Fisheries and Oceans Canada
Ecosystems and Oceans Science

Pêches et Océans Canada
Sciences des écosystèmes et des océans

Canadian Science Advisory Secretariat (CSAS)

Research Document 2023/031

Pacific Region

Recommendations on the design of a Multispecies Benthic Marine Invertebrate Dive Survey Program for Stock Monitoring in British Columbia

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July 2023

Foreword

This series documents the scientific basis for the evaluation of aquatic resources and ecosystems in Canada. As such, it addresses the issues of the day in the time frames required and the documents it contains are not intended as definitive statements on the subjects addressed but rather as progress reports on ongoing investigations.

Published by:

Fisheries and Oceans Canada
Canadian Science Advisory Secretariat
200 Kent Street
Ottawa ON K1A 0E6

http://www.dfo-mpo.gc.ca/csas-sccs/
csas-sccs@dfo-mpo.gc.ca

ISSN 1919-5044
ISBN 978-0-660-48392-4   Cat. No. Fs70-5/2023-031E-PDF

Correct citation for this publication:

Lochead, J., Schwarz, C.J., Rooper, C., and Bureau, D. 2023. Recommendations on the design of a Multispecies Benthic Marine Invertebrate Dive Survey Program for Stock Monitoring in British Columbia. Can. Sci. Advis. Sec. Res. Doc. 2023/031. ix + 99p.

Aussi disponible en français :

Lochead, J., Schwarz, C.J., Rooper, C. et Bureau, D. 2023. Recommandations sur la conception d’un programme de relevé en plongée de multiples espèces d’invertébrés marins benthiques pour le suivi des stocks en Colombie-Britannique. Secr. can. des avis sci. du MPO. Doc. de rech. 2023/031. ix + 113 p.

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ABSTRACT

A new multispecies benthic invertebrate monitoring program is being developed to quantitatively monitor stock abundance over time on the British Columbia (BC) coast. This dive survey is designed to monitor abundance of Green (*Strongylocentrotus droebachiensis*), Red (*Mesocentrotus franciscanus*) and Purple (*Strongylocentrotus purpuratus*) Sea Urchin, Giant Red Sea Cucumber (*Apostichopus californicus*), Northern Abalone (*Haliotis kamtschatkana*), Sunflower Star (*Pycnopodia helianthoides*) and Pacific Geoduck (*Panopea generosa*) populations, and also to collect detailed habitat information on substrate and algae. The survey protocol was developed in 2016 and is described in detail. Pilot surveys were conducted in different areas of the coast from 2016 to 2021. Data from these pilot surveys, along with data from single-species surveys (1978 to 2021), were analysed to make recommendations on optimal survey design for the new monitoring program. The methods included reviewing single-species analyses that informed sampling intensity on transects, looking at historic maximum transect lengths, investigating stratification variables, and using an acceptance sampling method to determine the minimum required number of transects, given predefined risks and probabilities associated with being above or below reference points. In addition, densities of the Giant Red Sea Cucumber and size and habitat subsets of Red Sea Urchin populations were estimated as an example of how these data could be used to assess stock status in the future. The recommendations on survey design are to: 1) Use the dive survey protocol described in Appendix A of the Research Document; 2) Exclude sections of shoreline with fetch values lower than 20,000 m or higher than 2.52 million m; 3) Ensure surveys occur at the same time of year to avoid introducing seasonal variation to the data; 4) Use the common (across species) coast wide standard deviation-to-mean ratio of density (animals per m^2) equal to 1.27 in calculations to determine the target number of transects to be sampled; 5) Conduct at least 240 transects coast wide to estimate stock status; 6) Implement a two-stage, random sampling design that minimizes the time required to cover the entire BC coast and optimizes the efficient use of available resources; and 7) Continue to explore pre- or post-stratification variables to improve survey precision, as data become available.

1. INTRODUCTION

Benthic marine invertebrates are important resources in British Columbia and are harvested in commercial and recreational fisheries, as well as for food, social and ceremonial (FSC) purposes by First Nations. Commercial dive fisheries for several species of benthic marine invertebrates, i.e., Northern Abalone (*Haliotis kamtschatkana),* Geoduck (*Panopea generosa*), Green Sea Urchin (GSU) (*Strongylocentrotus droebachiensis*), Red Sea Urchin (RSU) (*Mesocentrotus franciscanus*), and Giant Red Sea Cucumber (*Apostichopus californicus*), began in British Columbia (BC) in the 1970s. With the exception of Northern Abalone[1], dive surveys to assess stock densities for these species began in the 1990s as single-species surveys undertaken to estimate biomass in various portions of the BC coast to inform quota setting decisions by fishery managers.

Each dive fishery in BC is unique and individually managed. The Green Sea Urchin fishery is relatively small, and operates throughout the year on the south coast of BC (DFO 2022a). The Red Sea Urchin fishery occurs along most of the coast of BC, and also operates all year long (DFO 2022b). The Sea Cucumber fishery mostly takes place on the north and central coast of BC, as well as between Vancouver Island and the BC mainland (DFO 2022c). It typically opens on the first Monday in October and usually lasts 6 to 8 weeks (DFO 2022c). The Geoduck fishery is conducted coast wide, and it typically operates year-round (subject to acceptable biotoxin levels) with peaks in demand around Christmas and Lunar New Year (DFO 2022d, 2021a).

In 2009, "A fishery decision-making framework incorporating the precautionary approach" (DFO 2009) (the PA) became policy under the Sustainable Fisheries Framework. The PA, in general, is about being cautious when scientific information is uncertain, unreliable or inadequate and not using the absence of adequate scientific information as a reason to postpone or fail to take action to avoid serious harm to the resource (DFO 2009). The PA Policy outlines DFO's method to apply the precautionary approach to set harvest levels and make decisions respecting harvest levels in fisheries to reduce the risk of harm to fish populations. The PA framework specifies three stock status zones for fish stocks (Healthy, Cautious and Critical) that are delimited by reference points. The Upper Stock Reference (USR) is the boundary between the Healthy and Cautious zones. The Limit Reference Point (LRP) is the boundary between the Cautious and Critical zones and is defined as the point where serious harm to the stock is likely to occur.

Coast wide populations of Northern Abalone, Geoducks, GSU, RSU, and Giant Red Sea Cucumbers are each considered a single stock for management purposes because there is no strong evidence of genetic differentiation that would indicate multiple stocks in BC waters (but

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[1] The Northern Abalone is a traditional food that was harvested by coastal BC First Nations (Sloan and Breen 1988). A recreational fishery for Northern Abalone developed in BC in the 1950s with the advent of SCUBA diving (Campbell 1997). A commercial fishery for Northern Abalone took place in BC from the early 1900s to 1990; the commercial fishery was most active during the 1970s and 1980s (Fedorenko and Sprout 1982; Campbell 1997). All fisheries for Northern Abalone in BC were closed in 1990 due to conservation concerns. Northern Abalone in BC was assessed as Threatened by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) in 1999 and listed as Threatened on Schedule 1 under the *Species At Risk Act* (SARA) in 2003. Northern Abalone was re-assessed as Endangered by COSEWIC in 2009 and listed on Schedule 1 as Endangered under SARA in 2009 (Obradovich et al. 2021). Dive surveys to monitor Northern Abalone population density in BC began in the late 1970s and are ongoing.

see Xuereb et al. (2018) for spatial patterns of Giant Red Sea Cucumber genetic variation in BC). Therefore, reference points are applied coast wide. Reference points have been developed for all the benthic marine invertebrates that are commercially harvested by dive in BC (Zhang and Hand 2007; DFO 2021a [Geoducks]; DFO 2018 [GSU]; Lochead et al. 2019 [RSU]; Hajas et al. 2023 [Giant Red Sea Cucumbers]) (Table 1).

In 2019, Bill C-68 was passed, resulting in revisions to Canada's *Fisheries Act* (RSC, 1985, c. F-14). Changes to the *Act* include new Fish Stocks Provisions that require evidence to support management measures to:

1. maintain major fish stocks at or above the level to promote sustainability of the stock (6.1(1)),
2. maintain stocks above the Limit Reference Point (6.1(2)), and
3. develop plans to rebuild stocks that are below their Limit Reference Point in provided time periods (6.2),

while taking into account the biology of the species and environmental conditions affecting the stocks.

To achieve these requirements for the BC dive fisheries, Fisheries and Oceans Canada (DFO) Science has been developing a new coast wide multispecies benthic marine invertebrate dive survey. Previous survey methods, although useful for estimating biomass for setting fishing quotas, are not suitable for fulfilling the requirements of the Fish Stocks Provisions, i.e., determining stock status in relation to reference points. The single-species dive survey programs currently in place are designed to estimate biomass in various portions of the coast each year[2] (Duprey and Stanton 2018; Bureau 2017a; Duprey and Stanton 2015; DFO 2014; Duprey 2014; Duprey 2011; Waddell et al. 2010; Babuin et al. 2006; Hand and Dovey 2000; Bureau et al. 2000a, 2000b, 2000c, 2000d). As a result, biomass estimates change from year to year because different areas are surveyed each year, however, these changes do not necessarily reflect stock trends and are therefore not suitable for stock monitoring. A new monitoring program, designed to assess stocks of benthic marine invertebrates against reference points, is therefore required for the BC dive fisheries. Because single-species dive survey protocols are similar between species, using a multispecies survey approach results in a more efficient use of resources available for field work.

The multispecies benthic marine invertebrate dive survey is intended to quantitatively monitor stock abundance over time on the BC coast. The survey is designed to monitor abundance of Green, Purple (*Strongylocentrotus purpuratus*) and Red Sea Urchins, Giant Red Sea Cucumber, Northern Abalone, Sunflower Star (*Pycnopodia helianthoides*) and Geoducks. Detailed habitat data on substrate and algae are also recorded. The multispecies dive survey is intended to enable DFO to determine stock status in relation to reference points. The data collected on the multispecies dive surveys can also contribute to updates to current reference points, and potentially the eventual development of reference points for the other invertebrates assessed by the survey, and provide an additional tool to monitor the endangered Northern Abalone population. In addition to achieving the requirements of the Fish Stocks Provisions, the multispecies benthic marine invertebrate dive survey will help work towards an ecosystem-based-approach to stock assessment and fisheries management, which is a Departmental

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[2] Bazinet, A.C., Garner, G.D., and S.C. Hansen. *In prep*. Biomass estimates for sea cucumber (*Apostichopus californicus*) as determined through surveys conducted from 2014 to 2020. Can. Manuscr. Rep. Fish. Aquat. Sci.

priority (DFO 2007b). An ecosystem approach includes the need for rule-based and risk-based management strategies using reliable indicators, broadening the basis for science advice to include the regime status of the environment, and applying knowledge of how stock productivity is expected to change with population demographics and environmental conditions (DFO 2007b).

Table 1. Limit reference point (LRP) and upper stock reference (USR), size/age at first maturity, and minimum legal size (where applicable) for Giant Red Sea Cucumber, Green Sea Urchin, Red Sea Urchin, Pacific Geoduck, Purple Sea Urchin, Northern Abalone, and Sunflower Star.

Species	**Commercial Fishery in BC**	**Common Size Measurement**	**LRP**	**USR**	**Size/Age at first maturity**	**Minimum legal size**
Giant Red Sea Cucumber	Yes	Biosamples taken and split weight recorded starting in 2020	0.029 sea cucumbers/ m^2 on sea cucumber habitat	0.038 sea cucumbers/ m^2 on sea cucumber habitat	unknown; DFO uses Bensia pencil length for size at first maturity	n/a
Green Sea Urchin	Yes	test diameter (TD)	0.45 legal-sized (≥ 55 mm TD) urchins per m^2	0.9 legal-sized (≥ 55 mm TD) urchins per m^2	25 mm / ~2 - 3 years	55 mm
Red Sea Urchin	Yes	test diameter (TD)	0.3 mature (≥ 50 mm TD) urchins per m^2 on RSU habitat	0.6 mature (≥ 50 mm TD) urchins per m^2 on RSU habitat	50 mm / ~2 - 3 years	90 mm
Pacific Geoduck	Yes	shell length	40% coastwide unfished biomass	50% coastwide unfished biomass	2 years	n/a
Purple Sea Urchin	No	test diameter (TD)	n/a	n/a	16 - 25 mm / ~ 2 years	Fished under scientific permit from 1990 to 1992 and are no longer harvested commercially.
Northern Abalone	No	max shell length	n/a	n/a	50 mm	Fishery closed in 1990
Sunflower Star	No	n/a	n/a	n/a	unknown	n/a

The dive survey will also be an important tool for monitoring Sunflower Star (*Pycnopodia helianthoides*) abundance. *Pycnopodia* was listed as critically endangered by the International Union for Conservation of Nature (IUCN) in 2021 along its range in North America (Gravem et al. 2021). It is possible that *Pycnopodia* will be assessed by the Committee On the Status of Endangered Wildlife In Canada (COSEWIC) in the future. If this assessment leads to listing of *Pycnopodia* under the *Species At Risk Act*, then DFO will be responsible to monitor *Pycnopodia* populations in BC and the multispecies dive survey will fulfill this requirement. The data from the monitoring program will also be useful for habitat mapping, species distribution modeling, emergency response, as well as Marine Protected Area planning and monitoring programs. Furthermore, the results from this monitoring program may also be useful to Fisheries Managers for incorporating the Precautionary Approach and ecosystem considerations into their decision making.

The multispecies benthic invertebrate dive survey protocol was developed in 2016 and pilot surveys were then conducted around Vancouver Island, on the mainland North Coast and south east Haida Gwaii between 2016 and 2021. The purpose of this research document is to review the 2016-2021 pilot data and make recommendations on optimal survey design for the long-term, coast wide monitoring program. This is being undertaken to ensure the program can collect the data necessary to evaluate the status of several important benthic marine invertebrates to fulfill the requirements of the Fish Stocks Provisions in the revised *Fisheries Act*. The specific objectives are to:

1. Describe the methods used to collect multispecies benthic marine invertebrate dive data during the 2016-2021 pilot studies.
2. Summarize benthic marine invertebrate abundance and its variability by species and region for the 2016-2021 pilot surveys for Green, Purple and Red Sea Urchins, Giant Red Sea Cucumber, Geoduck, Northern Abalone, and Sunflower Star. Note any gaps or uncertainties arising from the design and/or implementation of the pilot studies.
3. Make recommendations on optimal survey design considerations such as survey effort (number of transects), survey frequency, distribution of survey effort (random, index sites, panel design), etc.
4. Make recommendations on types of environmental data that would inform relevant survey stratification and/or strengthen the interpretation of the species abundance results (e.g., Sea Otter, *Enhydra lutris,* presence/absence*,* occupancy time; fetch; etc.).
5. Identify knowledge gaps and key uncertainties that could be addressed to further improve the survey design.

This research document is comprised of eight major sections. Following Section 1 (Introduction), Section 2 (The Pilot Surveys) describes how the survey protocol was developed, including where the pilot surveys were conducted. It also provides the methods and results of invertebrate density estimation, variability in density, and expected precision from the pilot surveys. Section 3 (Using Pilot Survey Data to Inform Protocol Improvements) outlines how the pilot survey data were used to help identify the shoreline to be targeted (relative to fetch values), and examines four potential stratification variables. Section 4 (Sample Size and Survey Design) summarizes how a statistical process called 'Acceptance Sampling' was used to recommend the survey sample size (total number of dive transects). Section 4 also provides examples of random survey designs along with a recommendation on the preferred random survey design for the coast wide monitoring program. Section 5 describes the uncertainties, Section 6 provides a general discussion, Section 7 outlines areas for future work, and the science advice recommendations are provided in Section 8.

2. THE PILOT SURVEYS

2.1. DEVELOPING THE PROTOCOL

Conducting dive surveys poses logistical challenges, many of which are related to SCUBA diving time limits and safety considerations. These limitations must be considered when designing a dive survey protocol so that the protocol is safe, effective at gathering the desired data and logistically achievable. In addition, working in remote parts of the BC coast poses additional logistical and safety challenges that must also be taken into account in survey design.

Most dive surveys conducted by DFO to date used fixed or randomly placed transects that are conducted perpendicular to the shoreline[2] (DFO 2021b; Duprey and Stanton 2018; DFO 2018; Bureau 2017a; DFO 2016a; Duprey and Stanton 2015; DFO 2014; Duprey 2014; Duprey 2011; Waddell et al. 2010; Babuin et al. 2006; Hand and Dovey 2000; Bureau et al. 2000a, 2000b, 2000c, 2000d). Transects lines (made of 25 m sections of lead-core rope marked every 5 m with cable ties) were deployed from the dive boat at the pre-selected transect location before the dive. The deep end of a transect was marked with a float. Once the transect was deployed, divers got ready, then entered the water near the float, descended to the deep end of the transect and surveyed the transect from deep to shallow.

The same general concept was chosen for the multispecies dive surveys. Previous experience with dive survey design and the intended objectives of the multispecies dive survey informed the initial dive protocol. The initial protocol was tested near Nanaimo between June and August, 2016, so that the dive protocol could be refined and finalized before the first survey in September 2016.

The detailed multispecies dive survey protocol and required equipment are presented in Appendix A. The following sections outline the factors taken into consideration when designing the multispecies dive survey protocol.

2.1.1. Depth limits of the survey

Because of diving time restrictions and safety considerations, a maximum target depth for the deep end of transects must be chosen. Dive surveys for various benthic marine invertebrates in BC have used different depth limits. For Geoduck surveys, target depths are 3 m to 18 m relative to chart datum (i.e., corrected for tide height, Bureau et al. 2012) to a maximum actual depth of 21.3 m (70 feet). Green Sea Urchin dive surveys have target depths of 0 m to 10 m relative to chart datum (DFO 2014). The shallow limit for Abalone index site surveys is typically around -1 to 0 m relative to chart datum; Abalone index site survey plots are 7 m long (DFO 2016b) so that the maximum theoretical depth would be 7 m (relative to chart datum) if a site was a vertical wall. The depth limits for Sea Cucumber surveys are from the surface (0 m) to 15.2 m (50 feet) actual depth (Duprey and Stanton 2018), i.e., not relative to chart datum so that maximum depth surveyed on a transect (relative to chart datum) is dependent on tide height at the time the transect is surveyed. Similarly, habitat mapping survey transects were conducted from the surface to 18 m actual depth (Davies et al 2018), i.e., not relative to chart datum. Red Sea Urchin survey transects are surveyed from the surface (0 m) to 15 m (50 feet) actual depth (Lochead et al. 2019), i.e., not relative to chart datum.

For surveys that use target depths not relative to chart datum, the depth range surveyed after being corrected for tide height varies between transects making comparisons between transects more difficult. Making data more comparable between transects sometimes requires truncation of data for some of the deepest quadrats for transects surveyed at lower tides (because the survey would have gone deeper relative to chart datum), which can translate into wasted survey effort.

Surveying the same depth range across transects is desirable to make the data more comparable between transects and maximize the useability of the data. Because tide heights change during the day and between days, it is preferable to choose target maximum and minimum survey depths in relation to chart datum so that the same depth range (corrected for tide height) is surveyed on all transects irrespective of tide height at the time of the survey.

In DFO diving regulations, shallow dives are restricted to 18.3 m (60 feet) or shallower. Choosing a target maximum depth that will lead to actual survey depths ≤18.3 m most of the time was therefore desired. Tide amplitude in some portions of the BC coast can be as high as 7.6 m. A target maximum depth of 12.2 m (40 feet) below chart datum could thus translate in a maximum actual survey depth of 19.8 m, albeit infrequently (i.e., only when tide height is >6.1 m (20 feet) which would only occur at high tide in locations and days with highest tide amplitudes) and the maximum actual depth would be less than 18.3 m on most dives. The target maximum survey depth was therefore chosen as 12.2 m (40 feet) relative to chart datum for the multispecies dive survey program. This ensures that the maximum actual target depth (not corrected for tide) will be shallower than 18.3 m for most transects. The average maximum actual depth surveyed on multispecies dive transects conducted to date (where the target depth was reached) was 14.3 m (47 feet, n = 418) making the maximum actual depth of the multispecies dive transects on average similar to the maximum target depth for Sea Cucumber and Red Sea Urchin surveys.

Depth at which to end a transect at the shallow end may also be dependent on tide height. Since Northern Abalone can sometimes be found in the low intertidal zone, surveying partly into the intertidal zone is desirable. However, there is no need to survey the entire intertidal zone since species of interest are mostly not found there. The portion of the intertidal zone that is available to be surveyed by divers will be dependent on tide height. Therefore, it would not be possible to survey the entire intertidal zone at all sites. A target depth of -2 m (-6 feet, i.e., 2 m or 6 feet above chart datum) was chosen as a target minimum survey depth so that intertidal abalone are surveyed while avoiding surveying the high intertidal zone. It may not be possible to survey to the shallow target depth at all sites depending on weather conditions (see survey protocol, Appendix A).

2.1.2. Quadrat size

Urchin and abalone surveys conducted to date in BC used a 1 x 1 m (1 m^2) quadrat size and divers work together on each quadrat. Single-species Geoduck surveys use 1 x 5 m strip quadrats and sea cucumber single-species surveys use 2 x 5 m strip quadrats. For Geoduck and sea cucumber surveys, each of the two divers works independently surveying on each side of the transect line so that the total transect width for these surveys is 2 m and 4 m, respectively. Using a quadrat size greater than 1 m^2 for a multispecies survey would not be practical or logistically feasible. Consequently, a 1 m^2 quadrat size was chosen for multispecies dive surveys to be consistent with previous urchin and abalone surveys.

2.1.3. Sampling intensity on transects

Survey site locations were randomly selected, therefore, transects covered all habitat types available within the survey area. Because start and end of transects are defined in terms of depth relative to chart datum, transect length is variable between sites and dictated by the slope of the substrate at the site.

Skibo et al. (2008) compared sampling designs and sampling intensity for Red Sea Urchin dive surveys and showed that sampling variation within a transect was small compared to between transect variation after 20 to 30 quadrats had been sampled on a transect. They concluded that, for Red Sea Urchins, once a moderate number of quadrats are sampled, the greatest gains in precision of density estimates will be achieved by increasing the number of transects rather than increasing the number of quadrats sampled per transect.

Campbell et al. (1998) conducted analyses to optimize a two-stage sampling design for Geoduck surveys. A number of transect locations are randomly chosen in the first stage and a number of quadrats (along a transect) are surveyed in the second stage. Optimal sampling design for a two-stage sampling procedure is dependent on the allocation of available resources between each stage as well as the variability at each stage (Campbell et al. 1998). For Geoduck transects, surveying more transects with fewer quadrats per transect was preferable to surveying every quadrat along a transect and surveying fewer transects. It was therefore recommended to use a two-stage sampling design where primary sampling units (transects) are randomly chosen with systematically placed quadrats sampled along a transect; sampling interval between quadrats is dependent on transect length (Survey Type 4 in Campbell et al. 1998). This is the survey design currently used for Geoduck surveys. A similar survey design was used in DFO nearshore habitat mapping dive surveys (Davies et al. 2018).

For urchin surveys conducted up to 2010 in BC, some sampling (density, substrate, depth, algae within 1 m^2 quadrats) generally occurred at least every second metre (i.e., every second quadrat) along a transect, with sampling of size measurements occurring less frequently, depending on urchin density and transect lengths (e.g., Waddell and Perry 2012). This protocol required recording data at every second quadrat along a transect which, in the case of long transects, resulted in a large number of quadrats. This protocol was time consuming to survey and limited the number of transects that could be surveyed, thereby decreasing precision of survey density estimates. Furthermore, some preliminary data were required to determine the sampling frequency of size measurements. In 2011, the Green Sea Urchin survey protocol was modified to have a systematic quadrat skipping scheme that was dependent on transect length (DFO 2014). Analyses showed that similar density estimates were obtained from the subsampled datasets and the full datasets, and the modified survey protocol led to improved efficiency (DFO 2014).

Based on the research from Skibo et al. (2008), Campbell et al. (1998) and DFO (2014) reviewed above, a two-stage sampling design was selected for multispecies dive surveys, with randomly selected transect locations in the first stage and systematically spaced quadrats sampled along transects in the second stage.

It was expected that quadrats would take approximately two minutes each to survey. Having a sampling scheme that targets a maximum of 25 quadrats on most transects would translate into dives lasting up to 50 minutes, which is logistically feasible and should allow for completion of a transect on a single SCUBA tank in most cases. The spacing between quadrats was capped at sampling every 5th quadrat, which corresponds to stopping at every cable tie along the transect line. Therefore, the sampling scheme is dependent on transect length. For example, when the transect length is 0-25 m, every quadrat is sampled; when the transect length is 25-50 m, every 2nd quadrat is sampled; when the transect length is 50-75 m, every 3rd quadrat is sampled, and so on (Table 2).

Table 2.Sampling scheme, quadrat skipping pattern, minimum and maximum number of sampled quadrats as a function of transect length (m) and number of sections of lead line.

Transect Length (m)	# Sections of Lead Line	Sampling Scheme	Quadrat Skipping Pattern	Min # Quadrats	Max # Quadrats
0 – 25 m	1	every quadrat	No skip	14.0*	25
25 – 50 m	2	every 2nd quadrat	Skip 1	12.5	25
50 – 75 m	3	every 3rd quadrat	Skip 2	16.7	25
75 – 100 m	4	every 4th quadrat	Skip 3	18.8	25
100 – 125 m	5	every 5th quadrat**	Skip 4	20.0	25

**: For -2 m to 12 m target depth range, minimum transect length for vertical wall.*
***: Sample at every cable tie.*

Frequency distribution of transect length from nearshore habitat mapping surveys conducted between 2013 and 2015, Red Sea Urchin surveys completed between 1994 and 2015 and Sea Cucumber surveys conducted between 1997 and 2015 were used to guide the choice of maximum transect length. These survey types were selected because they use the same transect location assignment method as the multispecies dive survey, i.e., random transect placement along the shoreline.

Nearshore habitat mapping transects (Davies et al. 2018) were conducted down to 18.3 m (60 feet) gauge depth. The average transect length for 800 randomly placed transects conducted as part of the nearshore habitat mapping project between 2013 and 2015 was 88 m with a median of 65 m. The majority of transects (82%) were less than 125 m long (Table 3, Figure 1). Over half (52%) of transects were between 25 and 75 m long.

Table 3. Count and frequency distribution of dive survey transect lengths for 800 nearshore Habitat Mapping survey transects (2013 to 2015); 2,576 Red Sea Urchin survey transects (1994 to 2015) and 10,880 Sea Cucumber survey transects (1997 to 2015). "Average" columns are average value between survey types (non-weighted). "Cum." refers to cumulative frequency.

Transect Length (m)	Habitat Mapping			Red Sea Urchin			Sea Cucumber			Average	
	#	% Frequency		#	% Frequency		#	% Frequency		% Frequency	
	Trans	Bin	Cum.	Trans	Bin	Cum.	Trans	Bin	Cum.	Bin	Cum.
0 - 25	62	7.8	7.8	446	17.3	17.3	2,525	23.2	23.2	16.1	16.1
26-50	261	32.6	40.4	692	26.9	44.2	4,421	40.6	63.8	33.4	49.5
51-75	155	19.4	59.8	453	17.6	61.8	1,733	15.9	79.8	17.6	67.1
76-100	91	11.4	71.1	283	11.0	72.7	913	8.4	88.2	10.3	77.3
101-125	76	9.5	80.6	243	9.4	82.2	420	3.9	92.0	7.6	84.9
126-150	40	5.0	85.6	120	4.7	86.8	283	2.6	94.6	4.1	89.0
151-200	49	6.1	91.8	154	6.0	92.8	271	2.5	97.1	4.9	93.9
201-250	28	3.5	95.3	86	3.3	96.2	208	1.9	99.0	2.9	96.8
>250	38	4.8	100.0	99	3.8	100.0	106	1.0	100.0	3.2	100.0
Totals	**800**	-	-	**2,576**	-	-	**10,880**	-	-	-	-

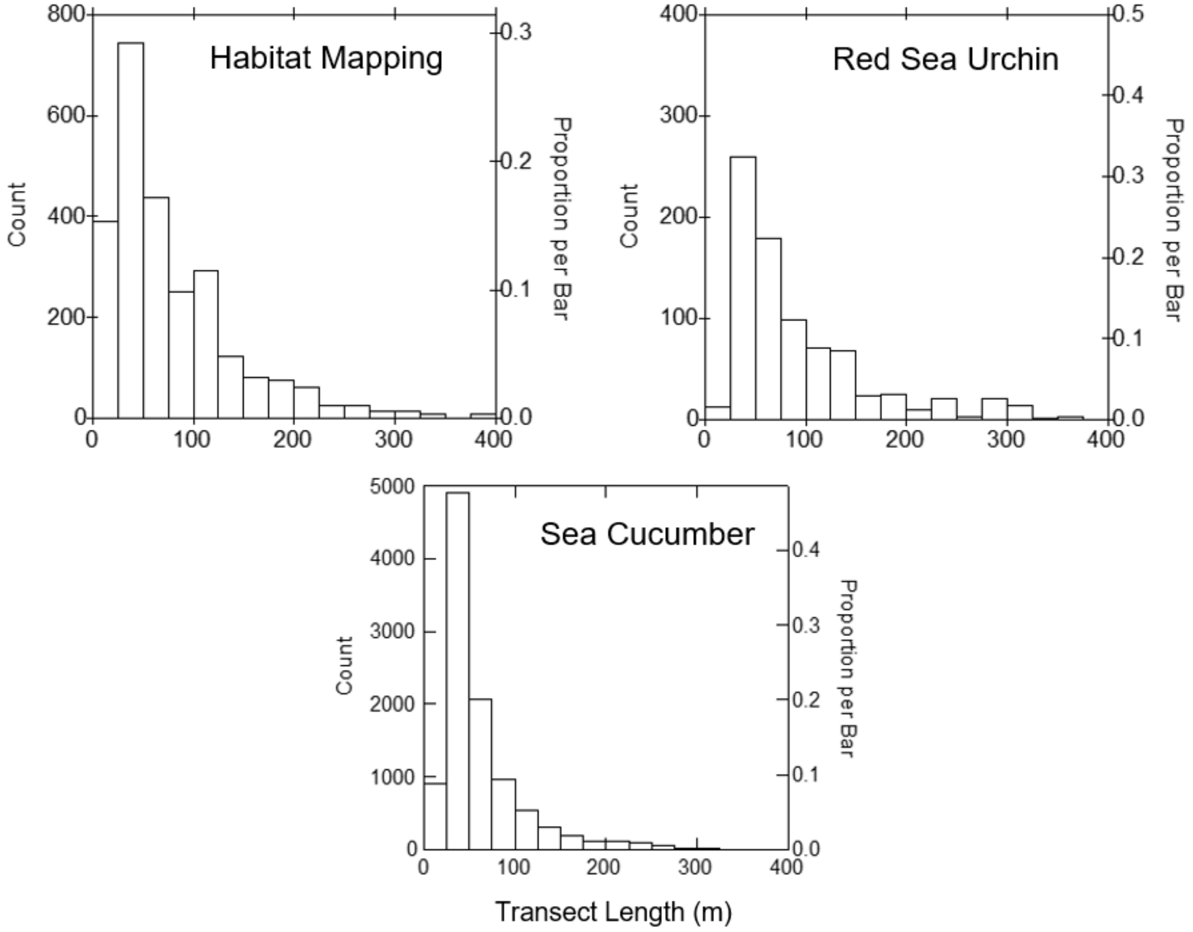

Figure 1. Transect length frequency distribution for Habitat Mapping survey transects (2013 to 2015); Red Sea Urchin survey transects (1994 to 2015) and Giant Red Sea Cucumber survey transects (1997 to 2015). Bins are 25 m wide.

For Red Sea Urchin surveys (Bureau et al. 2000a, 2000b, 2000c, 2000d) conducted throughout the BC coast between 1994 and 2015, average and median transect lengths were 84 m and 59 m, respectively, n = 2,576. Transect lengths for Red Sea Urchin surveys were less than 125 m for 82% of transects (Figure 1, Table 3).

Sea cucumber survey[2] (Duprey and Stanton 2018; Duprey and Stanton 2015; Duprey 2014; Duprey 2011) transects were on average the shortest (59 m, median = 40 m, n = 10,880; Figure 1, Table 3) of the survey types investigated. Transect lengths for sea cucumber surveys were less than 125 m for 92% of transects. Short transect lengths were expected for sea cucumber surveys because many of these surveys have taken place in steep inlets and channels.

Overall, across survey types, 85% of transects surveyed were 125 m or shorter. Maximum transect length for the multispecies dive survey was capped at 125 m, irrespective of whether the target maximum depth had been reached, based on past dive surveys and for diving logistical reasons, i.e., to be able to complete a transect on a single tank of air. Since transects are laid from the boat from shallow to deep, transects capped at 125 m run from the shallow target depth until the 125 m length limit is reached.

2.2. SURVEY LOCATIONS (2016-2021)

Multispecies benthic invertebrate pilot surveys were conducted using the survey protocol (Appendix A) on north- and south-eastern Vancouver Island, the mainland North Coast, south-east Haida Gwaii and the West Coast of Vancouver Island from 2016-2021 (Figure 2). The surveys were conducted in September each year. Trip IDs (numerical codes used to uniquely identify each survey in the database), year, survey locations, sublocations, number of transects and number of quadrats are summarized in Table 4.

In 2016, the first multispecies survey was conducted in the inside waters between northern Vancouver Island and mainland BC, in Pacific Fishery Management Area (PFMA) 12 (Sub-Areas 12-8, 12-10, 12-11, 12-13, 12-16, 12-41). A total of 131 transects were completed that year, 36 in the Deserters Group, 45 in the Gordon Islands, 44 in Wells Pass and 6 on the north coast of Malcolm Island (Figure 2). These areas were chosen by overlaying and visually inspecting the 2004-2014 commercial fishery footprints of the Red Sea Urchin, Green Sea Urchin, Giant Red Sea Cucumber and Geoduck fisheries. Areas were chosen based on overlap among the fisheries to ensure the species of interest were encountered on the survey and to represent a range of habitat types. Gordon Islands and the Deserters Group represent island habitats, Wells Pass represents inlet-type habitat and northern Malcolm Island represented sandy bay habitat.

In 2017, the multispecies dive survey was conducted in the inside waters between southern Vancouver Island and mainland BC and Washington in PFMAs 18 (Sub-Areas 18-2, 18-3, 18-4, 18-5, 18-6) and 20 (Sub-Areas 20-5, 20-6) (Figure 2). Forty-one transects were conducted in the Gulf Islands and 17 transects were conducted near Beecher Bay. The PFM Sub-Areas of interest were chosen by visually inspecting the fishery footprints and selecting areas with overlap between fisheries. In addition, the area near Beecher Bay was chosen because a single-species GSU survey was also being conducted at the same time in that area.

In 2018, the survey returned to the inside waters between northern Vancouver Island and the BC mainland, in PFMA 12 (Sub-Area 12-13) (Figure 2). Specifically, to the Deserters Group and Gordon Islands, which were surveyed in 2016. In 2018, 34 transects were surveyed in the Deserters Group: these consisted of 19 transects randomly selected from the 36 that were surveyed in 2016, plus an additional 15 new randomly placed transects. Fifty-four transects were surveyed around the Gordon Islands: 28 transects were randomly selected from the 45 that were surveyed in 2016, plus an additional 26 new randomly placed transects. This approach was taken to quantify interannual variability and inform recommendations on survey design (i.e., whether to repeat transect locations, re-randomize or some combination of the two; see Appendix C.3).

In 2019, the survey was conducted on BC’s mainland North Coast. A total of 86 transects were completed, 9 in PFMA 3 (Sub-Areas 3-1, 3-2, 3-4, 3-5), 61 in PFMA 4 (Sub-Areas 4-1, 4-2, 4-4, 4-5, 4-7, 4-8, 4-9, 4-10), and 16 in PFMA 5 (Sub-Area 5-10) (Figure 2). In addition to consideration of fishery footprint overlap as in previous years, areas of interest were also identified through engagement with Gixaala, Kitsumkalum, Lax Kw’alaams and Metlakatla Nations.

In 2020, the survey took place in south-east Haida Gwaii, in PFMA 2 (Sub-Areas 2-8, 2-9, 2-11, 2-12, 2-13, 2-14, 2-15, 2-16, 2-17, 2-18, 2-19) (Figure 2). Seventy-one transects were completed. DFO Science engaged with Parks Canada (PC) and the Archipelago Management Board (AMB) of the Haida Nation during survey planning. The Gwaii Haanas National Park Reserve, National Marine Conservation Area Reserve, and Haida Heritage Site was selected as the area of interest because new fishing closures were implemented in the ‘restricted access zones’ and the ‘strict protection zones’ of the park in 2018 (Gwaii Haanas Gina 'Waadluxan

KilGuhlGa Land-Sea-People Management Plan 2018) (Figure 2), thereby making 2019 optimal for the collection of baseline data. The number of transects allocated to the Gwaii Haanas 'strict protection zones' (where commercial fishing is prohibited) and the 'multiple use zones' (where some commercial fishing is allowed) was roughly proportional to the length of shoreline contained in each type (note that the 'restricted access zones' were excluded). Twenty-one transects were completed in 'strict protection zones' and 50 were completed in 'multiple use zones'. Ten of the 50 transects in the 'multiple use zones' were randomly selected out of 42 transects that were previously surveyed using the Red Sea Urchin survey protocol in 2010 and 2014.

