Fisheries New Zealand

Tini a Tangaroa

Candidate species selection for the development of environmental health measures for open ocean salmon aquaculture

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While open ocean aquaculture (OOA) is expected to offer benefits in environmental and species health over current inshore finfish farming methods, environmental effects are still likely to occur. Proposed open ocean farms sited in well-flushed locations potentially have a good capacity to disperse and dilute farm wastes. As a result, farm-derived organic enrichment at these sites is likely to be more diffuse but spread over a larger area, so will potentially cause far-field environmental effects. As industry expands into open ocean locations, farms are more likely to interact with mixed- and hard-bottom habitats that may support functionally different communities from those present in sheltered areas. While effects of organic enrichment on soft-sediment infauna are well studied, little is known about how OOA-derived impacts will manifest in previously unencountered open ocean environments.

Fisheries New Zealand has a multi-stage research project with the overall aim to understand how organic enrichment at levels anticipated for OOA impacts benthic species. The project will use key epifaunal species as case studies to describe potential impacts and identify possible indicators of environmental health in open ocean environments. This review aims to identify those key species and is the first of several project objectives to accomplish the overall aim.

Summary

To identify key epifaunal taxa likely to occur in environments that may be affected by OOA, the authors considered:

- A review of information from seabed effects assessments for proposed OOA finfish farms in New Zealand and reporting for dispersive salmon farms in Tory Channel.
- Species identified from seabed footage at proposed OOA and existing highly dispersive farm sites in New Zealand.
- A review of existing literature about the response of these species to organic enrichment.
- The ecological importance of species in the areas of interest and the ecosystem services they provide.
- The cultural importance of species (kaimoana and / or taonga species), or those that support species of importance (mahinga kai).

Candidate species choice:

- Using this information, we developed a framework to guide the systematic identification of key species appropriate for use in Objective 2 laboratory experiments. Candidate taxa were identified using a matrix that assigned a numerical score based on the framework categories. From the narrowed suite of potential candidates, logistical constraints ultimately influenced final taxa selection (for example, if the taxon is present in the field in sufficient numbers).
- Taxa chosen for use in future laboratory experiments include horse mussels, bryozoans, scallops, and erect sponges (species to be determined). Brachiopods will be used as a contingency species.
- Taxa chosen for use in field-based experiments include sea cucumbers, (opportunistic field sampling), bryozoans, brachiopods (field translocation experiments), erect sponges, horse mussels, and scallops (opportunistic or translocation depending on abundance across depositional gradient).

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1. INTRODUCTION

Finfish aquaculture in New Zealand is traditionally located within inshore coastal areas. Fisheries New Zealand has identified development of the open ocean[2] finfish aquaculture industry as a key priority for industry expansion (Giles et al. 2021), with the aim of reaching a goal of NZ$3 billion in annual sales by 2035[3]. While farming in open ocean locations is expected to result in a reduced magnitude of farm-derived environmental impact (albeit on a larger spatial scale), there are few examples of full-scale commercial open ocean aquaculture (OOA) operations (none of which are in New Zealand), and the environmental effects of these operations have not been well documented (Keeley et al. 2019a, 2020, Giles et al. 2021, Bennett et al. 2022, Fletcher et al. 2022). The tolerance of open ocean habitats to finfish farming-derived changes, such as organic enrichment, is also poorly understood. As such, little is known about how impacts from OOA will manifest in previously unstudied open ocean environments.

1.1 Project scope

Fisheries New Zealand has engaged the Cawthron Institute (Cawthron) in a multi-stage research project with the overall aim of understanding how organic enrichment at levels anticipated for OOA impacts benthic species. The project will use key species as case studies to describe potential impacts and identify possible indicators of environmental health in open ocean environments. This review aims to identify those key species and is the first (Objective 1) of several project objectives to accomplish the overall aim. We have focused on epifaunal organisms in this review, as the response of infauna to organic enrichment is relatively well understood and tools already exist in New Zealand to assess impact of aquaculture in soft-sediment environments (Keeley 2012).

To identify key species likely to occur in environments that may be affected by OOA, we made a list of epifaunal taxa present at proposed OOA and existing dispersive farm sites in New Zealand. Using this list, we considered the ecological importance of these species and the ecosystem services they provide, and also reviewed what is known about their responses to organic enrichment and sedimentation. We also considered whether the taxa are of cultural importance (kaimoana and/or taonga species), or support species of importance (mahinga kai). Using this information, we developed a framework to systematically identify key species appropriate for use in Objective 2, which will entail a series of laboratory and field-based experiments with the aim of identifying responses and tolerances of the candidate species to organic enrichment. In the third and final objective (Objective 3), the results from Objective 2 will be discussed in the context of addressing current information gaps related to OOA and assessing the potential magnitude and effect of organic enrichment from OOA on ecosystem health.

To satisfy the aims for Objective 1, this document includes:
- a review of information from seabed effects assessments for proposed OOA finfish farms in New Zealand[4] and reporting for dispersive salmon farms in Tory Channel, as well as a catalogue of epifauna identified from these locations;
- a literature review surrounding stress indicators[5] and known physiological response to enrichment, sedimentation, and/or general environmental change for species or related taxa identified from proposed OOA areas;
- a review of species important from a mātauranga Māori perspective;

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[2] Open ocean aquaculture (OOA) is defined by Fisheries New Zealand as "aquaculture outside of semi-enclosed bays and harbours or other sheltered locations around mainland New Zealand and larger offshore islands" (Giles et al. 2021). There is no established standard definition for OOA in the literature, and alternative terms such as 'exposed' and 'offshore aquaculture' are used interchangeably (Holmer 2010).

[3] The New Zealand Government Aquaculture Strategy: https://www.mpi.govt.nz/fishing-aquaculture/aquaculture-fish-and-shellfish-farming/aquaculture-strategy-for-new-zealand

[4] Blue Endeavour, New Zealand King Salmon; Hananui, Ngāi Tahu Seafood Resources; Project South, Sanford.

[5] A review of potential indicators of stress and/or physiological change in epifauna is provided in Appendix 3.

- provision of a framework for identifying the most appropriate candidate species for use in Objective 2 laboratory experiments;
- a list of possible stress indicators appropriate for the chosen candidate species.

This report focuses on the expected seabed impacts arising from OOA production; for information related to the likely impacts associated with initial site development and presence of farm structures, we refer the reader to Elvines et al. (2021) and Bennett et al. (2022). For expected effects on the water column, see Campos et al. (2019) and Newcombe et al. (2020).

1.2 Benthic impacts of organic enrichment from salmon aquaculture

Finfish farming in New Zealand is focused primarily on king (or Chinook) salmon (*Oncorhynchus tshawytscha*). The ecological effects of salmon farming in inshore environments are generally well known (Keeley et al. 2013). The primary seabed impact from salmon farm operations is the addition of farm-derived organic material, which primarily comprises faecal waste (Reid et al. 2009. Carvajalino-Fernandez et al. 2020), and, to a lesser extent, uneaten feed pellets (Schumann & Brinker 2020). The severity of organic enrichment is driven by both characteristics of the farm[6] and the physical characteristics of the receiving environment where the farm is sited.

Deep, well-flushed locations (hereafter 'dispersive') have increased dispersal and dilution of farm-derived organic waste compared with shallower sites with lower current speeds. As a result, the organic enrichment beneath the farm pens at dispersive sites is expected to be more diffuse than at non-dispersive sites but is likely to be spread over a larger area of seabed (Keeley 2020, Giles et al. 2021). This difference is due to the increased current transport of fine particles and flocculent material into the far field from the primary depositional footprint, as well as increased particle resuspension from the seabed (Keeley et al. 2019b). Sediment texture also has a large role in resuspension and dispersion of farm wastes (Law 2019); sandy substrates are less sticky and more mobile than finer, cohesive sediments (Law et al. 2016). Seabed effects typically manifest beneath the pens and decrease with distance from the farm. Open ocean farm sites will likely be sited in deep water (greater than 30 m) with strong current velocities (more than 15 cm per second), characteristics which influence the flushing potential of the site.

Sediment properties

Deposition and subsequent microbial decay of farm waste can significantly alter sediment chemistry, resulting in elevated total free sulphides[7], reduced redox potential[8], and depleted oxygen levels. In some instances, mat-forming sulphide oxidising bacteria can occur in conjunction with elevated sulphide levels (Preisler et al. 2007). Modifications to sediment chemistry are typically followed by predictable changes to infaunal communities (see below).

Accumulation of organic material may also result in increased dissolved nutrients such as nitrogen and phosphorus in sediments. As a result, algal growth may increase, although this is unlikely in deep locations. Excessive waste deposition on the seabed can also result in anoxic conditions in the overlying water, which may cause oxygen stress to epifaunal communities. The high flushing capacity expected in locations targeted for OOA is likely to reduce organic matter accumulation and increase sediment oxygenation when compared with sheltered, inshore farming sites (Findlay & Watling 1997).

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[6] For instance, the type of pen structures utilised, fish stocking density, type of feed and feeding system, settling velocity of fish faeces, and severity of water flow reduction caused by the farm structures (Keeley & Taylor 2011).

[7] An indication of the chemical oxidative state of the sediments and potential toxicity to organisms on the seabed. Increased concentrations are indicative of possible enrichment.

[8] A measurement of the oxic status or oxygen content of sediments; a decrease in these values is an indicator of possible enrichment.

Biological communities

The response of infaunal organisms[9] to organic enrichment has been well described at dispersive sites in the Marlborough Sounds (Keeley et al. 2013). The composition of these communities, and relative abundance of individuals within these communities, are commonly used as indicators of farm effects due to farms typically being sited over soft-sediment habitats (Keeley et al. 2013). Infaunal communities follow a largely predictable succession in response to organic enrichment at these sites (described by Pearson & Rosenberg 1978). As industry seeks expansion to open ocean locations, farms are more likely to interact with proximal mixed and hard-bottom habitats (Elvines et al. 2021, Bennett et al. 2022) instead of soft-sediment environments typical of inshore farm areas. Infaunal communities in OOA locations may be functionally different than those present in inshore farm locations[10], or may be present in very low numbers on hard substrate due to the presence of only relatively shallow layers of soft sediment. The use of infaunal community assessments as indicators of ecosystem health may therefore need to be redefined on a site-specific basis.

The tolerance of large epibiota to organic waste is poorly documented (Keeley & Taylor 2011) as these are typically site- and species-specific. Trophic subsidies provided by fish farms to these species may have positive effects, such as increased growth/body condition through an increase in food supply (George & Parrish 2015, Bergvik et al. 2019); or sub-lethal effects, such as reduced growth or reproductive output (White et al. 2016, Sardenne et al. 2020) through a reduction in seston/food quality possibly leading to an eventual reduction in densities. Increased turbidity may reduce light levels (Gonzalez 1996, Igwegbe et al. 2019), and this could compromise photosynthesis in plants such as seagrass and kelp and other seaweeds, along with maerl and rhodoliths that may form biogenic habitats. Increased sedimentation from farm-derived particulates smothers some organisms, potentially displacing mobile epifauna and impairing feeding efficiency in sessile organisms. Greater sediment deposition could also reduce settlement and survival of some key species through to their adult forms (see Section 3 for further detail), potentially having ecosystem effects.

It is significant to note that for the past decade, rocky reef community monitoring has been undertaken adjacent to salmon farms sited in dispersive environments in the Marlborough Sounds. These habitats are known to contain taxa sensitive to disturbance (MacDiarmid et al. 2013); however, results of this monitoring to date show there have been no discernible changes attributable to the farms with respect to abundance for individual groups or taxa (see Dunmore 2020). However, biodiversity surveys may have a limited to ability to detect sub-lethal effects and more subtle changes (Dunlop et al. 2021). Such changes may be revealed through biochemical, physiological, or molecular changes.

2. SPECIES PRESENT AT PROPOSED OPEN OCEAN AQUACULTURE SITES AND EXISTING HIGHLY DISPERSIVE SITES

2.1 Methods

A list of epifauna observed at the Blue Endeavour, Hananui, and Project South (Southland) farm applications were collated (Figure 1, Table 1). Data were derived from the video assessments completed for each site, where drop-camera or ROV footage captured the benthic habitat at locations across the potential farm areas. Epifauna were subsequently identified to the lowest taxonomic level possible. In some instances, it was not possible to identify an organism to species level, whether due to the nature of the animal (some taxa are difficult to identify without molecular tools) or due to video quality (for example, poor water visibility, very high current creating difficulties in capturing a video still).

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[9] Those living within the sediments.
[10] Though see Elvines et al. (2019), who reported that infaunal communities in soft-sediment habitats at the proposed Blue Endeavour farm location were largely comparable with those in highly dispersive sites in Tory Channel.

Although the Tory Channel farms are in sheltered, inshore environments, the hydrodynamic conditions in this area are highly dispersive. The infaunal communities are also functionally and taxonomically similar to the Blue Endeavour farm site and they share relatively comparable community metrics. Therefore, epifaunal community data from soft-sediment habitats and rocky reefs next to the Tory Channel farms were also reviewed.

Key habitat types present at proposed OOA sites and existing Tory Channel farm sites are described in the relevant sub-sections below. This information provides an overview of the benthic communities present at each site and the relative importance of taxa across habitat types and the areas of interest.
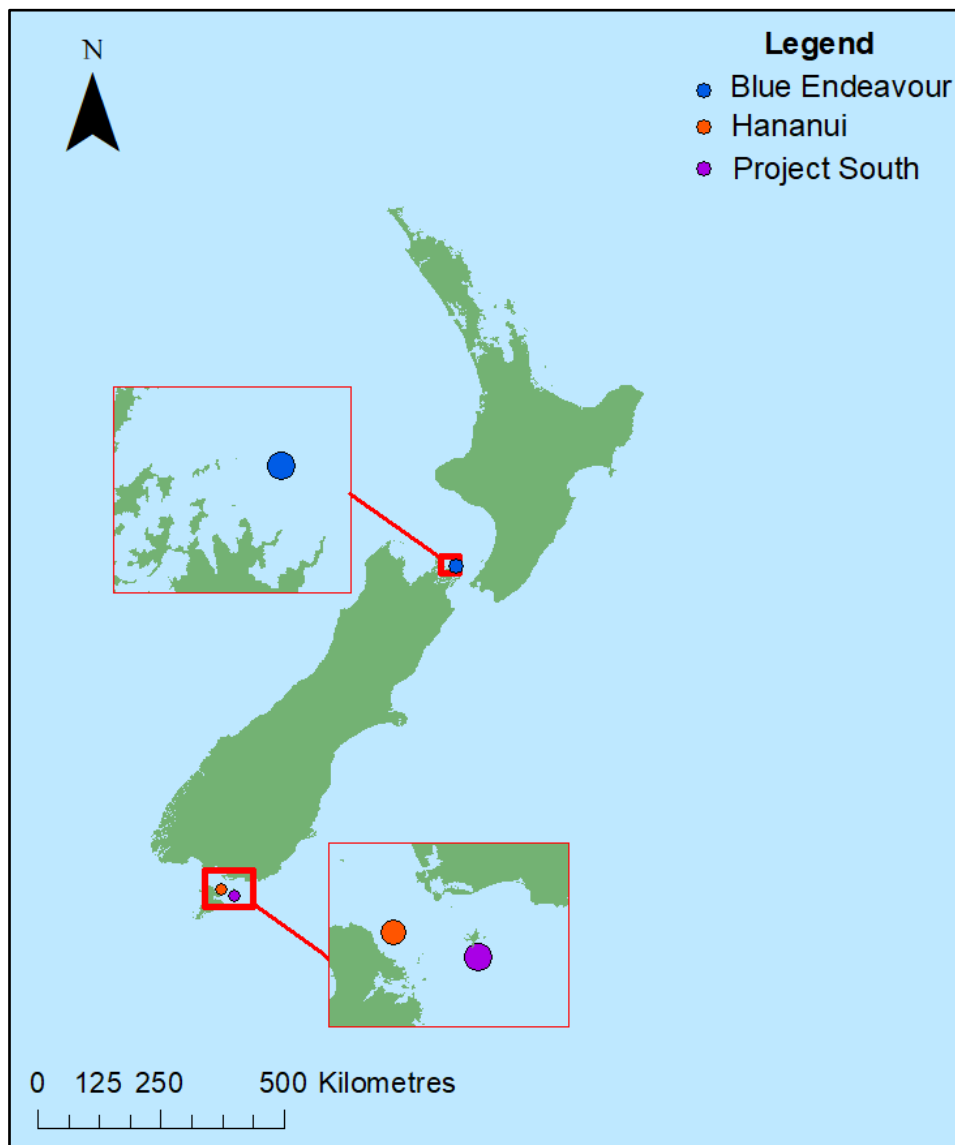
Figure 1: **Locations of the Blue Endeavour, Hananui, and Project South proposed sites for open ocean aquaculture in New Zealand.**

2.2   Blue Endeavour

Blue Endeavour was a consent application lodged by New Zealand King Salmon Ltd to develop an offshore salmon farm northeast of the Marlborough Sounds, due north of Cape Lambert and east of the Chetwode Islands (Figure 1). Consent for development of the Blue Endeavour site for OOA was granted in November 2022, but has subsequently been appealed. While collection of seabed footage at the Blue

Endeavour site is challenging due to water depth and current flow, multiple benthic surveys have been undertaken in an effort to map habitats in the area and characterise the ecological importance of the communities present (Elvines et al. 2019, Elvines et al. 2021). Habitat types described for the Blue Endeavour site include low-density epifauna—soft sediment (LDE-SS) and low-density epifauna—coarse sediment (LDE-CS), horse mussel brachiopod bed (HMBB), patch reef (PR), clump reef (CR), and mixed biogenic habitats (MB). Depths within the vicinity of the Blue Endeavour site range from 60 m to 110 m, and mid-depth current speeds are 40 cm/s (Newcombe et al. 2020).

LDE-SS comprised large (10s of metres) stretches where epifauna was sparse (50%[11] bare: 20–80%). Here, the seafloor had reasonably homogenous, muddy sediments with burrow holes and few to no epifauna visible. The most prevalent species seen in the sparsely populated areas were unidentified tube-dwelling anemones. Occasionally, small clusters of shell debris with encrusting organisms were seen, as well as a fine green algal-like growth.

Within LDE-CS habitat, small bryozoans were the prevalent epifauna (Elvines et al. 2019). The bryozoans seen were of various forms, but flexible branching (most likely Candidae) and bushy forms (likely Catenicellidae) dominated. All were soft bryozoans, as opposed to rigid, frame-building forms. Generally, this habitat comprised sandy sediments that appeared reasonably hard and consolidated. Where larger shell debris was more prevalent, there was typically more diverse and abundant epifauna, including increased soft bryozoan coverage. Soft bryozoan abundances had an estimated average of 2% coverage throughout this habitat type. In addition to bryozoans, encrusting taxa in these communities included sponges of various forms, feather and strand hydroids, and colonial ascidians. Fauna also included cushion stars, long-armed sea stars, gastropods (for example, *Calliostoma* sp., *Maoricolpus roseus*), fan worms, snake stars, sea slugs, large anemones, tube-dwelling anemones, crinoids (or crinoid-like forms; rare), scallops (*Pecten novaezelandiae*), and variegated scallops (for example *Chlamys* spp.). There were very few observations of horse mussels or brachiopods seen in video taken from this habitat, and large biogenic clumps were very rare.

HMBB habitat was characterised primarily by areas of intact horse mussels embedded in the sediment, brachiopods, and other mobile and encrusting taxa (Elvines et al. 2019, Elvines et al. 2021). Other fauna reported within this habitat type included brittle stars (*Ophiopsammus maculata*), sea cucumbers (Holothuroidea), gastropods (notably *Astraea heliotropium* and *Maoricolpus roseus*), large anemones (unidentified), fan worms (Sabellidae), feather hydroids (Aglaopheniidae), fan shells (Pectinidae), bryozoans (primarily bushy and flexible branching forms), cushion stars (Valvatida), long-armed sea stars (possibly *Cosmasterias dyscrita*), crabs (unidentified), sea slugs (Pleurobranchaea), sponges (various morphologies; encrusting, arborescent, globular, tube clustered), solitary (including *Styela clava*) and colonial ascidians (various), and tube-dwelling anemones (Ceriantharia).

