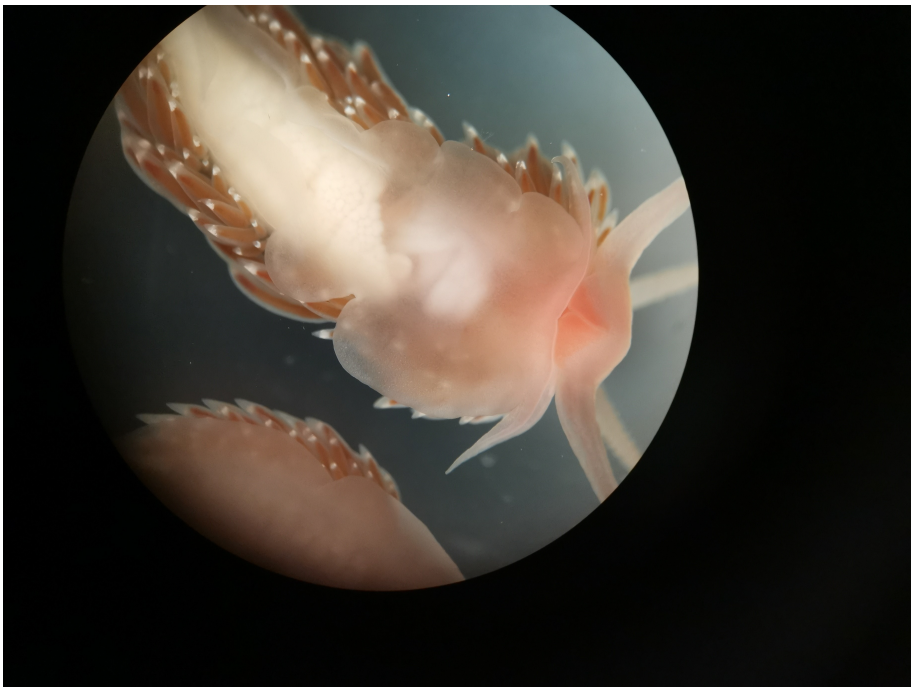
Master's thesis

NTNU
Norwegian University of Science and Technology
Faculty of Natural Sciences
Department of Biology

Live Agnethe Sørlundsengen Haugen

Feeding ecology and behavior of nudibranchs from the sublittoral zones of Trondheimsfjorden and interactions with fouling communities

Master's thesis in Ocean Resources
Supervisor: Nicole Aberle-Malzahn
Co-supervisor: Jussi Evertsen
May 2022



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Live Agnethe Sørlundsengen Haugen

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The picture on the front page is a *Facelina bostoniensis* attached to the water surface. Photo: Live Agnethe Sørlundsengen Haugen.

Abstract

Scyphozoan medusae pose a serious nuisance to fish welfare in aquaculture pens, to the recreational value of coastal areas, and to their own public image and reputation. Their benthic polyp stage is the origin of jellyfish blooming events that have the potential to cover several kilometers of sea-surface area with adult medusae. Limiting growth and reproduction due to predation of this particular life stage could reduce the bloom formation abilities of these populations and could thus act as natural bloom-controllers.

Bryozoan species have shown invasive characteristics in certain regions. They cover entire laminas of kelp and cause them to become brittle and less resistant to wave exposure. As kelp lamina are one of the wave breaking factors preventing shorelines from erosion, it is important that they do not become more fragile due to bryozoan epigrowth.

Nudibranch predation on scyphozoan polyps and bryozoan zooids can therefore aid in the tackling of these problems. The nudibranchs' predation potential on specific fouling organisms has been proven to exist by several authors and is also documented in this study.

As the oceans are likely to face an increase in surface temperatures in the near future, this study looked at finding any temperature-dependent relationship in nudibranch ingestion rates. This relationship was not found to be of significant manner for either of the species used in this study, *Facelina auriculata, Facelina bostoniensis* and *Onchidoris muricata*.

The age of the scyphozoan polyps (*Aurelia aurita*) used in these experiments had a significant effect on the carbon specific ingestion rates of the nudibranchs that were used in this study (*F. bostoniensis* and *F. auriculata*). Age is directly correlated with size, however, other studies have shown conflicting results on this matter thus needing further research. The two different species of nudibranchs were tested, and results showed a significant difference in carbon specific ingestion rates between them. This indicates a difference in predation potential on benthic scyphozoan polyps and hence their ability to combat scyphozoan blooms in marine ecosystems. It is unknown whether these species would choose to prey on scyphozoan polyps if other prey items were available, as fouling community studies are rare for nudibranchs. Conducting such studies is important if we want to widen our understanding of the feeding ecology and behavior of nudibranchs.

As mentioned, no significant effect of temperature on ingestion rates of *Onchidoris muricata* feeding on the bryozoan species *Electra pilosa* and *Membranipora membranacea* was found. Hence, further research is needed on bryozoans and their interactions with nudibranchs. It will be especially important to study the carbon concentration of the zooids and the carbon specific ingestion rates of several species of dorid nudibranchs. By comparing the carbon biomass of nudibranchs to the carbon concentration of their prey, a better understanding of the relationship between the two components can be achieved. As this study was conducted using only the ingestion rates (zooids min^{-1}), and not carbon specific ingestion rates (µgC prey $µg^{-1}$ C nudibranch min^{-1}), this could possibly have given a different result statistically.

Sammendrag

Stormaneter eller medusa i klassen Scyphozoa opptrer som en plage for fiskevelferd i akvakulturmerder, for rekreasjonsverdien i kystnære områder, og ovenfor deres eget rykte. Deres bentiske polyppstadie forårsaker oppblomstringer som har potensiale til å dekke flere kilometer av havoverflaten med voksne medusa. Ved å begrense vekst og reproduksjon ved hjelp av predasjon allerede på dette tidlige livsstadiet kan evnen disse polyppene har til å skape slike oppblomstringer reduseres på populasjonsnivå.

Arter i rekken Bryozoa har vist å ha invaderende egenskaper i enkelte regioner. Disse artene dekker hele tareblader (lamina) og forårsaker at de blir skjøre og mindre resistente mot påkjennelsen fra bølger. Tare er en av flere viktige faktorer som bidrar til at kyst- og strandlinjen ikke vaskes bort, ved at de skaper brytning i bølgene som kommer inn mot land. Dermed er det viktig at ikke taren langs kysten blir skjørere grunnet påvekstorganismer slik som mosdyr.

Predasjon fra nakensnegler på glassmanet-polypper og mosdyr-zooider kan bidra i bekjempelsen av disse problemene. Predasjonspotensialet til nakensnegler har blitt dokumentert av flere forskere rundt om i verden, og det har også blitt dokumentert i dette studiet.

Ettersom havet står ovenfor en potensiell økning i overflatetemperatur i relativ nær framtid har dette studiet forsøkt å finne ut om det er et temperaturavhengig forhold i nakensneglers matinntak. Et slikt forhold viste seg å ikke være signifikant for nakensnegleartene brukt i dette studiet, *Facelina auriculata*, *Facelina bostoniensis* og *Onchidoris muricata*.

Alderen til glassmanet-polyppene (*Aurelia aurita*) som ble brukt i disse eksperimentene hadde en signifikant effekt på den karbon-spesifikke inntaksraten til nakensnelgene *F. auriculata* og *F. bostoniensis*. Alder er direkte korrelert til størrelse, men det viser seg at andre studier har motstridende resultater på nettopp dette. I tillegg ble det funnet en signifikant forskjell i inntaksrate mellom de to artene av nakensnegler (*F. auiculata* og *F. bostoniensis*). Dette indikerer at det er en forskjell i predasjonspotensiale på bentiske polypper og dermed også deres evne til å bekjempe oppblomstring av maneter. Det er uvisst om disse artene ville valgt polypper som byttedyr dersom andre bytter hadde vært tilgjengelig, ettersom forsøk med generell påvekstflora og fauna så langt virker å være sjelden vare for nakensnegler. Å gjennomføre slike studier vil derfor være viktig dersom vi ønsker å utvide vår kunnskap om føde-økologi og atferd hos nakensnegler.

Som nevnt over ble det ikke funnet noen signifikant effekt av temperatur på inntaksraten hos *Onchidoris muricata* av mosdyr-artene *Electra pilosa* og *Membranipora membranacea*. Dermed vil det være nyttig å utføre videre research på mosdyr og deres interaksjon med nakensnegler. Det vil være spesielt viktig å studere karbonkonsentrasjonen til zooidene og den karbon-spesifikke inntaksraten hos flere arter av doride nakensnegler. Ved å sammenligne nakensneglenes karbon-biomasse til byttet deres vil en kunne få en bedre forståelse av forholdet mellom de to artsgruppene. Dersom karbon-analyser hadde vært mulig i dette studiet ville det kanskje ha gitt andre resultater som også kunne vært signifikante.

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List of Abbreviations

A. aurita	*Aurelia aurita*
C	carbon
cladobranch	species of the suborder Cladobranchia
dorid	species of the suborder Doridina
FA	*Facelina auriculata* 15 min
FB1	*Facelina bostoniensis* 60 min
FB2	*Facelina bostoniensis* 15 min
polyp	scyphistoma or scyphopolyps
SST	sea surface temperature
sp	species
TBS	Trondhjem Biological Station
WW	wet weight
µg	microgram

1 Introduction

1.1 Nudibranchia

Nudibranchia in the class Gastropoda of phylum Mollusca, refers to sea slugs characterized by their loss of shell (Wägele and Willan, 2000). They belong to the subclass Heterobranchia (Wägele et al., 2014) including both shell-less gastropods and gastropods who can withdraw partly or completely into their shell. There are approximately 30 000 described species in this subclass. Nudibranchia is a monophyletic group and all members have lost their shell (Wägele and Klussmann-Kolb, 2005). The lack of shell has led to the development of different defense mechanisms. Amongst them is camouflaging in the same colors and shapes as the nudibranch's prey, one mechanism that is very common. Others secrete toxic substances making them taste bad, while some adopt cnidocytes (kleptocnids) from other organisms incorporating them and use them as their own defense mechanism (Bakken et al., 2021c).

The main suborders of Nudibranchia are Cladobranchia and Doridina. They all have a muscular foot, head, mantle and gills, but the placement and shape of these compartments as well as gills and digestive system contribute to the distinction between the two suborders. Cladobranchia have a reduced mantle with cerata, and anus placed on the right side of the body. Doridina have a prominent mantle with tubercles, gill plumes and anus laterally positioned (Bakken et al., 2021c). All Heterobranchs are hermaphrodites (Raja-Salleh et al., 2019).

Nudibranch's diets are known to consist of specific life-stages of Porifera, Cnidaria, Bryozoa, Crustacea, Mollusca, and Ascidiacea, and most species are considered as specialists on one food item (Wägele and Willan, 2000). Those gastropods which depend on prey with short generation times often have several generations themselves throughout the year as e.g. hydroid-feeders (Moen and Svensen, 2020).

1.1.1 Cladobranchia

The Cladobranchia nudibranchs have dorsal fingerlike structures (cerata) often symmetrically positioned (Glaser, 1910). Through their diet, cladobranch species can store stinging cells as kleptocnids (Wägele et al., 2014) (unexploded nematocysts) (Salvini-Plawen, 1972) from prey in designated cnidosacs in the end of their cerata (Rudman, 1999a). Examples of organisms that have these nematocysts are hydroids, anemones and scyphozoans with a benthic polyp stage (Rudman, 1999a). These nematocysts are found within cnidocytes in its host organism. When they are ingested by cladobranch nudibranchs, the nematocysts are separated from the cnidocyte before it goes through the digestive system to the cnidosacs. The stinging cells become useful as a defense mechanism to the nudibranch only after a proton transfer has occurred in the cnidosacs (Goodheart et al., 2018).

Cerata contain extensions of a channel of the digestive gland (Picton and Morrow, 1994, Thompson, 1988, Rudman, 1999a) and is otherwise filled with blood allowing for gill function and other gas exchange (Rudman, 1999a). The cerata themselves can look quite different from species to species. Some are classic tubular, while others can be puffy, flat, or even branched and bushy. The color of the duct and hence also the overall color we

perceive, depends on what the nudibranch eats, as it often acquires its colors from prey (Rudman, 1999a).

As for their other morphology, cladobranch nudibranchs have two rhinophores on their head, functioning like nostrils (Herdman, 1890). These rhinophores are crucial for prey detection and are specifically designed with a large surface area to volume ration to detect smells. The rhinophores can be retracted into pockets on the nudibranch's head if predators approach (Rudman, 1999b).

Cladobranch nudibranchs are known to eat Cnidaria and particularly hydroids. Lambert (1991) found that the size of the body plan and the structure of radula tongue on the nudibranchs determine what part of the hydroid polyps they consume (e.g. suction from stem or biting of whole polyps) (Lambert, 1991). Organisms within the clade Anthozoa are also popular, with prey items often being anemones, stony corals and Octocorallia (Goodheart et al., 2017).

In Norwegian waters, and the Trondheimsfjord in particular, cladobranch species like *Facelina bostoniensis* (Couthouy, 1838), *Facelina auriculata* (Müller, 1776), *Catriona aurantia* (Alder & Hancock, 1842), *Aeolidia filomenae* Kienberger, Carmona, Pola, Padula, Gosliner & Cervera, 2016, *Aeolidia papillosa* (Linnaeus, 1761)*, Favorinus branchialis* (Rathke, 1806), *Dendronotus frondosus* (Ascanius, 1774), and *Coryphella verrucosa* (M. Sars, 1829) are common (Evertsen and Bakken, 2005, Evertsen and Bakken, 2013). All these species (except *Coryphella verrucosa*) have been found in the framework of this project. Although species mentioned above were found prior to experiments, the only two species used were *Facelina auriculata* and *Facelina bostoniensis* as they were the only ones found in larger amounts.

Facelina auriculata has lamellate rhinophores as one out of two species. It is easily distinguished from its sibling, *F. bostoniensis*, by the blue colors on the cerata. It can get up to 38 mm long with a slender body. The dorsal side is covered with five to seven groups of cerata, with a distinct gap between first and second group of cerata. The body is translucent white with pinkish cast in the head region between head tentacles and rhinophores. *Facelina auriculata* can be found alongside the Norwegian coast from Oslofjord in the south, to Finnmark in the north. The species is known to feed on hydrozoans of the genus *Clava* Gmelin, 1788, *Tubularia* Linnaeus, 1758, and *Eudendrium* Ehrenberg, 1834 amongst others (Bakken et al., 2021a), but has also been observed preying on other species of nudibranchs (Moen and Svensen, 2020). The species is often found on lamina of *Laminaria hyperborea* (Gunnerus) Foslie, 1884 and sometimes *Saccorhiza polyschides* (Lightfoot) Batters, 1902, however it is unknown which prey they were found associated with in that study (Andersen, 2011). This species can be found from the tidal zone down to 40 meters or more (Moen and Svensen, 2020).

Facelina bostoniensis is the other species with lamellate rhinophores, thus enabling identification to genus level easily. It can get up to 55 mm and has a robust snail shaped body. The back is covered with five to seven groups of long, thin cerata, where the first group of cerata can stretch over the entire body length. Its body is transparent white with a pink tint around the head. The intestine color is red-brown and visible in the cerata. White pigmentation is present several places, but most characteristically down the entire tail. Similar to *F. auriculata*, *F. bostoniensis* eats hydroids, but feeding on the polyps of *Aurelia aurita* (Linnaeus, 1758), Pennatulacea and Stauromedusae is also documented (Bakken et al., 2021b). Ringed tubularia (*Ectopleura larynx* (Ellis & Solander, 1786)) happens to be the preferred habitat and prey for the nudibranchs *F. bostoniensis* as well

as for *C. aurantia*. It can be found down to about 30 meters depth. The eggs of this species are laid in a coiled up helix (Moen and Svensen, 2020).

Coryphella verrucosa can become up to 62 mm but is usually found at 15-25 mm. Its body is white translucent. The tail is pulled out into a pointy end with a medial white line. Head and mouth tentacles have white pigmentation on its tips, often as a line. Head tentacles often have a pink tint. This species has one variant with extremely short cerata, while another has longer cerata, that are positioned in five to seven groupings on each side of the body in both variants. There is a wide white pigmentation below the tip of each cerata, while the tip itself lacks any color. Alimentary canals are red, red yellowish or red brownish. *C. verrucosa* is often found on exposed areas in shallow water, or in deeper waters with flood tides down to 300 meters (Moen and Svensen, 2020). This species forage on hydroids and is also known to prey on sessile stages of *A. aurita* (Hernroth and Gröndahl, 1985a) and jellyfish polyps of the genus *Cyanea* Péron & Lesueur, 1810 (Moen and Svensen, 2020).

1.1.2 Doridina

The suborder Doridina has around 2000 morphologically described species. Although these nudibranchs do not have any fossil records (Valdés, 2004), it is clear that they once had shells, but that they completely lost them or partly reduced their shells over the course of evolution. Seeing that a shell has protective abilities, these nudibranchs have, similarly to the Cladobranchia, evolved another form of protection against predators and other dangers (Wägele and Klussmann-Kolb, 2005). In light of this, dorid nudibranchs are known to have calcareous spicules covering their mantle (Penney, 2008), composed of calcite ($CaCO_3$) and brucite ($Mg(OH)_2$) (Wägele and Klussmann-Kolb, 2005). An individual's dry weight therefore often comprise spicule-components for the largest part (Penney, 2008). These spicules are visible as glassy structures giving the sluggish organism a tactical advantage over predators. Larry G. Harris (1973) found that the cephalaspidean species *Navanax* sp. Pilsbry, 1895 would reject all dorids with a spiculose mantle, although the species in this genus usually preys heavily on nudibranchs. Another defensive strategy is how some Doridina species possess the ability to produce and store sulfuric acids in its active form (Wägele and Klussmann-Kolb, 2005).

Other than their spicules, Doridina are classified by a rosette of gill plumes surrounding their anus, towards the end of their dorsal mantel (Moen and Svensen, 2020). They have rhinophores acting as sensory organs on their heads, which also contain spicules. Additional to the sensory function, the rhinophores contain a lymphatic channel (Lisova and Vortsepneva, 2022).

Examples of dorid nudibranchs that can be found in Norwegian waters, and Trondheimsfjorden more specifically are *Onchidoris muricata* (O. F. Müller, 1776), *Polycera quadrilineata* (O. F. Müller, 1776), *Doto coronata* (Gmelin, 1791), and *Jorunna artsdatabankia* Neuhaus, Rauch, Bakken, Picton, Pola & Malaquias, 2021. All have been found in the framework of this project.

Onchidoris muricata has a flat and oval body shape. It can get up to 20 mm, however most often found around 15 mm long. The white-yellowish body is covered in a cape with ball-shaped tubercles. These tubercles contain calcareous spicules protecting them from predators. *Adalaria proxima* (Alder & Hancock, 1854) can be confused with *O. muricata*, however the former has round and pointed tubercles (Bakken et al., 2021d). Two lamellate rhinophores are present at the head (Bakken et al., 2021d) at some distance from each

other (Lisova and Vortsepneva, 2022), and several feather-like gills are present around the anus (Bakken et al., 2021d).

Onchidoris muricata is a generalist when it comes to feeding on bryozoans (Harvell, 1984). It uses a suction mechanism to ingest their prey (Chadwick and Thorpe, 1981), which according to Todd (1979a) is strenuous and restricting their body growth. There is not yet a consensus on whether the nudibranch removes the frontal membrane of the zooecium before sucking out the polypide (Chadwick and Thorpe, 1981), but it has been suggested (Ryland, 1977).

Onchidoris muricata is a hermaphroditic species (Havenhand and Todd, 1988) that reproduces in late wintertime (Lambert et al., 2016). Its eggs are laid in helical bands (Moen and Svensen, 2020). Their planktonic larvae are found in summertime with a meroplanktonic life-stage (Lambert et al., 2016, Havenhand and Todd, 1988) estimated to last for about 60 days (7°C). Eggs of *O. muricata* hatch after approximately 20 days (5°C), while eggs from the resembling species, *A. proxima* hatch after about 47 days (7°C). The larval stage of the latter only lasts for two days (Todd, 1979b).

Studies conducted in Connecticut by Clark (1975) found that *O. muricata* has an optimum temperature of 16°C with an ambient temperature of 5°C in the boreal to subarctic regions. In Norwegian coastal waters around the island of Hitra, *O. muricata* is found on *L. hyperborea* more often than other kelp species (Andersen, 2011). The thesis of Andersen (2011) emphasize the dominance of this dorid species on kelp around Hitra in March, compared to other months of the year.

1.2 Scyphozoa polyps as prey

The common name "Jellyfish" refers to the medusae-stage of gelatinous zooplankton. Their body consist of approximately 96% water and 0.5% carbon content. Most jellyfish belong to the phylum Cnidaria, which also contain corals (Duarte et al., 2014). The jellyfish medusae passively drift in the water column, acquiring their food through heterotrophy. As heterotrophs, they rely on the uptake of food items and their abundance is limited by food availability (Lucas et al., 2012). If food is scarce, adult medusae will allocate most resources into reproduction (Lucas and Dawson, 2014). However, a peak in food resources often correspond to a population abundance peak, often called jellyfish blooms. Such blooms can be caused by changes in environmental conditions (Lucas et al., 2012), and enhanced by anthropogenic activity. Environmental conditions that are considered to enhance blooms are changes in salinity, oxygen content, pH, and temperature. Additionally, the magnitude of temperature change is important (Lucas and Dawson, 2014).

True blooms are caused by natural seasonal cycles. Such blooms can thus happen to any organism that has both sexual and asexual reproduction strategies (metagenesis) when favorable environmental conditions are present. Biomass increases are then caused by population increases due to reproduction, growth and anthropogenic actions (Lucas and Dawson, 2014).

When a jellyfish bloom occurs, it is generally not appreciated by the public but rather looked upon as a plague (Duarte et al., 2014). For most people, jellyfish are unambiguously tied to their ability to sting although not all species have the stinging ability. It is true that some species of Cnidaria have fatal stinging cells (cubozoans), however most stinging Cnidaria are harmless to humans (Doyle et al., 2014).

Jellyfish blooms negatively affect coastal industries like aquaculture of salmon and mussels, as well as fisheries. Fish in pens are harmed when stung by jellyfish and fish nets can be clogged. On the contrary, these industries stimulate jellyfish population growth as e.g. excess nutrients from fish farms can stimulate secondary production and aquaculture installations provide additional settling surfaces for polyp settlement, in addition to predators being removed e.g. by fisheries (Purcell et al., 2007). Such jellyfish blooms can also affect the recreational value of coastal zones, like for example beaches and fishing spots (Myrvold, 2020). As for their ecosystem services, jellyfish contributes notably to regulating the climate through carbon sequestration. They also help transport carbon from surface-layers to the ocean floor when they die (Doyle et al., 2014).

A stage-specific characteristic in most scyphozoan jellyfish is crucial when it comes to the magnitude of blooms, namely their early, benthic life stage which perform asexual reproduction. Not all scyphozoans have a benthic polyp stage but rather metamorphose directly into a mini-medusa (e.g. *Pelagica noctiluca* (Forsskål, 1775)) (Ceh et al., 2015). The budding rate of polyps are affected by temperature and food availability. Budding results in one new polyp every 1-3 days (Lucas and Dawson, 2014).

The jellyfish *Aurelia aurita* (Linnaeus, 1758) belongs in the class Scyphozoa of the phylum Cnidaria. Although *A. aurita* is a zooplankton which entails a drifting form of life, the species possesses two life stages occurring through both a pelagic medusae and a sessile, benthic polyp. The particular life cycle involves both sexual and asexual reproduction (Figure 1) (Lucas, 2001).

