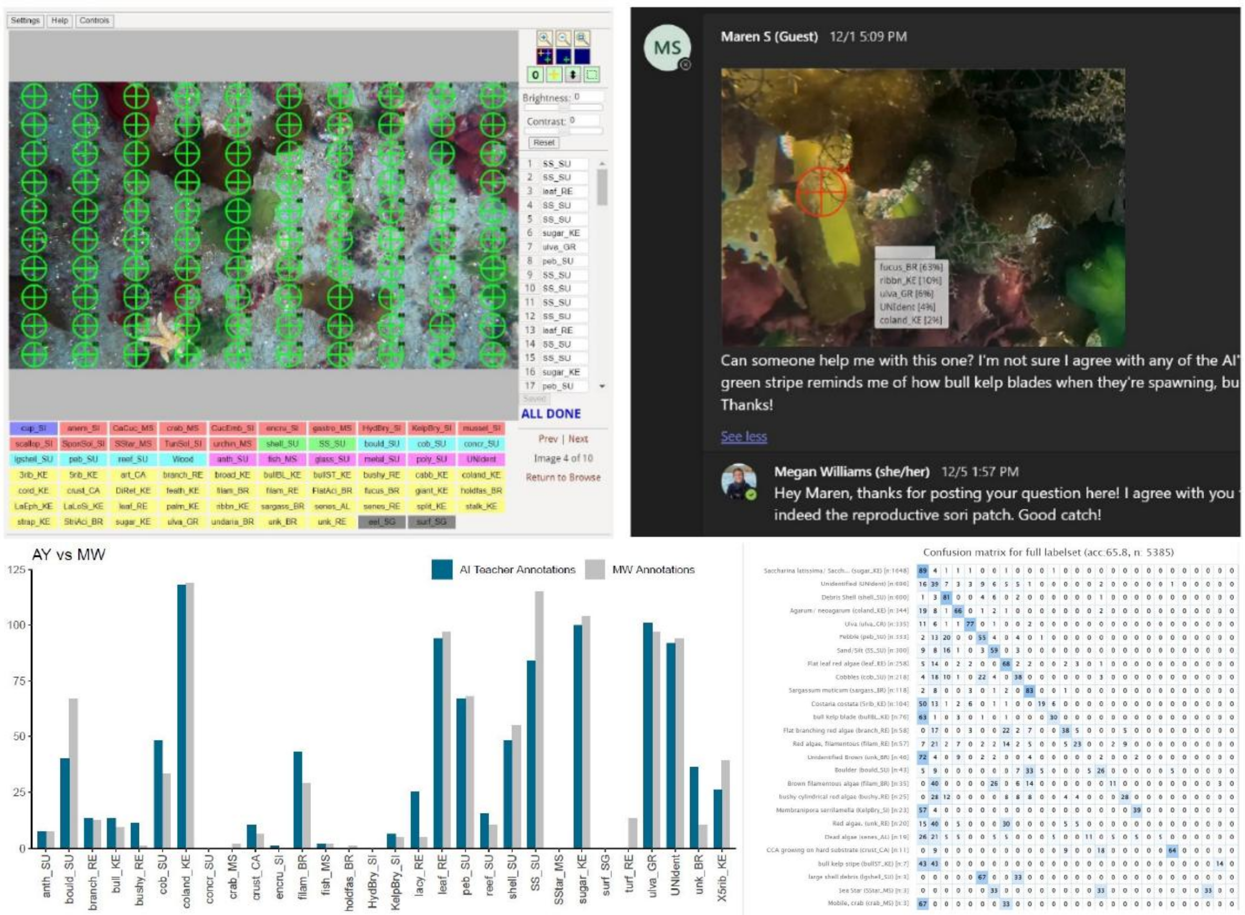

Figure 7. AI Teacher training process

Row 1. (Left) a training image with 100 points uniformly distributed that all AI Teachers annotate. (Right) our Microsoft Teams channel enabling questions to be answered. Row 2. (Left): an AI Teacher's training annotations (in blue) versus our ground truth annotations (in gray), allowing rapid identification of error. (Right) our CoralNet algorithm's confusion matrix. The diagonal denotes the probability of the algorithm accurately identifying specific percent-cover categories.

Furthermore, once our AI Teacher volunteers are literate in the 66 percent-cover categories, as well as the CoralNet program they will interface with, they then go through a standardization process. Specifically, each AI Teacher "annotates" (labels a data point with the specific category underlying the data point) 10 images, each containing 100 data points, 1000 data points total (see, e.g., **Figure 7** row 1, left). These data points are uniformly distributed across the image (in contrast to the random distribution of our "real" data points), enabling all AI Teachers to train on the same subset of data points. Once the training set is complete, Williams analyses the data and compares the AI Teacher annotations to her own via a *R* script, allowing her to view which categories were missed by the AI Teacher, or mislabeled (**Figure 7**, row 2, left). This enables us to identify any systematic errors or misunderstandings about the percent-cover categories.

Once this training procedure is complete, the AI Teacher graduates to labeling "real" ROV data from the field. However, again, Williams monitors the annotated data by reviewing all data points across every 10^{th} image to identify any trends in mislabeled data. If more than 10% of data are found to be mislabeled, we correct the image labels and have a conversation with the AI Teacher to help them understand correct labeling / identification.

Furthermore, note that we maintain a Microsoft Teams channel dedicated to our ROV methods development and AI training (**Figure 6** row 1, right). All of our Seattle Aquarium staff, volunteers, and Special Projects Volunteers are on this Teams channel, and this provides a communication platform through which AI Teachers can contact us with specific questions and obtain rapid feedback.

Note that our CoralNet AI algorithm has been trained on over 800 images, with over 80,000 data points generated across eight Urban Kelp ROV sites thus far. The AI algorithm is making predictions, thus our AI Teachers are now manually reviewing the algorithm's predictions by marking each point as "verified", if labeled correctly, or by providing the correct label if the algorithm's guess was incorrect. As noted previously, certain common labels are being predicted with relatively high percent accuracies (e.g., sugar kelp is being identified correctly 89% of the time, **Figure 6**, row 2, right).

13.2 Laboratory data verification

Not applicable.

13.3 Validation requirements, if necessary

Not applicable

13.4 Model quality assessment

The GIS analyses associated with this project will be completed iteratively and we will routinely share our progress and updates with HSIL as well as subject-matter experts involved in habitat suitability modeling with DFW and DNR as to solicit feedback and avenues to improve our model performance and approach.