In 2021, a total of 68 transects were completed in PFMAs 23 (Sub-Areas 23-4, 23-5, 23-7, 23-8, 23-9, 23-10), 24 (Sub-Areas 24-2, 24-4, 24-6, 24-8), 25 (Sub-Areas 25-4, 25-6, 25-12, 25-13, 25-15), 26 (Sub-Areas 26-2, 26-4, 26-5, 26-6) and 27 (Sub-Areas 27-3, 27-7) on the West Coast of Vancouver Island (WCVI) (Figure 2). DFO considered overlap in the fishery footprints in select PFM Sub-Areas to develop an initial suite of transect locations that were shared with the Ahousaht First Nation, Ehattesaht/Chinehkint First Nation, Hesquiaht First Nation, Hupacasath First Nation, Huu-ay-aht First Nation, Ka:'yu:'k't'h'/Che:k:tles7et'h' First Nation (Kyuquot), Mowachaht/Muchalaht First Nation, Nuchatlaht First Nation, Nuu-chah-nulth Tribal Council, Maa-nulth First Nations, Quatsino First Nation, Tla-o-qui-aht First Nation, Toquaht First Nation, Tseshaht First Nation, and Ucluelet First Nation, along with an invitation to provide feedback on transect locations. Any feedback that was received was incorporated into the final selection of the planned transects.

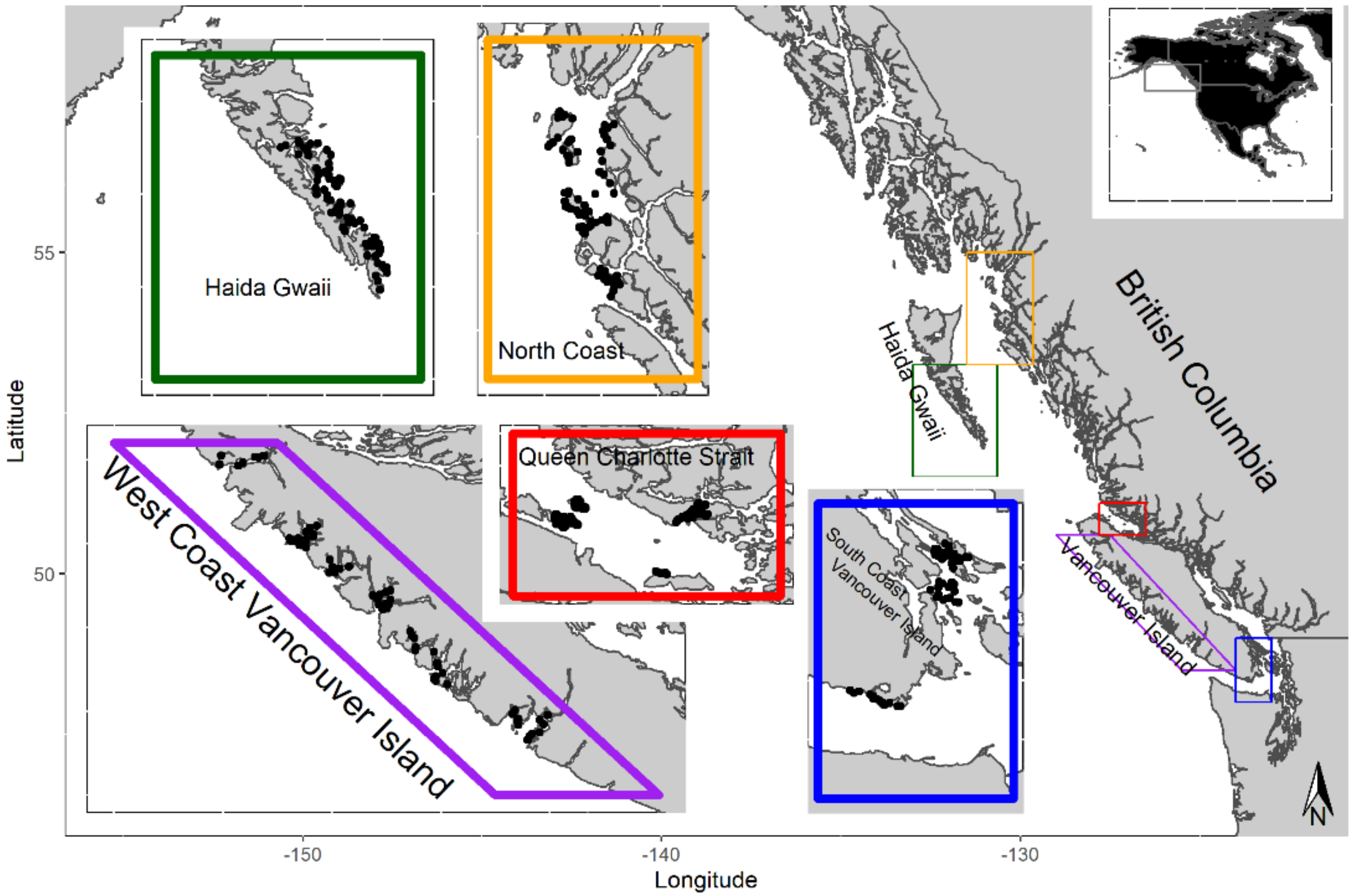

Figure 2. Map of survey regions and transect locations.

Table 4. Trip IDs (numerical codes used to uniquely identify each survey in the database), year, survey location, survey sublocations, total number of quadrats and total number of transects.

Trip ID	Year	Survey Locations	Sublocations	Number of transects	Number of quadrats
1	2016	North East Vancouver Island (Queen Charlotte Strait)	Deserters Group, Gordon Island, Malcolm Island, Wells Pass	131	2731
2	2017	South East Vancouver Island	Becher Bay, Gulf Islands	58	1290
3	2018	North East Vancouver Island (Queen Charlotte Strait)	Deserters Group, Gordon Island	88	1852
4	2019	Mainland North Coast	Kitkatla, Prince Rupert	80	1806
5	2020	Haida Gwaii	Gwaii Haanas	70	1541
6	2021	West Coast Vancouver Island	Barkley, Clayoquot, Nootka, Kyoquot, and Quatsino Sounds	68	1404
			Total:	**495**	**10624**

2.2.1. Transect locations (2016-2021)

Transect locations were randomly placed along the coastline for the survey areas of interest using the CHS_Pacific_High_Water_Coastline_Albers shapefile in ArcGIS prior to each survey. The anticipated total number of transects that could be completed on a survey was estimated by multiplying the number of dive days by 5 transects per dive skiff per day, and by the number of dive skiffs (one or two) plus 20. The 20 additional transects were to account for transects that would randomly be selected in locations where diving is not logistically feasible, e.g., intertidal areas, or locations considered unworkable due to high-currents or physical obstructions, such as log booms and docks.

Areas were selected for each survey (Section 2.2) and a mask of the survey area was created in ArcGIS. The shoreline for the area of interest (within the mask) was clipped, dissolved and random points were then assigned along the dissolved survey shoreline in ArcGIS. The randomly selected transect locations were then reviewed and transects in non-navigable areas (e.g., intertidal zone, rivers, tidal lagoons) were eliminated.

Fetch is defined as the distance traveled by wind or waves across open water and can be used as an estimate of exposure of a site to wave action. In areas of high fetch, large waves can routinely build making it too dangerous to conduct dive surveys. In areas of low fetch in the nearshore, soft muddy or silty sediments can accumulate and water can be relatively stagnant. In addition, the species of interest are often not found in very sheltered locations.

Fetch values have been estimated for points located every 50 m along the BC coastline and are available in the Fetch_all_BC geodatabase from the Open Data Portal (Fetch all BC Geodatabase). Total Fetch was estimated as the sum of the distance from a point to the nearest land at each of 72 compass bearings (every 5 degrees) to a maximum distance of 200 km for each direction.

Starting in 2020, the high-water coastline shapefile was masked to regions > 20,000 m fetch to remove low exposure areas since preliminary data from 2016 to 2019 surveys suggested that the probability of encountering the species of interest was low in these areas. This was anticipated to increase survey efficiency by eliminating areas of unsuitable habitat. A 26 m buffer was created around fetch points (halfway to the next fetch point + 1 m) to ensure adjacent buffers connected and dissolved together over the entire shoreline shapefile. Sections of shoreline with fetch < 20,000 m were clipped out. Then transects were randomly assigned to the remaining areas of interest as random points along the shoreline, using ArcGIS.

2.3. INVERTEBRATE DENSITIES

2.3.1. Invertebrate density estimation - Methods

The number of individuals for each invertebrate species was counted for each surveyed quadrat in a transect and the density on transect *i* was estimated as:

$$\widehat{D}_i = \frac{\sum_{quadrats} C_{ij}}{n}$$

where C_{ij} is the (adjusted for size categories in some cases) count on quadrat *j* on transect *i* and *n* is the number of surveyed quadrats on a transect and is equal to the area surveyed on each transect, since quadrats are 1 m^2.

The transect is considered as the sampling unit since quadrats along a transect may not be independent of one another.

There are two common ways to compute the overall density. First is a simple mean of the individual transect densities, which gives all transects equal weight. Second is a ratio estimator, which gives longer transects a higher weight (see Thompson et al. 1992 for estimator and estimated standard errors). If the transects are all the same length, then the estimators are identical.

We used the simple mean of the individual transect densities when deriving the sampling plans because of the greatly simplified computation. The results will be similar if the ratio estimator is used because transect-to-transect variation is large and the variation in transect-lengths is relatively small. For more accurate estimates of density, the ratio estimator should be used as is currently the practise in the analyses of dive data from DFO single-species surveys (Hajas et al. 2023; Lochead et al. 2015; Bureau et al. 2012; Hand et al. 2009).

In this document, we use the term “overall density” instead of “mean density” for the following reasons. Firstly, the density is already a mean (number of individuals per area). Secondly, there are instances where density is calculated for individual quadrats, and at the transect level, the site level, and the population level. The density at the transect level is a ratio of total counts to total area measured. The density at the site level is not a simple mean of the transect densities, for example if a ratio estimator is used to weight by transect length. The density at the population level could also be a weighted mean of the site densities by site area, etc.

2.3.2. Size-, habitat-, and zone-specific densities - Methods

Four species of invertebrates (Giant Red Sea Cucumbers, GSU, RSU, and Geoduck) surveyed have existing reference points (Table 1). Giant Red Sea Cucumbers and RSU have coast wide reference points and their stock status is intended to be evaluated using the multispecies benthic invertebrate monitoring program. Note that GSU reference points were derived from high density index sites that are located on the south coast, where the GSU fishery operates, and may not be appropriate coast wide. GSU stock status is currently evaluated using the index site data (i.e., stock status is not currently assessed coast wide) (DFO 2016a, 2018, 2021c). Also, due to the timing of the multispecies survey (generally in September) the survey will not be used to assess Geoduck stock status (more in Section 5: Uncertainties and Section 6: General Discussion). Therefore, only Giant Red Sea Cucumber and RSU densities are provided below, as an example of how the multispecies benthic invertebrate survey data can be used to evaluate coast wide stock status.

Giant Red Sea Cucumbers have reference points (LRP and USR) based on their density and habitat (Hajas et al. 2023). Size and habitat-based reference points (LRP and USR) have been defined for Red Sea Urchins based on the density of mature individuals on urchin habitat (Lochead et al. 2019). Specifically,

Giant Red Sea Cucumber:

LRP = 0.029 sea cucumbers per m^2 (on sea cucumber habitat)

USR = 0.038 sea cucumbers per m^2 (on sea cucumber habitat)

RSU:

LRP = 0.30 mature (≥ 50 mm test diameter(TD)) RSU per m^2 on urchin habitat.

USR = 0.60 mature (≥ 50 mm TD) RSU per m^2 on urchin habitat.

All quadrats were included in the computation of Giant Red Sea Cucumber density estimates, because Giant Red Sea Cucumber habitat was not considered in this paper (note that “on sea cucumber habitat” was added to the reference points for sea cucumber after much of this work had been completed, see DFO 2022e). Only quadrats defined as urchin habitat were included in the calculation of Red Sea Urchin density estimates. As described in Lochead et al. (2019), only including urchin habitat for Red Sea Urchins makes the estimates of mature RSU densities comparable to other studies where urchin habitat is targeted. We implemented the set of rules described in Lochead et al. (2019) to define urchin habitat from the substrate data and then removed non-urchin habitat quadrats within surveyed transects from the density calculations for Red Sea Urchins, only. Urchin habitat was defined as a substrate of gravel or larger, where mud is not the predominant substrate. These are substrates that urchins can hold onto with their tube feet. Specifically, this included any occurrence of smooth bedrock, bedrock with crevices, boulders, cobble, gravel, shell, crushed shell, or whole shell in the primary or secondary substrate categories. Any quadrats where the primary substrate was mud, regardless of other substrate categories, were removed.

Red Sea Urchin densities were further refined by size category. The number of individuals per quadrat was adjusted based on the proportion above size at maturity (≥ 50 mm TD). If no size-size frequency data was available for a quadrat, then the size-frequency data for that transect was used.

The density for each trip-transect-species was determined by the ratio of the total (adjusted) count of animals ≥ minimum-size over the quadrats surveyed divided by the number of quadrats surveyed.

As an example of how the multispecies data may also be useful for monitoring zone effectiveness in marine protected areas such as The Gwaii Haanas National Park Reserve, National Marine Conservation Area Reserve, and Haida Heritage Site, invertebrate densities (# per m^2) were also estimated by zone (i.e., multiuse zone where commercial harvest is allowed, and strict protection zone where commercial harvest is not allowed).

2.3.3. Density and size on each transect - Results

Northern Abalone results are not reported by transect, but are rolled up by region in this document. As illegal harvest is the most significant threat to Northern Abalone recovery, geospatial information that details Northern Abalone locations is considered sensitive information, and is not available for public release pursuant to Section 124[3] of the *Species at Risk Act* (SARA).

The distribution of density for all species is skewed with most transects showing low densities (Figure 3). Transects with densities of 5 per m^2 or higher were only observed for Green Sea Urchin and Red Sea Urchin.

Densities of Geoduck were generally lower in south east Vancouver Island and West Coast Vancouver Island compared to the other regions (Figure 4), however caution should be used in interpreting these results because September is not an optimum time for estimating Geoduck abundance (see Section 1). Densities of Geoduck ranged from 0 to 4.35 per m^2 and were highly variable over short distances of shoreline (Figure 4). The overall density was 0.241 (SE = 0.072) per m^2 in the North Coast of BC, but across all the surveys the median density was 0 per m^2 (Table 5).

The Giant Red Sea Cucumber density was typically less variable (indicated by a lower relative standard error) than the other species of invertebrates surveyed here (Table 5). Their densities were relatively high, with means ranging from 0.084 (SE = 0.018) per m^2 on the south east coast of Haida Gwaii to 0.283 (SE = 0.053) per m^2 on West Coast Vancouver Island (Table 5). The spatial variability in Giant Red Sea Cucumber densities was generally lower than that of the other species and the number of zero observations was less (Figure 5).

Northern Abalone densities were highest on the east coast of Haida Gwaii (mean 1.267 (SE 0.179) per m^2). Northern Abalone densities were the lowest in south east Vancouver Island and the West Coast Vancouver Island (Table 5). Variability of Northern Abalone density was relatively low, with relative standard errors (RSEs) of between 0.14 and 0.32.

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[3] The Minister, on the advice of COSEWIC, may restrict the release of any information required to be included in the public registry if that information relates to the location of a wildlife species or its habitat and restricting its release would be in the best interests of the species.

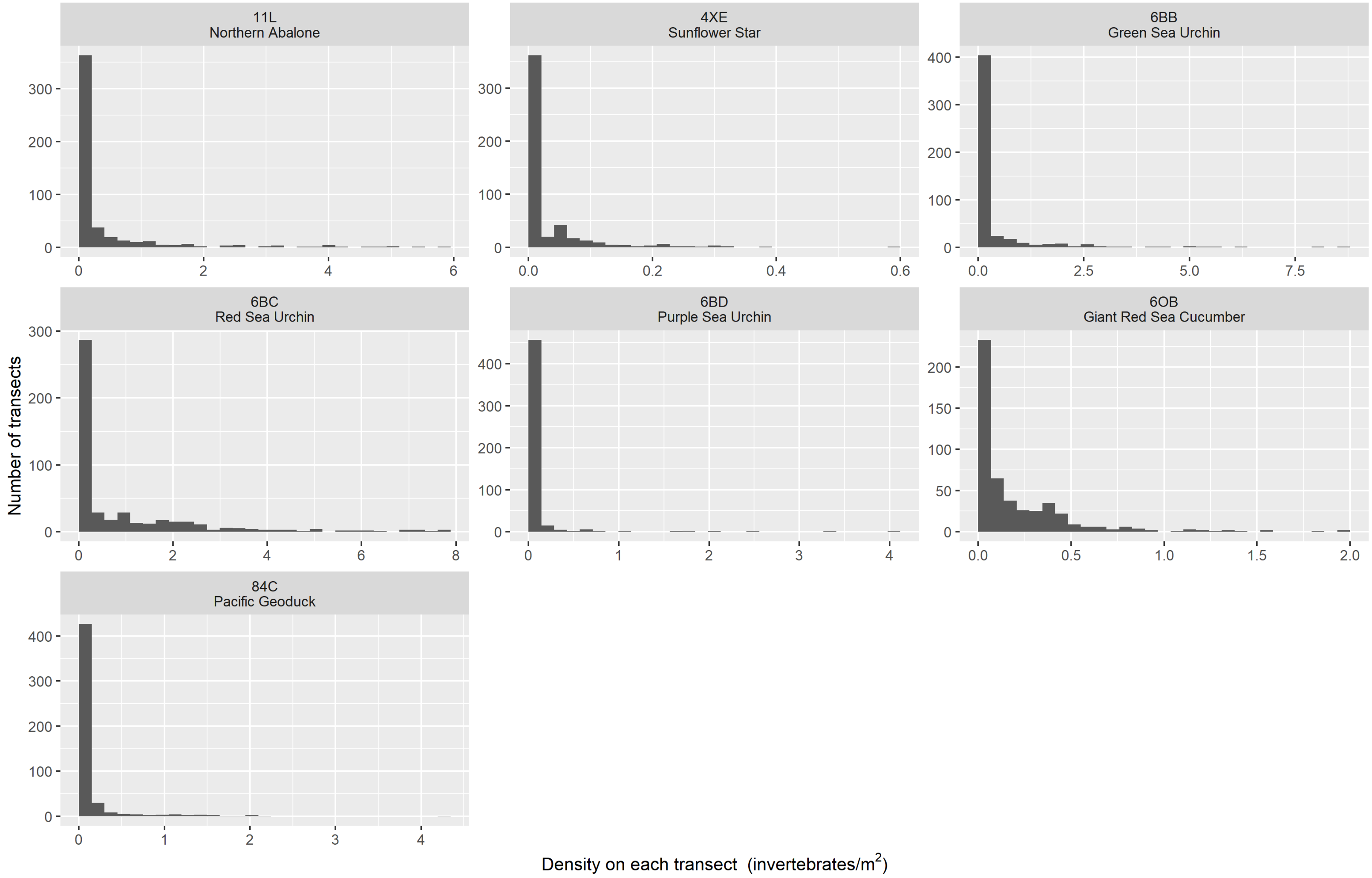

Figure 3. Distribution of transect densities for all species.

The range of mean densities for Red Sea Urchins was considerable across the different regions, with the lowest densities in parts of north east Vancouver Island, south east Vancouver Island and West Coast Vancouver Island (Figure 6). Mean Red Sea Urchin densities were higher in the North, i.e., on the east coast of Haida Gwaii (mean = 3.188 (SE 0.367) per m^2) and the North Coast (mean = 2.239 (SE 0.302) per m^2), than in southern BC (Table 5).

Green Sea Urchin densities were highest on the North Coast (mean = 0.926 (SE 0.095) per m^2) and on north east and south east Vancouver Island (mean = 1.219 (SE 0.145) per m^2) (Figure 7). Their densities were the lowest on West Coast Vancouver Island (mean = 0.008 (SE 0.003) per m^2) (Table 5).

The highest densities of Purple Sea Urchins (PSU) were found in southeast Haida Gwaii (mean = 0.177 (SE 0.066) per m^2), and West Coast Vancouver Island (mean = 0.173 (SE 0.070) per m^2), and PSU were virtually absent from the North Coast, and parts of north east and south east Vancouver Island (Figure 8). The density of Purple Sea Urchins on the North Coast (mean = 0.001 (SE 0.001) per m^2) was the lowest of any of the invertebrates surveyed (Table 5). The Purple Sea Urchin had the overall highest variability (RSE's ranging from 0.32 to 1.0) of the invertebrates surveyed.

As expected, due to the impacts of sea star wasting disease, Sunflower Stars were uncommon (Figure 9). The highest densities were observed on the North Coast (mean = 0.024 (SE = 0.006) per m^2) and north east and south east Vancouver Island (mean = 0.040 (SE 0.005) per m^2) (Table 5).

The size distributions of measured invertebrate species (Red Sea Urchin, Green Sea Urchin, Purple Sea Urchin, Northern Abalone, Sunflower Star and Giant Red Sea Cucumber) were similar across regions within species (see Appendix B for detailed size distributions). The median test diameter of Red Sea Urchins was 60 mm (range = 2 - 229 mm). The median size of Green Sea Urchins was 48 mm (range = 1 – 100 mm). The median size of Purple Sea Urchins was 52.5 mm (range = 12 - 152 mm). The median size of Northern Abalone was 55 mm (range = 4 - 139 mm). Sunflower Stars were the largest invertebrates observed, with a median diameter of 104 mm (range = 13 - 885 mm). The median of split weight of Giant Red Sea Cucumbers on the east coast of Haida Gwaii was 486.5 g (range = 38 – 1,384 g), and on West Coast Vancouver Island the median was 204.5 g (range = 18 – 460 g) (these were the only two surveys that collected split weight data; see Appendix A: Multispecies Dive Survey Protocol).

Table 5. Mean, median, standard error (SE), range, and relative standard error (RSE) of invertebrate densities (# per m^2) by survey location.

Species	Location	Transects	Mean	Median	SE	Range	RSE
Geoduck	SE Haida Gwaii	70	0.105	0	0.037	0, 2.00	0.35
	NE and SE Vancouver Island	277	0.081	0	0.016	0, 1.96	0.20
	North Coast BC	80	0.239	0	0.072	0, 4.35	0.30
	West Coast Vancouver Island	68	0.025	0	0.011	0, 0.61	0.44
Giant Red Sea Cucumber	SE Haida Gwaii	70	0.084	0.040	0.018	0, 0.91	0.21
	NE and SE Vancouver Island	277	0.194	0.105	0.015	0, 1.44	0.08
	North Coast BC	80	0.246	0.082	0.043	0, 2.00	0.17
	West Coast Vancouver Island	68	0.283	0.069	0.053	0, 1.94	0.19
Green Sea Urchin	SE Haida Gwaii	70	0.398	0.122	0.068	0, 3.04	0.17
	NE and SE Vancouver Island	277	1.219	0.167	0.145	0, 17.84	0.12
	North Coast BC	80	0.926	0.095	0.224	0, 13.43	0.24
	West Coast Vancouver Island	68	0.008	0	0.003	0, 0.10	0.38
Northern Abalone	SE Haida Gwaii	70	1.267	0.849	0.179	0, 5.95	0.14
	NE and SE Vancouver Island	277	0.091	0	0.014	0, 1.76	0.15
	North Coast BC	80	0.718	0.085	0.134	0, 5.13	0.19
	West Coast Vancouver Island	68	0.098	0	0.031	0, 1.50	0.32
Purple Sea Urchin	SE Haida Gwaii	70	0.177	0	0.066	0, 4.12	0.37
	NE and SE Vancouver Island	277	0.034	0	0.011	0, 2.11	0.32
	North Coast BC	80	0.001	0	0.001	0, 0.05	1.00
	West Coast Vancouver Island	68	0.173	0	0.070	0, 3.33	0.40
Red Sea Urchin	SE Haida Gwaii	70	3.188	2.092	0.367	0, 10.89	0.12
	NE and SE Vancouver Island	277	0.748	0.087	0.084	0, 8.44	0.11
	North Coast BC	80	2.228	1.250	0.302	0, 12.08	0.14
	West Coast Vancouver Island	68	0.385	0.047	0.092	0, 3.36	0.24
Sunflower Star	SE Haida Gwaii	70	0.001	0	0.001	0, 0.05	1.00
	NE and SE Vancouver Island	277	0.04	0	0.005	0, 0.60	0.13
	North Coast BC	80	0.024	0	0.006	0, 0.32	0.25
	West Coast Vancouver Island	68	0.009	0	0.003	0, 0.20	0.33

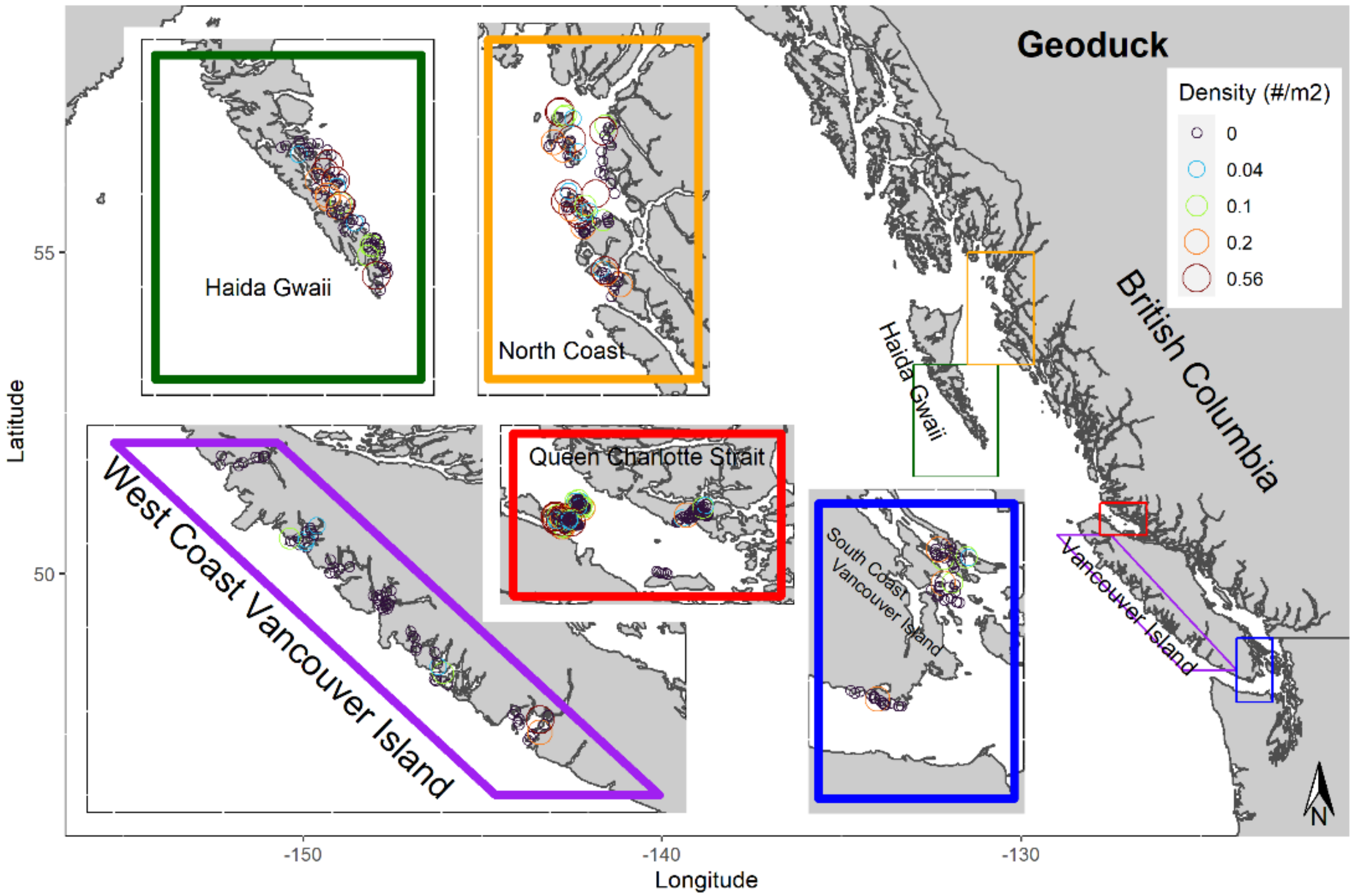

Figure 4. Geoduck densities by transect.

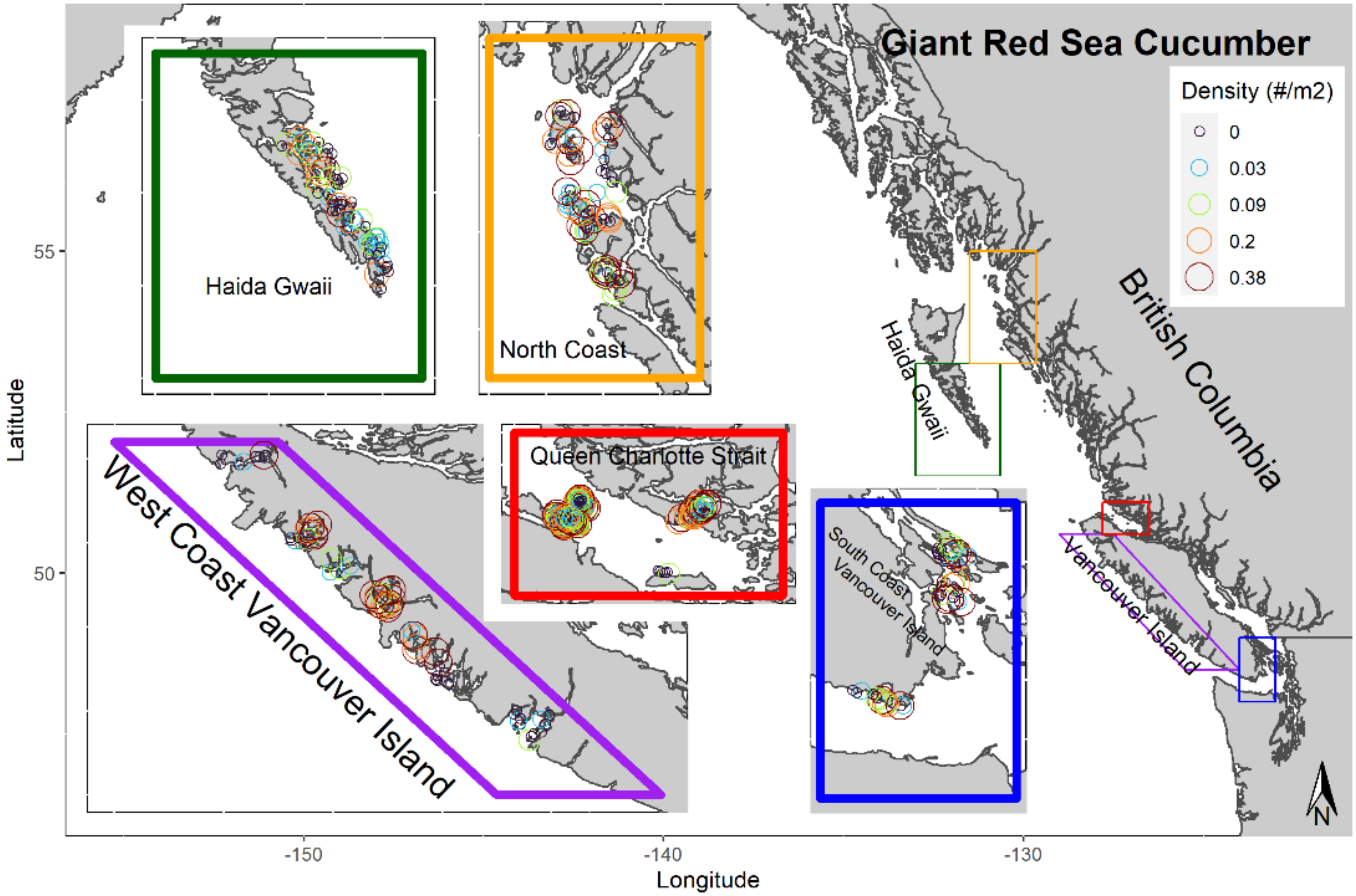

Figure 5. Giant Red Sea Cucumber densities by transect.

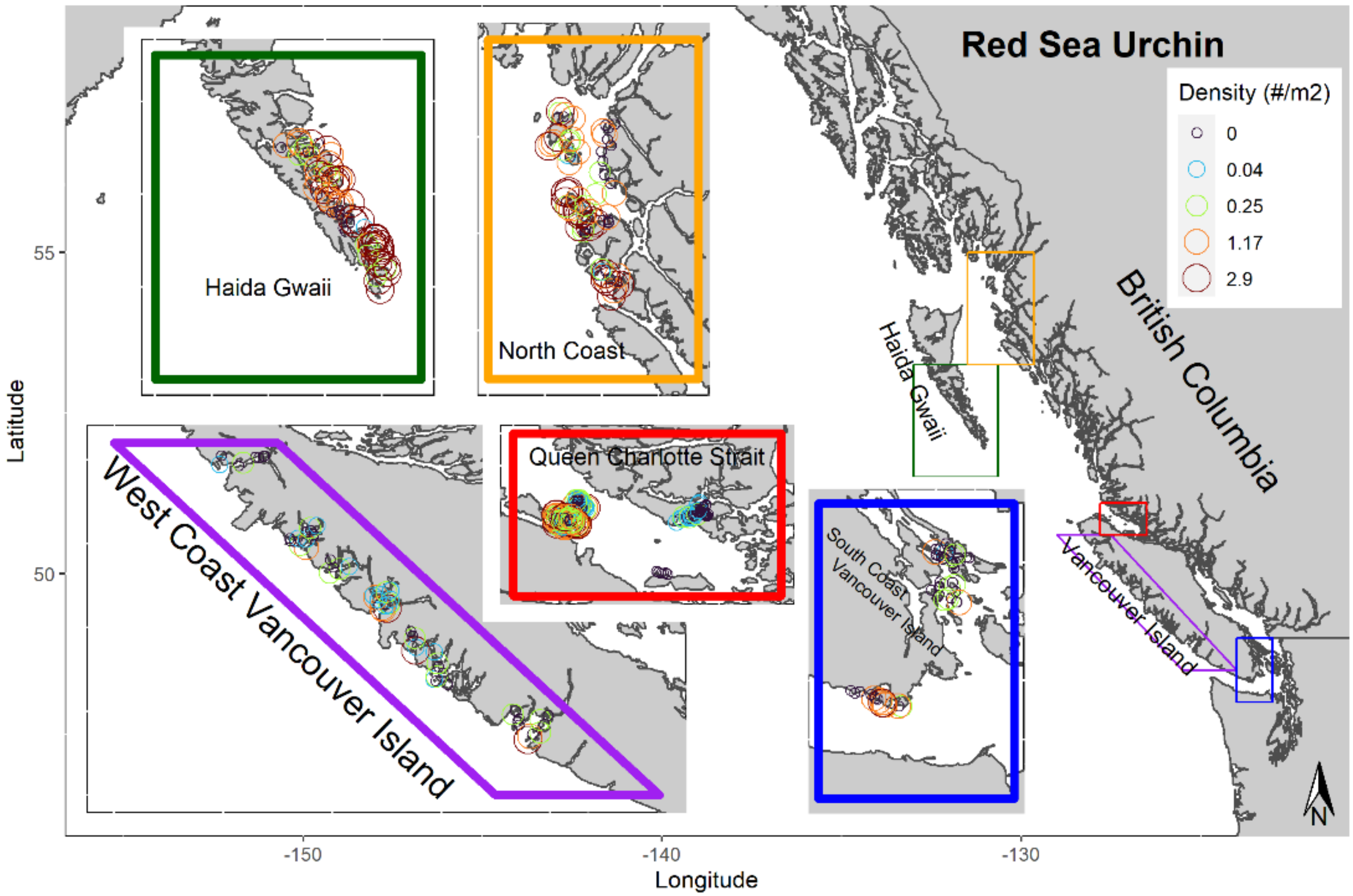

Figure 6. Red Sea Urchin densities by transect.

