University of Southampton

Faculty of Environmental and Life Sciences

Ocean and Earth Science

Grazing and Egg Production by *Calanus finmarchicus* across the Fram Strait: Implications for a Changing Arctic Ocean

by

Holly Elizabeth Jenkins

ORCID ID 0000-0002-1055-642

Thesis for the degree of Doctor of Philosophy

August 2023

University of Southampton

Abstract

Faculty of Environmental and Life Sciences

Ocean and Earth Science

Doctor of Philosophy

Grazing and Egg Production by *Calanus finmarchicus* across the Fram Strait: Implications for a Changing Arctic Ocean

by

Holly Elizabeth Jenkins

Copepods of the genus *Calanus* are fundamental to the biogeochemical and ecological functioning of the Arctic Ocean. They are an energy-rich food source for higher trophic levels, including commercially important fish stocks. They accumulate lipid stores and sequester them at depth during an overwintering period. Recent climatic warming has shifted the range of the typically Atlantic *Calanus finmarchicus* northwards into the Arctic. Rapid warming has also changed the timing, composition, distribution, and magnitude of the phytoplankton blooms on which *Calanus* spp. rely, altering the quantity and composition of the food available and potentially shifting towards flagellate-dominated communities.

To better understand the effects of the food environment on *C. finmarchicus* and the limits to its production, this thesis presents the composition of the plankton assemblage with rates of grazing and production of *C. finmarchicus* females across a spatiotemporal scale. Metabolic carbon budgets were created to investigate the relationships of ingestion and production. **Chapter 2** presents rates of ingestion, rates of production and gonad maturation in *C. finmarchicus* in spring 2018, finding a disconnect between ingestion and production and surplus carbon in the budget. The surplus carbon and highly variable grazing rates were attributed to the energetically expensive process of gonad maturation. **Chapter 3** presents rates of ingestion, rates of production and gonad maturation in *C. finmarchicus* in summer 2019 during post-bloom conditions. The microplankton assemblage was low in biomass and dominated by ciliates and flagellates. The animals were food limited, thus suggesting the project shift to a flagellate dominated system in the future Arctic may have negative consequences for *Calanus*. **Chapter 4** compares plankton taxonomic composition of the Fram Strait across seasons, revealed through metabarcoding to microscopic identification and to high-throughput flow imaging. The results provide evidence for the future shift to a plankton assemblage dominated by the smaller taxa. The flexible and diverse diet *C. finmarchicus* consumed in the high biomass plankton assemblage exceeded metabolic demand. However, when the plankton shifts towards its projected future composition, there are negative consequences for *C. finmarchicus.* The rates of grazing and production in *C. finmarchicus* presented here are vital measurements that form the basis of our understanding of phytoplankton-zooplankton interaction and will constrain future biogeochemical models.

Table of Contents

Table of Tables

Table of Tables

Table of Figures

Table of Figures

Table of Figures

Research Thesis: Declaration of Authorship

Print name: Holly Elizabeth Jenkins

Title of thesis: Grazing and Egg Production by *Calanus finmarchicus* across the Fram Strait: Implications for a Changing Arctic Ocean

I declare that this thesis and the work presented in it are my own and has been generated by me as the result of my own original research.

I confirm that:

1. This work was done wholly or mainly while in candidature for a research degree at this University;
2. Where any part of this thesis has previously been submitted for a degree or any other qualification at this University or any other institution, this has been clearly stated;
3. Where I have consulted the published work of others, this is always clearly attributed;
4. Where I have quoted from the work of others, the source is always given. With the exception of such quotations, this thesis is entirely my own work;
5. I have acknowledged all main sources of help;
6. Where the thesis is based on work done by myself jointly with others, I have made clear exactly what was done by others and what I have contributed myself;
7. Parts of this work have been published as:-

Jenkins et al. 2022

Jenkins et al. (in prep.)

Signature: ..... Date: 31/08/2023.....

