

# **An evaluation of benthic monitoring baseline assessments completed as part of the regulatory requirements for aquaculture finfish site applications on the south coast of Newfoundland**

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2017

**Canadian Technical Report of  
Fisheries and Aquatic Sciences 3222**



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Cat. No. Fs97-6/3222E-PDF ISBN 978-0-660-09047-4 ISSN 1488-5379

Correct citation for this publication:

Hamoutene D., Salvo F., Belley R., Lush L., Hendry C. and Marshall K. 2017. An evaluation of benthic monitoring baseline assessments completed as part of the regulatory requirements for aquaculture finfish site applications on the South Coast of Newfoundland. Can. Tech. Rep. Fish. Aquat. Sci. Fs97-6/3222E-PDF: v + 25 p.

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## ABSTRACT

Hamoutene D., Salvo F., Belley R., Lush L., Hendry C. and Marshall K. 2017. An evaluation of benthic monitoring baseline assessments completed as part of the regulatory requirements for aquaculture finfish site applications on the South Coast of Newfoundland. Can. Tech. Rep. Fish. Aquat. Sci. Fs97 6/3222E-PDF: v + 25 p.

In aquaculture impact studies, it is necessary to acquire knowledge of benthic communities prior to the initiation of the activity to determine potential effects on ecosystems. We used baseline environmental survey reports of data collected from 2003 to 2011 by underwater video to document the composition of the seafloor in bays on the South Coast of Newfoundland where aquaculture activities occur. Organisms were identified at high taxonomic levels and were generally sparse and patchily distributed. We observed low natural taxa richness with the presence of barren stations. The dominant groups reported were: anemones, coralline algae, brittle stars, and other algae. Targeted sites and bays have a high level of substrate patchiness and were sometimes located over some sensitive habitats. Groups such as coralline algae score high in their uniqueness and vulnerability despite being classified as low productivity areas. Other habitats such as kelps (mostly present in Little Bay) and sponges (in Little Passage) were also observed. Despite a depth effect, there was a lack of obvious vertical zonation in taxa presence, suggesting that vertical distribution might be also governed by substrates and reflect the patchiness of the area. Recommendations for future baseline surveys are detailed in this document. In particular, baseline surveys have to be designed with explicit consideration of the legislative construct.

## RÉSUMÉ

Dans les études d'impact en aquaculture, la connaissance approfondie des communautés benthiques présentes avant l'initiation des activités est nécessaire pour déterminer les effets potentiels de l'aquaculture sur les écosystèmes. Nous avons utilisé des données issues de rapports établissant ces états de référence ayant été collectés entre 2003 et 2011 par vidéo sous-marine afin de documenter la composition du fond marin des baies de la côte ouest de Terre-Neuve où se situent les activités aquacoles. Les organismes ont été identifiés à un niveau taxonomique élevé et étaient distribués de manière éparse et non uniforme. Nous avons observé une faible richesse taxonomique naturelle ainsi que la présence de stations sans aucun organisme visible (epifaune). Les groupes dominants dans cette étude étaient : les anémones, les algues encroûtantes type corallinales, les ophiures et autres algues. Les sites et baies étudiés se sont révélés composés par une hétérogénéité de substrats avec parfois des habitats potentiellement fragiles. Le groupe des algues encroûtantes type corallinales a une forte valeur écologique par sa singularité et vulnérabilité bien que ces habitats soient considérés comme des communautés à faible productivité. D'autres habitats tels que les varechs (principalement in Little Bay) et les éponges (Little Passage) ont également été recensés. En dehors d'un effet de la profondeur, nous n'avons pas observé de zonation particulière dans la présence des groupes taxonomiques, suggérant que la distribution verticale peut être également le résultat de l'hétérogénéité de substrats. Des recommandations pour la future surveillance des états de références sont détaillées dans ce document. Plus particulièrement, les états de références ont besoin d'être évalués avec la considération explicite de l'appareil législatif.

## INTRODUCTION

Understanding any modification of the environment necessitates a good knowledge of reference starting points, making baseline data collection essential (Borja et al. 2012). In aquaculture impact studies, it is necessary to acquire sufficient knowledge of benthic communities prior to aquaculture to determine the nature and extent of the potential effect on ecosystems. Since 2002, Fisheries and Oceans Canada (DFO) has required the submission of baseline surveys of the proposed lease area as part of the aquaculture site licensing process administered by the Government of Newfoundland and Labrador (NL). Baseline surveys performed by environmental companies using video monitoring and sediment grab sampling include observations of taxa (presence/absence), topography, and substrate types.

Similar to some Norwegian counties (Taranger et al. 2014) and British Columbia (Abo et al. 2013), Newfoundland and Labrador (NL) finfish aquaculture sites are installed over hard substrates where grab sampling to assess benthic habitat changes due to organic enrichment can be challenging (Anderson et al. 2005; Hamoutene 2014; Hamoutene et al. 2013, 2014, 2015, 2016). As a result, local NL conditions necessitate the use of visual approaches for baseline assessments and documenting any benthic changes resulting from aquaculture instead of sulfide/redox sampling (DFO 2012; Mabrouk et al. 2014). There are currently no published surveys of the natural benthic communities around proposed NL aquaculture sites (Hamoutene et al. 2015). A comprehensive evaluation of the benthos in areas of NL targeted for aquaculture production is important for determining potential impacts to fish habitats supporting fisheries. In this report, we used baseline environmental survey reports of data collected from 2003 to 2011 by underwater video to document the composition of the seafloor prior to the implementation of aquaculture operations. Using these data, the aim of this document is to provide a description of the taxa and substrate of benthic habitats in bays of the South Coast with aquaculture activities.

In 2015, the Aquaculture Activities Regulations (AAR) further described below came into effect (DFO 2015). These regulatory requirements highlight the importance of knowledge acquisition on benthic communities prior to deposition; as part of these regulations, the owner or operator must submit the following information to the Minister at least 300 days before making a first deposit of a deleterious substance in accordance with methodologies prescribed in a Monitoring Standard and accompanying guidance document (DFO 2015):



- a) The predicted contours of the footprint of the biochemical oxygen demanding matter that will be deposited by the facility, calculated in accordance with the Monitoring Standard;
- b) A survey conducted in accordance with the Monitoring Standard that identifies the fish and fish habitat on the seabed that is leased for the operations of the facility and the water column above the seabed;
- c) The bathymetry of the seabed that is leased for the operations of the facility, measured in accordance with the Monitoring Standard; and
- d) In the case of a facility located over a soft bottom, the additional information that is specified in the Monitoring Standard concerning the seabed that is leased for the operations of the facility.

This report provides a snapshot of the data collected so far as part of the past aquaculture regulatory framework and examines the quality of these data and the potential improvements to be implemented to ensure information is useable by regulators and stakeholders. The 22 baseline assessments analysed were intended to address statutory obligations under Section 35 of the *Fisheries Act* to avoid a harmful alteration, disruption, or destruction (HADD) of fish habitat. Since the implementation of baseline assessments, the Fisheries Act was amended to refocus Section 35 on protection of fish habitat supporting commercial, recreational, or aboriginal fisheries rather than HADD avoidance per se and, in 2015, a new regulatory construct, the AAR were implemented – these latter regulations set the conditions under which aquaculture may impact fish habitat and deposit deleterious substances, that would not constitute a violation pursuant to Section 35 and 36 of the *Fisheries Act*, respectively. The baseline assessment guidelines are not yet amended to reflect those different legislative and regulatory requirements and objectives.