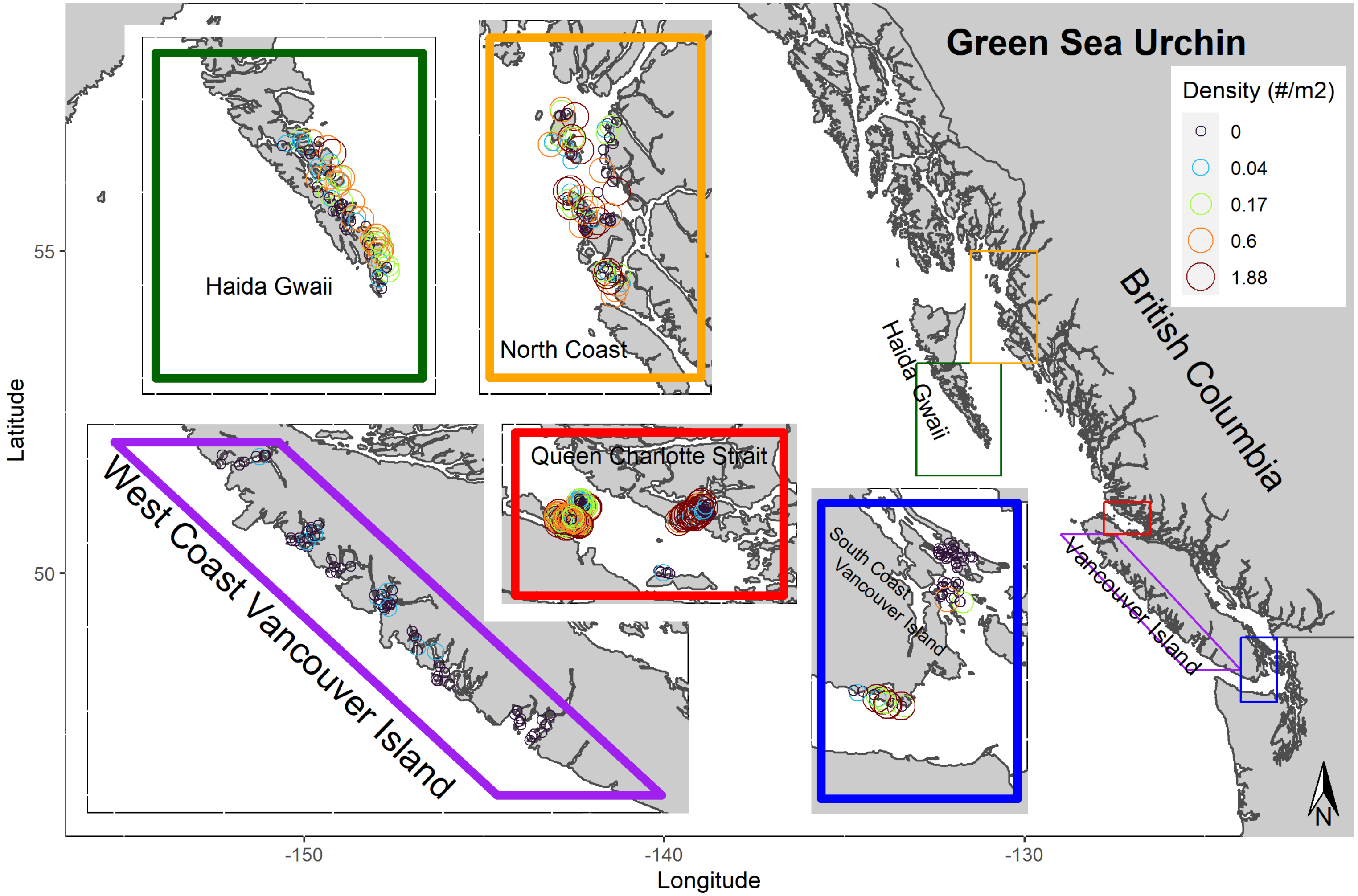

Figure 7. Green Sea Urchin densities by transect.

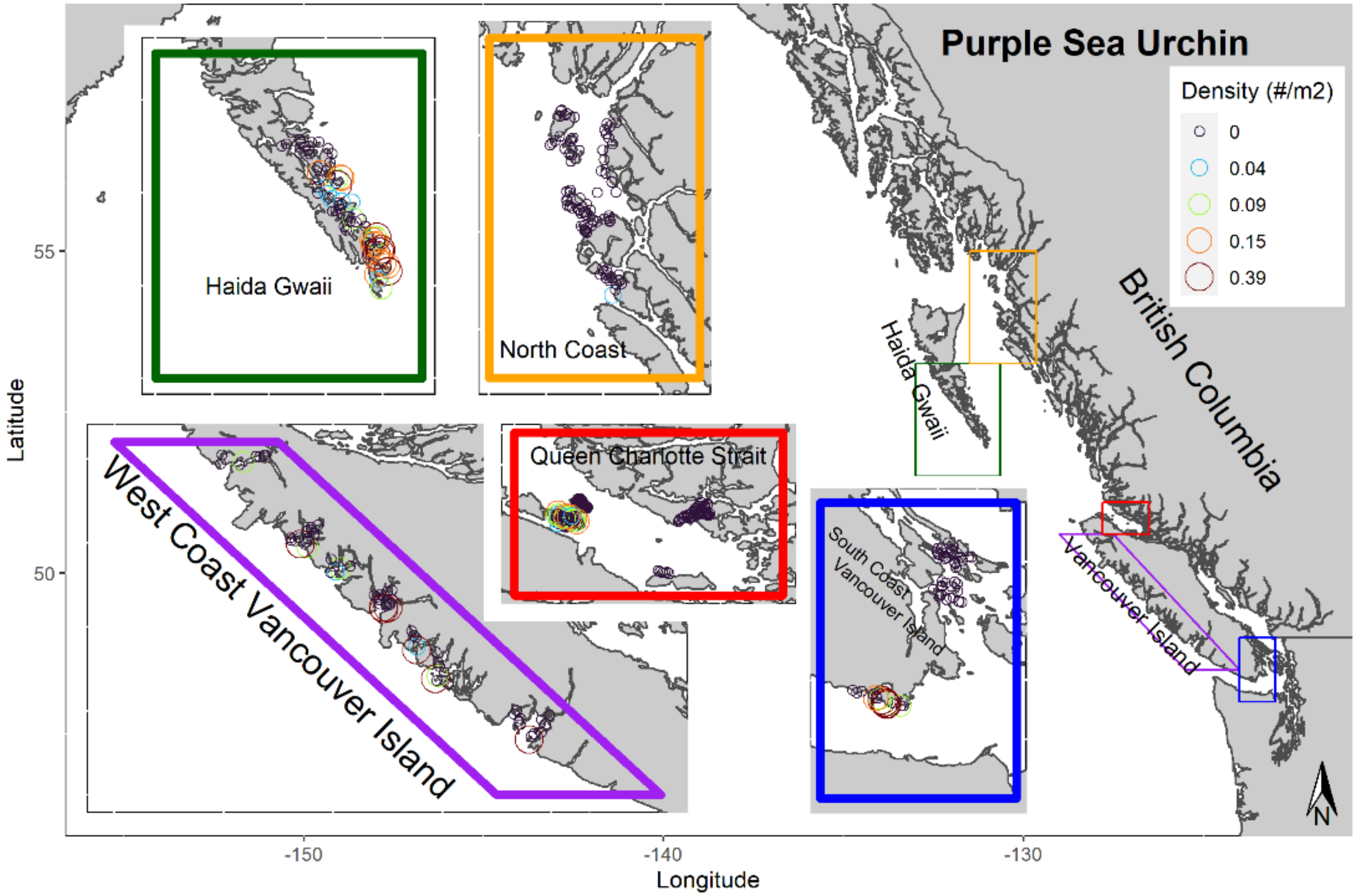

Figure 8. Purple Sea Urchin densities by transect.

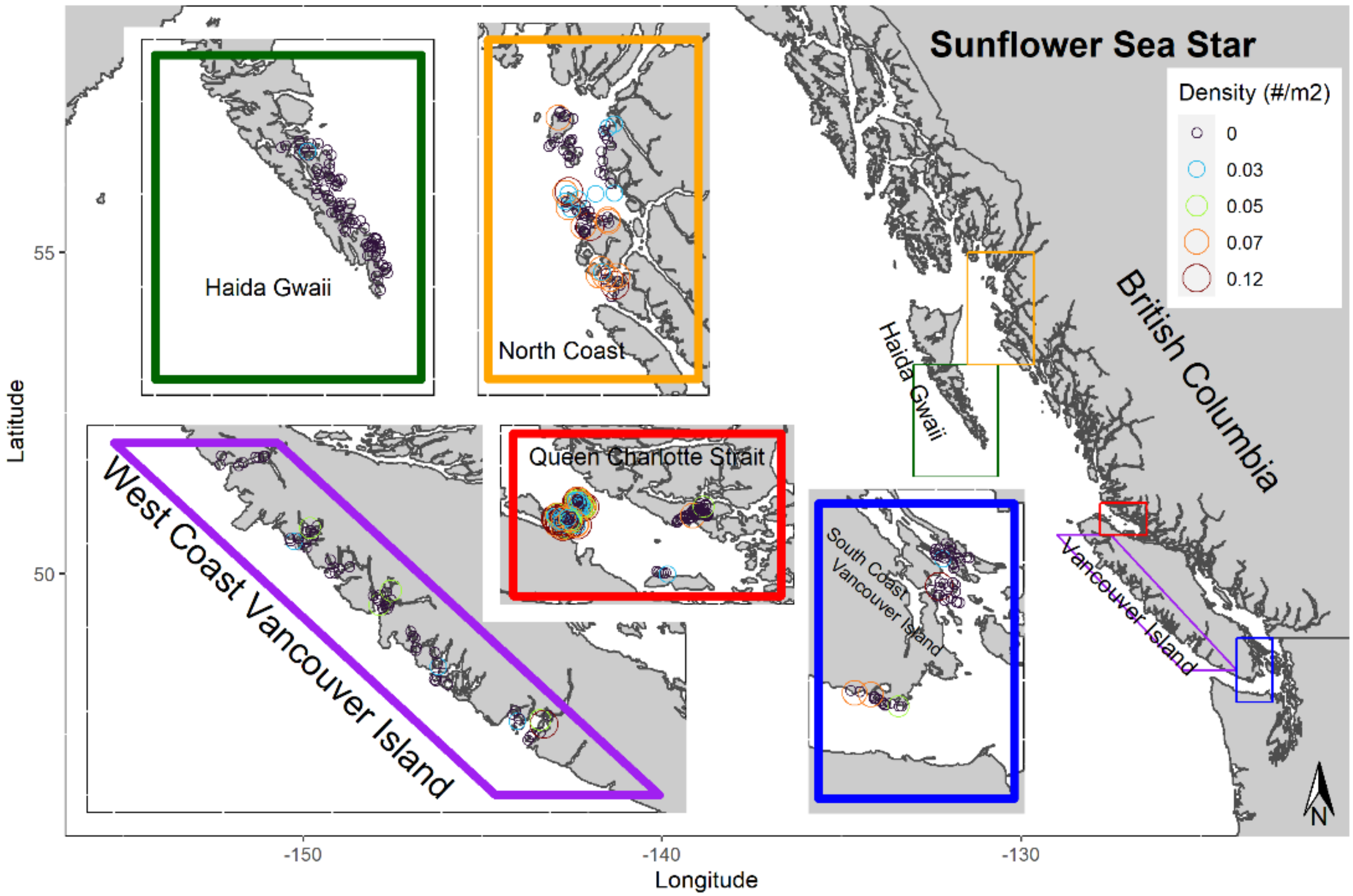

Figure 9. Sunflower Star densities by transect.

2.3.4. Size, habitat and zone-specific densities - Results

The coast wide estimate of Red Sea Urchin density was 0.90 per m^2 on urchin habitat, and the coast wide estimate of Giant Red Sea Cucumber density was 0.193 per m^2 (on all habitat) (Table 6; Figure 10).

Differences in species densities in multi-use and strict protection areas in The Gwaii Haanas National Park Reserve, National Marine Conservation Area Reserve, and Haida Heritage Site were tested using a 2-factor ANOVA, and the results showed that there were significant differences in overall density among species ($p < 0.0001$), but there was no overall difference in invertebrate density between multi-use and strict protection areas ($p = 0.997$), nor were there significant differences between multi-use and strict protection areas for individual species densities (tested by the interaction term, $p = 0.875$) (Figure 11).

Table 6. Species densities for Giant Red Sea Cucumber and portions of the Red and Green Sea Urchin populations that are assessed against the limit reference point (LRP) and upper stock reference (USR) for management purposes.

Species	Location	Transects	Mean	SE
Giant Red Sea Cucumber (# adults/m²)	East Haida Gwaii	70	0.082	0.018
	NE and SE Vancouver Island	277	0.186	0.014
	North Coast BC	80	0.241	0.043
	West Coast Vancouver Island	68	0.278	0.051
Red Sea Urchin (# ≥ 50 mm TD/m² on urchin habitat)	East Haida Gwaii	70	2.211	0.259
	NE and SE Vancouver Island	277	0.527	0.067
	North Coast BC	80	1.566	0.211
	West Coast Vancouver Island	68	0.271	0.076

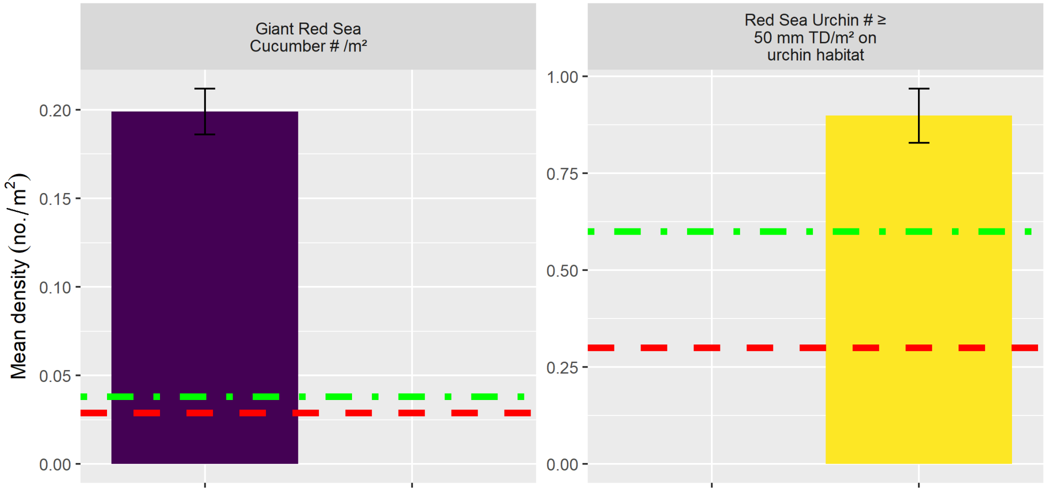

Figure 10. Densities of Giant Red Sea Cucumber and Red Sea Urchin with reference points pooled across regions and years (2016 to 2021). Error bars represent standard error, the USR for each species is shown as the green horizontal line and the LRP is shown as the red horizontal line. Note that the reference points for Giant Red Sea Cucumber should be applied to cucumber habitat only, however, here we have shown the density of Giant Red Sea Cucumber across all habitats.

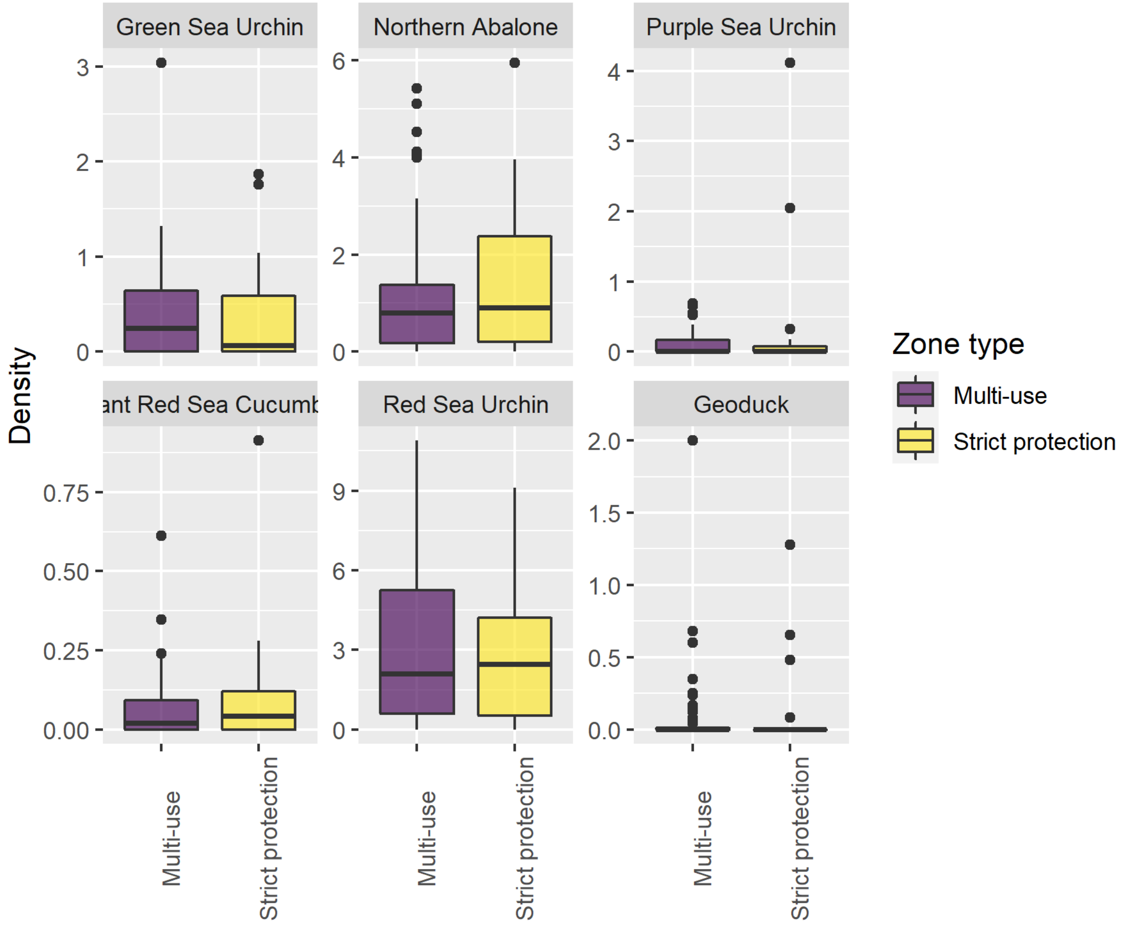

*Figure 11. Invertebrates densities (# per m^2) from surveys of the east coast of Haida Gwaii in strict protection zones (according to IUCN Standard II) and multi-use zones. Lines in the box and whisker plots are the mean densities, boxes represent the 25th and 75th percentiles, the whiskers are 1.5 * the distance between the first and third quartiles. Sunflower Star densities are not shown since there was only a single non-zero density for this species recorded during east coast Haida Gwaii surveys.*

2.3.5. Variability in density over all trips - Methods

The variation in observed transect densities among transects varies as a function of density, i.e., the variation in observed transect densities is smaller when density is low compared to when density is high. Derivation of the sampling plan (Section 4.1.1) will require information on how the transect-to-transect variation varies as a function of the density. The standard deviation (sd) and mean of the transect-level density estimates were calculated, and then the sd:mean ratio was estimated for each species, over all year-sublocations from Table 4 (except for 2021 West Coast Vancouver Island where sublocations were combined) using a simple linear model with no intercept. Differences among species were examined with a multiple comparison (a Tukey-like comparison).

2.3.6. Variability in density over all trips - Results

The relationship of the standard deviation of transect-specific densities (SD) for each year-sublocation to the mean density for each year-sublocation was similar for all species (Figure 12).

There is some evidence that the sd:mean ratio may vary among species. A multiple comparison showed evidence that the sd:mean relationship for RSU was different than the sd:mean relationship for GSU, but there was no evidence of differences among any of the other species (Appendix D: Supplementary Material). The differences in the sd:mean ratio were small, so a common sd:mean ratio was fit with the standard deviation of transect-specific densities estimated to be 1.27 times the mean density for all species. Again, note that the sd:mean ratio was only used in determining the sample plan (Section 4.1.1) so there was little need for any formal statistical analyses.

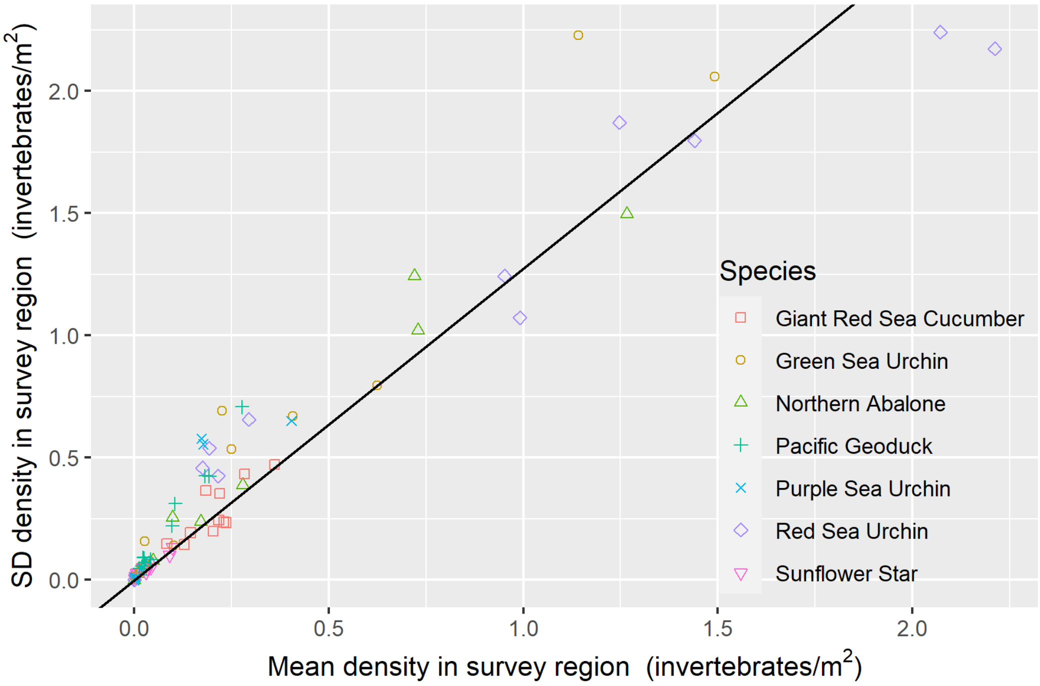

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Figure 12. Relationship between standard deviation (SD) of transect-specific densities in the survey region to mean density in survey region for all species. Different points for a given species represent unique year-sublocations from Table 4, except for 2021 West Coast Vancouver Island where sublocations were combined).

2.3.7. Expected precision - Methods

The relative standard error (RSE) for an estimator was computed as follows:

$$RSE = \frac{SE}{Estimate}$$

Many authors have suggested target RSE for certain purposes. For example, Krebs (1999) suggests targets of

> RSE = 25% for preliminary surveys so that the 95% CI is ± 50% of the estimate;
>
> RSE = 12% for management survey, so that the 95% CI is ± 25% of the estimate; and
>
> RSE = 5% for scientific surveys so that the 95% CI is ± 10% of the estimate.

Because the transect-to-transect variation depends on density, the sd:mean ratio derived above in Section 2.3.6 is needed to estimate the approximate relative standard error expected under different sample sizes using a simple mean estimator. Using a common (across species) sd:mean ratio of 1.27 derived from the sampling data (Figure 12), plots of relative standard error versus density (# invertebrates per m^2) were generated for sample sizes of 20, 30, 40, 50, 60, 100, 200, 250 and 300 transects, and compared to threshold values of RSE (5%, 12% and 25%) (Figure 13). These approximate estimates of RSE are useful for initial planning purposes to know the (approximate) sample size (number of transects) that will be needed to obtain estimates with a target RSE.

2.3.8. Expected precision - Results

Relative standard error decreases as sample size increases, and because the standard deviation in density among transects is a multiple of the density, the relative standard error also declines as density declines (Figure 13).

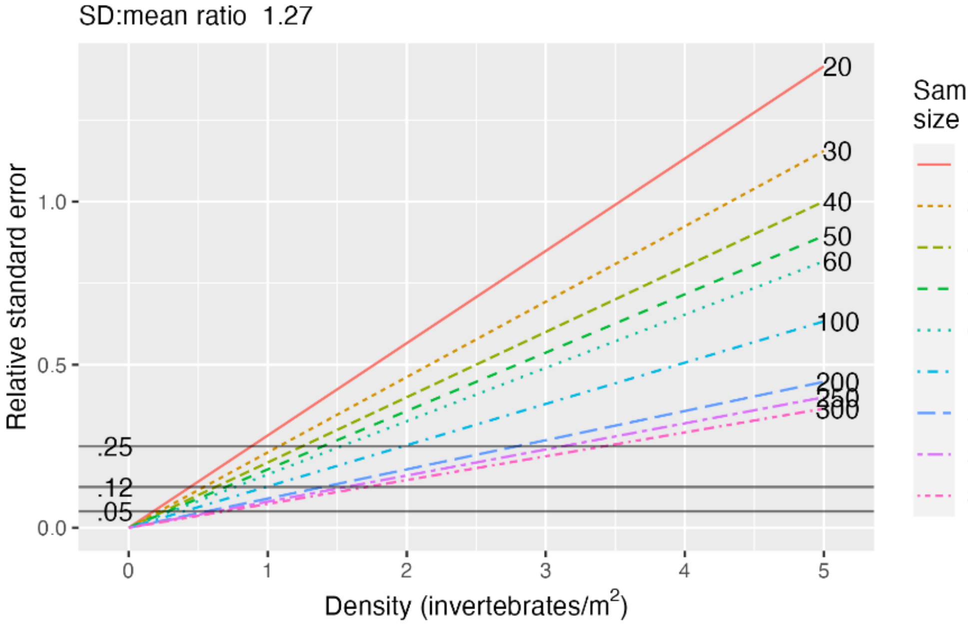

Figure 13. Relative standard error, with a standard deviation to mean ratio of 1.27, for sample sizes of 20, 30, 40, 50, 60, 100, 200, 250, and 300 transects. Horizontal black lines denote relative standard errors of 5%, 12% and 25%.

2.4. DISCUSSION

Design of the pilot surveys was based on previous dive survey data and experience. The protocol was tested through a series of pilot surveys conducted on a subset of areas of the BC coast each September from 2016-2021. The pilot surveys demonstrated the practical feasibility of the protocol, while also gathering preliminary information to guide the following protocol improvements (Section 3) and recommendations about the statistical design of the survey (Section 4).

3. USING PILOT SURVEY DATA TO INFORM PROTOCOL IMPROVEMENTS

3.1. ESTABLISHING FETCH CUTOFF VALUES

Fetch can be used to exclude sections of shoreline at the random transect location selection stage when preparing a survey. The species of interest are typically not found in areas of low fetch (e.g., mouths of rivers, intertidal areas), and furthermore, low fetch areas are often not navigable. Using a low fetch cutoff could then be useful when setting up a survey to exclude areas of unsuitable habitat. Similarly, sections of shoreline with high fetch values should not be considered for surveying, and should be excluded from the shoreline where transects are randomly placed, because diving at these locations is rarely possible due to wave action.

Fetch values are available for points located every 50 m along the entire BC coastline and are available in the Fetch all BC Geodatabase. The fetch data can be used at survey setup time to exclude sections of shoreline unsuitable for the survey, i.e., those sections of shoreline that are too sheltered or too exposed to wave action.

Preliminary data from the 2016 to 2019 pilot multispecies surveys (early version of Figure 17) were reviewed and expert opinion used to determine a low fetch cutoff value. A low fetch cutoff of 20,000 m was selected and sections of shoreline below the 20,000 m cutoff were excluded before selecting transect locations for the 2020 and 2021 pilot surveys.

An upper fetch cutoff was not explicitly determined prior to the pilot surveys, but was instead evaluated on a site-by-site basis based on weather conditions and the ability of divers to survey a site safely. A database of marine benthic invertebrate dive survey transects (n = 24,657) indicated that 95% of all invertebrate survey dives conducted in BC (across a range of species) from 1977-2021 had fetch values less than 2.52 million m (Figure 14). This suggests that an upper limit of 2.52 million m fetch could be considered as an upper limit to the sampling frame for the multispecies survey. The fetch value for the 95th percentile of dives, i.e., 2,520,000 m, is recommended as the upper fetch cutoff for setting up future multispecies dive surveys.

Using these values in combination with the fetch GIS layer and the coastline shape file, it is estimated that the distribution of fetch across the entire BC coast is from 0 to 9.87 million m (Figure 15). The median value of fetch for the BC coastline is 0.07 million m (mean = 0.35, SE = 0.80). About 76% of the coastline is between 20,000 m and 2.52 million m fetch and would be suitable for dive survey sampling (20.2% is less than 20,000 m and 3.8% is greater than 2.52 million m). An example of fetch cut off values along a section of the BC coastline is shown in Figure 16.

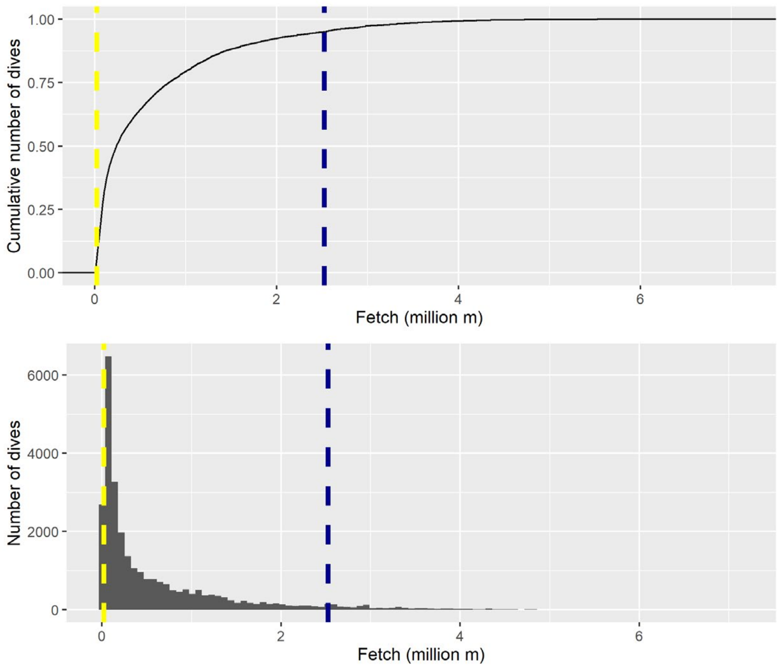

Figure 14. Cumulative number of dives (upper panel) and total number of dives (lower panel) conducted from 1977-2021 (n = 24,657) on DFO dive surveys in British Columbia. Yellow dashed line indicates a fetch value of 20,000 m and the blue dashed line indicates the upper 95th percentile of fetch values (2.52 million m) for all dives in the database.

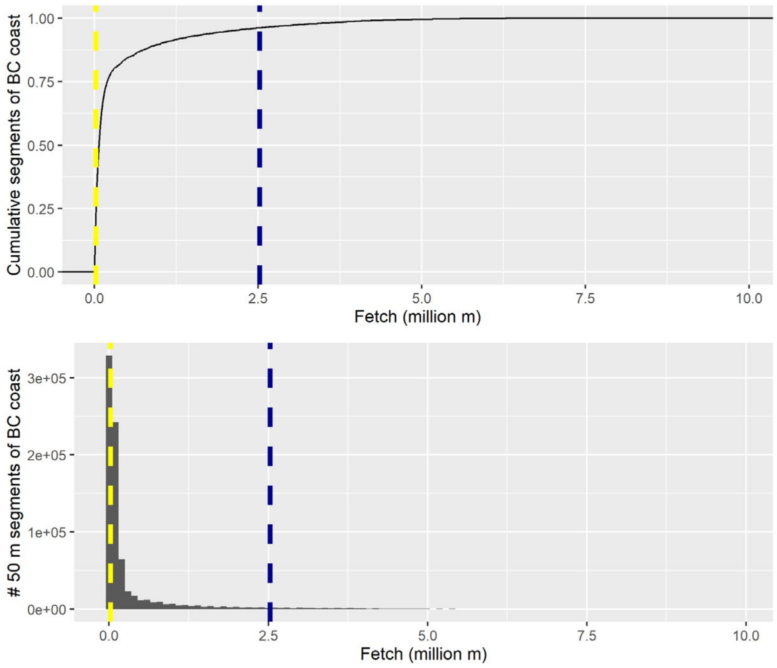

Figure 15. Cumulative number of 50 m shoreline segments along the British Columbia coast in fetch categories from 0 to 10 million m (upper panel) and total number of 50 m shoreline segments along the British Columbia coast in fetch categories from 0 to 10 million m (lower panel). Data were summarized from GIS layers of coastline fetch available on the Open Data Portal (Fetch all BC Geodatabase). Yellow dashed line indicates a fetch value of 20,000 m and the blue dashed line indicates the upper 95th percentile of fetch values (2.52 million m) for all dives in the database.

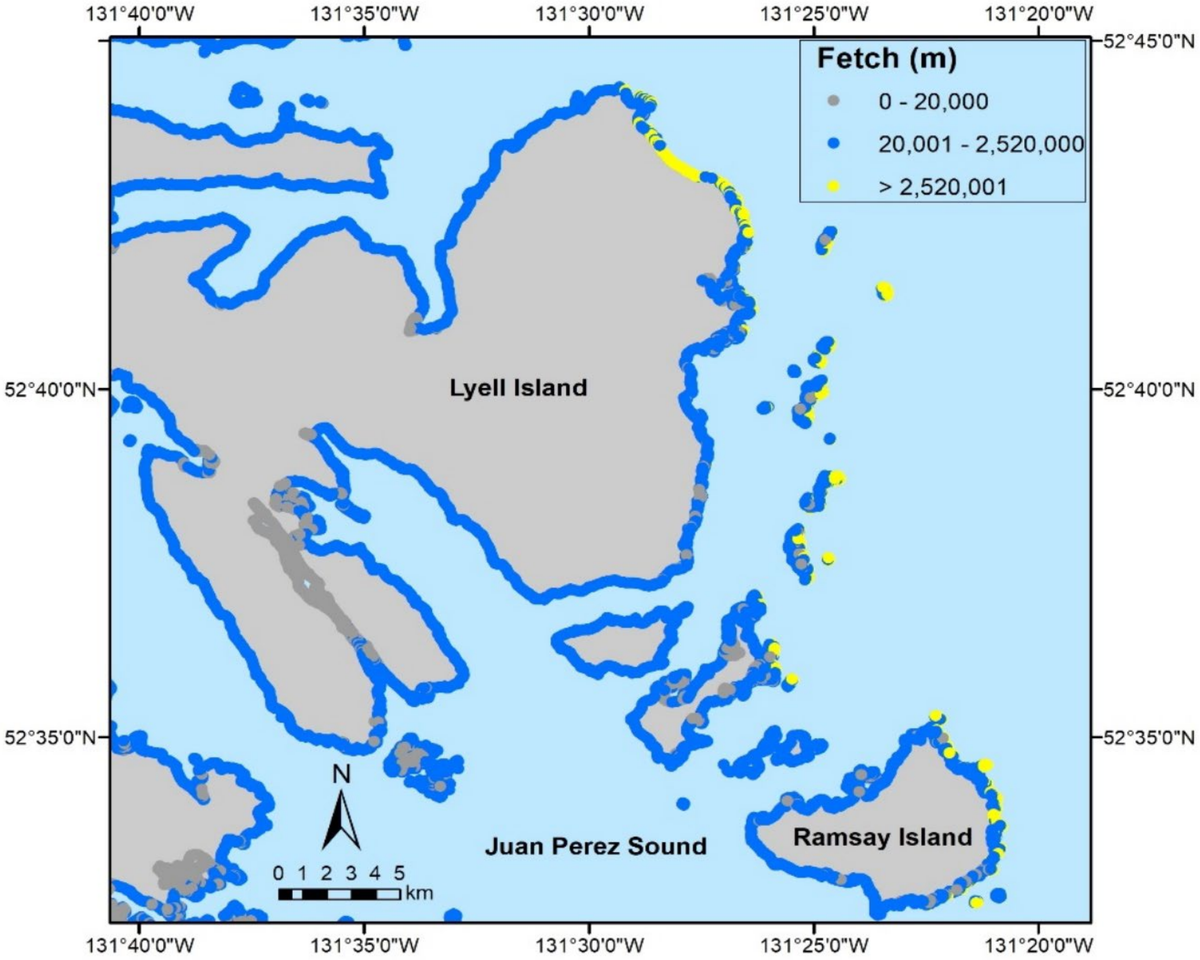

Figure 16. Example stretch of coastline in south east Haida Gwaii showing fetch values.

3.2. STRATIFICATION

3.2.1. Stratification methods

The limiting factor for sampling plans (Section 4.1.1) is the variation among transects in a survey area. This variation arises from:

- local variation in densities among transects due to local factors
- sampling variation in density on a transect because of natural variation along the transect
- sampling variation in density on a transect because of skip sampling the quadrats

In most cases, the last two sources of variation will be small and the first source of variation is typically the largest, e.g., Skibo et al. (2008), Campbell et al. (1998).

One way to account for local variation in density among transects is stratification, where a study area is divided into strata that are more homogeneous. A sample of transects is taken from each stratum; the density is estimated for each stratum; and a weighted average of the stratum-specific density estimates gives the final estimate of density.

If stratification is successful, the variability in transect-specific densities within strata is less than the variability among transect-specific densities over the entire study area and so the uncertainty of the final estimate will be smaller under stratification than without stratification.

Stratification can be done pre- or post-survey. With pre-survey stratification, the stratification must be done prior to surveying transects and the pre-survey stratification variable must be known for the entire coast. With post-stratification, the sample can be divided into strata after the fact, but the stratification variable is still needed for the entire study area so that suitable weights can be derived. The key advantage of pre-sample stratification is the ability to re-allocate effort among strata (e.g., sample more transects in strata that are larger and have higher within-stratum transect-specific variability) to further reduce the uncertainty in the final density estimate.