The adult, sexually reproducing medusa stage releases eggs developing into planktonic planula larvae after fertilization (Lucas et al., 2012). These planula larvae settle in sheltered areas on hard substrate (Rekstad, 2019) after drifting in the water masses from 12 hours up to one week (Lucas, 2001). Settled planula larvae turn into polyps, also called scyphistoma. In turn, these polyps asexually reproduce and develop into strobilae. Strobilae are an intermediate stage before they metamorphoses into juvenile medusa called ephyra (Lucas et al., 2012). Because of the ever-lasting life stage of polyps, these stages are crucial in bloom formation of adult medusae.

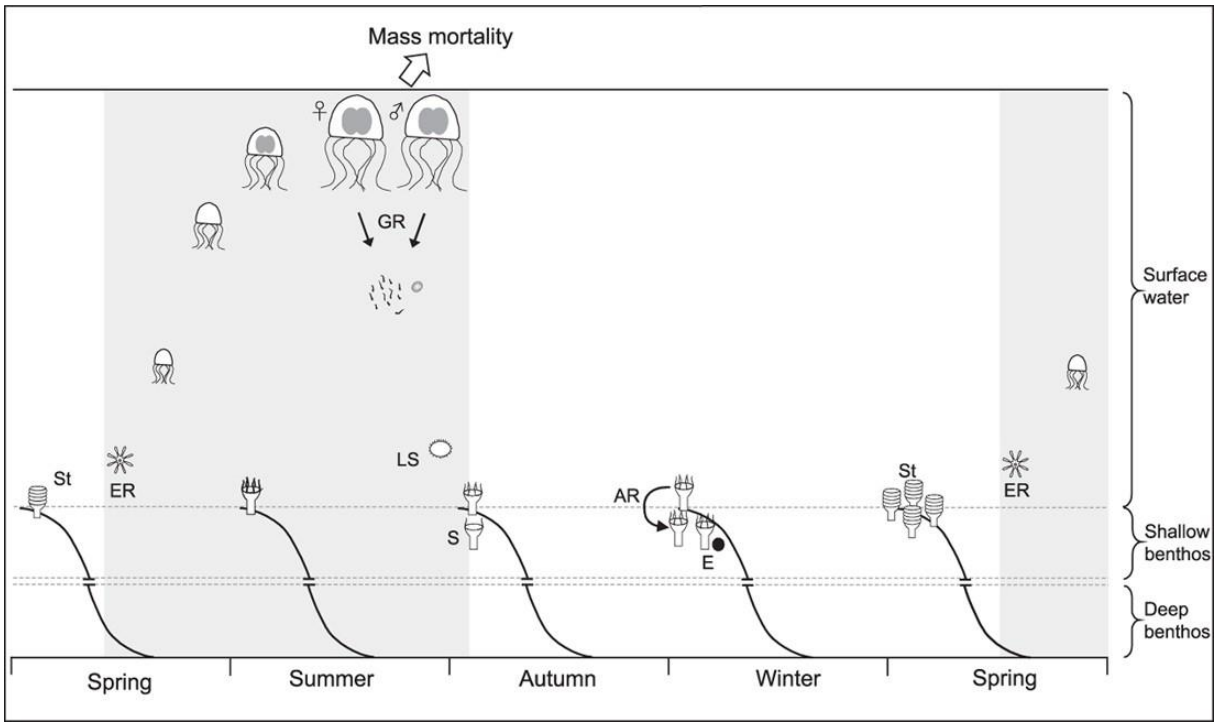

Figure 1: Illustration of The Metagenetic Life Cycle model for temperate regions described by Agassiz in 1860. Medusae develop from ephyra (ER) in early spring. Growth of medusae occur during summer until they reach sexual maturity (illustrated by two grey, oval gonads). Gametes are released (GR), and the fertilized planula larvae sink to the seafloor (LS). As the planula larvae settle (S), they metamorphose into polyps. Polyps reproduce asexually (AR) by strobilation (St) or budding (E, encystment). Polyps develop into strobilae and release ephyra in spring, completing the circle. Benthic stages are depicted with a white background, while the pelagic stages are depicted in grey background (Ceh et al., 2015).

1.3 Bryozoans as prey

The phylum Bryozoa of the clade Lophophorata, are divided into Gymnolaemata (WoRMS Editorial Board, 2022) which is the biggest class (Moen and Svensen, 2020) containing mostly marine species, and Stenolaemata – only marine species (WoRMS Editorial Board, 2022). Bryozoans are colony-forming animals (Moen and Svensen, 2020) that grow as epiphytes mostly on kelp but can also be found on other substrates (Harvell, 1984). Colonies of bryozoans consist of zooids with its tail section in a box-like structure called a zooecium. The zooids are made of chitin deposited in calcium (Aarnes, 2003). They are filter feeders aided by a retractable lophophore. Feeding occurs by creating a water-current where food items are carefully chosen depending on their size. As a colony, they help each other by creating a more amplified water current, again providing more food for all zooids (Shunatova and Ostrovsky, 2001).

Most bryozoans are hermaphrodites, meaning they have both reproductive organs (Moen and Svensen, 2020). Hence, they perform both sexual and asexual reproduction. Sexual reproduction releases a lecithotrophic larvae without the adult's characteristic external skeleton. When larvae settle, they perform metamorphosis in two steps. The first is securing a permanent attachment to the substrate and the other is transforming into an "ancestrula" which can bud into entire colonies (Stricker, 1989).

Bryozoans are prone to both direct and indirect predation, however, they have evolved some protection mechanisms. Some predators like Asteroidea will eat everything except the zooid skeleton around the animal (Gordon, 1972). The same is true for the feeding of the species *Onchidoris muricata* and *Adalaria proxima*, while *Polycera quadrilinieata* will consume the zooecium (skeleton-box) and the polypide (intestines) (Todd and Havenhand, 1989). Brittle stars on the other hand, will ingest large "sheets" of the entire bryozoan colony (Gordon, 1972).

1.3.1 *Membranipora membranacea* (Linnaeus, 1767)

Membranipora membranacea (Linnaeus, 1767), as well as *Electra pilosa* (Linnaeus, 1767) belong to the class Gymnolaemata, and the order Cheilostomata (WoRMS Editorial Board, 2022). They grow on *L. hyperborea* and *Laminaria digitata* (Hudson, J. V Lamouroux, 1813) but can also be found on other species of brown algae. *Membranipora membranacea* can be found down to 500 meters depth, and in brackish water. Zooids of *M. membranacea* are rectangle and form a typical brick-house pattern on its host substrate (Figure 2) (Moen and Svensen, 2020).

Membranipora membranacea larvae are planktonic for about four weeks, before they settle (Saunders and Metaxas, 2007). The initial settling of *M. membranacea* is enhanced when the larvae are in contact with algae. When it settles, it will spend a couple of days creating a twin ancestrula. This means there are two sources of asexual reproduction after settling (Stricker, 1989). In Nova Scotia, Canada, settling happens over the summer, with a maximum from July to September (Saunders and Metaxas, 2007). The same approximate period is true for the North-Atlantic in general, with a peak from June to August (Førde et al., 2016).

According to Saunders and Metaxas (2007), growing degree days and abundance of this bryozoan species indicate that an increase in global temperatures can cause a rise in *M. membranacea* abundance.

The colonies can grow several millimeters per day, as a response to predation (Moen and Svensen, 2020). *Membranipora membranacea* can grow spines on the skeleton of the zooid as a defense mechanism. However, studies suggests that it only prevents slow feeders including *O. muricata*. Additionally, the acquisition of these skeletal spines will most likely be at the expense of growth. Spines can be provoked after contact with a predator, and usually within two days according to Harvell (1984). The spines are made from chitin and can grow in the corner of the skeletal box (zooecium), or from the flexible frontal membrane (Harvell, 1984, Aarnes, 2003).

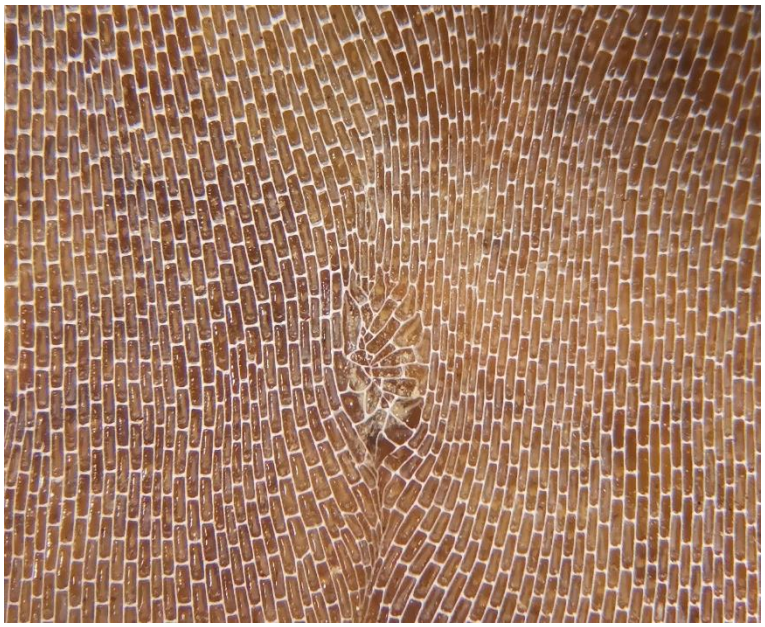

Figure 2: The bryozoan *Membranipora membranacea* and its rectangle physiology of the zooid. Photo: Live Agnethe Sørlundsengen Haugen.

1.3.2 *Electra pilosa* (Linnaeus, 1767)

Electra pilosa is found in the intertidal areas, usually in the first 50 meters (Hermansen et al., 2001). It can grow on brown and red algae but also on hydroids, other bryozoans, and crustaceans. If colonies get large, they can also detach from the epiphytes and start forming free-standing fringy colonies. Zooids are oval or rounded and have two thorns in one end of the zooid, and one large spike in the other end. The colonies form star-like shapes patchily distributed on its host substrate (Figure 3) (Moen and Svensen, 2020). Contrastingly to *M. membranacea*, *E. pilosa* forms only one ancestrula zooid per settled larvae (Stricker, 1989).

According to Førde et al. (2016), encrustation of *E. pilosa* at the islands of Frøya and Reksta at the Norwegian coast was more abundant on brown algae lamina around April, while *M. membranacea* takes over this role in late summer. The planktonic larvae (cyphonautes) are released in late summer (Førde et al., 2016) and are free-floating for about two months or more thereafter (Førde et al., 2016, Ryland and Stebbing, 1971). Settling of this species is also more likely to happen in close proximity to already settled colonies of *E. pilosa*, indicating some sort of chemical cues before hatching (Ryland and Stebbing, 1971).

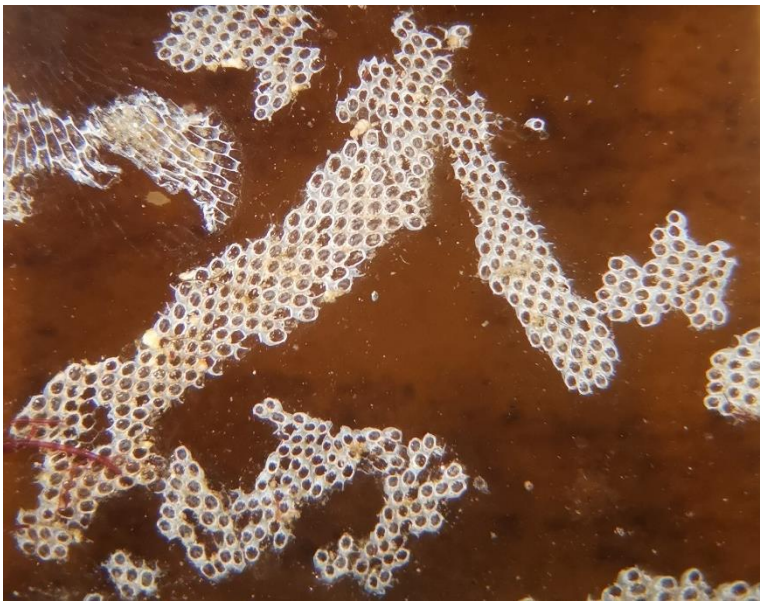

Figure 3: The bryozoan *Electra pilosa* and its oval physiology of the zooids, as well as a patchy, often star-shaped distribution of the colony. Photo: Live Agnethe Sørlundsengen Haugen.

1.4 Predator-prey interactions

1.4.1 *Aurelia aurita* polyps and cladobranch nudibranchs

From an evolutionary perspective, most Cladobranch nudibranchs have commonly been feeding on Hydrozoa. A complete taxon shift in feeding preference is therefore unlikely. However, based on ancestral prey preference, there is evidence for a prey preference shift to Scyphozoa polyps at some point (Goodheart et al., 2017). Studies showed that different species in particular regions have the potential to utilize *A. aurita* polyps as their main food source (Takao et al., 2014, Hernroth and Gröndahl, 1985a).

Seeing that *A. aurita* planula larvae settle on hard substrate, predation on the sessile polyp stage by nudibranchs could reduce the medusae population by 10-30 ephyra per polyp thus affecting their bloom potential considerably (Hernroth and Gröndahl, 1985b, Chi et al., 2019). As for recently settled polyps, predation before strobilation (asexual reproduction) could reduce the amount of ephyrae released from a particular polyp down to zero. This could contribute to a natural control of medusae populations that are able to reproduce sexually and thus alter the formation of local jellyfish blooms (Hernroth and Gröndahl, 1985b).

Metabolism in heterotrophic organisms is normally affected by increasing temperatures in the way that ingestion rates and growth rates are enhanced at higher temperature (metabolic theory of ecology) (Clarke, 2006, Brockington and Clarke, 2001). Thus, another angle to the problem is that polyp ingestion rates of nudibranchs might increase with temperature (Myrvold, 2020). Although nudibranchs are prone to large temperature variations in coastal, inter-tidal zones of e.g. Trondheimsfjorden, both nudibranchs and polyps can be affected by temperature increases. A basal form of knowledge would suggest that an increase in ingestion rate due to temperature, would result in an increase in nudibranch growth. Such a potential increase in ingestion rates and growth at elevated temperature conditions would lead to an increase in the nudibranch's demand for food

because of increased energetic demands, and potentially also increased nudibranch populations due to alternations in reproduction rates.

1.4.2 Bryozoa and dorid nudibranchs

Dorid nudibranchs have a wide range of prey items. In temperate regions, their preferred prey tends to be bryozoans, while sponges dominate dorid's feeding preference in tropical regions (Faulkner and Ghiselin, 1983). *Membranipora membranacea* and *E. pilosa* are native bryozoan species in Norwegian waters, preferred by the nudibranchs *Polycera quadrilineata* and *O. muricata* respectively (Pratt and Grason, 2007). Although bryozoans are not in the spotlight when it comes to media coverage, kelp often is. The aquaculture industry has begun increasing its kelp farming and thereby usage and development in regular household products. *Membranipora membranacea*, however, can cause macroalgae lamina to become more fragile and easily break during bad weather conditions (Dixon et al., 1981, Pratt and Grason, 2007, Scheibling et al., 1999, Lambert et al., 1992). Additionally, growth is impaired due to blockage from sun-light to reach the chlorophyll in the lamina (Pratt and Grason, 2007).

As *O. muricata* reproduces in wintertime, their juveniles appear from late spring to early summer. They have an annual life cycle and are therefore adults from December to March (Table 1) (Lambert et al., 2016).

Both *E. pilosa* and *M. membranacea* have an annual life cycle. *Membranipora membranacea* settle from May to September with a peak in September. This means that the colonies will be at their highest density at the same time as the growth stage of juvenile *O. muricata* (Denley et al., 2014). Life cycle investigations of *E. pilosa* are insufficient, however, Førde et al. (2016) found that the presence of this species was higher in April than later in the summer months. As senescence has been discovered in bryozoans (Bayer and Todd, 1997), it is likely that the death of *E. pilosa* zooids in early summer and a following settling of *M. membranacea* allows for niche differentiation between the two species (Denley et al., 2014, O'connor et al., 1979).

Studies have indicated that there might be a shift in prey preference for *O. muricata* over its annual life cycle. The species seems to be found on *E. pilosa* between December and May, and on *M. membranacea* between June and November (Lambert et al., 2016). This coincides with the results from Førde et al. (2016) in Norwegian waters. As *O. muricata* is a generalist with a preference towards Bryozoa, the bryozoans *M. membranacea* and *E. pilosa* are both highly prone to predation by the nudibranch *O. muricata*.

Nybakken and McDonald (1981) assessed the radula size and teeth of nudibranch species feeding on Bryozoa and found that *O. muricata* had 12 teeth and a relatively narrow radula structure. Additionally, the study emphasizes the generalization in feeding of *O. muricata* as it is recorded to feed upon bryozoans that are fleshy/soft, calcified, uncalcified, encrusting, as well as with and without avicularia (mandible). The exception to its feeding habits is vertically standing Bryozoa (see article for more details) (Nybakken and McDonald, 1981).

Table 1: Life cycle events throughout the year for the nudibranch *Onchidoris muricata* and its prey items, the bryozoans *Electra pilosa* and *Membranipora membranacea* (Lambert et al., 2016).

Month / Species	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**
Nudibranch *Onchidoris muricata*	Adults reproduce			Juveniles appear		Growth phase						Adults
	Found on *E. pilosa*					Found on *M. membranacea*						Found on *E. pilosa*
Bryozoa *Electra pilosa*				Abundance peak followed by senescence								
Bryozoa *Membranipora membranacea*						Settling phase						

1.5 Study locations in Trondheimsfjorden & Frohavet

The Trondheimsfjord is 126 km long from Agdenes (west) to Hjellbotnen (north-east). Trondheimsfjorden is the third longest fjord in Norway with a volume of 235 km^3, mean depth at 165 meters and deepest point at 617 meters in Ytterfjorden. Recreational value, transportation and work-related value are important aspects of the fjord due to its relevance in mid-Norway (Bakken, 2000).

Trondheimsfjorden has four sills dividing the fjord into three main basins: Ytterfjorden (outer part), Midtfjorden (middle part) and Beistadfjorden (inner part). Although the sills are quite shallow, they are still deep enough to allow for sufficient water exchange (Bakken, 2000). This is crucial for e.g, dispersal of nudibranch veliger larvae, Bryozoa larvae and Scyphozoa planula larvae. Average tidal range in Trondheimsfjorden is 1.6 meters (Bakken et al., 2000). The outermost sill creates a relatively shallow area between Storfosna and Agdenes, and between the island group of Hitra and the mainland. The fjord ends along the line between Storfosna and Agdenes (Bakken, 2000). The temperature conditions in Trondheimsfjorden are characterized by the inflow of deep water every year (Jacobson, 1983). These deep-water masses are between 6.5°-7.4°C when they arrive in February before stagnating between March and June. Jacobson (1983) summarized annual temperature variations in Trondheimsfjorden between 1963 and 1980, indicating a large fluctuating pattern between years as well as between the different depths of the fjord.

Most fjord ecosystems have hard, rocky substrates in steep slopes, while soft clay sediments characterize the basin areas. These sediments often come from terrestrial areas by wind and are carried by rivers. This is characteristic for most of the Trondheimsfjord as well (Bakken, 2000).

The area around Frøya is part of Frohavet, a more remote and exposed location (Bakken, 2000). Dragneset, Frøya, is characterized by sandy bottom. The sediments consist of arenaceous clay and muddy sand (Folk, 1954, Norges Geologiske Undersøkelse - NGU, 2021).

1.6 Aims of the thesis

In this study, the key questions regarding nudibranch feeding ecology that were addressed focused on potential prey items for nudibranchs in Trondheimsfjorden and Frohavet, and how their feeding behavior looks like. In the light of a global increase in sea surface temperatures, the question how changes in temperature affect the ingestion rates of nudibranchs on scyphozoan polyps and bryozoans was also addressed. Will this increase in sea surface temperatures enhance nudibranch ingestion? And will such an increase in sea surface temperatures cause a shift in trophic match between prey and predator?

The aims of this thesis were to understand the feeding ecology of nudibranchs living in the Trondheimsfjord and Frohavet, and how temperature affects their feeding potential and behavior, with special emphasis on the feeding on Scyphozoa polyps and Bryozoa zooids.

The hypotheses were (1) increasing sea surface temperatures influence nudibranch ingestion rates, (2) different nudibranch species show differences in ingestion rates when feeding on scyphozoan polyps, (3) nudibranch ingestion rates differ depending on the age and size of scyphozoan polyps, and (4) nudibranchs show temperature-dependent ingestion rates when feeding on different Bryozoa species.

2 Methods

The study was mainly conducted during two periods with set-up in spring 2021, two overlapping sequences of field work and lab experiments in the fall.

2.1 Study area: Frohavet and Trondheimsfjorden

Collection of nudibranchs and Scyphozoa were done in the intertidal areas of the Trondheimsfjord, Norway, e.g. just off Trondheim Biological Station (TBS) and Dragsneset at Frøya.

Nudibranchs of the species *F. bostoniensis* were collected on August 28, 2021, at Planktonic AS pier (63°26.57'N, 10°20.92'E). Nudibranchs of the species *F. auriculata* were collected from Dragsneset, Frøya (63°47.41'N and 8°45.12'E), but mainly at TBS (63°26.46'N and 10°20.93') on October 11, 2021.

Specimens of *A. aurita* were collected at Sjøbadet, Trondheim (63°26.13'N and 10°23.41'E) and TBS (63°26.46'N and 10°20.93'). Settling plates were deployed at TBS (63°26.46'N and 10°20.93') on June 28 as well as at Dragsneset (63°47.41'N and 8°45.12'E) on July 3, 2021.

2.2 Preparations

2.2.1 Settling plate rig set-up

The first part of the study was conducted by deploying settling plates at TBS and at Frøya. The settling plate design was adopted from the work of Mathias Rekstad (2019), and adjusted to previous set-ups (van Walraven et al., 2016). Round settling plates of 7.5 cm in diameter were cut out from grey PVC material (Figure 4). Plates were roughened using sandpaper, then attached using zip ties to the underside of regular bricks (28.5 cm x 8.5 cm x 8.5 cm). A line with buoys held ropes with bricks attached at the end. The line consisted of 15 bricks at 1 meter depth. A mooring connected to a large buoy with several meters of excess rope allowed for up to 3 meters difference in tides (Figure 5). To avoid fretting of the rope against the zip ties as well as the brick, electrical tape was wrapped around the rope in that area.

The set-up of bricks as well as the attachment to land was checked throughout the immersed period so that any entangling was detangled as soon as possible. Due to boat traffic to and from the pier at TBS, the brick set-up was moved twice after first assembly. Settling plates were left in the sea for 3.5 months, June 28-October 11, 2021.

When collected and brought into the lab, the bricks were dismantled, and the PVC plates and bricks were placed in separate aquariums. The fouling communities on the bricks, ropes and PVC plates were assessed and identified. Plates were planned to be used in a temperature gradient table with three different temperature treatments, in order to assess selective feeding and prey preference by recording removal of prey in a natural fouling community over time. The bricks served as an effective way of collecting nudibranch specimens from the field.