## **MATERIALS AND METHODS**

All data used in this study were extracted and compiled from baseline environmental reports. Data collection and reporting were performed by environmental companies using video monitoring and protocols established by DFO advice documents (DFO 2012; Mabrouk et al. 2014). Initial guidelines established by the DFO Habitat Protection Program suggest flexibility in how surveys used in this study (2003-11) were completed. In addition, due to surveying costs, logistical difficulties and the small number of stations over 100 m depth within most leases, surveys were limited by DFO Habitat Protection Program to 100 m depth contours.

### ***Video Sampling***

Video data were collected by environmental companies from 2003-11 as part of baseline environmental surveys, whose purpose is to provide knowledge of the seafloor composition prior to implementation of aquaculture operations/ site placement. Baseline environmental assessment protocols required video sampling according to a 100 m grid within the site boundaries/lease. Video footage of the bottom conditions was taken using an underwater camera (cameras were not always the same in these surveys), and at least one minute of video recording at the bottom was sampled at every station. At each station the following information was reported: site and station names, GPS coordinates, depth, bottom type (substrate), and an overall description of fauna and flora taxa. Twenty two reports were selected among the baseline reports available; some reports were not included as they described only partial information on grab sampling and/or provided very incomplete descriptions of stations sampled. The 22 sites selected were grouped in seven geographical regions (Figure 1) based on site proximity and geographical features.

### ***Data Extraction and Compilation***

There was a lack of consistency between reports in terms of data reporting and terminology. In order to ensure a more rigorous data interpretation we proceeded to the reclassification of the data (for every sampling location, i.e. station) as described below:

- The type of seafloor (i.e. substrate) was categorized in bedrock, coarse (boulder, rubble), medium (cobble, gravel), and fine (sand, mud), as described in Wentworth (1922).
- Organisms reported were compiled in presence / absence and classified at a high taxonomic level as per the main groups described in the reports to standardize the dataset.
- Organisms were categorized in corals, algae, coralline algae, kelp, anemones, sponges, sea stars, brittle stars, feather stars, urchins, sand dollars, mussels, scallops, shrimp, polychaetes, tunicates, crabs, and fish. Motile taxa (shrimp, crab, fish) were reported as their presence was noted when observed within the video segments.
- The dataset built for this study has the following information for each sampling station: station name, site name, GPS coordinates, depth (m), dominant substrate type and presence/absence of organisms.

Within the 22 selected reports, six baselines (in bold below) were completed at sites that had previous aquaculture activities. We have included these reports in our analyses

and have identified them in red in Table 1(a,b). As already stated, reports were grouped in seven geographical regions/bays (Figure 1) based on proximity of sites (Table 1a,b):

- Northern Arm (NTA): Sugarloaf Island, Wild Cove, Kilbuck Cove
- Upper Bay d'Espoir (UBE): Lou Cove, Arran Cove, Hardy Cove, Northwest Cove, Pomley Cove
- Little Passage (LP): Deer Cove, Blackfish Cove, Seal Nest Cove, Grip Cove, Robin Hood Cove
- Upper Hermitage Bay (UHB): Green Point, Herring Cove
- Little Bay (LB): Murphy Point
- Belle Bay West (BBW): Farmers Head, Chapel Island (two sites): AQ 1014 and 998, Grandy Rock, Bottle Hill
- Belle Bay East (BBE): Little Burdock Cove, Rencontre Island

### ***Data Description and Statistical Analysis***

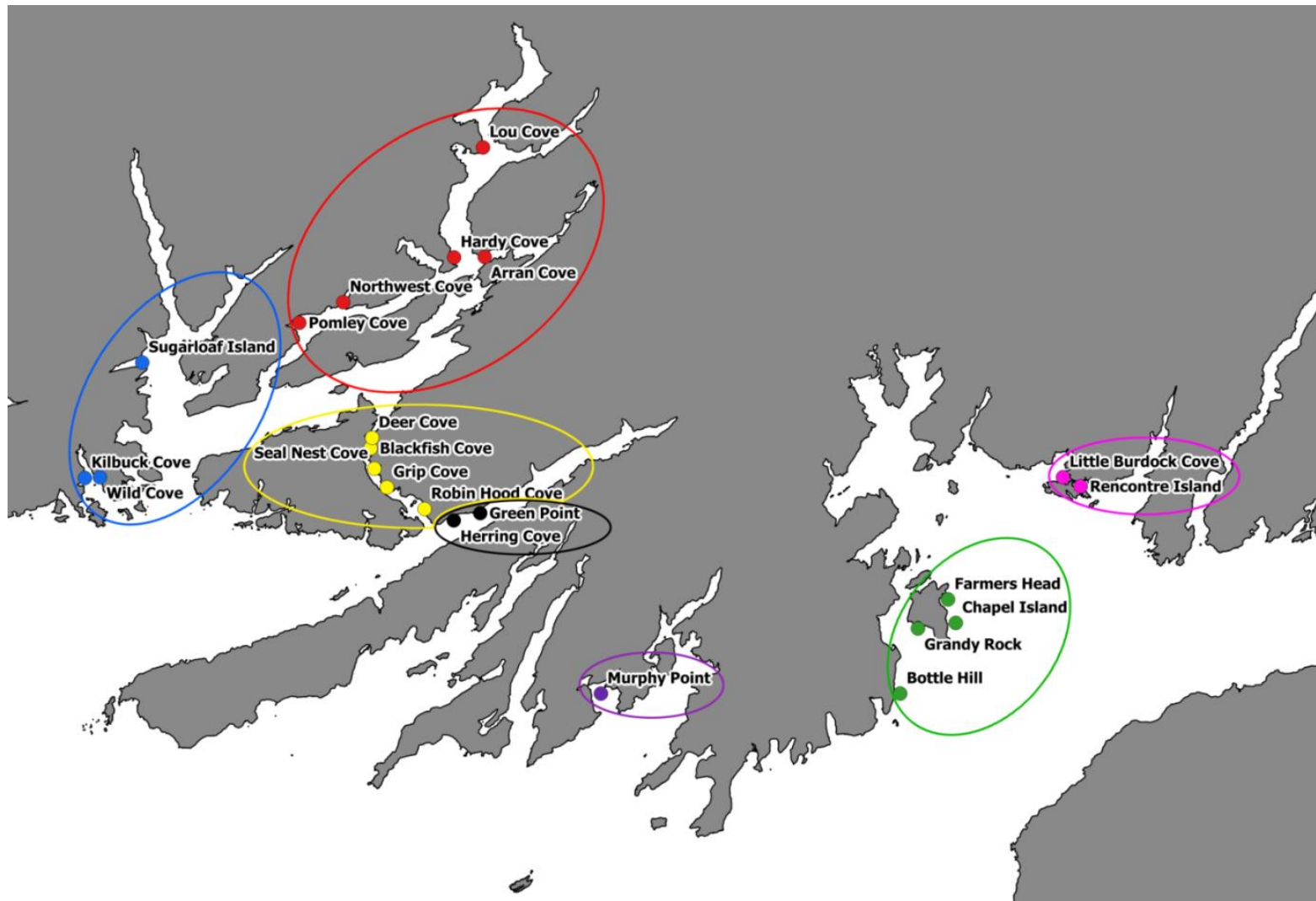
We used the software R version 3.3.1 (R Core Team 2016) to explore distribution of taxa with substrate and depth by representing taxa presence/absence data compiled per bay. Boxplots of taxa richness were also plotted per site.