The following stratification variables of interest were:

- Fetch
- Sea Otter occupation (false/true) and year since occupation
- Dominant substrate type
- Depth

3.2.1.1. Fetch - Methods

Fetch was calculated for points located every 50 m along the entire BC coast and therefore can be considered for pre-sample stratification.

As described in Section 2.2.1, a 26 m buffer was created around each fetch point so that a fetch value was available for every point along the BC coastline. Intercepting survey locations along the shoreline with the buffered fetch layer allowed to assign a fetch value to each transect. These fetch values were then used to explore the relationship between density of the various species of interest and fetch.

3.2.1.2. Impact of fetch as a stratification variable - Results

The distribution of fetch across all transects for all species shows considerable variability, however most transects have fetch values over around 30,000 m (or about $\log_{10}30{,}000 = 4.5$) (Figure 17).

The survey area for all surveys was divided into two strata based on various fetch cutoffs 20,000, 30,000, 50,000 and 100,000 m, (i.e., for each fetch value, the strata are defined as being above or below the particular value), the sd:mean ratio of the transect-specific densities in each stratum was calculated and plotted for all species (Figure 18).

A simple linear regression through the origin of the sd:mean of transect-specific densities for each fetch stratum gives a standard deviation to mean density ratio of 1.80 for the low fetch stratum and 1.25 for the high fetch stratum.

The standard deviation of the transect-specific densities has been reduced in the high fetch stratum compared to the unstratified ratio of 1.27. The sd:mean ratio for the low fetch stratum is larger, but would be applied against a lower overall density for each species.

If we assume that the current surveys were allocated at random relative to the fetch value, the proportion of transects in each fetch stratum will approximate the proportion of all possible transect locations in each fetch stratum (Table 7; note that 2020 Haida Gwaii and 2021 WCVI were excluded from the table because transects were not placed on shoreline with fetch < 20,000 m).

Table 7. Total numbers (#) and percent (%) of transects in high and low fetch strata with various fetch cutoff values.

Fetch cutoff value	**#/% in high stratum**	**#/% in low stratum**
20,000	327/92%	30/8%
30,000	318/89%	39/11%
50,000	301/84%	56/16%
100,000	243/68%	114/32%

So approximately 80-90% of transect locations would be in the *High* fetch stratum unless the Fetch cutoff was set over about 100,000 m and the remainder in the *Low* fetch stratum (Table 8), but this will vary over the different surveys (Table 8).

Table 8. Total number of transects (n) and proportion in low and high fetch strata with various fetch cutoff values by location-year.

Location	**Year**	**Fetch cutoff**	**Total number of transects (n)**	**Proportion in low fetch stratum**	**Proportion in high fetch stratum**
Queen Charlotte Strait	2016	20,000	131	0.11	0.89
Queen Charlotte Strait	2016	30,000	131	0.14	0.86
Queen Charlotte Strait	2016	50,000	131	0.21	0.79
Queen Charlotte Strait	2016	100,000	131	0.43	0.57
South Coast Vancouver Isl.	2017	20,000	58	0.03	0.97
South Coast Vancouver Isl.	2017	30,000	58	0.03	0.97
South Coast Vancouver Isl.	2017	50,000	58	0.07	0.93
South Coast Vancouver Isl.	2017	100,000	58	0.26	0.74
Queen Charlotte Strait	2018	20,000	88	0.08	0.92
Queen Charlotte Strait	2018	30,000	88	0.08	0.92
Queen Charlotte Strait	2018	50,000	88	0.10	0.90
Queen Charlotte Strait	2018	100,000	88	0.24	0.76
Mainland North Coast	2019	20,000	80	0.09	0.91
Mainland North Coast	2019	30,000	80	0.15	0.85
Mainland North Coast	2019	50,000	80	0.19	0.81
Mainland North Coast	2019	100,000	80	0.28	0.72

Again, in many cases, nearly all transects will appear in the high-fetch stratum.

We estimated the density (and standard error) for each species in each year’s transects using an unstratified and assuming a proportional allocation stratified design (using fetch) for each year to estimate the impact of stratification by fetch. Plots of the relative standard errors of density for all species with and without stratification show that there is virtually no benefit from stratification at the various fetch levels under proportional stratification, i.e., what would occur if you post-stratified by fetch after randomizing the transects (Figure 19). At the lower fetch levels, so few of the transects are in the lower fetch stratum that changing the number further (because this stratum has a higher variance on a lower mean) also would not confer any great advantage.

The majority of observations for all multispecies surveys and for all species were above the 20,000 m value ($\log_{10}$ fetch = 4.30 in Figure 17). However, Giant Red Sea Cucumbers and Sunflower Stars were observed at values below 20,000 m more frequently than other species (Figure 17).

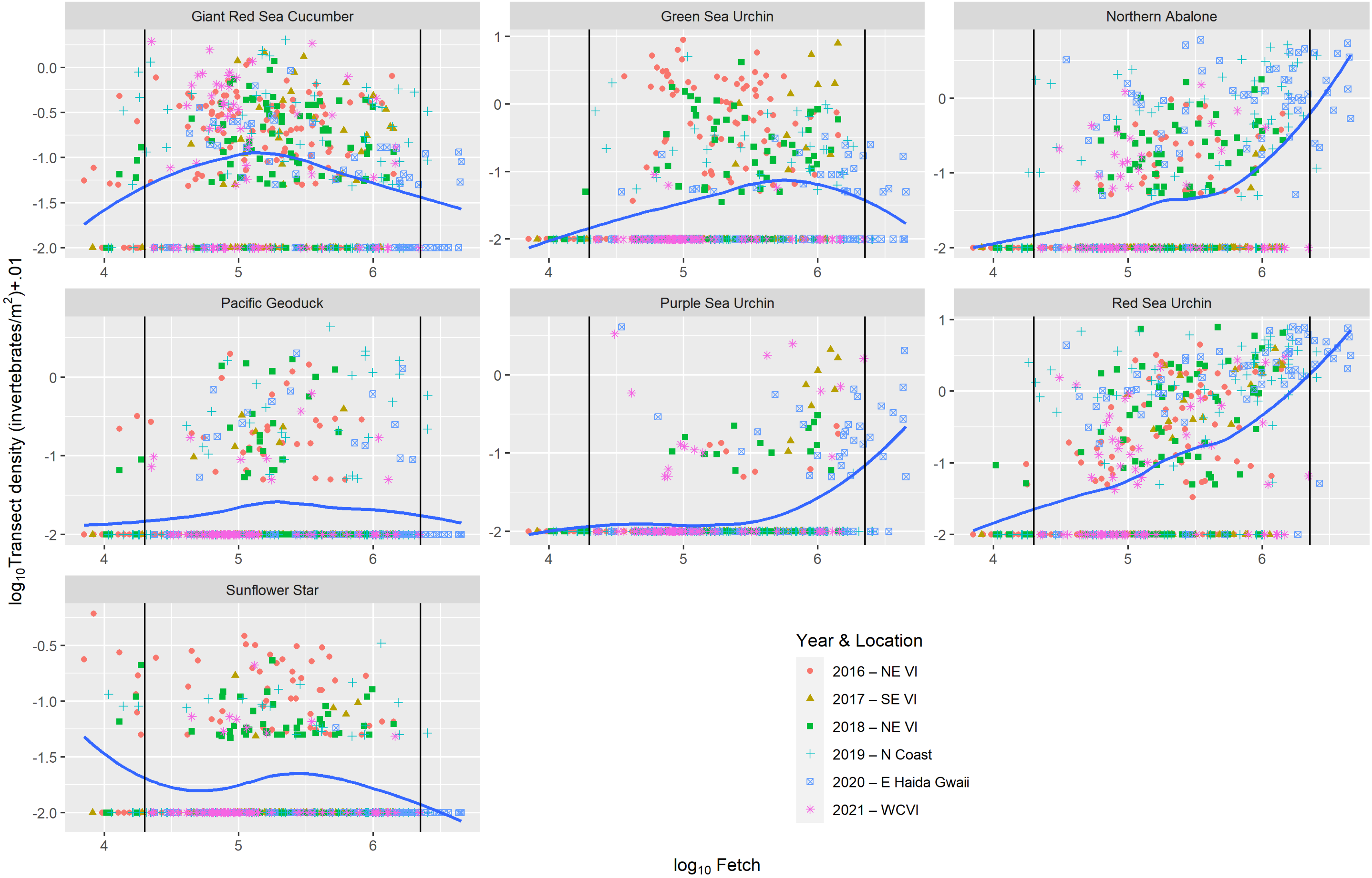

Figure 17. Distribution of density by fetch for all invertebrate species across all transects and years The solid blue line is the (smoothed) average on the log-scale. The solid black lines are the low (20,000 m or 4.30 on $\log_{10}$ scale) and high (2.52 million m or 6.40 on $\log_{10}$ scale).

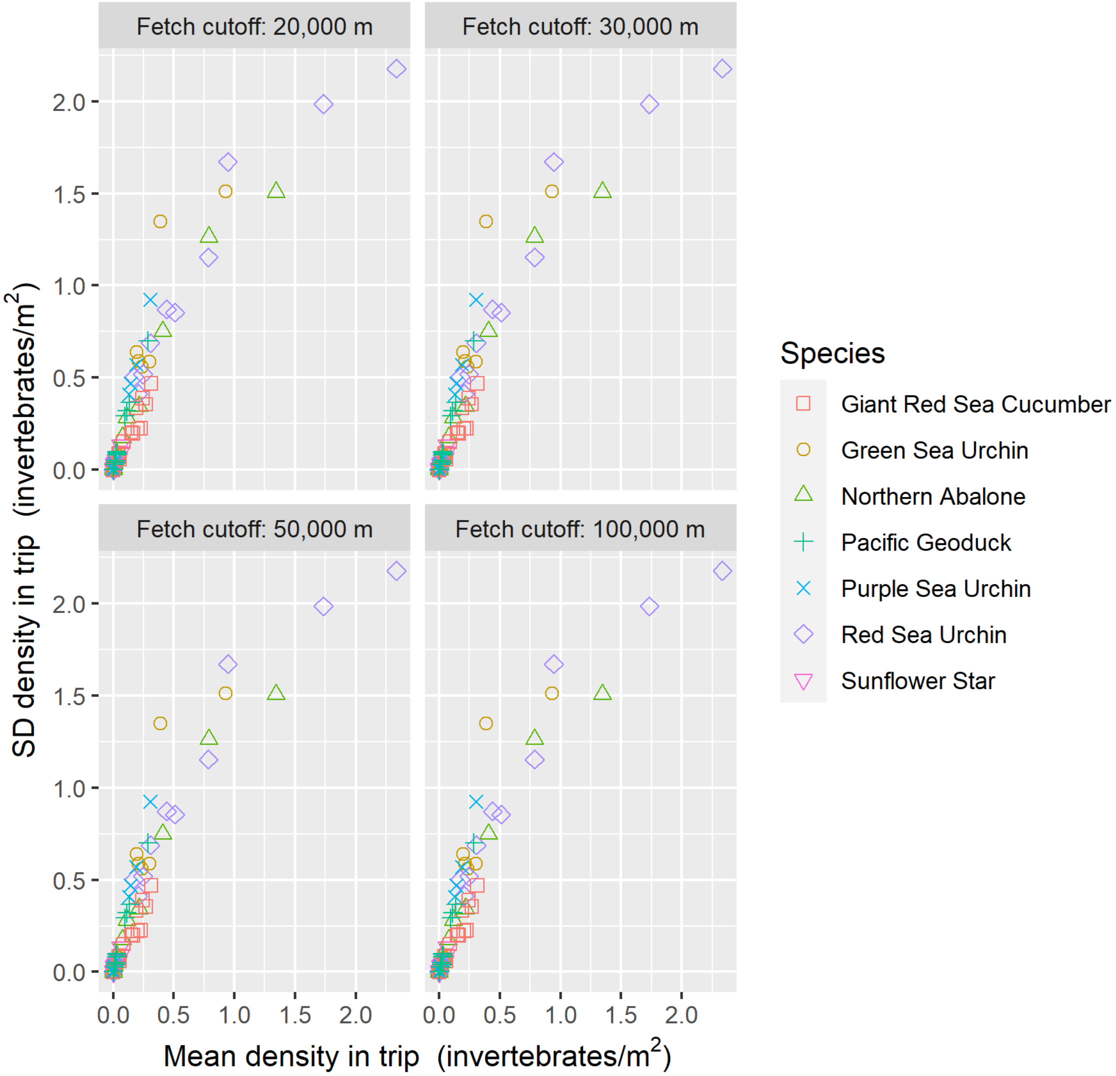

Figure 18. Relationship between the standard deviation of the transect-specific densities to mean density for all invertebrate species and all years with fetch cutoffs of 30,000 m, 40,000 m, 50,000 m and 100,000 m.

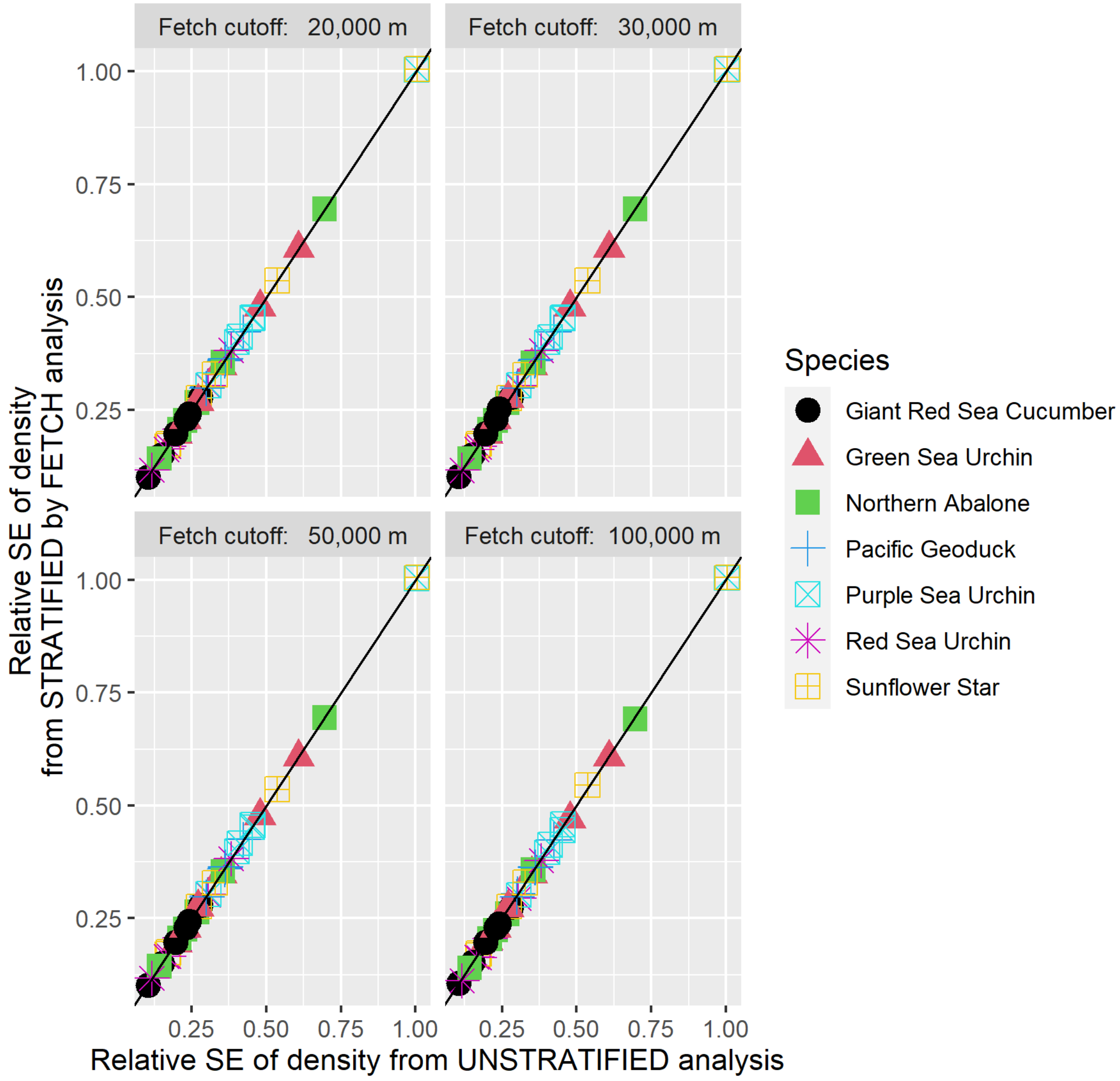

Figure 19. Relative standard errors of density for all invertebrate species with and without stratification with fetch cutoffs of 20,000 m, 30,000 m, 50,000 m and 100,000 m.

3.2.1.3. Sea Otter occupancy - Methods

Sea Otters (*Enhydra lutris*) prey on benthic invertebrates. They are important keystone predators that reduce urchin densities throughout their range (Burt et al. 2018; Lee et al. 2016; Watson and Estes 2011; Kvitek et al. 1989; Pearse and Hines 1987; Estes and Palmisano 1974; Lowry and Pearse 1973). Northern Abalone are also part of Sea Otters' diet, and the recovery of Sea Otters in BC has been shown to cause a 16-fold decline in Northern Abalone populations (Lee et al. 2016). Sea Otters also prey on Giant Red Sea Cucumbers (Laidre and Jameson 2006). In one study in southeast Alaska, sea cucumber density depended on the duration and magnitude of Sea Otter presence, with 100% decline in areas occupied by Sea Otters since 1994 (Larson et al. 2013). Since Sea Otter predation can have a strong influence on invertebrate densities, Sea Otter occupancy was considered as a stratification variable in this study.

Sea Otter occupancy for the entire coast of BC is available and therefore this variable may be suitable for pre-sample stratification. Sea Otter occupancy was derived from DFO's Sea Otter range-wide surveys that have been conducted every five years since 1977 (Nichol et al. 2015). At the time of this study, data up to and including the 2013 Sea Otter survey were available (Nichol et al. 2015). The criterion for occupancy was the presence of at least one raft of Sea Otters at the time of the survey (in spring or summer). Sea Otter occupancy estimates were available for each of the 24 survey segment polygons (Figure 2 in Nichol et al. 2015). Where there wasn't a segment, Sea Otters were not yet considered established. The exception was a small number of the multispecies transects that fell outside of the survey segments that were deemed likely to have been impacted by Sea Otter foraging, possibly in the winter, outside of the survey timeframe by subject matter expertise (Linda Nichol, Marine Mammal Biologist, DFO, Nanaimo, BC, 2022, pers. comm.). Specifically, this refers to 15 transects in Nootka Sound (PFMAs 25-4, 25-6, 25-15) and 10 transects Kyuquot Sound (PFMAs 26-2, 26-4, 26-5, 26-6) that were identified as being occupied by Sea Otters (by subject matter expertise) but do not have an occupancy year associated with them. Full Sea Otter survey details can be found in Nichol et al. (2015).

3.2.1.4. Impact of sea otter occupancy as a stratification variable - Results

The total number of transects in the Sea Otter unoccupied and occupied categories (Table 9), and occupancy year (Table 10) of Sea Otter recolonization were tabulated.

Table 9. Total number of multispecies dive transects in Sea Otter unoccupied and occupied categories by location-year.

Location	Year	Unoccupied (# transects)	Occupied (# transects)
Queen Charlotte Strait	2016	50	81
South Coast Vancouver Island	2017	58	0
Queen Charlotte Strait	2018	0	88
Mainland North Coast	2019	80	0
South East Haida Gwaii	2020	70	0
West Coast Vancouver Island	2021	14	54

Table 10. Total number of multispecies dive transects by year of Sea Otter occupancy by location-year.

Location	**Year**	**1972**	**1989**	**1991**	**1992**	**1996**	**2000**	**2001**	**2008**	**2009**	**Unoccupied**	**Occupied - Unknown Year**
Queen Charlotte Strait	2016	0	0	0	0	0	0	0	0	81	50	0
South Coast Vancouver Island	2017	0	0	0	0	0	0	0	0	0	58	0
Queen Charlotte Strait	2018	0	0	0	0	0	0	0	0	88	0	0
Mainland North Coast	2019	0	0	0	0	0	0	0	0	0	80	0
South East Haida Gwaii	2020	0	0	0	0	0	0	0	0	0	70	0
West Coast Vancouver Island	2021	1	2	1	1	1	12	8	3	0	14	25

Table 11. Total number of multispecies dive transects in Sea Otter unoccupied and occupied categories by year of Sea Otter occupancy.

Year of Sea Otter occupancy	**1972**	**1989**	**1991**	**1992**	**1996**	**2000**	**2001**	**2008**	**2009**	**Totals**
Unoccupied	0	0	0	0	0	0	0	0	0	272 transects unoccupied
Occupied	1	2	1	1	1	12	8	3	169	223 transects occupied (198 with known year and 25 with unknown year)

Essentially the year of occupancy is 2009 for the majority of transects that were occupied, so the binary (unoccupied, occupied) was a better surrogate for the presence of Sea Otters because the occupancy year had too little data.

In most cases, transects in a small survey region were all either occupied or not occupied by Sea Otters and so stratification by Sea Otter occupancy does not matter at local scales. We see that at the survey level, the same is true except for Queen Charlotte Strait in 2016 and WCVI in 2021.

Plots of density versus Sea Otter occupied (false/true) for all years (Figure 20) do not show any obvious differences between the two categories.

A linear regression through the origin of the standard deviation of the transect-specific densities to mean density for each Sea Otter stratum (unoccupied and occupied) gives a ratio of 1.25 for unoccupied and 1.73 for occupied.

The standard deviation of transect-specific densities was lower in the stratum without Sea Otters compared to the stratum with Sea Otters.

If we assume that the current surveys were allocated at random relative to Sea Otter occupancy, the proportion of transects in a stratum will approximate the proportion of all possible transect locations in each Sea Otter occupancy stratum (Table 12).

Table 12. Total numbers (#) and percent (%) of transects in Sea Otter unoccupied and occupied strata.

#/% in unoccupied	#/% in occupied
272/55%	223/45%

Approximately 55% of transect locations would be in the unoccupied stratum, and the remainder in the occupied stratum (Table 12), but this will vary over the different location-years (Table 13).

Table 13. Total number of transects (n) and proportion in Sea Otter unoccupied and occupied strata by location-year.

Location	Year	Total number of transects (n)	Proportion in unoccupied	Proportion in occupied
Queen Charlotte Strait	2016	131	0.38	0.62
South Coast Vancouver Island	2017	58	1.00	0.00
Queen Charlotte Strait	2018	88	0.00	1.00
Mainland North Coast	2019	80	1.00	0.00
South East Haida Gwaii	2020	70	1.00	0.00
West Coast Vancouver Island	2021	68	0.21	0.79

All surveys except Queen Charlotte Strait in 2016 and WCVI in 2021 (Table 13) have all transects either occupied or not occupied, so stratification by Sea Otter occupancy is only relevant for those surveys.

We estimated the density (and standard error) for each species in each of the above surveys using an unstratified and assuming a proportional allocation stratified design (using Sea Otter not occupied/occupied) for each year where there appears that stratification by Sea Otter occupancy would be sensible.

Plots of the relative standard errors of density for all species with and without stratification by Sea Otter occupancy (unoccupied/occupied) shows that there is virtually no benefit from this stratification (Figure 21).

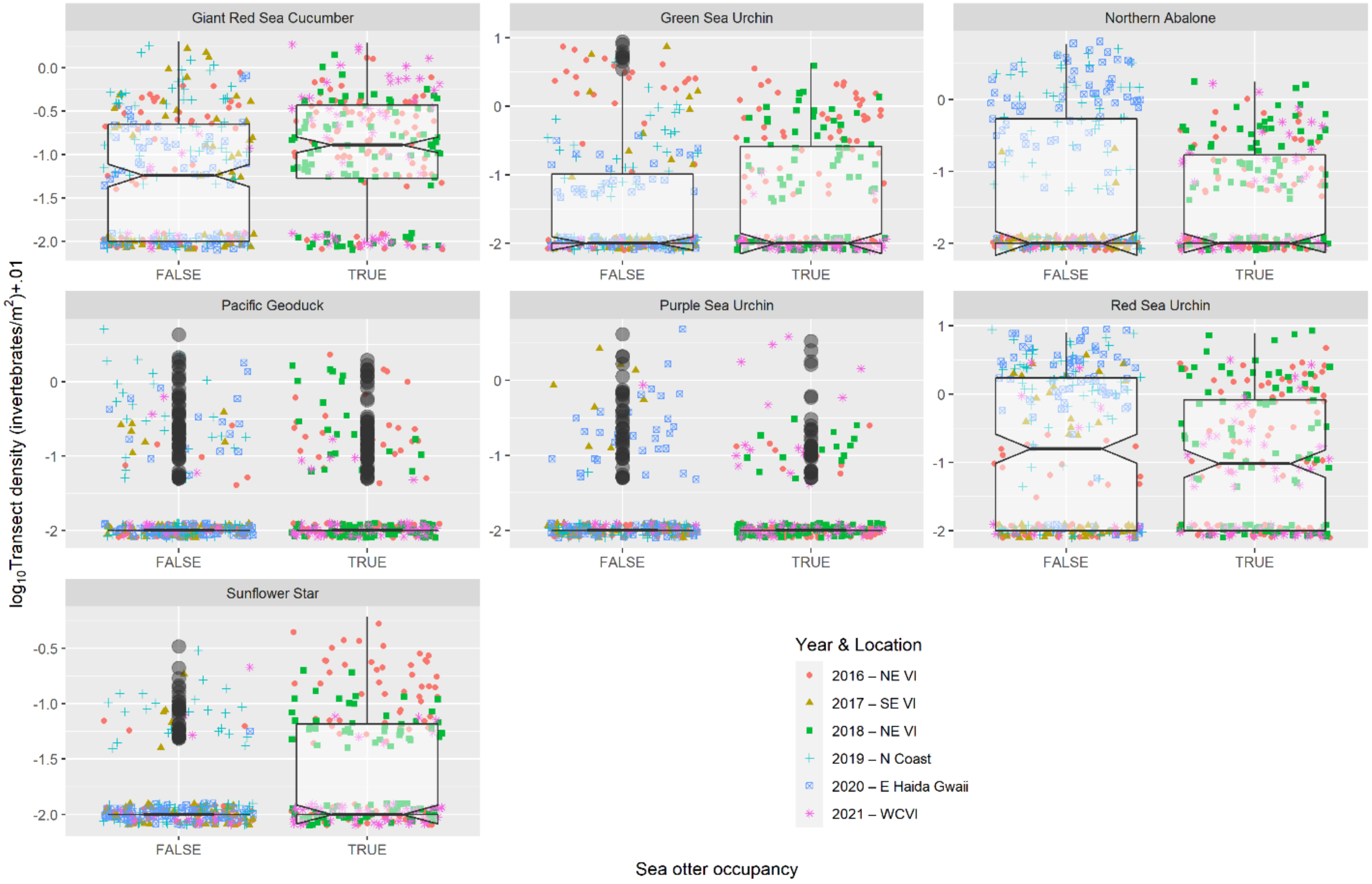

Figure 20. Density versus Sea Otter occupancy (false = unoccupied; true = occupied) for species and all location-years.

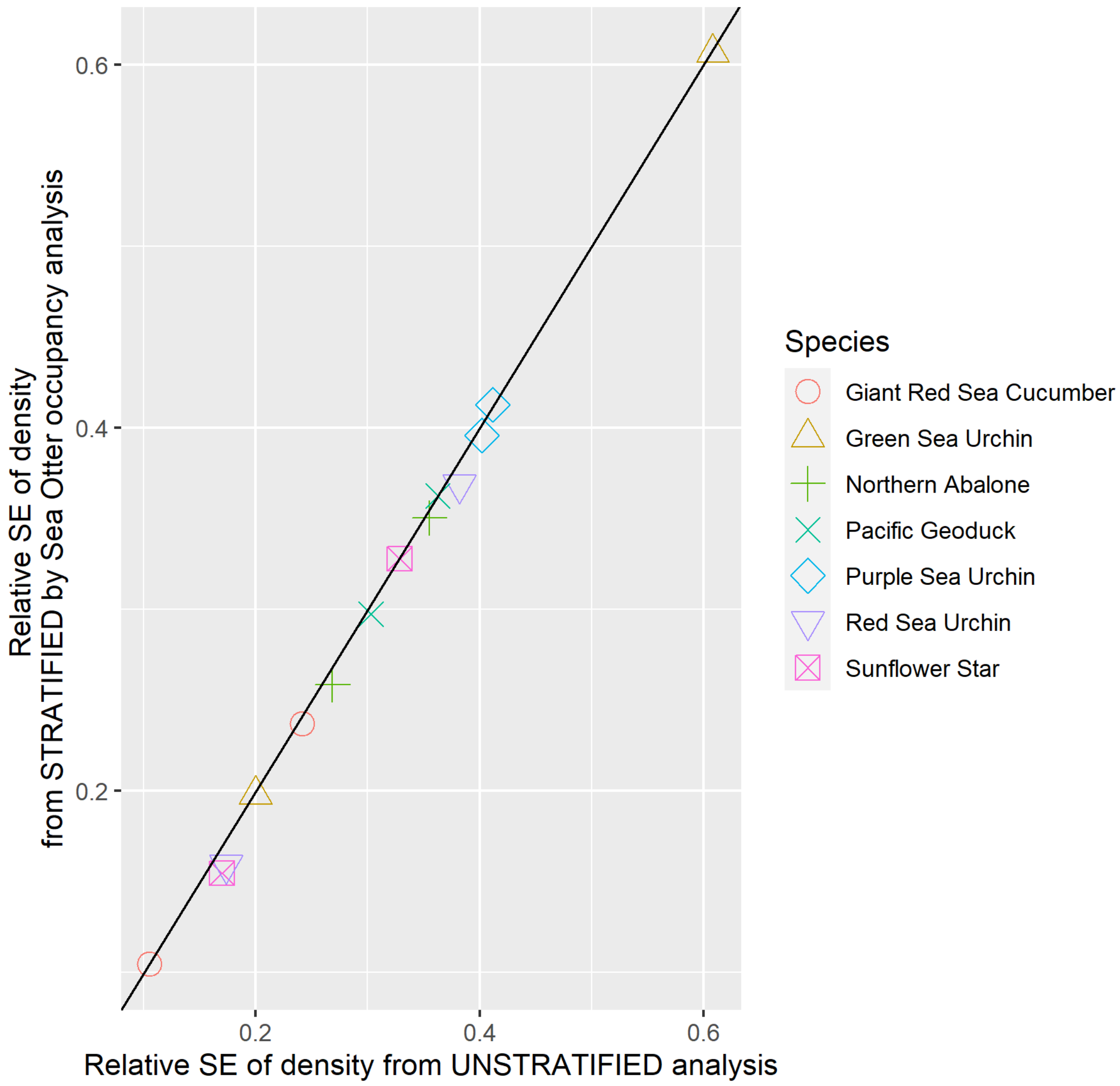

Figure 21. Relative standard errors of density for all species with and without stratification by Sea Otter occupancy (false = unoccupied; true = occupied) for all species.

3.2.1.5. Dominant substrate type - Methods

The first, second and third most dominant substrate types within each quadrat were recorded by divers as one of eleven codes, as described in Appendix A.3: 0 = wood; 1 = smooth bedrock; 2 = bedrock with crevices; 3 = boulders (> 30 cm); 4 = cobble (between 7.5 cm and 30 cm); 5 = gravel (between 2 cm and 7.5 cm); 6 = pea gravel (between 0.25 cm and 2 cm); 7 = sand; 9 = mud; 10 = crushed shell; and 11 = whole shell. The dominant substrate type was investigated as a possible stratification variable.

3.2.1.6. Impact of dominant substrate category as a stratification variable - Results

There is considerable variability across surveys (location-years) in the mean densities reflecting local effects of locations surveyed, but there appears to be some general common trends across trips (Figure 22). There does not appear to be a common substrate suitable for stratification for all species. For example, boulders are generally good for sea urchins but poor for Sunflower Star; vice versa for pea gravel. Additionally, if substrate type was to be used as a pre-stratification variable, its distribution over the survey area would be needed which is currently not available at the necessary resolution.

Substrate preferences for different species were apparent, densities of Northern Abalone, Green Sea Urchins and Purple Sea Urchins were greatest on hard substrates (bedrock to gravel) and near zero for other substrates (Figure 22). In contrast, and as expected, Geoducks were associated with soft substrates.

Here the raw primary substrate data was used to explore stratification. Further stratification options could look at summarizing substrate data from the three dominant substrate types into more general categories, e.g., using the methods of Gregr et al. (2013) or a similar approach.

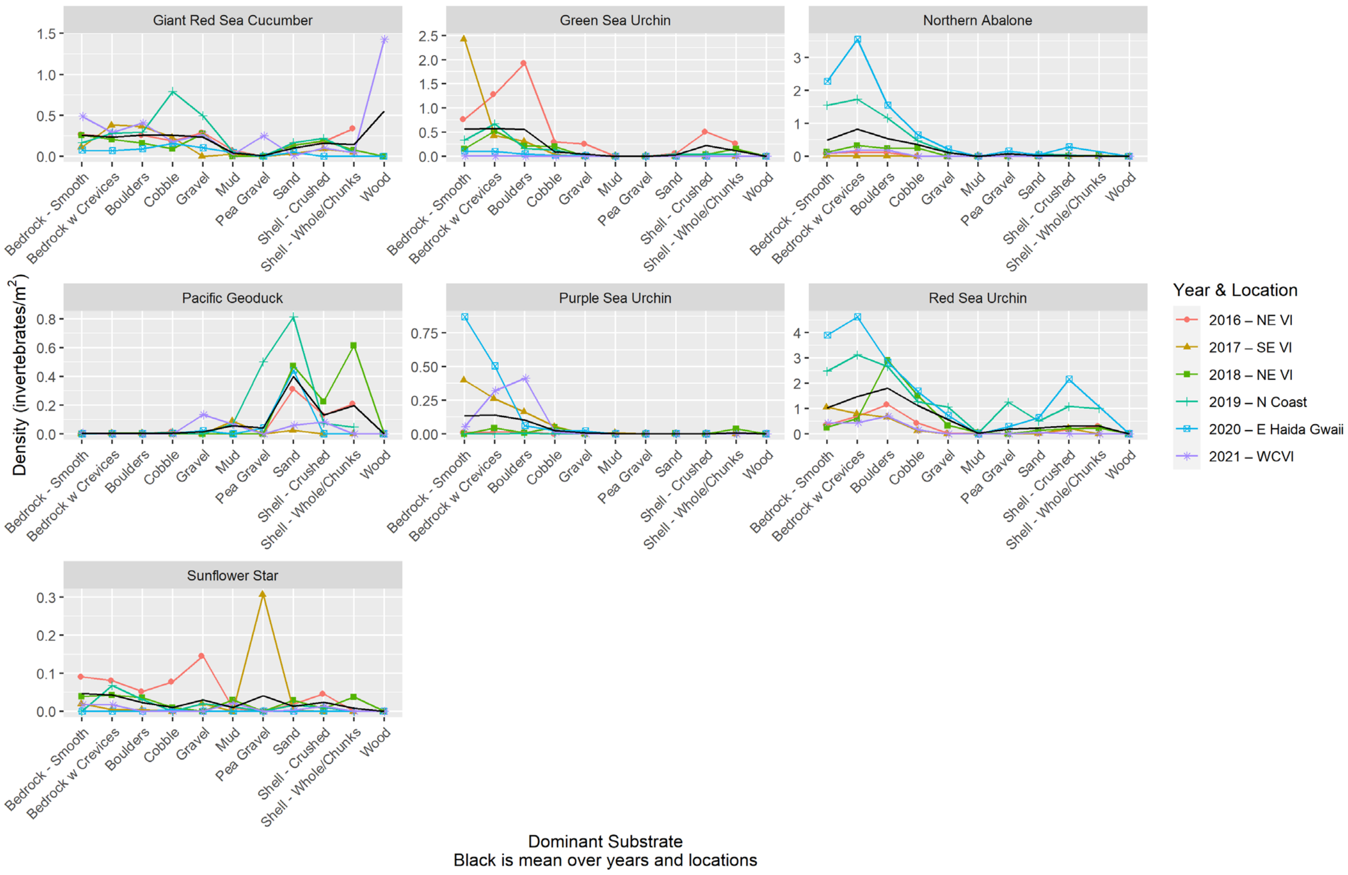

Figure 22. Density by dominant substrate category for all invertebrate species and trips. Black line indicates mean density over all years and locations.

3.2.1.7. Tide corrected depth - Methods

Gauge depth (i.e., the depth recorded from the dive computer during the dive) was corrected to chart datum for each quadrat following the survey by subtracting tide height. Negative values for corrected depths are possible. Water levels (tide height) are most often above 0 m corrected depth (a.k.a. chart datum), and can be up to several metres above 0 m corrected depths. Divers can therefore survey the lower intertidal zone on most dives. These surveys target -2 m corrected depth as the shallow end to ensure all the abalone habitat is covered. The depth values recorded by divers then become negative for quadrats in the intertidal zone once the tide height value is subtracted from the recorded gauge depth. Tide corrected depth was investigated as a possible stratification variable.