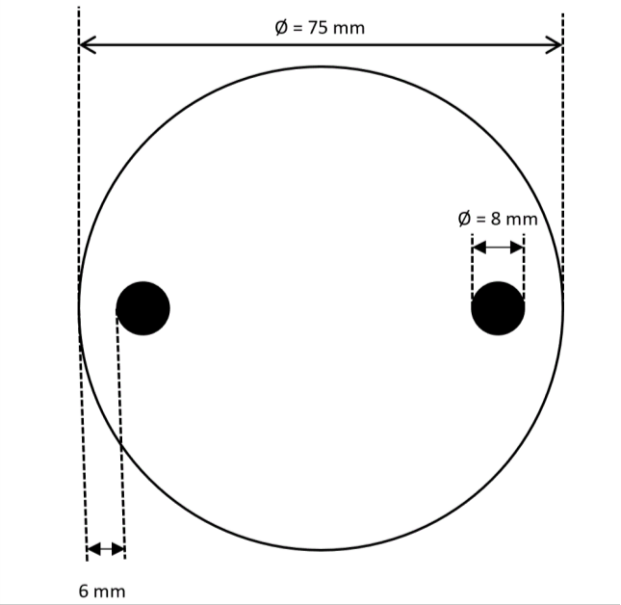

Figure 4: Illustration of the PVC plates used for settling experiments. Two parallel holes of 8 mm width were made with a distance of 6 mm from the edge of the circle, for the zip ties to be fastened in. Figure: Rune Bjørgum.

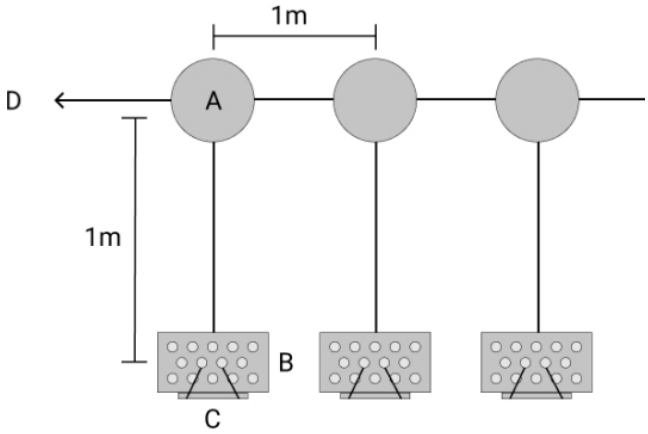

Figure 5: Plate rig set-up adjusted from Rekstad (2019). Buoys (A) attached to a long rope with 1 meter between each buoy. Bricks (B) hanging 1 meter below each buoy, allowing for them to follow the tides. PVC-plates (C) were attached with zip ties underneath each brick. A mooring point (D) was set in the end of the rope.

2.2.2 Nudibranch collection

Numerous enquiries were put out to local professional and recreational divers starting in May 2021. Despite this, there were no nudibranchs collected over the summer (perhaps due to murky waters (Shi and Wang, 2010)). The bay at TBS was scanned for nudibranchs multiple times starting in May by snorkeling, as well as other locations around Trondheim and Frøya.

The first nudibranch sighting happened on August 18, 2021, among *Ectopleura larynx* colonies on artificial substrate at the pier of Planktonic AS in Trondheim. The surface was densely covered with these hydroids, and more specimens were found on August 28, 2021.

Specimens of *F. bostoniensis*, *F. auriculata* and *C. aurantia* were collected there. All but one specimen of *F. auriculata* were found on the settling plate rigs in the intertidal areas just off TBS. They were all found among hydrozoans that had settled on the rough surface of the bricks. *Onchidoris muricata* inhabiting lamina of *L. hyperborea*, *Laminaria digitata* and S*accharina latissima* (Linnaeus) C.E.Lane, C.Mayes, Druehl & G.W.Saunders, 2006 were collected by snorkeling in the bay at TBS.

An indoor flow-through aquarium was set up at TBS for interim storage of collected specimens. The outlet of the aquarium was covered with a plankton gaze with a mesh size of 230 µm. Specimens were fed with hydroids and Bryozoa *ad libitum*.

2.2.3 Specimens collection and planula larvae extraction

Adult ripe female medusae of the moon jellyfish *Aurelia aurita* (Scyphozoa) were collected in the Trondheimsfjord off the coast of TBS and Trondheim city. The search for medusae occurred on several occasions, but in general densities were low and specimens not close enough to the shore to be easily collected. Female medusae of *A. aurita* were spotted at Ilsvika beach on August 28 and by the TBS pier on August 29, 2021. Female individuals were collected from the shore outside of Sjøbadet (Brattørkaia) on August 30, at TBS pier on August 31 and from TBS beach at night on August 31, 2021 (SST 12.4°C). Their bell diameter were 19 cm, 20 cm, and 17 cm respectively.

Specimens were brought to the wet lab at TBS in large buckets and immediately put into a large indoor aquarium with running seawater. Hereafter, seawater refers to seawater pumped from a depth of 80 meters, right off TBS, then filtered through a sand filter and a 20 µm gauze. The bell size of the medusae was measured using a ruler. Afterwards, individual medusae were kept in a shallow tray with seawater over night and until all or most of its planula larvae were released. Using a plastic pipette, planula larvae were sucked from the oral arms and transferred to a clean bucket. The bucket containing planula larvae and seawater was filtered through a 125 µm sieve, rinsing out any large waste particles. Thereafter, the seawater containing planulae was poured through a 35 µm sieve, hence collected in the sieve. Planula larvae were then transferred to a smaller container creating a more concentrated solution. Three subsamples of 1 mL were extracted from the concentrated solution using a Finnpipette. The subsamples were put into separate Eppendorf tubes, before Lugol's iodine solution was added to fix the larvae. Afterwards, the subsamples were counted using a counting chamber and an inverted plankton microscope (LEICA DM IRB) to achieve an estimate of the total number of planula larvae. The calculations were done according to formula I-IV (Appendix 1).

$$\text{Mean of planula larvae } 1\ \text{mL}^{-1} = \frac{\text{subsample 1+subsample 2+subsample 3}}{3} \quad \text{(I)}$$

$$\text{Planulae larvae in solution} = 90\ \text{mL} \times \text{mean of planula larvae } 1\ \text{mL}^{-1} \quad \text{(II)}$$

$$\text{Planula larvae in each beaker} = \frac{\text{planula larvae in solution}}{\text{\# of beakers}} \quad \text{(III)}$$

$$\text{Planula larvae } 1\ \text{mL}^{-1} \text{ in each beaker} = \frac{\text{added planula larvae solution}}{\text{mL in beaker}} \quad \text{(IV)}$$

Colonies of Bryozoa were found together with their predators, *O. muricata* on brown algae lamina and collected by snorkeling at the bay of TBS. Lamina of *L. hyperborea*, *Laminaria*

digitata and S*accharina latissima* were cut from the stamen and holdfast and put in aquariums with running seawater.

2.2.4 Settlement of planula larvae

Settlement of planula larvae was acquired by using plastic petri dish lids (5.4 cm diameter) and a temperature gradient table. The petri dish lids were roughened using sandpaper and marked for easier counting (Figure 6). 4.5 mL of planula larvae solution was transferred to 15 sterile urine beakers (100 mL) on September 1, 2021 and filled up to the 100 mL mark by adding filtered seawater. All beakers had a final planulae concentration of ca. 7 planulae mL^{-1} (formula I-IV, Appendix 1).

The urine beakers were then transferred to 800 mL glass beakers in the temperature gradient table, containing 200 mL of acclimatized seawater. The 15 urine beakers were placed in three different temperature treatments (11°C, 13°C, 15°C), with five replicates in each (see section 2.2.6 Temperature gradient table). The temperature regimes were the same for all experiments, where the intermediate temperature was chosen based on the ambient sea surface temperatures at the time of specimens' collection (13.8°C). The other two temperatures were chosen to represent the possible changes in sea surface temperature during the year, with one reduced temperature at 11°C and one elevated temperature at 15°C.

On September 2, 2021, petri dish lids were placed floating on the water surface with the roughened surface facing downward (Figure 7). The settling experiment was continued until no more free-swimming planula larvae were visible in the beaker (September 6). After settling, the petri dish lids were gently removed from the urine beakers and photographed while emersed in a glass petri dish (9 cm) filled with filtered seawater (September 7). Photographs of petri dish lids covered half of each lid (e.g. section 1-4, and 5-8), to maximize the resolution of the planulae. The digital camera used was a Nikon D3000 with a Nikon AF-S DX NIKKOR 18-55mm f/1:3.5-5.6 G lens. Individual photographs were uploaded into the software Inkscape 1.1, settled polyps were marked, and then counted with the aid of the software. The seawater was changed before the petri dish lids were put back into the urine beaker (September 7). September 8, the urine beakers were removed to provide polyps with a greater volume of seawater in addition to enhanced oxygen flow. They were then kept in the 800 mL glass beakers, which were filled with approximately 600 mL of filtered seawater.

The remaining planula larvae that were not used to set-up the initial experiment were transferred to shallow plastic trays filled with filtered seawater. Rough plastic sheets (Olmec Inc Printing Sheets, 21x30 cm) were placed floating on the water surface so that the remaining planulae could settle on the plastic sheets (see Figure 10 for settlement example). Once settled (September 6), seawater was changed (September 7), and polyps were fed *Artemia* sp. Leach, 1819 *ad libitum* (September 12).

For all polyp settlements, water exchange and feeding occurred on alternating days, every other day starting September 12, 2021.

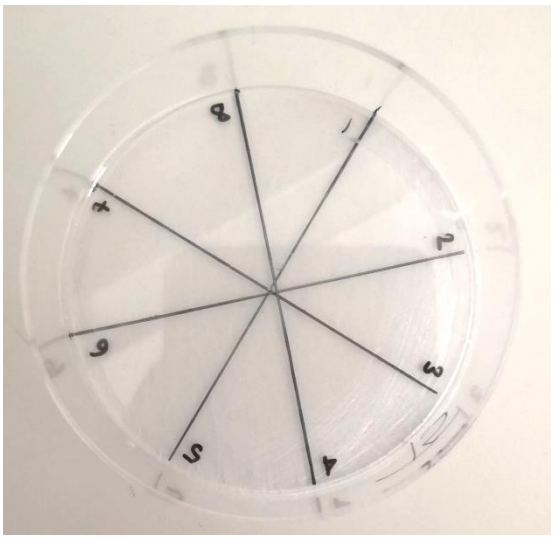

Figure 6: Petri dish lids divided into sections and numbered to facilitate polyp counting. Lids were marked with treatment name on the edges. Photo: Live Agnethe Sørlundsengen Haugen.

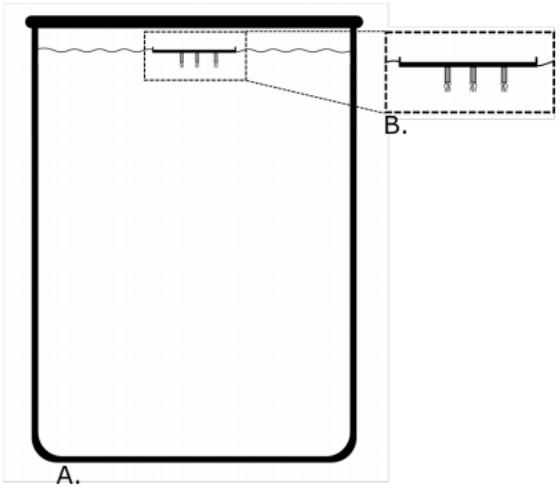

Figure 7: A. Beaker set-up in temperature gradient table for settling of planula larvae. B. Petri dish lids with *Aurelia aurita* polyps hanging upside down after settlement (according to Myrvold, 2020).

2.2.5 Cultivation of *Artemia* sp. feed for polyps

Cultures of *Artemia* sp. were created on September 8, 2021, and every other day until experiments finished. An 800 mL glass beaker was filled with 550 mL filtered seawater in a temperature (15.2°C ± 0.61°C) and light controlled room (1.5 µmol m^{-2} s^{-1}). A tip of a teaspoon with unhatched cysts was added to the beaker. Air bubbling was added to secure that all *Artemia* sp. were suspended in the water column. Cysts hatched after two days (September 10). As the lipid content of the *Artemia* sp. was too high to serve as a diet for the polyps immediately, they were fed only after two days of hatching (Stian Myrvold, personal communication September 9, 2021). To guarantee a continuous culture throughout the experimental periods, another culture was set up every other day.

2.2.6 Temperature gradient table

A temperature gradient table was used to standardize temperatures across and within experiments and treatments. Two Julabo CORIO CD-300F Refrigerated-Heating Circulators were placed on opposite sides of an aluminum table like the one illustrated in Figure 8. Five 800 mL glass beakers were used in three temperature regimes aligned horizontally in the table. The three different temperatures used were as mentioned in section 2.2.4 Settlement of planula larvae, based on the ambient sea surface temperature at the time of specimens collection (13.8°C, hence 13°C), with one elevated (15°C) and one reduced temperature treatment (11°C) in addition. The temperature treatments were the same for all experiments using cladobranch nudibranchs preying on scyphozoan polyps and for all experiments using dorid nudibranchs preying on bryozoans. All temperature measurements are found in Appendix 2: Temperatures, Table 3.

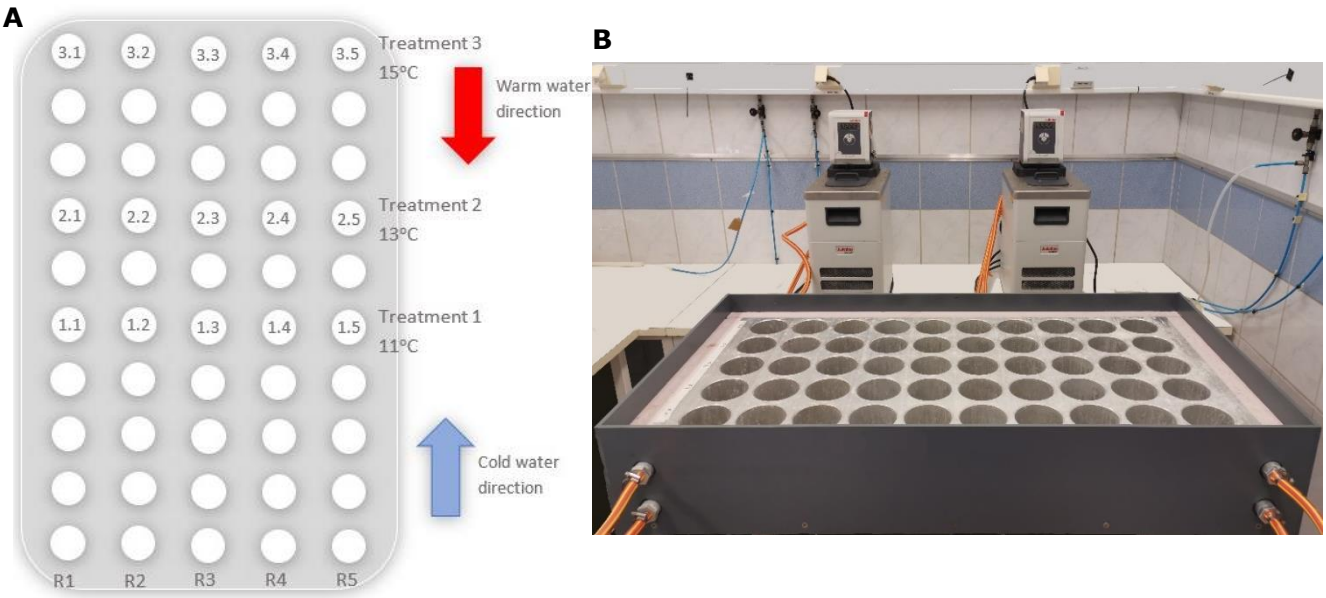

Figure 8: A. Set-up of temperature gradient table with three treatments at different temperatures (11°C, 13°C and 15°C), and replicates in horizontal rows (R1-R5). Arrows indicating direction of warm water and cold water flow along the aluminum plate of the temperature gradient table using the two Julabo temperature regulators. Figure adjusted from Myrvold (2020). B. Julabo temperature regulators with hoes leading into the aluminum plate creating the temperature gradient across the treatments. Photo: Live Agnethe Sørlundsengen Haugen.

2.3 Temperature-dependent ingestion rates of nudibranchs preying on scyphozoa polyps and bryozoan zooids

Acclimation of nudibranch specimens prior to the experiments was crucial to avoid unnecessary temperature stress of the specimens. This was ensured by keeping specimens of nudibranchs in indoor tanks for several days before the experimental trials (Takao et al., 2014). 24 hours prior to the experiments, specimens to be used were moved to a lab with controlled light (1.5 µmol m^{-2} s^{-1}) and temperature regimes (15.2 ± 0.61°C). Additionally, specimens were left in a tank without food and hence starved for 24 hours during the acclimatization period prior to the experiments (Takao et al., 2014, Àvila et al., 1998).

The length of nudibranchs was measured using a tape measurer from the base of their head tentacles to the tip of their tail before adding them to the experimental units. Nudibranchs were randomly allocated to a treatment. One hour before the experiments started, the specimens to be used were placed in their respective temperature regime (11°C, 13°C or 15°C) to be further acclimated to the conditions in the experimental units. Temperature of seawater in beakers was checked and noted before each trial.

2.3.1 Experiments with cladobranch nudibranchs preying on scyphozoa polyps

To distinguish if there was a difference between ingestion rates by certain species of cladobranch nudibranchs, an experiment was conducted using planulae settled on artificial substrates in the lab. The species used were *Facelina bostoniensis* and *Facelina auriculata*. Nudibranchs and polyps were kept in separate containers until the experiments started. During the cultivation and acclimation period prior to the experiments, nudibranchs were not exposed to polyps as food item at any point.

Before the experiments started, all petri dish lids were photographed and counted again, in case any polyps had died since last counting. After measuring the length of each nudibranch, individual nudibranchs were then gently transferred using a spoon and placed individually to the bottom of each urine beaker (100 mL) (Hoover et al., 2012). The urine beakers were put inside the 800 mL glass beakers in the temperature gradient table (five replicates in each treatment), filled with 200 mL seawater. Thereafter, the petri dish lids with settled polyps on were placed floating on the surface (Figure 9).

Experiment 1

For the 1st experiment with *F. bostoniensis* (September 15), each nudibranch specimen stayed in the beaker for 60 minutes from first contact with the food items before it was taken out (referred to as FB1 in figures). Due to the short period inside the beakers and to avoid disturbance of nudibranchs during feeding, no aeration stick was used. Three treatments were used, temperature regime 1, 2 and 3. Petri dish lids with remaining polyps were put back into the temperature gradient table for further growth. Between experiments, the urine beakers were cleaned with freshwater to remove nudibranch slime and potential chemotaxis from either animal (Hoover et al., 2012).

Experiment 2

For the 2nd experiment with *F. bostoniensis* (September 22) each nudibranch specimen was kept in the beaker for 15 minutes from the first contact with polyp prey (referred to as FB2 in figures). The petri dish lids with the most polyps remaining were used. Two treatments were used, temperature regime 1 and 3.

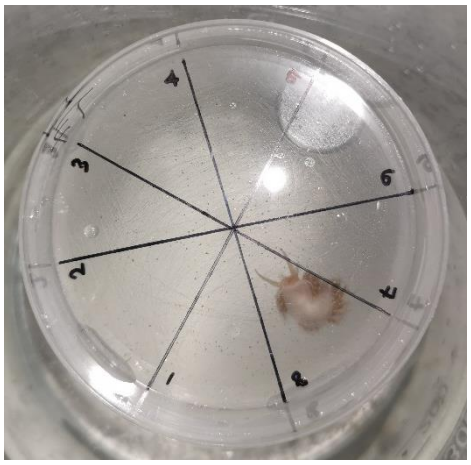

Figure 9: The nudibranch *Facelina bostoniensis* feeding on polyps of *Aurelia aurita*, hanging upside-down under a petri dish lid. Photo: Live Agnethe Sørlundsengen Haugen.

Experiment 3

In the 3rd experiment (October 13, 2021), the same experimental set-up as September 22 was used with the species *F. auriculata* (referred to as FA in figures). As the petri dish lids were mostly empty after two experimental runs, the plastic sheets where additional polyps had been settled were used. 10 semi-round cut-outs with a diameter of 8 cm were made where density of polyps was highest. A stencil marked with a cross from end to end was used to separate into compartments for easier counting. Plastic sheet cut-outs were placed emersed in water in a glass petri dish, with the stencil underneath (Figure 10). They were photographed, marked, and counted before experiments started. Due to the light weight of the plastic sheets, the cut-outs were attached to the underside of plastic petri dish lids like the ones used in previous experiments, using waterproof Tesa Extra Power Universal Tape.

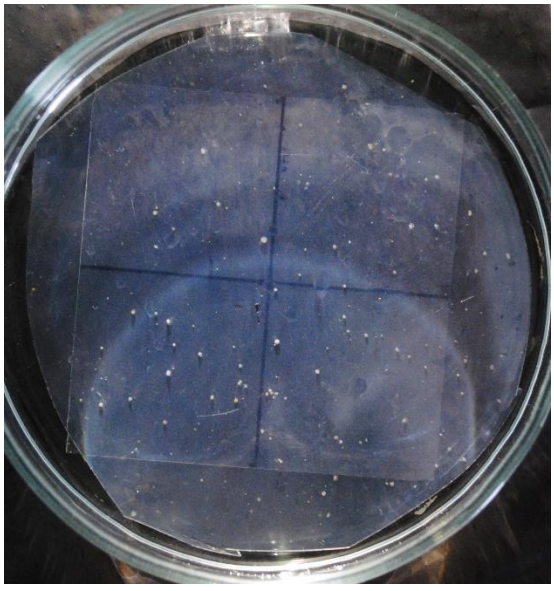

Figure 10: Plastic sheets (Olmec inc printing) with settled polyps of *Aurelia aurita*. Stencil with sections placed underneath the glass petri dish for easier counting. Photo: Live Agnethe Sørlundsengen Haugen.

Ingestion rates were calculated after the petri dish lids were photographed and polyps were counted.

2.3.2 Experiments with dorid nudibranchs preying on Bryozoa zooids

To distinguish if there was a difference between ingestion rates of the nudibranch species *O. muricata* on different species of bryozoans, an experiment was conducted using bryozoans inhabiting lamina of *L. hyperborea*, *L. digatata* and *S. latissima.* The different species of bryozoans used as prey items were *M. membranacea* and *E. pilosa*.

Experiment 4

For the 4th experiment (November 11, 2021), 2x2cm squares were cut out from the lamina of *M. membranacea* where dense patches of bryozoans were present. Cut-outs were photographed while emersed in seawater in a glass petri dish. Using the software Inkscape 1.1, the empty bryozoan zooids were marked and counted (Figure 11). Thereafter, the cut-outs were placed at the bottom of the urine beakers filled with seawater, in the same set-up as previous experiments. Temperature regime 1 and 3 served as the different treatments. The nudibranch specimens of *O. muricata* were placed on top of the cut-outs and left there for 4.5 hours untouched. Afterwards, the predators were removed, and the cut-outs photographed while emersed in seawater. Empty zooids were counted and ingestion rates calculated.

Experiment 5

For the 5th experiment, the same procedure as in experiment 4 was conducted where the nudibranch *O. muricata* was offered the bryozoan *E. pilosa* as prey items on November 16, 2021.