Analyses of benthic assemblages were performed in PRIMER 7.0. and PERMANOVA+ add-on package (Clarke and Gorley 2015, Anderson et al. 2008). All data in PRIMER were entered as presence/absence and analyses completed as described below:

- To explore composition and similarity of substrate between sites and bays, the four categories of substrates were entered as presence/absence per station. Then, similarity percentage analyses (SIMPER) using Bray-Curtis distances for resemblance were completed to inform on the homogeneity of the substrate within a site and at the bay scale. Percentages of similarity were based on dominant substrate types within stations for sites and within sites for bay scale calculations.
- Likewise, similarity in terms of taxa between sites and bays was calculated using SIMPER analysis (Bray-Curtis distances for resemblance). In addition, we extracted from SIMPER results taxa responsible for most of the similarity (i.e. dominant taxa in terms of presence) within a site and its associated percent contribution to the similarity.
- Using the same taxa dataset, non-metric multidimensional scaling (nMDS) were used to explore benthic assemblages and permutational multivariate analysis of variance (PERMANOVA) to assess factors responsible for taxa distribution using simple matching similarity measures for binary data. PERMANOVA were used to test the effect of bay, site, and substrate on taxa composition; substrate was

considered as random and site and bay as fixed factors, site being nested within bay; and depth was set up as covariate (rounded by 10 m). PERMANOVA were set-up with 9999 permutations and built under unrestricted permutation of raw data model. After the model was created, estimates of variation (square root) can give an idea of the relative importance of different terms in the model. Estimates of components of variation are used as a basis for comparing the relative importance of different terms in the model towards explaining overall variation (Underwood and Petraitis 1993). Pair-wise test were run when a factor was significant using the same set-up. Afterwards, SIMPER procedures were also run to explore the taxa contributing to the group similarity or differences.

Total richness (i.e. total count of all taxa groups) was calculated using presence/absence of taxa at each station. Average richness was calculated for sites and bays but also by depth categories (by 10 m) and substrate type. Differences in taxa richness between bays, substrates and depths were evaluated using Kruskal-Wallis test with multiple comparisons testing when significant using the software R 3.3.1 (R core Team 2016).



**Figure 1:** Geographical regions of finfish aquaculture sites with baseline reports. Blue: Northern Arm, Red: Upper Bay d'Espoir, Yellow: Little Passage, Black: Upper Hermitage Bay, Purple: Little Bay (Great Bay de l'Eau), Green: Belle Bay West, Pink: Belle Bay East.

## RESULTS

### TOPOGRAPHY AND SUBSTRATE TYPE

A total of 752 stations were analyzed with depth ranging from 2 to 100 m. Each site had a different depth range with values from 34 m (Pomley Cove) to 97 m (Lou Cove). In average Grandy Cove was the deepest site (average  $\pm$  SD =  $77 \pm 16$  m) and Arran Cove ( $32 \pm 13$  m) the shallowest. Overall, the sites sampled in Belle Bay West (BBW) were the deepest ( $58 \pm 20$  m) while sites in Belle Bay East (BBE) were within the shallowest area ( $31 \pm 17$  m) (Table 1a,b).

All sites had mixed substrates: most substrate types were represented within a site and often at the sampling station level. On the 22 sites reported, 11 were dominated by fine substrates (Table 1a,b). SIMPER analysis revealed a few trends in substrate composition among and within bays. The proportion of dominant substrate varied within sites and the proportion of similarity between sampling stations ranged from 25 to 81%. Hardy Cove was the most homogeneous site in terms of substrate composition and Grip Cove the most heterogeneous. Similarly, dominant substrates varied considerably within geographic regions/bays with some regions having different dominant substrates in every site. Overall similarity percentages ranged from 29 to 79%, with Little Passage being the most heterogeneous area and Little Bay the most homogeneous with fine sediments as a dominant substrate (88.0% of stations); however, only one baseline report (i.e. 1 site) is available for Little Bay (Table 1a,b).

**Table 1a:** Overall site description of baseline data (substrate, depth, taxa) collected at 22 sites as well as SIMPER results (percentages of similarity).

	Northern Arm			Upper Bay d'Espoir					Little Passage				
Site Name	Sugarloaf	Wild	Kilbuck	Lou	Arran	Hardy	North West	Pomley	Deer	Blackfish	Seal Nest	Grip	Robin Hood
# of Stations	75	19	23	41	35	31	49	13	42	18	32	20	22
Depth (m) avg $\pm$ SD	45 $\pm$ 20	45 $\pm$ 20	51 $\pm$ 22	40 $\pm$ 31	32 $\pm$ 13	43 $\pm$ 27	43 $\pm$ 27	74 $\pm$ 11	41 $\pm$ 23	61 $\pm$ 17	46 $\pm$ 18	40 $\pm$ 20	54 $\pm$ 24
range	3 - 77	18 - 80	14 - 89	2 - 99	11 - 64	5 - 95	8 - 98	53 - 87	2 - 73	24 - 78	9 - 65	8 - 62	14 - 95
Richness avg $\pm$ SD	2.7 $\pm$ 1.1	2.7 $\pm$ 1.0	3.0 $\pm$ 0.9	2.3 $\pm$ 0.9	2.9 $\pm$ 1.0	2.6 $\pm$ 1.0	2.5 $\pm$ 1.1	2.4 $\pm$ 0.8	2.9 $\pm$ 0.9	2.8 $\pm$ 0.9	2.9 $\pm$ 1.1	3.4 $\pm$ 1.8	2.1 $\pm$ 0.9
range	0-6	1-5	1-4	0-4	1-5	1-5	0-5	1-4	1-4	1-4	0-5	1-7	1-3
avg regional $\pm$ SD	2.8 $\pm$ 1.1			2.5 $\pm$ 1.0					2.8 $\pm$ 1.2				
% stations = 0	1.3	0	0	3.4	0	0	6.1	0	0	0	3.1	0	0
Taxa similarity (%)	32.3	32.1	28.5	36.6	61.9	50.9	28.9	43.9	28.4	35.9	35.8	40.6	19.0
	29.8			34.1					26.1				
Dominant taxon (% similarity)	AN (69.8)	COR (46.9)	COR (45.6)	BR (78.8)	BR (73.0)	BR (63.1)	AN (41.1)	AN (55.8)	COR (51.7)	BR (47.3)	AN (39.6)	COR (48.2)	SP (24.7)
Dominant substrate (% presence in stations)	F (78%)	B (47%)	C (43%)	F (76%)	F (50%)	F (90%)	F (47%)	C (61%)	B (88%)	M (56%)	F (56%)	F (40%)	F (41%)
Substrate similarity (%)	35.4	29.2	28.9	61.1	39.2	81.5	32.1	41.0	78.0	35.3	44.3	24.7	28.6
	39.1			43.5					29.4				

AN: Anemones, COR: Coralline Algae, BR: Brittle Stars, SP: Sponges, POL: Polychaetes, K: Kelp, ALG: Algae

F: Fine, M: Medium, C: Coarse, B: Bedrock. avg: average

**Table 1b:** Overall site description of baseline data (substrate, depth, taxa) collected at 22 sites as well as SIMPER results (percentages of similarity).