PR habitat comprised patches of broken shell debris (often non-living horse mussels) in areas of otherwise homogenous soft, muddy sediments. Brachiopods and horse mussels (live, ~10 %) were seen occasionally in these areas. Conspicuous encrusting and mobile taxa were very similar to those in the horse mussel/brachiopod beds. Between the patches of shell debris and biogenic structure, there were large (10s of metres) stretches where epifauna was sparse. Here, the seafloor had reasonably homogenous, muddy sediments with burrow holes and few to no epifauna visible. The most prevalent species seen in the sparsely populated areas was an unidentified species of tube-dwelling anemone. Occasionally, small clusters of shell debris with encrusting organisms were seen, as well as a fine green algal-like growth.

Within CR habitat, aggregates of biogenic material were prevalent, ranging from small (0.1–0.5 m) to large (more than 1 m) high-relief biogenic clumps, sometimes appearing to form more extensive 'bands' (reef-forming biogenic clumps). Within this habitat, the 'biogenic clump reef' structures were patchy, punctuated by large (20–70 m or more) areas of more homogenous seabed with low-density epifauna and no large biogenic structures. Diverse epifaunal assemblages were associated with the biogenic

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[11] Commonality is defined as % of time viewed along transect at Blue Endeavour. See also Table 1.

structure and included nudibranchs, anemones (various), sea cucumbers, large parchment worms, tree and feather hydroids, colonial ascidians, encrusting and small and large sponges (various forms), bryozoans (frame building, flexible, bushy), gastropods (including *Astraea heliotropium*) and bivalves (including *Chlamys* spp.). Brachiopods were positively identified occasionally, and possible *Atrina zelandica* were rare. Clump reef represents habitat that can be important during vulnerable life stages of marine species as, like patch reef, it contains a relatively high diversity of organisms compared to LDE habitats.

Mixed biogenic habitat comprises biogenic communities, dominated by sponges, soft bryozoans, brachiopods, and other mixed taxa. Biogenic components of this community tend to be present in densities covering > 30% of the seafloor.

2.3   Hananui

Hananui is a consent application lodged by Ngāi Tahu Seafood Resources Ltd to develop an offshore salmon farm 2–6 km offshore of northern Stewart Island/Rakiura (hereafter Stewart Island). Historical records of bottom sediments show the general Foveaux Strait region to be a mixture of pebbly gravel, medium to fine sandy pebble gravel, and well- and poorly sorted fine sand (Cullen 1962). A recent analysis of local knowledge of biogenic habitats reports a number of habitat types throughout Foveaux Strait and around Stewart Island with patches of bryozoans, sponges, 'coral', large bivalve beds, tubeworms, sea tulips, and complex reef identified (Jones et al. 2018). A rich variety of macroalgae are also reported for shallow rocky reefs in the Foveaux Strait region, while Foveaux Strait rock-wall communities have a very high diversity of encrusting invertebrates (Kettles et al. 2017). Depths within the vicinity of the Hananui site range from 25 m to 40 m, and mid-depth current speeds range from 38 cm/s to 44 cm/s.

'Mullock' reefs, areas of complex biogenic reef habitat, exist in areas of strong current flow in Foveaux Strait. The reefs comprise the bryozoan *Cinctipora elegans*, together with other encrusting bryozoan species, ascidians, sponges, and polychaete worms (Cranfield et al. 1999). Large bivalves, including dredge oysters (*Ostrea chilensis*) and mussel species, were historically known to be abundant in areas of mullock reef.

An assessment of effects, including habitat mapping, has been conducted for the proposed Hananui site (Bennett et al. 2022), and this report describes communities within each habitat type. Habitat types include sand (S), sandy shell hash (SSH), coarse gravel and sand (CGS), bushy bryozoan thickets[12] (BBT), and bryozoan-sponge reefs (BS). While percentage cover was assessed only for the proposal area at Hananui, it is important to note that additional habitat exists outside this area but within the predicted depositional footprints of the proposed farm. Therefore, the composition of habitat that may be affected by the farm is likely to differ from that presented here.

Sand is the dominant habitat type present at the proposed Hananui site, at ~77% of the total proposal area. Bennett et al. (2022) reported observing the following taxa in video surveys: brittle stars (mostly *Ophiopsammus maculata*), some red and brown drift algae, occasional tufts of bushy bryozoans and feather hydroids, isolated encrusting bryozoans, sponges, ascidians (such as sea tulip, *Pyura pachydermatina*), sea cucumbers (*Australostichopus mollis*), 11-armed sea stars (*Coscinasterias muricata*), cushion stars (*Patiriella regularis*), occasional gastropods including saw shells (*Astraea heliotropium*) and turret shells (*Maoricolpus roseus*), brachiopods (likely *Neothyris lenticularis* or *Magasella sanguinea*), and large bivalve species including the dredge oyster (*Ostrea chilensis*), scallops (*Pecten novaezelandiae*), dog cockles (likely *Tucetona laticostata*), bearded horse mussel (*Modiolus areolatus*), tuatua (*Paphies subtriangulata*), and long trough shell (*Oxyperas elongata*). Similar taxa were observed in SSH (11% of the proposal area) and CGS (2.7% of the proposal area) habitats.

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[12] Bushy bryozoan thickets occur where bryozoans are interspersed with calcareous tubeworm mounds. Bryozoan thickets may also be referred to as bryozoan beds.

Bushy bryozoan thickets covered approximately 0.3% of the proposed Hananui site, and included areas of abundant bushy bryozoans (*Orthoscuticella innominata*); calcareous tube worms (likely *Galeolaria hystrix*), often in mounds; a moderate abundance of small erect bryozoans (mainly *Cinctipora elegans*) and sponges in some places; a moderate number of feather hydroids; sparse foliose red algae; some red and brown drift algae; occasional individual or small patches of large bivalves, including dredge oysters (sparse to patchy); scallops (sparse); bearded horse mussels (sparse) and dog cockles (sparse to abundant); occasional patches of brachiopods; occasional kina (*Evechinus chloroticus*), sea tulips, sea cucumbers, brittle stars, cushion stars, and 11-armed sea stars; and gastropods, including saw shells.

Bryozoan-sponge reefs covered approximately 9.1% of the proposed Hananui site and included biogenic clumps formed by the massive encrusting bryozoan *Celleporaria agglutinans* and erect bryozoan *Cinctipora elegans*; a number of other erect and encrusting species of bryozoan, including fragile lacy forms such as *Hornera foliacea* and possibly *Hornera robusta*; abundant encrusting and erect sponges (including *Dactylia varia*, *Iophon minor*, and *Crella incrustans*); colonial ascidians (including *Botrylloides* sp., *Botryllus* sp., and *Eudistoma circumvallatum*); calcareous tubeworms (likely *Galeolaria hystrix*); a moderate abundance of bushy bryozoan and feather hydroids; sparse foliose and encrusting red algae; occasional individual or small patches of large bivalves, including dredge oysters (sparse to patchy), scallops (sparse), bearded horse mussels (sparse), and dog cockles (sparse to abundant); occasional patches of brachiopods; patches of sea anemones (*Anthothoe albocincta*) in some areas; and occasional kina, sea tulips, sea cucumbers, brittle stars, cushion stars, 11-armed sea stars and gastropods, including saw shells.

The conspicuous biota common among all habitat types (although varying in abundance) were the brittle star (*Ophiopsammus maculata*) and blue cod (*Parapercis colias*). Areas of bushy bryozoan thickets and bryozoan-sponge reefs had significantly higher biodiversity than sand-dominated areas. Some taxa were observed in all habitat types, including brachiopods (likely either *Neothyris lenticularis* or *Magasella sanguinea*) and large bivalve species such as dredge oysters (*Ostrea chilensis*), scallops (*Pecten novaezelandiae*), dog cockles (likely *Tucetona laticostata*), and bearded horse mussels (*Modiolus areolatus*). Abundances of these taxa were generally greater within bushy bryozoan thickets or bryozoan-sponge reefs than in sandy areas.

2.4   Project South (Southland)

Project South is a consent application lodged by Sanford Ltd to develop an offshore salmon farm in eastern Foveaux Strait, to the southeast of Ruapuke Island. An assessment of ecological effects has been completed for the site (James et al. 2020); however, this provides only high-level descriptions of the benthic habitats within the area of interest. A habitat map of the area around the northern part of Stewart Island and bottom of the South Island by Michael (2010) shows the dominant habitat type present at the application site is sand ripple habitat. North and northwest of the application site, diverse habitats are present and include rocky patch reef with epifauna, flat gravels with clean (usually *Ostrea chilensis*, *Pseudoxyperas elongata,* and *Glycymeris modesta*) or encrusted (usually bound by small encrusting bryozoans) shell, flat gravels with red algae and sea tulips (*Pyura pachydermatina*), gravel waves or lowly undulating gravels with clean shell in the troughs, flat sand and gravel with and without biogenic patches, large sand waves, and biogenic areas (along the coastline of Stewart Island).

Drop-camera and video footage collected by James et al. (2020) found no indication of biogenic reefs within the vicinity of the Project South area of interest. Sparse epifauna comprising a few brittle star (*Ophiopsammus maculata*) and other sea stars were noted on the seabed around the west and northern part of the application site. Oysters and other bivalves, as well as crinoids, were observed in the eastern part of the application site. Sea urchins were observed around the western end of the application area. Solitary ascidians were seen in the shallower area to the east and northern part of the application site, and a number of anemones were noted throughout much of the application site.

Overall, the seabed in the application site appears to be relatively homogeneous, comprising sand, occasionally mixed with mud or shell hash, and sparse epifauna and infauna. Ecological diversity was

described as low by James et al. (2020). They found the ecologically sensitive habitats that have previously been reported in Foveaux Strait, such as bryozoan reefs or oyster reefs, were absent from the proposed farm site. While the proposed site for Project South was predicted as suitable habitat for bryozoan species by Wood et al. (2013), James et al. (2020) did not find the area to contain ecologically sensitive bryozoan reefs. As the described habitats at this site were high level and the taxa identified were also reported at Blue Endeavour and Hananui, we did not include this site in further consideration for the remainder of the report.

2.5   Tory Channel

There are currently three salmon farms operating in Tory Channel: Te Pangu Bay, Clay Point, and Ngāmahau Bay, all of which are considered high flow sites (average current flow more than 10 cm s^{-1}) (Fletcher et al. 2022). Average current speeds are 15 cm s^{-1} at Te Pangu, 19.6 cm s^{-1} at Clay Point, and 22 cm s^{-1} at Ngāmahau (Fletcher et al. 2021, McGrath et al. 2021). At the time of this report, the dispersive capability of the Tory Channel farming environment is more like the open ocean environment than any other existing farm area.

Habitats beneath and immediately adjacent to these farms typically consist of soft sediment and are found at depths of 20–40 m. Sediment can range from very fine and homogenous to coarse shell hash or cobbles. Burrow holes and trail marks from mobile fauna are common. Epifauna is generally sparse and includes brittle stars (*Ophiopsammus maculata*), sponges, anemones (often *Anthothoe albocincta*), sea cucumbers (*Australostichopus mollis*), bryozoans, tubeworms (Serpulidae), 11-armed sea stars (*Coscinasterias muricata*), apricot stars (*Sclerasterias mollis*), cushion stars (*Patiriella regularis*), kina (*Evechinus chloroticus*), horse mussels (*Atrina zelandica*), hydroids, fan worms, ascidians (solitary), and hermit crabs.

The Clay Point, Te Pangu Bay, and Ngāmahau Bay salmon farms also have rocky reefs nearby. Overall, monitoring of these reef habitats has not shown obvious alterations to communities at farm sites relative to reference locations. Important ecosystem changes can manifest as more than just changes in the abundance or distribution of a species. Subtle changes in resource use, food web dynamics, or organism physiology and health may take a long time to result in population changes that can be detected through a visual assessment. Biochemical, physiological, or molecular approaches may reveal more subtle, but nevertheless important, changes in populations as a result of farm-derived organic matter.

Species found to be ubiquitous across the Tory Channel farm reef sites include snake stars (*Ophiopsammus maculata*), cushion stars (*Patiriella regularis*), kina (*Evechinus chloroticus*), sea cucumbers (*Australostichopus mollis*), solitary ascidians, encrusting ascidians, gastropods (including *Calliostoma* sp., *Trochus* sp., and *Lunella smaragda*/cat's eye snail), grey cup sponges (*Ecionemia alata*), encrusting bryozoans, and tree hydroids. Other species found in rocky reef environments near farms in the Marlborough Sounds include orange sponges (*Crella encrustans*), blue encrusting sponges, duck's bill limpets (*Scutus breviculus*), yellow colonial ascidians (*Aplidian phortax*), purple ascidians (*Botrylloides leachii*), coral (colonial stony), Cook's turbans (*Cookia sulcata*), white stripped anemones (*Anthothoe albocinta*), colonial sea squirts (*Aplidium phorax*), tubeworm mounds (formed by *Galeolaria hystix*), tubeworms (Serpulidae), limpets (*Cellana stellifera* and *Siphonaria* sp.), flat oysters (*Ostrea chilensis*), and 11-armed sea stars (*Coscinasterias calamaria*).

Table 1: Summary of taxa present and their frequency of occurrence at each location of interest and commonality (if known). Habitat codes are as follows: HMBB = horse mussel brachiopod beds, PR = patch reef, CR = clump reef, LDE-CS = low-density epifauna – coarse sediment, LDE-SS = low-density epifauna – soft sediment, BBT = bushy bryozoan thickets, BS = bryozoan-sponge (mullock), S = sand, SR = sand ripple, FG = flat gravel, SS = soft sediment, RF-PS = reef Pelorus Sound, RF-TC = reef Tory Channel. Codes for commonality at the Blue Endeavour site are as follows: (R) = rare < 5%, (O) = occasional 5–20%, (U) = uncommon 20–50%, (C) = common 50–80%, (A) = abundant 80–100%, (?) = commonality not previously reported. Codes for commonality at the Hananui site are as follows: (I) = isolated individuals, (P/O) = patchy/occasional 5–10% coverage or 2–3 individuals in close proximity, (M) = moderate 10–40% coverage or several individuals in close proximity, (A) = abundant > 40% coverage or dense aggregations. Commonality is defined as % of time viewed along transect at Blue Endeavour and as % cover or through individual counts at each station for Hananui. Biogenic habitats are italicised. (Continued on next 3 pages)

Species	Functional role	Blue Endeavour	Hananui	Project South (commonality not reported)	Tory Channel (commonality not reported)
Horse mussels (*Atrina zelandica*)	Suspension-feeding bivalves – epifaunal	*HMBB (R-A), PR (R-O),* LDE-CS (R), LDE-SS (R), *CR (R)*			SS
Talochlamys		*HMBB (O-U), PR (O),* LDE-CS (?), *CR (O)*			
Fan shells (Pectinidae)		*HMBB (?), PR (?)*			
Bearded horse mussels (*Modiolus areolatus*)			*BBT (P/O), BS (P/O),* S (P/O)		
Green-lipped mussels (*Perna canaliculus*)					SS
Blue mussels (*Mytilus galloprovincialis*)					SS
Dredge/flat oysters (*Ostrea chilensis*)			*BBT (P/O), BS (P/O),* S (P/O)	SR (?), FG (?)	*RF-PS*
Scallops (*Pecten novaezelandiae*)		LDE-CS (?)	*BBT (P/O), BS (P/O),* S (P/O)		SS
Brachiopods (Blue Endeavour: *Notosaria nigricans*, *Magasella sanguinea*, *Calloria inconspicua*; Hananui: either *Neothyris lenticularis* or *Magasella sanguinea*)	Suspension-feeding brachiopods – mid-water column	*HMBB (U-A), PR (O-UO-U),* LDE-CS (R), LDE-SS (R), *CR (R-U)*	*BBT (P/O), BS (P/O),* S (P/O)		
Tube-dwelling anemones (Ceriantharia)	Sessile suspension feeders – anemones	*HMBB (R-A), PR (U),* LDE-CS (U-C)			
Giant wandering anemone (*Phlyctenactis tuberculosa*)		*HMBB (R), PR (R),* LDE-CS (R)			

Species	Functional role	Blue Endeavour	Hananui	Project South (commonality not reported)	Tory Channel (commonality not reported)
White striped anemone (*Athothoe albocincta*)			*BS (P/O)*		*RF_PS, SS*
Anemones – undefined		*HMBB (O), PR (O), CR (O)*		SR (?)	
Feather hydroid (Aglaopheniidae) / hydroid tree	Sessile suspension feeders – hydroids	*HMBB (O-U), PR (O),* LDE-CS (R/O), LDE-SS (?), *CR (O)*	*BBT (M–A), BS (M-A),* S (P/O)		*RF,* SS
Solitary cup coral	Sessile suspension feeders – coral	*PR (?),* LDE-CS (R), LDE-SS (R), *CR (?)*			
Colonial stony coral					*RF_PS*
Tubeworm mounds / calcareous tubeworms from family Serpulidae (*Galeolaria hystrix* at Hananui)	Sessile suspension feeders – habitat-building tubeworms	*PR (R-O),* LDE-CS (R), LDE-SS (R), *CR (?)*	*BBT (A), BS (?)*		*RF_PS,* SS
Tubeworm (Chaetopteridae)		LDE-CS (R)			
Giant tubeworms		*HMBB (R), PR (R-U),* LDE-CS (R), *CR (O)*			
Fan worm (Sabellidae)	Sessile suspension feeders – habitat-building tubeworms	*HMBB (O-R), PR (R-O),* LDE-CS (?)			SS
Large parchment worm		*CR (?)*			SS
Bryozoans (bushy and flexible branching forms, *Orthoscuticella innominata*, *Hornera foliacea* and possibly *Hornera robusta* at Hananui)	Sessile suspension feeders – bryozoans, bushy and flexible	*HMBB (R-C), PR (O-U),* LDE-CS (O-U), LDE-SS (?), *CR (A)*	*BS (M-A), BBT (A),* S (I-P/O)		
Erect bryozoans (mainly *Cinctipora elegans*)	Sessile suspension feeders – bryozoans, erect frame building	CR (R-U)	*BBT (M-A), BS (?),*		
Massive encrusting bryozoan (*Celleporaria agglutinans*, encrusting brown in Tory Channel)	Sessile suspension feeders – bryozoans, encrusting		*BS (?)*		*RF,* SS
Yellow encrusting sponge	Sessile suspension feeders – sponge, encrusting	*HMBB (O), PR (O),* LDE-CS (R), *CR (O)*			
Orange sponge (*Crella encrustans*)			*BS (A)*		*RF*
Blue encrusting sponge					*RF*
Encrusting sponges		*HMBB (R-C), PR (O),* LDE-CS (O), *CR (?)*			