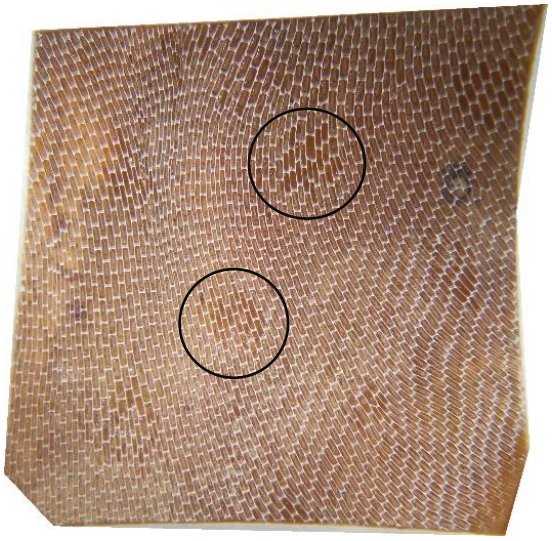

Figure 11: Example of a cut-out of kelp lamina from *Laminaria hyperborea* inhabiting the Bryozoa *M. membranacea* before experiments started. Circles are surrounding areas with dark, empty zooids of Bryozoa. Photo: Live Agnethe Sørlundsengen Haugen.

2.4 Data processing

2.4.1 Carbon and nitrogen analysis

After the experiments, predator specimens were collected for carbon:nitrogen analyses. 20 individual specimens of *O. muricata* were weighed (wet weight in mg) and their length measured (mm), before being transferred to 1.5 mL Eppendorf tubes. The Eppendorf tubes were marked, then placed on ice, slowly decreasing the body temperature of the specimen, before transferring them to a -20°C freezer. Thereafter, the Eppendorf tubes were removed from the freezer, put in a tray, and oven dried at 60°C for 48 hours. In between drying and further weighing, the tubes were put in a desiccator preventing moisture to reappear. The dried matter was scraped into premeasured tin capsules, and the dry weight was measured (mg). Tin capsules were packed tightly on a metal plate using forceps to avoid having any air inside. Each packed sample was then put into a 96 plastic well plate with a lid. Using standard C:N methods and an organic elemental analyzer (vario EL cube, Elementar), the nudibranch samples were burned to obtain the carbon and nitrogen equivalents.

The same procedure was done with 20 samples of the nudibranch *F. bostoniensis*. The carbon values for these nudibranch samples were plotted against their size. Thereafter, the trend line was extrapolated to extract the sizes of nudibranchs along the regression line.

Data on polyp carbon concentrations of *A. aurita* polyps were gathered from published data. Carbon values from Cawood (2012), Myrvold (2020), Kamiyama (2011), Ikeda et al. (2017), and Chi et al. (unpublished data) were extracted. The values were plotted in a scatterplot with carbon concentration (µg C polyp^{-1}) as a function of polyps age (days from settling) and extrapolated to apply for the ages of the polyps used in these experiments (Figure 19). The formula of the trend line was used to find the carbon concentration of polyps at the different ages (formula V-VII).

$$y = 1.2288\text{x} - 3.3045 \quad \text{(V)}$$

$$y = 1.2288\,(9\,days) - 3.3045 \quad \text{(VI)}$$

$$y = 7.7547 \approx 7.755 \quad \text{(VII)}$$

2.4.2 Ingestion rate calculations

Ingestion rates were calculated in Excel using data on amount of prey consumed divided by the experimental time (providing prey min^{-1}). Formula VIII-IX show the calculations.

$$\text{Ingestion rate} = \frac{\#\text{ prey consumed}}{\text{experimental time (minutes)}} \quad \text{(VIII)}$$

$$\text{Ingestion rate} = \text{prey min}^{-1} \quad \text{(IX)}$$

Carbon specific ingestion rates were calculated using the amount of prey consumed per minute, the carbon concentration of nudibranchs (µg C nudibranch^{-1}) and the carbon concentration of polyps (µg C polyp^{-1}) (formula X-XI). This provided the unit of carbon specific ingestion rate: µg C polyp µg^{-1} C nudibranch min^{-1}.

$$\text{Carbon specific ingestion rate} = \mu g\ C\ prey^{-1} \times \mu g\ C\ nudibranch^{-1} \times prey\ min^{-1} \quad (X)$$

$$\text{Carbon specific ingestion rate} = \mu g\ C\ \mu g^{-1}\ C\ min^{-1} \quad (XI)$$

Mean estimates were given with ± sample standard deviation (σ) (formula XII).

$$\sigma = \sqrt{\frac{\sum_i^n (X_i - \bar{X})^2}{n-1}} \quad (XII)$$

2.4.3 Statistical analyses

For the statistical analysis of the data that has been collected, RStudio version 4.0.4 (2021-02-15) (RStudio Team, 2020) was used. All plots were made using the packages "ggplot2" and "ggpubr". Model selection was based on biological significance and the Akaike Information Criterion (AIC).

The data and its residuals were assumed to be independent of each other, normally distributed and with equal variance around the mean. Hence, a linear model (lm) with a simple linear regression was used. An analysis of variance (ANOVA) was run to check for significance. The hypotheses were rejected at a level of significance of $p \leq 0.05$.

3 Results

3.1 Settling plate experiment

When settling plate rigs were collected on October 11, 2021, ropes were densely overgrown by green and red algae at both locations. Some bricks also had anemones (Actiniaria) on them. One brick at TBS had been fretted off and lost, so only 29 bricks were brought back into the lab. Despite initial expectations, no scyphozoan polyps were found on either of the plates. As a result of this, the planned selective feeding of nudibranchs on natural fouling communities could not be conducted as there had been no polyp settlement.

During the time the settling plates were immersed at 1 meter depth at Frøya, they were colonized by *Spirobranchus triqueter* (Linnaeus, 1758) (Polychaeta), *Spirorbis* sp. Daudin, 1800 (Polychaeta), *Mytilus edulis* Linnaeus, 1758 (Bivalvia), and *Pododesmus* sp. Philippi, 1837 (Bivalvia). Some plates were almost clean, while others were more densely covered with fouling organisms (Figure 12).

The plates at TBS, however, were colonized by hydroids, bryozoans (*M. membranacea* and *E. pilosa*), *Caprella linearis* (Linnaeus, 1767) (Amphipoda) and the dorid nudibranch *O. muricata.* Additionally, eggs from nudibranchs were found on the plates. Several species of nudibranchs were found on the settling plate rigs themselves, and the fouling community on the bricks. As mentioned, many *O. muricata* were present on the plates close to the Bryozoa. Approximately 15 individuals of *F. auriculata* were found in between the red algae inhabiting hydroids, both on the ropes and in the cracks and holes of the bricks. This was the main source of this particular nudibranch species used during the ingestion experiments on nudibranch predation on Scyphozoa polyps. Several individuals of the dorid nudibranch *Polycera quadrilineata* were found where there was brown algae and bryozoans. The cladobranch nudibranchs *Aeolidia filomena*, *Aeolidia papillosa*, and *Dendronotus frondosus*, as well as *Aplysia punctata* (Cuvier, 1803) (Aplysiida, Mollusca) were also found in relation to the bricks.

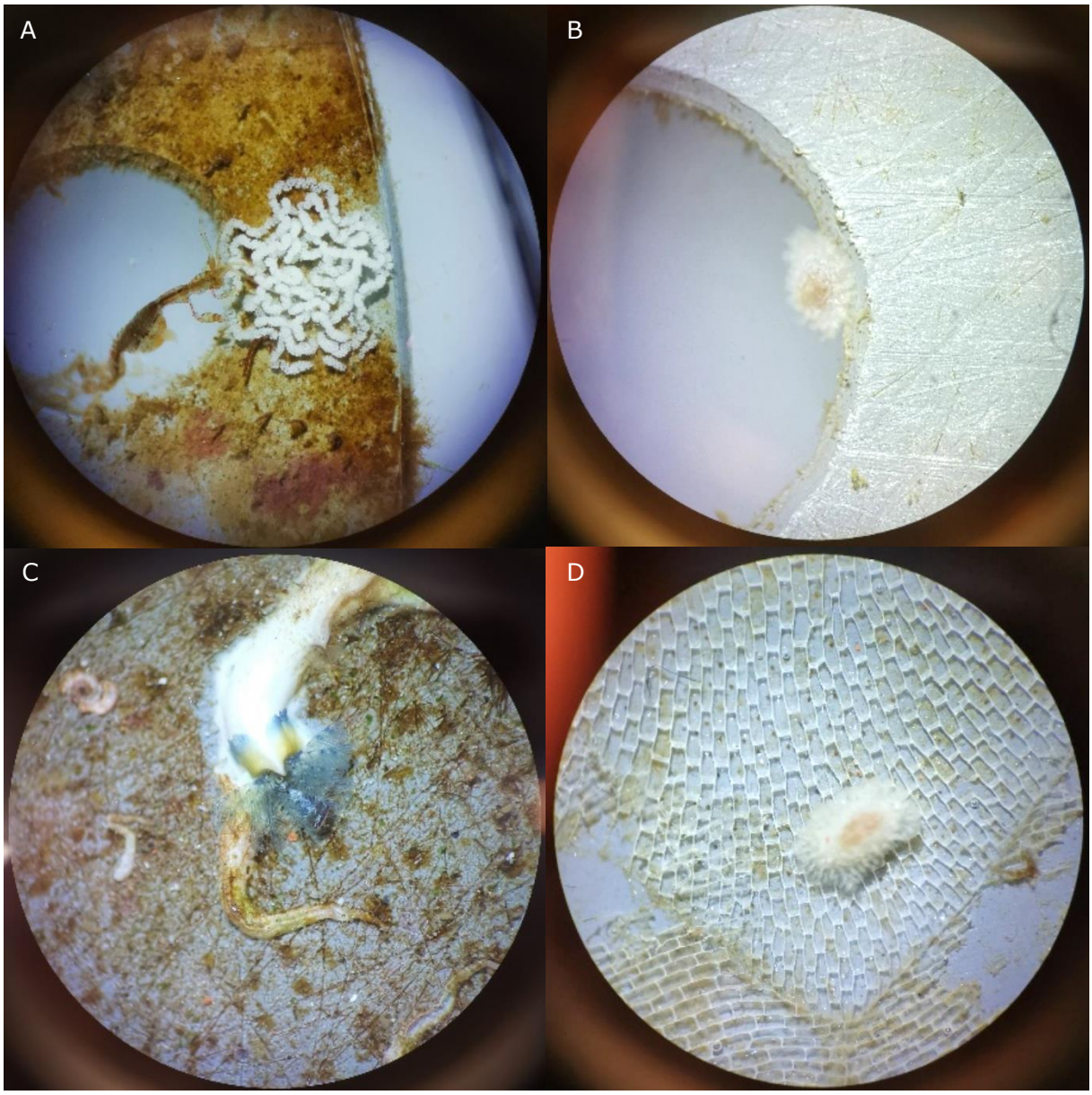

Figure 12: Fouling organisms on some of the plates that had been deployed at TBS and at Frøya. A. *Caprella linearis*, nudibranch eggs and other fouling organisms. B. *Onchidoris muricata* on an uninhabited plate. C. *Spirorbis* sp., *Spirobranchus triqueter* and other fouling organisms. D. *O. muricata* on *M. membranacea*. Photo: Live Agnethe Sørlundsengen Haugen.

3.2 Temperature-dependent ingestion rates of nudibranchs preying on scyphozoa polyps and bryozoan zooids

3.2.1 Experiments with cladobranch nudibranchs preying on Scyphozoa polyps

Nudibranchs in experiment 1-3 were preying on the polyps of *A. aurita*. The ingestion rates at different temperatures are shown in Figure 13. Ingestion rates depending on nudibranch size showed no particular pattern (Figure 14).

For experiment 1, the mean number of polyps eaten per minute was of 2.78 ± 0.91 (11°C), 2.39 ± 0.76 (13°C) and 2.11 ± 0.78 (15°C). This was for *F. bostoniensis*, run for 60 minutes (FB1). There was no significant difference in ingestion rates between the temperature treatments ($p>0.05$). There was no clear relationship between the nudibranch size (mm) and their ingestion rates (Figure 14). The polyps used were 9 days old (post-settlement) at the time of the experimental trial and had an estimated carbon concentration of 7.76 µg C $polyp^{-1}$ (Figure 19).

For experiment 2, the mean number of polyps eaten per minute was 4.49 ± 2.40 (11°C) and 4.9 ± 5.59 (15°C). This was for *F. bostoniensis* run for 15 minutes (FB2). There was no significant difference in ingestion rates between the temperature treatments ($p>0.05$). There was no clear relationship between the nudibranch size (mm) and their ingestion rates (Figure 14). The polyps used were 16 days old at the time of experimental trial and had an estimated carbon concentration of 16.36 µg C $polyp^{-1}$ (Figure 19).

For experiment 3, using *F. auriculata* for 15 minutes (FA), the mean number of polyps eaten per minute was 2.91 ± 2.0 in the 11°C treatment, and 2.74 ± 1.69 in the 15°C treatment. There was no significant difference in ingestion rates between the temperature treatments ($p>0.05$). There was no clear relationship between the nudibranch size (mm) and their ingestion rates (Figure 14). These polyps were 37 days old at the time of experimental trial and had an estimated carbon concentration of 42.16 µg C $polyp^{-1}$ (Figure 19).

When conducting a statistical analysis between experiment 1, 2 and 3, there was no significant effect on ingestion rates (polyps min^{-1}) of the species of nudibranchs used (Figure 13). Additionally, there was no significant difference between the age of the polyps that were used (ANOVA).

Carbon specific ingestion rate (µg C of polyps $µg^{-1}$ C nudibranch min^{-1}) was calculated (Figure 16). The carbon specific ingestion rates for each temperature treatment are shown in Figure 15.

For experiment 1, the mean carbon specific ingestion rate for *F. bostoniensis* (60 min. experimental trial) was 44 675.35 ± 14 783.02 µg C of polyps $µg^{-1}$ C nudibranch min^{-1} in the 11°C treatment. The mean was 38 271.01 ± 17 460.60 µg C of polyps $µg^{-1}$ C nudibranch min^{-1} in the 13°C treatment. Carbon specific ingestion rate was 31 564.77 ± 9563.18 µg C of polyps $µg^{-1}$ C nudibranch min^{-1} in the 15°C treatment. There was no significant difference in carbon specific ingestion rate between the temperature treatments ($p>0.05$).

For experiment 2, using *F. bostoniensis* (15 min. experimental trial), the mean carbon specific ingestion rate was 108 356.61 ± 54 020.55 µg C of polyps $µg^{-1}$ C nudibranch min^{-1} in the 11°C treatment, and 161 115.70 ± 179 004.79 µg C of polyps $µg^{-1}$ C nudibranch

min^{-1} in the 15°C treatment. There was no significant difference in carbon specific ingestion rate between the temperature treatments in this experiment ($p>0.05$).

For experiment 3, the mean carbon specific ingestion rate for *F. auriculata* was 296 867.62 ± 188 397.53 µg C of polyps $µg^{-1}$ C nudibranch min^{-1} in the 11°C treatment and 241 110.34 ± 114 094.87 µg C of polyps $µg^{-1}$ C nudibranch min^{-1} in the 15°C treatment. No significant effect of temperature treatment on carbon specific ingestion rate was found ($p>0.05$).

When comparing the carbon specific ingestion rates of the three experiments (1-3) to each other, there was a significant difference between the species used on the carbon specific ingestion rate ($p<0.01$) (Table 2). A significant effect of polyp age (days) on the carbon specific ingestion rate ($p<0.01$) was detected. See Figure 22 and Figure 23 in Appendix 5: Statistical analysis output for R script.

Table 2: Analysis of variance table (ANOVA) for the statistical tests on carbon-specific ingestion rates of nudibranchs in relation to species, polyp age, temperature treatment and nudibranch size.

Source of variation	DF	Sum of squares	Mean square	F	P
Species	2	3.1975^{11}	1.5988^{11}	16.056	0.00002263*
Polyp age	1	3.1257^{11}	3.1257^{11}	31.697	0.000004415*
Temperature	2	5.2945^{10}	2.6473^{10}	1.3585	0.2735
Size	1	4.652^{10}	4.652^{10}	2.442	0.1288

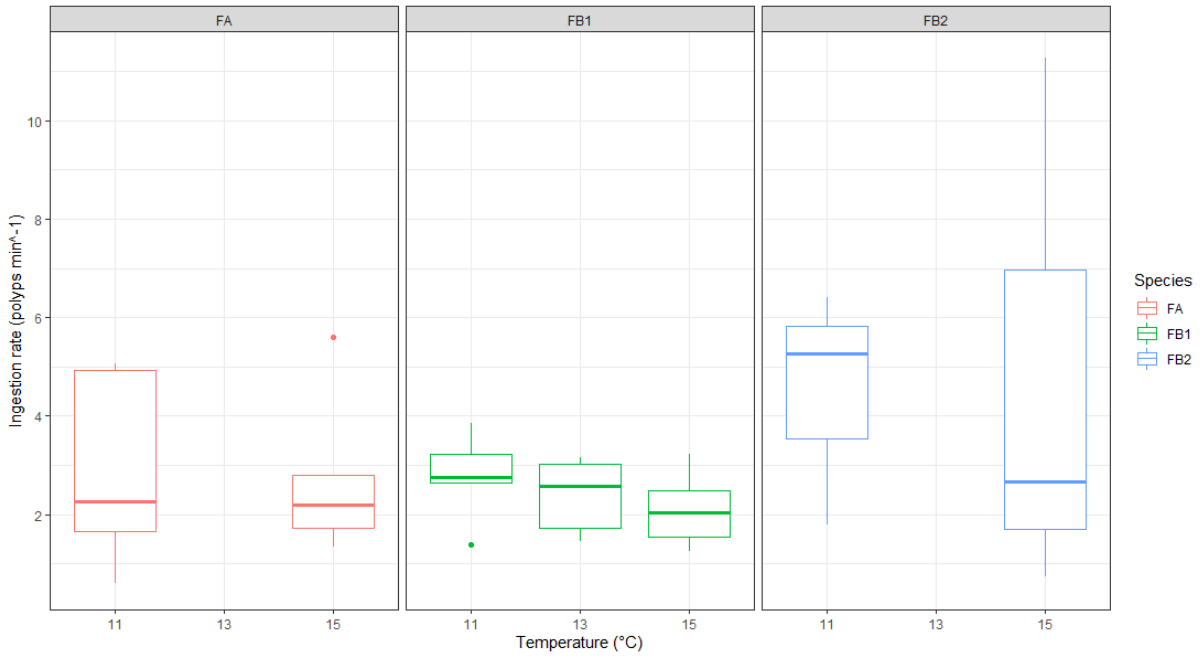

Figure 13: Ingestion rates of nudibranchs preying on *Aurelia aurita* polyps (polyps min^{-1}) as a function of temperature (°C). The three nudibranch species used were FA *Facelina auriculata*, FB1 *Facelina bostoniensis*, and FB2 *Facelina bostoniensis* at different temperature conditions (11°C, 13°C and 15°C).

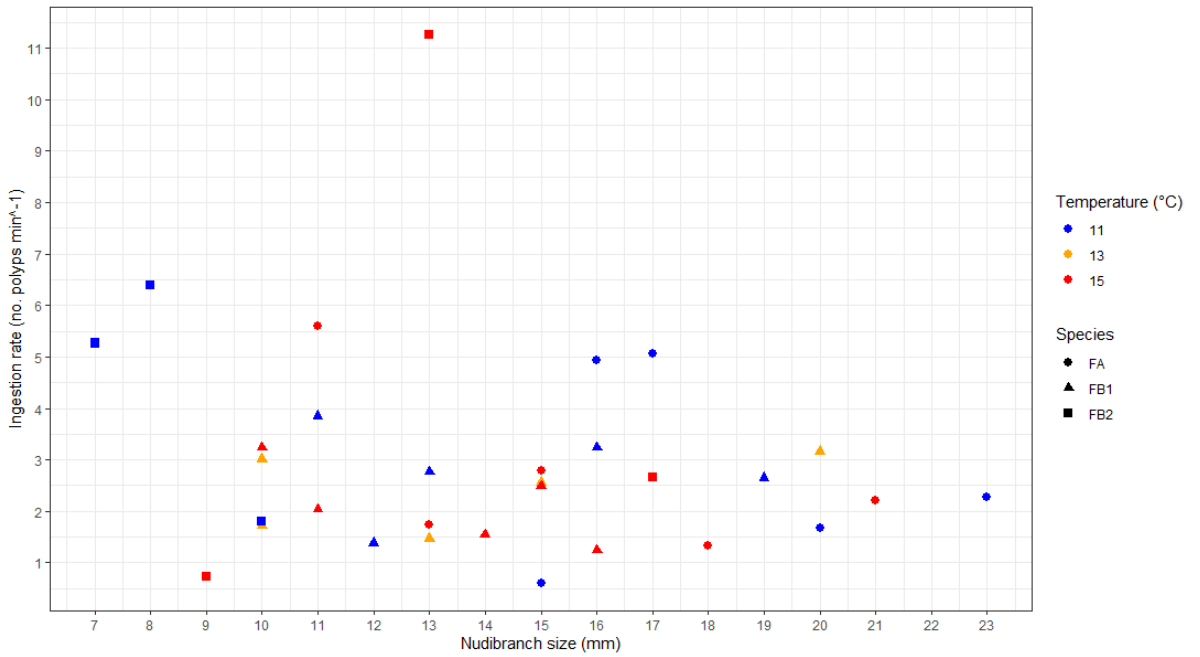

Figure 14: Ingestion rates on *Aurelia aurita* polyps (polyps min^{-1}) as a function of nudibranch size (mm) for three different species. Species are indicated by different shapes: FA *Facelina auriculata*, FB1 *Facelina bostoniensis*, FB2 *Facelina bostoniensis*. Treatments are indicated by different colors: temperatures 11°C, 13°C, and 15°C.

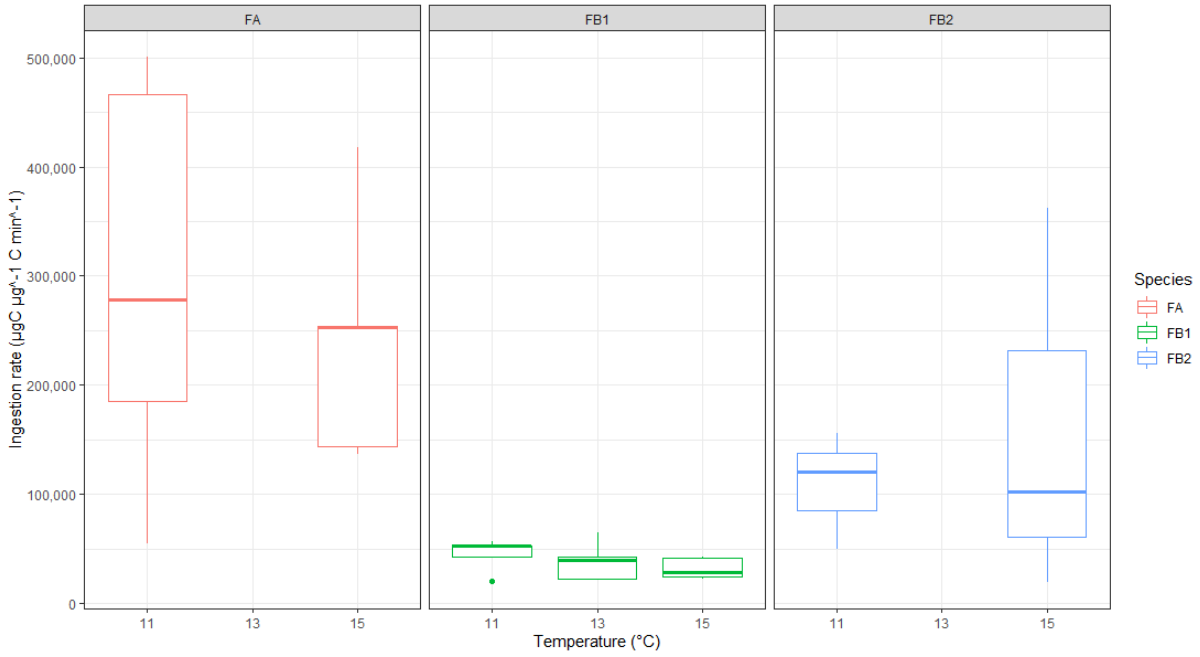

Figure 15: Carbon specific ingestion rates of nudibranchs preying on *Aurelia aurita* polyps (µg C polyp µg^{-1} C nudibranch min^{-1}) as a function of temperature (°C). The three nudibranch species used were FA *Facelina auriculata*, FB1 *Facelina bostoniensis*, and FB2 *Facelina bostoniensis* at different temperature conditions (11°C, 13°C and 15°C).