	Upper Hermitage Bay		Little Bay	Belle Bay West				Belle Bay East	
Site Name	Green	Herring	Murphy Point	Grandy	Farmers	Chapel	Bottle	Little Burdock	Rencontre
# of Stations	15	25	78	17	34	31	32	54	54
Depth (m) avg $\pm$ SD	45 $\pm$ 29	33 $\pm$ 30	39 $\pm$ 13	77 $\pm$ 16	41 $\pm$ 7	68 $\pm$ 22	56 $\pm$ 13	36 $\pm$ 19	26 $\pm$ 13
range	3-80	3-93	9-62	50-100	30-65	35-100	30-80	5-64	3-42
Richness avg $\pm$ SD range	2.1 $\pm$ 1.9 0-5	2.8 $\pm$ 1.6 0-5	2.9 $\pm$ 1.0 1-6	1.8 $\pm$ 1.4 0-4	1.9 $\pm$ 0.9 0-4	1.7 $\pm$ 1.1 0-4	1.7 $\pm$ 1.0 0-4	2.1 $\pm$ 0.9 1-5	2.3 $\pm$ 0.9 1-5
avg regional $\pm$ SD	2.6 $\pm$ 1.7		2.9 $\pm$ 1.0	1.8 $\pm$ 1.1				2.3 $\pm$ 0.9	
% stations = 0	33.3	12.0	0	17.6	5.9	9.7	6.2	0	0
Taxa similarity (%)	14.1	34.8	31.5	30.8	27.7	34.2	41.5	42.3	31.4
	25.9		31.5	29.6				36.4	
Dominant taxon (% similarity)	AN (35.6) COR (47.7) COR (30.2)	COR (47.7)	POL (32.0) K (26.2)	AN (49.3)	ALG (63.4)	AN (69.8)	ALG (88.2)	ALG (66.5)	ALG (57.4)
Dominant substrate (% presence in stations)	M (47%)	C (40%)	F (88%)	F (65%)	M (58%)	B (51%)	M (75%)	F (43%)	F (48%)
Substrate similarity (%)	32.4	31.3	79.3	43.4	49.7	38.4	61.3	30.0	38.0
	32.2		79.3	34.5				34.0	

AN: Anemones, COR: Coralline Algae, BR: Brittle Stars, SP: Sponges, POL: Polychaetes, K: Kelp, ALG: Algae

F: Fine, M: Medium, C: Coarse, B: Bedrock. avg: average



## TAXA DISTRIBUTION

The most frequent taxa reported were anemones (39%), algae (28%), coralline algae (27%), brittle stars (26%), sea stars (14%) and kelp (14%) (Table 2). Percentages were calculated by dividing the number of occurrences of particular taxa by the total number of stations. Tunicates and sand dollars were rarely observed compared to the other organisms (2 to 3 sites). We noted that sea urchin presence was higher when site had experienced aquaculture (Arran Cove, Blackfish Cove, Seal Nest Cove or Hardy Cove). Mussels were more present at Blackfish Cove again when aquaculture had occurred. Shrimp was reported in only two sites (Sugarloaf Island and Lou Cove).

Restrictions with depth were apparent for kelp, mussels and fish with no occurrences found at depths higher than 75 m (Figure 2). Some taxa were more frequently reported in deeper habitats (>40 m) such as corals, anemones, sponges, brittle and feather stars whereas other occurred more often in shallower stations (i.e. photic zone) such as urchins, mussels, algae and coralline algae (Figure 2, Table 2).

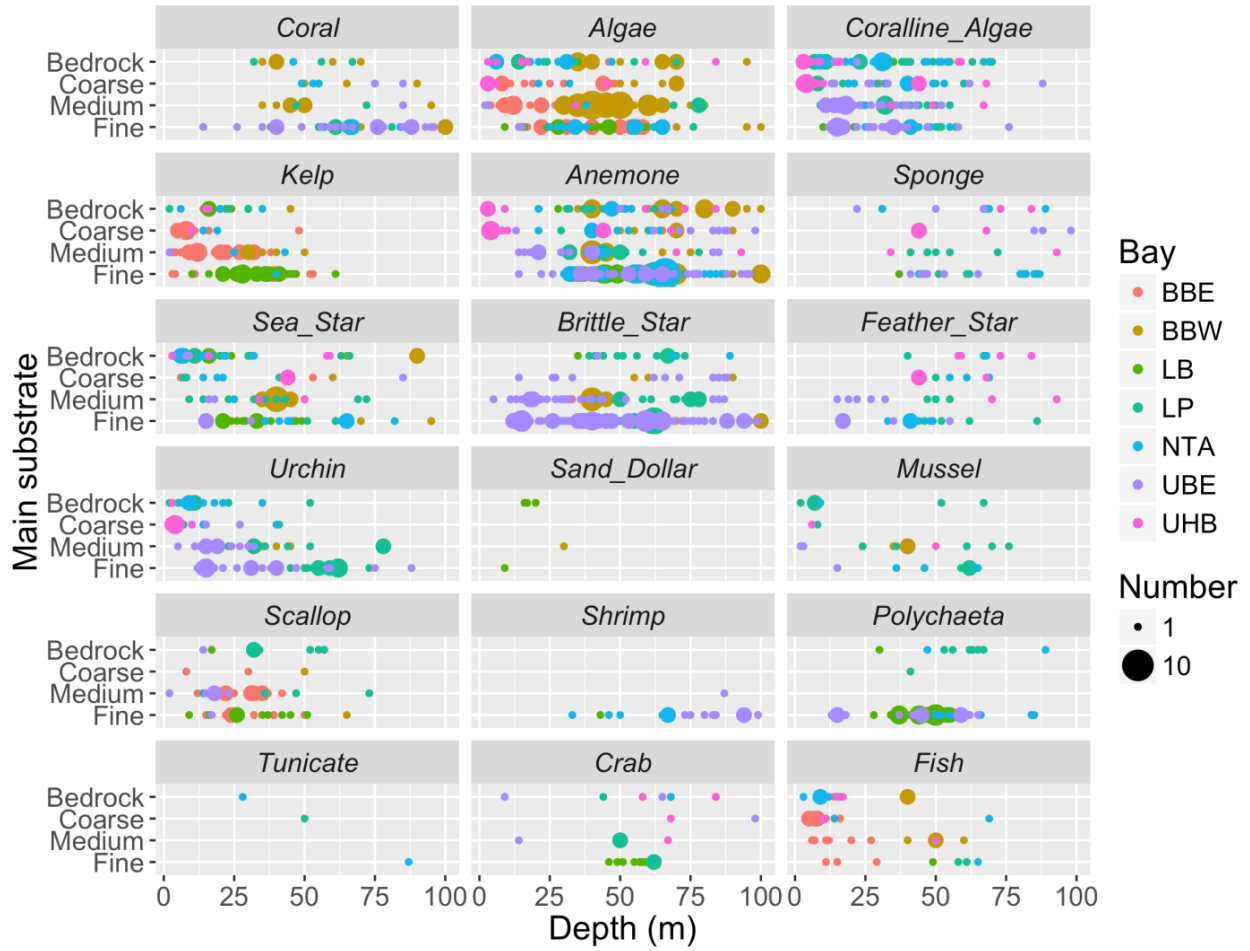
Restrictions with substrate can be observed for shrimp or sand dollar as they were not reported in stations with bedrock or coarse dominant substrates; similarly, no polychaetes or tunicates were observed on medium substrates (Figure 2).