Furthermore, prior to incorporating real ROV-derived abundance and percent-coverage data into the existing habitat suitability model, we will experiment extensively with the model via simulated data. Specifically, we will use the programming language *R* to simulate columns (the various percent-cover categories such as substrate type and red and brown algae, and abundances of conspicuous species such as sea urchins, sea stars, bull kelp stipes, and fishes, etc.) and rows, with each row corresponding to a (simulated) image gathered by the ROV. We will simulate the precise data structure of our real Urban Kelp Research Project data with the Port of Seattle that we have extracted from CoralNet and combined with metadata. That is to say, we will simulate the actual transect length i.e. the number of images sampled per transect (the number of rows) as well as the actual number of transects per site (x4 per site, with x8 sites total). These simulations will allow us to have full control over the underlying patterns comprising the ROV data, thus allowing us to experimentally evaluate model performance and output prior to incorporating "real" data.

For example, once the various environmental model layers are updated and behaving as expected, we will incorporate randomly distributed ROV data (e.g., uniformly distributed) such that there is no signal, or substantial underlying pattern present. Our prediction would be that these ROV data would not substantially affect model output. Next, we will simulate certain anticipated patterns, such as relatively high bull kelp abundances along with high percent-cover counts of hard substrate. We would anticipate seeing these patterns of association influence model output positively with respect to predicted bull kelp habitation. We return to our use of data simulation below in **13.4.2 Analysis of sensitivity and uncertainty.**

Fast forward, once these simulation-based investigations have been completed and we are confident that we understand how our model behaves in response to ROV-derived data, we will switch from simulated to real ROV-derived estimates of percent-coverages and abundances.

13.4.1 Calibration and validation

Please see **7.3 Modeling design and analysis** and **13.4 Model quality assessment** for the bulk of our narrative regarding calibration and validation. We will further note here however that each environmental data layer listed in **Tables 6** and **7** will be added one at a time, such that we can better understand the relative contribution of each to model output. We likewise will be able to "toggle" these layers on and off in the output product, such that we can overlay the layer's patterns, as well as the output generated as a product of the inclusion or exclusion of the environmental model in question (also described in **13.4.2 Analysis of sensitivity and uncertainty**).

13.4.1.1 Precision

Not applicable.

13.4.1.2 Bias

Not applicable.

13.4.1.3 Representativeness

Not applicable.

13.4.1.4 Qualitative assessment

While not specifically in scope for this project, the Seattle Aquarium is committed to advancing our CCR research as part of a long-term monitoring and research program. We therefore are committed to using the finer-scale predictions of our expanded bull kelp habitat suitability model to inform and guide future ROV surveys, even if those take place outside the scope of this HSIL project.

13.4.2 Analysis of sensitivity and uncertainty

The first step of our bull kelp habitat suitability model expansion is to update the various environmental layers in the existing model, as in certain cases newer data are available. Furthermore, we will add additional layers that were not a part of the original model. As part of this process, we will add the ability to toggle various layers “on” and “off”, as to perform a sensitivity analysis to better understand the relative effects of individual layers (environmental variables such as temperature, nutrients, current) upon model behavior and predicted outputs. We will carefully record (via saving model output in our DropBox) how model performance and predictions vary as individual layers are added.

Once the layers are updated and model behavior is better understood, we will establish a pipeline to incorporate ROV-derived percent-cover and abundance data into the model. As noted above in **13.4.1 Calibration and validation**, we will first incorporate simulated ROV data. These simulations will provide powerful insight into the sensitivity of the model to various underlying patterns in the ROV data. Flipping the data simulation around, we will also hold the input data constant, and instead simulate with various instantiations of the weightings attributed to the environmental and ROV layers. Modifying these weightings in an experimental fashion will provide a better sense of model sensitivity and behavior, and we can use these trials to tune our approach.

14.0 Data Quality (Usability) Assessment

14.1 Process for determining project objectives were met

The processes for determining whether project objectives are met when we:

- Successfully complete ROV field work when intended as well as properly storing, processing, and color correcting the ROV survey imagery.
- Successfully annotate all ROV field images as to further train our AI algorithms.
- Successfully update HSB model layers as well as incorporate simulated and real ROV-derived data into the HSB.
- Successfully derive finer-scale predictions about what locations contain the optimal substrate, benthic community, and environmental conditions to support persistent bull kelp populations.

14.2 Treatment of non-detects

Not applicable. Please see **6.2.2.2 Representativeness** and **6.2.2.3 Completeness** for discussions about what our ROV surveys can and cannot capture, and how we are structure our sampling design and methodology accordingly.

14.3 Data analysis and presentation methods

Aside from the analyses that need to place using ArcGIS (such as the HSB), all statistical analyses will take place using *R*. Certain ROV telemetry file analyses will take place in Python. Please note that all code used as part of this and any other CCR project will be publicly available (e.g., see here for *R* and Python code associated with our research).

- In order to compare the ROV's abundance data to Reef Check's abundance data we can utilize a variety of approaches. For example, to make a pair-wise comparison (e.g., is the number of crabs the ROV saw significantly different than the number of crabs divers recorded along the same transect?), we will use a Wilcoxon signed-rank test (Rosner et al., 2006). This is a non-parametric test (an analog of the t-test) that will be utilized due to the anticipated differences in sample size between the ROV- and diver-data, which precludes the use of more standard (parametric) methods. Additionally, because we are analyzing two groups from the same transect (ROV and diver data), the ROV and diver data will not be independent, again violating standard assumptions of parametric methods and thus necessitating the use of a non-parametric approach.
- Comparing ROV's percent-coverage data to Reef Check's percent-coverage UPC data. This will be slightly more complex, though the solution is largely the same as comparing abundances above. Because the ROV gathers imagery and we annotate those points in CoralNet, with 30 photos and 50 data points per photo, we will have 1500 percent-cover data points relative to Reef Check's 90 data points across a 30m transect. Therefore, due to the substantial differences in sample size, we will compare proportions, i.e., the

proportional representation of red algae captured by the ROV vs the diver along each individual transect. This once more can be achieved by the non-parametric Wilcoxon signed-rank test.