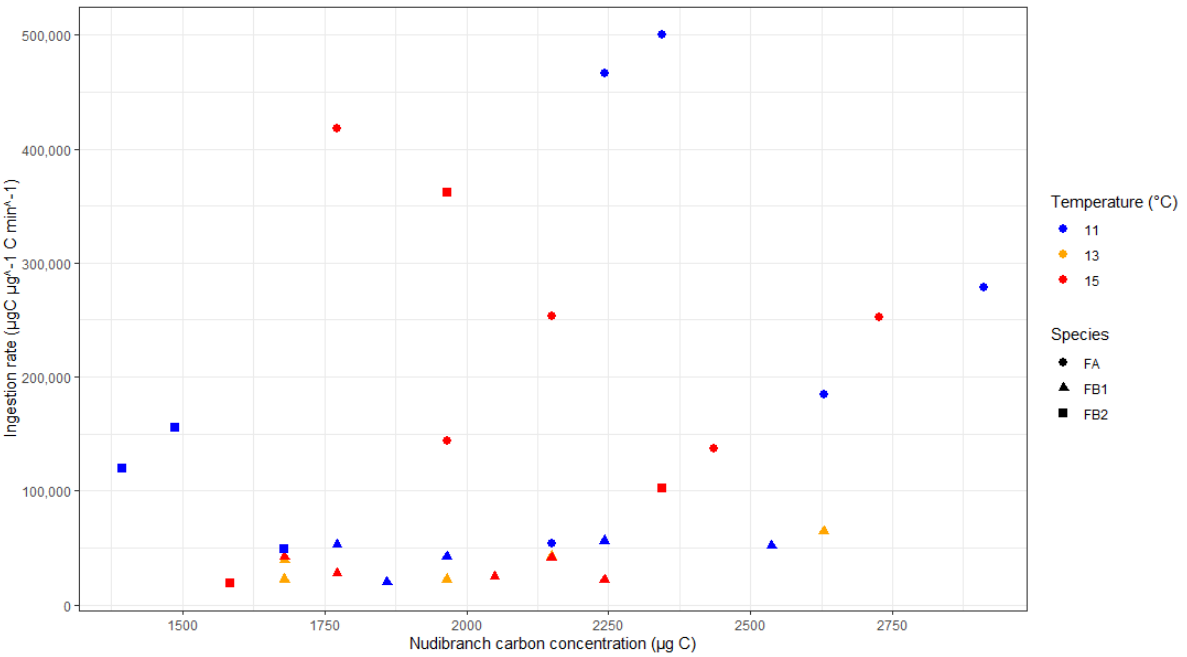

Figure 16: Carbon specific ingestion rates of nudibranchs preying on *Aurelia aurita* polyps (µg C polyp µg $^{-1}$ C nudibranch min^{-1}) as a function of nudibranch carbon concentration (µg C). Species are indicated by different shapes: FA *Facelina auriculata*, FB1 *Facelina bostoniensis*, FB2 *Facelina bostoniensis*. Treatments are indicated by different colors: temperatures 11°C, 13°C, and 15°C.

3.2.2 Experiments with dorid nudibranchs preying on Bryozoa zooids

The nudibranch *Onchidoris muricata* preyed on bryozoan zooids from both *M. membranacea* and *E. pilosa* in experiment 4 and 5.

In experiment 4, the mean number of *M. membranacea* zooids eaten per minute was 0.25 ± 0.13 (11°C) and 0.31 ± 0.19 (15°C) (Figure 17). There was no significant difference in ingestion rates (zooids min^{-1}) between the temperature treatments ($p>0.05$). Additionally, there was no clear relationship between the ingestion rate and the nudibranch size (Figure 18).

In experiment 5, the mean number of *E. pilosa* zooids eaten per minute was 0.34 ± 0.26 (11°C) and 0.37 ± 0.24 (15°C) (Figure 17). There was no significant difference in ingestion rates (zooids min^{-1}) between the temperature treatments ($p>0.05$). There was no clear relationship between the nudibranch size (mm) and the ingestion rates of bryozoan zooids (Figure 18).

When comparing the two experiments 4 and 5 statistically, there was no significant difference in ingestion rates (zooids min^{-1}) between the two species of Bryozoa, *E. pilosa* and *M. membranacea* ($p>0.05$, ANOVA). Carbon specific ingestion rates were not calculated due to lack of carbon values for the zooids.

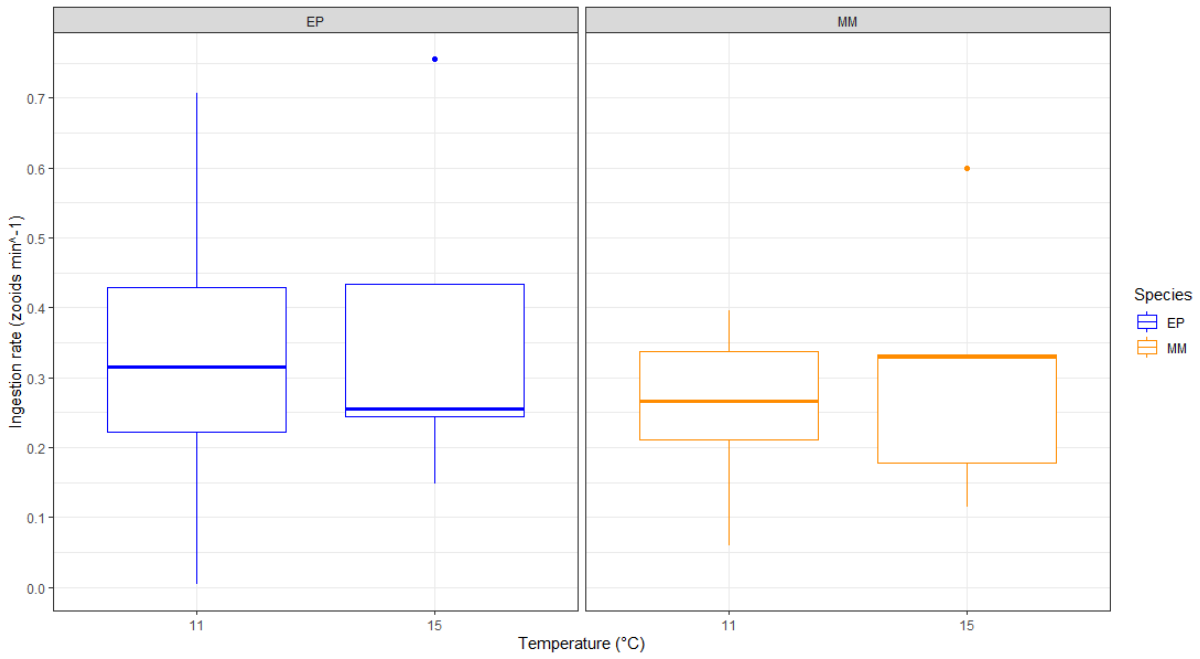

Figure 17: Ingestion rates on *Electra pilosa* (left) and *Membranipora membranacea* (right) (zooids min^{-1}) as a function of temperature (°C) for the nudibranch *Onchidoris muricata*. The two temperature treatments were 11°C and 15°C.

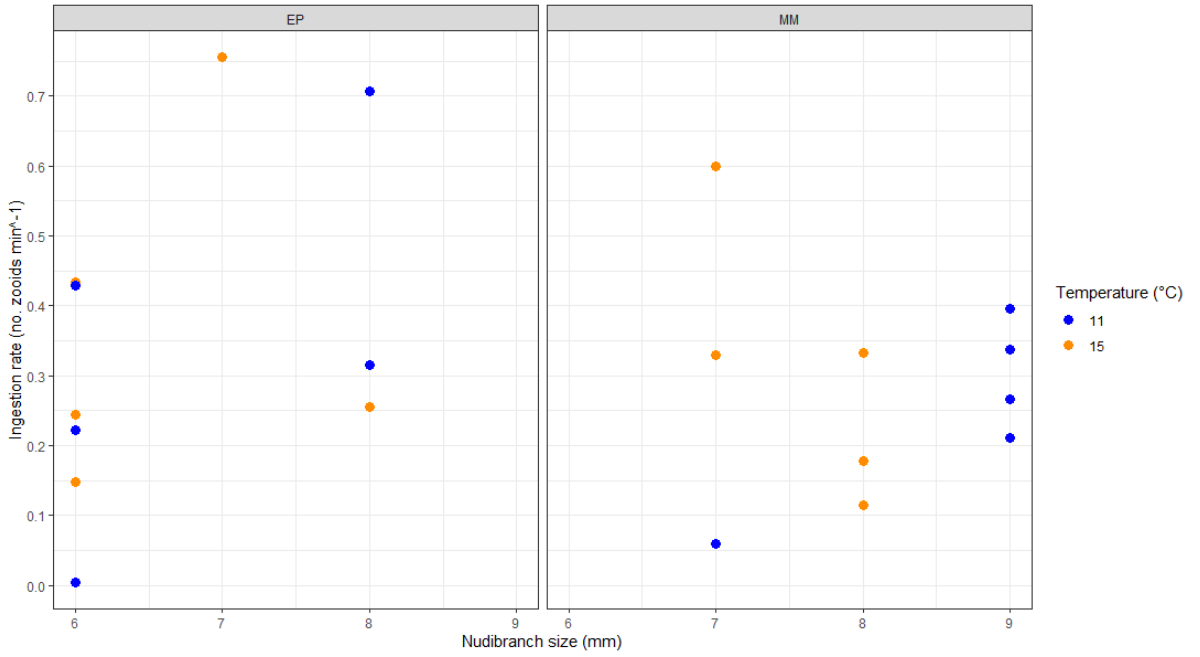

Figure 18: Ingestion rates of the nudibranch *O. muricata* preying on bryozoan zooids of *Electra pilosa* (left) and *Membranipora membranacea* (right) (zooids ingested min^{-1}) as a function of nudibranch size (mm). Temperature treatments were 11°C and 15°C.

3.2.3 Carbon analysis

Polyp carbon concentration was gathered from Cawood (2012), Myrvold (2020), Kamiyama (2011), Ikeda et al. (2017), and Chi et a. (unpublished data). The formula of the trend line was y=1.2288x – 3.3045 and R^2 was 0.16 (Figure 19). The carbon estimates of polyps were extrapolated from the regression line providing carbon values of 7.55 µg C polyp^{-1} (9

days old polyps), 16.36 µg C polyp^{-1} (16 days old polyps) and 42.16 µg C polyp^{-1} (37 days old polyps).

Nudibranchs of the species *F. bostoniensis* and *O. muricata* were analyzed for carbon concentration (Appendix 4: Carbon weight analysis, Table 5). Carbon concentration of the cladobranch nudibranchs (*F. bostoniensis* and *F. auriculata*) plotted against extrapolated nudibranch sizes illustrated what the relationship looked like after the extrapolation (Figure 20). Carbon concentrations of *O. muricata* plotted against nudibranch size (mm) showed a positive trend (Figure 21).

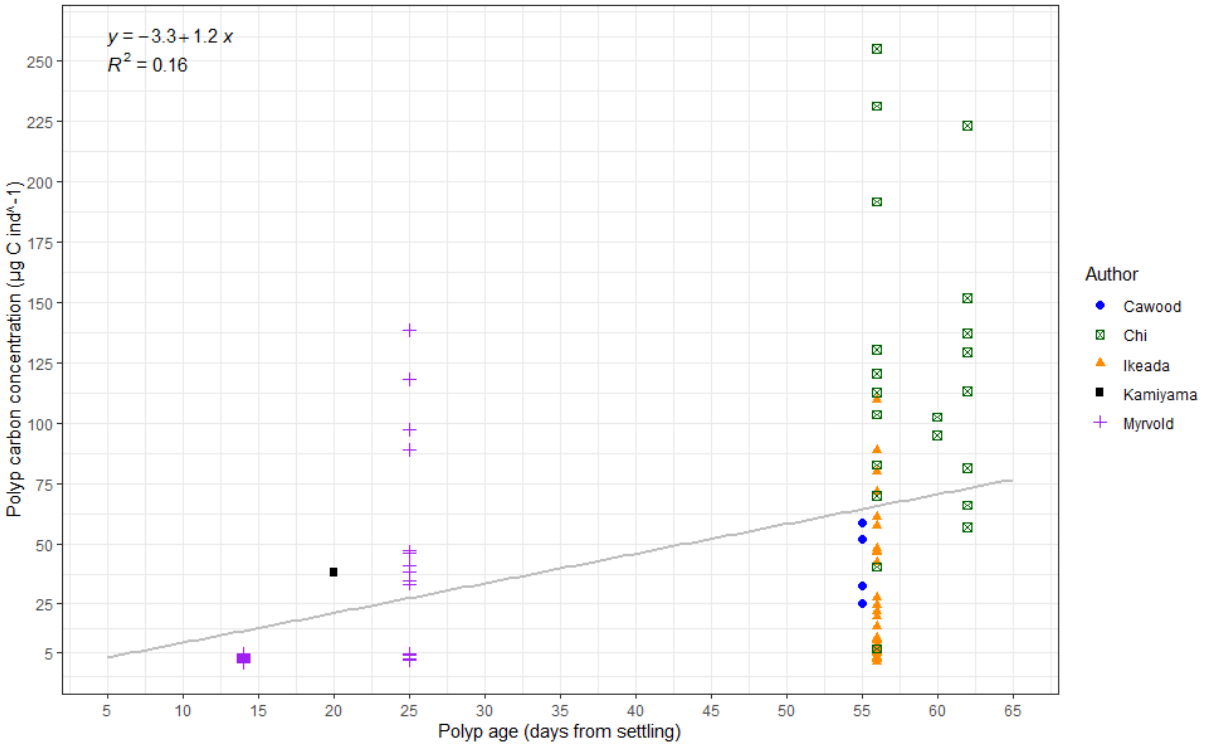

Figure 19: Polyp carbon concentration (µg C ind^{-1}) as a function of polyp age (days from settling) for *Aurelia aurita* scyphopolyps. The C-values were extracted from data obtained from Cawood (2012), Myrvold (2020), Kamiyama (2011), Ikeda et al. (2017), and Chi et al. (unpublished data).

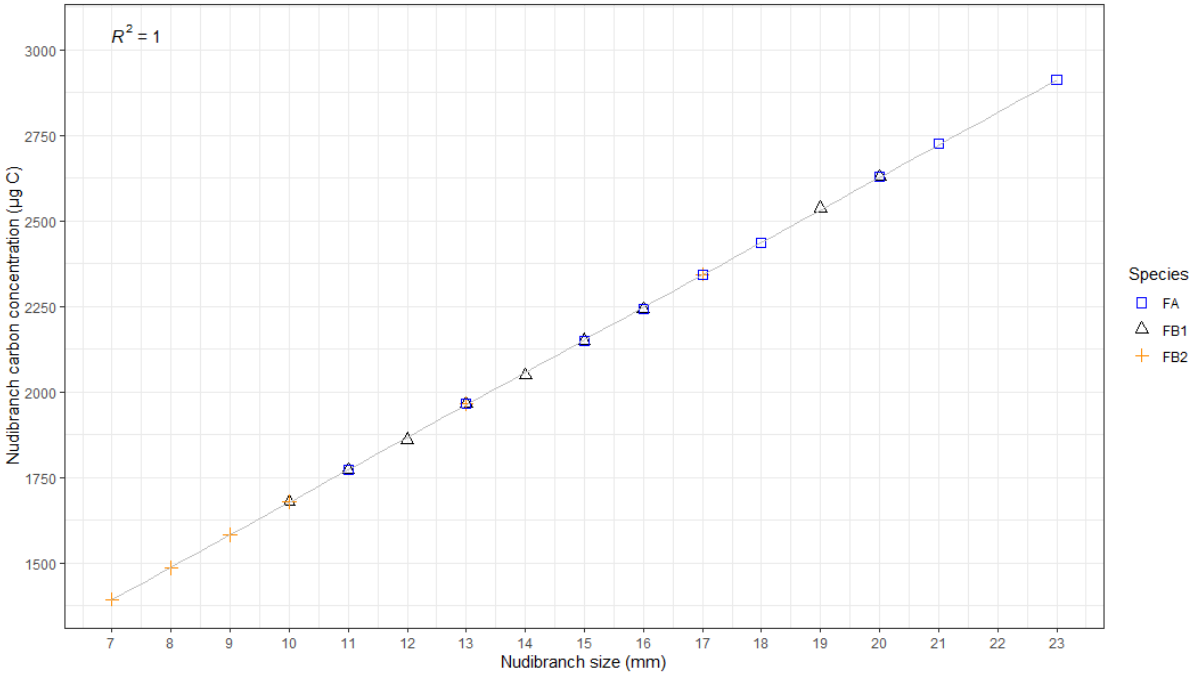

Figure 20: Nudibranch carbon concentration (µg C) as a function of nudibranch size (mm). Cladobranch species of nudibranchs indicated by different shapes; FA *Facelina auriculata*, FB1 *Facelina bostoniensis*, FB2 *Facelina bostoniensis*.

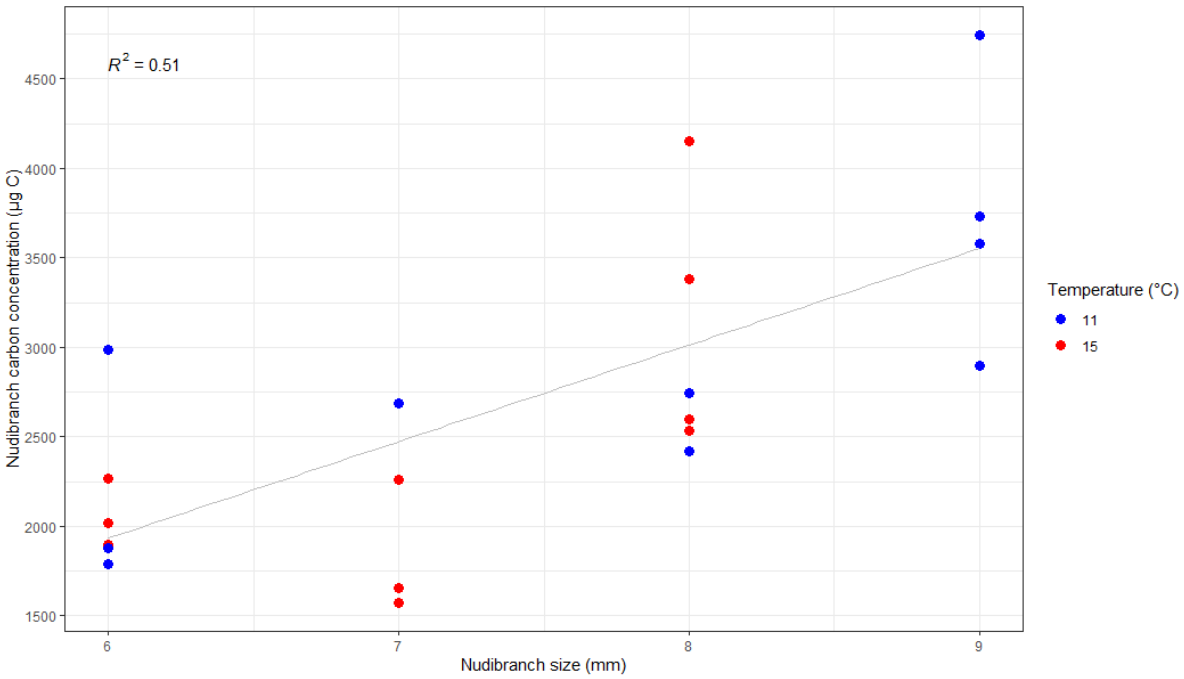

Figure 21: Carbon concentration (µg C) of *O. muricata* as a function of nudibranch size (mm). The temperature treatments are indicated by color, blue being 11°C and red being 15°C.

4 Discussion

4.1 Settling plate experiment

Even though the settling plates were deployed during the peak season of scyphopolyp settlement that usually happens during the period from August to September in Trondheimsfjorden, no scyphopolyps were found on the settling plates, neither at Frøya nor at TBS.

One brick and hence settling plate was lost in the experiment, although measures had been taken to avoid it. However, it might have been caused by wave exposure, natural corrosion by seawater or by a collision with nearby boat activity.

Although polyps were not found on the plates, a diverse fouling community had settled to the plates during the period of exposure (Actiniaria, Amphipoda, Aplysiida, Bivalvia Bryozoa, Chlorophyceae, Cladobranchia, Doridina, Hydrozoa, Phaeophyceae, Polychaeta, Rhodophyceae). There were nudibranch eggs on several of the plates that were deployed close to TBS pier. In addition, several adult nudibranchs were found on the bricks themselves. Seeing that there were no polyps present on any of the plates while nudibranchs were present, one possible explanation could therefore be that the plates were in the sea for too long. This could have allowed nudibranchs to have already eaten all the polyps that had freshly settled on the plates after the release of planula larvae in August or early September.

Another explanation could be that the moving of the settling plate rigs as well as the nearby boat traffic had disturbed the plates too much during the settling period. However, this is unlikely as the other fouling organisms on the plates seemed to not be affected by this and scyphopolyps are considered to colonize a variety of natural and anthropogenic hard substrates efficiently (Rekstad et al., 2021).

Aurelia aurita polyps are deemed to be very robust and tolerant to changing environmental conditions e.g. changes in salinity (Holst and Jarms, 2010). As we struggled to collect adult medusae of this species over the summer, the absence of *A. aurita* polyps on the plates could also be related to the low abundance of *A. aurita* medusae. Researchers from the Norwegian Institute of Marine Research reported large bloom activity in 2021, however, mainly in the south-western parts of Norway (Falkenhaug, 2021) and not during the warmest summer months (Lorentzen, 2022). This supports the fact that only minor *A. aurita* blooms occurred along the northern part of the Norwegian coast in summer 2021. When comparing recorded sightings of scyphozoan medusae between 2020 and 2021, there seemed to be a clear difference in scyphozoan blooms (data from Dugnad for Havet, a citizen observatory from the Norwegian Institute for Marine Research). The first record of any medusa in 2020 was made on May 31 and was an *A. aurita* bloom. Thereafter, *A. aurita* blooms were continuously reported until the end of June, whereas blooms of *Cyanea lamarckii* Péron & Lesueur, 1810 seemed to take over from July. On the contrary, the first recorded scyphozoan in 2021 was *C. lamarckii* on July 7, and not a single *A. aurita* was reported over the entire summer period of 2021. It is believed that citizen sightings are not recorded unless it is a considerably large occurrence or bloom, hence it is reasonable to think that this indirectly reflects the lack of larger blooms in the Trøndelag region in 2021.