Taxa distribution at the site scale was heterogeneous with percentages of similarity between stations in terms of taxa composition ranging from 14 to 61% and rarely exceeding 35%. Anemones were recorded as the dominant taxa at 32% of studied sites while coralline algae were the most prevalent taxa in 23% of the sites. When focussing on the dominant taxa only, similarity percentages within a site varied from 31 to 88%.

A total of 24 stations (4 bedrock, 1 coarse, 6 fine and 13 medium substrates) were found deprived of fauna or flora, with the highest proportions of barren stations in UHB and BBW (Table 1).

**Table 2:** Frequency of presence of each taxon according to depth ranges as reported in the 22 reports.

Depth (10 m range)	Corals	Algae	Coralline algae	Kelp	Anemones	Sponges	Sea stars	Brittle Stars	Feather stars	Urchins	Sand dollars	Mussels	Scallops	Shrimp	Polychaetes	Tunicates	Crabs	Fish
<b>0</b>	0	39	39	26	30	0	13	0	0	35	0	13	4	0	0	0	0	4
<b>10</b>	1	28	54	36	13	0	28	15	0	31	1	9	7	0	1	0	3	30
<b>20</b>	0	30	50	30	10	1	27	22	6	23	4	2	17	0	4	0	0	7
<b>30</b>	2	44	46	28	21	3	17	22	3	10	1	0	15	1	3	1	0	2
<b>40</b>	7	32	26	13	44	5	14	28	7	9	0	4	7	1	10	0	1	2
<b>50</b>	9	27	20	7	48	7	10	18	6	7	0	3	6	2	23	1	5	4
<b>60</b>	17	24	11	1	53	3	6	38	7	11	0	5	2	0	12	0	9	3
<b>70</b>	16	21	7	0	60	10	10	30	7	1	0	4	2	6	5	0	5	2
<b>80</b>	16	16	3	0	45	13	3	39	3	8	0	3	0	11	3	0	3	0
<b>90</b>	38	0	5	0	62	33	14	57	10	5	0	0	0	14	10	5	0	0
<b>100</b>	40	20	0	0	60	7	7	47	0	0	0	0	0	7	0	0	7	0



**Figure 2:** Taxa occurrence according to depth, substrate and geographical location.

## MULTIVARIATE ANALYSIS

No clear pattern was observed using nMDS (representation not shown, stress of representation >0.2) therefore no distinct assemblages could be determined based on presence/absence data. PERMANOVA analyses (Table 3) showed a significance effect of depth ( $p < 0.001$ ) on taxa distribution as well as site, bay, and substrate and the interaction depth x substrate ( $p < 0.05$ ).

**Table 3:** PERMANOVA results using 9999 permutations (perm) of taxa presence/absence using simple matching similarity matrix data testing bay, substrate and site with depth as covariate and some interactions. Site is nested within bay and both considered as fixed factors whereas substrate is designated as random.

Source	df	MS	Pseudo-F	P(perm)	Estimates of components of variation Sq.root
Depth	1	13500.0	87.669	0.0001	4.213
Bay	6	5363.4	33.252	0.0001	7.103
Site (Bay)	15	776.3	6.1387	0.0001	4.685
Substrate	3	516.3	4.7772	0.0001	1.900
Depth x Substrate	3	482.7	4.4667	0.0001	1.591
Residuals	723	108.1			10.396
Total	751				4.213

Significant differences in taxa assemblages were detected when comparing substrates (Table 4). However, differences were not explained by strict associations between a particular taxon and substrate type but by differences in frequency of dominant taxa. Taxa similarity analysis by substrates showed that medium substrates are similar in taxa at 18.5% with algae contributing to 39% of the similarity, bedrock at 23% with coralline algae contributing to 39%, fine at 21.7% with anemones contributing to 46% of the similarity and coarse at 25.2% with coralline algae contributing to 35% of the similarity. Detailed pairwise SIMPER comparisons are not presented in order to not overload the document. The main observations (i.e. taxa with % of contribution to dissimilarity higher than 10%) related to differences in dominant substrates are as follows:

- Bedrock and Medium substrates: differed mainly by a higher proportion of coralline algae and anemones on bedrock, and higher occurrence of algae on medium substrates.
- Bedrock and Fine substrates: higher proportion of anemones on fine sediment and higher occurrence of coralline algae on bedrock.

- Bedrock and Coarse substrates: both are dominated by coralline algae however more anemones were found over coarse substrates whereas sea stars were more often present over bedrock substrates.
- Coarse and Medium substrates: higher occurrence of algae in medium substrates and greater proportion of coralline algae and anemones in coarse substrates.
- Coarse and Fine substrates: more anemones in fine sediments and higher presence of coralline algae over coarse sediment.
- Medium and Fine substrates: higher proportion of anemones and brittle stars on fine substrates and more algae in medium sediments.

**Table 4:** Level of similarity within and between substrates and significance level (p-values) of taxa comparisons per substrate using pairwise PERMANOVA on substrate.

	<b>Bedrock</b>	<b>Coarse</b>	<b>Medium</b>	<b>Fine</b>
<b>Bedrock</b>	82.3			
<b>Coarse</b>	81.9 <sup>*</sup>	81.5		
<b>Medium</b>	82.3 <sup>*</sup>	81.8 <sup>***</sup>	83.4	
<b>Fine</b>	81.8 <sup>***</sup>	81.5 <sup>*</sup>	82.7 <sup>*</sup>	83.4

\*\*\*:p<0.001, \*:p<0.05

When comparing taxa occurrence in different bays, pairwise PERMANOVA comparisons (Table 5) showed that all bays differed from each other with the expectation of UHB with LB and NTA.