- Furthermore, if we want to compare ROV-diver data in a more holistic modeling framework, e.g., to compare ROV-diver counts across different sites representing different benthic substrate environments as to account for the effects of the different sites, we could use a linear mixed model framework, with survey type (ROV and diver) as a fixed effect, and site and transect as random effects. Similar designs have been conducted comparing ROVs and divers (Boavida et al., 2016; Maslin et al., 2021; Wetz et al., 2020).
- To compare our counts of bull kelp stipes along the PSRF outplanted location at Jefferson Head to the PSRF's diver counts from the same area, we can once more use the Wilcoxon signed-rank non-parametric approach, in the same manner and for the same reasons described above in the first bullet point of this section.
- To compare benthic and community structure between PSRF's bull kelp enhancement site vs a control site, we will once more use a non-parametric approach. Specifically, we will use non-metric multidimensional scaling (NMDS), a standard approach for analyzing high-dimensional community structure data, and then we will use PERMANOVA to test for differences in "centroid positioning", i.e., differences in the mean of the two groups, i.e., two sites (Larson et al., 2022; Randell et al., 2022).

14.4 Sampling design evaluation

Not applicable.

14.5 Documentation of assessment

Data usability will be documented through our AI Teacher QA/QC process.

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Hyperlinks embedded in the text:

- GitHub repository containing information about our Coastal Climate Resilience (CCR) research: https://github.com/zhrandell/Seattle_Aquarium_ROV_development
- Northwest Straits Commission Kelp Plan landing page: https://nwstraits.org/our-work/kelp/
- WA Floating Kelp Indicator landing page: https://kelp-canopy-vital-sign-for-puget-sound-wadnr.hub.arcgis.com/pages/Indicator%20Creation
- WA kelp working groups: https://view.officeapps.live.com/op/view.aspx?src=https%3A%2F%2Fwww.nwstraits.org%2Fmedia%2F3470%2Fpuget-sound-kelp-project-inventory_2020-2022.xlsx&wdOrigin=BROWSELINK
- Northwest Straits Commission kelp policy recommendations: https://nwstraits.org/media/3514/kelp-policy-advisory-group-recommendations.pdf
- Senate Bill 5619 for kelp conservation and recovery: https://app.leg.wa.gov/billsummary?BillNumber=5619&Year=2021
- Kelp Plan 2023 Status Update: https://nwstraits.org/media/3483/kelp-plan-status-update_2023oct.pdf
- PISCO subtidal field protocols: https://piscoweb.org/kelp-forest-sampling-protocols
- Reef Check Kelp Forest Program: https://www.reefcheck.org/kelp-forest-program/
- Reef Check map of Washington survey sites: https://www.google.com/maps/d/u/0/viewer?ll=47.98706287825558%2C-122.34608633188746&z=9&mid=1WN7Eo4l8bRnMdJsHsiDKi3-qQ-x5sjI
- Blue Robotics' BlueROV2: https://bluerobotics.com/store/rov/bluerov2/
- Coastal Climate Resilience (CCR) research on GitHub: https://github.com/zhrandell/Seattle_Aquarium_ROV_development
- Tether de-tangling from bull kelp: https://drive.google.com/file/d/1a-SRlrAFt43HIkNrAGRNxlSQBMEUnt3K/view
- Another instance of de-tangling tether from bull kelp: https://drive.google.com/file/d/1qdtdPwB8RHkjL27RRzu95MTenNgszDAC/view
- Side-by-side video from the ROV: https://www.dropbox.com/s/ri6zqosdktrsbpj/Pier62_sidebyside.mp4?dl=0
- 2023 year-end report to the Port of Seattle: https://www.dropbox.com/scl/fi/r9h8rwams3n2h39q5hpib/2023_YearEnd_Report_Urban-Kelp-Research-Project.pdf?rlkey=35ua56ufnezqcyuomed3wgnep&dl=0
- CoralNet, an open-source AI program: https://coralnet.ucsd.edu/

- Northwest Straits Commission kayak survey protocol: https://www.nwstraits.org/media/3380/mrc-kelpkayaksurveyprotocol-2023update.pdf
- Kelp forest surveys along Washington's Strait: https://wadnr.maps.arcgis.com/apps/MapSeries/index.html?appid=6b32b37740a443cb8e8848a8614879a2
- Urban Kelp research in the Seattle Times: https://www.seattletimes.com/sponsored/seattle-partnership-dives-into-the-mysteries-of-elliott-bays-bull-kelp/
- Urban Kelp research on YouTube: https://www.youtube.com/watch?app=desktop&v=WdsnsdwxIYs
- Information about VIAME: https://www.star.nesdis.noaa.gov/star/documents/meetings/2020AI/presentations/202008/20200827_Dawkins.pdf
- ROV data associated with our Urban Kelp research: https://www.dropbox.com/scl/fi/gojf42mdtod3omq61smvu/species_richness_curves.csv?rlkey=q03kl78df4537gt9y0ykin16o&dl=0
- Signed contract with the HSIL: https://www.dropbox.com/scl/fi/fsd1ykos6eqvadcltc5h8/23-23658-Agreement-combined-compressed.pdf?rlkey=ikxx4rzdkq883y6uj0znqt9u1&dl=0
- PI Randell's Ph.D. public dissertation defense: https://www.youtube.com/watch?v=PpVOXf6_830&t=198s
- PI Randell's Ph.D. dissertation: https://www.researchgate.net/publication/358729664_Grazer-resource_behavior_interactions_and_feedbacks_control_kelp-forest_dynamics_and_stability
- University of Washington Geographic Information Systems credential: https://www.pce.uw.edu/certificates/geographic-information-systems
- Calculate area of seafloor filmed by our ROV: https://github.com/zhrandell/Seattle_Aquarium_ROV_telemetry_imagery_analysis/issues/9
- The Underwater GPS system we use: https://waterlinked.com/underwater-gps
- The Doppler Velocity Log we use: https://waterlinked.github.io/dvl/dvl-a50/
- *Surftrak*, a custom autonomous altitude-holding software develop by Clyde McQueen: https://github.com/clydemcqueen/ardusub_surftrak
- *R* code generating species richness curves: https://www.dropbox.com/scl/fi/2jsrtvbya0qwey5lcoprk/spp_richness_curves.R?rlkey=nncg9pjy7t3wt56yh40q6ks0n&dl=0