4.2 Temperature-dependent ingestion rates of nudibranchs preying on scyphozoa polyps and bryozoan zooids

4.2.1 Experiments with cladobranch nudibranchs preying on Scyphozoa polyps

Selective feeding and behavior of nudibranchs

A well-known fact about scyphozoans is that they sting. As they can sting humans when in contact, they can also sting other marine animals. When experiments were run, the overall behavior of the nudibranchs were occasionally filmed and assessed. The planula larvae had settled all over the petri dish lids and no clear settling patterns could be observed. Because of this, some areas of the petri dish lid were more densely populated than others, leading to a more frequent encounter rate for the nudibranch while feeding. Some nudibranchs moved rather quickly, clearly searching for food by moving their entire head or just moving their tentacles. If either tentacle came across a polyp, the nudibranch seemed to flinch as if it was stung. An immediate retraction of the tentacle towards the body plan usually followed after the sting, before the nudibranch would direct the mouthparts towards the polyp and ingest it. When they encountered a polyp, the mouthparts started to move in a churning manner. If they were already busy eating another polyp when they were obviously stung, they kept their tentacles perpendicular to the body, instead of horizontally, to avoid being stung again. As the substrate on which the experiments were run was transparent, the feeding behavior of the nudibranchs could nicely be assessed. No pronounced difference in the feeding behavior between *F. auriculata* and *F. bostoniensis* could be observed (own personal observations).

The same feeding behavior as the one observed in the current study could be observed by others for the nudibranch *Spurilla neapolitana* (Delle Chiaje, 1841) feeding on anemones. The authors described it as a behavior comprising four steps; approaching, first contact, retracting, and re-approaching (Conklin and Mariscal, 1977). Several explanations on how nudibranchs handle the ingestion of cnidarian prey items successfully have been published previously, among them studies stressing the relevance of acclimation over time (Conklin and Mariscal, 1977), mucus secretion as protection (Glaser, 1910, Harris, 1973, Harris, 1971, Russell, 1942, Waters, 1973), and cells in the epidermis preventing cnidocytes from entering (Edmunds, 1966, Graham, 1938).

In the current study, a third species was originally considered to be tested, in addition to *F. auriculata* and *F. bostoniensis*, since the nudibranch *Catriona aurantia* was found amongst the barnacles and *Ectopleura larynx* at the pier of Planktonic AS, Trondheim, as well. However, *C. aurantia* seemed not to be as interested in polyps as the other two species and had a more evident response to the stinging from polyps. As the experimental design was not optimal for this species, these experiments were terminated.

There is an ongoing discussion of whether nudibranchs feed on different prey when in captivity compared to natural settings (Martinsson et al., 2021). Swennen (1961) found that *F. auriculata* fed on another opistobranch, *Elysia viridis* (Montagu, 1804), as well as feeding on several different hydroids when kept in captivity. However, when the specimens were provided with enough food, they showed no tendency to cannibalism (Swennen, 1961). According to the abovementioned study, the cladobranch nudibranch turned green after ingesting the Saccoglossa. Colors of feed items therefore seems to directly be portrayed in their cerata and can be looked at as an indicator of which feed items they

have consumed. Other experiments have also come to the conclusion that *F. auriculata* is not a picky eater (Swennen, 1961).

In retrospect, keeping *F. auriculata* and other nudibranchs in the same aquarium might have caused unforeseen deaths of specimens and competitive interactions, even though feed items were added regularly. Specimens were assessed regularly by looking at the color of their dorsal cerata since the color provides an indicator for food shortage.

When considering ingestion to take place without any constraints from other limiting factors, ingestion rate should increase proportionally with food concentration. Limiting factors could be mechanical and physiological constraints when processing food, capturing, and ingesting prey. Also, there is belief that predators lose interest in prey when food abundance is high (Båmstedt et al., 2000) or too low. The former was described by Keen (1991) and Hoover et al. (2012) for the nudibranch *Hermissenda crassicornis* (Eschscholtz, 1831) feeding on *A. aurita*- and *Aurelia labiata* Chamisso & Eysenhardt, 1821 polyps respectively. For MacLeod and Valiela (1975), the number of hydroid polyp prey where the ingestion rates stagnated was at 50 polyps, meaning that beyond this number of prey, the nudibranch *C. verrucosa* would not continue feeding at the same pace. In the case of scyphozoan polyps, this might not be a direct lack of interest but instead a defense mechanism where nudibranchs try to avoid getting stung when prey density is high. According to MacLeod and Valiela (1975), some species of cladobranch nudibranchs are opportunistic in the way that they ingest as much hydroid polyps as they can get, and then choose to move on to the next colony as soon as the prey density gets low. This being when other polyp predators are present (MacLeod and Valiela, 1975), which is the likely scenario *in situ*. This example follows the aspect of optimal foraging theory, where the benefits of foraging needs to exceed the costs of it (Campbell et al., 2015).

Reported ingestion rates for nudibranchs on scyphopolyps are e.g. 43 polyps day^{-1} for small *H. crassicornis* (>0.5 g WW) and 535 polyps day^{-1} for larger *H. crassicornis* (2 g WW). *Sakuraeolis enosimensis* (Baba, 1930) of sizes 0.07-0.48 g had ingestion rates from 8-45 polyps day^{-1} according to Takao et al. (2014). They found a significant effect of nudibranch body weight (wet weight) and the ingestion rate when preying on *A. aurita* polyps. The same was true for Hoover et al. (2012), and the nudibranch *H. crassicornis*' consumption of *A. labiata* polyps increased with increasing nudibranch size. Hernroth and Gröndahl (1985a) reported ingestion rates for *Coryphella verrucosa* (1-15 mm long) up to 200 polyps day^{-1}. In the current study, the nudibranch sizes ranged from 7 to 23 mm, with ingestion rates ranging between 0.6-11.3 *A. aurita* polyps min^{-1}. No difference in ingestion rates for different sizes of nudibranch was found, and no difference in ingestion rate between the two cladobranch nudibranch species was found. However, carbon specific ingestion rates showed a significant difference between the two species. Therefore, hypothesis 2, of the current study assuming that *different nudibranch species show differences in ingestion rates when feeding on scyphozoan polyps* could be confirmed.

Similar to what other authors did, Folino (1993) found that there was a positive correlation between nudibranch size and the amount of the hydroid *Hydractinia echinata* (Fleming, 1828) that were consumed by *Cuthona nana* (Alder & Hancock, 1842). However, small *C. nana* only preyed on parts of the hydroid polyps or the stolons of the hydroid, while the large ones fed on whole hydroid polyps (Folino, 1993). Because of the differences in radula of nudibranchs and whether mandibles are present or not, the parts of the hydroid tissue they are able to feed on differs. This creates niches in feed preferences because larger species are often able to eat on different areas of the prey than smaller species. *Facelina*

sp. usually feed on hydroid polyps as it has the body size, mandibles and a radula that allows feeding on not only polyps of one specific species but up to ten different hydroid species. *Catriona aurantia*, however, might be limited to sucking or biting on the stem of hydroids, potentially explaining why it would not feed on *A. aurita* polyps in the current study's experiments (Jussi Evertsen, personal communication May 10, 2022).

Prey size is a crucial factor when it comes to selective feeding of predators. Larger individuals may be seen easier from a distance and encounter rates can be increased (Båmstedt et al., 2000). Optimal foraging theory says that a predator should choose the prey that gives optimal energy gain and energy expenditure (Begon et al., 1990). This indicates that it is profitable to choose larger prey that are close by to optimize the gain/expenditure ratio. Furthermore, size-dependent grazing and the difference between species was accounted for. This was ensured by using polyps of three different ages/sizes as prey and two different species of nudibranchs as predators. The age ranges from the youngest to the oldest polyps that served as prey items were 28 days while age can be directly correlated with size. In the current study, there was a higher mean carbon specific ingestion rate when polyp prey was larger than when they were just a couple of days old. It is hence likely that the nudibranchs in the latter experiments were able to optimize their gain/expenditure ratio better. Hoover et al. (2012), however, did not find any significant effect of the polyp size on consumption by nudibranchs.

Although the sizes of the polyps were not measured in this study directly, age-size conversion and carbon estimates were extracted from several papers (Cawood (2012), Myrvold (2020), Kamiyama (2011), Ikeda et al. (2017), and Chi et al. (unpublished data)). Scyphopolyps show either exponential growth (Willcox et al., 2007) or sigmoid growth (Coyne, 1973). Willcox et al. (2007) experienced exponential growth in the last days of their experiments (after day 21), which could mean it had not reached its growth plateau yet. Polyp growth is in general considered to be limited by space (Willcox et al., 2007) and food availability (Coyne, 1973). Polyps used in these experiments were not space limited yet, hence they could have shown an exponential or sigmoidal growth curve if the growth phase was continued. There was no indication of exponential growth in the literature values that were gathered on polyp carbon concentration (Figure 19). Before deciding on which growth pattern described the data best, an exponential and a linear trend line was fitted to the data. The R^2-values for both trend lines were below 0.2, meaning neither of them were particularly more fit than the other. To further test the potential difference in using a linear or exponential approach, the polyp carbon values for the polyp ages were calculated based on the linear and the exponential trend line. As assumed, the exponential trend line provided polyp carbon estimates that were on a different range thus potentially affecting the statistical tests applied. As R^2 of 1 indicates a perfect linear relationship (Ratner, 2009), the values in this study for both the linear and the exponential approach for age/size and carbon conversions were far from that. In this case, the decision to use a linear regression to extract and extrapolate polyp carbon concentration from the graph was taken. Due to the pros and cons of using either approach, the extrapolated data should be treated with caution. During the study, C:N analyses of the polyps were not measured directly, although the amount of polyp carbon estimates in published literature was also limited.

The statistical models with polyp age as a predictor variable and carbon specific ingestion rate as response variable showed that polyp age (possibly correlated to polyp size as well) does have an effect on nudibranch feeding. Hypothesis 3 stating that *nudibranch ingestion rates differ depending on the age and size of scyphozoan polyps* is therefore accepted.

Temperature selectivity of nudibranchs and the impact on ingestion rates

The aim of this study was to increase our knowledge on nudibranch's selective feeding behavior and effectiveness of different species especially in the light of changes in temperature conditions.

Temperature tolerance ranges for nudibranchs are summarized for some species in certain geographical regions by Clark (1975). The optimum temperature seems to differ significantly between species, and might be more connected to the region itself than to the species. Armstrong et al. (2019) found that maximum heat tolerance differed between species, but that it was the same for any species within the same location. They also emphasized the fact that an increase in sea surface temperatures would be fatal for most nudibranchs (Armstrong et al., 2019). Seeing the previously described temperature variations in Trondheimsfjorden (Jacobson, 1983), nudibranchs found in that area are probably well adapted to such fluctuations. Nevertheless, a global increase in SST could still have fatal consequences for them.

The nudibranch *A. papillosa* is apparently restricted to a specific temperature regime in the water from 10-38 meters depth (Miller, 1961). Franz (1970) suggested that 25°C would be somewhere around the upper temperature tolerance of sub-arctic species of nudibranchs for reproduction to occur. This was true for the two species *C. aurantia* and *Tergipes tergipes* (Forsskål in Niebuhr, 1775) in Massachusetts. Others have suggested that 22°C was the optimal temperature for reproduction in the species *Armina maculata* Rafinesque, 1814 (Pires, 2012). When it comes to feeding at different temperatures, Pires (2012) found higher ingestion rates at 18°C than at higher temperature regimes. Temperatures of 24°C have been reported to be outside of the tolerable limits for *H. crassicornis* (Tyndale et al., 1994).

Optimal temperature is likely correlated to both species and location, hence can therefore not be defined within any simple frames.

The Cladobranchian species *A. maculata* was found to significantly reduce its foraging behavior when temperatures increased (Pires, 2012). This was when feeding on the Pennatulacea species *Veretillum cynomorium* (Pallas, 1766) of quite some size. Increased mortality was also found when temperatures increased to outside their optimal range (Pires, 2012). The ingestion rates from the abovementioned study are, however, not directly comparable to the current study because of the size differences between species and prey sizes.

Acclimation is a crucial component in experiments in marine ecology. For the present experiments, all specimens were acclimated to the same temperature condition for 24 hours prior to the experiments. Thereafter, the specimens were put into their respective temperature regime an hour before feeding experiments started. This could bear a bias towards the upper temperature regime and the specimens used there, as those specimens had 25h in approximately the same temperature while the others did not.

Temperature is just as crucial in feeding experiments. Temperature has a large impact on metabolic activities, especially growth rate (metabolic theory of ecology) (Brown et al., 2004). Animals usually need more energy at elevated temperature conditions. The same goes for feeding activities, digestion rate and mobility. This would imply that foraging efforts and food demand would increase, and feeding and digestion would be faster in warmer environments than in colder ones (Båmstedt et al., 2000). Light also plays a role (Båmstedt et al., 2000), as nudibranchs have light-sensoring eyes. For nudibranchs,

however, most prey detection occurs using their rhinophores and tentacles. Despite the strong temperature-dependency of metabolic processes, no significant effects of temperature on ingestion rates or carbon specific ingestion rates could be found in either of the three feeding experiments. Seeing that previous studies have had a larger gap between the temperature treatments, this might have caused the lack of significant difference in the present experiments. Hypothesis 1 stating that *increasing sea surface temperatures influence nudibranch ingestion rates*, therefore needs to be rejected.

Phenology regarding prey and predator occurrence is an important aspect in trophic ecology. Phenology is the study of life- and seasonal cycles in the light of climate (Ji et al., 2010). In her thesis, Hoett (2019) describes the possibility of trophic mismatch situations between nudibranchs in Portugal and their prey. Although temperature did not play a significant part in the present experiments, temperature dependent organisms may be severely affected by changes that occur in specific seasons. This can again cause cascading effects across the food web (Goddard and Pearse, 2011). Differences in phenology and annual prey densities can therefore cause nudibranch populations to differ between years (Hoett, 2019). For example, reproduction in *F. auriculata* is paused during the winter. They are believed to not thrive in cold water, and would also not spawn in aquariums with low temperatures (Swennen, 1961).

During the first experimental trial (FB1 for 60 min), two individuals had not been in contact with the polyps at all. They were replaced with two new individuals. When removed, it was noted that one had been laying eggs instead of eating. This indicates that nudibranchs do not eat constantly (Pratt and Grason, 2007, Båmstedt et al., 2000, Folino, 1993). This was the case for Folino (1993) and others, who over a 24 hour period documented nudibranchs who occasionally stopped feeding and engaged in copulating or spawning activities. In the present study, during the time that the nudibranchs were in the aquarium tank in the lab, they seem to frequently reproduce seeing that they are all hermaphrodites (Raja-Salleh et al., 2019). Hence, there were new clusters of eggs reappearing on a regular basis meaning their ability to spawn was not impaired when kept in temperatures of approximately 9°C.

During the course of experiment 2 (FB2 for 15 min), one specimen of nudibranch had not been attached to the substrate inhabiting prey the entire time. The individual timer was still run for 15 minutes to standardize across beakers. This lack of interest in food foraging was considered abnormal activity, despite the starvation period prior to the experiment. Cruciality of starvation prior to an experiment was described for nudibranchs by Àvila et al. (1998). They found a significant different result for the cladobranch nudibranch *H. crassicornis* eating hydroid polyps when individuals had been starved compared to when they had not been starved. Although all individuals in this study had been starved for 24 hours before the experiments started, there might be individual differences that led to those nudibranchs being uninterested in foraging. Takao et al. (2014) starved their specimens for 3 hours, before they were allowed to feed for 1-3 days, suggesting that a starvation period of 24 h might have been too long and not representative of the natural conditions.

Seeing that both nudibranchs were actively feeding on *A. aurita* polyps during the feeding trials, the probability is high that they would feed on polyps *in situ* as well. In the future, similar feeding trials should be conducted with the polyps of *Cyanea* species in addition as they pose a larger threat to recreation and tourism by limiting swimming and other water activities as well as interfering with aquaculture and fishery activities to a larger extent. The development and intensity of jellyfish blooms could potentially be controlled by natural

predators of scyphopolyps, thereby efficiently reducing the local population of adult medusae thereafter (Hernroth and Gröndahl, 1985b). Previous studies even looked into the possibility of biocontrolling the benthic polyp stages of jellyfish by transplanting natural predators to specific environments to reduce the risk of jellyfish bloom occurrences (Takao et al., 2014). A biocontrol by nudibranchs on scyphozoan polyps is, regardless, plausible.

However, as mentioned above, the naturalness of feeding in captivity after being starved can be questionable. Alqudah et al. (2016) observed what they called "abnormal activity" in the family Phyllidiidae when keeping them in an aquarium. Therefore, it is not unlikely to think that other families of nudibranchs are affected the same way when kept in captivity.

4.2.2 Experiments with dorid nudibranchs preying on Bryozoa zooids

Compared to the cladobranch species used in the previous experiments, *O. muricata* is a much slower mover and feeder. Hardly any movement was detected when just looking at the nudibranch from above in the temperature gradient table. However, when the 4.5 hours had passed, it was clear that the nudibranch had moved and eaten (visible as darker patches of empty zooids). According to Harvell (1984), the spines induced on Bryozoa through predator exposure formed within two days. Since these experiments were only run for 4.5 hours, it is likely to think that spines had not had the chance to form yet. However, looking at this from the perspective of the optimal foraging theory, studies have found that dorid nudibranchs will most likely choose to move on to the next colony instead of preying on a colony with spines already induced (Adler and Harvell, 1990). This might serve a reason for why some of the dorid *O. muricata* used in the current study did not dig into the food items right below them. If the lamina cut-out given to them were already induced with spines from beforehand, the cost might have exceeded the benefit of feeding.

Pratt and Grason (2007) found that *O. muricata* prefers *E. pilosa* over *M. membranacea* when given the choice. However, the ingestion rates were not constant as *O. muricata* was observed not engaging in feeding on several occasions. In the present study, no significant difference in the ingestion rates when preying on the two different species of Bryozoa, *E. pilosa* and *M. membranacea*, were found in relation to temperature. Hypothesis 4 stating that *nudibranchs show temperature-dependent ingestion rates when feeding on different species of Bryozoa* can thus be rejected. This applies also for hypothesis 1 that can be rejected for experiment 4 and 5 as well.

Previous studies on nudibranch interactions with Bryozoa provided conflicting results as e.g. ingestion rates in the range between 0.09-30 polypides per hour being reported earlier (Pratt and Grason, 2007). In the current study, ingestion rates ranging between 0.2-45.3 polypides per hour were found for *E. pilosa* and 3.6-36.0 polypides per hour for *M. membranacea*. Several studies have found a relationship between *O. muricata* feeding and their body size (Pratt and Grason, 2007, Seroy and Grünbaum, 2018), however it was not possible to conduct a statistical test on this in the current study's experiments.

Seroy and Grünbaum (2018) used nudibranchs of an average size of 5.3 mm in their study while Pratt and Grason (2007) used nudibranchs with an average size of 6.5 mm. In the current study, nudibranchs with an average size of 7.4 mm were used. It has also been suggested earlier that once *O. muricata* specimens reach a certain size, their ingestion rates are considered to be no longer directly related to their body size (Todd and Havenhand, 1989, Pratt and Grason, 2007). This might have applied in the current study since nudibranchs had already reached a large size when they were used in the present experimental trials.

A thin coating of some sort of sticky matter formed on the lamina of the kelp a couple of days after being harvested. The liveliness of the zooids of Bryozoa was therefore best the first day after the lamina had been collected from the sea. The first experiment with *M. membranacea* used lamina that was collected one day before. The experiment with *E. pilosa* used lamina that was collected five days before. For the latter experiment, it was harder to detect filter feeding activities from the zooids. In a similar experiment conducted by Todd and Havenhand (1989), fresh bryozoans were collected every two weeks and experiments were still conducted successfully. Hence, this should not necessarily have affected the current feeding trials significantly.

Some nudibranchs only eat the internal structures of the Bryozoa. A question that arises is whether this will help to better secure photosynthesis for the kelp underneath? Other nudibranchs, like *Polycera quadrilineata*, eat both the hard skeleton (zooecium) and the polypide (the internal animal). This indicates a difference in grazing potential between the species of dorid nudibranchs with different implications on kelp performance when colonized by Bryozoa. The same factors are viable for dorid nudibranchs as for cladobranch nudibranchs when it comes to prey choice and niche differentiation. Certain morphological structures like radula and mandible creates differences in species, which also creates differences in the feeding methods they have evolved to use. Some might be adapted to puncturing the zooids of Bryozoa and sucking out the content, while others might crush the zooids and scrape out the content (Jussi Evertsen, personal communication May 10, 2022).

Studies have shown that it differs not only how many individual zooid each nudibranch feeds, but also in which region the Bryozoa-eating nudibranchs feed (Todd and Havenhand, 1989, Best and Winston, 1984). Some have suggested that the middle parts of the colonies are more calcified than the outermost growing regions. This could reduce the nudibranch's ability to eat as much as it wants if placed in the middle regions in experiments. This might pose a limitation to this study design, as sections of colonized lamina were cut out, leaving growing zooids out. The nudibranch *P. quadrilineata* has been documented to constantly feed on the growing margins of *M. membranacea* after each re-initiation of experiments (Todd and Havenhand, 1989). As the specimens in this study was put directly in the middle of the lamina cut-out at the beginning of the experiment, this might not have had any effects. There does not seem to be a pattern of where the nudibranchs have eaten, other than around the area where they were placed. However, if the central zooids would have been more calcified, it might have restricted the nudibranch's feeding efficiency, in regard to time spent and amount of food they were able to ingest.

When Bryozoa colonies are preyed upon, and the marginal growth-zooids are eaten, they lose their ability to asexually reproduce. This will then stop the growth of that bryozoan colony completely (Todd and Havenhand, 1989). It is unlikely that an entire Bryozoa colony will be fully consumed and thus removed from one kelp individual, however, the large number of nudibranchs grazing on the same kelp lamina simultaneously exert a predation pressure of great intensity.

4.3 Methodological constraints

Despite the original plan, selective feeding of nudibranchs on natural fouling communities could not be analyzed in the present study due to the lack of polyps on the settling plates. Therefore, predation preference experiments with a greater variety of prey are necessary to understand the predation potential of cladobranch nudibranchs. The fouling community on the settling plates would have needed to be dense and colonized by a variety of potential prey species. However, the structure of the build-up needs to be limited to 2D rather than 3D, as nooks and crannies are hard to photographically quantify. Additionally, transparent plates, rather than the grey PVC plates that were used in this experiment, would be preferable. This is because when using a temperature gradient table with solid sides, it is necessary to visually see when nudibranchs approach prey items and when they are voluntarily not eating.

When it comes to the temperature treatments chosen for the current experiments, they could profitably have included more extreme values to provoke any difference between the ingestion rates. Additionally, the period of acclimation could have been done at the nudibranch's respective temperature regimes, instead of keeping all of the specimens in one unanimous tank. Prior to the experiments, the fact that the nudibranchs needed to be starved was chosen to be of greater importance. Both starving and temperature acclimating the nudibranch specimens was, however, not feasible as they would have had to be kept in 800 mL beakers without the possibility of any aeration. This would not have been in line with ethics and poses animal welfare issues although invertebrates do not fall under regulations regarding animal experiments as e.g. vertebrates do.