**Table 5:** Similarity between and within bays (%) and significance level (p-values) of comparisons of taxa collected in bays using pairwise PERMANOVA. BBE = Belle Bay East, BBW = Belle Bay West, LB = Little Bay, LP = Little Passage, NTA = Northern Arm, UBE = Upper Bay.

	BBW	BBE	UBE	LB	NTA	LP	UHB
BBW	88.3						
BBE	87.0 <sup>*</sup>	89.5					
UBE	83.2 <sup>***</sup>	80.9 <sup>***</sup>	85.9				
LB	82.8 <sup>*</sup>	83.6 <sup>***</sup>	79.0 <sup>***</sup>	82.6			
NTA	84.4 <sup>*</sup>	82.6 <sup>***</sup>	81.9 <sup>***</sup>	80.5 <sup>***</sup>	84.0		
LP	82.2 <sup>***</sup>	80.1 <sup>***</sup>	83.0 <sup>***</sup>	78.3 <sup>**</sup>	81.8 <sup>***</sup>	81.5	
UHB	82.0 <sup>***</sup>	81.1 <sup>***</sup>	80.0 <sup>***</sup>	78.3 <sup>ns</sup>	82.5 <sup>ns</sup>	80.4 <sup>***</sup>	81.9

\*\*\*:p<0.001, \*\*: p<0.01, \*: p<0.05, ns: non-significant

Overall, SIMPER analyses of similarity (results not shown) revealed that the dominant taxa (Table 1) are the main contributors in determining differences between bays; only kelp which is not a dominant group appeared to be also contributing to differences in benthic assemblages in LB (26%) and BBE (24%).

## **RICHNESS**

Taxa richness ranged between 0 and 7 in our dataset (Table 1a,b, Figure 3).

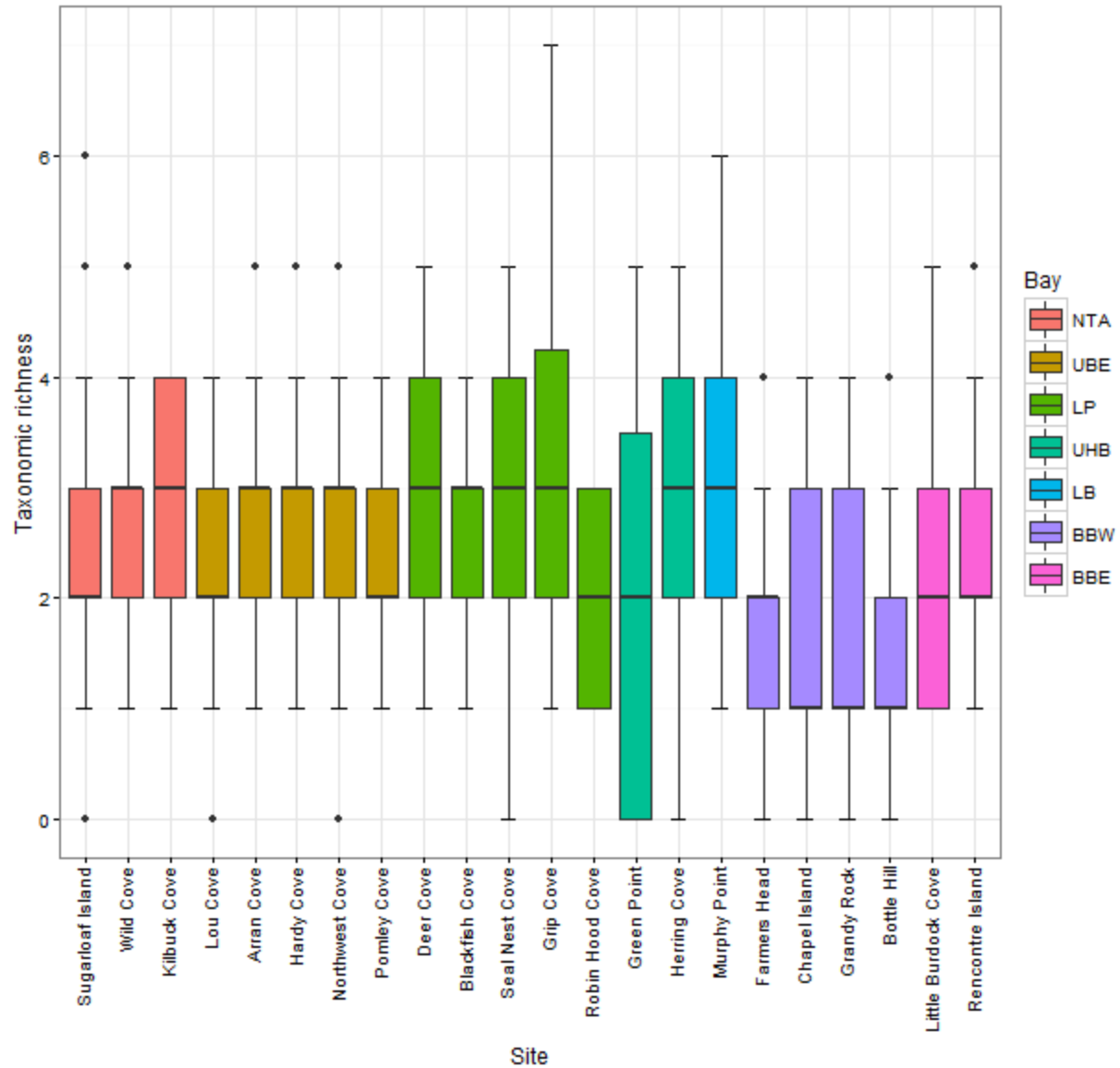
Taxa richness was significantly different between bays (Kruskal-Wallis chi-squared = 71.893,  $df = 6$ ,  $p$ -value < 0.0001) and sites (Kruskal-Wallis chi-squared = 91.969,  $df = 21$ ,  $p$ -value < 0.0001). BBW and BBE had the lowest average richness. BBE was significantly less rich than LB, LP and NTA; BBW was significantly less rich than all the other bays with the exception of BBE (Table 1a,b).

Kruskal-Wallis test on ranks showed that depth had a significant effect on richness (Kruskal-Wallis chi-squared = 52.424,  $df = 10$ ,  $p$ -value < 0.0001) but without any particular depth patterns (Table 6).

Substrate type did not influence richness when all data were considered together (Kruskal-Wallis chi-squared = 4.267,  $df = 3$ ,  $p$ -value = 0.234). On the other hand, substrate types influenced richness when bay data were considered separately with statistically significant differences between substrates in UHB (Kruskal-Wallis chi-squared = 15.057,  $df = 2$ ,  $p$ -value < 0.001), BBE (Kruskal-Wallis chi-squared = 11.887,  $df = 3$ ,  $p$ -value = 0.008), and LB (Mann Whitney U statistics (only two types of substrates),  $p$ -value = 0.018).