Species	Functional role	Blue Endeavour	Hananui	Project South (commonality not reported)	Tory Channel (commonality not reported)
Finger sponge	Sessile suspension feeders – sponge, erect	*HMBB (O-U), PR (O), LDE-CS (R), CR (O)*			SS
Orange erect sponge		*HMBB (O), PR (O)*			
Sponge (*Dactylia varia*)			*BS (A)*		
Sponge (*Iophon minor*)			*BS (A)*		
Grey cup sponges (*Ecionemia alata*)					*RF*
Sponges (other or nondescript)	Sessile suspension feeders – sponge, other	*CR (O)*	*BBT (M – A)*, S (I)		SS
Solitary ascidian (including *Styela clava*)	Sessile suspension feeders – solitary ascidian	*HMBB (R)*		SR (?)	*RF, SS*
Sea tulip (*Pyura pachydermatina*)			*BBT (P/O), BS (P/O)*, S (I)	FG (?)	SS
Colonial ascidians	Sessile suspension feeders – colonial ascidian	*HMBB (O-C), PR (O), LDE-CS (O), CR (O)*			*RF, SS*
Colonial ascidians (*Botrylloides* sp.)			*BS (?)*		*RF*
Colonial ascidians (*Botryllus* sp.)			*BS (?)*		
Colonial ascidians (*Eudistoma circumvallatum*)			*BS (?)*		
Yellow colonial ascidian (*Aplidium phortax*)					*RF_PS*
Burrowing sea cucumber	Sessile suspension feeders – sea cucumber	*HMBB (O),* LDE-CS (R)			
Crinoids	Sessile suspension feeders – crinoids	LDE-CS (?)	*BS (P/O)*	SR (?)	
Brittle stars (*Ophiopsammus maculata*)	Mobile deposit feeders	*HMBB (O), PR (O-U), LDE-CS (O), CR (O)*	*BBT (P/O), BS (P/O)*, S (P/O)	SR (?)	*RF, SS*
Sea cucumber (*Australostichopus mollis*)		*HMBB (O), PR (R-O), CR (O)*	*BBT (P/O), BS (P/O)*, S (I)		*RF, SS*
Turret shell (*Maoricolpus*)	Mobile suspension feeder	*HMBB (O), PR (O),* LDE-CS (?), CR (O)	S (O)		SS
Kina (*Evechinus chloroticus*)	Mobile grazers		*BBT (P/O)*	SR (?)	*RF*
Cat's eye snail (*Lunella smaragda*)					*RF*
Duck's bill limpet (*Scutus breviculus*)					*RF*
Cook's turban (*Cookia sulcata*)					*RF_PS*
Limpet (*Cellana stellifera*)					*RF_PS*
Limpet (*Siphonaria* sp.)					*RF_PS*

Species	Functional role	Blue Endeavour	Hananui	Project South (commonality not reported)	Tory Channel (commonality not reported)
Calliostoma sp.		LDE-CS (?)			*RF*
Trochus sp.					*RF*
Cushion stars (*Patiriella regularis*)	Mobile scavenger / predator	*HMBB (O), PR (?), LDE-CS (?)*	*BBT (P/O), BS (P/O)*, S (I)		*RF*, SS
11-armed sea star (*Coscinarius muricata*)		*HMBB (R),*	*BBT (P/O), BS (P/O)*, S (I)		*RF_PS*, SS
7-armed sea star (*Astrostole scabra*)		*HMBB (R-O), PR (O)*			
Long-armed sea star (possibly *Cosmasterias dyscrita*)		*HMBB (O), PR (?), LDE-CS (?)*			
Apricot star (*Sclerasterias mollis*)					SS
Crab, nondescript (Brachyura)		*HM(R), PR (?)*			SS
Hermit crab (*Pagurus novizealandiae*)					SS
Hairy seaweed crab (*Notomithrax ursus*)					SS
Penion sp.		*HMBB (R-O)*			
Nudibranch (Pleurobranchaea)		*HMBB (R-O), PR (O), LDE-CS (O), CR (O)*			
Circular saw-shell (*Astraea heliotropium*)	Mobile, undefined	*HMBB (R-O), PR (O), CR (O)*	*BBT (P/O), BS (P/O)*, S (P/O)		SS

3. ECOLOGICAL AND CULTURAL SIGNIFICANCE OF TAXA

This section provides an introduction to the key epifaunal taxa present at proposed OOA sites in New Zealand. It also discusses the ecological importance of taxa at the Blue Endeavour and Hananui sites, as well as the significance of these taxa to local iwi[13,14]. This section also covers whether the taxa meet the definition of sensitive marine benthic habitats as defined by MacDiarmid et al. (2013). Sensitivity considers the tolerance of a species or habitat to damage from an external factor, and the time taken for recovery from such damage. Throughout the remainder of this report, we have focused on sessile and mobile epifauna and have excluded infauna from our framework assessment, as the response of infauna to organic enrichment is relatively well understood and tools already exist in New Zealand to assess the impact of aquaculture in soft-sediment environments. Such tools are routinely used to assess compliance with consent conditions (see McGrath et al. 2022a, 2022b for more information).

3.1 Mobile deposit feeders

Deposit feeders ingest particles associated with sediments, or they ingest the sediment particles themselves and remove organic material (including microbes) associated with the sediment grains. Species are often generalists, extracting any available material with nutritional value; however, they may feed on particular grain sizes and at particular depth layers in the sediment. Deposit feeders, opportunistic taxa, and those with benthic/pelagic larval development are typically more common in soft-sediment habitats at aquaculture sites when compared with reference sites (Macleod et al. 2007).

Mobile deposit-feeding taxa, such as snake stars, long-armed sea stars, and sea cucumbers, have been consistently observed within areas proposed for OOA development but were not usually densely distributed (Section 2.5, Table 1). While these organisms are not habitat formers, they can provide important ecosystem services. Deposit feeders are major bioturbators[15] in marine sediments, influencing nutrient cycling, pollutant burial, and sediment–water exchange (Snelgrove 2013). Sea cucumbers, for instance, can assist in reducing organic loadings under farms by consuming waste feed and fish faeces (Zamora et al. 2016). As primary consumers, sea cucumbers, brittle stars, and sea urchins (kina) also serve an important role in capturing organic carbon and nutrients for use by higher-trophic-level organisms and reducing unfavourable, low-oxygen conditions. Furthermore, kina are culturally important for local iwi as kaimoana[16].

Some deposit feeders observed at offshore sites are known prey for blue cod/rāwaru (hereafter blue cod). The impact of the proposed Hananui development on blue cod was raised as a concern by whānau[17]. While the proposed site is not a known blue cod fishery, sightings of blue cod were common throughout site assessments, suggesting they may feed in the area and could be indirectly impacted through effects to prey species.

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[13] While iwi consultation will be ongoing throughout the duration of this project, for the purposes of this report existing documentation relevant to iwi perspectives on the current proposed aquaculture developments were reviewed to get a better understanding on species of importance to Māori.

[14] For the Blue Endeavour site, documentation included information from submissions (see Appendix 1). For the Hananui application, the *Ngā Hua o Āpiti Hono Tātai Hono* assessment (https://www.epa.govt.nz/assets/Uploads/Documents/Fast-track-consenting/Hananui/the-application/Appendix-B_Nga-Hua-o-Apiti-Hono-Tatai-Hono-report.pdf) and cultural impact assessment (https://www.epa.govt.nz/assets/Uploads/Documents/Fast-track-consenting/Hananui/the-application/Appendix-A_Cultural-impact-assessment.pdf) were reviewed.

[15] Bioturbation is the biogenic transport of sediment particles and pore water. The activity reduces sediment stratification and influences the chemical and physical properties of the sediment.

[16] Food that has been gathered from the sea.

[17] Extended family, family group.

3.2 Bivalves

Bivalves fulfil a variety of roles integral to ecosystem functioning (MacDiarmid et al. 2013, Anderson et al. 2019) and are an important part of coastal and shelf ecosystems in New Zealand (Anderson et al. 2019). Living bivalves provide resources to other organisms through assimilation and cycling of organic matter and nutrients, which can then flow through to higher-trophic-level organisms (Schlieman et al. 2022) and as filter feeders they influence water quality (Rothschild et al. 1994). Non-living and living shells of bivalves, particularly large species, can form complex structures that provide shelter and substrate on which larvae may settle (Cranfield et al. 2004), in turn supporting the development of more diverse benthic communities (MacDiarmid et al. 2013, Fletcher 2015, Anderson et al. 2019). The role of large bivalve species as ecosystem engineers and providers of other ecosystem services makes them ecologically important. Many large bivalve species are also of commercial value and are culturally important kaimoana[18] and mahinga kai[19] resources.

The definition of a bivalve bed is 30% or higher coverage of both living and dead specimens from imaging surveys covering 100 m^2 or more (MacDiarmid et al. 2013). At Hananui, cover of live large bivalve specimens within the survey area was rarely greater than 30%, with the exception of dog cockle abundance at a small number of sites. However, in some areas the percentage cover of empty large bivalve shells (predominantly oyster shells) was greater than 30%, so in these areas habitat formed by oysters was also classed as a bivalve bed (Bennett et al. 2022)[20]. At the Blue Endeavour site, intact horse mussels were the predominant epifauna in HMBB habitat, where they occupied up to 30% of a 100 m^2 area, with ~50% of these estimated to be alive. In PR habitat, horse mussel shells covered 20–80% of the seabed in patch reef habitat (live animals were only occasionally present at ~10% or less). These descriptions classify these habitats as horse mussel beds/patches following MacDiarmid et al. (2013). We considered the presence of large bivalve beds to be of ecological significance to an area, given their contribution to energy cycling within food webs.

The cultural impact assessment undertaken for the Hananui application noted concern raised by whānau regarding potential damage to oyster/tio (hereafter oyster) beds from the proposed operation. The dredge oyster (*Ostrea chilensis*) was occasionally observed at both the proposed Hananui and Project South sites, though oysters were absent from the proposed Blue Endeavour site. As filter feeders, oysters, along with other suspension feeders, provide an important ecosystem service by reducing water turbidity and generating more favourable conditions for other valuable species, such as pāua (*Haliotis iris*) and crayfish/kōura (*Jasus edwardsii*). Biogenic habitat formed by oysters is potentially important for settling larvae and juvenile fish. Scallops/tupa were also identified as important kaimoana species at the Blue Endeavour site, as the farm location is located adjacent to an important scallop fishing ground (see Appendix 1 for more information).

3.3 Brachiopods

Brachiopods are part of an ancient phylum that have been largely unchanged for 500 million years and are therefore of particular scientific, in addition to conservation, importance (MacDiarmid et al. 2013). Dense, stable beds of brachiopods are important contributors to benthic ecosystems and support a variety of organisms (Morrison et al. 2014). Acting as ecosystem engineers, brachiopods provide hard substrate for other organisms to settle on (Sprinkle & Rodgers 2010), and as filter feeders they fix carbon and contribute to the cycling of nutrients and organic matter through food webs (Rudwick 1962).

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[18] Seafood, shellfish.

[19] The value and protection of food resources that sustain life, including species, natural habitats and materials, and the practices used for harvesting food.

[20] The imaging survey areas at both Hananui and Blue Endeavour were greater than 100 m^2 and therefore the definition of a bivalve bed proposed by MacDiarmid (et al. 2013) is appropriate.

A brachiopod bed is defined as one or more specimens occurring per square metre of sampling[21] (MacDiarmid et al. 2013). Brachiopod densities at Hananui ranged from sparse (isolated individuals) to abundant (dense aggregations, at two sites only), although rarely were more than five observed at one site. These densities were not high enough for the habitat to be defined as brachiopod bed following MacDiarmid et al. (2013). Brachiopods were abundant in some areas of HMBB habitat at the Blue Endeavour site, where they exceeded densities of one per square metre, thus constituting the stratum as a brachiopod bed. Brachiopods were also occasionally recorded in patch reef habitat at Blue Endeavour, though densities there were fewer than one per square metre. At both the Hananui and Blue Endeavour sites, brachiopods could be sparse or abundant, but distribution within habitat types was typically patchy (Table 1).

While brachiopods themselves were not specified as species of importance to Māori in the documents consulted, extensive beds of this species, as observed at the Blue Endeavour site, provide valuable habitat for larval settlement and for succession of biota requiring hard substrate on which to attach.

3.4   Bryozoans

Bryozoans are colonies of very small filter-feeding animals called zooids. Zooids possess tentacle crowns that are extended for feeding on living and non-living organic particles from the surrounding water column (Gordon & Mills 2016). In addition to feeding zooids, bryozoans include other types of zooids that perform roles such as reproduction and nutrient storage (Gordon & Mills 2016). Bryozoans exist in a large variety of forms, including encrusting, branching and flexible, and reef-forming/frame-building species. The latter are of particular significance due to their ability to form complex three-dimensional structures in areas that are otherwise relatively barren and sandy. These structures provide habitat for a multitude of organisms, helping support and maintain local biodiversity and consequently enhance ecosystem services (Wood 2005, MacDiarmid et al. 2013). Organisms that make use of habitat provided by bryozoans include ophiuroids, annelids, decapods, sponges, ascidians, and bivalve molluscs. These structures also provide shelter and a place for mobile fauna, such as fish and sea stars, to live and reproduce.

Bryozoan beds, or thickets, can be deemed to exist when colonies of large frame-building bryozoans (greater than 50 mm in three dimension) form more than 4% mean cover over large areas (10–100s km^2) or dominate the seabed (over 50% per square metre) in smaller areas (10–100 m^2) (MacDiarmid et al. 2013). At Hananui, average reef-forming bryozoan cover was approximately 30%, and in places reef-forming bryozoan cover was estimated to be greater than 50%. Based on these descriptions, bryozoan beds or thickets were present within the survey area. Coverage of the bushy clown-hair bryozoan was estimated to be up to 80% where it formed thickets (ca. 22% coverage on average where it occurred). At the Blue Endeavour site, small bryozoans were the prevalent epifauna within low-density epifauna–coarse sediment habitats; however, the coverage of bryozoans was generally very sparse. Flexible branching and bushy types dominated this habitat, while frame-building species were rare. As such, the soft bryozoans seen within the Blue Endeavour survey area were not considered significant in terms of ecosystem services and habitat provision and did not meet the definition of bryozoan beds or thickets.

Bryozoans form extensive habitats at the Hananui site and, although the taxa themselves are not listed as taonga or used directly as kaimoana, they have an important role in enhancing biodiversity and food web complexity within the area of interest. Frame-building bryozoans are particularly important habitat formers that support mahinga kai resources such as oysters (Cranfield et al. 1999, 2004) or provide nursery habitats and breeding grounds for juvenile fish, including blue cod (Cranfield et al. 2001, Carbines et al. 2004).

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[21] Seabed imaging methods to determine the presence (or absence) of brachiopod beds is cautioned against by MacDiarmid et al. (2013). However, data collected from the analysis of seabed imagery are the only data available for the areas of interest; therefore, these data have been used to define habitat presence in this review.

3.5   Sponges

Sponges are the most common marine invertebrate around the New Zealand coastline and fulfil many important functional roles in benthic ecosystems, including providing nursery grounds for juvenile fish, stabilising substrate, and facilitating nutrient-cycling processes (Bell 2008). They feed by filtering water using specialised cells called choanocytes. Choanocytes propel a one-directional water current through the sponge body: water enters through small inhalant pores on the surface and exits through several large exhalent holes. Food is captured in a fringe surrounding the base of the choanocyte flagella and is passed back through the cell body to other cells that distribute it around the sponge. Excretory products exit in the water current as they leave the body. Sponges often host diverse and species-specific communities of microorganisms that are thought to contribute to many aspects of the sponge's physiology and ecology (Webster & Thomas 2016). This microbial community is known as a 'microbiome' and, together with the host organism, as a 'holobiont'.

A sponge garden may be defined as 25% or greater cover of one or more species in either uniform or clumped distribution over an area of 100 m^2 or more (MacDiarmid et al. 2013). Based on this description, sponge gardens exist within the Hananui survey area (ca. 28% coverage on average where they occurred). Sponges were observed at the Blue Endeavour site; however, percentage cover did not meet the criteria to be characterised as a sponge garden.

Through provision of food and habitat resources to a range of species, sponges enhance ecosystem functioning and likely support mahinga kai and kaimoana populations at the Hananui site, including blue cod and crayfish. Large sponges with high morphological and taxonomic variability are also present at the proposed Blue Endeavour site, and serve a similar role, although traditional mahinga kai and kaimoana resources such as pāua, crayfish, oysters, and blue cod are unlikely to occur here due to the greater depths at this site.

3.6   Tubeworms

Tubeworm species in the family Serpulidae secrete tubes of calcium carbonate and grow as either individuals or in colonies (Anderson et al. 2019). Tubeworm species are important biomineralisers and provide three-dimensional habitat for a variety of organisms, often resulting in biodiversity hotspots (MacDiarmid et al. 2013). Species found to be associated with these mounds include polychaete worms such as *Spirobranchus latiscapus*, burrowing anemones (*Cerianthus* sp.), octopus, blue cod, and tarakihi (*Nemadactylus macropterus*) (Davidson et al. 2011). Low-relief[22] tubeworm mounds, known as Chaetopteridae[23] worm fields, can also be correlated with increases in fish densities (Stoner et al. 2007).

A tubeworm mound is defined as a raised reef-like structure 1–100 m in diameter, while a thicket is present where one or more mounds occur, or intertwined tubes account for over 10% of the catch in dredge or beam trawls, or where intertwined tubes are collected in a single grab sample (MacDiarmid et al. 2013, Anderson et al. 2019). Chaetopteridae worm fields are present when worm tubes and associated epifaunal species occupy 25% or more of the visual field in underwater imagery of areas greater than 500 m^2 (MacDiarmid et al. 2013). Where present at the Hananui site, tubeworm abundance ranged from patchy (a few tubes present in video footage) to abundant (present in over 60% of the area covered in video footage). Following the definition by MacDiarmid et al. (2013), tubeworm mound habitat was present where tubeworms were considered abundant. Tubeworm mounds were also present, but rare, at the Blue Endeavour site.

Although tubeworms were not specified as valuable to iwi at either the Blue Endeavour or Hananui sites, they are habitat formers and provide substrate for larval settlement and attachment.

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[22] Those that project only slightly from the ground/flat surface.
[23] Chaetopteridae may be classed as infaunal when tubes are constructed within sediment, and epifaunal when tubes are attached to rocky reef substrate.

3.7 Cnidarians

Cnidarians have specialised stinging cells called nematocysts that they use to capture prey. This phylum includes sea anemones, true corals, soft corals, sea pens, and hydrozoans. Some members of this group are able to attach into soft-sediment seafloors via a pedal disc (Kelley et al. 2021).

3.8 Ascidians

Ascidians are common fouling organisms around the world, settling and growing in great abundance on both natural and artificial substrates. Most endemic species are present in relatively low numbers in intertidal and subtidal environments around New Zealand, while introduced species can be present in great abundance and in densities that exclude other species. Ascidians feed by filtering water through their body via inhalant and exhalant siphons. Both solitary and colonial (groups of individuals) ascidians have a short-lived free-living larval stage prior to settlement. Ascidians can be a crucial factor in structuring motile fauna in seabed communities through modification of physical habitat structure (Yakovis et al. 2005).

Ascidians are not defined as forming sensitive marine benthic habitats by MacDiarmid et al. (2013); however, these organisms are reliant on habitat-forming organisms for settlement substrate at open ocean sites.

4. EPIFAUNAL RESPONSE TO ORGANIC ENRICHMENT AND SEDIMENTATION

The primary seabed impact from salmon farm operations is the addition of farm-derived organic material, which primarily comprises faecal waste and, to a lesser extent, uneaten feed pellets. The particle size, density, and organic composition of salmon faecal waste is largely dependent on diet (Reid et al. 2008, 2009)[24], while the nutrient properties of waste/uneaten food particles will influence the level of possible trophic subsidy to epifauna. The amount of farm-derived waste that reaches the seafloor will be influenced by farm production and physical characteristics of the receiving environment.

Few studies have directly examined the possible effects of salmon farm-derived waste on epifauna in the context of OOA. As the effects from large farm-derived particles may be similar to those of sedimentation (e.g., smothering) and small particles similar to those of suspended sediment (e.g., clogging of filtering apparatus), previous literature surrounding the effects of sedimentation as well as organic enrichment on relevant taxa is presented.