- Existing Bull Kelp Habitat Suitability Model: https://experience.arcgis.com/experience/b11daaa83ff045f1a9d88b2b926e1f75/page/About/
- Suitability Modeler tool in ArcGIS: https://pro.arcgis.com/en/pro-app/latest/help/analysis/spatial-analyst/suitability-modeler/what-is-the-suitability-modeler.htm
- Benthic Terrain Modeler tool: https://www.arcgis.com/home/item.html?id=b0d0be66fd33440d97e8c83d220e7926#!?TB_iframe=true
- Weighted Overlay tool: https://pro.arcgis.com/en/pro-app/latest/tool-reference/spatial-analyst/weighted-overlay.htm
- BlueROV2 Electronics Enclosure: https://bluerobotics.com/store/rov/bluerov2-components-spares/brov2-asm-electronics-r1/
- Some data associated with Urban Kelp research: https://www.dropbox.com/scl/fi/gojf42mdtod3omq61smvu/species_richness_curves.csv?rlkey=q03kl78df4537gt9y0ykin16o&dl=0
- Draft HSIL Initial Factsheet: https://www.dropbox.com/scl/fi/tpvs1ibtyioiw8pmblzu9/2023-HSIL-Initial-Factsheet_V2.docx?rlkey=534cv9x115sqpjrstd7u3wrbv&dl=0
- GitHub repository for ROV development: https://github.com/zhrandell/Seattle_Aquarium_ROV_development
- GitHub repository for ROV code and analysis: https://github.com/zhrandell/Seattle_Aquarium_ROV_telemetry_imagery_analysis
- GitHub repository for recent Seattle Aquarium publication: https://github.com/zhrandell/Seattle_Aquarium_Hawaii_coral_reef_fish_monitoring
- GitHub page with CCR *R* and Python code: https://github.com/zhrandell/Seattle_Aquarium_ROV_telemetry_imagery_analysis/tree/main/code
- CCR hardware components: https://github.com/zhrandell/Seattle_Aquarium_ROV_development/blob/main/documents/hardware_software.md
- Underwater GPS system: https://waterlinked.github.io/underwater-gps/introduction/
- GNSS Satellite Compass: https://www.advancednavigation.com/inertial-navigation-systems/satellite-compass/gnss-compass/
- BlueOS Extension: https://github.com/clydemcqueen/wl_ugps_external_extension

- Downward-facing survey imagery: https://www.dropbox.com/s/13hnw5mpjuwhczs/2022_10_06_T1_downwards.mp4?dl=0
- Pre- vs post-processed survey imagery: https://www.dropbox.com/s/9prk20bc521m76u/Native_2_color.mp4?dl=0
- Extract stills from video using Python code: https://github.com/zhrandell/Seattle_Aquarium_ROV_telemetry_imagery_analysis/blob/main/code/cutvideo8sec.py
- CoralNet web tutorial: https://www.dropbox.com/scl/fi/itvhzl20ohdulmx8whn82/CoralNet_tutorial.mov?rlkey=ui3kp0t8iqvi6r9wwzlsqjhhs&dl=0
- Washington Department of Ecology's Environmental Information Management System Search for Marine Sediments: https://apps.ecology.wa.gov/eim/search/MonitoringProgramDefault.aspx?StudyMonitoringProgramUserId=MarineSediment&StudyMonitoringProgramUserIdSearchType=Equals
- National Centers for Environmental Information's Bathymetric Data Viewer: https://www.ncei.noaa.gov/maps/bathymetry/
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- Washington State Department of Ecology Marine Water Column Monitoring, 2018: https://ecology.wa.gov/Research-Data/Monitoring-assessment/Puget-Sound-and-marine-monitoring

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- Washington State Department of Ecology Marine Water and Sediment Monitoring, 2018:

  https://ecology.wa.gov/Research-Data/Monitoring-assessment/Puget-Sound-and-marine-monitoring
- CoralNet source with benthic images from ROV surveys: https://coralnet.ucsd.edu/source/3737/

16.0 Appendices

Appendix A. SOP for ROV navigation and survey flights

Central to our ROV surveys is the ability to consistently and precisely record the geospatial positioning of the vehicle at all times. Across the latter half of 2022 and throughout 2023 we have implemented a SOP using three key sensors and open-source software to provide highly precise ROV positioning information, including a redundancy in case the primary acoustic systems exhibits error. The SOP, along with relevant links, can be found below. Please see here for a complete list of ROV, sensor, and topside parts and components associated with our work.

- Once the vessel is anchored securely above a ROV survey location, we first set up WaterLinked's Underwater GPS system. We use the U1 wireless acoustic transmitter affixed in a vertical position on the BlueROV2.
- Deploy the x4 D1 acoustic receivers in a square/rectangular grid shape around the vessel (e.g., deploy D1 receivers off the port stern, the starboard stern, port midship, and starboard midship). Plug the receiver cables into the G2 box, and power the G2 box. The depth of the receivers is set at increments of 0.5m, with x2 receivers at 1.5m depth and x2 receivers at a depth of 3m being advisable.
- The GNSS Satellite Compass should be mounted on the vessel with the front facing the bow. Plug the Power over Ethernet (PoE) into the GNSS Compass, and plug the other end into an Ethernet switch. The GNSS Compass, G2 box, and ROV laptop all need to be on the same network (192.168.2.1), and this can be achieved by plugging them all into a powered Ethernet switch.
- Measure the distances between the GNSS Compass to each of the D1 receivers mounted around the vessel, and enter those distances (including D1 receiver depth in the water column) into the UGPS GUI.
- Once powered, confirm the GNSS Compass is receiving a signal. In BlueOS (a program we use to fly the ROV), pull up wl_ugps_external_extension—a custom, open-source piece of software that will feed GPS positioning and compass heading information from the GNSS Compass to the UGPS G2 box (thereby enabling far more precise GPS and compass information than the G2 box is able to obtain by itself). Once the extension is running in BlueOS and all four indicators are green, the vehicle is ready to fly and coordinates will be automatically recorded in QGroundControl's .csv telemetry log.
- In order to fly a mission, load a preprogrammed Mission Plan in QGroundControl. These Mission Plans can take variety of forms, but in short, they provide the specific tracklines across which we want to fly the ROV and gather imagery. The Mission Plans are to be configured ahead of time via latitude/longitude coordinates provided either manually in QGroundControl or via a .csv.

- Power the ROV, time-sync the cameras (see **Appendix B. SOP for camera settings and imagery processing**), power the externally mounted lights, and deploy the ROV over the side of the vessel. Fly the ROV to either the Mission Plan's individual transect survey (in the summer) or a grid pattern (in the winter). Proceed with the survey, i.e., fly the vehicle such that the ROV "icon" in QGroundControl (denoting the precise GPS location of the vehicle) is atop the tracklines. Activate *surftrak* as to autonomously keep the ROV 1m above the substrate. Fly the vehicle at no more than 0.3m/s. Once the survey is completed, retrieve the vehicle and download the .csv telemetry file from QGroundControl containing the GPS coordinates from the mission, as well as the GPS coordinates generated by the DVL.
- In the event there is some sort of issue with the navigational system, we can always fall back to using information from the WaterLinked [Doppler Velocity Log] (DVL) to plot our surveys. This redundancy ensure our surveys always have precise spatial tracking.