**Table 6:** Taxonomic richness (mean  $\pm$  SD) for each depth range (10 m) for all geographic regions combined.

<b>Depth range</b>	<b>Richness (mean <math>\pm</math> SD)</b>
<b>0</b>	2.52 $\pm$ 1.31
<b>10</b>	2.99 $\pm$ 0.96
<b>20</b>	2.83 $\pm$ 1.06
<b>30</b>	2.76 $\pm$ 1.06
<b>40</b>	2.58 $\pm$ 1.19
<b>50</b>	2.42 $\pm$ 1.11
<b>60</b>	2.33 $\pm$ 1.12
<b>70</b>	2.19 $\pm$ 1.09
<b>80</b>	1.79 $\pm$ 1.21
<b>90</b>	2.81 $\pm$ 0.93
<b>100</b>	1.93 $\pm$ 1.22



**Figure 3:** Box and whisker plot of taxa richness per site. The thick line within boxes represents the median, while the upper and lower edges of the box correspond to the 25<sup>th</sup> and 75<sup>th</sup> percentiles. The lower and upper whiskers extend to lowest data points within 1.5 inter-quartile ranges. Outliers are represented by dots.

## DISCUSSION

### DATA LIMITATIONS AND RECOMMENDATIONS

The data analyzed in this study were extracted from reports completed prior to the establishment of detailed protocols as part of the AAR in 2015. Data are inconsistent, leading us to summarize the information and reducing the power of the analysis. Even when the video methodology is similar, data collected between 2003 and 2011 differed in terms of sampling times, quality of camera used, sampling gear, operators, and lighting conditions. Another constraint in the data analysis is the referencing of baseline data for sites that have already experienced aquaculture (20% of the sites analyzed). In term of characterizing larger bay areas, a limitation also worth noting is that the number of sites differed among bays; for example, in the Little Bay region only one study site was investigated.

The following points detail data limitations and represent recommendations for future surveys:

- Reports did not provide information on the surface area surveyed per station. Sampling surface was not standardized in term of transect lengths or distances between the camera and the seafloor. Each video recording lasted at least 1 min but boat speed is not provided, therefore the estimation of the benthic area covered was not possible. The benthic area covered during each recording is usually estimated based on the product of quadrat width, recording duration, and boat speed (Hamoutene et al. 2015). Lack of information on surface covered (in addition to the absence of abundances) dictated the usage of presence/absence data.
- Reports did not contain species or genus information but only high level taxonomic identification. While grouping unclassified taxa to the next highest taxonomic level has the potential to be misleading (Borja et al. 2008; Keeley et al. 2012), this was the only approach available to us. Other studies focusing on environmental impacts of aquaculture have also used similar high taxonomic level identifications to record epibenthic organisms (Crawford et al. 2001). A study on the effect of bottom fishing on benthic ecosystems has revealed that many differences in macrofaunal community structure between sediments inside and outside protected areas were still evident at higher levels in the taxonomic hierarchy (Barrio Froján et al. 2012). However, there is value in ensuring the lowest level of identification possible is attained especially when characterizing reference communities.
- Video description tables did not include abundances but only a description of taxa observed in video segments. Considering the absence of counts we could only calculate richness thus limiting our ability to properly characterize communities (especially for a before/after comparison). Species richness is a relatively insensitive



community attribute that considers all species to be equal and so treats rare and common species similarly (Magurran 2004). It is of importance to collect counts/abundance (or percentage cover) of fauna and flora in order to better evaluate epibiotic richness (Hamoutene et al. 2014; DFO 2014) and overall community description.

- There was a lack of precision in substrate type determination in terms of the terminology used in reports. One of the main sources of observer error reported by Mabrouk et al. (2014) resided in the distinction between substrate classes. This suggests the necessity of limiting the categories in order to get a more consistent evaluation by observers. Possible categories of substrate classification could be modified from previous classification schemes (Mabrouk et al. 2014). These classes would include bedrock (continuous solid rock), coarse (boulders greater than 250 mm and rubble ranging from 130-250 mm), medium (cobble ranging from 30-130 mm, and gravel ranging from 2-30 mm), and fines (sands and mud/silt/clay grouping less than 2 mm) as modified from the Wentworth scale (Wentworth 1922).
- We have used data collected in reports to give a snapshot of benthic communities at the bay scale. However, it is important to note that these data were not collected for that purpose. Care is needed in the identification of appropriate spatial scales for sampling before conclusions can be reached about differences in organisms from one area to another (Morrissey et al. 1992). Sampling designs will need to be adapted accordingly for a bay management purpose in terms of benthic communities' sensitivity/vulnerability.
- Previous monitoring campaigns did not include sampling at depths greater than 100 m by design which limited access to data in deeper areas. This limitation is not supported by the AAR standards and will not be an issue in future sampling campaigns.

## **SITE CHARACTERISTICS**

All lease areas referenced in this analysis covered a high range of depths (from 2 to 100 m), substrates and slopes. Overall, the sites sampled in BBW were the deepest ( $58 \pm 20$  m) while sites in BBE were within the shallowest area ( $31 \pm 17$  m). All sites encompassed most substrate types but proportions of each substrate varied among and between sites. Interestingly most of the sites and bays included fine sediments. However, attempts to complete grabs in many of those areas had low success suggesting that only a thin layer of sediment is deposited. SIMPER analyses highlight the substrate patchiness observed at the station (i.e. sampling point), site and bay scales. This patchiness can be quantified by the proportion of similarity between sampling stations ranging from 25 to 81% for sites and from 29 to 79% within bays. These values underscore the challenges of surveying the environment of the South

Coast of NL, with its large variability in bathymetry and presence of rock walls and boulders, as well as the patchiness of its habitat and substrate (Hamoutene et al. 2015). Little Passage was found to be the most heterogeneous area and Little Bay the most homogeneous dominated by 88.0% fine sediments. However, there is only one baseline report considered for Little Bay. The spatial heterogeneity observed in areas such as Little Passage can also be the result of the presence of specific groups of organisms. For example, some sessile suspension feeders such as sponges (dominant taxa in Robin Hood Cove site in LP) can exert a major influence on species biodiversity by locally increasing habitat heterogeneity (Buhl-Mortensen et al. 2010). We could hypothesize that this heterogeneity might represent a factor of vulnerability of associated fauna and flora in terms of potential recovery and an added difficulty in the monitoring of anthropogenic impacts.