4.1 Mobile deposit feeders

Known sensitivity to organic enrichment and sedimentation

Mobile deposit feeders are generally more tolerant of moderate to high levels of deposition when compared with sessile organisms, both in New Zealand and overseas. For example, urchins (Hall-Spencer et al. 2006, White et al. 2018) and snake stars (*Ophiopsammus* sp.; D. Elvines, personal observation, Govier & Bennett 2007) have been noted to increase in abundance in response to increased bio-deposition within the primary footprint of finfish farms. Scavenging fauna, including crabs, whelks, and starfish, are also positively correlated with enrichment. In Scotland, abundances 10- to 100-fold higher were recorded in fish farms located over maerl beds due to a build-up of organic matter in the benthos (Hall-Spencer et al. 2006), and in British Columbia, shrimp, squat lobster, and urchins were abundant within zones of high modelled carbon flux at aquaculture sites (Sutherland et al. 2018, Baltadakis et al. 2020, Sardenne et al. 2020). Predators such as *Coscinasterias muricata* (Crawford et al. 2001) and *Aterias rubens* (Dunlop et al. 2021) have been observed at higher densities near finfish farms, likely due to the drop-off in biofouling organisms.

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[24] Although research reports on salmon faecal pellet settlement rates are difficult to compare due to differences in fish size, methodologies, water characteristics, and diet (Reid et al. 2008).

While many mobile epifauna appear to be directly (deposit feeders due to increased organic content) or indirectly (predators due to increased prey abundance) attracted to organic enrichment, tolerance varies and is species-specific, with densities of some species peaking in areas exposed to moderate levels of organic enrichment. Echinoidea (urchins), surface-dwelling brittle stars (*Ophiocomina nigra*), and crabs of the family Lithodidae varied in distribution along the enrichment gradient from farms in Norway but were attracted to areas 250–1000 m from farms (Keeley et al. 2020, Markussen 2022). These patterns suggest that the organisms benefit from an increase in organic enrichment to a point, past which environmental conditions may be less favourable. Similarly, at the Clay Point and Te Pangu salmon farms in Tory Channel, snake stars have been observed to aggregate in a 'halo' around farms but are less common beneath the pens (McGrath et al. 2022a, 2022b). Other species, such as green-lipped mussel (*Perna canaliculus*), blue mussel (*Mytilus galloprovincialis*), and white striped anemones (*Anthothoe albocincta*), and to a lesser extent cushion stars (*Patiriella regularis*), solitary ascidians, and hermit crabs occur frequently beneath pen sites in soft-sediment habitats within Tory Channel. For mussels, this distribution may be a result of biofouling drop-off from pen nets; however, other organisms may exhibit different spatial distributions due to their differing response to farm-derived sedimentation and enrichment.

Known physiological responses

Sea stars and brittle stars are likely to benefit from increased levels of organic enrichment. In many situations these species are non-selective and feed on sediment but do not expend high amounts of energy on particle sorting prior to ingestion, as is common in bivalves (Ward & Shumway 2004). Organically enriched environments may support increased somatic growth and gonad development in brittle stars *Amphiura filiformis* and *A. chiajei* (Sköld & Gunnarsson 1996, Nilsson 1999).

Sea cucumbers are known to enhance mineralisation and nutrient cycling in organically enriched sediments (MacTavish et al. 2012) and have been suggested as suitable candidates to mitigate organic matter loading in marine sediments (Zamora et al. 2016). Juvenile *Australostichopus mollis* have exhibited capability to select for sediments higher in organic content in laboratory trials (Slater et al. 2011), while enhanced growth and survival has been observed for *Apostichopus japonicus* and *Parastichopus californicus* cultivated under finfish aquaculture sites (Hannah et al. 2013). Sea cucumbers have not only been shown to reduce organic load, but their feeding also results in horizontal redistribution and bioturbation of sediments (Uthicke & Karez 1999, Slater & Carton 2009, Yuan et al. 2016).

4.2   Sessile epifauna

Sessile epifauna are generally more sensitive to organic enrichment and other forms of anthropogenic disturbance when compared with mobile epifauna. Their sedentary nature means they are unable to move away from unfavourable environmental conditions and are susceptible to smothering. However, a sessile mode of life can also result from individuals lacking the need for mobility due to them being well adapted to tolerate variable environmental conditions. Sessile epifauna are most often suspension feeders, and changes in the quality of suspended particulates on which they feed can result in changes to the efficiency of feeding and alter energetic costs associated with obtaining organic matter. Due to their sessile nature, ultimate consequences are displacement in unfavourable conditions; however, sub-lethal effects can also occur before this.

Bivalves

Known sensitivity to organic enrichment and sedimentation

Bivalve beds are considered sensitive habitats[25], but little research has directly examined the effect of salmon farm deposition on these organisms. Based on information for other filter-feeding organisms, the general expectation is that fish farm sedimentation will enhance bivalve food supply at very low levels of deposition. If the bivalve populations are food limited, then small increases in organic matter in the particle size utilised by a species may have a beneficial effect (Jørgensen 1990, Bergvik et al. 2019). At higher levels of deposition, organic matter may reduce bivalve density or completely displace populations (through a variety of mechanisms). There will be a species-specific 'threshold' at which these effects will manifest. It is unknown whether chronic 'sub-lethal' effects (e.g., compromised reproduction and growth; White et al. 2016) will occur along the impact spectrum, but changes in food quality (e.g., filtering requirements, particle size of farm-derived organic material, food to sediment ratio, nutritional composition) are likely to be a key driver of such effects should they manifest. The threshold for such effects is unknown and is likely to be species-specific.

Known physiological responses

Recent studies suggest that possible chronic implications of farm-related trophic subsidies to marine organisms include reduced reproductive fitness in urchins (White et al. 2016, 2018), increased body condition in fish (Fernandez-Jover et al. 2007), decreased condition in crustaceans (Drolet et al. 2022) and bivalves (Both et al. 2012), and potential for enhanced growth (notably in the absence of an abundant natural food source) (Cranford et al. 2013, Irisarri et al. 2015). While the net effects of trophic subsidies provided by fish farms remains to be assessed, understanding the potential for fish waste assimilation and potential physiological implications on native species associated with fish farming is the first step in assessing broader ecosystem risks.

Mussels

Being predominantly organic and rich in lipids, farm-derived sedimentation represents a nutrient-rich potential food source to mussels. However, the nutritional quality of farm subsidies differs from marine seston, and the resulting shift in body composition from this trophic subsidy may have physiological consequences for some organisms (Fernandez-Jover et al. 2011, White et al. 2017), particularly if they are unable to compensate for the loss of important fatty acids in their diet.

Feeding behaviours in response to food availability have been investigated in a range of mussel species. Parameters used to investigate such behaviours are varied and have included clearance rates, filtration rates, and organic content of ingested matter. The feeding response of mussels to suspended organic matter is influenced by the organic content (quality) of seston (Bayne et al. 1993, Hawkins et al. 1998); for example, in *Mytilus edulis* feeding efficiency tends to exponentially decrease with decreasing proportions of organic/inorganic ratio of food (Bayne et al. 1989, Navarro et al. 1991). However, *M. edulis* also appears to be capable of acclimating to high concentrations of seston with low organic content, with net rates of assimilation increasing over a 12-day period of consistent exposure (Bayne et al. 1993).

Aquaculture has the potential to change particle size composition in seston, a factor mussels are known to be sensitive to. The ability of bivalves to be selective about the particles ingested will have consequences on their energy yield from the environment at different levels of food availability and quality (Navarro & Iglesias 1993). Velasco & Navarro (2002) found that *Mytilus edulis* and *M. chilensis* were able to selectively enrich the organic content of ingested matter relative to filtered matter by preferentially rejecting inorganic matter prior to ingestion. While increased suspended sediments can result in increased respiration in bivalves (Gibbs et al. 2005), the ability to selectively filter particles is a potential mechanism of resilience to changes in sedimentation or food quality due to finfish

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[25] Sensitivity is defined as the tolerance of a species or habitat to damage from an external factor, and the time taken for its subsequent recovery from damage sustained as a result of an external factor (MacDiarmid et al. 2013).

aquaculture. Previous research has described signs of stress and a significant drop in condition (biomass) in horse mussels in response to increased suspended sediment concentrations (Ellis et al. 2002). Observations of increased stress were attributed to the reduced quality of food intake and were associated with energy demands of increased filtering requirements from higher levels of non-food particulates.

Horse mussels have been identified as a key species at the proposed Blue Endeavour site and are known to exist in proximity to existing farms in the Marlborough Sounds, where they are subject to a low level of farm-related organic enrichment (less than 1 kg m^{-2} yr^{-1}) (Keeley et al. 2013). The persistence of horse mussels at dispersive sites where farms have been operating close to maximum consented limits for almost 10 years (such as Te Pangu Bay; McGrath et al. 2022b) indicates that they can tolerate at least a low level of exposure to farm-related organic deposition. Research is currently being undertaken to explore uptake and utilisation of salmon faeces by horse mussels to better understand potential effects of fish farm trophic subsidies to these ecosystem engineers (Elvines in prep.).

Oysters

Evidence suggests that oysters may be less equipped than mussels to select food particles. For example, the oyster *Pinctada margarifera* exhibits no differential retention of particles on the gill and little selective rejection through production of pseudofaeces (Hawkins et al. 1998). Similarly, the oyster *Crassostrea gigas* is not as efficient as mussels in the net selection of organic matter or in digesting and/or assimilating ingested organics (Hawkins et al. 1998), resulting in lower rates of net energy gain and slower growth. This physiological difference may result in a reduced capacity of oysters to tolerate reductions in seston quality when compared with some mussel species.

Oyster habitat is degraded through human activities such as pollution, poor water quality, fishing pressure, and increased suspended sediment loading. Increased levels of sedimentation that may arise from finfish farming could cause direct burial of oyster beds or reductions in filtration efficiency, respiration rates, and/or reproduction and settlement (Wilber & Clarke 2010). Juvenile and adult oysters may experience physiological impacts such as reductions in respiration and feeding rates. Increased levels of suspended sediments could reduce pumping rates in oysters (Loosanoff & Tommers 1948). Some information on physiological responses of oysters to suspended sediment exposures can be derived from responses to concentrations used in lab-based studies. For instance, Chu et al. (2002) found no significant effects on several physiological parameters in oysters exposed to up to 1 g L^{-1} of suspended clay particles for 40 days. However, sub-lethal effects on oysters have been noted at suspended sediment concentrations of 800 mg L^{-1}, with glycogen content reduced in oysters held in these conditions for 5 weeks (Volety et al. 2006). Burial of adult oysters in more than 5 cm of sediment was reported to cause mortality (Lunz Jr 1938, Galtsoff 1964, Rose 1973), while oysters buried in less than 1.25 cm of sediment could usually clear their gills of sediment via pumping (Dunnington 1968).

Brachiopods

Known sensitivity to organic enrichment and sedimentation

Brachiopod beds are considered 'sensitive' habitats, but there have been no studies examining their response to salmon farm deposition and very little information can be found on brachiopod sensitivity to organic enrichment or siltation. It is likely that effects from increased organic matter will manifest in a similar manner to bivalve species: enhanced food supply possibly at low levels of deposition, and reductions in density (through lower feeding efficiency and associated stress, or recruitment effects) and complete exclusion at higher levels of deposition.

Known physical responses

Very little is known about the physiological response of brachiopods to organic enrichment. Fossil records have indicated that brachiopods favour oligotrophic to mesotrophic environments at water depths of 60–90 m (García-Ramos et al. 2020). Local variations in water depth, temperature, oxygenation productivity, and organic enrichment are likely to be tolerated; however, sediment-stirred, current-swept offshore habitats are probably preferred. As with other filter feeders, we expect that

increases in organic enrichment may enhance food availability to a point, after which it may be associated with higher energy demands for the organism.

Bryozoans

Known sensitivity to organic enrichment and sedimentation

As bryozoans are slow-growing animals, recovery from widescale impact can take decades (Batson & Probert 2000). The widespread presence of bryozoans at some offshore sites in New Zealand with more dynamic particle flux suggests that some species within the phylum can tolerate environments with high current flow and relatively high levels of sediment deposition (Anderson et al. 2019, 2020). Areas of the Blue Endeavour site are particularly dynamic with regards to sediment movement, and the presence of small flexible and bushy bryozoans in these areas may indicate a tolerance of these species to high particle flux (Elvines et al. 2021). Furthermore, bryozoans have been observed settling and growing on artificial structures under and near salmon farms overseas (Angel et al. 2002, Israel et al. 2017) and on seaweeds exposed to salmon farm waste discharge (Haugland et al. 2021). Less is known about the susceptibility of frame-building bryozoans to organic enrichment and sedimentation from salmon farming. As for other filter feeders, the feeding efficiency of bryozoans is likely to be affected by changes in the quality or particle size composition of seston.

Known physical responses

Little information exists on the stress response of bryozoans to organic enrichment; however, general stress responses in this group have been associated with changes in colony morphology and skeleton rigidity (Casoli et al. 2017), growth rates, reproductive investments, and skeletal composition (Swezey et al. 2017). Antioxidant enzymes and detoxifying enzymes involved in the detoxification of environmental pollutants have also been proven as effective biomarkers of oxidative stress in the freshwater bryozoan *Lophopus crystallinus* (Elia et al. 2007).

Sponges

Known sensitivity to organic enrichment and sedimentation

Tolerance to organic enrichment and sedimentation likely varies among different species of sponge (see Laroche et al. 2022). However, it is known that some sponges are sensitive to anthropogenic stressors, including sedimentation due to the clogging of their aquiferous system (Sutherland et al. 2018). In Tory Channel, long-lived and potentially deposition-sensitive sponges have remained present in similar numbers over time; however, some changes in the appearance of these organisms have been noted since monitoring began. These changes included the appearance of filamentous red algae cover on some sponges and some changes in sponge size and presence/absence over time. Although these changes have been observed at both farm and reference sites in the Marlborough Sounds, they do highlight sponges as sensitive to environmental change and good candidates for studying organic enrichment effects.

Known physical responses

Stress responses in sponges are relatively well understood when compared with many other epifauna taxa identified at the proposed OOA sites in New Zealand. Sponges appear to be affected by sedimentation and organic enrichment; however, studies generally indicate the phylum possesses some tolerance to such conditions.

The clogging of inhalant canals and the aquiferous system by suspended sediments is likely the most important direct impact of sedimentation on sponges (Gerrodette & Flechsig 1979, Tompkins-MacDonald & Leys 2008). Unlike bivalves, sponges have little selective control over their filtering intake and are vulnerable to clogging by fine sediments. Such clogging can have detrimental effects on the acquisition of nutrients and feeding efficiency (Reiswig 1971, Gerrodette & Flechsig 1979). Sediment can also have an abrasive effect on sponge tissue and can negatively influence light-dependent larval settlement (Whalan et al. 2008). Reduction of pumping activity by sponges is a known response to increased sedimentation. Gerrodette & Flechsig (1979) found that an increase in sediment loading to 95 mg L^{-1} caused a reduction in the pumping rate of the tropical marine sponge *Verongia lacunosa* by

up to 41% over a short-term study. Variation in pumping rates can have consequences for respiration and food filtration by sponges (Gerrodette & Flechsig 1979).

Sedimentation also influences respiration rates in sponges. Lohrer et al. (2006) found oxygen consumption to be reduced in response to both low and high sediment treatment over a period of 3 weeks for the sponges *Aaptos rosacea* and *A. globosum*. Sediment grain size and composition, as well as concentration, have been reported as influencing the respiration response of sponges to sedimentation (Bannister et al. 2012, McGrath et al. 2017). Short-term changes in respiration by sponges may represent defence mechanisms, while longer-term responses may reflect a decline in sponge health (Bell et al. 2015). Some sponges produce mucus as a sediment-clearance mechanism; this process is poorly understood but expected to be energetically costly (McGrath et al. 2017).

Evidence of impaired pumping activity and respiration rates in sponges due to sedimentation suggests that feeding efficiency, growth, and reproduction will also be impaired by this stressor (Bell et al. 2015). Burial by settling sediment results in sponges expending more energy to cope with stress and less on growth and reproduction. Sponge symbiont productivity has also been shown to change in response to higher sedimentation, where a reduction in symbiont productivity can in turn reduce the amount of nutrients available to the sponge host for growth (Wilkinson & Vacelet 1979). Many sponges do possess adaptations that allow them to tolerate, or even thrive, in sedimented environments. Such adaptations may include spicule arrangements, gross morphology, mucus production, and flow reversal (McGrath et al. 2017, Dahihande & Thakur 2021).

Less is known about the response of sponges to organic enrichment when compared to sedimentation. Laroche et al. (2021) investigated the response of *Weberella bursa* to finfish farming in Norway and found the most obvious response was present in the fatty acid profile of the organism, with higher proportions of terrestrial fatty acids and long saturated and monosaturated fatty acids present in organisms at farm-impacted sites. The same study also indicated that potentially harmful elements such as sulphite, nitrite, and pesticides may be neutralised and degraded by a particularly enriched set of bacteria in the *W. bursa* microbiome, suggesting this species is more resistant to organic enrichment than anticipated. Apoptosis[26] has also been identified as a suitable biomarker to monitor unfavourable conditions in sponges (Wagner et al. 1998). Specifically, the apoptotic gene MA-3 is known to undergo increased expression in organisms within the Porifera phylum in response to certain stress stimuli. Similarly, Laroche et al. (2021) found that the sponge *W. bursa* had reduced ribosomal activity while having increased expression of genes controlling cell apoptosis when exposed to organically enriched environments. A second study by Laroche et al. (2022) found sponge respiration rates to show no clear patterns with distance from finfish farm pens; however, farming activities appeared to have a species-specific effect on microbiome diversity, with most pathways for metabolic function negatively associated with fish farming. Several additional studies have also employed lipids, fatty acids (Bennett & Elvines 2018) and gene expression (Webster et al. 2013, Guzman & Conaco 2016) to study the stress response of sponges.

Tubeworms

Known sensitivity to organic enrichment and sedimentation

Little is known about the sensitivity or response of tubeworms to sedimentation or organic enrichment. Serpulidae worms have a specialised operculum that blocks the entrance of their tubes when they withdraw into them. This behaviour may afford some species higher tolerance to increased sedimentation as feeding appendages can be kept clear of settling particulates. However, efficiency of feeding may in turn be diminished if suitable windows for feeding are reduced for the species. Ongoing settlement by larvae is usually enhanced by chemical and physical cues provided by the presence of live adult worms (Brougham 1984, Gibson et al. 2001). However, increased sedimentation may reduce the ability of tubeworm larvae to settle and, in turn, the expansion of these habitat-forming species.

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[26] Biochemical events that lead to characteristic cell changes and, ultimately, cell death.

Little information exists on the stress response of tubeworms to organic enrichment. We may, however, assume the physiological stress response of this group will be similar to that of polychaetes in general. While polychaetes are a diverse taxonomic grouping and possess a wide range of sensitivities/tolerances to organic enrichment, their physiological responses to enrichment and approaches used to detect these are relevant to this review. It should be noted that differences likely exist between the response of sediment-dwelling polychaetes, which are exposed to changes in sediment chemistry and organics, and those that are epifaunal.

Carregosa et al. (2014) investigated the physiological and biochemical response of the polychaete *Diopatra neapolitana* to organic enrichment and found such conditions reduced the capacity of these species to regenerate their body and led to increased glycogen and protein levels in organism tissue. Oxidative stress also increased with increasing organic enrichment. Specifically, lipid peroxidation (an indicator of cellular damage) oxidised glutathione and glutathione S-transferase activity (indicators of redox status), and antioxidant enzymes catalase and superoxide dismutase activity (indicators of antioxidant enzyme activities) increased in organisms from areas with higher organic enrichment. Similarly, Bocchetti et al. (2004) concluded that analysis of antioxidant defences in polychaetes can reveal important information about the ability of species to adapt to stressful environmental conditions and biological disturbances.