Undoubtedly, feeding preference studies benefit from having a large sample size. For most experiments in the current study, the replicate number for each temperature treatment was 5, however, due to unforeseen challenges with the nudibranchs, some experiments only had 3 replicates. The extent of damage from the loss of one or two replicates could have been minimized if the current study had a larger sample size. However, the number of nudibranch species and the number of individuals found of each species can vary from season to season (Jussi Evertsen, personal communication May 10, 2022). Hence, it is difficult to anticipate how many one can find prior to the actual sampling, which may lead to an unfavorable statistical result. By using several other nudibranch species as well, the extent of predation potential on benthic scyphozoans could be better interpreted. Perhaps one could do experiments with a natural fouling community of bryozoan species as well.

Additionally, carbon analysis for the nudibranchs were done a while after experiments were conducted. This meant that the treatment that had been applied to each nudibranch was unknown, as they had been mixed after the experiments, as well as being used during several consecutive experiments might having biased the outcome. The carbon concentration and size values were therefore extrapolated thus making it impossible to account for the correct variations between individuals. A perhaps bigger flaw to this part of the experiments was that only one of the two species of cladobranch nudibranchs were used to extract carbon concentration. Individuals of *F. auriculata* and *F. bostoniensis* were quite similar in size as well as circumference, however as they are not the same species and potentially have different prey preferences, their carbon concentrations are not necessarily equal. However, when the time of the C:N analyses came, too many of the species *F. auriculata* had died. As a result of this, only *F. bostoniensis* was analyzed and the data provided from the C:N analyses were used in statistical analyses for both species regardless. This bears a bias to these experiments. If similar studies were to be conducted

in the future, actual analyses of the carbon concentrations for the specimens (both prey and predator) used during the experiments should be prioritized.

As for the Bryozoa settling issue on kelp lamina, several different studies regarding this can further deepen the understanding of the issue. Another angle here could be to use e.g. the species *P. quadrilineata* and *Limacia clavigera* (O. F. Müller, 1776), which are frequently found feeding on Bryozoa (Jussi Evertsen, personal communication May 10, 2022). Also, conducting the same experiments as above, but doing the C:N analysis of the prey items would be useful. It seems as if there is a greater chance of getting a significant result when using carbon specific ingestion rates, than when regular ingestion rates are used.

There has been a trend of identifying nudibranchs based on what they feed on, as they were believed to be specialists (Jussi Evertsen, personal communication May 10, 2022). However, recent studies have found that this hypothesis does not hold anymore as their feeding choices are wider than believed (e.g. *Doto* sp. Oken, 1815) (Martinsson et al., 2021). Hence, conducting experiments with nudibranchs in captivity could lead to drawing conclusions that are not true, or even underdetermining the results that were found. *In situ* studies might be hard to conduct but would provide useful information towards the discussion of whether nudibranchs are really opportunistic species, rather than specialists like first believed based on experiments in captivity.

5 Conclusion and future perspective

Despite original assumptions, nudibranchs have a wider prey preference than first believed. Cladobranch nudibranchs have successfully been shown to feed on scyphozoan polyps, while dorid nudibranchs have shown that their prey can comprise several species of Bryozoa. Although any temperature-dependent ingestion rates were not found to be significant in this study, it is important to keep future warming trends in mind to assess temperature impacts on trophic ecology on longer terms. In the future, conducting studies with more species of scyphozoans will be interesting, especially for *Cyanea* sp. as they are more hazardous to humans as well as negatively interfering with aquaculture and fisheries. By looking at the possibility of transplanting natural predators of scyphopolyps into areas where blooms frequently occur, one could speculate that local populations of adult medusae could be reduced, or at least it would contribute to minimizing the risk of large jellyfish blooms. Possible areas of application could be e.g. artificial substrates introduced by humans like aquaculture pens, feed barges, wind parks or piers. This way of potentially applying biocontrol could be done in the proximity of seaweed farms as well, where dorid nudibranchs could be used to clean the kelp lamina from fouling organisms like bryozoans. Acting in the same way as cleaner fish, the dorid nudibranchs could enhance seaweed production as well as prolong the growth period. Biocontrolling, using native predators to stimulate production of commercially important species or to control mass occurrences of nuisance species could therefore act at a remedy in several industries, eventually increasing the economic gain of those industries.

Bibliography

ADLER, F. R. & HARVELL, C. D. 1990. Inducible defenses, phenotypic variability and biotic environments. *Trends in Ecology & Evolution,* 5**,** 407-410.

ALQUDAH, A., SAAD, S., SUSANTI, D., HADRY, N. F., KHODZORI, M. F. A., YUSOF, M. H. & RANI, M. H. 2016. Observations on nudibranch behaviour patterns under laboratory conditions. *Jurnal Teknologi,* 78.

ANDERSEN, H. K. 2011. *Gastropods Associated with Laminaria hyperborea and Saccorhiza polyschides in a Norwegian Kelp Forest: Comparison of Sampling and In Situ Imaging Techniques.* Master's Thesis, NTNU.

ARMSTRONG, E. J., TANNER, R. L. & STILLMAN, J. H. 2019. High heat tolerance is negatively correlated with heat tolerance plasticity in nudibranch mollusks. *Physiological and Biochemical Zoology,* 92**,** 430-444.

ÀVILA, C., TYNDALE, E. & KUZIRIAN, A. M. 1998. Feeding behavior and growth of *Hermissenda crassicornis* (Mollusca: Nudibranchia) in the laboratory. *Marine & Freshwater Behaviour & Physiology,* 31**,** 1-19.

BAKKEN, T. 2000. Topografien i Trondheimsfjorden *In:* SAKSHAUG, E. & SNELI, J.-A. (eds.) *Trondheimsfjorden* Trondheim: Tapir forlag

BAKKEN, T., EVERTSEN, J. & SKAUGE, C. 2021a. *Facelina auriculata* (Müller, 1776) [Online]. Available: https://www.artsdatabanken.no/Pages/308075/ [Accessed January 25 2022].

BAKKEN, T., EVERTSEN, J. & SKAUGE, C. 2021b. *Facelina bostoniensis* (Couthouy, 1838) [Online]. Available: https://www.artsdatabanken.no/Pages/308076/ [Accessed January 25 2022].

BAKKEN, T., EVERTSEN, J. & SKAUGE, C. 2021c. Nakensnegler Nudibranchia Ducrotay-Blainville, 1814 [Online]. Available: https://artsdatabanken.no/Pages/301001/Nakensnegler [Accessed February 11 2022].

BAKKEN, T., EVERTSEN, J. & SKAUGE, C. 2021d. *Onchidoris bilamellata* (Linnaeus, 1767) [Online]. Available: https://www.artsdatabanken.no/Pages/313967/ [Accessed January 25 2022].

BAKKEN, T., HOLTHE, T. & SNELI, J.-A. 2000. Strøm, vannutveksling og tidevann. *In:* SAKSHAUG, E. & SNELI, J.-A. (eds.) *Trondheimsfjorden.* Trondheim: Tapir forlag.

BAYER, M. M. & TODD, C. D. 1997. Evidence for zooid senescence in the marine bryozoan *Electra pilosa*. *Invertebrate Biology***,** 331-340.

BEGON, M., HARPER, J. L. & TOWNSEND, C. R. 1990. *Ecology: individuals, populations and communities,* Punjab, National Agro Industries.

BEST, B. A. & WINSTON, J. E. 1984. Skeletal strength of encrusting cheilostome bryozoans. *The Biological Bulletin,* 167**,** 390-409.

BROCKINGTON, S. & CLARKE, A. 2001. The relative influence of temperature and food on the metabolism of a marine invertebrate. *Journal of Experimental Marine Biology and Ecology,* 258**,** 87-99.

BROWN, J. H., GILLOOLY, J. F., ALLEN, A. P., SAVAGE, V. M. & WEST, G. B. 2004. Toward a metabolic theory of ecology. *Ecology,* 85**,** 1771-1789.

BÅMSTEDT, U., GIFFORD, D. J., IRIGOIEN, X., ATKINSON, A. & ROMAN, M. 2000. Feeding. *In:* HARRIS, R. P., WIEBE, P. H., LENZ, J., SKJODAL, H. R. & HUNTLEY, M. (eds.) *ICES zooplankton methodological manual.* London, UK: Elsevier Academic Press.

CAMPBELL, N. A., REECE, J. B., URRY, L. A., CAIN, M. L., WASSERMAN, S. A., MINORSKY, P. V. & JACKSON, R. B. 2015. *Biology: A global Approach* Harlow, Pearson Education Limited.

CAWOOD, A. M. 2012. *Laboratory and in situ investigations of factors affecting the growth and survivorship of the Scyphozoan jellyfish Aurelia sp1.* Doctor of Philosophy, University of California
CEH, J., GONZALEZ, J., PACHECO, A. S. & RIASCOS, J. M. 2015. The elusive life cycle of scyphozoan jellyfish–metagenesis revisited. *Scientific reports,* 5**,** 1-13.
CHADWICK, S. R. & THORPE, J. P. 1981. An investigation of some aspects of bryozoan predation by dorid nudibranchs (Mollusca: Opisthobranchia). *Recent and Fossil Bryozoa. Olsen & Olsen, Fredensborg***,** 51-58.
CHI, X., MUELLER-NAVARRA, D. C., HYLANDER, S., SOMMER, U. & JAVIDPOUR, J. 2019. Food quality matters: Interplay among food quality, food quantity and temperature affecting life history traits of *Aurelia aurita* (Cnidaria: Scyphozoa) polyps. *Science of the Total Environment,* 656**,** 1280-1288.
CLARK, K. B. 1975. Nudibranch life cycles in the Northwest Atlantic and their relationship to the ecology of fouling communities. *Helgoländer Wissenschaftliche Meeresuntersuchungen,* 27**,** 28-69.
CLARKE, A. 2006. Temperature and the metabolic theory of ecology. *Functional Ecology,* 20**,** 405-412.
CONKLIN, E. J. & MARISCAL, R. N. 1977. Feeding behavior, ceras structure, and nematocyst storage in the aeolid nudibranch, *Spurilla neapolitana* (Mollusca). *Bulletin of Marine Science,* 27**,** 658-667.
COYNE, J. A. 1973. An investigation of the dynamics of population growth and control in scyphistomae of the scyphozoan *Aurelia aurita*. *Chesapeake Science,* 14**,** 55-58.
DENLEY, D., METAXAS, A. & SHORT, J. 2014. Selective settlement by larvae of *Membranipora membranacea* and *Electra pilosa* (Ectoprocta) along kelp blades in Nova Scotia, Canada. *Aquatic Biology,* 21**,** 47-56.
DIXON, J., SCHROETER, S. C. & KASTENDIEK, J. 1981. Effects of the Encrusting Bryozoan, *Membranipora membranacea*, on the Loss of Blades and Fronds by the Giant Kelp, *Macrocystis pyrifera* (Laminariales) 1. *Journal of Phycology,* 17**,** 341-345.
DOYLE, T. K., HAYS, G. C., HARROD, C. & HOUGHTON, J. D. R. 2014. Ecological and societal benefits of jellyfish. *Jellyfish blooms.* Dordrecht: Springer.
DUARTE, C. M., PITT, K. A. & LUCAS, C. H. 2014. Introduction: Understanding jellyfish blooms. *Jellyfish Blooms.* Dordrecht: Springer.
EDMUNDS, M. 1966. Protective mechanisms in the Eolidacea (Mollusca Nudibranchia). *Zoological Journal of the Linnean Society,* 46**,** 27-71.
EVERTSEN, J. & BAKKEN, T. 2005. Nudibranch diversity (Gastropoda, Heterobranchia) along the coast of Norway. *Fauna norvegica,* 25**,** 1.
EVERTSEN, J. & BAKKEN, T. 2013. Diversity of Norwegian sea slugs (Nudibranchia): new species to Norwegian coastal waters and new data on distribution of rare species.
FALKENHAUG, T. 2021. Flere maneter til Sørlandet. *NRK*, Available: https://www.nrk.no/sorlandet/flere-maneter-til-sorlandet-1.15502413 [Accessed May 5, 2022].
FAULKNER, D. J. & GHISELIN, M. T. 1983. Chemical defense and evolutionary ecology of dorid nudibranchs and some other opisthobranch gastropods. *Marine ecology progress series. Oldendorf,* 13**,** 295-301.
FOLINO, N. C. 1993. Feeding and growth of the aeolid nudibranch *Cuthona nana* (Alder and Hancock, 1842). *Journal of molluscan studies,* 59**,** 15-27.
FOLK, R. L. 1954. The distinction between grain size and mineral composition in sedimentary-rock nomenclature. *The Journal of Geology,* 62**,** 344-359.
FRANZ, D. R. 1970. Zoogeography of Northwest Atlantic opisthobranch molluscs. *Marine Biology,* 7**,** 171-180.
FØRDE, H., FORBORD, S., HANDÅ, A., FOSSBERG, J., ARFF, J., JOHNSEN, G. & REITAN, K. I. 2016. Development of bryozoan fouling on cultivated kelp (*Saccharina latissima*) in Norway. *Journal of applied phycology,* 28**,** 1225-1234.
GLASER, O. C. 1910. *The nematocysts of eolids*, Williams & Wilkins.
GODDARD, J. & PEARSE, J. 2011. Long-term faunal changes in California nudibranchs: climate change and local ocean health. Santa Barbara: University of California

GOODHEART, J. A., BAZINET, A. L., VALDÉS, Á., COLLINS, A. G. & CUMMINGS, M. P. 2017. Prey preference follows phylogeny: evolutionary dietary patterns within the marine gastropod group Cladobranchia (Gastropoda: Heterobranchia: Nudibranchia). *BMC Evolutionary Biology,* 17**,** 1-14.
GOODHEART, J. A., BLEIDIßEL, S., SCHILLO, D., STRONG, E. E., AYRES, D. L., PREISFELD, A., COLLINS, A. G., CUMMINGS, M. P. & WÄGELE, H. 2018. Comparative morphology and evolution of the cnidosac in Cladobranchia (Gastropoda: Heterobranchia: Nudibranchia). *Frontiers in zoology,* 15**,** 1-18.
GORDON, D. P. 1972. Biological relationships of an intertidal bryozoan population. *Journal of natural History,* 6**,** 503-514.
GRAHAM, A. 1938. IX.—The structure and function of the alimentary canal of aeolid molluscs, with a discussion on their nematocysts. *Earth and Environmental Science Transactions of The Royal Society of Edinburgh,* 59**,** 267-307.
HARRIS, L. G. 1971. Nudibranch associations as symbioses. *In:* CHENG, T. C. (ed.) *Aspect of the Biology of Symbiosis.* Baltimore, MD: University Park Press.
HARRIS, L. G. 1973. Nudibranch associations. *In:* CHENG, T. C. (ed.) *Current Topics in Comparative Pathobiology.* Elsevier.
HARVELL, C. D. 1984. Predator-induced defense in a marine bryozoan. *Science,* 224**,** 1357-1359.
HAVENHAND, J. N. & TODD, C. D. 1988. Physiological ecology of *Adalaria proxima* (Alder et Hancock) and *Onchidoris muricata* (Müller)(Gastropoda: Nudibranchia). I. Freeding, growth, and respiration. *Journal of experimental marine biology and ecology,* 118**,** 151-172.
HERDMAN, W. A. 1890. Memoirs: On the Structure and Functions of the Cerata or Dorsal Papillæ in some Nudibranchiate Mollusca. *Journal of Cell Science,* 2**,** 41-64.
HERMANSEN, P., LARSEN, P. S. & RIISGÅRD, H. U. 2001. Colony growth rate of encrusting marine bryozoans (*Electra pilosa* and *Celleporella hyalina*). *Journal of Experimental Marine Biology and Ecology,* 263**,** 1-23.
HERNROTH, L. & GRÖNDAHL, F. 1985a. On the biology of *Aurelia aurita* (L.) 3. Predation by *Coryphella verrucosa* (Gastropoda, Opisthobranchia), a major factor regulating the development of *Aurelia* populations in the Gullmar Fjord, western Sweden. *Ophelia,* 24**,** 37-45.
HERNROTH, L. & GRÖNDAHL, F. 1985b. On the biology of *Aurelia aurita* (L.): 2. major factors regulating the occurrence of ephyrae and young medusae in the Gullmar Fjord, western Sweden. *Bulletin of Marine Science,* 37**,** 567-576.
HOETT, S. 2019. *Ecological study of nudibranchs in the Armona biodiversity hotspot.* Master's Thesis, Univeristy of Algarve.
HOLST, S. & JARMS, G. 2010. Effects of low salinity on settlement and strobilation of Scyphozoa (Cnidaria): is the lion's mane *Cyanea capillata* (L.) able to reproduce in the brackish Baltic Sea? *Hydrobiologia,* 645**,** 53-68.
HOOVER, R. A., ARMOUR, R., DOW, I. & PURCELL, J. E. 2012. Nudibranch predation and dietary preference for the polyps of *Aurelia labiata* (Cnidaria: Scyphozoa). *Hydrobiologia,* 690**,** 199-213.
IKEDA, H., MIZOTA, C. & UYE, S.-I. 2017. Bioenergetic characterization in *Aurelia aurita* (Cnidaria: Scyphozoa) polyps and application to natural polyp populations. *Marine Ecology Progress Series,* 568**,** 87-100.
JACOBSON, P. 1983. Physical oceanography of the Trondheimsfjord. *Geophysical & Astrophysical Fluid Dynamics,* 26**,** 3-26.
JI, R., EDWARDS, M., MACKAS, D. L., RUNGE, J. A. & THOMAS, A. C. 2010. Marine plankton phenology and life history in a changing climate: current research and future directions. *Journal of plankton research,* 32**,** 1355-1368.
KAMIYAMA, T. 2011. Planktonic ciliates as a food source for the scyphozoan *Aurelia aurita* (sl): feeding activity and assimilation of the polyp stage. *Journal of Experimental Marine Biology and Ecology,* 407**,** 207-215.
KEEN, S. L. 1991. *Clonal dynamics and life history evolution in the jellyfish Aurelia aurita.* Doctor of Philosophy, University of California, Davis.

LAMBERT, W. J. 1991. Coexistence of hydroid eating nudibranchs: do feeding biology and habitat use matter? *The Biological Bulletin,* 181**,** 248-260.
LAMBERT, W. J., BELL, G. R. R. & HARRIS, L. G. 2016. Growth and reproduction of the dorid nudibranch *Onchidoris muricata* fed native and invasive Bryozoan prey. *American Malacological Bulletin,* 34**,** 40-50.
LAMBERT, W. J., LEVIN, P. S. & BERMAN, J. 1992. Changes in the structure of a New England (USA) kelp bed: the effects of an introduced species? *Marine ecology progress series. Oldendorf,* 88**,** 303-307.
LISOVA, E. D. & VORTSEPNEVA, E. V. 2022. New data on nudibranchs rhinophore morphology and their spicule complex in *Onchidoris muricata* (Doridina, Gastropoda). *Zoologischer Anzeiger,* 296**,** 58-70.
LORENTZEN, E. A. 2022. -Mykje brennmaneter, sa du? *Dykking.* Available: https://dykking.no/nyheter/79-nyheter/880-mykje-brennmaneter-sa-du [Accessed May 5, 2022] Havforskningsinstituttet.
LUCAS, C. H. 2001. Reproduction and life history strategies of the common jellyfish, *Aurelia aurita*, in relation to its ambient environment. *Hydrobiologia,* 451**,** 229-246.
LUCAS, C. H. & DAWSON, M. N. 2014. What are jellyfishes and thaliaceans and why do they bloom? *In:* PITT, K., LUCAS, C. (ed.) *Jellyfish blooms.* Dordrecht: Springer.
LUCAS, C. H., GRAHAM, W. M. & WIDMER, C. 2012. Chapter Three - Jellyfish Life Histories: Role of Polyps in Forming and Maintaining Scyphomedusa Populations. *In:* LESSER, M. (ed.) *Advances in Marine Biology.* Academic Press.
MACLEOD, P. & VALIELA, I. 1975. The effect of density and mutual interference by a predator: a laboratory study of predation by the nudibranch *Coryphella rufibranchialis* on the hydroid *Tubularia larynx*. *Hydrobiologia,* 47**,** 339-346.
MARTINSSON, S., MALMBERG, K., BAKKEN, T., KORSHUNOVA, T., MARTYNOV, A. & LUNDIN, K. 2021. Species delimitation and phylogeny of *Doto* (Nudibranchia: Dotidae) from the Northeast Atlantic, with a discussion on food specialization. *Journal of Zoological Systematics and Evolutionary Research,* 59**,** 1754-1774.
MILLER, M. C. 1961. Distribution and food of the nudibranchiate Mollusca of the south of the Isle of Man. *The Journal of Animal Ecology,* 30**,** 95-116.
MOEN, F. E. & SVENSEN, E. 2020. *Dyreliv i havet, Norsk marin fauna,* Norge, Kolofon.
MYRVOLD, S. 2020. *Effect of temperature on the settling rate, survival, ingestion rate and biochemical composition of the polyp stages of A. aurita.* Master's Thesis, NTNU.
NORGES GEOLOGISKE UNDERSØKELSE - NGU. 2021. *Bunnsedimenter (kornstørrelse)*. Available: https://geo.ngu.no/kart/marin_mobil/
NYBAKKEN, J. & MCDONALD, G. 1981. Feeding mechanisms of west American nudibranchs feeding on Bryozoa, Cnidaria and Ascidiacea, with special respect to the radula. *Malacologia,* 20**,** 439-449.
O'CONNOR, R. J., SEED, R. & BOADEN, P. J. S. 1979. Effects of environment and plant characteristics on the distribution of Bryozoa in a *Fucus serratus* L. community. *Journal of Experimental Marine Biology and Ecology,* 38**,** 151-178.
PENNEY, B. K. 2008. Phylogenetic comparison of spicule networks in cryptobranchiate dorid nudibranchs (Gastropoda, Euthyneura, Nudibranchia, Doridina). *Acta Zoologica,* 89**,** 311-329.
PICTON, B. E. & MORROW, C. C. 1994. *A field guide to the nudibranchs of the British Isles*, Immel Publishing.
PIRES, V. L. 2012. *Impact of global warming on the spawning success, metabolism and embryogenesis of a specialized soft coral-feeding nudibranch, Armina maculata.* Master's Thesis, Universidade de Lisboa.
PRATT, M. C. & GRASON, E. W. 2007. Invasive species as a new food source: does a nudibranch predator prefer eating an invasive bryozoan? *Biological Invasions,* 9**,** 645-655.
PURCELL, J. E., UYE, S.-I. & LO, W.-T. 2007. Anthropogenic causes of jellyfish blooms and their direct consequences for humans: a review. *Marine Ecology Progress Series,* 350**,** 153-174.