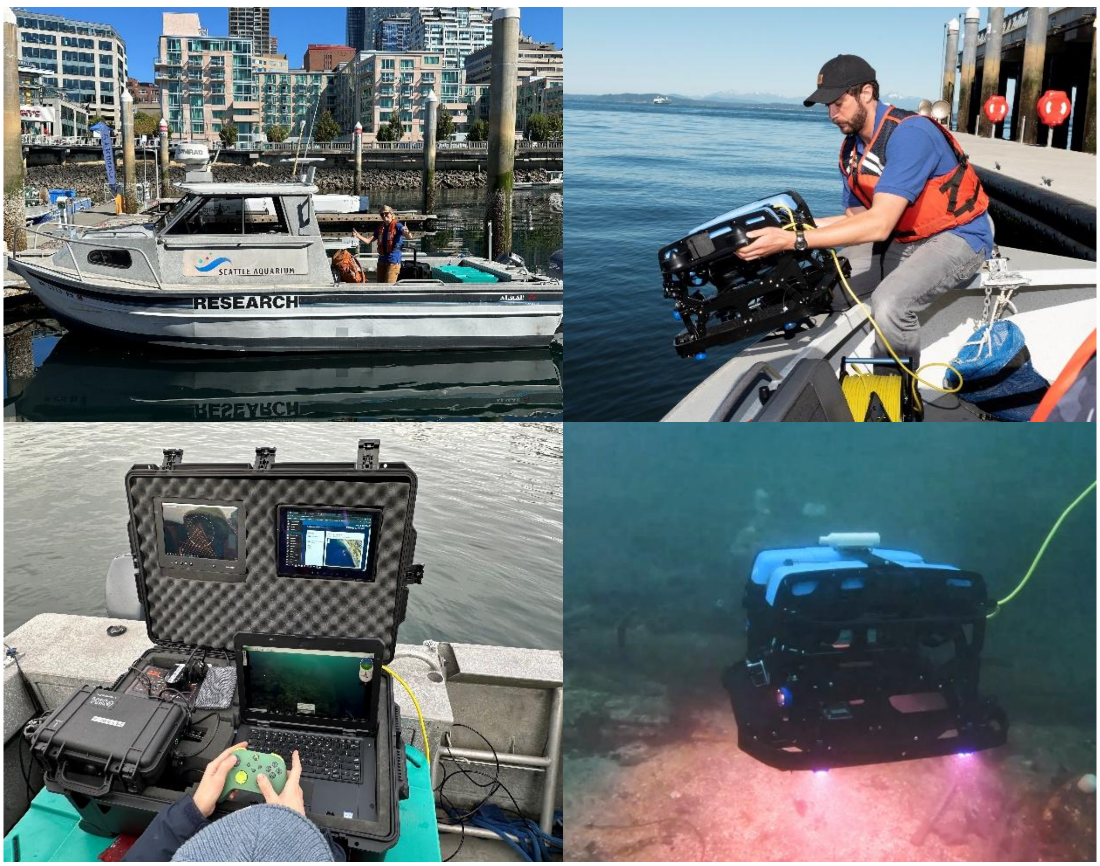

Figure A1. Seattle Aquarium vessel and field deployment of the ROV. Row 1. (left) the Seattle Aquarium vessel, a 24' Aluminum Almar; Randell and Williams are trained vessel operators. (right) Randell deploying ROV *Nereo* off the side of a vessel. Row 2. (left) the Command Console used to track and fly the ROV. (right) ROV Nereo illuminating and filming the seafloor beneath Pier 59 along the Seattle waterfront.

Appendix B. SOP camera settings and imagery processing

We use the following settings on our GoPro HERO 12 video cameras, as well as the following steps to edit the resulting imagery:

- GoPro video settings: 4K resolution 30fps frame rate, Wide lens, ISO = 100-1600, Native White Balance, Flat Color, High Sharpness, 0.0 EV Comp.
- Produce video clips for each transect in Davinci Resolve 18 by combining and trimming videos to isolate the beginning and ending of each transect. We delete the interim "travel" period (flying the ROV to and from the survey location) once the transect has been extracted.
- Adjust the color balance of the video to achieve a natural white balance in Davinci Resolve 18. Use pre-configured white balance settings to generate realistic colors from the Native White balance setting (akin to shooting Raw photos). You can see the difference between the "Native" video we shoot vs the processed videos here.
- Extract the sharpest image at intervals of 8 seconds using a Python code (linked here)
- Proceed with analysis of still images (see **Appendix C. SOP to train CoralNet and extract data**).

We use the following setting on our GoPro HERO 12 to shoot time lapse photos of the seafloor, as we have found the photos are of higher quality than stills extracted from video:

- Timelapse photo, Wide, 1 fps, JPG, Natural color, native WB, ISO 100-400, shutter speed 1/250, zoom 1.x. Default mode when camera is turned on: timelapse; Orientation lock (landscape).
- Customize the settings using GoPro labs firmware website (specific setting customized here).
- Manually add a shutter speed minimum and maximum command: !MEXPN=250! MEXPX=250.
- Show the generated QR code to the cameras and verify the settings adjustment.
- Time sync command introduced to the cameras stereo pair just before the survey begins.
- Transfer the data from the SD card to the DropBox folder corresponding to the date of the survey.
- Import photos to Adobe Lightroom software for images editing.
- Process the photos by adjusting overall exposure, highlights, shadows; apply dehaze effect, enhance texture and clarity; color correct by adjusting a tone curve.

Appendix C. SOP to train CoralNet and extract data

The following steps are required to initiate a project source in CoralNet and begin training an AI algorithm with user uploaded information. To see a web tutorial recorded by Williams for use by our AI Teachers, click here.