Cnidarians (anemones, cup corals, hydroids)

Known sensitivity to organic enrichment and sedimentation

Cnidarians are a diverse phylum, and so sensitivity to organic enrichment and sedimentation is likely to be highly varied across the grouping. It has been suggested that increased levels of organic enrichment and sedimentation may benefit anemones through reduction in competition from other species and enhancement of photosynthetic efficiency (Liu et al. 2015). Plumose anemones, for example, have been found to be more numerous proximal to aquaculture net pens than reference sites (Sutherland et al. 2016, 2018). Little is reported on the sensitivity of hydroids to organic enrichment. However, this group has been used as a potential indicator of environmental disturbance (Cullain et al. 2018), suggesting some species of hydroid may be somewhat tolerant to organic loading. Variable responses by different hydroid species to increases in organic content have been noted (Perry 2018).

Known physical responses

Anemones

Nutrient enrichment may result in increased food availability and hence increased growth for anemones. Alternatively, excess enrichment may result in bacterial or algal mats that smother the benthos to the detriment of the species. Photosynthetic efficiencies of symbionts in anemones may be improved in response to sediment loading and organic loading. Liu et al (2015) reported that the maximum yield of photosystems was significantly higher in anemones from a sediment-enriched mesocosm compared with controls over a 1-month study period, and the rate of asexual reproduction increased under elevated nutrient exposure. Sutherland et al. (2018) observed plumose anemones to respond positively to an increase in depositional carbon flux, with densities peaking between a flux of 2 and 4 g C m^{-2} day^{-1} and the species tolerant of carbon flux above 5 g C m^{-2} day^{-1}.

Hydroids

While information specific to the stress response of hydroids to organic enrichment is lacking, reduced growth, low total protein content, high metabolic rates, and high protein carbonyl are considered to be indicative of stress in this taxonomic grouping (Eder et al. 2018).

Ascidians (solitary and colonial)

Known sensitivity to organic enrichment and sedimentation

Evidence suggests that ascidians are tolerant of nutrient and organic enrichment (Readman 2016, Readman & Hiscock 2016), with enriched sediments leading to increased ascidian recruitment on nearby hard substrate (Lawes et al. 2017). However, colonial ascidians may respond negatively to metal-contaminated sediments that are possibly caused by high-production finfish farming operations (Lawes et al. 2017).

Known physical responses

Little information exists on the physiological stress response of ascidians to organic enrichment; however, many ascidians do appear to tolerate or even favour increased levels of enrichment (Readman 2016, Readman & Hiscock 2016). Transcriptomics and proteomics appear to be common approaches used to understand stress responses in ascidians and for developing biomarkers of environmental stressors in these species (Azumi et al. 2004, Fujikawa et al. 2010, Kuplik et al. 2019). For example, the ascidian *Botryllus schlosseri* was concluded to be a good indicator of the ecological status of a lagoon in Italy, where the transcription of genes for antioxidant protein activity and the activity of enzymes involved in the oxidative stress response changed in response to declining environmental conditions (Tasselli et al. 2017). Additional physiological and phenotypic responses to stress have also been noted for ascidians; signs of stress in *Botrylloides nigrum* and *Botryllus planus* include higher mortality, lower heart rates, and limited asexual reproduction (Dijkstra & Simkanin 2016).

5. FRAMEWORK FOR SPECIES SELECTION

Using information from the reviews undertaken in Sections 2 and 3, we developed a comprehensive framework that aims to provide suitability criteria to assist in identifying key species likely to occur in environments that may be affected by OOA (Figure 2). The framework considers how much information exists for the habitats of interest, the scale of interest, the assemblage composition (for example, frequency of occurrence, density, biomass, and commonality), ecological function, sensitivity to enrichment, logistical considerations for execution of experimental trials, and value to iwi[27]. The framework also provides a guide for the selection of potential stress indicators[28] that should be considered in tandem with species selection. Flexibility is allowed for in its application; it can be used as a general guide or checklist and applied to any circumstance where identifying species of interest is of use.

The framework advises what factors should be considered when identifying species that would be best suited to achieve the defined objective. We used the framework as a guide for developing a comprehensive matrix that allows a quantitative approach to evaluate which of the taxa present within the proposed open ocean sites/Tory Channel would be most appropriate for use in Objective 2 experiments. Matrix development is discussed in Section 5.1, application of the matrix to the list of possible taxa is discussed in Section 5.2, results from species scoring is detailed in Section 5.3, and discussion of the outcomes of this approach can be found in Section 5.4.

5.1 Methodology for development of species selection matrix

To assist in identifying key species on which to investigate the effects of organic enrichment from OOA, we used a detailed matrix that allowed for a quantitative application of the suitability criteria presented in the framework (Appendix 2). A numerical score was assigned to each taxon in the matrix, and each

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[27] Specifically focusing on taxa that were identified as important to customary fishing and practices, fishing resources or fisheries, as identified in the application/consenting process for the Blue Endeavour and Hananui proposals. This includes taxa that are either directly important kaimoana, mātaitai, and mahinga kai, or that indirectly support these resources.

[28] A review of potential indicators of stress and/or physiological change in epifauna is provided in Appendix 3.

was ranked for further consideration. The appropriate potential stress indicators were then considered for the highest-scoring taxa. While the primary goal was to identify species for inclusion in Objective 2 laboratory experiments, species were also considered for field translocation studies and opportunistic sampling at existing highly dispersive farms.

Each taxon was scored for 15 factors across five broad categories (Appendix 2, Table A2.1), derived from the framework presented in Figure 2. Existing benthic assessments for proposed OOA in New Zealand were reviewed (Sections 2.2 to 2.4), along with information for soft-sediment and reef communities present at existing dispersive inshore salmon farms (Section 2.5). The cultural importance of each taxon from a mātauranga Māori perspective was also considered[29]. This information was used to identify species present at locations relevant to OOA and to characterise their frequency of occurrence, when possible.

Categories were: preliminary data/existing information, biotic assessment, ecological function, sensitivity to salmon farming, and value to iwi. Generally, very little information exists on the response of the species listed in Table 1 to sedimentation and organic enrichment from salmon farming. However, useful information on species' responses can be drawn from related taxa or from their physiological response to other environmental factors.

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[29] As identified in the application/consenting process for the Blue Endeavour and Hananui proposals.

OBJECTIVES
- Create a list of candidate species on which to investigate the effects of organic enrichment from open ocean aquaculture on epifauna
- Identify appropriate techniques for measuring physiological, molecular and/or biochemical responses to enrichment

PRELIMINARY DATA/EXISTING INFO
- Species presence within areas of interest:
  - Assessment of effects for proposed Blue Endeavour, Hananui, and Southland OOA sites
  - Reef and benthic monitoring reports for existing inshore salmon farms
  - Raw data from Blue Endeavour and Hananui habitat assessments
- Information on species' physiological response in peer-reviewed literature

BIOTIC ASSESSMENT (presence, sensitivity, commonality)
- Factors to be considered under biotic assessment include:
  - Species presence in biogenic habitat types
  - Sensitive marine benthic species and habitats, as defined by MacDiarmid et al. (2013)
  - Frequency of occurrence

ECOLOGICAL FUNCTION
- Ecosystem services: greater weighting will be given to species that are considered ecosystem engineers, being resource or habitat providers to other organisms or providing access to resources through habitat modification i.e. filtration, assimilation of primary production, nutrient cycling
- Greater weighting will be given to species known to act as ecological indicators of environmental change (keystone species)

SENSITIVITY TO ORGANIC ENRICHMENT AND SEDIMENTATION
- Little is known about the sensitivity of most species to organic enrichment and sedimentation; however, species sensitivity will increase the weighting of this category, where information is available
- Species responses may be expected to be either positive (e.g. enhanced growth through greater food supply), or negative (reduced food quality, increased sedimentation, higher nutrient load)

VALUE TO IWI
- Cultural assessments of Hananui applications by iwi groups across New Zealand and submissions by Ngati Kuia, Te Runanga o Ngati Kuia, and Te Ohu Kaimoana on the Blue Endeavour application
- Identification of kaimoana or taonga species (or those important to mahinga kai)
- Consideration of the value of habitat forming species and ecosystem service providers to supporting key kai moana and taonga species e.g. mullock provides important blue cod habitat
- Extends far beyond species selection and will guide the development of broader outcomes from the project

OTHER LOGISTICAL CONSIDERATIONS
- It must be feasible to move species for laboratory and translocation trials from their place of origin without causing long-term stress or damage to the organism
- For species included in tank trials, it must be possible to provide an environment in which the organism can carry out normal physiological functions and behaviors
- Species need to be present at densities high enough for opportunistic sampling or for an adequate sample size to be reliably collected for lab trials or field translocations

SPECIES MEETING SELECTION CRITERIA

Laboratory experiment   Opportunistic field sampling   Field translocations

INDICATOR SELECTION
- Indicators must consider species physiology, sampling logistics, timing of responses, methodology and experimental design

SPECIFIC CRITERIA FOR INDICATOR SELECTION
- Indicators should be measurable in a laboratory environment
- Indicators should have an associated method appropriate for species physiology, body size, and fragility
- Some analytical approaches will ideally capture change through time (through conservative sampling)
- The indicators will be expected to detect a change over the experimental time period (3-6 months)
- Approaches should be collaborative in that, in theory, they offer a way to link physiological changes to broader ecosystem effects
- Analytical approaches should be sensitive enough (i.e. low enough replicate variability) to detect change through analysis of 5 or fewer replicates
- Approaches should consider time and funding available and how this fits within the project budget

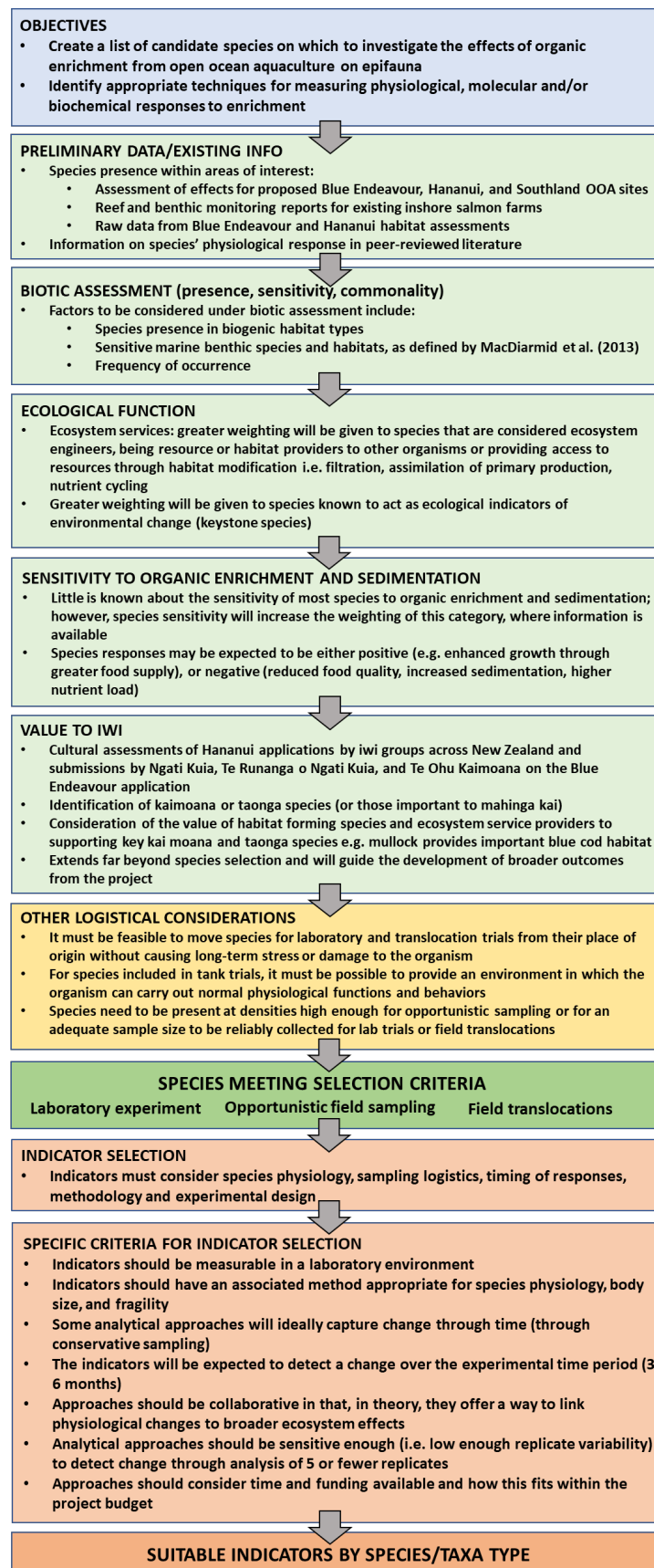**SUITABLE INDICATORS BY SPECIES/TAXA TYPE**

Figure 2: Overview of framework developed for the selection of species for inclusion in laboratory and field trials to investigate the physiological response of epifauna to organic enrichment and sedimentation from salmon farming.

5.2　Scoring

Scores were developed for all 15 factors for all the taxa identified at the proposed Blue Endeavour, Hananui, and Project South open ocean sites, and at existing Tory Channel farm sites (Section 2, Appendix 2, Table 2). Higher scores for taxa and functional groups equated to a greater likelihood of being selected for laboratory and field experiments. Some factors were scored 'yes' (score = 1) or 'no' (score = 0). Where a binary assignment was not considered appropriate, the factor was allocated a score between 0 and 1 (see Table 2 for the potential scores allocated for each factor). Cells were left blank where insufficient information existed to allocate a score. Scores were added, and taxa with the highest score were identified as more relevant to OOA environments and therefore more likely to be chosen for use in Objective 2 (laboratory/field experiments).

After allocating scores for taxa identified in Table 1, taxa from the open ocean sites were allocated to functional groups, and these groups were scored. Typically, scores for functional groups were calculated as the average for each taxon per factor for ranked factors (calculation of averages considered blanks cells to be equivalent to no data as opposed to zero values), while for binary factors functional groups were scored as Y (=1) if any taxa were present within the group. However, in some cases, where information was lacking for species groups within the functional group, best professional judgement was used to allocate a score for the functional group. Best professional judgement considers knowledge on the functional group more broadly and can be supported by published literature or through personal communications. For example, very little information is published on any of the erect species of sponges observed at sites of interest; however, significant information exists on the response of other sponge species to sedimentation and, to a lesser extent, organic enrichment. Therefore, the broader functional group 'erect sponges' is allocated a score for the factor 'Information on species physiological response', despite each individual species lacking sufficient information to allocate a score. Where best professional judgement was used to allocate a score to a functional group, scores are highlighted pink in Table A2.1 of Appendix 2.

Equal score weighting was given to each factor included in the matrix and an overall total score was calculated for each taxon, as well as functional groups, by adding the scores for all 15 factors across each row (see Table A2.1). Taxa and functional groups were then ranked according to their overall score. Logistical and geographical constraints were then used to further refine the candidate species following assessment of taxa and functional group rankings. Scoring for frequency of occurrence differed between Blue Endeavour and Hananui due to differences in the methods used to characterise habitats at each location. At Blue Endeavour, frequency of occurrence is defined as % of time viewed along transect, and, at Hananui, frequency of occurrence is defined as % cover or through individual counts at each station. The ranges for percentage cover described in the respective assessments of environmental effects for the two sites also differed slightly, meaning there is some inconsistency in the definitions for maximum frequency of occurrence between the sites (see Table 2).

Table 2: Definition of values used to assign factor scores for taxa and functional groups in selection framework. (Continued on next page)

Factor	Score	Definition
Present at Blue Endeavour	Y/N	
Present at Hananui	Y/N	
Information on species physiological response	Blank cell	Insufficient information / unknown
	0.25	Some information on physiological responses, but nothing specific to sedimentation or organic enrichment
	0.5	Some information on physiological responses specific to sedimentation[30] or organic enrichment[31], but nothing specific to aquaculture
	0.75	Some information on physiological responses specific to sedimentation or organic enrichment from aquaculture
	1	Information on physiological responses specific to sedimentation or organic enrichment from aquaculture in New Zealand
Present in biogenic and other hard-substrate habitat types	Y/N	
Is percentage cover/density considered sensitive benthic habitat at each OOA site?	Blank cell	Not classified as a sensitive environment[32] by MacDiarmid et al. (2013)
	0.5	Not classified as sensitive by MacDiarmid et al. (2013), but present at high enough densities to make an important contribution to some aspect of ecosystem functioning
	1	Meets requirements for classification as sensitive habitat by MacDiarmid et al. (2013)
Maximum frequency of occurrence[33]	Blank cell	Insufficient information / unknown
	0.05	< 5% of the time at Blue Endeavour; isolated individuals at Hananui
	0.2	5–20% of the time at Blue Endeavour; 5–10% coverage or 2–3 individuals in close proximity at Hananui
	0.5	20–50% of the time at Blue Endeavour; 10–40% coverage or several individuals in close proximity at Hananui
	0.8	50–80% of the time at Blue Endeavour; >40% coverage or dense aggregations at Hananui
	1	80–100% of the time at Blue Endeavour
Habitat-forming species	Y/N	
Filter feeders	Y/N	
Bioturbators	Y/N	
Nutrient and organic matter cycling	Blank cell	Insufficient information/unknown
	0.33	Minimal contribution, low biomass, not a primary consumer
	0.66	Moderate contribution, may have high biomass, or a key position in the food web
	1	High contribution, large biomass, high feeding rate, primary consumer
Indicator species	Y/N	
	Blank cell	Insufficient information / unknown
	0.33	Tolerant

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[30] The process of allowing particles in suspension to settle out of the fluid in which they are entrained and come to rest.

[31] The discharge of organic matter or nutrients, which can lead to the reduction or disappearance of sensitive organisms.

[32] Sensitivity is defined as the tolerance of a species or habitat to damage from an external factor, and the time taken for its subsequent recovery from damage sustained as a result of an external factor (MacDiarmid et al. 2013).

[33] See caption to Table 1 for further explanation on how scores are defined. Max. freq. of occurrence scores are allocated based on the site/s at which species have the highest frequency of occurrence.

Factor	Score	Definition
Susceptibility to organic enrichment and sedimentation	0.66	Moderately tolerant/sensitive
	1	Sensitive
Is the species taonga, kaimoana or a mahinga kai resource?	Y/N	
Does the species support taonga species or kaimoana and mahinga kai resources?	0.33	Not significantly important for kaimoana and/or taonga species
	0.66	Does not provide crucial habitat but provides other ecosystem services to support kaimoana and/or taonga species
	1	Expected to provide crucial habitat for kaimoana and / or taonga species

Once the taxa were ranked according to their individual scores (Appendix 2) they were evaluated based on the logistical criteria below. Those unlikely to meet the criteria were excluded (see Table 4).

- It must be feasible to move taxa for laboratory and translocation trials from their place of origin without causing long-term stress or damage to the organism.
- For taxa included in laboratory experiments, it must be possible to provide an environment in which the organism can carry out normal physiological functions and behaviours.
- Taxa need to be present at sufficient densities to provide an adequate sample size for lab trials and experimental trials (for example, opportunistic sampling and field translocations).

5.3   Results

The species selection matrix scored each factor presented in the framework (see Appendix 2) for relevant functional groups (Table 3) and all taxa (Table 4) present within the proposed open ocean sites and Tory Channel.

Table 3:   Overall scores for functional groups identified at the proposed open ocean aquacultures sites and existing farms located in dispersive environments in Tory Channel. Functional groups are ranked from highest to lowest scoring.