RAJA-SALLEH, T. S. A., SIANG, H. Y., YUSUF, Y. & NORAINY, M. H. 2019. Embryonic and larval development of the nudibranch *Phyllidiella nigra*. *Aquaculture, Aquarium, Conservation & Legislation,* 12**,** 2085-2092.
RATNER, B. 2009. The correlation coefficient: Its values range between+ 1/− 1, or do they? *Journal of targeting, measurement and analysis for marketing,* 17**,** 139-142.
REKSTAD, M., MAJANEVA, S., BORGERSEN, Å. L. & ABERLE-MALZAHN, N. 2021. Occurrence and Habitat Characteristics of *Aurelia* sp. Polyps in a High-Latitude Fjord. *Frontiers in Marine Science,* 8.
REKSTAD, M. E. 2019. *Distribution, interspecific variation and habitats of scyphozoan polyp colonies in Trondheimsfjorden.* Master's thesis, NTNU.
RSTUDIO TEAM 2020. RStudio: Integrated Development Environment for R. *In:* RSTUDIO (ed.) *PBC.* Boston, MA.
RUDMAN, W. B. 1999a. *Cerata (ceras) in aeolids.* [Online]. Sea Slug Forum: Australian Museum, Sydney. Available: http://www.seaslugforum.net/find/ceras [Accessed February 11 2022].
RUDMAN, W. B. 1999b. *Rhinophore in nudibranchs* [Online]. Sea Slug Forum: Australian Museum. Available: http://www.seaslugforum.net/showall/rhinonud [Accessed May 10 2022].
RUSSELL, H. D. 1942. Observations on the feeding of *Aeolidia papillosa* L., with notes on the hatching of the veligers of Cuthona amoena A. and H. *Nautilus,* 55**,** 80-82.
RYLAND, J. S. 1977. Physiology and ecology of marine bryozoans. *Advances in marine biology,* 14**,** 285-443.
RYLAND, J. S. & STEBBING, A. R. D. 1971. Settlement and orientated growth in epiphytic and epizoic bryozoans. *In:* CRISP, D. J. (ed.) *Fourth European marine biology symposium.* Cambridge: Cambridge University Press.
SALVINI-PLAWEN, L. V. 1972. Cnidaria as food-sources for marine invertebrates. *Cahiers de Biologie Marine,* 13**,** 385-400.
SAUNDERS, M. & METAXAS, A. 2007. Temperature explains settlement patterns of the introduced bryozoan *Membranipora membranacea* in Nova Scotia, Canada. *Marine Ecology Progress Series,* 344**,** 95-106.
SCHEIBLING, R. E., HENNIGAR, A. W. & BALCH, T. 1999. Destructive grazing, epiphytism, and disease: the dynamics of sea urchin-kelp interactions in Nova Scotia. *Canadian Journal of Fisheries and Aquatic Sciences,* 56**,** 2300-2314.
SEROY, S. K. & GRÜNBAUM, D. 2018. Individual and population level effects of ocean acidification on a predator-prey system with inducible defenses: bryozoan-nudibranch interactions in the Salish Sea. *Marine Ecology Progress Series,* 607**,** 1-18.
SHI, W. & WANG, M. 2010. Characterization of global ocean turbidity from Moderate Resolution Imaging Spectroradiometer ocean color observations. *Journal of Geophysical Research: Oceans,* 115.
SHUNATOVA, N. N. & OSTROVSKY, A. N. 2001. Individual autozooidal behaviour and feeding in marine bryozoans. *Sarsia,* 86**,** 113-142.
STRICKER, S. A. 1989. Settlement and metamorphosis of the marine bryozoan *Membranipora membranacea*. *Bulletin of Marine Science,* 45**,** 387-405.
SWENNEN, C. 1961. Data on distribution, reproduction and ecology of the nudibranchiate molluscs occurring in the Netherlands. *Netherlands Journal of Sea Research,* 1**,** 191-240.
TAKAO, M., OKAWACHI, H. & UYE, S.-I. 2014. Natural predators of polyps of *Aurelia aurita* sl (Cnidaria: Scyphozoa: Semaeostomeae) and their predation rates. *Plankton and Benthos Research,* 9**,** 105-113.
THOMPSON, T. E. 1988. Molluscs: benthic Opisthobranchs: Mollusca, Gastropoda: keys and notes for the identification of the species. *In:* PLATT, H. M. & WARWICK, R. M. (eds.) *Free living Marine Nematodes - Part II - British Chromadorids.* London: The Bath Press.
TODD, C. D. 1979a. The annual cycles of two species of *Onchidoris* (Opisthobranchia: Nudibranchia). *In:* NAYLOR, E. & HARTNOLL, R. G. (eds.) *Cyclic Phenomena in Marine Plants and Animals.* Pergamon Press.

TODD, C. D. 1979b. Reproductive energetics of two species of dorid nudibranchs with planktotrophic and lecithotrophic larval strategies. *Marine Biology,* 53**,** 57-68.
TODD, C. D. & HAVENHAND, J. N. 1989. Nudibranch-bryozoan associations: the quantification of ingestion and some observations on partial predation among Doridoidea. *Journal of Molluscan Studies,* 55**,** 245-259.
TYNDALE, E., AVILA, C. & KUZIRIAN, A. M. 1994. Food detection and preferences of the nudibranch mollusc *Hermissenda crassicornis*: experiments in a Y-maze. *The Biological Bulletin,* 187**,** 274-275.
VALDÉS, Á. 2004. Phylogeography and phyloecology of dorid nudibranchs (Mollusca, Gastropoda). *Biological Journal of the Linnean Society,* 83**,** 551-559.
VAN WALRAVEN, L., DRIESSEN, F., VAN BLEIJSWIJK, J., BOL, A., LUTTIKHUIZEN, P. C., COOLEN, J. W. P., BOS, O. G., GITTENBERGER, A., SCHRIEKEN, N. & LANGENBERG, V. T. 2016. Where are the polyps? Molecular identification, distribution and population differentiation of *Aurelia aurita* jellyfish polyps in the southern North Sea area. *Marine biology,* 163**,** 1-13.
WATERS, V. L. 1973. Food-preference of the nudibranch *Aeolidia papillosa*, and the effect of the defenses of the prey on predation. *The Veliger,* 15**,** 174-192.
WILLCOX, S., MOLTSCHANIWSKYJ, N. A. & CRAWFORD, C. 2007. Asexual reproduction in scyphistomae of *Aurelia* sp.: Effects of temperature and salinity in an experimental study. *Journal of Experimental Marine Biology and Ecology,* 353**,** 107-114.
WORMS EDITORIAL BOARD. 2022. *Bryozoa* [Online]. Available: https://marinespecies.org/aphia.php?p=taxdetails&id=146142 [Accessed January 31 2022].
WÄGELE, H. & KLUSSMANN-KOLB, A. 2005. Opisthobranchia (Mollusca, Gastropoda)–more than just slimy slugs. Shell reduction and its implications on defence and foraging. *Frontiers in Zoology,* 2**,** 1-18.
WÄGELE, H., KLUSSMANN-KOLB, A., VERBEEK, E. & SCHRÖDL, M. 2014. Flashback and foreshadowing—a review of the taxon Opisthobranchia. *Organisms Diversity & Evolution,* 14**,** 133-149.
WÄGELE, H. & WILLAN, R. C. 2000. Phylogeny of the Nudibranchia. *Zoological Journal of the Linnean Society,* 130**,** 83-181.
AARNES, H. 2003. *Zoologi - om dyr og dyreliv* [Online]. UiO Institutt for biovitenskap. Available: https://www.mn.uio.no/ibv/tjenester/kunnskap/plantefys/leksikon/m/mosdyr.html [Accessed October 2021].

Appendices

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Appendix 1: Formulas

The calculations of approximate amount of *A. aurita* planula larvae in each beaker (planulae mL^{-1} $beaker^{-1}$) was done using formulas I-IV.

$$\text{Mean of planula larvae 1 mL}^{-1} = \frac{113+121+104}{3} = 112.66 \text{ planulae mL}^{-1} \quad \text{(I)}$$

$$\text{Planulae larvae in solution} = 90 \text{ mL} \times 112.7 \text{ planulae mL}^{-1} = 10\ 143 \text{ planulae} \quad \text{(II)}$$

$$\text{Planula larvae in each beaker} = \frac{10\ 143}{15 \text{ urine beakers}} = 676.2 \text{ beaker}^{-1} \quad \text{(III)}$$

$$\text{Planula larvae 1 mL}^{-1} \text{ in each beaker} = \frac{676.2 \text{ beaker}^{-1}}{104.5 \text{ mL in beaker}} = 6.47 \text{ planulae mL}^{-1} \text{ beaker}^{-1} \quad \text{(IV)}$$

Appendix 2: Temperatures

Temperature measurements (Table 3) in the temperature gradient table were taken every day during the cultivation of polyps, as well as for the days of experimental trials. Measurements were either taken in the treatment beakers, or in the beakers containing the replacement water. Using a digital thermometer, temperatures were given as °C (± 0.1). Room temperature was provided by the cooling device in the laboratory. Anomalous temperatures and NAs are bold.

Table 3: Temperature measurements in the temperature gradient table for all experimental trials (1-5). Temperatures are given in °C (± 0.1). Mean temperature per treatment is given at the bottom with standard deviation.

Treatment (°C)	11					13					15					Room temperature (°C)
Replicate	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5	
01.sep	12.0	**NA**	**NA**	**NA**	**NA**	13.4	**NA**	**NA**	**NA**	**NA**	14.7	14.8	**NA**	**NA**	**NA**	**NA**
02.sep	11.2	11.2	11.2	11.3	11.3	12.8	12.8	12.9	12.8	12.8	14.7	14.7	14.7	14.7	14.7	15.0
04.sep	11.3	11.3	11.3	11.2	11.2	12.7	12.7	12.7	12.8	12.8	14.6	14.6	14.7	14.7	14.7	**NA**
06.sep	11.1	11.2	11.2	11.2	11.2	12.8	12.7	12.8	12.8	12.8	14.6	14.6	14.6	14.6	14.7	15.4
07.sep	11.4	11.4	11.4	11.4	11.4	12.8	12.8	12.9	12.9	12.9	14.6	14.6	14.6	14.6	14.6	15.3
08.sep	11.5	11.4	11.4	11.4	11.4	13.0	13.0	13.0	13.0	12.9	14.7	14.6	14.7	14.7	14.7	15.8
09.sep	11.3	11.3	11.4	11.4	11.3	12.8	12.8	12.9	12.8	12.9	14.6	14.7	14.7	14.7	14.6	16.0
10.sep	11.3	11.3	11.3	11.3	11.2	12.9	12.8	12.9	12.8	12.8	14.7	14.7	14.7	14.7	14.7	16.0
11.sep	11.4	11.3	11.4	11.3	11.3	12.8	12.8	12.9	12.9	13.0	14.7	14.7	14.7	14.8	14.7	15.8
12.sep	11.3	11.3	11.3	11.2	11.2	12.9	12.8	12.9	12.8	12.9	14.7	14.7	14.7	14.7	14.7	15.4
13.sep	11.1	11.2	11.1	11.1	11.1	12.6	12.7	12.7	12.7	12.7	**14.1**	14.7	14.7	14.7	14.7	15.3
14.sep	**10.1**	**9.9**	**9.9**	**9.9**	**9.9**	**12.1**	**12.0**	**12.0**	**12.1**	**12.0**	**14.0**	**14.1**	**14.0**	**14.0**	**14.0**	16.0
15.sep	11.1	11.1	11.1	11.1	11.2	12.7	12.7	12.7	12.7	12.8	14.6	14.7	14.6	14.7	14.6	15.8
16.sep	**10.3**	**10.2**	**10.2**	**10.1**	**10.2**	**11.8**	**11.8**	**11.9**	**12.0**	**11.9**	**14.4**	**14.4**	**14.4**	**14.3**	**14.2**	13.5
17.sep	11.1	10.9	10.9	10.9	10.8	12.5	12.5	12.5	12.6	12.6	14.7	14.6	14.6	14.6	14.6	15.4
18.sep	10.8	10.7	10.7	10.7	10.8	12.2	12.1	12.1	12.2	12.1	14.2	14.2	14.1	14.0	14.0	15.3
19.sep	10.9	10.9	10.8	10.8	10.8	12.6	12.6	12.5	12.5	12.5	14.6	14.6	14.6	14.5	14.5	14.5
20.sep	10.9	10.9	10.8	10.8	10.8	12.5	12.5	12.5	12.5	12.5	14.6	14.6	14.6	14.6	14.5	14.8
21.sep	10.9	10.9	10.8	10.8	10.8	12.5	12.5	12.5	12.6	12.6	14.6	14.6	14.6	14.6	14.6	15.2
22.sep	10.3	10.3	10.3	10.3	10.3	**NA**	**NA**	**NA**	**NA**	**NA**	14.0	14.0	14.0	14.0	14.0	14.9
12.okt	11.5	11.5	11.4	11.4	11.4	12.9	12.9	13.0	13.0	13.0	14.7	14.6	14.6	14.6	14.6	14.7
13.okt	11.5	11.5	11.5	11.4	11.4	**NA**	**NA**	**NA**	**NA**	**NA**	14.8	14.7	14.7	14.7	14.7	14.6
11.nov	11.5	11.5	11.4	11.4	11.4	**NA**	**NA**	**NA**	**NA**	**NA**	14.7	14.7	14.8	14.7	14.7	15.3
15.nov	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	**NA**	14.5
16.nov	11.4	11.4	11.3	11.3	11.4	**NA**	**NA**	**NA**	**NA**	**NA**	14.5	14.6	14.6	14.6	14.6	**NA**
Mean	11.1 ± 0.43°C					12.6 ± 0.32°C					14.6 ± 0.22°C					15.2 ± 0.61°C

Appendix 3: Salinities

Salinity in the beakers was measured every day simultaneously with temperatures (Table 4). However, as there was little deviance from day to day, the salinity was only checked and not written down.

Table 4: Salinities measured in the temperature gradient table over the experimental period, - indicating lacking data. Mean and standard deviation are given at the bottom.

Treatment (°C)	11					13					15				
Replicate	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
07.sep	37.4	-	-	-	-	-	-	-	-	-	-	-	-	-	-
08.sep	34.7	34.7	34.7	34.6	34.6	34.8	34.7	34.7	34.7	34.7	34.8	34.9	34.8	34.9	34.8
09.sep	34.9	-	-	-	-	-	-	-	-	-	-	-	-	-	-
10.sep	34.7	-	-	-	-	34.8	-	-	-	-	35.0	35.2	35.0	35.2	35.1
11.sep	34.6	34.8	-	-	-	-	-	-	-	-	35.2	-	-	-	-
12.sep	34.6	34.5	-	-	-	-	-	-	-	-	-	-	-	-	-
13.sep	34.5	-	-	-	-	34.7	-	-	-	-	35.5	-	-	-	-
14.sep	34.5	-	-	-	-	-	-	-	-	-	-	-	-	-	-
12.okt	34.9	34.9	34.9	35.0	35.0	35.1	35.1	35.1	35.0	35.0	35.2	35.3	35.4	35.3	35.3
13.okt	35.0	34.9	34.9	35.1	35.0	-	-	-	-	-	35.5	35.5	35.6	35.5	35.5
Mean	34.9 ± 0.56					34.9 ± 0.18					35.2 ± 0.26				

Appendix 4: Carbon weight analysis

The C:N analysis provided results given below (Table 5).

Table 5: Values from C:N analysis. Sample ID represent species and replicate number, Fb being *Facelina bostoniensis*, and Om being *Onchidoris muricata*. Sample weight (mg) is how much was put into the C:N analyzer. µg C and N was used to calculate the C/N ratio.

Sample ID	Size (mm)	Sample weight (mg)	µg C nudibranch^{-1}	µg N nudibranch^{-1}	C/N ratio
Fb01	14	3.961	1325.927137	336.3542192	3.9420559
Fb02	10	5.114	1890.759551	204.0049469	9.2682044
Fb03	14	6.323	2118.346925	157.2199106	13.473783
Fb04	9	5.673	1995.298339	180.314814	11.065637
Fb05	12	5.423	1834.792573	181.7731262	10.09386
Fb06	21	16.569	6103.237719	317.0687844	19.248939
Fb07	10	2.883	822.626726	175.1363176	4.6970653
Fb08	14	2.118	623.953197	207.9632228	3.0003055
Fb09	8	10.095	3760.849437	125.4644185	29.975426
Fb10	10	2.261	702.5609133	168.559032	4.1680407
Fb11	10	8.227	2797.608379	93.56011897	29.901719
Fb12	12	2.368	747.7559537	1350.574048	0.5536579
Fb13	9	3.932	1330.895075	834.5924398	1.5946647
Fb14	11	9.224	3713.427678	440.1555808	8.4366252
Fb15	8	2.116	724.3670806	498.7483757	1.4523698
Fb16	7	2.439	821.0879843	471.9808445	1.7396638
Fb17	7	4.46	1537.060502	446.3668312	3.4434918
Fb18	6	1.42	510.8337038	640.9268536	0.7970234
Fb19	11	2.01	663.3889473	834.5108224	0.7949435
Fb20	9	1.187	339.8135605	367.5431067	0.9245543
Om01	9	12.418	3576.418911	505.1962451	7.0792666
Om02	9	11.099	2897.121395	462.2498856	6.2674356
Om03	9	18.48	4745.982702	346.0107722	13.716286
Om04	7	10.1	2687.230613	392.7430366	6.8422107
Om05	9	14.089	3731.592475	372.2459105	10.024536
Om06	8	14.316	4148.872682	523.6229544	7.9233973
Om07	8	9.514	2536.895905	308.3883416	8.2263029
Om08	8	11.32	3381.670281	457.0738437	7.3985207
Om09	7	7.85	2263.143431	472.1583088	4.7931878
Om10	7	6.53	1655.514458	366.6262078	4.5155377
Om11	6	7.33	2017.352566	362.6628385	5.5626117
Om12	6	7.332	1897.760893	585.0256006	3.2438938
Om13	8	9.51	2597.194963	534.7144728	4.8571623
Om14	7	5.839	1572.417279	711.4958526	2.2100161
Om15	6	7.781	2266.731618	579.5061283	3.9114886
Om16	8	9.265	2418.879533	874.2971527	2.7666561
Om17	6	6.366	1879.119819	505.8458012	3.7148076
Om18	6	6.375	1788.71499	755.5722759	2.3673645
Om19	6	9.959	2987.30099	783.6290753	3.8121365
Om20	8	9.976	2742.647844	680.2950149	4.0315566

Appendix 5: Statistical analysis output

The outputs for the statistically significant models run in R Studio are shown below. Linear model of carbon specific ingestion rate as a function of species (Figure 22), and carbon specific ingestion rate as a function of polyp age (Figure 23).

```
Call:
lm(formula = C.spec.feed ~ factor(Species), data = nudidata)

Residuals:
    Min      1Q  Median      3Q     Max
-214598  -17324  -10231   14345  231603

Coefficients:
                  Estimate Std. Error t value Pr(>|t|)
(Intercept)         268989      31555   8.524 2.88e-09 ***
factor(Species)FB1  -230819      40738  -5.666 4.51e-06 ***
factor(Species)FB2  -134253      51529  -2.605   0.0145 *
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 99790 on 28 degrees of freedom
Multiple R-squared:  0.5342,    Adjusted R-squared:  0.5009
F-statistic: 16.06 on 2 and 28 DF,  p-value: 2.263e-05
```

Figure 22: Carbon specific feeding as a function of species. P-values are less than 0.05. Intercept represents FA. Estimates for the other two species (FB1 and FB2) equals the difference in mean estimate from Intercept.

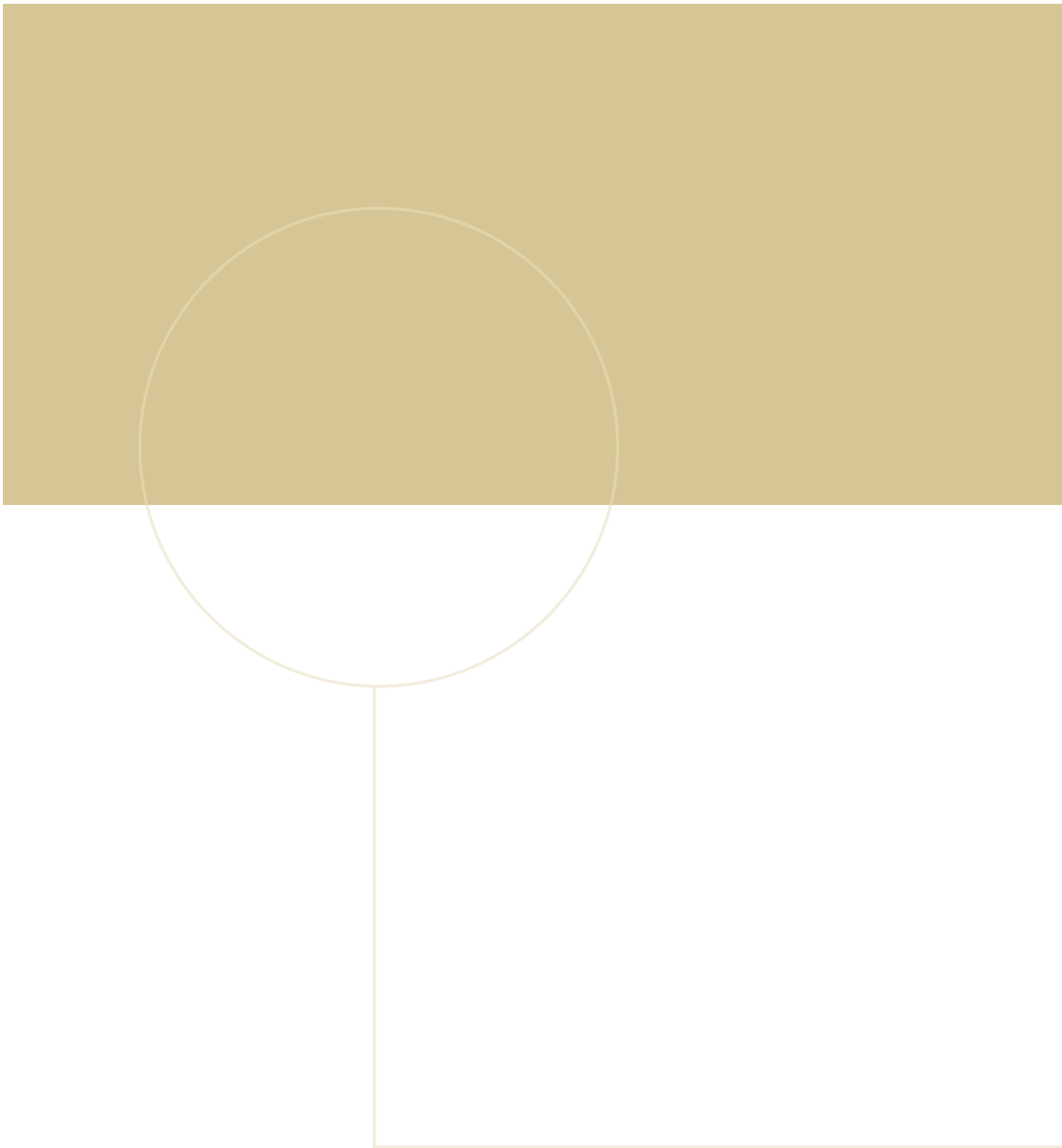
```
Call:
lm(formula = C.spec.feed ~ polypage, data = nudidata)

Residuals:
    Min      1Q  Median      3Q     Max
-219216  -25360   -5999    7160  258195

Coefficients:
            Estimate Std. Error t value Pr(>|t|)
(Intercept)   -25303      33046  -0.766     0.45
polypage        8079       1435   5.630 4.41e-06 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 99300 on 29 degrees of freedom
Multiple R-squared:  0.5222,    Adjusted R-squared:  0.5057
F-statistic:  31.7 on 1 and 29 DF,  p-value: 4.415e-06
```

Figure 23: Carbon specific feeding as a function of polyp age (days). Intercept represents carbon specific ingestion rate at polyp age of 0 days. P-value is less than 0.01.



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