- A public CoralNet source (project named CPP_ROV) was created for this project.
- Downward facing images extracted from the downward facing video transects are uploaded into the CoralNet source.
- A .csv file is generated containing the image name and metadata for each image. The metadata associated with the images comprises the date of the transect from which the image was extracted, the season (summer or winter), site name, and transect number.
- A label set containing 66 categories of substrate, algae, and invertebrates was created. General red and green algae categories were made, while distinct categories were made for kelp species. Washington state kelp species list was obtained from the Puget Sound Kelp Forests report prepared by Max Calloway for Washington Department of Fish and Wildlife (Calloway, 2022).
- CoralNet generates a set number of points overlaid onto each image through a straightforward random selection method. We originally used 100 points, and have since switched to 50 points per each image.
- On the annotation tool page, points are labeled with one of the 66 categories within the label set, determined by the object or feature the point is positioned on top of.
- The CoralNet classification engine is built on transfer learning approach. A feature extractor that takes an image patch (224 x 224 pixels) centered on the point overlaid onto the image and outputs a feature vector. The classifier takes the feature vector as input and outputs the *n* scores. The classifier is trained repeatedly on a per source basis with the manually labeled points from our source. Once 20 images have been manually labelled, a classifier is trained using the precomputed features as input with the output compared to the manual labels. CoralNet can now use the trained classifier to suggest potential labels that are verified or manually corrected. For every 10% more images labelled another classifier is trained and replaces the old classifier if it is 1% or more accurate.
- After the images have been labeled and/or corrections to the classifier predictions have been made a .csv file containing the percent coverage for each category in the label set in each image can be downloaded.

Appendix D. SOP to conduct ROV-scuba diver methods overlap

These ROV-diver methods have been practiced off of the Seattle Aquarium vessel with support from Seattle Aquarium divers, NOAA National Marine Fisheries Service divers (on their own vessel) as to complete a "shakedown" field day to see how best to choreograph these methods. Based on that preliminary sampling day that took place offshore of Centennial Park in April of 2023, the day-of procedure will consist of the following:

- Two Seattle Aquarium scientific scuba divers trained in Reef Check survey methods will operate off the Seattle Aquarium vessel, as will x2 ROV operators and x2 Primary Boat Operators.
- Assuming topside surface and water conditions are favorable (<20kt wind, <3' swell, flooding or slack high tide, 3m minimum visibility), the Seattle Aquarium vessel will deploy from Elliott Bay Marina breakwater and proceed to, e.g., Centennial Park. We will anchor and stage equipment. The Aquarium vessel will remain anchored at all times.
- The x2 Seattle Aquarium divers will splash, descend, and spool out a 100m tape along the benthos at a depth of ~7m running parallel to shore, in this case north to south. They will use small rocks to weigh down the meter tape every 5-10m as to hold the meter tape in place along the benthos. They will deploy a Pelican float at the 0m and 100m mark to designate the sampling area for surface personnel and to enable the ROV to navigate towards the survey area. Note that the 100m tape will allow us to capture x3 30m transects with 5m spacing.
- Once the 100m tape is deployed, the Seattle Aquarium divers will not collect the benthic Reef Check data but will instead ascend and return to the vessel.
- Once the divers are back on board, the ROV team will deploy the ROV, which will proceed to the 0m mark along the seafloor (designated by one of the x2 pelican floats). Once on station at the 0m, the ROV will lock in an altitude above the seafloor of 1m, and will proceed forward—centered above the meter tape—at a speed of 0.10 meters per second. The ROV will complete x2 passes—out and back—prior to ascending to the surface (though only the first pass is of primary interest to the methods comparison, as the divers likewise only make a single pass).
- Once the ROV is back on board, the divers will once more deploy, and this time they will gather their Reef Check invertebrate and algal swath data, as well as their Uniform Point Contact data. Once complete, they will spool up the meter data, retrieve the Pelican lines, safely ascend, and return to the vessel.
- Once divers are back onboard and the site has been cleaned up, we will pull anchor and depart the site.

Appendix E. SOP for PSRF ROV monitoring

ROVs have yet to be used to monitor bull kelp restoration, nor have Seattle Aquarium personnel participated before in this bull kelp restoration, so the first few instances of our participation in these activities will be experimental such that we intentionally work to identify how best to use our ROV to supplement the diver surveys that monitor these outplanted locations. That being said, certain field protocols are already established, especially regarding the before-and-after full site surveys along the “restored” (or enhanced) vs control locations:

- Prior to the deployment of the bull kelp seed, Seattle Aquarium personnel will motor to the Jefferson Head enhancement site and deploy anchor as close as possible to the site.
- We will deploy our ROV (no divers) along with all navigational equipment (our acoustic GPS system, GNSS Satellite Compass) as to provide precise tracking. Using information from PSRF, we will have programed the center of the enhancement site into our navigational system such that we can see that waypoint and navigate accordingly.
- We will run the ROV systematically such that we survey “back and forth”, specifically in a reciprocal transect manner (with 1m spacing between reciprocal transects), as to obtain benthic-facing imagery that slightly overlaps between adjacent transects. This will allow us to image the same “spot” multiple times between adjacent transects, thereby enabling the construction of 3D models. These overlapping transects will encompass at minimum a 500 square meter grid, providing a comprehensive accounting of the seafloor and fine-scale substrate considerations. This back-and-forth sampling will proceed with a grid overlayed on our screens on the vessel, such that we simply fly the ROV along a pre-determined path.
- These full site surveys will take place at the enhancement sites (at Jefferson Head) as well as a simple, “control” site at Centennial Park where bull kelp is known to control. The purpose of this is to compare and contrast the restored location to a control location as to disentangle any possible differences in local factors such as, e.g., substrate type.
- Furthermore, these full site surveys will take place in March, prior to deployment of the bull kelp seed, and in June, at the conclusion of the PSRF monitoring, as to provide a before and after comparison.

In addition to the beginning and end of the bull kelp monitoring season surveys, we will supplement PSRF’s diver based surveys of the outplanted bull kelp by surveying during the “off weeks”. That is to say, PSRF’s divers survey every other week, thus we will supplement these surveys by surveying via the ROV when the divers are not present. This will provide greater temporal resolution regarding the growing patterns of the outplanted bull kelp.

Appendix F. Reef Check benthic survey protocols

A complete guide to Washington Reef Check's benthic survey protocols, including their benthic invertebrate and algae swath, as well as their Uniform Point Cover survey, can be found at the following link:

Reef Check Washington Student Guide

Appendix G. Glossaries, Acronyms, and Abbreviations

Glossary of General Terms

Dissolved oxygen (DO): A measure of the amount of oxygen dissolved in water.

Diurnal: Of, or pertaining to, a day or each day; daily. (1) Occurring during the daytime only, as different from nocturnal or crepuscular, or (2) Daily; related to actions which are completed in the course of a calendar day, and which typically recur every calendar day (e.g., diurnal temperature rises during the day, and falls during the night).