Functional group	Overall score from matrix
Suspension-feeding bivalves (epifaunal)	11.76
Suspension-feeding brachiopods	10.28
Sessile suspension feeders – sponge erect	9.91
Sessile suspension feeders – habitat-building tubeworms	8.73
Mobile deposit feeders	8.49
Sessile suspension feeders – bryozoans erect	8.22
Sessile suspension feeders – sponge encrusting	8.10
Mobile grazers	7.37
Sessile suspension feeders – hydroids	6.97
Sessile suspension feeders – anemones	6.91
Sessile suspension feeders – colonial ascidian	6.87
Mobile scavengers/predators	6.64
Sessile suspension feeders – bryozoans bushy and flexible	6.58
Mobile suspension feeder	6.20
Sessile suspension feeders – solitary ascidian	6.04
Sessile suspension feeders – coral	5.63
Sessile suspension feeders – bryozoans encrusting	5.42
Sessile suspension feeders – sea cucumber	5.28
Sessile suspension feeders – non-habitat-building tubeworms	5.12
Sessile suspension feeders – crinoid	5.12
Mobile undefined	4.53

Table 4: Overall scores for taxa identified at the proposed open ocean aquaculture sites and existing farms located in dispersive environments in Tory Channel. Taxa are ranked from highest to lowest scoring. Species that were excluded from selection due to logistical constraints have been eliminated (struck through). (Continued on next page)

Taxon	Overall score from matrix
Horse mussels (*Atrina zelandica*)	11.08
Brachiopods (Blue Endeavour: *Notosaria nigricans*, *Magasella sanguinea*, *Calloria inconspicua*), (Hananui: either *Neothyris lenticularis* or *Magasella sanguinea*)	10.28
Erect bryozoans (mainly *Cinctipora elegans* at Hananui)	9.22
~~Tubeworm mounts/ calcareous tubeworms from family Serpulidae (*Galeolaria hystrix* at Hananui)~~	9.05
Scallops (*Pecten novaezelandiae*)	8.95
Dredge oysters (*Ostrea chilensis*)	8.78
Sea cucumber (*Australostichopus mollis*)	8.74
Green-lipped mussel (*Perna canaliculus*)	7.92
Brittle stars (*Ophiopsammus maculata*)	7.24
Kina (*Evechinus chloroticus*)	7.07
~~Feather hydroid (*Aglaopheniidae*) / hydroid tree~~	6.97
Blue mussel (*Mytilus galloprovincialis*)	6.92
Bryozoans (bushy and flexible branching forms, *Orthoscuticella innominata*, *Hornera foliacea* and possibly *Hornera robusta* at Hananui)	6.58
~~Sponge (*Dactylia varia*)~~	6.30
~~Sponge (*Iophon minor*)~~	6.30
Turret shell (*Maoricolpus*)	6.20
~~Colonial ascidians~~	6.07
Crab, nondescript (*Brachyura*)	5.88
~~Giant tubeworms~~	5.85
~~Orange sponge (*Crella encrustans*)~~	5.80
Finger sponge	5.70
11-armed sea star (*Coscinarius muricata*)	5.62
Bearded horse mussels (*Modiolus areolatus*)	5.45
Cushion stars (*Patiriella regularis*)	5.45
Massive encrusting bryozoan (*Celleporaria agglutinans*, encrusting brown at Tory Channel)	5.42
~~Tube-dwelling anemones (Ceriantharia)~~	5.33
~~Encrusting sponges~~	5.30
White striped anemone (*Athothoe albocincta*)	5.28
~~Burrowing sea cucumber~~	5.28
~~Talochlamys~~	5.12
~~Fan worm (Sabellidae)~~	5.12
~~Crinoids~~	5.12
Solitary ascidian (including *Styela clava*)	4.97
~~Large parchment worm~~	4.92
~~Colonial ascidians (*Botrylloides* sp.)~~	4.92
Anemones – undefined	4.83
Orange erect sponge	4.70
~~Grey cup sponges (*Ecionemia alata*)~~	4.67
Circular saw-shell (*Astraea heliotropium*)	4.53
~~Solitary cup coral~~	4.38
~~Giant wandering anemone (*Phlyctenactis tuberculosa*)~~	4.30
~~Tubeworm – Chaetopteridae~~	4.30
~~Yellow encrusting sponge~~	4.20
~~Sea tulip (*Pyura pachydermatina*)~~	4.12
Fan shells (Pectinidae)	3.92
~~Colonial ascidians (*Botryllus* sp.)~~	3.92
~~Colonial ascidians (*Eudistoma circumvallatum*)~~	3.92
Hermit crab (*Pagurus novizealandiae*)	3.83
~~Nudibranch (Pleurobranchaea)~~	3.82
7-armed sea star (*Astrostole scabra*)	3.62

Taxon	Overall score from matrix
Hairy seaweed crab (*Notomithrax ursus*)	3.58
Penion sp.	3.47
Long-armed Sea star (possibly *Cosmasterias dyscrita*)	3.45
Calliostoma sp.	3.42
~~Colonial stony coral~~	3.33
~~Blue encrusting sponge~~	3.00
~~Yellow colonial ascidian (*Aplidium phortax*)~~	2.92
Cat's eye snail (*Lunella smaragda*)	2.42
Cook's turban (*Cookia sulcata*)	2.42
~~Limpet (*Cellana stellifera*)~~	2.42
~~Limpet (*Siphonaria* sp.)~~	2.42
Trochus sp.	2.42
Apricot star (*Sclerasterias mollis*)	2.25
Duck's bill limpet (*Scutus breviculus*)	2.08

5.4 Summary of species suitability assessment for Objective 2 experimental trials

The top 10 ranking species identified from the species selection matrix were (in order): horse mussels (*Atrina zelandica*), brachiopods, tubeworm mounds/calcareous tubeworms, erect bryozoans, sea cucumbers (*Australostichopus mollis*), scallops (*Pecten novaezelandiae*), dredge oysters (*Ostrea chilensis*), brittle stars (*Ophiopsammus maculata*), green-lipped mussels (*Perna canaliculus*), and feather hydroids/hydroid trees. The top-ranking functional groups were primarily represented by these high-ranking taxa, the exception being suspension-feeding erect sponges. This is due to the lack of information on the species observed within the areas of interest. However, erect sponges in general are known to be a good indicator of change in the surrounding environment and their response to sedimentation, and to a lesser extent organic enrichment, has been relatively well studied. We therefore included them as appropriate candidate species for Objective 2 experimental trials.

Horse mussels

Presence in areas of interest: Horse mussels are present at Blue Endeavour and sites in Tory Channel and in the wider Marlborough Sounds and Tasman Bay/Golden Bay area. Anderson et al. (2020) reported often seeing horse mussels heavily fouled by reef-building bryozoans in Queen Charlotte Sound. Sparsely distributed horse mussels have also been observed in mid to deep slopes at Perano Shoal in Queen Charlotte Sound, and at Separation Point in Port Underwood. Horse mussels were not present at Hananui; however, bearded horse mussels (*Modiolus areolatus*) were found.
Biotic assessment: Live horse mussels form a key component of HMBB at the Blue Endeavour site, and non-living horse mussel shells are an important component of the shell debris present in areas of patch reef habitat, providing hard substrate on which community succession may occur. Horse mussels were of ecological importance at Blue Endeavour, where their presence was at times abundant in HMBB habitat and rare to occasional in patch reef habitat.
Ecological function: The species plays a key role in supporting higher relative diversity in biogenic habitat at Blue Endeavour. The larvae of many benthic organisms, both encrusting and erect, require a hard substrate on which to settle. The large surface area provided by horse mussel shells among large areas of relatively barren sediment is therefore an important feature of this offshore area. As filter feeders, horse mussels also play an important role in the provision of nutrients and organic matter to benthic food webs and are known to be good indicators of environmental change.
Sensitivity to organic enrichment and sedimentation: Previous research on horse mussels and observation of horse mussels beneath existing farm sites in New Zealand suggests the species are tolerant to some degree of organic enrichment. However, little is known about how and at what levels of exposure sub-lethal effects will manifest.
Value to iwi: Yes. Not a kaimoana or taonga species, but beds of horse mussels are recognised as important habitat for the settlement of larvae for a range of species, and as nursery grounds for juvenile fish.

Logistical considerations: Horse mussels are typically robust to transport and house in laboratory conditions; however, care should be taken not to damage shells while removing animals from sediment. Horse mussels have been previously housed at the Cawthron Aquaculture Park (CAP), with excellent survival rates. Horse mussels have also been well studied by the physiology team at CAP, meaning the chance of obtaining meaningful results from this species is high.

Brachiopods

Presence in areas of interest: Brachiopods have been observed in high densities at the proposed Blue Endeavour site and in lower densities at the proposed Hananui site. While they have not been observed at existing farm sites in the Marlborough Sounds, several species have been recorded from the Marlborough Sounds, including four common endemic species (*Magasella sanguinea*, *Calloria inconspicua*, *Neothyris lenticularis*, and *Liothyrella neozelanica*) (see Davidson & Richards 2015, Davidson et al. 2011, Morrisey et al. 2015). While brachiopods are widely distributed in the Marlborough Sounds, their occurrence within sites is localised. Anderson et al. (2020) found that the middle of Queen Charlotte Sound and the deep areas of Cook Strait had the highest percentage of brachiopod sites, while Tory Channel supported the fewest. Although the average depth of brachiopods was typically greater than 25 m at most sites, they were seen as shallow as 10–15 m, indicating that collection of organisms by divers would be possible.

Biotic assessment: Live brachiopods form a key component of HMBB at the Blue Endeavour site and are considered to be ecologically important within this area. Although occasionally present at Hananui, brachiopod densities were not considered to be bed-forming, although they did warrant protection from potentially adverse activity.

Ecological function: Like horse mussels, brachiopods play a key role in supporting increased diversity in biogenic habitat at Blue Endeavour. They act as a substrate on which larvae can settle and have a role in bentho-pelagic coupling.

Sensitivity to organic enrichment and sedimentation: Very little is known about how brachiopods tolerate organic enrichment and sedimentation. Inclusion of a species from this understudied group would fill an important knowledge gap with regards to the potential impacts of OOA on epifauna.

Value to iwi: Yes. Not a kaimoana or taonga species, but bed-forming densities are recognised to provide important habitat for the settlement of larvae, and as nursery grounds for juvenile fish.

Logistical considerations: Collection of brachiopods will involve removal of the hard substrate to which they are attached and should be straightforward if the animals are attached to smaller cobbles or are cemented to soft sediment. Brachiopods have been observed to be robust to translocation and lab conditions (Williams 2022).

Bryozoans

Presence in areas of interest: Bryozoans were present at both the Blue Endeavour and Hananui sites. Sparsely distributed bushy and flexible bryozoans were most common at Blue Endeavour, although frame-building species were rare or uncommon at the site. Erect/frame-building bryozoans were abundant at Hananui, as were branching forms; encrusting bryozoans were observed but their frequency was not noted. A mix of erect and encrusting bryozoan species has been noted at existing Tory Channel sites. Gordon et al. (2009) noted that several reef-forming species of bryozoan are known to occur within the broader Marlborough Sounds area. These include *Celleporaria agglutinans*, *Cinctipora elegans*, *Galeopsis porellanicus*, *Diaperoecia purpurascens*, and *Hornera robusta*. Anderson et al. (2020) found bryozoan reef cover to be highest at the entrance to Queen Charlotte Sound, particularly on the eastern side of the channel at depths ranging from 13 m to 68 m.

Biotic assessment: Bryozoans form a key component of BS and BBT habitats at Hananui, with *Celleporaria agglutinans*, *Cinctipora elegans*, and *Orthoscuticella innominata* being of particular importance and forming bryozoan beds within this area of interest. Bryozoans of varying forms ranged from rare to abundant across varying habitat types at Blue Endeavour but were not observed to form bryozoan beds, as defined by MacDiarmid et al. (2013).

Ecological function: Rigid, frame-building bryozoans can form extensive kilometres-long reef structures that support the settlement and growth of diverse communities, including sponges, ascidians, and bivalves.

Sensitivity to organic enrichment and sedimentation: As filter feeders it is expected this group may respond to changes in the quantity or quality of seston in the water column; however, the level of tolerance they will exhibit to such conditions is unknown.

Value to iwi: Yes. Not a kaimoana or taonga species but recognised for forming critical habitat for high-value species such as blue cod and crayfish at the Hananui site and within the Marlborough Sounds.

Logistical considerations: Bryozoans are likely to be the most challenging species currently proposed for selection in laboratory and/or field-based trials. Challenges may arise during collection, given they are often located near the limit of scuba depths. Removing the organisms from the substrate to which colonies are attached may also be challenging; however, it may be possible to collect a piece of the hard substrate along with the bryozoan or reattach the organisms to hard substrate in the laboratory or field using cable ties. Given bryozoans are colonies of zooids, damage to a part of the structure is unlikely to compromise all organisms that make up the structure.

Sea cucumbers

Presence in areas of interest: The sea cucumber *Australostichopus mollis* is typically sparsely distributed (rare to occasional) across all biogenic habitat types. However, the species is also found in areas of soft sediment, both at offshore sites and at existing inshore farm sites. Anderson et al. (2020) noted the species to be prevalent across most of the Marlborough Sounds and to be most common on sediment slopes and rocky reefs in depths ranging from 2 m to > 100 m. Research has been conducted on the role of sea cucumbers in recycling farm-derived organic waste and on the potential for use of these deposit feeders in multi-trophic aquaculture (Zamora et al. 2016).

Biotic assessment: *Australostichopus mollis* was not noted as a potentially sensitive benthic species in the review by MacDiarmid et al. (2013) and was not reported as abundant within the areas of interest. However, the species is notable for its widespread distribution.

Ecological function: Sea cucumbers are distinct from the other candidate species presented here as they are deposit feeders, capable of extracting organic matter from sediment and potentially reducing organic loading, and act as bioturbators.

Sensitivity to organic enrichment and sedimentation: When compared to filter feeders, mobile deposit feeders are considered to be more tolerant of sedimentation and organic enrichment. These organisms are generalists, making use of all organic matter available within the sediment. They also have sorting mechanisms, allowing for rejection of inorganic material during digestion. Finally, their mobility allows them to move away from unfavourable conditions and/or towards favourable conditions, meaning sea cucumbers may be found to aggregate in areas with optimum levels of depositional flux and residual solids.

Value to iwi: Yes. While not a kaimoana or taonga species, sea cucumbers can be prey items to large fish. They also help to support a healthy ecosystem by making carbon and nutrients available to higher-trophic-level species and reducing organic loading of sediments which can reduce the likelihood of anaerobic conditions developing.

Logistical considerations: Few logistical challenges are anticipated in collecting and transporting sea cucumbers. It is unlikely these organisms will be used in field translocations given they are already distributed across depositional gradients from existing farms. As with scallops, replicating an environment in which treatment conditions can be linked to levels of depositional flux observed in the field may be more challenging, given this species will require a sediment layer on which to feed.

The mobile nature of sea cucumbers presents some challenges in terms of assessing the potential enrichment these organisms are exposed to. However, previous research indicates that sea cucumbers are attracted to sediments with higher organic loading (Zhang et al. 1990, Zamora & Jeffs 2011, Zamora et al. 2016) and therefore we may expect that populations of sea cucumbers remain resident around operational salmon farms. Their mobile nature, however, means there will be a greater range of uncertainty when estimating the levels of residual solids or depositional flux an organism has been exposed to.

Scallops

Presence in areas of interest: Scallops (*Pecten novaezelandiae*) were occasionally noted across BBT, BS, and S habitat types at Hananui and rarely found in LDE-CS at Blue Endeavour. They have also been observed at existing inshore salmon farm sites. Scallops have also been noted to be moderately abundant at the Perano Shoal site among *Galeolaria* tubeworm mounds in Pelorus Sound and within Kaipākirikiri Bay, Grove Arm, and mid-slope shell debris fields in Queen Charlotte Sound (Anderson et al. 2020).

Biotic assessment: While scallops are noted only as rare to occasional at the Blue Endeavour and Hananui sites, by feeding at the sediment–water column interface they represent a different feeding strategy when compared with horse mussels, green-lipped mussels, and brachiopods, which feed at a more elevated position in the water column. This species was not reported at sufficient densities to constitute sensitive benthic habitats at any of the OOA sites of interest.

Ecological function: As filter feeders, scallops have an important role in capturing nutrients and organic matter from pelagic systems and making it available for benthic food webs.

Sensitivity to organic enrichment and sedimentation: Studies from overseas suggest that the species may be capable of clearance and retention of salmon faeces and feed—and assimilation of this material into tissues. They have also been described as a potential candidate species for integrated multi-trophic aquaculture, suggesting the species could assist in mitigating build-up of waste from salmon farming or in validating the extent of the depositional footprint of a farm (Bergvik et al. 2019).

Value to iwi: Scallops are important kaimoana and once significant beds have been heavily impacted by fisheries dredging. Concern was expressed by iwi groups over the potential impact the proposed open ocean farms at Blue Endeavour and Hananui would have on this species.

Logistical considerations: Collection of scallops in sufficient numbers for laboratory experiments, and transportation of organisms, should not be a significant logistical constraint. Provision of a habitat suitably replicating that experienced in the field may be more challenging for this species given it lives at the sediment–water column interface and therefore requires an appropriate level of sediment into which to settle in experimental tank systems.

Erect sponges

Presence in areas of interest: Species of erect sponge have been observed across the Blue Endeavour, Hananui, and Tory Channel sites. The physiological and molecular response of these organisms has been studied in response to sedimentation, and organic enrichment.

Biotic assessment: The group forms an important component of sponge-bryozoan beds at Hananui, and mixed habitat, HMBB, patch reef, and clump reef at Blue Endeavour. Sponge gardens meeting the MacDiarmid et al. (2013) criteria for a sensitive benthic habitat have been noted at Hananui, and, though sponges are not as abundant at Blue Endeavour, they similarly form an important component of biogenic habitats.

Ecological function: Important for habitat provision (nursery grounds for juvenile fish), bentho-pelagic coupling/transfer of energy, nutrients, and organic matter through food webs; some species are known to be good indicators of proximal environmental conditions.

Sensitivity to organic enrichment and sedimentation: Shown to be highly varied among species of sponges.

Value to iwi: Yes. Not a kaimoana or taonga species but recognised for their importance as nursery grounds for juvenile fish.

Logistical considerations: Sponges have been successfully housed in laboratory conditions and can be relocated by removing the substrate on which they attach.

Species proposed for experimental trials

Laboratory trials

Laboratory experiments will be used to determine the tolerance and response of organisms to levels of organic enrichment (salmon faeces) along an exposure gradient predicted at proposed OOA sites in New Zealand. Controlled laboratory experiments allow the isolation of effects from a key stressor of

interest, compared with *in situ* research where an array of factors influence physiological and gene-level responses. Laboratory experiments also provide the opportunity to identify molecular or compound-specific biomarker/s in organisms that have been exposed in isolation to known and controlled levels of organic compounds (see Appendix 3 for a review of potential stress indicators), unlike field sampling approaches. This will enable us to pinpoint effects of low levels of organic enrichment on the health of key species. Laboratory experiments also allow for the determination of complex physiological stress responses that require specific conditions for sample handling and analysis.

To understand how key taxa respond to organic enrichment, we propose the inclusion of horse mussels, bryozoans, scallops, and erect sponges in laboratory trials. Brachiopods will be collected as a back-up for bryozoans, which are considered medium risk for inclusion in laboratory trials. This selection captures a range of feeding strategies and functional groups, while considering species' cultural importance and ecological significance.

Field trials

Organisms *in situ* are subject to a suite of natural processes that cannot be fully re-created in laboratory settings, even with the most elaborate state-of-the-art systems. Furthermore, the specific processes associated with salmon farm waste deposition and resuspension are particularly challenging to re-create in a laboratory setting. As a result, the laboratory experiments described above focus primarily on the effects of organism exposure to fish faeces, rather than the effects of organically enriched sediments and associated processes. To support the laboratory experiments, we propose using existing enrichment gradients from salmon farms in dispersive environments in the Marlborough Sounds[34] to validate the relevance and detectability of indicators/anticipated effects to key species in a real-world system where other influences are also at play.