3.2.1.8. Impact of tide corrected depth category as a stratification variable - Results

Tide corrected depths were divided into 2 m intervals from -6 m to 18 m, with the last 3 intervals (12-14 m, 14-16 m, 16-18 m) folded into a single interval of 12+ m to avoid small sample sizes in those intervals.

There is considerable variability across trips (years/regions) in the density reflecting local effects of the locations surveyed, but there appears to be some general common trends across trips (Figure 23). Urchins and Sunflower Stars appear to have a higher density in the 0-6 m interval, while sea cucumbers and Geoducks appears to have a higher density as depth increases. It is unclear if stratification will improve precision in estimating density since the transects are conducted perpendicular to shore (i.e., across contour lines) and so automatically include many of the depth strata.

Relationships between species density and depth were apparent. Northern Abalone and Purple Sea Urchin densities were greatest between -2 and 2 m depth and then generally declined (Figure 23). Giant Red Sea Cucumber and Geoduck densities generally increased to a depth of 14 m with no data for greater depths (Figure 23).

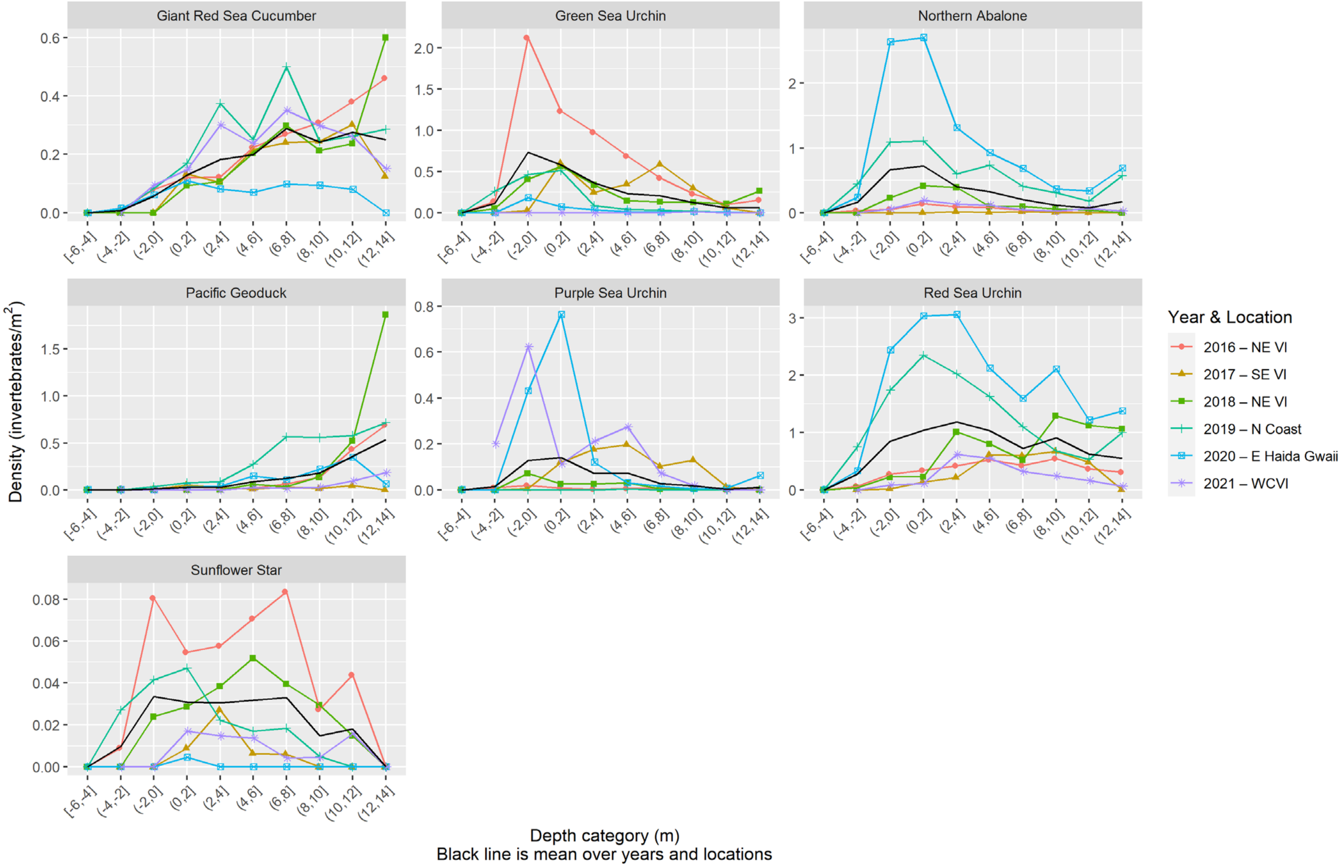

Figure 23. Density by tide corrected depth category for all invertebrate species and trips. The notation for the depth intervals indicates the interval excludes the left value and includes the right value, e.g. (0,2] indicates that depths of 0 are excluded and depths of 2 are included. The last interval (12-14] includes all depths above 12 m. Black line indicates the mean density overall years and locations.

3.3. INDEX SITES VS RE-RANDOMIZATION

An important consideration when designing a time-series survey is whether the same locations or a new set of random locations are surveyed on each visit. Because shallow water benthic habitats are variable over small distances, using index sites and returning to the same sites year after year may minimize site differences that would occur if a new set of random sites was chosen each time, thereby potentially improving the ability to detect changes in population abundance. Generally speaking, a positive correlation in density measurements on replicated transects between two surveys (i.e., between two years) will lead to improved estimates of change (i.e., a smaller standard error) and improved power to detect a change in the density between surveys.

3.3.1. Index sites vs re-randomization methods

Some transects that were surveyed in Queen Charlotte Strait in 2016 were randomly selected to be resurveyed in 2018. In the Deserters Group, 19 transects were surveyed in both years, and in the Gordon Islands, 28 transects were surveyed in both years. Estimation of the potential benefit of using index sites or re-randomization was carried out using the 19 repeated transects from the Deserters Group and the 28 repeated transects from the Gordon Islands. We fit a model to estimate the variance components needed to determine the impact of replicating transects on the two surveys. For simplicity, we will assume that transects are approximately equal length so that the estimate of the density at a site is simply the mean of the transect-specific densities.

The model fit (for each combination of site and species) is

$$D_{ij}=\mu_j+T_i(R)+\epsilon_{ij}$$

where

- D_{ij} is the transect-specific density on transect i in year j
- μ is the density over all transects in year j
- T_i is the (random) effect of transect around the overall density with variance σ_T^2. The (R) indicates a random effect.
- ϵ_{ij} is the transect-year random noise with variance σ^2.

The correlation between the transect counts in the two years if all transects were replicated in the two years is the intra-class correlation. In cases where some transects are not replicated, or the transect variance is estimated as zero, the intra-class correlation may differ slightly from the raw correlational value:

$$\rho=\frac{\sigma_T^2}{\sigma_T^2+\sigma^2}$$

The gain in efficiency if all transects are replicated in each of two years, called the design effect, was calculated as follows:

The standard error (SE) of the density across transects in any survey year is found as

$$SE_{mean} = \frac{\sqrt{\sigma_T^2+\sigma^2}}{\sqrt{n}}$$

where

- n is the number of transects run in that survey year.

There is no benefit in replicating transects if interest lies only in estimating the year-specific density.

The SE of change in density if new transects are run in each of two years is

$$SE_{diff\ mean,no\ rep} = \frac{\sqrt{2 \times (\sigma_T^2+\sigma^2)}}{\sqrt{n}}$$

where

- n is the number of transects run in each of the two years,

The SE of the change in density if all transects are replicated in each of two years is:

$$SE_{diff\ mean,rep} = \frac{\sqrt{2 \times (\sigma^2)}}{\sqrt{n}}$$

where

- n is the number of transects run in each of the two years that are replicated.

The gain in efficiency, or the design effect, is computed as the square of the ratio of these two standard errors

$$DE = \frac{\sigma_T^2}{\sigma_T^2+\sigma^2}$$

The design effect is relative to the intraclass correlation:

$$DE = \frac{1}{1-\rho}$$

A design effect of 1 indicates no improvement from replicating transects across surveys. A design effect > 1 indicates it is more efficient (i.e., less transects required to detect a certain size effect) by this factor i.e., *DE = 2* implies that the sample size would be halved when using replicated transects compared to using new transects each year to detect the same size difference in density.

3.3.2. Index sites vs re-randomization results

Estimated transect-specific densities from the first visit to the transect locations (2016) were plotted against the estimated transect-specific densities from the second visit (2018) for each invertebrate species (Figure 24). Correlations were modest for most species, often caused by a small number of transects where the transect-specific densities appear to be quite different between the two years. In some cases, a single transect appears to be quite “different” in the relationship between the two years of the survey. This may indicate that the replicated transect did not quite match the location of the transect in the previous visit.

The raw correlation, the intra-class correlation, and the design effect were calculated for each invertebrate species for the resurveyed transects in Deserters Group and Gordon Islands (Table 8). The design effect for most species was between 1 and 1.9, but was 2.5 for Green Sea Urchins in the Gordon Islands, and 4.6 for Giant Red Sea Cucumbers in Deserters Group. This suggests that resurveying the same locations (index sites) may be beneficial over short time intervals for some species.

The impact of replicating transects on longer term trends is more difficult to determine because it seems reasonable to assume that the correlation of density among replicated transects would decline as the period of time between visits increases. For example, there may be a very high correlation in the density when transects are measured 1 month apart, which could decline to a moderate correlation when the same transects are measured 2 years apart, which could then decline to 0 when the same transects are measured 5 years apart.

Unfortunately, we have insufficient information to know how fast the correlations decline. We have some information for the Giant Red Sea Cucumber for repeated transects in control sites from the Experimental Fishery Areas (Hajas et al. 2023; Hand et al. 2009), however we do not have this type of information for all the invertebrate species of interest, which makes it difficult to optimize the survey design using temporal correction of density for multiple species.

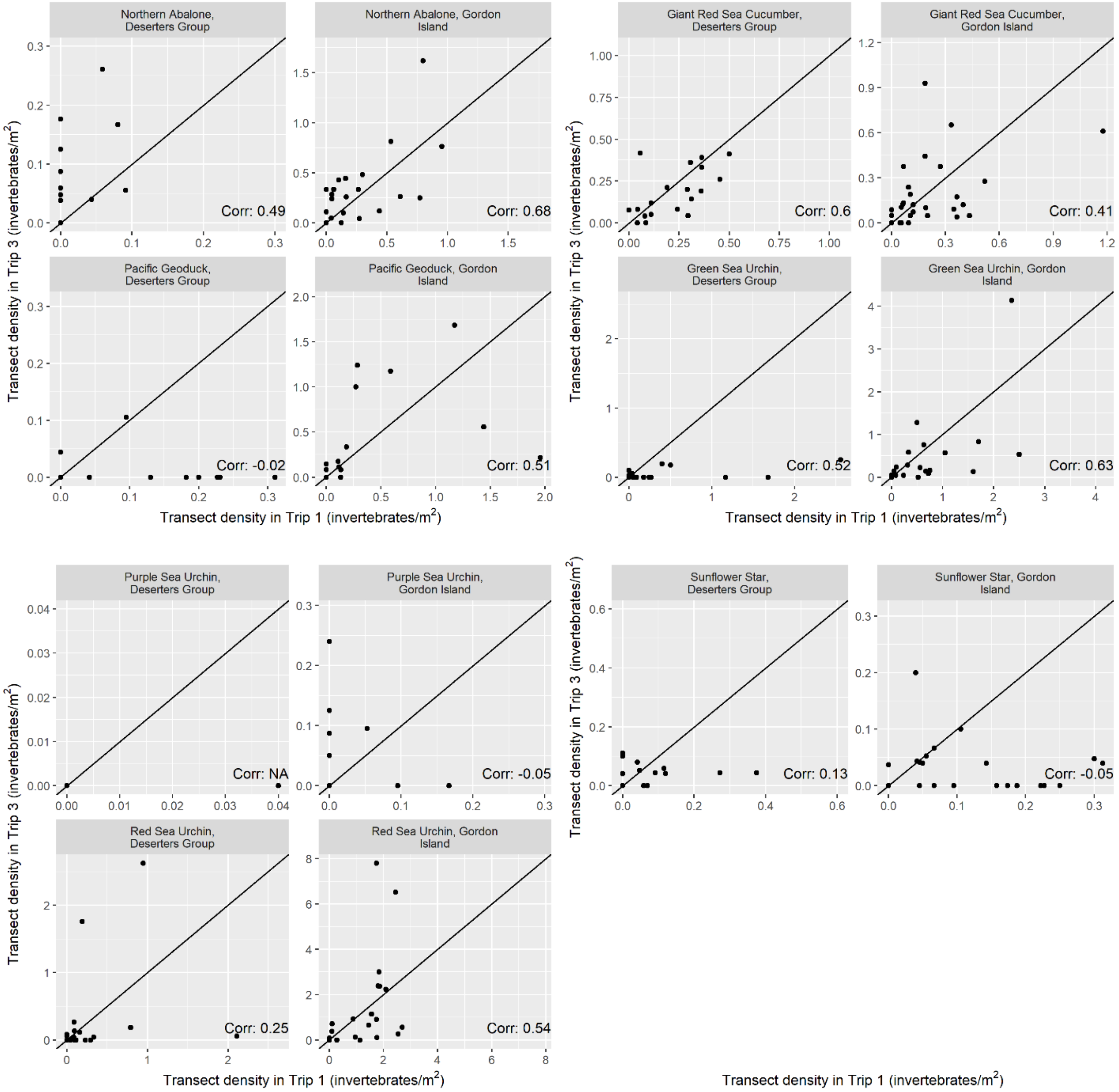

Figure 24. Estimated density of invertebrates on transects that were surveyed on Trip 1 (Queen Charlotte Strait, 2016, Deserters Group and Gordon Islands) and then resurveyed on Trip 3 (Queen Charlotte Strait, 2018, Deserters Group and Gordon Islands) and the raw correlation coefficients. Black diagonal lines show 1 to 1 relationship. Trip 1 and Trip 3 are unique identifiers from the database.

Table 14. Raw corrections, transect standard deviations (SD), residual standard deviations (SD), intra-class correlations and design effect for each invertebrate species for the transects that were surveyed in both 2016 and 2018 in the Deserters Group and Gordon Islands in Queen Charlotte Strait (Note that Purple Sea Urchin in Deserters Group was only observed on one transect in 2016).*

Location	Species	Raw correlation	Transect SD	Residual SD	Intra-class correlation	Design effect
Deserters Group	Northern Abalone	0.49	0.026	0.054	0.18	1.2
Deserters Group	Sunflower Star	0.13	0.059	0.087	0.32	1.5
Deserters Group	Green Sea Urchin	0.52	0.046	0.397	0.01	1.0
Deserters Group	Red Sea Urchin	0.25	0.211	0.429	0.19	1.2
Deserters Group	Purple Sea Urchin	*	0	0.005	0.00	1.0
Deserters Group	Giant Red Sea Cucumber	0.60	0.189	0.100	0.78	4.6
Deserters Group	Pacific Geoduck	-0.02	0	0.072	0.00	1.0
Gordon Islands	Northern Abalone	0.68	0.280	0.185	0.70	3.3
Gordon Islands	Sunflower Star	-0.05	0	0.082	0.00	1.0
Gordon Islands	Green Sea Urchin	0.63	0.564	0.466	0.59	2.5
Gordon Islands	Red Sea Urchin	0.54	1.063	1.148	0.46	1.9
Gordon Islands	Purple Sea Urchin	-0.05	0	0.061	0.00	1.0
Gordon Islands	Giant Red Sea Cucumber	0.41	0.161	0.179	0.45	1.8
Gordon Islands	Pacific Geoduck	0.51	0.283	0.308	0.46	1.8

3.4. DISCUSSION

The pilot surveys provided a rich dataset with which to explore potential protocol improvements such as fetch cutoff values (single-species survey data were also examined for this), stratification, and overall survey design (i.e., index sites versus re-randomization of transects). Results indicate that future surveys should exclude sections of shoreline with fetch values lower than 20,000 m or higher than 2.52 million m when randomly placing transects to ensure sampling effort is focused on suitable habitat while also avoiding potentially unsafe areas for diving.

There was insufficient evidence from the present analyses to suggest that stratification by fetch, Sea Otter occupancy, substrate type, or depth improved survey precision at this time, however the dynamic ecosystems occupied by the invertebrate species of interest implies that these or other variables could become relevant to the survey design in the future. It is therefore recommended to continue to explore pre- or post-stratification variables to improve survey precision, as data become available.

The impact of replicating transects (i.e., index sites) was minimal, as only moderate positive correlations between repeated density measurements were observed with no substantial gains in precision when compared with randomly selecting new transect positions for each survey. Furthermore, it is often difficult to return to the exact transect location on repeat visits. On the other hand, re-randomizing the selection of transect locations prior to each survey could: allow the collection of data from more locations over time that may contribute to improving stratification, enable the use of different models to estimate density (e.g., spatio-temporal models), lead to better knowledge about coast wide spatial and temporal variability in the populations, and readily incorporate additional environmental and/or ecosystem information over time. It is therefore recommended to re-randomize transect locations for future surveys.

4. SAMPLE SIZE AND SURVEY DESIGN

4.1. OPTIMIZING SAMPLE SIZE

4.1.1. Acceptance sampling methods

Acceptance sampling is a common statistical method used in manufacturing contexts (Lawson 2021). It is also relevant when evaluating stocks against reference criteria (i.e., LRPs and USRs) in fisheries science because it uses statistical techniques for early detection and prevention of problems, rather than correction of problems that have already occurred.

Tradition hypothesis testing is unwieldly in this context. For example, the Fishery Manager may wish to protect the stock from overfishing. Consequently, the null hypothesis would be set that the stock density is below the LRP and no fishing should occur unless the data provide evidence that the stock density is above the LRP (if this null hypothesis was rejected or if the lower bound of the confidence interval for the density exceeded the LRP). However, producers may wish to fish unless the stock density is below the USR. In this case, the null hypothesis would be set that the stock density is above the USR and fishing should occur unless the data provide evidence that the stock density is below the USR. The usual concepts of Type I and Type II errors will now depend on the viewpoint when the hypothesis was set up.

Consequently, acceptance sampling works with two risks, called the consumer and producer risks. These names come from the role of acceptance sampling in manufacturing contexts. In the context of this paper, the consumer risk is the probability of accepting an area as meeting the reference criteria (i.e., of density being above the LRP) when in fact it is not (i.e., a false positive). Conversely, the producer risk is the probability of an area failing the reference criteria (i.e., of density being below the USR) when in fact it is acceptable (above the USR) (i.e., a false negative).

Serious harm to the stock may occur and measures (e.g., closing a fishery) may be triggered when a stock falls below the LRP. The sampling plan should therefore have a high power to detect a situation where the density of the stock falls below the LRP. Fisheries Managers were consulted to set the risk of not detecting if the density is below the LRP; i.e. the consumer risk. The risk of not detecting a density below the LRP was set at 0.05.

The USR is the value above which the stock is deemed ‘healthy’ and no action is needed to be taken. The sampling plan should have a low probability of falsely determining that the density is below the USR when in fact, the density is above the USR; i.e., the producer risk. Fisheries Managers set the risk of falsely identifying that a stock density is below the USR when the actual density is at the USR or higher also at 0.05.

As mentioned in the Introduction, LRPs and candidate USRs have been developed for Giant Red Sea Cucumber, Geoduck, Green Sea Urchin (GSU) and Red Sea Urchin (RSU) (Hajas et al. 2023; Lochead et al. 2019; DFO 2018) (Table 1). The reference points for Giant Red Sea Cucumber and RSU are intended to be applied coast wide, and accordingly, sampling plans were developed for these two species. It is important to note that the Green Sea Urchin reference points were derived from fishery-independent survey data at two high density index sites, one off north east Vancouver Island and one off south east Vancouver Island (DFO 2018), and these reference points may not be applicable coast wide (currently, GSU are only assessed in north east Vancouver Island and south east Vancouver Island, where they are fished (DFO 2021b; DFO 2018; DFO 2016a; DFO 2014)). Green Sea Urchins were still included in the acceptance sampling analyses and a sampling plan was developed (Section 4.1), in case coast wide assessments become desirable in the future. Also, due to the timing of the multispecies survey (generally in September) the survey will not be used to assess Geoduck stock status (more in Section 5: Uncertainties and Section 6: General Discussion). Therefore, a sampling plan was not generated for Geoduck.

The set of LRP, consumer risk, USR, and producer risk enable the development of a "single-sample variables acceptance sampling plan" (Lawson 2021). The term "single-sample" refers to design of the sample where a single sample is taken, the density is estimated, and a decision is made to accept or reject. The term "variables acceptance" implies that numerical values (e.g., density) are used to make a decision.

These plans have a value *k* and a sample size *n*. In this plan, the density is determined using a sample of *n* transects.

- If the estimated density is < *k*, then the stock density is considered to be unhealthy and measures must be taken.
- If the estimated density is > *k*, then the stock density is considered to be healthy, and no further measures are taken.

4.1.1.1. The single sampling plan

A single sampling plan was developed for Green Sea Urchin, Red Sea Urchin and Giant Red Sea Cucumber by solving for *n* and *k* such that

$$P(\widehat{Density} < k | \text{Density} \leq \text{LRP}) = 1 - \text{Consumer risk} = 0.95$$

$$P(\widehat{Density} < k | \text{Density} \geq \text{USR}) = \text{Producer risk} = 0.05$$

Sample sizes (number of transects) were assumed to be large enough that a normal approximation to the sampling distribution of $\widehat{Density}$ was applicable with the standard error of the estimates being a function of the standard deviation (sd, or 'σ') which in turn is a function of the mean (μ), i.e., $\sigma = 1.27 \times \mu$. This latter ratio (sd:mean of 1.27) was derived from estimating the standard deviation of transect-specific densities to the overall density across species (Figure 12).

Analytical formula are not available to determine the appropriate sample size, but the equations used to derive the plans are presented in Appendix D: Supplementary Material.

Sensitivity of the single sampling plans to the sd:mean ratio was inspected by constructing single sampling plans for Green Sea Urchin, Red Sea Urchin and Giant Red Sea Cucumber when the sd:mean ratio was increased by 10%, 25% and 50%.

4.1.1.2. Double sampling plans

Double sampling plans were also explored. In double-sampling plans a smaller initial sample is taken and it is determined if a decision can be made based on this smaller sample, or whether additional sampling is required. These types of plans can be useful when the stock is well above the USR or well below the LRP. It is not considered feasible at this time, and is presented in Appendix C: Double Sampling Plans.

4.1.2. Acceptance sampling results

Using the same standard deviation of density to mean density ratio for all species (1.27) and the same producer and consumer risks for all species, the *k* values (cutoff) and the number of transects (*n*) that need to be surveyed in single sampling plans are shown below in Table 9.

Table 15. Summary of single sampling plans.

Species	k	# of transects (n)
Giant Red Sea Cucumber	0.033	244
Green Sea Urchin	0.600	40
Red Sea Urchin	0.400	40

Because the USR is double the LRP for the two urchin species, the sample size (number of transects) is the same for both species with the values of '*k'* about halfway between their respective *LRP* and *USR*. Because the USR/LRP ratio is lower for the Giant Red Sea Cucumber, the number of transects that need to be surveyed in single sampling plans (n) is much larger than for the urchin species.

Operating Characteristic (OC) curves were developed to show the probability of declaring that the density is acceptable or unacceptable at various levels of actual density as a way to measure the performance of the sampling plans for Giant Red Sea Cucumber, Green Sea Urchin and Red Sea Urchin (Figure 25).

The typical width of a confidence interval for the given sample sizes are shown in Figure 26. As the sample size is reduced, this is reflected in increased risk of not detecting if the density is below the LRP (reduced power), and increased risk of falsely declaring that the density is low when the actual density is higher than the USR (increased false positive rate).

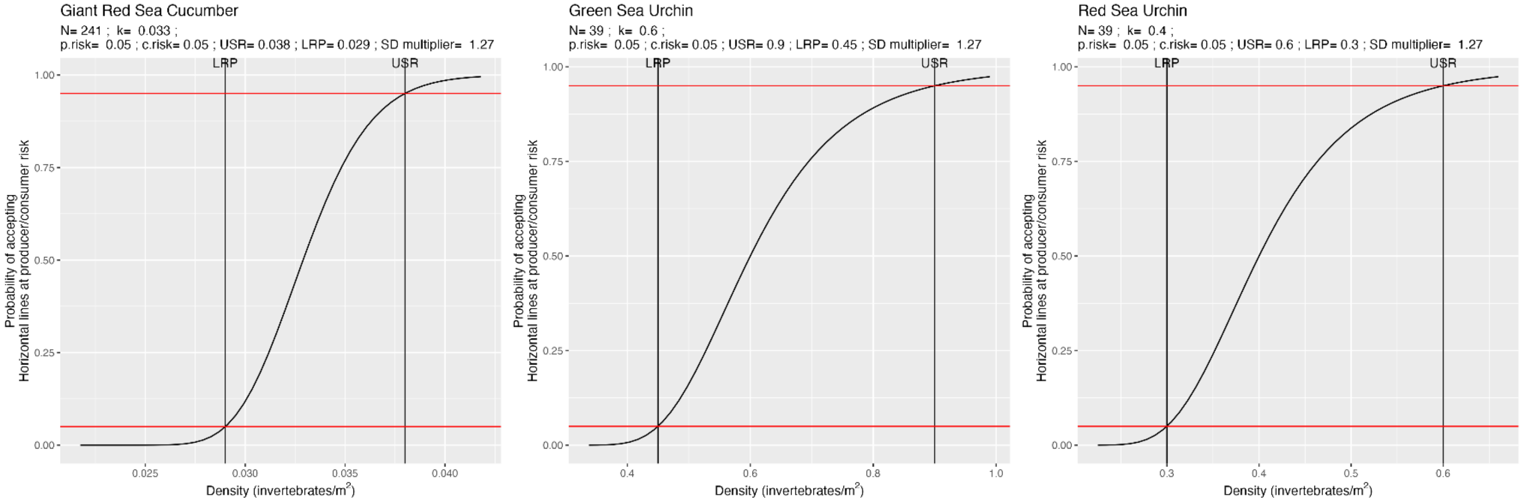

Figure 25. Operating Characteristic (OC) curve for Giant Red Sea Cucumber, Green Sea Urchin and Red Sea Urchin, showing the probability of declaring that the density is below the critical value at various levels of actual density.

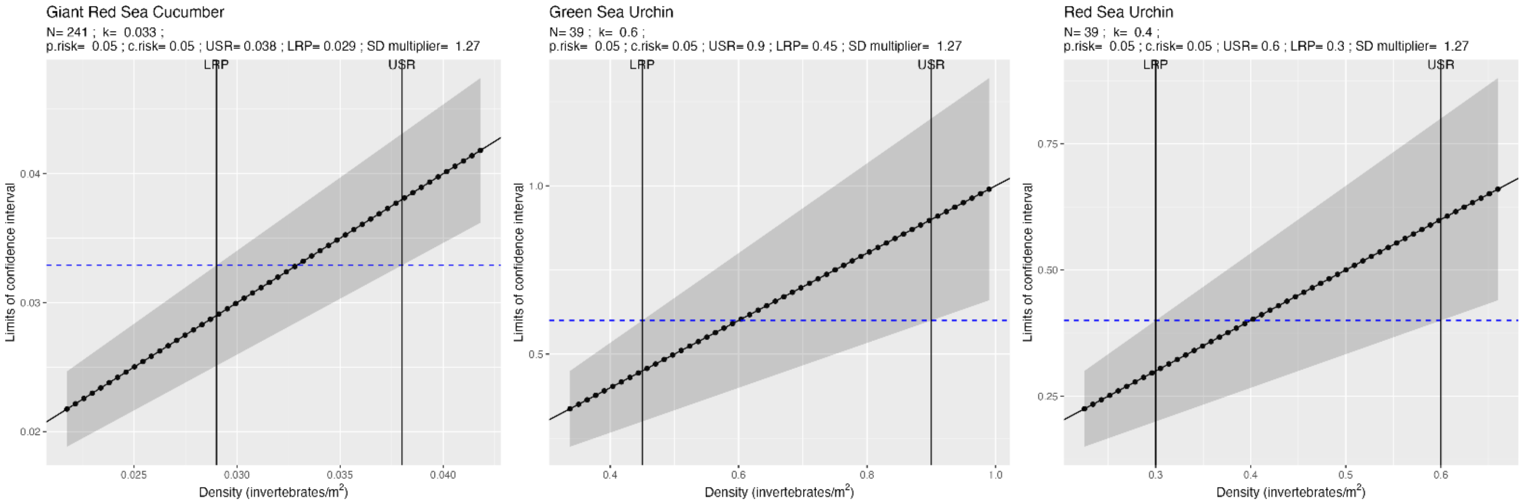

Figure 26. Plot of a typical 90% confidence interval for Giant Red Sea Cucumber, Green Sea Urchin and Red Sea Urchin density (invertebrates per m^2). Blue dashed lines represent the value of k.

4.1.3. Sensitivity to sd:mean multiplier

Table 16. The single sampling plans for Green Sea Urchin, Red Sea Urchin and Giant Red Sea Cucumber with sd:mean ratios of density increased by 10%, 25% and 50% from 1.27.

Species	**% increase**	**sd:mean ratio**	**K (animals per m^2)**	**n (number of transects)**
Green Sea Urchin	0	1.27	0.60	40
	10	1.39	0.60	48
	25	1.58	0.60	62
	50	1.90	0.60	89
Red Sea Urchin	0	1.27	0.40	40
	10	1.39	0.40	48
	25	1.58	0.40	62
	50	1.90	0.40	89
Giant Red Sea Cucumber	0	1.27	0.03	244
	10	1.39	0.03	295
	25	1.58	0.03	380
	50	1.90	0.03	548

As the standard deviation of the transect-specific densities (through the sd:mean ratio) increases, the required sample size increases but the value of *k* is unchanged.

4.2. OPTIMIZING TRANSECT PLACEMENT

4.2.1. Random sampling designs

The objective of the multispecies invertebrate dive survey is to produce unbiased estimates of the population abundance for the entire coast of British Columbia for the selected group of invertebrates. This survey would use the diver survey protocols described in Appendix A. Based on the analysis conducted in the previous sections, a sample size of ~240 transects would be suitable for measuring the coastwide population with sufficient precision to balance the consumer and producer risks required by the population reference points. Therefore, the ideal dive survey would be an annual survey that covered the entire sampling frame (the BC coastline with fetch values from 20,000 - 2.52 million m; Figure 27) with 240 randomly selected transects.

This survey would produce an unbiased (relative to the sampling frame) annual index of abundance for each of the seven invertebrates with reasonable precision.

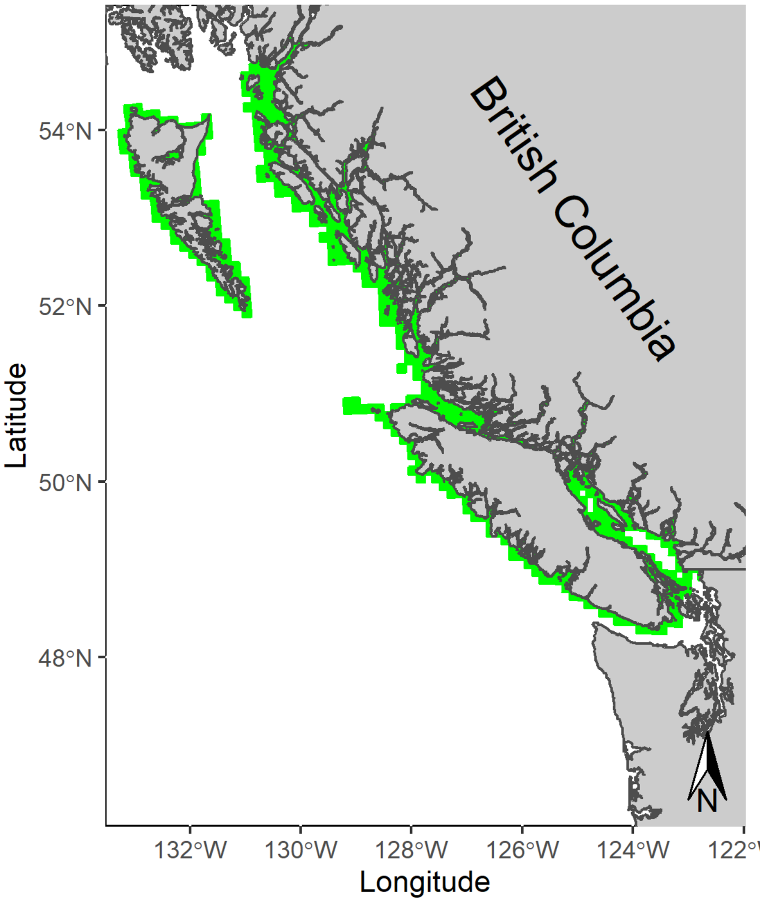

Figure 27. Coastline of British Columbia. Areas shown in green are regions of coastline that have fetch values between 20,000 m and 2.52 million m.

However, logistical considerations limit the available time and effort that can be assigned to the multispecies invertebrate dive survey. Currently the logistical limitations are determined by available vessel time and availability of qualified divers. Only about 14-21 days of vessel time and two teams of divers have been available in recent years in September. In practice, this limits the number of transects that can be completed to about 80 to 120 per year. The examples given below were developed to accommodate 80 transects per year, but the same concepts would hold for any other total number of transects deemed achievable in a given year.

4.3. POTENTIAL SURVEY DESIGNS AND RESULTS

We developed example scenarios for implementation of the dive survey using three different survey designs for selection of transect locations; simple random sampling, two-stage sampling and a panel design with two-stage sampling. Note that there is an additional sampling stage, consisting of systematically sampling quadrats along transects, regardless of the method used to select transect locations. In these examples we imposed a 1 km by 1 km grid over the entire BC coastline for simplicity. In these examples, the 1 km by 1 km grid cells were the sampling units where individual diver transects would be placed. In the pilot surveys, every metre of shoreline within the survey mask was eligible for transect placement. The density and sampling design are the same regardless of whether a 1 km by 1 km grid or every metre of shoreline is used as the fundamental unit for transect placement.

A simple random sample of 1 km by 1 km grid cells (n = 80) in a year resulted in a wide spread of individual transects across the BC coast (Figure 28).

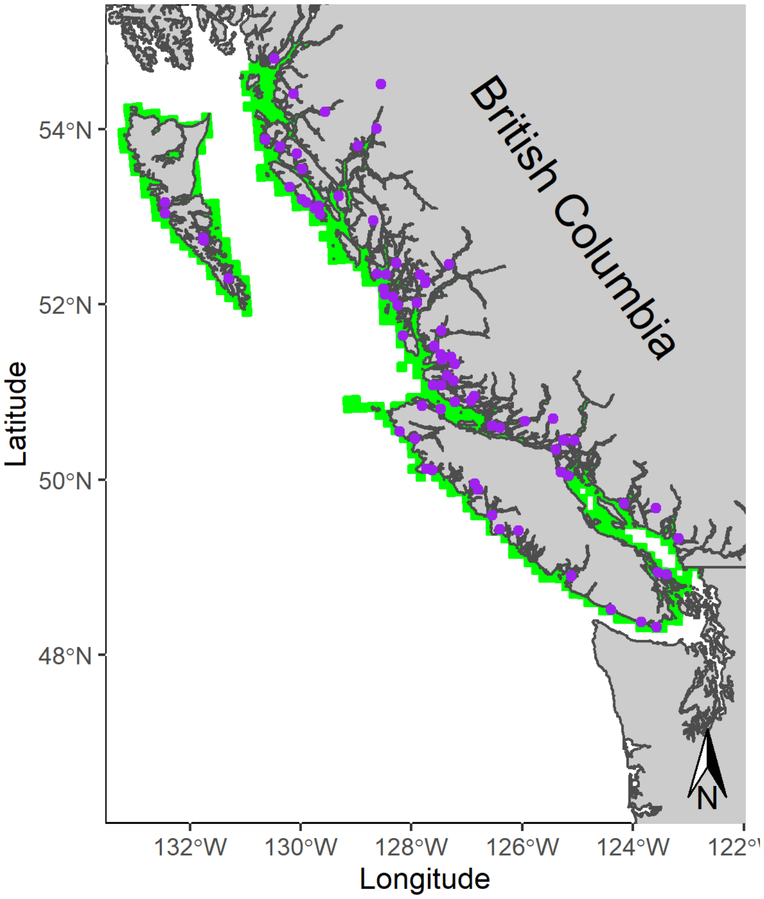

Figure 28. Example selection of 80 random locations, shown as purple dots, using simple random sampling to cover the entire coast. In this case dive survey transects were chosen from individual 1 km by 1 km sections of shoreline.

There are two common estimators to estimate the density: simple mean of transect-specific densities giving each transect equal weight or, more commonly a ratio estimator where transects are weighted based on transect length. The ratio estimator is used to estimate density for the DFO single-species surveys (Hajas et al. 2023; Lochead et al. 2015; Bureau et al. 2012; Hand et al. 2009). If all transects have equal length, the two estimators are identical.