## TAXA DESCRIPTION AND RICHNESS

Organisms were identified at high taxonomic levels and were generally sparse and patchily distributed similarly to other surveys completed on the South Coast of the island (e.g. Hamoutene et al. 2015). We could not identify obvious benthic assemblages as per the nMDS representation though the absence of abundance data might have precluded us from doing so. The dominant groups reported are: anemones, coralline algae, brittle stars, and other algae sometimes described as seaweeds, *Laminaria* sp., *Ulva* sp. or *Ptilota* sp. Depressions and gentle slopes in the deeper portions of subarctic rocky bottoms often collect mixed bioclastic and siliciclastic sediments, including a dense cover of coralline algal buildups (*Lithothamnium glaciale* and *Lithothamnium tophiiforme*) (Adey et al. 2015). In shallower waters (max. occurrence at 8 m) another crustose coralline algae *Clathromorphum compactum* has been observed in NL attaining a thickness of up to 3 cm and a life span of ~100 years (Gamboa et al. 2010). Rhodoliths (generic name for certain coralline algae (Rhodophyta, Corallinaceae)) can form extensive beds or aggregations overlying calcareous sediments produced by the accumulation of eroding rhodolith fragments. These beds typically occur in environments where water motion (waves and currents) or bioturbation are strong enough to move individual rhodoliths within beds, thereby preventing burial by sediments or overgrowth by other organisms, but not so high as to cause their destruction (Kenchington 2014). Rhodolith beds score high in their uniqueness and vulnerability as per the Ecological and Biological Significant Areas (EBSAs) criteria despite being classified as low productivity areas (DFO 2011; Kenchington 2014). Some other sensitive habitats were also observed such as kelps mostly present in Little Bay and sponges in Robin Hood Cove site (Little Passage). Sponge grounds may, have functions similar to those of coral reefs (Løkkeborg and Fosså, 2011), providing justification for conservation for fisheries habitat in accordance with the Biodiversity Convention.

The taxa richness revealed by the data provided in the reports was especially low, as already documented in the South Coast of NL (Hamoutene et al. 2014, 2015). This low taxa richness is partly due to the high taxonomic level identifications used in the baseline reports. The highest values were recorded in Little Passage and Little Bay and the lowest value in Belle Bay (BBW and BBE). Local environment and physical conditions likely drive those differences as further discussed below.

### ***Substrate effect***

Substrate appeared in the multivariate analysis to be a factor affecting taxa assemblages. Differences in benthic communities between substrates were due to differences in frequency of the main taxa and not types of assemblages. The substrate effect was the lowest in term of variance component and the interaction depth x substrate was found significant suggesting a combination of both factors in determining taxa distribution. It was also noted that substrate did not seem to influence taxa richness when all data were grouped for univariate analyses. On the other hand, when bays were considered separately, significant differences were observed between substrates (BBE, UHB, LB). These results are similar to Hamoutene et al. (2015) where richness was influenced by substrate; though no particular ranking in richness values associated with a substrate type could be detected. We could note some substrate preferences with coralline algae being mostly associated with bedrock and coarse substrates, algae mostly observed on medium substrates while brittle stars and anemones were reported mostly on fine substrates (and coarse as well for anemones). Only a few taxa seemed restricted to one type of substrate such as shrimp, and polychaetes with clear higher frequency on fine substrates. However, some polychaetes were reported in areas with bedrock; these might have been observed between boulders or represent another type of worms such as serpulids that develop on hard bottom substrates.

### ***Depth effect***

Taxa distribution and richness also showed a significant effect of depth but comparisons did not reveal a clear vertical pattern. Kelp, sponges and some motile taxa such as fish and shrimp did appear restricted by depth. Kelp (or likely kelp debris described as kelp) was not reported deeper than 75 m and sponges were reported only below 25 m. Shrimp were only present at depths >30 m while fish were observed only at less than 75 m. However, observations of more motile species (fish or shrimp in comparison with echinoderms or anemones) have to be taken with caution considering that reports and surveys are not detailed enough to provide an accurate description of depth distributions. Differences in fish behaviour according to species may also exist. Depth and substrate differences were also observed in Hamoutene et al. (2015; 2016). Depth and substrate type influenced community structure; overall, total abundances and taxon richness both increased (albeit slightly) with depth, while substrate type mostly

influenced taxa richness with almost no effect on abundances (Hamoutene et al. 2015). Depth had the strongest effect on assemblage structures partly explained by the dependence of algae and coralline algae *Lithothamnium* sp. on light as well as the fact that other organisms (e.g. sea stars, anemones) appeared to prefer greater depths (Hamoutene et al. 2015). The absence of a defined vertical depth zonation in this report might be due to the overall low data quality and the fact that data were grouped, therefore not allowing the identification of specific depth distributions related to a bay and/or site. It is worth noting that Haedrich and Gagnon (1991) found a similar lack of obvious vertical zonation in the Bay d'Espoir Fjord system and concluded that vertical distribution might be more governed by available substrates. The significant interaction between depth and substrate as highlighted in the previous section confirms this finding though it is worth noting that relatively to the site and bay effect the depth and substrate influence was lower.

### ***Effect of sites/bays***

In addition to an effect of substrate and depth, PERMANOVA analyses (Tables 3 and 4) showed a significant effect of sites and bays on taxa distributions. This geographical effect was found to represent the strongest influence on the data analyzed. The significant difference in assemblages at the site level can be explained by the specific topography, exposure and substrate of each site dictating the presence of taxa. The significant differences observed between bays were mostly driven by differences in frequency of the main reported taxa. Strict comparisons of diversity measurements from different regions of variable size and/or habitat may be hazardous (Magurran 2004). However, we can note a higher occurrence of impoverished stations at sites in BBW and UHB, in comparison with other regions. Hamoutene et al. (2015) observed a high proportion of bare stations at fallow sites. They postulated the possibility that these stations might be bare prior to any aquaculture development and naturally-occurring hypoxic or anoxic conditions could exist before the commencement of production. Bare stations were mostly found over fine and medium substrates. On fine substrates, video monitoring may have failed to detect organisms that could be living within the substrate and/or camera resolution may have affected the ability to observe and report all organisms. Moreover, some of the sites have experienced production when sampled and could drive some of the differences observed between bays and sites, as for instance mussels and urchins appeared more frequently at those sites.

## CONCLUSION

In addition to the recommendations for baseline data collection listed in the first section of the discussion, we can summarize our findings in the following points:

- Baseline surveys showed that the targeted sites and bays have a high level of patchiness as well as some structure-forming benthic biogenic habitats that will need to be considered in terms of vulnerability (i.e. conservation) and adapted monitoring approaches and sampling designs.
- Taxa richness detected by video monitoring is low including areas with barren stations suggesting low productivity habitats.
- A significant influence of geographical locations was observed on assemblages. This effect was found to be stronger than the depth and substrate effect though detailed analyses of abundances and a better spatial coverage of data collection might result in a different hierarchy of factors.
- The presence of barren stations prior to aquaculture has to be taken into account when assessing change through AAR reports completed at peak biomass and highlight the necessity of performing baseline surveys before installation of aquaculture.
- The baseline assessment guidelines are not yet amended to reflect the legislative and regulatory requirements described in the AAR. This is a de facto deficiency in the baseline assessments design. Baseline surveys were undertaken without the benefit of predictions (part of the current AAR regime) of where BOD deposits would occur. However, this will require a regional validation of predictive tools to better guide baseline assessments and subsequent effect monitoring.
- Consideration should be given to bay approaches in documenting changes, advising on licensing authorizations, and eventually include an understanding of potential cumulative effects.
- Finally, a more thorough analysis of various habitats on the South Coast of NL is necessary to determine the composition of the seafloor and its natural assemblages, in terms of taxa species, abundance and richness using standardized techniques.

## ACKNOWLEDGEMENTS

This work was supported by the Program for Aquaculture Regulatory Research (PARR).

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