We propose the inclusion of sea cucumbers, erect sponges, horse mussels, and scallops in opportunistic field sampling. Where possible, opportunistic field sampling is preferred over translocation of organisms, given collection of organisms directly from wild populations will better incorporate effects from long-term exposure to farm-related wastes and most realistic account of effects.

For field translocation experiments (if not found in sufficient numbers along depositional gradients), we propose the inclusion of bryozoans, brachiopods, erect sponges, horse mussels, and scallops. Given their known distributions within the Marlborough Sounds, bryozoans and brachiopods are unlikely to be found in sufficient densities across existing depositional gradients. Therefore, translocation of individuals from these groups will be necessary to understand responses in a real-world system and validate results observed in a laboratory setting.

Previous surveys and reports for benthic habitats in the Marlborough Sounds indicate that local collection of these taxa is possible. However, habitat distribution and species densities can change because of disturbance, environmental change, or seasonal variability. Inclusion of the species listed above is thus dependent on access to enough numbers for the relevant sampling component.

Indicators proposed for use in the study of these species are provided in Table 5 and described in Appendix 3.

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[34] We will communicate with Maritime New Zealand and New Zealand King Salmon prior to deploying fixtures near farms.

Table 5: **Physiological, molecular, and biochemical indicators proposed for measuring response of organisms to farm-related enrichment and sedimentation.**

Taxon	Proposed analytical indicators
Sponge	Transcriptomics, DNA methylation, host microbiome, respiration, feeding rates, scope for growth, digestion efficiency
Horse mussel	Transcriptomics, DNA methylation, oxidative stress, fatty acids, zinc, flow cytometry, respiration, growth performance, feeding rates, scope for growth, digestion efficiency, valve activity
Brachiopod	Transcriptomics, DNA methylation, oxidative stress, fatty acids, zinc, flow cytometry, respiration, growth performance, feeding rates, scope for growth, digestion efficiency
Bryozoan	Transcriptomics*, DNA methylation, respiration, feeding rates, scope for growth, digestion efficiency
Scallop	Transcriptomics, DNA methylation, flow cytometry, oxidative stress, fatty acids, zinc, respiration, growth performance, feeding rates, scope for growth, digestion efficiency
Sea cucumber	Transcriptomics, DNA methylation, host microbiome, oxidative stress, fatty acids, zinc, flow cytometry*, respiration, growth performance**, feeding rates, scope for growth, digestion efficiency

* These analytical approaches are tentative because they are dependent on our ability to extract the appropriate tissue type from the candidate organism.
** Measuring scope for growth in this organism is tentative because it is dependent on the extent to which seasonal fluctuations in growth mask changes due to exposure treatments.

6. FULFILMENT OF BROADER OUTCOMES

6.1 Iwi engagement

Consultation with iwi groups is vital to understanding susceptibility of cultural keystone taonga species to aquaculture, which will assist sustainable management and protection of these species and will generate positive outcomes for Māori communities. Our aim is to engage with our network of iwi partners and collaborators for input on the design and outcome of this project as it progresses. To initiate this process, we have collaborated with Te Kāhui Āio, Cawthron's Māori Business Development Team, who have a number of aquaculture research projects supporting iwi groups and are working to develop their smart Māori aquaculture sector. Using their existing relationships with iwi partners, Te Kāhui Āio are engaging in discussions surrounding existing work/work in development relevant to this space currently used by iwi, as well as the nature of input iwi would like to have in this project. These conversations are ongoing.

Our team has also engaged with the Social Science Team at Cawthron to ensure that we are following the appropriate human ethics protocol for engaging with iwi.

6.2 Development of international research network

We are collaborating with external researchers and organisations based both domestically and internationally. These collaborators include researchers from the University of Waikato, University of Tasmania, and the Institute of Marine Research in Norway. The development of a wider research network provides opportunities for skill development and research expertise by students and early-career researchers. Through these collaborations, the capacity and capability to conduct research within Cawthron and New Zealand will grow.

Our team has been in regular communication with our collaborators Dr Camille White (University of Tasmania) and Dr Joanne Ellis (University of Waikato) regarding their continued involvement in the

project as it moves forward. We anticipate engaging with Dr Nigel Keeley (Institute of Marine Research in Norway) as the project progresses.

6.3 PhD student

The proposal is designed to support at least one PhD student, who will be supervised by Dr Camille White, Dr Joanne Ellis, and Dr Anastasija Zaiko. The student(s) will primarily be based at Cawthron and, alongside adding value to the core project outcomes, their inclusion will assist in developing further expertise on environmental effects of aquaculture in new environments in New Zealand. A PhD position advertisement has been disseminated through the Cawthron website, New Zealand Marine Science Society list server, and to university connections across New Zealand. We have received numerous applications and have begun conducting interviews for the position.

7. ACKNOWLEDGEMENTS

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APPENDIX 1. REVIEW OF SUBMISSIONS FROM THE BLUE ENDEAVOUR FARM APPLICATION

Te Ohu Kaimoana Trustee Limited (Te Ohu Kaimoana) is a representative organisation working on behalf of 58 mandated iwi organisations that represent all Māori throughout Aotearoa. Te Ohu Kaimoana supported[35] the Blue Endeavour application conditionally. The submission emphasised the importance of an OOA application working on the basis that it operates within the environmental capacity for the site and the surrounding area. Points highlighted in the submission include the lack of a thorough understanding of the effects of the proposed development to the environment beneath the site and surrounding area. Te Ohu Kaimoana also highlighted the lack of understanding surrounding acceptable standards for key parameters of this site and ambiguity surrounding how the staged development will occur in the absence of a thorough understanding of environmental impact (for example, what are undesirable situations?). They raised concerns over how management would occur in the event of exceedance of environmental limits, as well as the cause of those exceedances to prevent further environmental impact and to allow recovery.

Ngāti Kuia are one of the oldest and largest iwi of the South Island. Their trust, Te Runanga o Ngāti Kuia, put forth a submission stating they support the Blue Endeavour application. They raised several concerns around the application, however, including the fishing effort displacement and potential loss or alteration of traditional fishing grounds from farm impact (for example, via deposition and accumulation). While the submission notes that Ngāti Kuia has a special relationship with all species found in the Ngāti Kuia Fisheries Protocol Area[36], tupa (scallop) is specifically mentioned in the context of the Blue Endeavour application site, which is near one of the biggest, most abundant tupa grounds important to the iwi. They also expressed concerns surrounding mahinga kai decline in the form of decreased seaweed populations due to kina barrens[37]. In addition, Ngāti Kuia raised concerns surrounding environmental protection and kaitiakitanga[38]. The submission states concern regarding the adverse effect of the seabed impact beneath the farm, as well as in the footprint, as a result of feed input allocated. Additional concerns were raised surrounding displacement of species from the farm area, such as fish, seals, sharks, and seabirds. Of particular importance noted are kaimoana species, including kōura (crayfish), tāmure (snapper), haku (kingfish), and hāpuka (groper) due to increased predators and seabird activities.

To address these concerns, New Zealand King Salmon agreed to develop a cultural indicators framework incorporating baseline information (by 31 December 2021[39]). These would include:

- 'cultural health indicators in accordance with Ngāti Kuia core values;
- monitoring methods to assess effects against the cultural indicators and timing for monitoring;
- tikanga based responses to any negative effects'.

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[35] We note that the first submission dated 20 December 2019 raised additional concerns regarding farm operation and absence of open ocean best management practice guidelines. The subsequent revised submission dated 14 October 2021 revoked those concerns due to information provided by New Zealand King Salmon in the interim.

[36] As recognised by the Crown under the Fisheries Act 1996.

[37] https://eservices.marlborough.govt.nz/download/files/wxpJxx49VRtL6NnmI0TMXvS6HIVq6o1oaup1Hqfyw Yow.

[38] Guardianship or protection.

[39] As per the hearing notes, relevant consent conditions are condition 96 through 100. As at the time of writing, it was unclear whether such a cultural impact statement has been prepared.

APPENDIX 2. SPECIES SELECTION MATRIX AND SCORING CRITERIA

Table A2.1: **Species selection matrix. (continued over next 7 pages)**

Key
BE: Present at Blue Endeavour
HA: Present at Hananui
TC: Present at Tory Channel site
SPRes: Information on species physiological response
BIOG: Present in biogenic habitat types
SBH: Does percentage cover/density qualify as a sensitive benthic habitat?
FREQ: Frequency of occurrence
HAB: Habitat-forming species
FF: Filter feeders
BIOTU: Bioturbators
RECYC: Nutrient and organic matter cycling
IND: Indicator species
SUS: Sensitivity to organic enrichment and sedimentation
KAI: Is the species kaimoana or taonga?
KAI +: Does the species support kaimoana or taonga species (or is it important to mahinga kai)?

Cells with italicised numbers in the table indicate cases where best professional judgement was used to allocate a score to a functional group.

Species	Preliminary data/existing info				Biotic assessment			Ecological function					Sensitivity to salmon farming	Value to iwi		Overall score
	BE	HA	TC	SPRes	BIOG	SBH	FREQ	HAB	FF	BIOTU	RECYC	IND	SUS	KAI	KAI+	
Horse mussels (*Atrina zelandica*)	Y	N	Y	0.75	Y	1.00	1.00	Y	Y	N	1.00	Y	0.67	N	0.67	11.08
Talochlamys	Y	N	N	0.25	Y	0.00	0.20	Y	Y	N	0.33	N	0.00	N	0.33	5.12
Fan shells (Pectinidae)	Y	N	N	0.25	Y	0.00	0.00	N	Y	N	0.33	N	0.00	N	0.33	3.92
Bearded horse mussels (*Modiolus areolatus*)	N	Y	N	0.25	Y	0.00	0.20	Y	Y	N	0.67	N	0.00	N	0.33	5.45
Green-lipped mussels (*Perna canaliculus*)	N	N	Y	0.75	N	0.00	0.00	Y	Y	N	0.67	Y	0.50	Y	1.00	7.92
Blue mussels (*Mytilus galloprovincialis*)	N	N	Y	0.75	N	0.00	0.00	Y	Y	N	0.67	Y	0.50	N	1.00	6.92
Dredge oysters (*Ostrea chilensis*)	N	Y	N	0.25	Y	1.00	0.20	Y	Y	N	0.67	N	0.67	Y	1.00	8.78
Scallops (*Pecten novaezelandiae*)	Y	Y	Y	0.25	Y	0.00	0.20	N	Y	N	0.67	N	0.83	Y	1.00	8.95
Suspension-feeding bivalves (epifaunal)	**Y**	**Y**	**Y**	**0.44**	**Y**	**1.00**	**0.36**	**Y**	**Y**	**N**	**0.63**	**Y**	**0.63**	**Y**	**0.71**	11.76
Brachiopods (Blue Endeavour: *Notosaria nigricans, Magasella sanguinea, Calloria inconspicua*), (Hananui: either *Neothyris lenticularis* or *Magasella sanguinea*)	Y	Y	N	0.25	Y	1.00	1.00	Y	Y	N	0.67	Y	0.70	N	0.67	10.28
Suspension-feeding brachiopods	**Y**	**Y**	**N**	**0.25**	**Y**	**1.00**	**1.00**	**Y**	**Y**	**N**	**0.67**	**Y**	**0.70**	**N**	**0.67**	10.28
Tube-dwelling anemones (Ceriantharia)	Y	N	N	0.50	Y	0.00	1.00	N	Y	N	0.50	N	0.00	N	0.33	5.33

Species	Preliminary data/existing info				Biotic assessment			Ecological function					Sensitivity to salmon farming	Value to iwi		Overall score
	BE	HA	TC	SPRes	BIOG	SBH	FREQ	HAB	FF	BIOTU	RECYC	IND	SUS	KAI	KAI+	
Giant wandering anemone (*Phylctenactis tuberculosa*)	Y	N	N	0.25	Y	0.00	0.05	N	Y	N	0.67	N	0.00	N	0.33	4.30
White striped anemone (*Athothoe albocincta*)	N	Y	Y	0.25	Y	0.00	0.20	N	Y	N	0.50	N	0.00	N	0.33	5.28
Anemones – undefined	Y	N	N	0.50	Y	0.00	0.20	N	Y	N	0.50	N	0.30	N	0.33	4.83
Sessile suspension feeders – anemones	**Y**	**Y**	**Y**	**0.38**	**Y**		**0.36**	**N**	**Y**	**N**	**0.54**	**N**	**0.30**	**N**	**0.33**	6.91
Feather hydroid (Aglaopheniidae) / hydroid tree	Y	Y	Y	0.50	Y	0.00	0.80	N	Y	N	0.33	N	0.00	N	**0.33**	6.97
Sessile suspension feeders – hydroids	**Y**	**Y**	**Y**	**0.50**	**Y**		**0.80**	**N**	**Y**	**N**	**0.33**	**N**		**N**	**0.33**	6.97
Solitary cup coral	Y	N	N	0.50	Y	0.00	0.05	N	Y	N	0.50	N	0.00	N	**0.33**	4.38
Colonial stony coral	N	N	Y	0.50	N	0.00	0.00	N	Y	N	0.50	N	0.00	N	**0.33**	3.33
Sessile suspension feeders – coral	**Y**	**N**	**Y**	*0.75*	**Y**		**0.05**	**N**	**Y**	**N**	**0.50**	**N**		**N**	**0.33**	5.63
Tubeworm mounds / calcareous tubeworms from family Serpulidae (*Galeolaria hystrix* at Hananui)	Y	Y	Y	0.25	Y	1.00	0.80	Y	Y	N	0.67	N	0.00	N	**0.33**	9.05
Tubeworm – Chaetopteridae	Y	N	N	0.25	N	0.00	0.05	Y	Y	N	0.67	N	0.00	N	**0.33**	4.30
Giant tubeworms	Y	N	N	0.25	Y	0.00	0.60	Y	Y	N	0.67	N	0.00	N	**0.33**	5.85
Sessile suspension feeders – habitat-building tubeworms	**Y**	**Y**	**Y**	**0.25**	**Y**	**1.00**	**0.48**	**Y**	**Y**	**N**	**0.67**	**N**		**N**	**0.33**	8.73
Fan worm (Sabellidae)	Y	N	Y	0.25	Y	0.00	0.20	N	Y	N	0.33	N	0.00	N	**0.33**	5.12

Species	Preliminary data/existing info				Biotic assessment			Ecological function					Sensitivity to salmon farming	Value to iwi		Overall score
	BE	HA	TC	SPRes	BIOG	SBH	FREQ	HAB	FF	BIOTU	RECYC	IND	SUS	KAI	KAI+	
Large parchment worm	Y	N	Y	0.25	Y	0.00	0.00	N	Y	N	0.33	N	0.00	N	**0.33**	4.92
Sessile suspension feeders – non-habitat-building tubeworms	**Y**	**N**	**Y**	**0.25**	**Y**		**0.20**	**N**	**Y**	**N**	**0.33**	**N**		**N**	**0.33**	5.12
Bryozoans (bushy and flexible branching forms, *Orthoscuticella innominata*, *Hornera foliacea* and possibly *Hornera robusta* at Hananui)	Y	Y	N	0.25	Y	0.50	1.00	N	Y	N	0.50	N	0.00	N	**0.33**	6.58
Sessile suspension feeders – bryozoans bushy and flexible	**Y**	**Y**	**N**	**0.25**	**Y**	**0.50**	**1.00**	**N**	**Y**	**N**	**0.50**	**N**		**N**	**0.33**	6.58
Erect bryozoans (mainly *Cinctipora elegans* at Hananui)	Y	Y	N	0.25	Y	1.00	0.80	Y	Y	N	0.50	Y	0.00	N	**0.67**	9.22
Sessile suspension feeders – bryozoans erect	**Y**	**Y**	**N**	**0.25**	**Y**	**1.00**	**0.80**	**Y**	**Y**	**N**	**0.50**	**N**		**N**	**0.67**	8.22
Massive encrusting bryozoan (*Celleporaria agglutinans*, encrusting brown at Tory Channel)	N	Y	Y	0.25	Y	0.00	0.00	N	Y	N	0.50	N	0.00	N	0.67	5.42
Sessile suspension feeders – bryozoans encrusting	**N**	**Y**	**Y**	**0.25**	**Y**			**N**	**Y**	**N**	**0.50**	**N**		**N**	**0.67**	5.42
Yellow encrusting sponge	Y	N	N	0.00	Y	0.00	0.20	N	Y	N	0.67	N	0.00	N	0.33	4.20
Orange sponge (*Crella encrustans*)	N	Y	Y	0.00	N	1.00	0.80	N	Y	N	0.67	N	0.00	N	**0.33**	5.80

Species	Preliminary data/existing info				Biotic assessment			Ecological function					Sensitivity to salmon farming	Value to iwi		Overall score
	BE	HA	TC	SPRes	BIOG	SBH	FREQ	HAB	FF	BIOTU	RECYC	IND	SUS	KAI	KAI+	
Blue encrusting sponge	N	N	Y	0.00	N	0.00	0.00	N	Y	N	0.67	N	0.00	N	0.33	3.00
Encrusting sponges	Y	N	N	0.50	Y	0.00	0.80	N	Y	N	0.67	N	0.00	N	**0.33**	5.30
Sessile suspension feeders – sponge encrusting	**Y**	**Y**	**Y**	**0.50**	**Y**	**1.00**	**0.60**	**N**	**Y**	**N**	**0.67**	**N**		**N**	**0.33**	8.10
Finger sponge	Y	N	Y	0.00	Y	0.00	0.20	N	Y	N	0.83	N	0.00	N	**0.67**	5.70
Orange erect sponge	Y	N	N	0.00	Y	0.00	0.20	N	Y	N	0.83	N	0.00	N	**0.67**	4.70
Sponge (*Dactylia varia*)	N	Y	N	0.00	Y	1.00	0.80	N	Y	N	0.83	N	0.00	N	**0.67**	6.30
Sponge (*Iophon minor*)	N	Y	N	0.00	Y	1.00	0.80	N	Y	N	0.83	N	0.00	N	**0.67**	6.30
Grey cup sponges (*Ecionemia alata*)	N	N	Y	0.00	N	0.00	0.00	N	Y	N	1.00	Y	0.00	N	**0.67**	4.67
Sessile suspension feeders – sponge erect	**Y**	**Y**	**Y**	*0.88*	**Y**	**1.00**	**0.50**	**N**	**Y**	**N**	**0.87**	**Y**		**N**	**0.67**	9.91
Solitary ascidian (including *Styela clava*)	Y	N	Y	0.25	Y	0.00	0.05	N	Y	N	0.33	N	0.00	N	**0.33**	4.97
Sea tulip (*Pyura pachydermatina*)	N	Y	Y	0.25	N	0.00	0.20	N	Y	N	0.33	N	0.00	N	**0.33**	4.12
Sessile suspension feeders – solitary ascidian	**Y**	**Y**	**Y**	**0.25**	**Y**		**0.13**	**N**	**Y**	**N**	**0.33**	**N**		**N**	**0.33**	6.04
Colonial ascidians	Y	N	Y	0.50	Y	0.00	0.20	N	Y	N	0.33	N	0.70	N	**0.33**	6.07
Colonial ascidians (*Botrylloides* sp.)	N	Y	Y	0.25	Y	0.00	0.00	N	Y	N	0.33	N	0.00	N	**0.33**	4.92
Colonial ascidians (*Botryllus* sp.)	N	Y	N	0.25	Y	0.00	0.00	N	Y	N	0.33	N	0.00	N	**0.33**	3.92