Let

n be the number of survey transects,

L_i be the length of the *i*th transect; and

y_i be the density of the invertebrates at transect *i*, and

Then the estimated overall density and standard error computed as a ratio estimator (Thompson et al. 1992) is:

$$\widehat{D}_{SRS} = \frac{\sum_{i=1}^{n} y_i L_i}{\sum_{i=1}^{n} L_i}$$

$$\widehat{se(D_{SRS})} = \sqrt{\frac{1}{\bar{L}^2} \frac{\sum_{i=1}^{n} \left(y_i L_i - \widehat{D} L_i\right)^2}{n}}$$

where

$\bar{L}$ is the mean transect length.

Notice that the finite population correction is ignored as for all intents and purposes it is 0.

Although simple random sampling across the entire coast would result in an unbiased estimate of the density of invertebrates, the distance between individual transects would be too great to be logistically feasible. Logistically, this would result in inefficient use of the sampling time, as completing multiple transects per day would not be possible in most cases due to the travel time between survey locations.

A more efficient method of placing 80 dive survey transects in a year would be to use a two-stage sampling design (Thompson et al. 1992). In this sample design a primary (or first) stage of sampling would be to choose a larger sampling unit at random that contains multiple smaller (secondary) sampling units. As an example, Figure 29 shows the 1 km by 1 km grid cells along the coastline (now termed secondary sampling units) within a larger 50 km by 50 km grid cell (primary sampling unit) imposed on the coastline of BC.

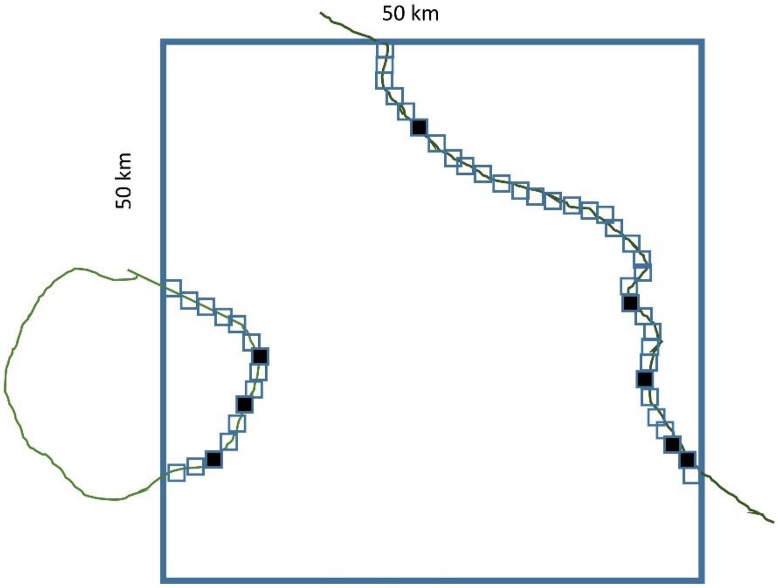

Figure 29. Example section of shoreline showing a larger (primary) sampling area (e.g., 50 km by 50 km squares) which is the first stage of sampling and the second stage (1 km by 1 km sections of shoreline) where dive transects would be placed. In this example, eight individual second stage samples were chosen at random within the first stage sample, which would have also been chosen at random.

In this example of a two-stage sample design, primary sampling units are chosen at random (n = 10) and then secondary sampling units (n = 8) are randomly chosen within each of the primary units for a total of 80 diver transects per year (Figure 30).

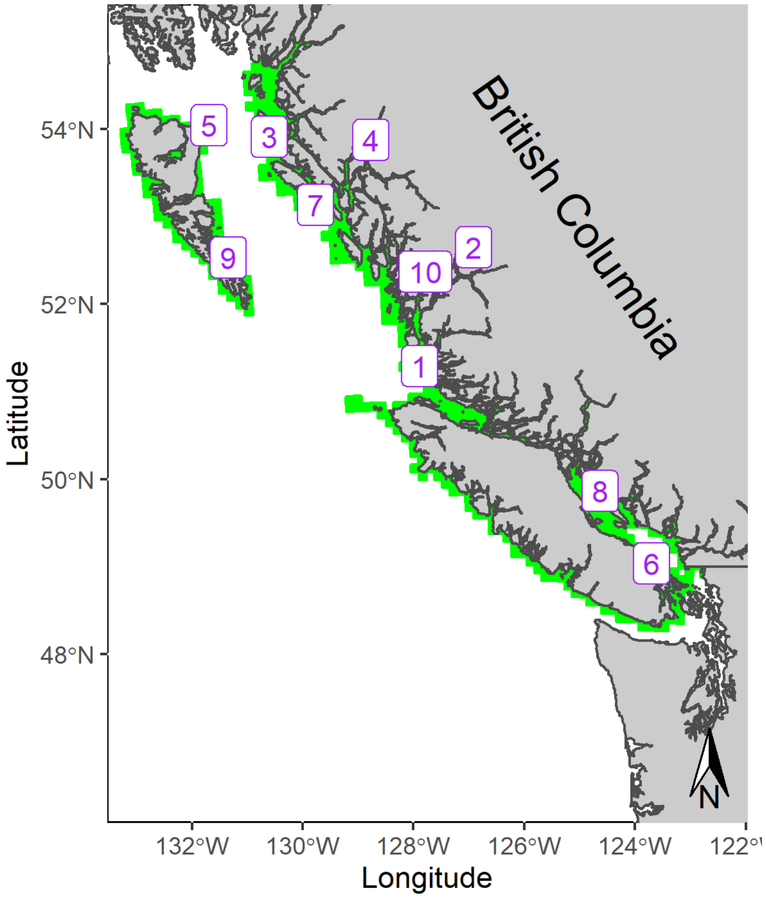

Figure 30. Example of two stage sampling where the primary stage are randomly selected 50 km by 50 km grid cells represented by the numbers 1-10 on the map and each of these primary units contains a random selection of 8 secondary stage sampling units (1 km by 1 km grid cells) of coastline where transects are placed. In this case 80 transects would be carried out in each year across the entire coast of BC.

For two-stage sample designs, y_{ij} is the density for an invertebrate species on second stage sampling units (dive transects i), and *j*th location (primary sampling unit). The ratio estimator is again used to estimate the density on each primary sampling unit ($\widehat{D}_j$) and associated standard error $se(\widehat{D}_j)$. The overall density estimate for an invertebrate species is thus the mean of primary stage density estimates:

$$\bar{y} = \frac{\sum_{j=1}^{n} \widehat{D}_j}{n}$$

where n is the number of primary locations sampled in a year. The estimated standard error is found by combining the variation among the primary and secondary units, as shown in Thompson et al. (1992).

There are advantages to the two-stage sample design over the simple random sampling design. Both provide an unbiased estimate of the coastwide density in each year but the two stage design is also more practical from a logistical standpoint and allows for better use of available survey time (through reduced travel time), as each of the randomly selected primary sampling units would contain eight transects that are within a fairly small distance from each other allowing multiple transects to be completed in a single day. The disadvantage of this sample design is that the primary sampling units are still very far distances apart. However, this issue is less of a concern than great distances between transects since travel between primary sampling could happen at night if a suitable vessel is available.

Other scenarios that could be considered consist of using a two-stage sampling design, but applied over two or three years (Figure 31). The further advantage of these examples would be to make surveying more efficient by reducing the travel distance between primary sampling units. The disadvantage of these examples would be that the entire coast would not be covered in a single year. This could result in a time-lag between changes in population size that would not be captured on an annual basis. For example, a large recruitment or mass mortality event occurring in a single year may not be detected until either 2 or 3 years later due to the rotational nature of sampling. A two-year lag (Figure 31; left panel) may be acceptable, especially as there may some differences in oceanography and biology between north and south BC that could put these invertebrate populations on differing trajectories. However, a three-year lag may be less desirable.

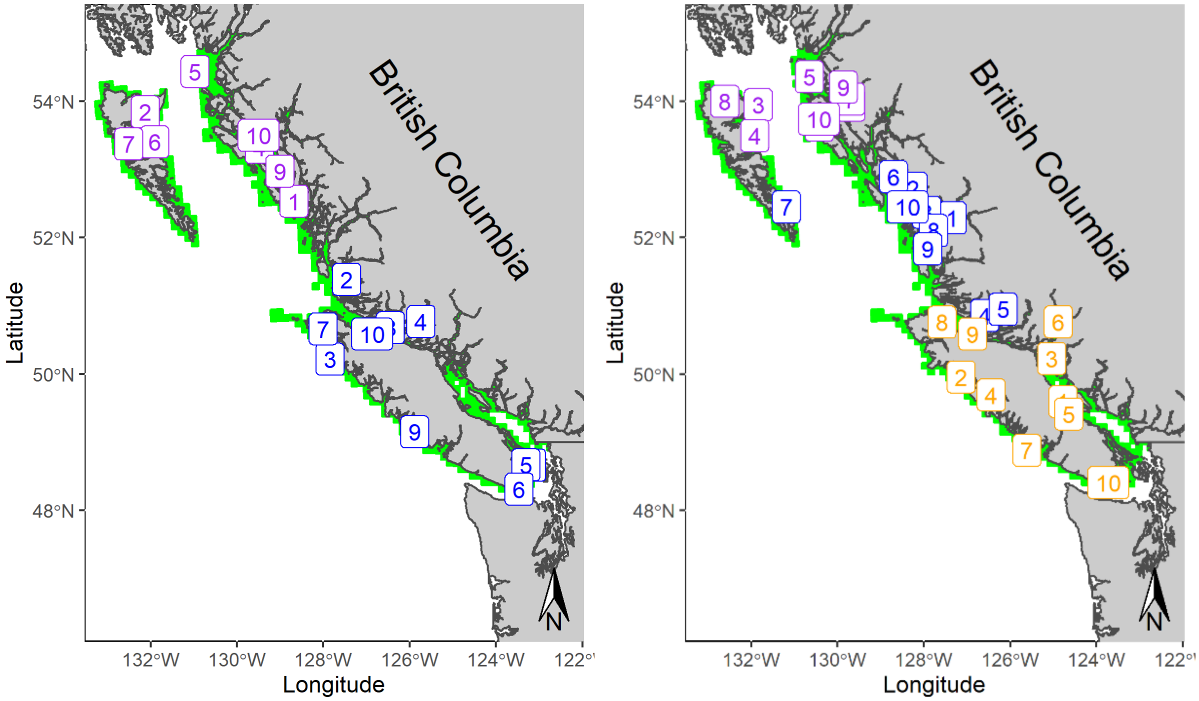

Figure 31. The left and right panels are examples of two stage sampling where the primary stage are the randomly selected 50 km by 50 km grid squares represented by the numbers 1-10 on the map and each of these primary samples contains a random selection of 8 secondary stage samples (1 km by 1 km grid cells) of coastline where transects are placed. On the left panel, the coast of BC is divided into a northern and a southern half and 80 transects would be carried out in each year so that the entire coast is sampled with 160 transects on a two-year cycle. On the right panel, the BC coast is divided into northern, central and southern portions and 80 transects would be carried out in each year so that the entire coast is sampled with a total of 240 transects on a three-year cycle.

Finally, a panel design was considered as another example. As implemented in this example, the panel design was a modification of the two-stage sampling protocol with a three-year cycle. In the panel design, a random sample of the previous year's locations would be chosen in each subsequent year. In this way, each year would repeat sampling at two primary sampling locations from the previous year (Figure 32). There are no simple analytical estimators and a model-assisted estimate would be necessary (Thompson et al. 1992). The advantage of the panel design would be that the repeated samples at some primary units would help to determine which part of a population density change in a given year was the result of temporal processes, like recruitment or mortality events and which portion was related to the spatial process (sampling in a new area in each year).

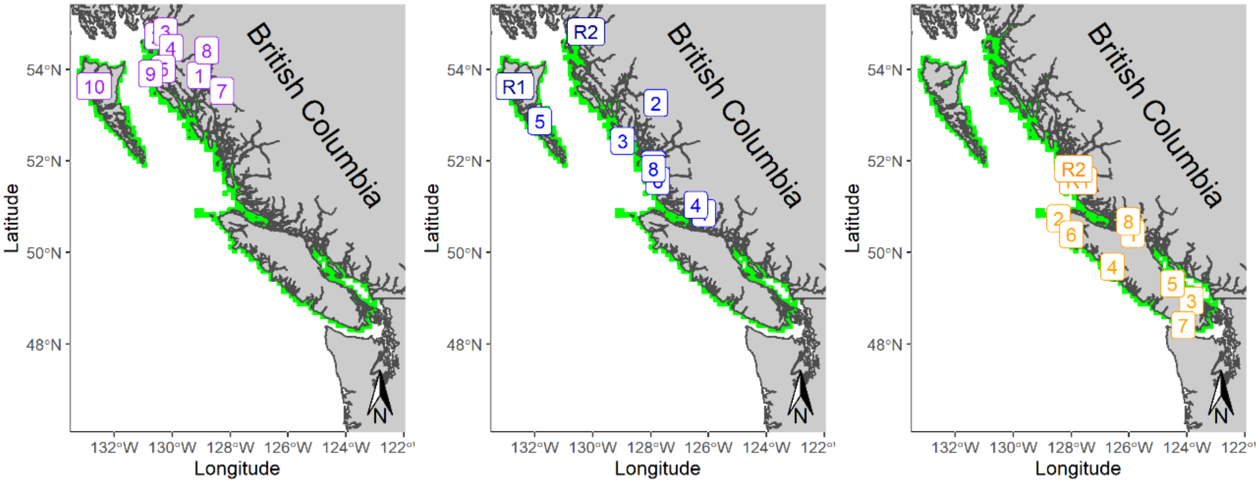

Figure 32. Example of two stage sampling with a panel design where the primary stage are the randomly selected 50 km by 50 km grid squares represented by the numbers 1-10 on the map and each of these primary samples contains a random selection of 8 secondary stage samples (1 km by 1 km grid cells) of coastline where transects are placed. In this case 80 transects would be carried out in each year and two of the primary sampling locations (represented by R1 and R2) from the previous year would be resampled in the following year. Here again, the coast of BC is divided into a northern, central and southern portion so that the entire coast is sampled on a three-year cycle.

4.4. DISCUSSION

Based on analyses of the multispecies survey data, the logistical considerations and the goal of collecting unbiased estimates of population density for invertebrate species, it is recommended to use a two-stage random sampling design that minimizes the time required to cover the entire BC coast and optimizes the efficient use of available resources. Ideally, the entire coast would be covered in a single year (roughly 42 days of ship time required). A realistic alternative would be to divide the coast in two or three sections and rotate through the sections over two or three years (requiring roughly 23 days or 18 days of ship time per year, respectively). Should the available resources (such as ship time, etc.) dictate a rotation longer than two years, then it is recommended that a panel design be considered so that temporal and spatial processes could be accounted for.

5. UNCERTAINTIES

5.1. DENSITY ESTIMATES

5.1.1. Depth coverage

Most of the species of interest to the multispecies dive survey extend to depths greater than the maximum survey depth (18.3 m uncorrected for tide) and also greater than the depths targeted by the dive fisheries (typically < 20 m). Geoducks have been documented down to 110 m depth (Jamison et al. 1984) but Mcdonald et al. (2015) observed low Geoduck densities between 30 and 60 m depth at four sites in Hood Canal, WA. Northern Abalone density decreases with depth (Figure 21) and generally occupy depth of less than 10 m (Sloan and Breen 1988) but have been observed to 40 m depth in the southern part of their range (Neuman et al. 2018). A submersible survey in central California observed Giant Red Sea Cucumbers down to a depth of 248 m, and the highest densities were found between 50-100 m (Blaine 2011). Green Sea Urchins can be found as deep at 300 m, but generally prefer the shallow subtidal zone from 0 to 50 m (Propp 1971; Scheibling and Hatcher 2013). Red Sea Urchins have been reported to depths up to 125 m in the central Salish Sea (Mortensen 1943) and a recent submersible survey near San Juan Island, WA, documented a Red Sea Urchin at 284 m, more than doubling this species’ known depth range (Lowe and Galloway 2020). Purple Sea Urchins are most common intertidally, however samples have been collected from as deep as 160 m (Morris et al. 1980). The depth range of Sunflower Star is from the intertidal the continental shelf break (~500 m) (Lambert 2000; Gravem et al. 2021).

Therefore undocumented biomass of the species of interest exists at depths greater than the survey and fishery depths. The multispecies survey may therefore provide an estimate of density or stock status for the portions of the stocks that are within survey/fishing depths but may not reflect status of the entire stocks since biomass below survey depths is undocumented.

5.1.2. Abalone

The search time for quadrats in the multispecies benthic invertebrate surveys is less than the search time typically spent on quadrats when conducting Northern Abalone surveys. Small abalone, especially those < 20 mm shell length, are less likely to be detected during multispecies surveys. In addition, abalone index site surveys and the multispecies survey use different protocols, i.e., Breen method for abalone (Obradovich et al. 2021) vs. random transects for multispecies. Breen survey plots measure seven metres perpendicular to shore; the maximum depth reached on a Breen survey plot is therefore 7 m, if the shallow end is placed at 0 m corrected for tide and the site is a vertical wall. The target maximum depth for multispecies surveys is 12 m corrected for tide. The depth range covered by the two types of surveys is therefore different. Consequently, Northern Abalone results from the multispecies survey are not directly comparable to those of the abalone index site surveys. However, the abalone results from the multispecies survey may provide a separate index of abundance for Northern Abalone and also provide further data on abalone distribution along the BC coast.

Cryptic sampling has been used on a portion of quadrats in some abalone surveys and consists of turning over rocks to look for hidden abalone (Hansen et al. 2020). Cryptic sampling can only be done in small boulder and/or cobble habitats and adds considerable time to a survey. For the purpose of multispecies dive surveys, the decision was made to not do cryptic sampling. Only animals that can be seen without moving the substrate are counted and/or measured. Therefore, the estimates obtained through these surveys may not be a true estimate of abundance but rather an index of abundance.

Cryptic behavior of Northern Abalone has been shown to differ in areas with and without Sea Otters; in areas without Sea Otters the probability of an abalone being cryptic decreased with increasing size while the probability of an abalone being cryptic did not decline with increasing size in areas occupied by Sea Otters, i.e., there was a higher probability of larger abalone being cryptic in areas occupied by Sea Otters (Obradovich et al. 2021). Although the multispecies dive survey will not sample for cryptic abalone, cryptic sampling is still done on the Northern Abalone index site surveys. The Northern Abalone population monitoring program will be ongoing and data on cryptic behavior of abalone will be available through that data set.

5.1.3. Geoducks

Geoduck show factor, the proportion of Geoducks that can be visually detected by survey divers, varies seasonally (Wayne Hajas, Quantitative Assessment Methods, DFO, Nanaimo, BC, 2022, pers. comm.). Geoduck show factor is highest between April and July and quantitative stock assessment surveys for Geoducks in BC are therefore scheduled during these months. Due to differences in survey methodologies, the Geoduck data from multispecies dive surveys cannot feed directly in the current methods to estimate Geoduck biomass. Depending on timing of multispecies dive surveys in the future, the Geoduck data may or may not be useful for quantitative stock assessment purposes. The Geoduck data however will be informative for determining locations where Geoducks are found outside of documented Geoduck beds and provide an additional index of Geoduck abundance.

5.1.4. Multiple years to cover entire coast

If the target number of transects and the coast of BC cannot be surveyed in a single survey/year, careful considerations to survey design should be given to balance priorities/objectives (e.g., detecting long term trends over time, estimating yearly densities, detecting rapid changes, etc.).

Using more than one year of data to determine coast wide densities relies on the assumption that density estimates for each region are stable during that period. This assumption may be valid most of the time for long-lived species with slow turnovers but may not be valid if a dramatic change in density occurs in one year. If more than one year of data will be needed to determine stock status, a lag effect will exist in the estimate of stock status. If a dramatic change in density occurs in a given year for a species, the stock status may not immediately reflect the magnitude of the decline since the density values for the previous year(s) used in estimating stock status would be from surveys that occurred pre-decline. This lag effect is unfortunately unavoidable without some resurveying of areas across years.

5.2. CLIMATE CHANGE

Climate change is having diverse impacts on the oceans and marine organisms. One such impact, ocean acidification (OA), is caused by increased atmospheric CO_2 from anthropogenic sources dissolving into the ocean resulting in increased concentration of CO_2 and thereby decreased pH in the ocean. Decreased ocean pH may hinder the ability of animals, including molluscs and echinoderms, to lay down shell/calcium carbonate structures at both larval and post-settlement life stages. This may consequently have impacts on survival and growth. Although much is still unknown about ocean acidification impacts, current research indicates potential negative direct and indirect effects to benthic marine invertebrates (Haigh et al. 2015). For example, in a laboratory experiment with the sea urchin *Paracentrotus lividus*, increased concentrations of CO_2 caused reduced shell thickness (Asnaghi et al. 2014). Red Sea Urchins were shown to require higher sperm concentration to achieve fertilization success with increased concentrations of CO_2 (Reuter et al. 2011). Female Purple Sea Urchins produced larger offspring (gastrula stage progeny) when experimentally conditioned to low pH levels (Wong et al. 2018). Reduced pH levels have been show to increase energetic demand in Pacific Geoduck larvae which resulted in smaller larvae and delayed development (Timmins-Schiffman et al. 2020). Huo et al. (2019) showed decreased larval hatching, survival, growth and metamorphosis rates at lower pH in *Panopea japonica*. In other studies, genetic markers have been used to infer possible physiological effects of OA. O'Donnell et al. (2009) measured the change in expression of a molecular protein in RSU, and linked the change to a reduced ability to handle temperature stress under OA. There are no known studies on OA effects for the Giant Red Sea Cucumber, however a study on a reef-dwelling sea cucumber species (*Holothuria* sp.) found impaired sperm motility at low pH values (Morita et al. 2010).

As climate change brings warmer water temperatures and instances of 'warm anomalies' (Bond et al. 2015), this may lead to increased occurrences of disease, such as sea star wasting disease (Harvell et al. 2019), as well as reduced growth and recovery rates of kelp (Krumhansl et al. 2017), an important food source for urchins and abalone. Given the important role Sea Stars have as predators, sea urchins have as grazers and sea cucumbers have as nutrient recyclers, any direct or indirect impacts of climate change on these species could have impacts on the ecosystem as a whole. The multispecies benthic invertebrate monitoring program, which also collects detailed information on algae, will be an important tool for monitoring potential climate change impacts.

The species of interest to the multispecies dive survey have broad geographical ranges. Geoduck and Northern Abalone range from Alaska to Mexico (Obradovich et al. 2021, Bureau 2017b). GSU are circumpolar in the Northern Hemisphere inhabiting all boreal and arctic marine regions (Sainte-Marie and Paille 2020). RSU are found on the west coast of North America as far south as the tip of Baja California (including the Gulf of California), northward to the Aleutian Islands, Alaska, and along the Asiatic coast as far south as the southern tip of Hokkaido Island, Japan (Campbell and Harbo 1992). PSU are distributed from the Kenai Peninsula in Alaska to the western coast of the Baja Peninsula (Olivares-Bañuelos et al. 2008; Field and Walker 2003). Sunflower Stars are commonly found in marine waters ranging from the Aleutian Islands, Alaska, south to San Diego, California (Gravem et al. 2021). Temperature changes related to climate change may therefore have relatively small direct impacts on the distribution range of these species within BC. However, potential impacts on depth distribution or other localized impacts are unknown.

6. GENERAL DISCUSSION

Long-term monitoring programs for marine species and their habitats are important tools for detecting and understanding changes in the nearshore ecosystem. Typically monitoring programs focus on a single species and are developed without clear objectives or much attention to survey design (Legg and Nagy 2006). The survey design, however, is key because it will to a large extent determine the accuracy of the abundance estimates and the interpretation of the results. Given the resources required to carry out long-term monitoring programs, it is prudent to define objectives and demonstrate that the data will be collected in a way to achieve those objectives, at the outset of any new monitoring program.

DFO Science, Pacific Region, is developing a long-term, coast wide, benthic marine invertebrate dive survey to move towards an ecosystem approach to stock monitoring, improve efficiency and allow the determination of stock status in relation to reference points for key BC marine invertebrate stocks. The species of interest include those species targeted by dive fisheries (i.e., Green Sea Urchin, Red Sea Urchin, Giant Red Sea Cucumber and Geoducks), the Purple Sea Urchin (whose abundance and distribution has shown dramatic shifts in

California in recent years (Rogers-Bennett and Catton 2019)), the endangered Northern Abalone, and an important mesopredator, the Sunflower Star (Burt et al. 2018; Schultz et al. 2016; Duggins 1983) whose population on the BC coast nearly collapsed between 2014 and 2015 due to sea star wasting disease (Hewson et al. 2014).

The Sunflower Star was listed in 2021 as critically endangered along its range in the eastern Pacific by the International Union for Conservation of Nature (IUCN) (Gravem et al. 2021). The multispecies dive survey will allow monitoring of the ecosystem effects of the *Pycnopodia helianthoides* reduction and also its population recovery. Of particular interest are the potential impacts of the decrease in abundance of the Sunflower Star on the density of its common prey species, which include sea urchins, sea cucumbers and abalone. Using the multispecies dive survey data to monitor Sunflower Stars and their prey is an example of how the multispecies survey can contribute to Ecosystem Based Fisheries Management.

Before the first multispecies benthic invertebrate survey, information from pre-existing benthic marine invertebrate dive surveys and in-water trials were used to develop and refine the dive survey protocol. The resulting sampling scheme that was based on transect length (Table 2), and the maximum transect length of 125 m, proved to repeatable and efficient. The dive survey protocol worked well within the logistical constraints of safe diving, with one transect generally being completed using a single tank of air per diver. Moving forward, it is recommended to continue with the protocol described in Appendix A, with the flexibility to add additional species and/or variables as needed. Further changes may be made as tools improve, e.g., new underwater data logging calipers.

Developing a survey design to meet objectives for multiple species can be challenging and will involve trade-offs. To help understand the variability in density among the different species we began by examining the ratio of the standard deviation of the transect-specific densities to the overall density. The sd:mean ratio for Red Sea Urchins was different than that for Green Sea Urchins, but there was no evidence of differences among any other species (Appendix D: Supplementary Material). Plots of standard deviation of the transect-specific densities against the overall density for the different survey regions showed that this difference, although statistically significant, was not dramatic and we chose to use a common sd:mean ratio of 1.27 for all species in determining a sampling plan.

The relative standard error across species and surveys suggested that the randomized multispecies survey provides a precise estimate of overall density. Although RSEs ranged from 0.08 to 1.0, the median RSE was 0.23 and in only two cases was the RSE > 0.5 (Table 5). Multispecies surveys have typically targeted a RSE of 0.20 to 0.40 across groundfish species[4] (Sinclair et al. 2003; Stanley et al. 2004; O’Driscoll and Ballara 2019; Blaine et al. 2020). Surveys for herring spawn were also designed to target a RSE of 0.25-0.30 (Schweigert et al. 1985; Boldt et al. 2015). Stanley et al. (2004) suggested a ranking system for RSEs that indicated RSEs < 0.20 as excellent, 0.20-0.30 as good and 0.30-0.40 as adequate for tracking population changes across time. The precision of the results from the invertebrate dive multispecies survey are consistent with a good ability to detect change in population size over time.

The limiting factor for sampling plans is the variability among transects in a survey area. This variability arises from local variation in densities among transects due to local factors, sampling variation in density on a transect because of natural variation along the transect and sampling

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[4] Bryan, D.R., Williams, K., and Rooper, C.N. In review. The design of a camera-based fisheries-independent survey for untrawlable habitat in the Gulf of Alaska. Fisheries Research.

variation in density on a transect because of skip sampling the quadrats. In most cases, the last two sources of variability will be small and the first source of variability is typically the largest (Skibo et al. 2008; Campbell et al. 1998).

One way to account for local variability in density among transects is stratification, whereby the study area is divided into strata that are more homogeneous. A sample of transects is taken from each stratum; the density is estimated for each stratum; and a weighted average of the stratum-specific density estimates gives the final estimate of density. If stratification is successful, the variation in density among transects within strata is less than the variation among transects over the entire study area and so the uncertainty of the final estimate will be smaller under stratification than without stratification.

Stratification can be done pre- or post-survey. With pre-survey stratification, the pre-survey stratification variable must be known for the entire coast when setting up the survey. With post-stratification, the sample can be divided into strata after the fact, but the stratification variable is still needed for the entire study area so that suitable weights can be derived. The key advantage of pre-survey stratification is the ability to re-allocate effort among strata (e.g., sample more transects in strata that are larger and have higher within-stratum variability) to further reduce the uncertainty in the final density estimate.

Four stratification variables of interest were evaluated: fetch, Sea Otter occupancy, dominant substrate and depth. For fetch, various cutoffs were evaluated, and the sd:mean ratio was similar for each cutoff. Plots of the relative standard errors for each species-year using an unstratified and assuming a proportional allocation stratified design indicated virtually no benefit from stratification at the various fetch cutoffs (Figure 19). Furthermore, approximately 80-90% of transects would be in the ‘high’ fetch stratum unless the fetch cutoff was set above 100,000 m.

Although Sea Otters can have a dramatic impact on the abundance of some benthic invertebrates (Lochead et al. 2019; Burt et al. 2018; Lee et al. 2018; Lee et al. 2016; Watson and Estes 2011; Kvitek et al. 1989; Pearse and Hines 1987; Lowry and Pearse 1973; Faro 1970; Ebert 1968), it was difficult to disentangle the impact of Sea Otter occupancy because it is highly confounded with the region. Year of occupancy was not a useful variable because the majority of transects were occupied in 2009, and there wasn’t enough data for other years. The binary (occupied, false/true) was investigated, and although the standard deviation of among-transect densities was reduced in the occupied-true stratum, the effect was not large (Figure 20). A comparison of the relative standard errors with and without stratification by Sea Otter occupied (false/true) did not show any benefit to stratification (Figure 21).

The dominant substrate variable was also not found to be useful for stratification. Due to the diversity in habitat preferences among the invertebrate species of interest, there was not a common dominant substrate suitable for stratification for all species (Figure 22). Additionally, if substrate type was to be used as a stratification variable, its distribution of the entire survey area would need to be known. At present, substrate distribution at the fine resolution needed is not known for the entire coast, but this information may become available in the future.

Similar to dominant substrate, depth distribution varied by species (Figure 23), and due to the lack of high resolution depth information for the nearshore, this variable was also found not to be useful for stratification at present. Although the substrate and depth data was not useful for post-stratification purposes, these data are still informative with regards to species depth distribution and substrate preferences.

The results of the comparisons of repeated transects to complete randomization between surveys were informative for designing future surveys. Although there was moderate correlation between repeated measurements, there did not appear to be substantial gains in precision that could be realized with repeated sampling. In addition to the absence of support based on the data collected here, other studies (e.g., Ryan and Heyward 2003; Perkins et al. 2019) have demonstrated the difficulty in performing repeated transects over even short time and spatial scales and the impact of spatial patterns on abundance estimates. Re-randomizing dive survey transects in each year will also allow the collection of data from new locations over time that may be able to be evaluated to improve stratification (e.g., data collection over dissimilar values of fetch may help to resolve density differences among categories) or allow switching to different models for estimating abundance over the survey areas. An example of the latter could be the application of spatio-temporal models (e.g., Anderson et al. 2022; Thorson et al. 2019) that could take advantage of spatial structure in the survey data to produce a better time series of abundance that was corrected for extrinsic factors (e.g., substrate patterns in the survey area). Re-randomizing transect locations, although perhaps marginally more costly in the number of transects needed to survey the population, will lead to better knowledge about the inherent spatial and temporal variability in the populations and is easier to implement in the field.

In light of the new legislated requirements of the Fish Stocks Provisions in the revised *Fisheries Act*, it is important that the data from this monitoring program can be used to determine stock status in relation to reference points for benthic marine invertebrate stocks that will be passed into legislation, such as Giant Red Sea Cucumber and Red Sea Urchin. Green Sea Urchin is currently assessed in the relatively small areas on the south coast where it is fished, but the multispecies survey data may become important should a move to a coast wide assessment become desirable in the future.

For Red Sea Urchin, the reference points are specific to densities of certain size categories on urchin habitat, and Giant Red Sea Cucumber reference points are specific to sea cucumber habitat. For example, the Red Sea Urchin reference points (LRP = 0.3, USR = 0.6) are specific to densities of individuals > 50 mm TD on sea urchin habitat (Lochead et al. 2019). Similar subsets of the total density are used for some other species (Table 1). Using the habitat, size and density data from the pilot multispecies surveys, Red Sea Urchin densities were higher than the coast wide Upper Stock Reference of 0.60 per m^2, at 0.90 per m^2 on urchin habitat coast wide (Table 6; Figure 10). The coast wide estimate of Giant Red Sea Cucumber density was 0.193 per m^2 (on all habitat), which also exceeds the coast wide USR of 0.038 per m^2 (Table 6; Figure 10). Green Sea Urchin reference points are specific to high density index sites in the inside waters of north east and south east Vancouver Island and may not be appropriate as coast wide reference points, therefore a coast wide example comparison is not provided.

Similarly, the survey data can be used to assess changes in invertebrate populations with changes in management structures. For example, the Gwaii Haanas Gina 'Waadluxan KilGuhlGa Land-Sea-People Management Plan 2018 set aside multiple usage zones for commercial harvesting within the Gwaii Haanas National Marine Conservation Area. These included multi-use areas (where some commercial harvest is allowed), strict protection areas (in accordance with IUCN II regulations; where commercial harvest is prohibited) and restricted access zones (which were excluded from this project). The multispecies dive survey found no significant differences in species densities between transects in multi-use and strict protection areas ($p = 0.875$), which was expected since the survey in Haida Gwaii took place one year after the closures were implemented, while there were significant differences in density among species ($p < 0.0001$, Figure 11). Future multispecies surveys can be used to monitor changes in invertebrate densities over time in multi-use areas to determine if harvest is having an impact on population densities through comparisons with strict protection and to monitor changes in invertebrate populations in un-harvested areas.

The development of the recommended number of transects for the sampling plan was largely dependent on the ability to detect with 95% confidence (level of risk determined by Fisheries

Managers) that the stock is below the LRP or above the USR, for the two species for which coast wide stock status will be evaluated via the monitoring program, Giant Red Sea Cucumber and Red Sea Urchin. These stocks are managed as one coast wide stock with many management subunits, and accordingly will be subject to one coast wide LRP and USR. The sampling plan was therefore developed for the whole BC coast. The total number of required transects was different between species, 39 for Red Sea Urchin and 240 for Giant Red Sea Cucumber, owing to the fact that the LRP is relatively much closer to the USR for Giant Red Sea Cucumber than for RSU. Therefore, to accommodate the species that requires the most sampling, we recommend that at least 240 transects be conducted over the whole coast of BC. It is not likely that a minimum of 240 transects, distributed across such a large geographical area, could be completed in one year with the current resources and limited ship time available for surveys. Logistical considerations, previous survey experience and recent ship time availability suggest that surveying approximately 80 to 120 transects could be achievable on a survey in a given year. Therefore, if 120 transects per year are possible, then two surveys/years would be required to reach the target minimum of 240 transects; it is therefore suggested that the survey effort along the BC coast be divided across two years. The coast could be divided into two sections of roughly equal suitable shoreline length with at least 120 transects conducted each section. Assignment of transects within sections of BC coast could follow the two-stage sampling plan described in Section 4.3. The resulting section densities could then be weighted based on the amount of suitable shoreline per section to provide a coast wide estimate of density using two years of data for each species/size class. These average coast wide density values could then be compared to reference points to determine stock status. Since two years of data would be required to determine coast wide stock status, the stock status could be determined once every two years, or, if annual stock status estimates are desired, a rotating average, using the latest two years of data, could be implemented.

For logistical and safety considerations, when planning future surveys, it is suggested to place a minimum of eight to ten transects within the primary sampling unit (i.e., the number that can be completed in a single day with two dive boats working). The size of units chosen for the primary sampling stage should therefore be large enough to accommodate 8 to 10 transects while also being small enough to ensure that travel distances between transects are reasonable to ensure 4 to 5 transects per boat per day can be completed. The survey areas chosen for the pilot surveys were not intended to be used for the long term monitoring program. Rather, they were chosen to provide data from a range of locations on the BC coast to inform the analyses performed here (e.g., common across species SD/mean ratio of 1.27) and inform future program design and needs (240 transects coastwide). Implementation of the long term monitoring program will involve defining the areas used for long term monitoring as well as the size and number of sampling units in the sampling plan.

Such multiyear plans are more suitable for species with slow turnovers so that the organisms present in the population in a given year are mostly those that were present the previous year, excluding losses (mortality and harvest) and including new recruits. The benthic invertebrates monitored with the multispecies dive survey have slow turnovers because of their long lifespans. Geoducks have life spans of at least 168 years (Bureau et al. 2002), Red Sea Urchins can live over 100 years (Ebert and Southon 2003), Purple Sea Urchins are thought to live over 50 years (Workman 1999) and Green Sea Urchins are estimated to live over 50 years (Russell et al. 1998). Longevity of the Sunflower Star is estimated to be up to 68 years, although 11-14 years might be more common (Gravem et al. 2021). Giant Red Sea Cucumber age is unknown, but year classes can be distinguished through analysis of length frequency data for the first three years, age at recruitment to the fishery is thought to be at least 4 years (DFO 2022e). Spreading the total number of required transects across multiple years will require care in interpretation of the density values. For example, such a multiyear survey will be slower to detect “disasters”, e.g., a widespread mortality event, since the coast wide density would be influenced by the previous years' data (prior to the die-off).