Eutrophic: Nutrient rich and high in productivity resulting from human activities such as fertilizer runoff and leaky septic systems.

Nutrient: Substance such as carbon, nitrogen, and phosphorus used by organisms to live and grow. Too many nutrients in the water can promote algal blooms and rob the water of oxygen vital to aquatic organisms.

pH: A measure of the acidity or alkalinity of water. A low pH value (0 to 7) indicates that an acidic condition is present, while a high pH (7 to 14) indicates a basic or alkaline condition. A pH of 7 is considered to be neutral. Since the pH scale is logarithmic, a water sample with a pH of 8 is ten times more basic than one with a pH of 7.

Pollution: Contamination or other alteration of the physical, chemical, or biological properties of any waters of the state. This includes change in temperature, taste, color, turbidity, or odor of the waters. It also includes discharge of any liquid, gaseous, solid, radioactive, or other substance into any waters of the state. This definition assumes that these changes will, or are likely to, create a nuisance or render such waters harmful, detrimental, or injurious to (1) public health, safety, or welfare, or (2) domestic, commercial, industrial, agricultural, recreational, or other legitimate beneficial uses, or (3) livestock, wild animals, birds, fish, or other aquatic life.

Primary contact recreation: Activities where a person would have direct contact with water to the point of complete submergence including, but not limited to, skin diving, swimming, and water skiing.

Rugosity: A variation of vertical relief

Substrate: Surface or material on which an organism can grow or attach to that is covered with water (for. Example, ocean or estuary)

Acronyms and Abbreviations

AI	Artificial Intelligence
BAG	Bathymetric Attributed Grid
BPI	Bathymetric Position Index
BTM	Benthic Terrain Modeler
DEM	Digital Elevation Model
DIN	Dissolved inorganic nitrogen
DVL	Doppler Velocity Log
e.g.	For example
et al.	And others
GIS	Geographic Information System software
GPS	Global Positioning System
HSIL	Habitat Strategic Initiative Lead
HSM	Habitat Suitability Model
i.e.	In other words
ML	Machine learning
MLLW	Mean Lower Low Water
MOTC	Motorboat Operator Training Course
MQO	Measurement Quality Objective
NCEI	National Centers for Environmental Information
NOAA	National Oceanic and Atmospheric Administration
NOAA NMFS	NOAA National Marine Fisheries Service
NOS	National Ocean Service
PBO	Primary Boat Operator
QA	Quality assurance
QC	Quality control
ROV	Remotely operated vehicle
SOP	Standardized Operating Procedures
SSM	Salish Sea Model
UGPS	Underwater GPS
USGS	United States Geographical Survey

WDFW	Washington Department of Fish and Wildlife
WDNR	Washington Department of Natural Resources
VIAME	Video and Image Analytics for Marine Environments

Units of Measurement

°C	Degrees centigrade
ft	Feet
km	Kilometer
m	Meter
cm	Centimeter
uM	Micron
ppt	Parts per trillion

Quality Assurance Glossary

Accreditation: A certification process for laboratories, designed to evaluate and document a lab's ability to perform analytical methods and produce acceptable data (Kammin, 2010). For Ecology, it is defined according to WAC 173-50-040: "Formal recognition by [Ecology] that an environmental laboratory is capable of producing accurate and defensible analytical data."

Accuracy: The degree to which a measured value agrees with the true value of the measured property. USEPA recommends that this term not be used, and that the terms *precision* and *bias* be used to convey the information associated with the term *accuracy* (USEPA, 2014).

Analyte: An element, ion, compound, or chemical moiety (pH, alkalinity) which is to be determined. The definition can be expanded to include organisms, e.g., fecal coliform, Klebsiella (Kammin, 2010).

Bias: Discrepancy between the expected value of an estimator and the population parameter being estimated (Gilbert, 1987; USEPA, 2014).

Blank: A synthetic sample, free of the analyte(s) of interest. For example, in water analysis, pure water is used for the blank. In chemical analysis, a blank is used to estimate the analytical response to all factors other than the analyte in the sample. In general, blanks are used to assess possible contamination or inadvertent introduction of analyte during various stages of the sampling and analytical process (USGS, 1998).

Calibration: The process of establishing the relationship between the response of a measurement system and the concentration of the parameter being measured (Ecology, 2004).

Check standard: A substance or reference material obtained from a source independent from the source of the calibration standard; used to assess bias for an analytical method. This is an obsolete term, and its use is highly discouraged. See Calibration Verification Standards, Lab Control Samples (LCS), Certified Reference Materials (CRM), and/or spiked blanks. These are all check standards but should be referred to by their actual designator, e.g., CRM, LCS (Kammin, 2010; Ecology, 2004).

Comparability: The degree to which different methods, data sets and/or decisions agree or can be represented as similar; a data quality indicator (USEPA, 2014; USEPA, 2020).

Completeness: The amount of valid data obtained from a project compared to the planned amount. Usually expressed as a percentage. A data quality indicator (USEPA, 2014; USEPA 2020).

Continuing Calibration Verification Standard (CCV): A quality control (QC) sample analyzed with samples to check for acceptable bias in the measurement system. The CCV is usually a midpoint calibration standard that is re-run at an established frequency during the course of an analytical run (Kammin, 2010).

Control chart: A graphical representation of quality control results demonstrating the performance of an aspect of a measurement system (Kammin, 2010; Ecology 2004).

Control limits: Statistical warning and action limits calculated based on control charts. Warning limits are generally set at +/- 2 standard deviations from the mean, action limits at +/- 3 standard deviations from the mean (Kammin, 2010).

Data integrity: A qualitative DQI that evaluates the extent to which a data set contains data that is misrepresented, falsified, or deliberately misleading (Kammin, 2010).

Data quality indicators (DQI): Commonly used measures of acceptability for environmental data. The principal DQIs are precision, bias, representativeness, comparability, completeness, sensitivity, and integrity (USEPA, 2006).

Data quality objectives (DQO): Qualitative and quantitative statements derived from systematic planning processes that clarify study objectives, define the appropriate type of data, and specify tolerable levels of potential decision errors that will be used as the basis for establishing the quality and quantity of data needed to support decisions (USEPA, 2006).

Data set: A grouping of samples organized by date, time, analyte, etc. (Kammin, 2010).

Data validation: The process of determining that the data satisfy the requirements as defined by the data user (USEPA, 2020). There are various levels of data validation (USEPA, 2009).