Species	Preliminary data/existing info				Biotic assessment			Ecological function					Sensitivity to salmon farming	Value to iwi		Overall score
	BE	HA	TC	SPRes	BIOG	SBH	FREQ	HAB	FF	BIOTU	RECYC	IND	SUS	KAI	KAI+	
Colonial ascidians (*Eudistoma circumvallatum*)	N	Y	N	0.25	Y	0.00	0.00	N	Y	N	0.33	N	0.00	N	**0.33**	3.92
Yellow colonial ascidian (*Aplidium phortax*)	N	N	Y	0.25	N	0.00	0.00	N	Y	N	0.33	N	0.00	N	**0.33**	2.92
Sessile suspension feeders – colonial ascidian	**Y**	**Y**	**Y**	**0.30**	**Y**		**0.20**	**N**	**Y**	**N**	**0.33**	**N**	**0.70**	**N**	**0.33**	6.87
Burrowing sea cucumber	Y	N	N	0.25	Y	0.00	0.20	N	Y	Y	0.50	N	0.00	N	**0.33**	5.28
Sessile suspension feeders – sea cucumber	**Y**	**N**	**N**	**0.25**	**Y**		**0.20**	**N**	**Y**	**Y**	**0.50**	**N**		**N**	**0.33**	5.28
Crinoids	Y	Y	N	0.25	Y	0.00	0.20	N	Y	N	0.33	N	0.00	N	**0.33**	5.12
Sessile suspension feeders – crinoid	**Y**	**Y**	**N**	**0.25**	**Y**		**0.20**	**N**	**Y**	**N**	**0.33**	**N**		**N**	**0.33**	5.12
Brittle stars (*Ophiopsammus maculata*)	Y	Y	Y	0.88	Y	0.00	0.20	N	N	N	0.50	Y	0.33	N	0.33	7.24
Sea cucumber (*Australostichopus mollis*)	Y	Y	Y	0.88	Y	0.00	0.20	N	N	Y	1.00	Y	0.33	N	0.33	8.74
Mobile deposit feeders	**Y**	**Y**	**Y**	**0.88**	**Y**		**0.20**	**N**	**N**	**Y**	**0.75**	**Y**	**0.33**	**N**	**0.33**	8.49
Turret shell (*Maoricolpus*)	Y	Y	Y	0.00	Y	0.00	0.20	N	Y	N	0.33	N	0.00	N	0.67	6.20
Mobile suspension feeder	**Y**	**Y**	**Y**		**Y**		**0.20**	**N**	**Y**	**N**	**0.33**	**N**		**N**	**0.67**	6.20
Kina (*Evechinus chloroticus*)	N	Y	Y	0.50	Y	0.00	0.20	N	N	N	0.67	N	0.70	Y	1.00	7.07
Cat's eye snail (*Lunella smaragda*)	N	N	Y	0.25	N	0.00	0.00	N	N	N	0.50	N	0.00	N	0.67	2.42

Species	Preliminary data/existing info				Biotic assessment			Ecological function					Sensitivity to salmon farming	Value to iwi		Overall score
	BE	HA	TC	SPRes	BIOG	SBH	FREQ	HAB	FF	BIOTU	RECYC	IND	SUS	KAI	KAI+	
Duck's bill limpet (*Scutus breviculus*)	N	N	Y	0.25	N	0.00	0.00	N	N	N	0.50	N	0.00	N	0.33	2.08
Cook's turban (*Cookia sulcata*)	N	N	Y	0.25	N	0.00	0.00	N	N	N	0.50	N	0.00	N	0.67	2.42
Limpet (*Cellana stellifera*)	N	N	Y	0.25	N	0.00	0.00	N	N	N	0.50	N	0.00	N	0.67	2.42
Limpet (*Siphonaria* sp.)	N	N	Y	0.25	N	0.00	0.00	N	N	N	0.50	N	0.00	N	0.67	2.42
Calliostoma sp.	Y	N	Y	0.25	N	0.00	0.00	N	N	N	0.50	N	0.00	N	0.67	3.42
Trochus sp.	N	N	Y	0.25	N	0.00	0.00	N	N	N	0.50	N	0.00	N	0.67	2.42
Mobile grazers	**Y**	**Y**	**Y**	**0.28**	**Y**		**0.20**	**N**	**N**	**N**	**0.52**	**N**	**0.70**	**Y**	**0.67**	7.37
Cushion stars (*Patiriella regularis*)	Y	Y	Y	0.25	Y	0.00	0.20	N	N	N	0.33	N	0.33	N	0.33	5.45
11-armed sea star (*Coscinarius muricata*)	Y	Y	Y	0.25	Y	0.00	0.20	N	N	N	0.50	N	0.33	N	0.33	5.62
7-armed sea star (*Astrostole scabra*)	Y	N	N	0.25	Y	0.00	0.20	N	N	N	0.50	N	0.33	N	0.33	3.62
Long-armed sea star (possibly *Cosmasterias dyscrita*)	Y	N	N	0.25	Y	0.00	0.20	N	N	N	0.33	N	0.33	N	0.33	3.45
Apricot star (*Sclerasterias mollis*)	N	N	Y	0.25	N	0.00	0.00	N	N	N	0.33	N	0.33	N	0.33	2.25
Crab, nondescript (Brachyura)	Y	N	Y	0.50	Y	0.00	0.05	N	N	Y	0.33	N	0.33	N	0.67	5.88
Hermit crab (*Pagurus novizealandiae*)	N	N	Y	0.50	N	0.00	0.00	N	N	Y	0.33	N	0.33	N	0.67	3.83
Hairy seaweed crab (*Notomithrax ursus*)	N	N	Y	0.25	N	0.00	0.00	N	N	Y	0.33	N	0.33	N	0.67	3.58

Species	Preliminary data/existing info				Biotic assessment			Ecological function					Sensitivity to salmon farming	Value to iwi		Overall score
	BE	HA	TC	SPRes	BIOG	SBH	FREQ	HAB	FF	BIOTU	RECYC	IND	SUS	KAI	KAI+	
Penion sp.	Y	N	N	0.25	Y	0.00	0.05	N	N	N	0.33	N	0.50	N	0.33	3.47
Nudibranch (Pleurobranchaea)	Y	N	N	0.25	Y	0.00	0.20	N	N	N	0.33	N	0.70	N	0.33	3.82
Mobile scavengers / predators	**Y**	**Y**	**Y**	**0.30**	**Y**		**0.16**	**N**	**N**	**Y**	**0.37**	**N**	**0.39**	**N**	**0.43**	6.64
Circular saw-shell (*Astraea heliotropium*)	Y	Y	Y	0.00	Y	0.00	0.20	N	N	N	0.00	N	0.00	N	0.33	4.53
Mobile undefined	**Y**	**Y**	**Y**		**Y**		**0.20**	**N**	**N**	**N**		**N**		**N**	**0.33**	4.53

APPENDIX 3: REVIEW OF POTENTIAL STRESS INDICATORS

Several biological indicators can be used to determine stress or physiological changes in organisms. However, the effectiveness and response of these indicators is usually specific to the species and stressor of interest. Below, physiological indicators have been grouped into two broad categories: whole-body indicators and targeted indicators. Whole-body indicators typically provide a metric that incorporates measures of the functioning of the entire organism, thus capturing 'net' responses of the stressed animal. These indicators are critical to understanding or extrapolating individual stress response observations to potential real-world implications. Meanwhile, targeted indicators generally focus on a specific bodily function relating to a type of tissue, cell, or organic molecule, helping us to understand the mechanisms behind stress responses and often providing more easily measurable stress metrics for field applications. The markers considered also recognise that localised enrichment may be *beneficial*, rather than stressful to individual organisms, improving growth and energy balance.

Using a combination of targeted and whole-body indicators for each candidate species will provide a robust assessment of physiological change associated with applied stress (for example, organic enrichment relevant to OOA). This approach will also allow for molecular and biochemical metrics to be linked to whole-body responses. Establishing correlations between these two types of indicators is important as molecular and biochemical metrics are generally easier to analyse after an organism has been euthanised or removed from its natural environment. Below we include a brief review of the potential indicators that may be applied to candidate species.

A3.1 Targeted indicators

Gene response

Specific genes may be up-regulated or down-regulated in response to stress from organic enrichment and could serve as valuable and informative molecular biomarkers of an organism's response to stress. Using a two-step approach, ideal biomarkers would be identified using transcriptomics as a screening methodology, followed by the development of targeted digital droplet PCR (ddPCR) assays for fast and economic quantification of the biomarkers' expression. Transcriptomics involves RNA[40] sequencing of the mRNA for overall profiling of the host's transcriptome[41]. This approach does not require a priori genetic information of the study organism, allowing for the assessment of complete *de novo* (previously undescribed) transcriptomes. On the other hand, ddPCR is a very precise and sensitive tool that can be used for cost-efficient quantification of RNA from a few genes of interest. RNA can be extracted from plasma of live animals and therefore allows for repeated sampling of individual organisms and the collection of time-series data on species responses. Gene annotation on non-model species can be challenging, therefore inclusion of a model species (*Perna canaliculus*, the New Zealand green-lipped mussel) as one of our study organisms would be preferred when using this approach[42].

Transcriptomics as an approach is universal in its application, given it does not require the genome of the target organism to have been sequenced and can be used for a variety of tissue types with little biological material needed. Changes in gene expression in response to stress occur very quickly and therefore this method can be an effective way of providing a snapshot of an organism's physiological state.

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[40] Carries genetic information that is translated by ribosomes into various proteins necessary for cellular processes.

[41] The full set of coding and non-coding RNAs that are transcribed at a specific developmental stage and /or in response to various physiological conditions within a cell type or tissue.

[42] We have made provisions to undertake transcriptomics work with one species within the timeframe and budget of this project. However, samples will be collected from all species should additional funding become available.

Global DNA methylation

Phenotypic plasticity, the ability of individual genotypes to adapt to exposed environmental conditions by producing different phenotypes, is partly regulated by epigenetic modifications through DNA methylation. Changes in DNA methylation across the genome may determine adaptive phenotypic divergences in epifauna that could be passed onto future generations and have long-term implications (Beal et al. 2018). Here, we propose using a simple, global DNA methylation approach to determine whether an overall trend can be observed in specimens exposed to a range of organic enrichment. While this methodology has low resolution, it could represent a useful and cost-efficient epigenetic proxy of stress.

DNA methylation is universal in its application, but it provides less sensitivity than transcriptomics. This approach could be useful for detecting stress responses in short-lived organisms given DNA methylation can be conserved across generations.

Host microbiome[43]

This method involves sampling the external and internal tissues to obtain a sample of the microbial community hosted by the study organism. The microbiome is analysed using microbial DNA metabarcoding[44] (16S rRNA). Microbiome turnovers are known to follow physiological alterations and pathological conditions in epifauna such as sponges and corals in response to anthropogenic activities (Meron et al. 2011, Morrow et al. 2012, Jessen et al. 2013, Gissi et al. 2019, Ziegler et al. 2019, Laroche et al. 2021, 2022). This approach would allow us to characterise changes in host microbiota across different levels of organic enrichment and link this to physiological (identified through oxidative stress, flow cytometry, and whole-organism approaches) and molecular (identified through transcriptomics) changes.

The host microbiome approach is more targeted as strong evidence of the relationship between the microbiome and physiological change in the host exists for only a subset of organisms, including sponges (Laroche et al. 2021), corals, and sea cucumbers (Wei et al, 2022). Relationships between the physiological state of other organisms of interest and their host microbiome have not been as well investigated for bivalves, brachiopods, bryozoans, tubeworms, and ascidians, making it difficult to apply this as a general approach across species.

Oxidative stress

Environmental stress can disrupt the balance between reactive oxygen species (ROS) (potentially damaging derivatives of molecular oxygen) and antioxidants (compounds that inhibit oxidation). Accumulation of ROS within cells results in oxidative stress, which causes oxidative damage to macromolecules (proteins, lipids, DNA). Oxidative stress markers are measured through a suite of biochemical assays[45], which involve the extraction of macromolecules and determination of levels of oxidation and enzymatic antioxidant activity by reading reactions in a plate reader[46]. These assays provide information on the levels of oxidative damage in the tissues due to a known stressor (such as lipid peroxidation[47]), and the capacity of enzymatic antioxidants to neutralise / reduce the damage produced.

Oxidative stress can be assessed using a specific tissue type such as haemolymph or can be applied to a homogenised aliquot of whole-organism tissue. While the use of oxidative species and antioxidants has not been proven across all species of interest in this review, the techniques appear to be successful

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[43] The organisms occurring inside and on host surfaces.
[44] A technique that allows for the simultaneous identification of many taxa within the same sample.
[45] An analytical *in vitro* (outside of the living body) procedure used to detect and quantify the binding or activity of a biological molecule.
[46] Instruments that are used to detect biological, chemical, or physical events of samples.
[47] The chain of reactions of oxidative degradation of lipids.

across a range of species, including bivalves (Delorme et al. 2021) ascidians (Azumi et al. 2004), polychaetes (Carregosa et al. 2014), and bryozoans (Elia et al. 2007).

Fatty acids

Fatty acid values in feed can be used to trace farm-derived particulates into the marine environment (sediment, or organisms themselves) (White et al. 2019, Woodcock et al. 2019, Laroche et al. 2021, McMullin et al. 2021). These tools/tracers are also useful for detecting physiological responses related to ecological stress and dietary changes. Omega-3 and omega-6 polyunsaturated fatty acids (PUFAs), for instance, are thought to provide increased stress resistance across a range of taxa, and depletion of these in sponges and corals has been associated with deteriorating health (Bachok et al. 2006, Bennett et al. 2018), while monounsaturated fatty acids (MUFAs), which represent a readily available energy source, have been found to be reduced in stressed sponges to compensate for energy deficit (Bennett et al. 2018). These biomarkers could also be applied to understand how an organism metabolises farm-derived organic and nutrient waste (for example, White et al. 2017) and extrapolate these to potential wider ecosystem or long-term effects. The use of fatty acids in this way is currently a focus of the research being done for the New Zealand horse mussel (Elvines in prep.). Measuring fatty acids involves derivatisation[48] of lipids from homogenised samples, extraction of fatty acids, and quantification using gas chromatography.

Fatty acid analysis can be applied to any lipid-containing tissue from all species of interest. Analysis requires a sample of ~10 mg dry tissue weight and therefore generally involves destructive sampling of an organism. This technique generally reflects mid- to long-term changes in tissue composition, and turnover of fatty acids in tissue is influenced by the metabolism and growth of an animal. Changes in lipid content or fatty acid composition in response to diet of stress should occur within a 3–6-month experimental time frame.

Zinc

Zinc is an additive in salmon feeds and may accumulate in the tissues of organisms that use fish waste as a trophic subsidy. At high levels, zinc can have toxic effects, so understanding whether organisms accumulate and/or may be adversely affected by zinc is important for assessing sub-lethal effects, particularly for longer-lived organisms that may accumulate it over longer time frames. A gap in understanding the effects of potential contaminants such as zinc on longer-lived organisms that interact with salmon farms was identified through the Board of Inquiry salmon farm consenting process in 2015. This project presents a good opportunity to advance knowledge in this space. Zinc can be reliably analysed through a range of methods, including inductively coupled plasma mass spectrometry, in a range of sample matrices.

Heavy metals such as zinc can take a significant period of time to bioaccumulate in an organism's tissues. Therefore, this analytical approach would be best applied to organisms that have inhabited the area within the depositional footprint of a farm for an extended period. Zinc is unlikely to bioaccumulate in tissues over the duration of lab trials or field translocation experiments; however, opportunistic sampling across depositional gradients at existing farms could provide useful insights into the levels of zinc and other potentially toxic heavy metals occurring at dispersive salmon farming sites.

Flow cytometry

Flow cytometry is an approach used to characterise various functional and immune parameters of cells and allows for the health assessment of organisms. The protocol for flow cytometry involves running a single-cell suspension (haemolymph/blood, sperm, eggs, microalgae) in front of a laser. The technique can measure up to 11 different properties of the cells based on the reflection and emission of the light. These properties include cell concentration, cell morphology, proportion of different cell types, cell viability, reactive oxygen species production (oxidative stress marker), mitochondrial membrane

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[48] The process of chemically altering an analyte.

potential, esterase[49] production, phagocytic capabilities[50], neutral lipid content, lysosomal[51] content, and apoptosis[52]. One of the advantages of this approach is that it is non-lethal, so can be used to sample the same organism through time.

This approach is limited to organisms from which extraction of haemolymph or other cell suspensions is possible. Solid tissues that can be dissociated into single cells such as mucosal tissues can also be characterised by flow cytometry. Haemolymph can be extracted from molluscs such as bivalves, brachiopods, and gastropods (Vignier et al. 2017, Rolton & Ragg 2020), but extractions of cell suspensions from sponges, tubeworms, cnidarians, and ascidians is not well documented and therefore this method cannot be relied upon to characterise physiological stress or changes in these taxonomic groups.

A3.2 Whole-body indicators

Respiration rates

Respiration experiments involve placing organisms in individual respirometry cores/chambers and measuring oxygen concentration over time. This method is applied to live organisms, can be measured repeatedly throughout the experiment, and will provide information about organism metabolic rates. Changes in respiration rates in response to stress from organic enrichment have been observed for species such as the soft coral *Duva florida*, which was found to display high respiration rates after collection from sites closer to farm cages (Laroche et al. 2022).

Measuring respiration rates should be possible for most species of interest, assuming respiratory chambers of an appropriate size are available (Jansen et al. 2009, Maldonado et al. 2012). The requirement for separate chambers for each individual does increase the space and resources needed to conduct such measurements, but individuals could be moved to and from respiratory chambers as required.

Growth performance

Physical growth is tracked through measurements of size and/or wet weight over time. This method provides information about the overall condition of organisms and whether they are receiving enough food/energy to sustain or increase their body size/weight.

For many species it can be challenging to take meaningful measurements of growth. In molluscs, for example, shell growth can easily be measured without compromising health of the individual, but condition, a more robust measure of growth/size, requires destructive sampling. Mobile epifauna such as sea stars and brittle stars can be weighed and measured to assess growth, given they do not retain significant amounts of water, while sea cucumbers appear to follow seasonal growth patterns (Poot-Salazar et al. 2014) and display weight variability based on seawater retention. For other taxonomic groups such as sponges, ascidians, and bryozoans, quantifying growth is likely to be logistically challenging without sacrificing the organism.

Energetics

Stress can present as a change in feeding rate or through increased processing costs associated with feeding. For example, as inorganic sediment levels increase, mussels must either sort and reject sediment particles or accept sediment particles and potentially reduce digestion efficiency as the extra 'roughage' passes through the gut. Even relatively modest sustained levels of sediment loading may outweigh the benefit of the food, effectively creating starvation dynamics even when food is available.

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[49] An enzyme that plays a role in the degradation of natural materials and pollutants.
[50] The phagocyte system is an essential component of innate immunity.
[51] Lysosomes are vesicles containing enzymes that can break down many kinds of biomolecules and are involved in various cell processes.
[52] A mechanism that allows cells to self-destruct when stimulated by the appropriate trigger.

Useful laboratory approaches for understanding how organic enrichment affects feeding energetics in key species are described below.

Feeding rates

Measuring feeding rates involves quantifying the amount of food an animal consumes over time. This approach provides information about the overall condition of the animal/s. Increased levels of stress typically reduce the feeding rate. This approach may require optimisation to accommodate differences in species feeding behaviours.

Scope for growth

Measuring scope for growth involves quantifying rates of food intake and waste production, the energy content of both, and the resting metabolic rate (through measures of respiration/oxygen consumption). Subtracting the energy lost through waste and metabolism from the energy ingested produces an estimate of 'scope for growth'. Scope for growth can be measured over a range of organic enrichment levels. A negative scope for growth value indicates an unsustainable energy balance.

Digestion efficiency

Digestion efficiency can be determined by measuring the organic to inorganic ratio of food used by the organism and of faeces/waste produced by the organism. A reduction in digestion efficiency can occur due to a reduction in food quality, which may be expected across an enrichment gradient.

Approaches to understand species energetics in response to farm-derived organic enrichment should be possible across a wider range of species. These approaches to understanding stress and physiological change can be time consuming, and can require that individuals are held in separate tanks or that an average value is calculated for multiple species held within a single aquarium.