In British Columbia, changes in the nearshore ecosystem are occurring at relatively fine, incremental scales through the impacts of climate change (Bond et al. 2015), ocean acidification (Haigh et al. 2015), and Sea Otter recolonization (Lee et al. 2016; Nichol et al. 2015), as well as at broader scales through abrupt and unexpected impacts of phenomena such as sea star wasting disease (Burt et al. 2018; Schultz et al. 2016; Hewson et al. 2014). With more incremental and likely unexpected changes on the horizon, the multispecies benthic invertebrate monitoring program will be a powerful tool for reliably detecting changes in populations and evaluating status of benthic marine invertebrate species.

7. FUTURE WORK

Continued work to fill knowledge gaps and the incorporation of new information into future assessments and management procedures will be an on-going process. Although the results from the analyses using the candidate stratification variables did not show any benefit to stratification, stratification based on these and other variables (independently or combined) may still prove to be useful and could be revisited in the future. For example, future stratification could be based on probability of presence from species distribution models, or habitat suitability index models, as well as whether the area is open or closed to commercial harvest. Use of substrate data for stratification could be evaluated by summarizing raw substrate data into more general habitat categories (e.g., Gregr et al. 2013). Continuing to collaborate with the DFO Marine Mammal group through data sharing on Sea Otter range expansion and impacts to benthic invertebrate, as well as future work on climate change impacts, will be vital as the near-shore ecosystem continues to be impacted by these processes. Additionally, future work could utilize species distribution models and/or habitat suitability index models (e.g., Nephin et al. 2020) to further define the survey's sampling frame. Future comparisons of multispecies survey trends to trends observed on other surveys would be interesting to look for convergence/divergence, and could provide insight into both fine and broad scale population trends. Future research on larval dispersal and settlement could help provide a better understanding of source/sink dynamics of broadcast spawning benthic marine invertebrates. Additionally, investigations into invertebrate densities and distribution deeper than diveable depths (i.e., approximately 18 m) using cameras, remotely operated vehicles or other tools would provide important information on the stocks in their full depth ranges. Assessing stock status of Giant Red Sea Cucumber and Red Sea Urchin will be an important next step, as will discussions about the spatial scale of assessment for Green Sea Urchin.

8. RECOMMENDATIONS

1. Use the dive survey protocol described in Appendix A, including the quadrat skipping sampling scheme that is dependent on transect length, a target depth range of -2 m to 12.2 m relative to chart datum, and a maximum transect length of 125 m.
2. Exclude sections of shoreline with fetch values lower than 20,000 m or higher than 2.52 million m when randomly placing transects to ensure sampling effort is focused on suitable habitat while also avoiding potentially unsafe areas for diving.
3. Ensure surveys occur at the same time of year to avoid introducing seasonal variation to the data.
4. Use the common (across species) coast wide standard deviation-to-mean ratio of density (animals per m^2) equal to 1.27 (i.e., $\sigma / \mu = 1.27$) derived from the pilot surveys in

calculations to determine the initial target number of transects to be surveyed. The number of transects to be surveyed may be revised in the future if the standard deviation-to-mean ratio of density observed, once coastwide data is available, differs substantially from the 1.27 estimate derived from pilot surveys.

5. Survey at least 240 transects coast wide to estimate stock status to adequately manage the risks of making incorrect stock status determinations. This target number of transects is based on an Acceptance Sampling analysis, and uses the current reference points for Red Sea Cucumber (RSU) (Lochead et al. 2019) and Giant Red Sea Cucumbers (Hajas et al. 2023), predetermined risk tolerances, and field-derived estimates of variability from the pilot surveys. The target number of transects could change in the future as new data become available or if reference points, observed variability in the data, and/or risk tolerances change.
6. Implement a two-stage, random sampling design that minimizes the time required to cover the entire BC coast and optimizes the efficient use of available resources. Ideally, the entire coast would be covered in a single year (roughly 42 days of ship time required). A realistic alternative would be to divide the coast in two or three sections and rotate through the sections over two or three years (requiring roughly 23 days or 18 days of ship time per year, respectively). Should the available resources (such as ship time, etc.) dictate a rotation longer than two years, then it is recommended that a panel design be considered.
7. Continue to explore pre- and post-stratification variables to improve survey precision, as data become available. Although there is insufficient evidence from the present analyses to suggest that stratification by fetch, Sea Otter occupancy, substrate type, or depth improve survey precision at this time, the dynamic ecosystem occupied by these species implies that these or other variables could become relevant to the survey design in the future.

9. ACKNOWLEDGEMENTS

We would like to thank the many divers who assisted in developing the multispecies dive survey protocol and the numerous divers that assisted in the surveys. We would also like to thank the crews of the CCGS *Vector* and the CCGS *J. P. Tully* for their logistical support while conducting the surveys. We are grateful for the support and feedback provided by many First Nations during survey planning: Haida, Hesquiaht, Huu-ay-aht, Hupacasath, Kitkatla, Kitsumkalum, Lax Kw'alaams, Toquaht, Maa-nulth, Metlakatla, and Ucluelet Nations. Thank you to Rob Flemming of the Shellfish Data Unit for his expertise in invertebrate data management. We would also like to sincerely thank the two formal reviewers for this paper, Joanne Lessard and Mackenzie Mazur, along with all the participants of the regional peer review meeting. The valuable comments we received helped us improve the Research Document.

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APPENDIX A. MULTISPECIES DIVE SURVEY PROTOCOL

The goal of the multispecies dive survey is to determine quantitative estimates of density and sizes of selected species of benthic marine invertebrates (Red, Green and Purple Sea Urchins, Northern Abalone, Giant Red Sea Cucumber, Geoduck and Sunflower Star) along with recording associated habitat characteristics (substrate and algae community data). The data gathered through these surveys will be used primarily to monitor stock status of selected species over time.

A.1. SELECTION OF RANDOM TRANSECT LOCATIONS

Selection of transect locations occurs before the survey using GIS. Transect locations are randomly selected along the coastline for the survey areas of interest using the CHS_Pacific_High_Water_Coastline_Albers shapefile in ArcGIS. The anticipated total number of transects that can be completed on a survey is estimated by multiplying the number of dive days available by 5 transects per dive skiff per day, and by the number of dive skiffs (one or two) plus 20. The 20 additional transects are to account for transects that would randomly be selected in locations where diving is not logistically feasible, e.g., intertidal areas, or locations considered unworkable due to high-currents or physical obstructions, such as log booms and docks.

Once the survey areas of interest have been selected, a mask of each survey area is created in ArcGIS. The shoreline for the area of interest (within the mask) is clipped and dissolved into a single line.

Fetch is defined as the distance traveled by wind or waves across open water and can be used as an estimate of exposure of a site to wave action. In areas of high fetch, large waves can routinely build making it too dangerous to conduct dive surveys. In areas of low fetch, soft muddy or silty sediments can accumulate, water can be relatively stagnant and the species of interest are generally not present. Low and high fetch cutoffs, 20,000 m and 2,520,000 m respectively, were recommended for the multispecies dive survey to increase survey efficiency by eliminating some unsuitable habitats.

The Fetch_all_BC geodatabase is available on the Open Data Portal (Fetch All BC Geodatabase). The fetch geodatabase has a fetch estimate for points located every 50 m along the BC coastline. Total Fetch is estimated as the sum of the distance to land at each of 72 compass bearings (every 5 degrees) to a maximum distance of 200 km for each direction. A 26 m buffer is created around fetch points (halfway to the next fetch point + 1 m) to ensure adjacent buffers connect and dissolve together over the entire shoreline shapefile section of interest. Sections of shoreline with fetch < 20,000 m and >2,520,000 m are clipped out. Then transects are then randomly assigned to the remaining sections of shoreline as random points along the shoreline, using ArcGIS. The randomly selected transect locations are then reviewed and transects in non-navigable areas (e.g., intertidal zone, rivers, tidal lagoons) are eliminated.

A.2. FINDING TRANSECT LOCATIONS

Randomly selected transect locations are loaded onto the boat’s chart plotter and/or navigation computer before the survey. Once the dive boat arrives at a site, the survey team evaluates the conditions and determines if there are any safety concerns with the site location. Sites that are highly exposed to swell may not be surveyable depending on the conditions. If a site is deemed to be too exposed then it is skipped. Similarly, sites in extreme current areas, located upstream from tidal rapids or in un-navigable areas (e.g. lagoons where the entrance dries at low tide) are avoided.

A.2.1. Laying transects

Once the decision is made that a site can be surveyed, a transect line is laid. Transects are made of 25 m sections of lead-core rope that are marked every 5 m with a cable tie and transect sections are joined together by “C” connecting links.

When ready to lay a transect, the tide height is looked up on the navigation computer or tide charts to determine the shallow and deep target depths. Shallow target depth is -2 m chart datum (-6 ft, to cover all abalone habitat, i.e., 6 ft above chart datum) and deep target depth is 12.2 m (40 ft) below chart datum, but not exceeding 18.3 m (60 ft). For example, if tide height is 10 feet the target depth range is 4 feet to 50 feet. If tide height was 15 feet, then the target depth range is 9 feet to 55 feet.

The position of the shallow end of the transect should be as close to the randomly selected point as possible. Once the location of the shallow end of the transect is identified, the boat noses up to shore and the shallow end of the transect. A 2.27 kg or 5 pound cannon ball attached with a longline clip is thrown as close to shore as possible. At high tide it may only be necessary to drive towards shore until the shallow target depth is reached and drop the shallow cannon ball there. The latitude and longitude of the shallow end of the transect are recorded on the dive log sheet. The boat then backs away from shore in a perpendicular direction until the target maximum depth is reached on the depth sounder or a transect length of 125 m is reached (maximum transect length for multispecies dive surveys) or a depth of 18.3 m (60 feet) is reached, whichever comes first. The person deploying the transect must count sections of lead line laid out to determine transect length and the quadrat skipping pattern to be used by the divers. Once the target depth (or 125 m transect length) is reached, a float line (with a 4.54 kg or 10 pound cannon ball attached with a longline clip) is attached to the transect line to mark the deep end of the transect and the transect line sections are then separated at the next CC link segment. Latitude and longitude of the deep end of the transect are recorded on the dive log sheet.

The sampling interval between quadrats along a transect is dependent on the transect length. The purpose of the variable sampling interval is to ensure that enough quadrats are sampled on short transects while long transects do not take too long to survey. Quadrat skipping pattern along the transect is determined by the transect length as described in the table below.

Table A-1. Sampling scheme, quadrat skipping pattern, minimum and maximum number of sampled quadrats as a function of transect length (m) and number of sections of lead line for multispecies dive survey transects.

Transect Length (m)	# Sections of Lead Line	Sampling Scheme	Quadrat Skipping Pattern	Min # Quadrats	Max # Quadrats
0 – 25 m	1	every quadrat	No skip	14.0*	25
25 – 50 m	2	every 2nd quadrat	Skip 1 m	12.5	25
50 – 75 m	3	every 3rd quadrat	Skip 2 m	16.7	25
75 – 100 m	4	every 4th quadrat	Skip 3 m	18.8	25
100 – 125 m	5	every 5th quadrat**	Skip 4 m	20.0	25

**: For -2 m to 12 m target depth range, minimum transect length for vertical wall.*
***: Sample at every cable tie.*

For very long transects or when wind and/or current are very strong it may be easiest to lay transects with the boat driving away from shore in a forward direction to provide better steerage and ensure transects are perpendicular to shore. In these cases, transects should be laid on the upwind (or upstream) side of the boat.

A.3. DIVE SURVEY TRANSECT PROTOCOL

After the transect has been laid, the boat tender prepares the data sheets (Appendix A.4) for the divers and records date, divers, transect number, transect length, skipping pattern, target depths and digital underwater caliper serial number on the invertebrate data sheet. The header portion of the algae sheet is also filled out before the dive. After divers get ready, the boat drops divers off near the float tied to the deep end of the transect. After a surface check, divers descend together to the desired target start depth of the transect. Divers place the quadrat on the right-hand side of the transect line at the starting depth. If surveying in kelp beds, three-sided quadrats may be easier to handle than four-sided quadrats. If using three-sided quadrats the transect line can be used as the fourth side of the quadrat.

Each diver is assigned a different role. For each quadrat, Diver 1 carries a double-sided clipboard with both invertebrate and algae data sheets (Appendix A.4) and records quadrat number, quadrat depth, time of day, substrate codes (0 = wood; 1 = smooth bedrock; 2 = bedrock with crevices; 3 = boulders (> 30 cm); 4 = cobble (between 7.5 cm and 30 cm); 5 = gravel (between 2 cm and 7.5 cm); 6 = pea gravel (between 0.25 cm and 2 cm); 7 = sand; 9 = mud; 10 = crushed shell; and 11 = whole shell) and percentages for the three dominant substrate types, algae species found in the quadrat (see species list on data sheet) and the percent cover of algae in the following categories: canopy (>2 m tall), understory (30 cm to 2 m tall/long), turf (0 to 30 cm tall/long) and encrusting on the algae dive sheet.

While Diver 1 is recording substrate and algae data, Diver 2 counts Giant Red Sea Cucumbers and Geoducks and provides those counts to Diver 1. Adult and juvenile (defined as those smaller than a pencil) Giant Red Sea Cucumbers are counted and recorded separately; with juvenile counts recorded in brackets, e.g., for a quadrat with 2 adult and 1 juvenile Giant Red Sea Cucumber, data is recorded as: 2(1).

Diver 2 then starts measuring the other species of interest using electronic underwater data-logging calipers. Each invertebrate (of the target species) observed in a quadrat is measured if possible. The calipers used (ZebraTech Dive Calipers) allow for direct logging of two types of measurements. Red and Green Sea Urchin test diameters are recorded using the underwater electronic data logging calipers since those two species are the most frequently encountered. Green Sea Urchins measurements are logged using button 1 on the calipers while Red Sea Urchin measurements are logged using button 2. Once an urchin has been measured it must be moved outside the quadrat to ensure each urchin is only measured once and to be able to see if small urchins or abalone are hiding under the large urchins.

Measurements for other species (Purple Sea Urchin, Northern Abalone and Sunflower Star (*Pycnopodia*)) are shown to Diver 1 who records them on the invertebrate data sheet. Diver 1 looks at the measurement on the display and writes it down on the datasheet with “A”, “P” or “Py” before the measurement for Abalone, Purple Sea Urchins and Sunflower Star (*Pycnopodia*), respectively (e.g., A74, P35, Py122).

Underwater digital calipers are turned on by pressing one of the two buttons. The calipers turn off automatically after a period of 2 minutes of inactivity. When turning calipers on, close the caliper jaws and ensure the measurement reads 0.0 mm. If not, tare the calipers by pressing and holding both buttons simultaneously until the display reads “tare” (this takes several seconds). Ensure the calipers are on (display active) before starting measurements. If calipers have turned off, turn them back on by pressing one of the buttons before starting measurements.

Diver 1 also fills out the invertebrate data sheet, which requires communication between divers. Diver 2 notifies Diver 1 if there are Red and/or Green Sea Urchins in a quadrat and gives Diver

1 the count of Geoducks and Giant Red Sea Cucumbers in the quadrat. Diver 2 shows measurements to Diver 1 for abalone (shell length), Purple Sea Urchins (test diameter) and Sunflower Star (diameter, arm tip to arm tip) and Diver 1 records them on the invertebrate sheet. Diver 2 also shows Diver 1 the calipers when Diver 2 logs the end-of-quadrat sequence (required to determine which measurements belong to what quadrats, close the calipers and press buttons 1 and 2 in sequence to record two zero measurements in the file) and Diver 1 records the last measurement logged in the calipers for a given quadrat. The end-of-quadrat sequence must be recorded for all quadrats along a transect, including those quadrats where there are no animals to measure. This data is later used in the data processing steps to assign measurement to the correct quadrats.

If an animal cannot be measured (e.g., located in a crevice) then Diver 2 indicates to Diver 1 to record a +1 for the species counts (e.g., +2 RSU if two RSU were seen but could not be measured in the quadrat).

When filling out data sheets, divers must write down zeros or put a line through a cell when the count for a particular cell is zero. Missing/null values do not equal zeros and it is essential to be able to differentiate between missing data (blank cell) and zeros (zeros or line through cell).

Once both divers complete their tasks in the quadrat, one diver moves the quadrat ahead as required based on skipping pattern for the transect. When divers move the quadrat underwater, they must ensure that they do not move the quadrat more than 1 m ahead for every skip. Because quadrat skipping pattern is driven by transect length, skipping more than 1 m at a time would result in fewer quadrats being sampled on a transect than what was intended. The best way to ensure proper skipping is to note the position of the forward edge of the quadrat, pick up the quadrat, move it so that the back edge of the quadrat now rests in position where the forward edge was previously, touch the forward edge to the substrate to locate next position to move to and so on. This technique is usually faster and less cumbersome than rolling the quadrat. In dense kelp it may be necessary to roll the quadrat instead.

Divers keep surveying using the specified sampling interval (based on transect length) until the shallow target depth or shallow cannon ball is reached. Another consideration regarding the depth at which to stop surveying at the shallow end of a transect is exposure to weather, waves and swell. For sites in exposed portions of the coast, it may not be safe to survey to -2m chart datum depth and the divers should judge the safe depth at which to stop surveying.

After the last quadrat on a transect is surveyed, one diver retrieves the weight from the shallow end of the transect and brings it back to the boat. Divers should swim well away from shore before surfacing and ensure that they surface together.

At the end of the dive, Diver 1 estimates and records the relative abundance (N [None] = 0, F [Few] = 1-10, M [Many] = 11-100, A [Abundant] > 100; for the whole dive, not just the sampled quadrats) of the following species: Green, Purple and Red Sea Urchins, Northern Abalone, Geoduck, and Giant Red Sea Cucumbers.

Once divers are back in the boat, the boat tender must ensure that data sheets are completely filled out, especially the transect length, skipping pattern and relative abundance of various species on the Invertebrate sheet. The dive tender should then enter dive start and end times on each underwater data sheet.

After the boat picks up the divers, the transect line is retrieved. The boat travels to the marker float at the deep end of the transect. The float is picked up and the float line and then the transect line are hauled back aboard. This is most easily done with the help of an electric line hauler (a.k.a. prawn trap puller).

A.3.1. Is an animal inside or outside the quadrat?

Animals that are located completely inside the quadrat are without question inside the quadrat. For animals that lie under the edge of the quadrat however, a set of rules must be used to ensure accuracy of density estimates and consistency between divers. Quadrats are built so that the visible area inside the quadrat edges is 1 m. The quadrat arms therefore are located outside (just at the edge of) the area to survey.

For sea urchins, an urchin is considered to be inside the quadrat if the center portion of the top of the urchin is inside the quadrat. If an urchin is partially in a quadrat but the center portion of the top of the test is outside the quadrat then the urchin is considered outside the quadrat.

Similarly, a Sunflower Star is considered to be inside the quadrat if the center portion of the top of the Sunflower Star is inside the quadrat. If a Sunflower Star is partially in a quadrat but the center portion of the top is outside the quadrat then the Sunflower Star is considered outside the quadrat.

For Geoducks, if more than half of the siphon is located in the quadrat then the Geoduck is counted as being in the quadrat.

If an abalone is more than half inside a quadrat, then it is counted as in the quadrat. Abalone with less than half the animal in the quadrat are considered to be outside the quadrat. If an abalone is half in and half out of the quadrat, only count it as inside the quadrat if the head of the abalone is in the quadrat.

Giant Red Sea Cucumbers are sometimes long, so the chance of being under the edge of the quadrat is higher than for other species of interest. If a sea cucumber is more than half inside a quadrat, then it is counted as in the quadrat. Giant Red Sea Cucumbers with less than half the animal in the quadrat are considered to be outside the quadrat. For Giant Red Sea Cucumbers that are half in and half out of the quadrat, only count it as inside the quadrat if the head of the cucumber is in the quadrat.

A.3.2. Sea Cucumber biological samples

The average weight and weight-frequency distribution of Giant Red Sea Cucumbers are determined from biological samples (“biosamples”). Biological samples are collected from a randomly selected sub-set of survey transects; one transect is randomly selected from every group of 10 transects in a subarea, or if the subareas are small and do not contain 10 transects, then one transect is selected for every group of 10 transects within a group of subareas. Starting in 2021, if no sea cucumbers are observed on a biosample transect, samples are collected from other transects (not initially selected as biosample transects) when sea cucumbers were observed (opportunistic sampling).

After a biosample transect is surveyed, the divers handpick the first 25 sea cucumbers observed from the transect line and surrounding area up to a maximum depth of 15.2 m (50 feet) (Duprey and Stanton 2018, 2015; Duprey 2014, 2011) and put the sea cucumbers in a pick bag (i.e., a mesh bag). The cucumbers are brought onboard the dive skiff and held in buckets of sea water.

At the end of the day, when back onboard the Coast Guard vessel, the sea cucumbers are longitudinally split with a utility knife, any guts, gonads and respiratory trees are removed, and liquid is drained. Each sea cucumber is then individually weighed and “split weights” are recorded on sea cucumber biological sample sheets to the nearest gram.

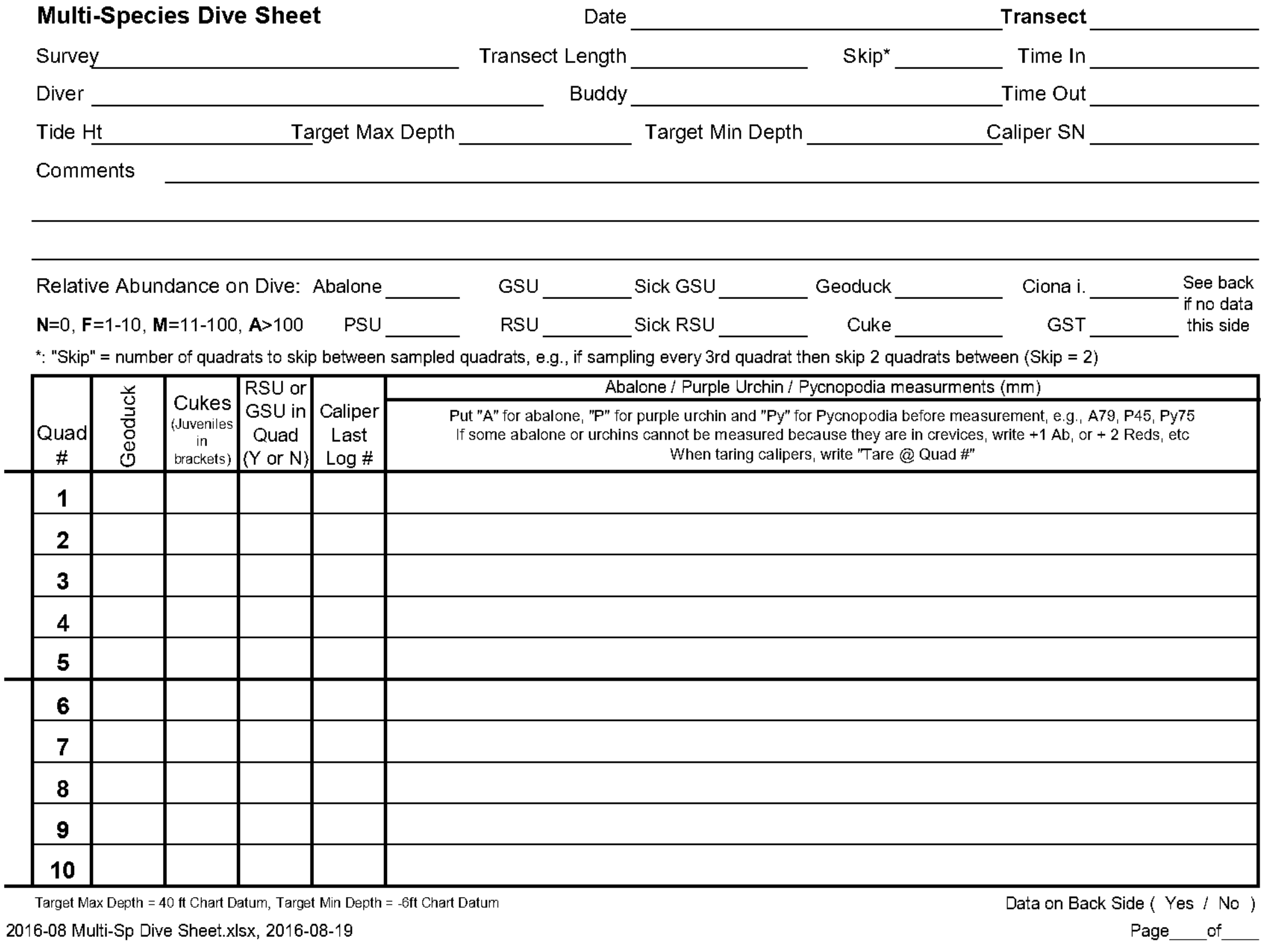
A.3.3. At the end of the survey day

At the end of a survey day, all data sheets are rinsed in freshwater to remove salt and hung to dry. Once the data sheets are dry, the diver who did the recording for each sheet checks their data to ensure all data is complete and legible and makes corrections as necessary. Someone then ensures all dive sheets for that day are accounted for and stored in a safe location. Data from the underwater data logging calipers is downloaded, checked and backed up on a separate storage unit (memory stick or external hard drive). Data on the calipers is then deleted and the calipers put on the charger overnight so that they are ready the next morning.

Table A-2. Equipment required to conduct multispecies dive surveys.

- Dive skiff and associated safety equipment (mandatory safety equipment, VHF radios, depth sounder, chart plotter, EPIRB)
- Navigation computer (rugged laptop with built-in GPS, to locate transect locations) or chart plotter
- Dive gear and associated safety equipment
- Transect lines (in 25 m sections connected with CC links and marked with cable ties every 5 m)
- Lead weights (cannon balls) for shallow end of transects (5 lbs) and deep end of float lines (10 lbs) with longline clips to attach them to the transect lines
- Float lines (100' long) to attach to deep end of transects with crab float, 10 lbs cannon ball and longline clip
- Electric line hauler (aka prawn trap hauler, e.g., Ace Line Hauler) to retrieve transects
- 3 and/or 4 sided 1 m x 1 m quadrat, two per boat so that a spare is available if one is lost
- Electronic (data logging) underwater calipers (ZebraTech Dive Calipers), two per boat so that a spare is available if one is lost
- Small plastic calipers or clear plastic ruler to measure abalone in crevices
- Underwater clipboards, clips, bungees and Bensia pencils, two per boat so that a spare is available if one is lost
- Underwater data sheets (Habitat (Substrate and Algae) and Invertebrates) and dive logs
- Clipboard for dive tender to keep completed dive sheets secure

A.4. MULTISPECIES SURVEY DATA SHEETS



Multi-Species Dive Sheet Date ______ **Transcept** ______

Survey ______ Transect Length ______ Skip* ______ Time In ______

Diver ______ Buddy ______ Time Out ______

Tide Ht ______ Target Max Depth ______ Target Min Depth ______ Caliper SN ______

Comments ______

Relative Abundance on Dive: Abalone ______ GSU ______ Sick GSU ______ Geoduck ______ Ciona i. ______ See back if no data this side

N=0, **F**=1-10, **M**=11-100, **A**>100 PSU ______ RSU ______ Sick RSU ______ Cuke ______ GST ______

*: "Skip" = number of quadrats to skip between sampled quadrats, e.g., if sampling every 3rd quadrat then skip 2 quadrats between (Skip = 2)

Quad #	Geoduck	Cukes (Juveniles in brackets)	RSU or GSU in Quad (Y or N)	Caliper Last Log #	Abalone / Purple Urchin / Pycnopodia measurments (mm) Put "A" for abalone, "P" for purple urchin and "Py" for Pycnopodia before measurement, e.g., A79, P45, Py75 If some abalone or urchins cannot be measured because they are in crevices, write +1 Ab, or + 2 Reds, etc When taring calipers, write "Tare @ Quad #"
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					

Target Max Depth = 40 ft Chart Datum, Target Min Depth = -6ft Chart Datum

Data on Back Side (Yes / No)

2016-08 Multi-Sp Dive Sheet.xlsx, 2016-08-19

Page____of____

Figure A-1. Invertebrate data sheet front page.

Comments ______________________________ **Transect** __________

Relative Abundance on Dive: Abalone ______ GSU ______ Sick GSU ______ Geoduck ______ Ciona i. ______

N=0, **F**=1-10, **M**=11-100, **A**>100 PSU ______ RSU ______ Sick RSU ______ Cuke ______ GST ______

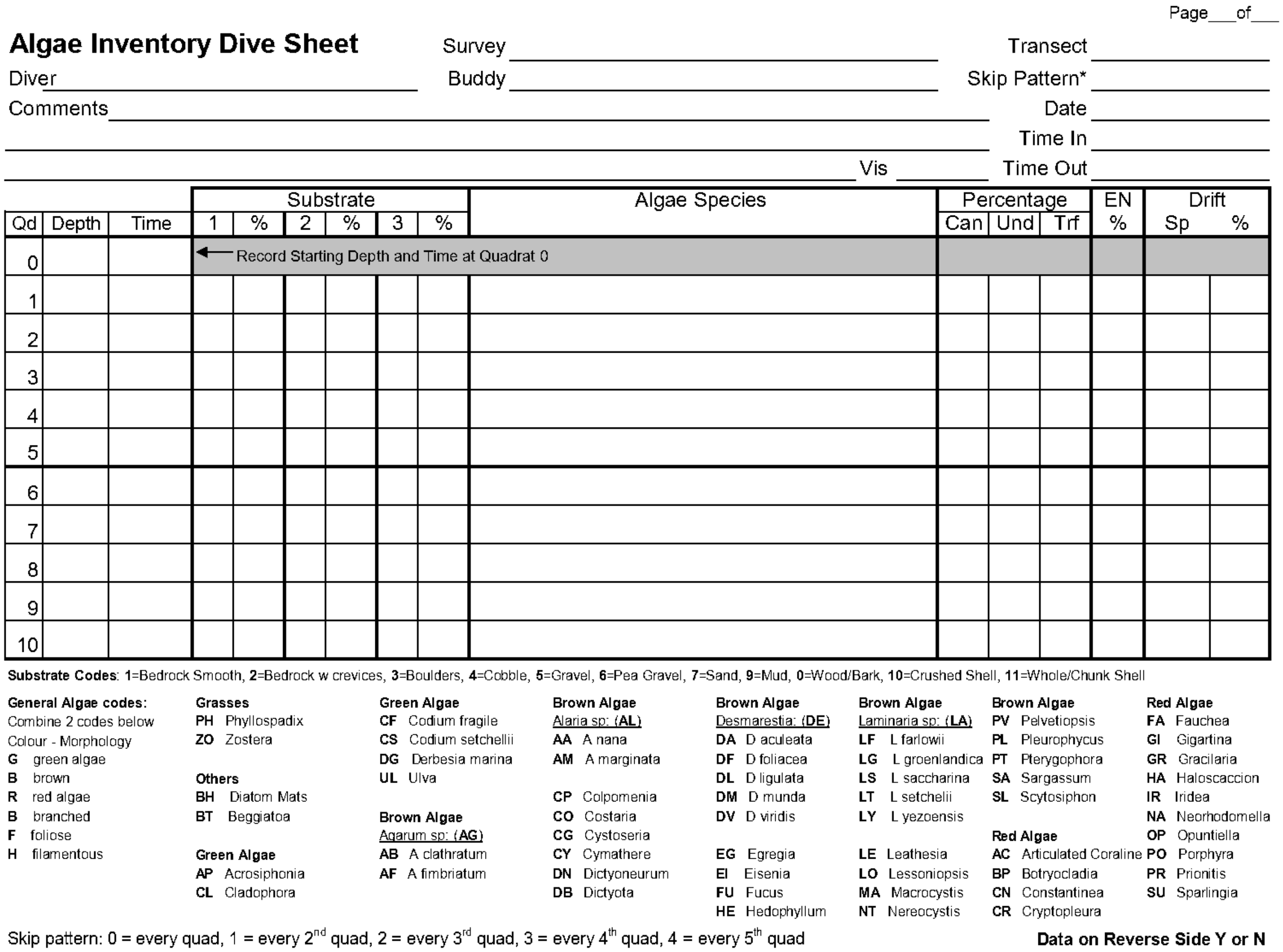
Quad #	Geoduck	Cukes (Juveniles in brackets)	RSU or GSU in Quad (Y or N)	Caliper Last Log #	Abalone / Purple Urchin / Pycnopodia measurments (mm) Put "A" for abalone, "P" for purple urchin and "Py" for Pycnopodia before measurement, e.g., A79, P45, Py75 If some abalone or urchins cannot be measured because they are in crevices, write +1 Ab, or + 2 Reds, etc When taring calipers, write "Tare @ Quad #"
11					
12					
13					
14					
15					
16					
17					
18					
19					
20					
21					
22					
23					
24					
25					

Data Continued on Another Sheet (Yes / No)

2016-08 Multi-Sp Dive Sheet.xlsx, 2016-08-19

Page____of____

Figure A-2. Invertebrate data sheet back page.