Data verification: Examination of a data set for errors or omissions, and assessment of the Data Quality Indicators related to that data set for compliance with acceptance criteria (MQOs). Verification is a detailed quality review of a data set (Ecology, 2004).

Detection limit (limit of detection)**:** The concentration or amount of an analyte which can be determined to a specified level of certainty to be greater than zero (Ecology, 2004).

Duplicate samples: Two samples taken from and representative of the same population, and carried through and steps of the sampling and analytical procedures in an identical manner. Duplicate samples are used to assess variability of all method activities including sampling and analysis (USEPA, 2014).

Field blank: A blank used to obtain information on contamination introduced during sample collection, storage, and transport (Ecology, 2004).

Initial Calibration Verification Standard (ICV): A QC sample prepared independently of calibration standards and analyzed along with the samples to check for acceptable bias in the measurement system. The ICV is analyzed prior to the analysis of any samples (Kammin, 2010).

Laboratory Control Sample (LCS)/LCS duplicate: A sample of known composition prepared using contaminant-free water or an inert solid that is spiked with analytes of interest at the midpoint of the calibration curve or at the level of concern. It is prepared and analyzed in the same batch of regular samples using the same sample preparation method, reagents, and analytical methods employed for regular samples. Monitors a lab's performance for bias and precision (USEPA, 2014).

Matrix spike/Matrix spike duplicate: A QC sample prepared by adding a known amount of the target analyte(s) to an aliquot of a sample to check for bias and precision errors due to interference or matrix effects (Ecology, 2004).

Measurement Quality Objectives (MQOs)**:** Performance or acceptance criteria for individual data quality indicators, usually including precision, bias, sensitivity, completeness, comparability, and representativeness (USEPA, 2006).

Measurement result: A value obtained by performing the procedure described in a method (Ecology, 2004).

Method: A formalized group of procedures and techniques for performing an activity (e.g., sampling, chemical analysis, data analysis), systematically presented in the order in which they are to be executed (USEPA, 2001).

Method blank: A blank prepared to represent the sample matrix, prepared and analyzed with a batch of samples. A method blank will contain all reagents used in the preparation of a sample, and the same preparation process is used for the method blank and samples (Ecology, 2004; Kammin, 2010).

Method Detection Limit (MDL): The minimum measured concentration of a substance that can be reported with 99% confidence that the measured concentration is distinguishable from method blank results (USEPA, 2016). MDL is a measure of the capability of an analytical method of distinguished samples that do not contain a specific analyte from a sample that contains a low concentration of the analyte (USEPA, 2020).

Minimum level: Either the sample concentration equivalent to the lowest calibration point in a method or a multiple of the method detection limit (MDL), whichever is higher. For the purposes of NPDES compliance monitoring, EPA considers the following terms to be synonymous: "quantitation limit," "reporting limit," and "minimum level" (40 CFR 136).

Parameter: A specified characteristic of a population or sample. Also, an analyte or grouping of analytes. Benzene and nitrate + nitrite are all parameters (Kammin, 2010; Ecology, 2004).

Population: The hypothetical set of all possible observations of the type being investigated (Ecology, 2004).

Precision: The extent of random variability among replicate measurements of the same property; a data quality indicator (USGS, 1998).

Quality assurance (QA): A set of activities designed to establish and document the reliability and usability of measurement data (Kammin, 2010).

Quality Assurance Project Plan (QAPP): A document that describes the objectives of a project, and the processes and activities necessary to develop data that will support those objectives (Kammin, 2010; Ecology, 2004).

Quality control (QC): The routine application of measurement and statistical procedures to assess the accuracy of measurement data (Ecology, 2004).

Relative Percent Difference (RPD): RPD is commonly used to evaluate precision. The following formula is used:

$$RPD = [Abs(a-b)/((a + b)/2)] * 100\%$$

where "Abs()" is absolute value and a and b are results for the two replicate samples. RPD can be used only with 2 values. Percent Relative Standard Deviation is (%RSD) is used if there are results for more than 2 replicate samples (Ecology, 2004).

Relative Standard Deviation (RSD): A statistic used to evaluate precision in environmental analysis. It is determined in the following manner:

$$RSD = (100\% * s)/x$$

where s is the sample standard deviation and x is the mean of results from more than two replicate samples (Kammin, 2010).

Replicate samples: Two or more samples taken from the environment at the same time and place, using the same protocols. Replicates are used to estimate the random variability of the material sampled (USGS, 1998).

Reporting level: Unless specified otherwise by a regulatory authority or in a discharge permit, results for analytes that meet the identification criteria (i.e., rules for determining qualitative presence/absence of an analyte) are reported down to the concentration of the minimum level established by the laboratory through calibration of the instrument. EPA considers the terms "reporting limit," "quantitation limit," and "minimum level" to be synonymous (40 CFR 136).

Representativeness: The degree to which a sample reflects the population from which it is taken; a data quality indicator (USGS, 1998).

Sample (field): A portion of a population (environmental entity) that is measured and assumed to represent the entire population (USGS, 1998).

Sample (statistical): A finite part or subset of a statistical population (USEPA, 1992).

Sensitivity: In general, denotes the rate at which the analytical response (e.g., absorbance, volume, meter reading) varies with the concentration of the parameter being determined. In a specialized sense, it has the same meaning as the detection limit (Ecology, 2004).

Spiked blank: A specified amount of reagent blank fortified with a known mass of the target analyte(s); usually used to assess the recovery efficiency of the method (USEPA, 2014).

Spiked sample: A sample prepared by adding a known mass of target analyte(s) to a specified amount of matrix sample for which an independent estimate of target analyte(s) concentration is available. Spiked samples can be used to determine the effect of the matrix on a method's recovery efficiency (USEPA, 2014).

Split sample: A discrete sample subdivided into portions, usually duplicates (Kammin, 2010).

Standard Operating Procedure (SOP): A document which describes in detail a reproducible and repeatable organized activity (Kammin, 2010).

Surrogate: For environmental chemistry, a surrogate is a substance with properties similar to those of the target analyte(s). Surrogates are unlikely to be native to environmental samples. They are added to environmental samples for quality control purposes, to track extraction efficiency and/or measure analyte recovery. Deuterated organic compounds are examples of surrogates commonly used in organic compound analysis (Kammin, 2010).

Systematic planning: A step-wise process which develops a clear description of the goals and objectives of a project, and produces decisions on the type, quantity, and quality of data that will be needed to meet those goals and objectives. The DQO process is a specialized type of systematic planning (USEPA, 2006).

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