Page___of___

Algae Inventory Dive Sheet Survey ____________________ Transect ________

Diver ________________ Buddy ____________________ Skip Pattern* ________

Comments ______________________________ Date ________

Time In ________

Vis ______ Time Out ________

			Substrate						Algae Species	Percentage			EN	Drift	
Qd	Depth	Time	1	%	2	%	3	%		Can	Und	Trf	%	Sp	%
0			← Record Starting Depth and Time at Quadrat 0												
1															
2															
3															
4															
5															
6															
7															
8															
9															
10															

Substrate Codes: **1**=Bedrock Smooth, **2**=Bedrock w crevices, **3**=Boulders, **4**=Cobble, **5**=Gravel, **6**=Pea Gravel, **7**=Sand, **9**=Mud, **0**=Wood/Bark, **10**=Crushed Shell, **11**=Whole/Chunk Shell

General Algae codes:
Combine 2 codes below
Colour - Morphology
G green algae
B brown
R red algae
B branched
F foliose
H filamentous

Grasses
PH Phyllospadix
ZO Zostera

Others
BH Diatom Mats
BT Beggiatoa

Green Algae
AP Acrosiphonia
CL Cladophora

Green Algae
CF Codium fragile
CS Codium setchellii
DG Derbesia marina
UL Ulva

Brown Algae
Agarum sp: (**AG**)
AB A clathratum
AF A fimbriatum

Brown Algae
Alaria sp: (**AL**)
AA A nana
AM A marginata

CP Colpomenia
CO Costaria
CG Cystoseria
CY Cymathere
DN Dictyoneurum
DB Dictyota

Brown Algae
Desmarestia: (**DE**)
DA D aculeata
DF D foliacea
DL D ligulata
DM D munda
DV D viridis

EG Egregia
EI Eisenia
FU Fucus
HE Hedophyllum

Brown Algae
Laminaria sp: (**LA**)
LF L farlowii
LG L groenlandica
LS L saccharina
LT L setchelii
LY L yezoensis

LE Leathesia
LO Lessoniopsis
MA Macrocystis
NT Nereocystis

Brown Algae
PV Pelvetiopsis
PL Pleurophycus
PT Pterygophora
SA Sargassum
SL Scytosiphon

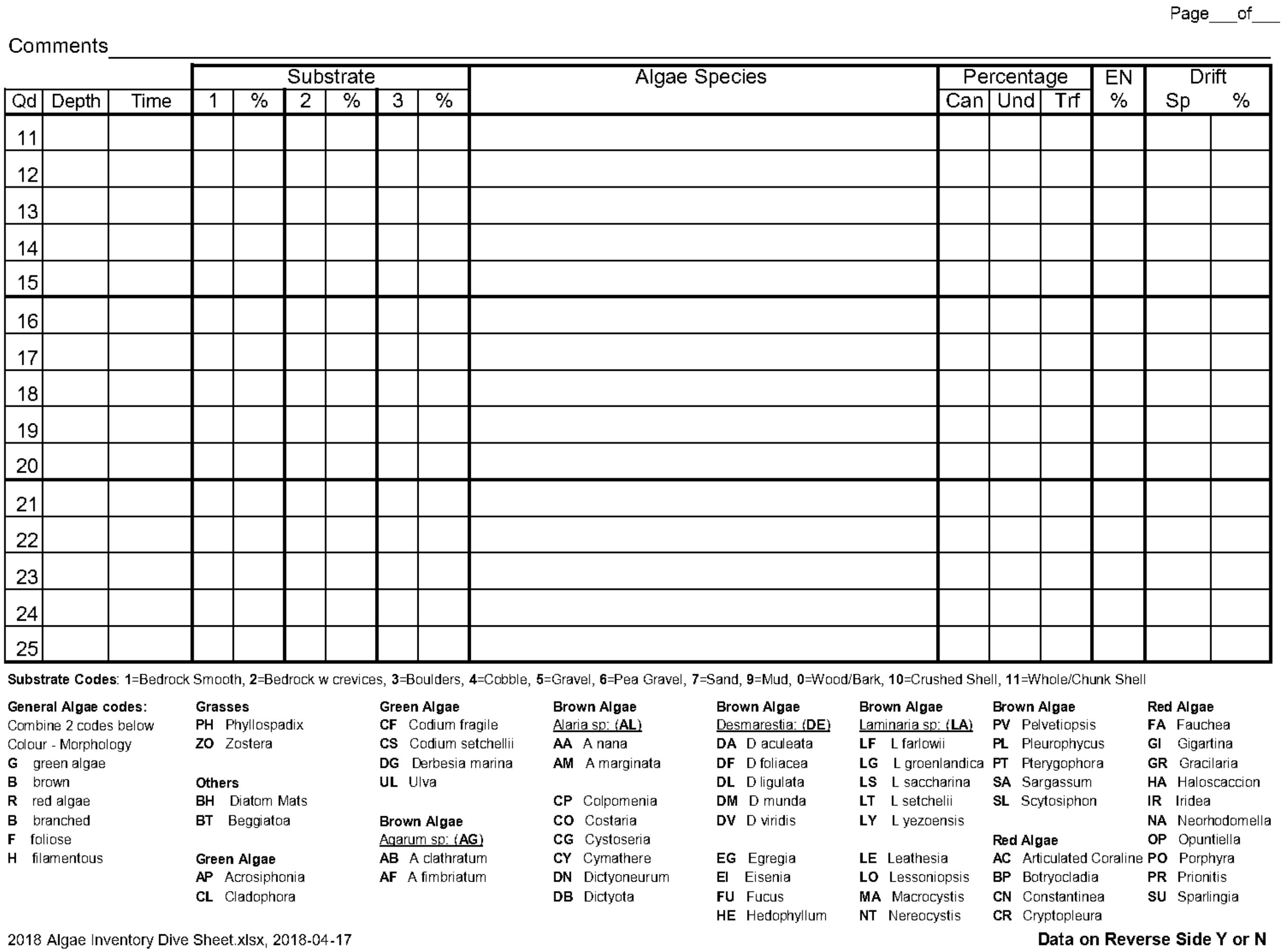
Red Algae
AC Articulated Coraline
BP Botryocladia
CN Constantinea
CR Cryptopleura

Red Algae
FA Fauchea
GI Gigartina
GR Gracilaria
HA Haloscaccion
IR Iridea
NA Neorhodomella
OP Opuntiella
PO Porphyra
PR Prionitis
SU Sparlingia

Skip pattern: 0 = every quad, 1 = every 2nd quad, 2 = every 3rd quad, 3 = every 4th quad, 4 = every 5th quad

Data on Reverse Side Y or N

Figure A-3. Algae and substrate data sheet front page. Depth and time of the first quadrat should be recorded under Quadrat 1 on the data sheet (do not record data in Quadrat 0 which is used for nearshore habitat mapping surveys only)



Page___of___

Comments____

			Substrate						Algae Species	Percentage			EN	Drift	
Qd	Depth	Time	1	%	2	%	3	%		Can	Und	Trf	%	Sp	%
11															
12															
13															
14															
15															
16															
17															
18															
19															
20															
21															
22															
23															
24															
25															

Substrate Codes: **1**=Bedrock Smooth, **2**=Bedrock w crevices, **3**=Boulders, **4**=Cobble, **5**=Gravel, **6**=Pea Gravel, **7**=Sand, **9**=Mud, **0**=Wood/Bark, **10**=Crushed Shell, **11**=Whole/Chunk Shell

General Algae codes:
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B brown
R red algae
B branched
F foliose
H filamentous

Grasses
PH Phyllospadix
ZO Zostera

Others
BH Diatom Mats
BT Beggiatoa

Green Algae
AP Acrosiphonia
CL Cladophora

Green Algae
CF Codium fragile
CS Codium setchellii
DG Derbesia marina
UL Ulva

Brown Algae
Agarum sp: **(AG)**
AB A clathratum
AF A fimbriatum

Brown Algae
Alaria sp: **(AL)**
AA A nana
AM A marginata

CP Colpomenia
CO Costaria
CG Cystoseria
CY Cymathere
DN Dictyoneurum
DB Dictyota

Brown Algae
Desmarestia: **(DE)**
DA D aculeata
DF D foliacea
DL D ligulata
DM D munda
DV D viridis

EG Egregia
EI Eisenia
FU Fucus
HE Hedophyllum

Brown Algae
Laminaria sp: **(LA)**
LF L farlowii
LG L groenlandica
LS L saccharina
LT L setchelii
LY L yezoensis

LE Leathesia
LO Lessoniopsis
MA Macrocystis
NT Nereocystis

Brown Algae
PV Pelvetiopsis
PL Pleurophycus
PT Pterygophora
SA Sargassum
SL Scytosiphon

Red Algae
AC Articulated Coraline
BP Botryocladia
CN Constantinea
CR Cryptopleura

Red Algae
FA Fauchea
GI Gigartina
GR Gracilaria
HA Haloscaccion
IR Iridea
NA Neorhodomella
OP Opuntiella
PO Porphyra
PR Prionitis
SU Sparlingia

2018 Algae Inventory Dive Sheet.xlsx, 2018-04-17

Data on Reverse Side Y or N

Figure A-4. Algae and substrate data sheet back page.

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Sea Cucumber BIOSAMPLES Sheet

Area	Sub-Area	Transect Number	Year	Month	Day	Sampler 1 Initials	Sampler 2 Initials	Sampler 3 Initials

Min Gague Depth	Max Gague Depth	Visibility

Cucumber Number	Split Weight (g)	Cucumber Number	Split Weight (g)
1		14	
2		15	
3		16	
4		17	
5		18	
6		19	
7		20	
8		21	
9		22	
10		23	
11		24	
12		25	
13			

Comments:

Figure A-5. Sea cucumber biological sample sheet.

APPENDIX B. SIZE FREQUENCY OF SURVEYED INVERTEBRATES

Invertebrate sizes were measured by divers in each quadrat for five species: Red , Green and Purple Sea Urchins, Northern Abalone and Sunflower Stars. Split weights for Giant Red Sea Cucumbers were also measured aboard the research vessel for a sample of individuals collected on dive survey transects. Details of the data collection can be found in Appendix A. To generate size frequency distributions for each species, the size measurements from a transect were weighted by the density of the species on that transect to obtain a size frequency histogram that reflected the size distribution for the sampled population. The size frequency distributions were similar for each species across areas where sufficient size data were collected. Red Sea Urchin size frequency distributions showed a peak at about 50 mm, with very few individuals observed at sizes above 150 mm (Figure A-1). Green Sea Urchins (Figure B-2) and Purple Sea Urchins (Figure B-3) were similar with peak sizes around 50 mm. However, Purple Sea Urchins in the inshore Vancouver Island regions also showed another peak in size distribution around 80 mm, but there were fewer individuals observed and measured for this species, so the size distribution may be less certain. Similarly very few Sunflower Stars were measured (relative to Red and Green Sea Urchins), so the size frequency distributions are discontinuous in most areas, with the exception of the inshore Vancouver Island surveys where a peak size of about 100 mm was observed (Figure B-4). Northern Abalone sizes peaked around 55 mm, with very few individuals larger than 100 mm, except in the west coast of Vancouver Island survey (Figure B-5). Giant Red Sea Cucumber split weights were measured during the east coast of Haida Gwaii and the west coast of Vancouver Island surveys. The median weight of Giant Red Sea Cucumbers in the surveys was 252 g (ranging from 18-1,384 g). The survey of the east coast of Haida Gwaii observed much larger sizes (ranging to the maximum of 1,384 g) while sea cucumbers on the west coast of Vancouver Island were only observed at sizes approaching 500 g (Figure B-6).

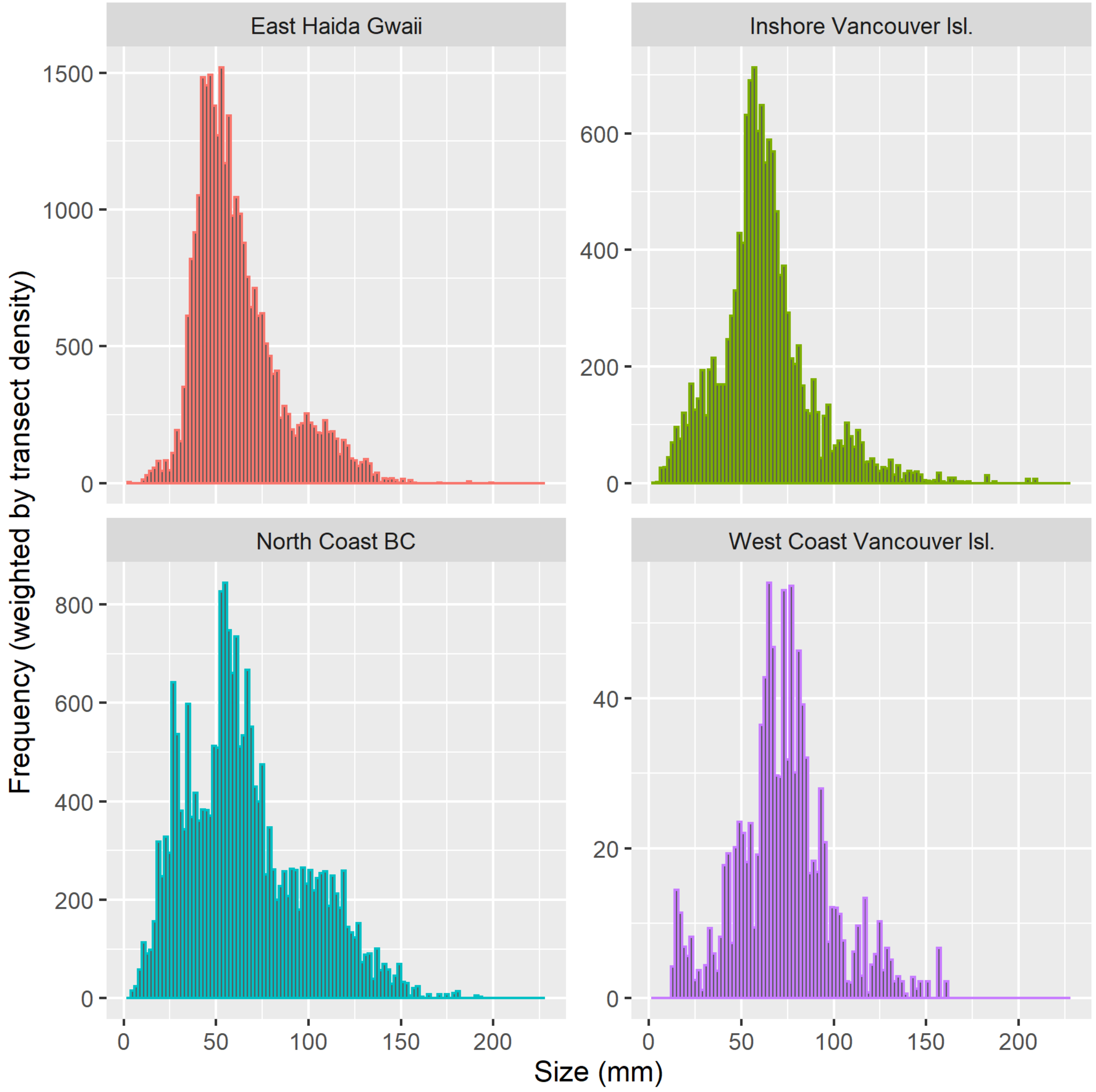

Figure B-1. Size frequency (test diameter) of Red Sea Urchins measured during surveys of the east coast of Haida Gwaii, inshore Vancouver Island, the north coast of BC and the west coast of Vancouver Island. Size frequencies are weighted by the density of individuals on each transect.

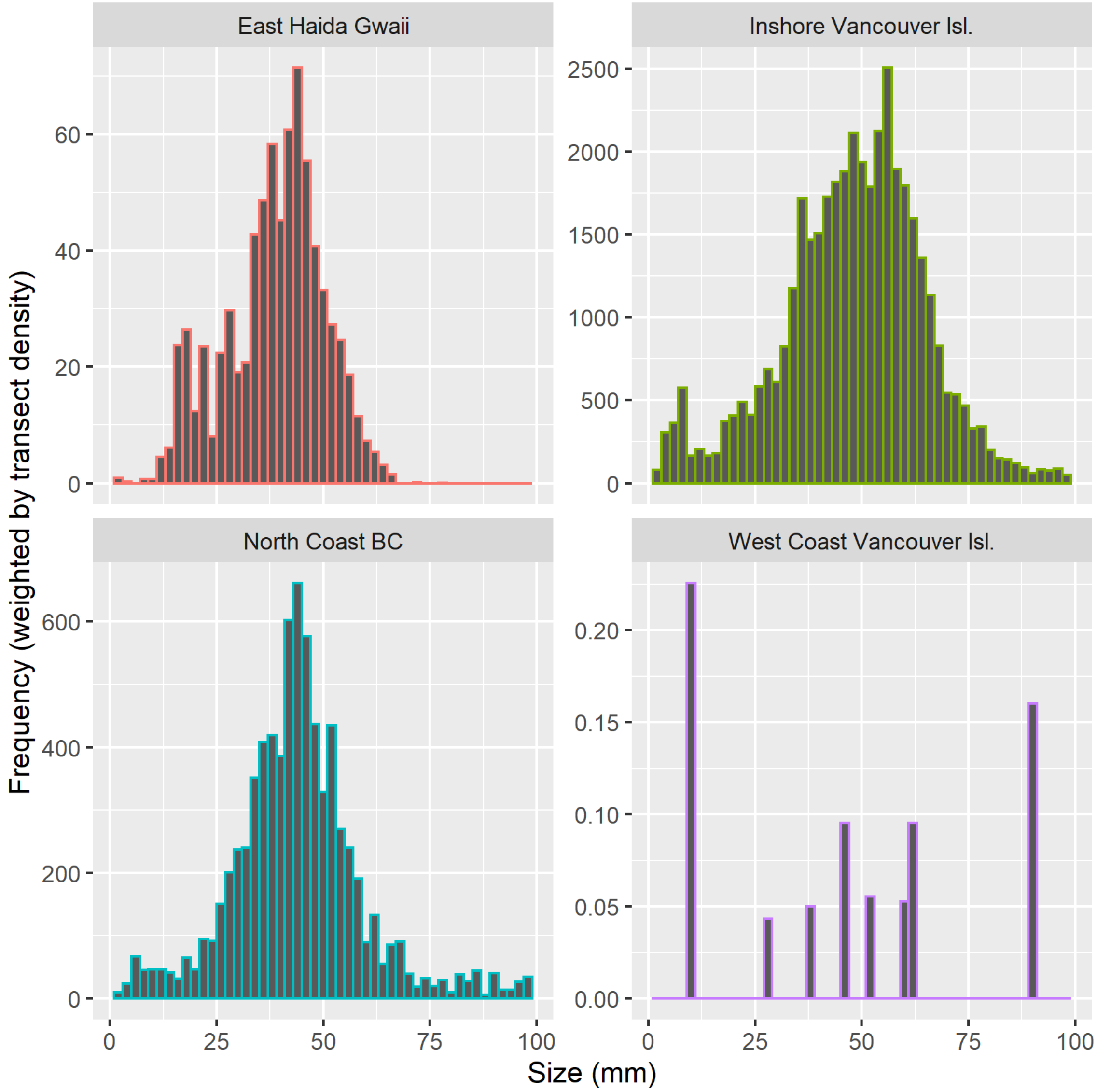

Figure B-2. Size frequency (test diameter) of Green Sea Urchin measured during surveys of the east coast of Haida Gwaii, inshore Vancouver Island, the north coast of BC and the west coast of Vancouver Island. Size frequencies are weighted by the density of individuals on each transect.

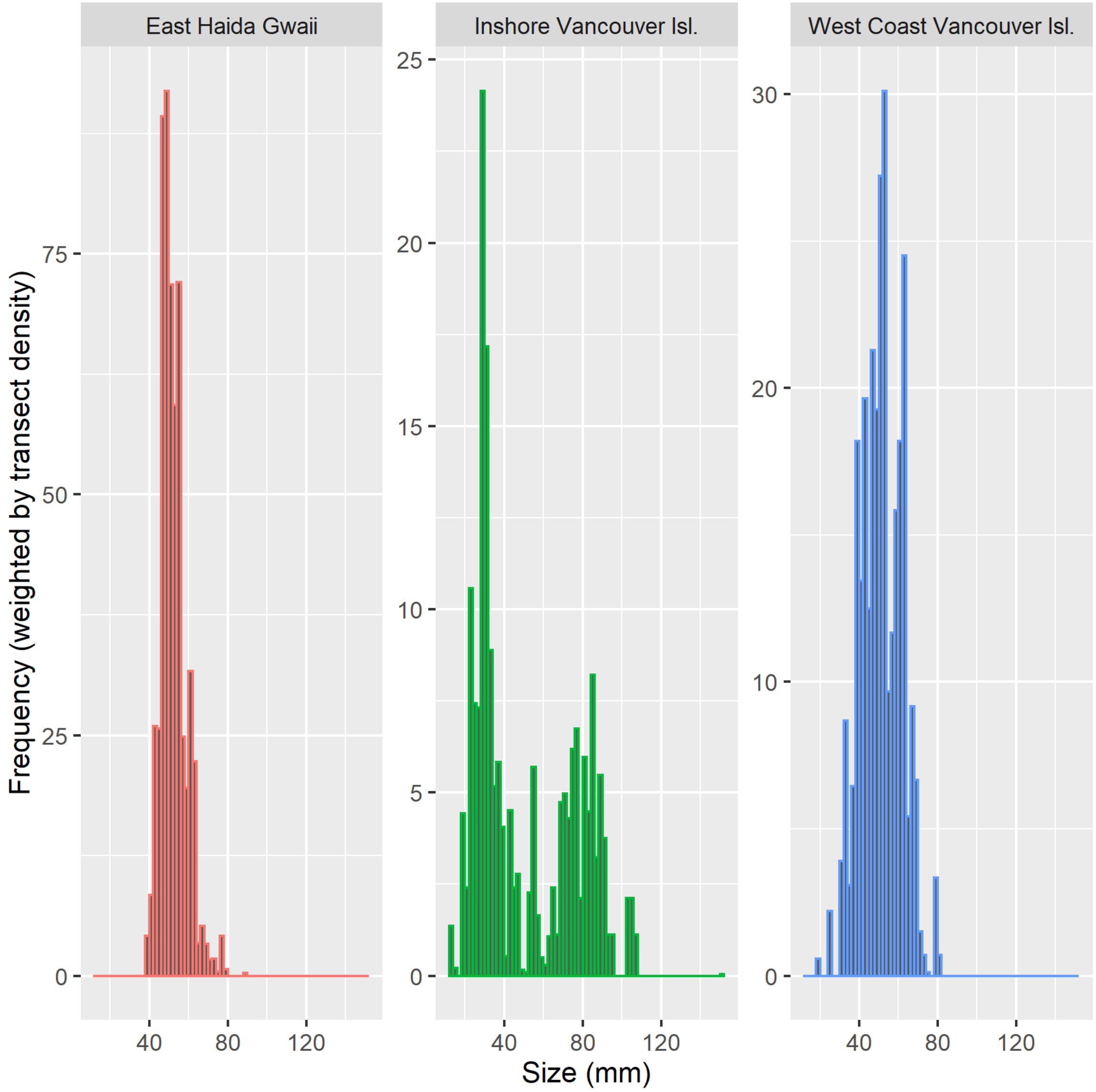

Figure B-3. Size frequency (test diameter) of Purple Sea Urchins measured during surveys of the east coast of Haida Gwaii, inshore Vancouver Island, and the west coast of Vancouver Island. No purple sea urchins were measured on the north coast of BC. Size frequencies are weighted by the density of individuals on each transect.

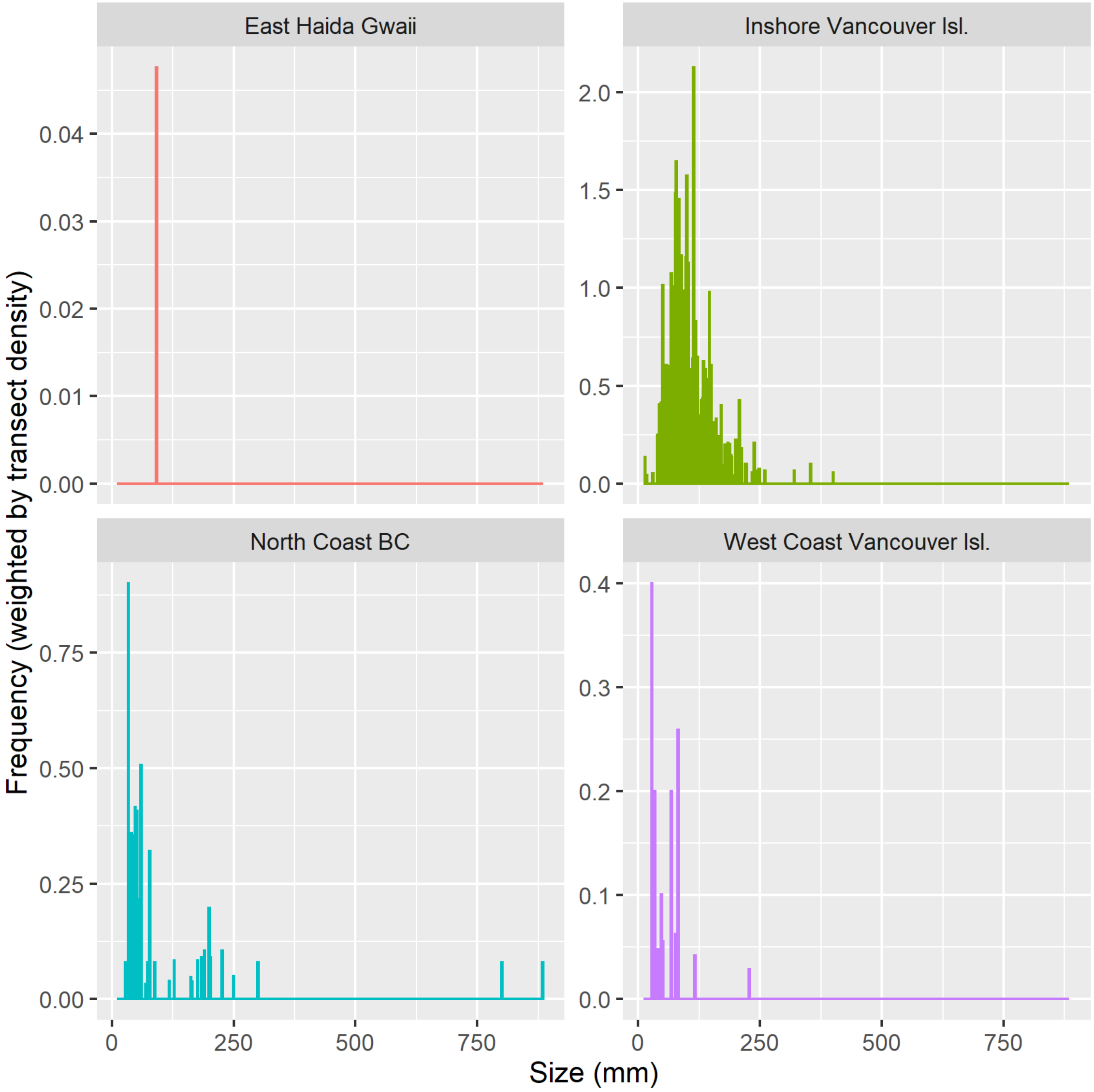

Figure B-4. Size frequency (total diameter) of Sunflower Stars measured during surveys of the east coast of Haida Gwaii, inshore Vancouver Island, the north coast of BC and the west coast of Vancouver Island. Size frequencies are weighted by the density of individuals on each transect.

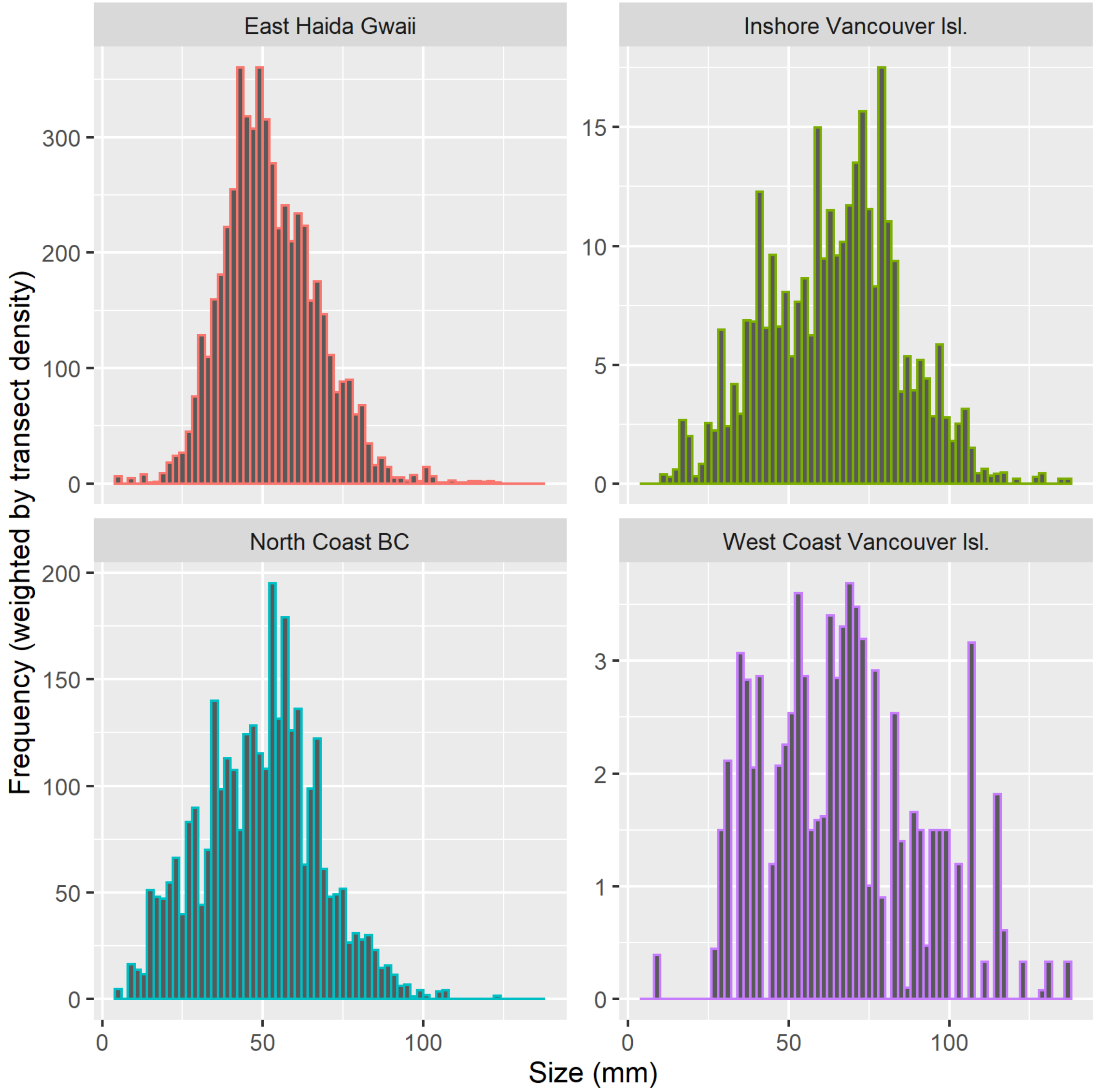

Figure B-5. Size frequency (shell length) of Northern Abalone measured during surveys of the east coast of Haida Gwaii, inshore Vancouver Island, the north coast of BC and the west coast of Vancouver Island. Size frequencies are weighted by the density of individuals on each transect.

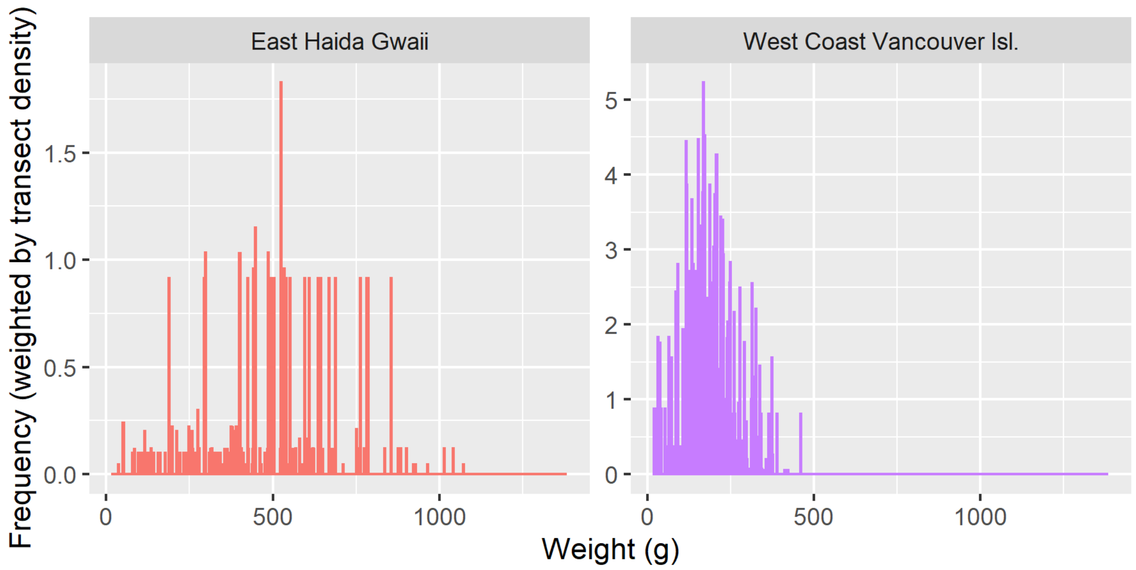

Figure B-6. Size frequency (split weight in grams) of Giant Red Sea Cucumbers weighed during surveys of the east coast of Haida Gwaii and the west coast of Vancouver Island. Giant Red Sea Cucumbers were not weighed during norther east Vancouver Island, south east Vancouver Island or north coast of BC dive surveys. Size frequencies are weighted by the density of individuals on each transect.

APPENDIX C. DOUBLE SAMPLING PLANS

The recommended minimum of 244 transects coastwide is an example of a single sampling plan where the sample size is fixed regardless of the underlying density. Double sampling plans were also explored. In a double-sampling plan, a smaller initial sample is taken and data are analyzed to determine if a decision (in this case stock status) can be made based on this smaller sample, or whether additional sampling is required. In a double sampling plan, an initial sample size of n_1 transects are taken, and the sample density at stage 1, $\widehat{Density}_1$ is determined.

Then,

> If $\widehat{Density}_1 < k - b$, then the stock status is deemed to be unhealthy and measures must be taken; or
>
> If $\widehat{Density}_1 > k + b$, then the stock status is considered to be healthy, and no further measures are taken,

The values of *k* and *b* again cannot be determined analytically, but require numerical methods as outlined in Appendix D: Supplementary Materials.

If the density at stage 1 is between *k-b* and *k+b, i.e.,*

> If $k - b < \widehat{Density}_1 < k + b$,

then a second sample of size n_2 is taken and the new density of the combined sample is determined, or $\widehat{Density}_{all}$.

A second decision is used,

> If $\widehat{Density}_{all} < k$, then the stock status is deemed to be unhealthy and measures must be taken; or
>
> If $\widehat{Density}_{all} > k$, then the stock status is considered to be healthy, and no further measures are taken.

The construction of the double plan used the same consumer and producer risks as for the singe-sample plan.

Double sampling plans were developed for Giant Red Sea Cucumber, Green Sea Urchin, and Red Sea Urchin (Table C-1; Figure C-1), with the same consumer and producer risks and LRP and USR values as the single sampling plan. In these plans, an initial small sample is taken, the density is calculated, and the sample density is evaluated against reference points to determine whether to stop or to continue sampling. For the double sampling plans, an initial sample of 19 transects are conducted for GSU and RSU, and 120 transects for GRSC. If the density in these first set of transects is below the stage 1 lower bound, then the density is considered to be too low and sampling stops. If the density in the first set of transects is above the stage 1 upper bound then the density is considered to be acceptable and sampling stops. If the density in the first set of transects is between the two cutoffs, then a second set of 39 is chosen for GSU and RSU, and a second set of 240 is chosen for GRSC. If the density from the combined sample is below *k*, the density is considered to be too low and conversely if the density from the combined set of transects is above *k*, the density is considered to be acceptable.

Table C-1. Summary of double sampling plans that match the single sampling plans developed in the main body of the document.

Species	n1	k	b	Stage 1 lower bound	Stage 1 upper bound	n2
Green Sea Urchin	19	0.60	0.078	0.522	0.678	39
Red Sea Urchin	19	0.40	0.052	0.348	0.452	39
Giant Red Sea Cucumber	120	0.03	0.002	0.031	0.035	240

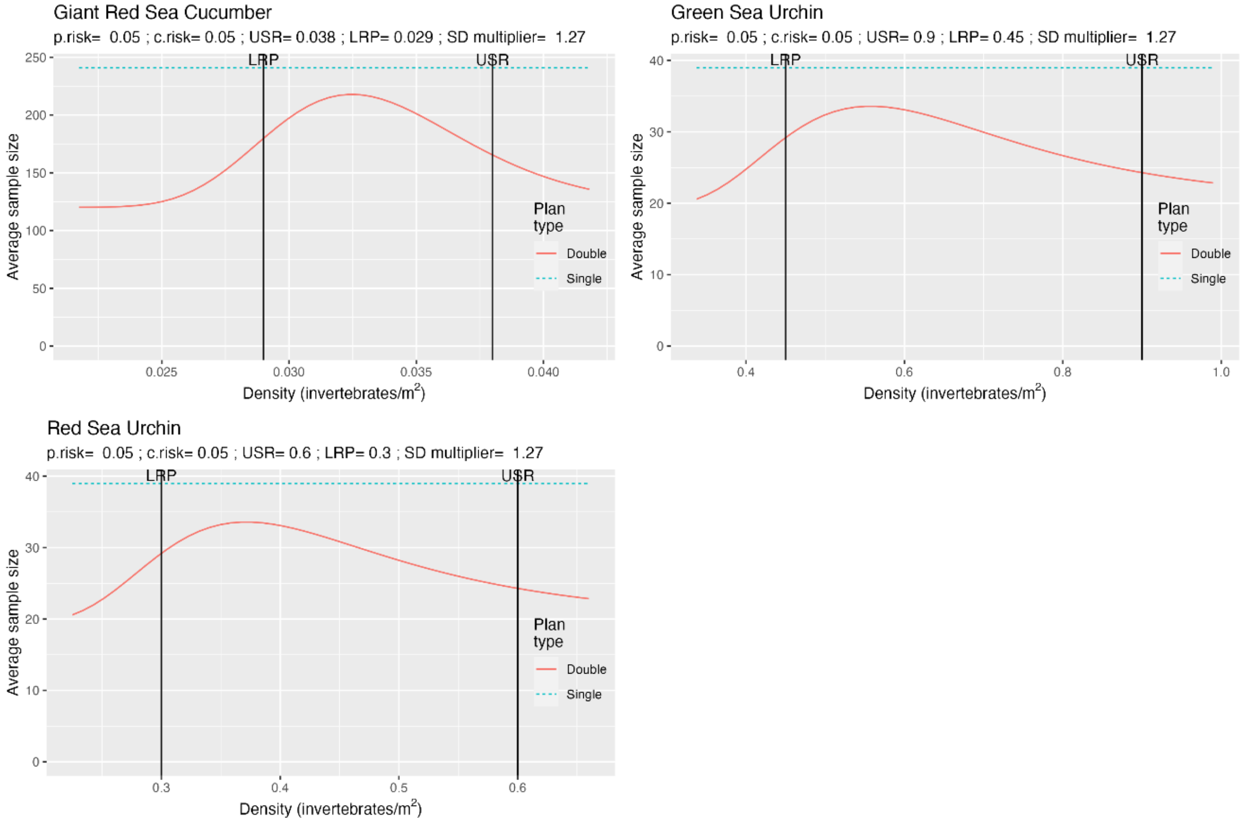

Figure C-1. Average sample size under single (blue dashed line) and double (red line) sampling plans for Giant Red Sea Cucumber, Green Sea Urchin and Red Sea Urchin.

This type of plan can be useful if the stock is well above the USR or well below the LRP because in those situations you may be able to reduce your sampling effort (i.e., the total number of transects). For the Giant Red Sea Cucumber, the total number of transects could be reduced to about 130 if densities are below 0.025 sea cucumbers per m^2 or above 0.043 sea cucumbers per m^2. Although attractive because it reduces the overall required resources, the double sampling plan will not be logistically feasible at this time because it requires real time density estimation during surveys, which would not be practical as data are recorded on underwater data sheets and there is not sufficient time while in the field to digitize and analyze data. A further issue with a double sampling plan is that, ideally, the reduced first sample would cover the entire coast but, as discussed previously, this is not logistically feasible with the multispecies dive survey. Consequently, double sampling plans are better suited to small geographic areas where randomization over the entire study area on each phase is logistically feasible. In addition, a double sampling plan would require the flexibility to add a survey in-season, if indicated by the results of the first survey, which is not feasible because ship time allocations are set months in advanced of the field season and cannot be modified in season.

APPENDIX D. SUPPLEMENTARY MATERIAL

Link to supplementary material.