Acknowledgements

I am deeply grateful to my primary supervisor Dr Daniel J. Mayor for his never-wavering patience, never-ending corrections, and never-fading enthusiasm for this project – despite my still being here years after the planned completion date. Dan's immense knowledge and experience has been essential to the shape of this PhD thesis, and the dedication he shows to science as a whole has been inspirational. I would also like to thank Dr Kathryn Cook for her technical support and for sharing her incredible zooplankton knowledge, and Professor Thomas Anderson for his thorough feedback and insight. My gratitude extends to the Faculty of Natural and Environmental Sciences – Ocean and Earth Science and the UKRI Changing Arctic Ocean Programme who allowed me the opportunity to do this work by funding the project. I would not have been able to submit this thesis without the help of Professor Andrew Cundy, Professor Martin Solan and Mary Smith. The data underpinning this thesis relied on the crew and scientists aboard cruises JR17005 and JR18007, the other Changing Arctic Oceans scientists, and the entertainment of Flo – so thank you! I am also grateful for the flexibility and support of the team at SOS-UK.

Finally, I would like to thank my family and friends for all their support and for not forgetting about me while I've been trapped behind a desk. Bagel, for getting me out walking, making me smile, and for the nose bumps if I ever dared work later than her walk time; Sam, for his dog sitting services, spelling and grammar check, for housing me during the cost of living crisis, for funding our pizza habit and for generally being a great person; Salma, for her love, positively, encouragement and feedback (and Hélias' adorable hands, noises, everything); Rhi, Liz, Ellen, Jon, Julian, and Jonny for their encouragement in my endeavour to leave Boomtown for a day of thesis writing and their celebration on my return; Lottie for bringing me plentiful snacks, teas, vitamins, medications, and her, Will and Sarah's tasty food; and my parents for caring for me in the final push.

In memory of Dr Barry Thornton

Definitions and Abbreviations

C .....Carbon

DHA .....Docosahexaenoic acid (22:6(n-3))

DNA .....Deoxyribonucleic acid

EGC.....East Greenland Current

EPA .....Eicosapentaenoic acid (20:5(n-3))

EPR .....Egg production rate

GGE .....Gross growth efficiency

Mol .....Mole

N.....Nitrogen

nSSU .....Nuclear small subunit

OUT .....Operational taxonomic unit

PCR .....Polymerase chain reaction

PUFA.....Polyunsaturated fatty acid

rRNA .....Ribosomal ribonucleic acid

WSC.....West Spitsbergen Current

Absorption efficiency .....the proportion of ingested food absorbed across the gut wall of the consumer, i.e. the availability of organic nutrients to meet requirements for metabolism and growth. Note that this term is sometimes used interchangeably with assimilation efficiency, but by definition assimilation efficiency is absorption minus respiration (Bochdansky, Deibel & Rivkin, 1999). Where the term assimilation has been used by others in place of absorption, the latter is quoted here.

Chapter 1 General introduction

1.1 The changing Arctic Ocean

The Arctic Ocean is the smallest of the world's oceans and the least connected, joining the Pacific Ocean via the Bering Strait and the Atlantic Ocean via the Fram Strait (Jones, 2001). The Arctic Ocean may be the smallest, but its size is not proportional to its global significance. The lower-than-average water salinity and temperature of this isolated pole drive global thermohaline circulation, making these sole two connecting straits areas of high scientific interest.

We are currently witnessing a monumental alteration to the ecosystems, biology, and biogeochemistry of the Arctic Ocean. Warming in parts of the Arctic is nearly four times the global mean rate (Rantanen et al., 2022), and this human-forced change will have global implications in terms of resources such as gas, oil and fish (Hassol, 2004; Degen et al., 2018), global ocean circulation and stratification (Greene & Pershing, 2007) and the associated nutrient cycles (Tremblay et al., 2015). The Arctic ecosystem is particularly vulnerable; with short growing seasons, lower biodiversity than temperate or tropical regions, and a highly variable climate (Hassol, 2004). Small temperature differences can have large effects on the extent and thickness of the sea ice, which organisms base their life cycles on (Smetacek & Nicol, 2005). Models predict that if climate change is not curbed, summer conditions in the Arctic will be ice-free by mid-century (Hartmann, Tank & Rusticucci, 2013).

The difference between the global warming rate and the Arctic warming rate is referred to as polar amplification (Holland & Bitz, 2003; Dai et al., 2019). Globally, the oceans have absorbed over 80 % of the heat added to the climate system, and despite having a larger thermal capacity than first thought, there has been a long term increase in temperature down to at least 700 m (Poloczanska et al., 2013; Caldeira & Wickett, 2003). In the Arctic, this anthropogenic climate warming first leads a reduction in sea ice thickness and extent. The darker colour of the open ocean compared to the ice causes the ocean to absorb more sunlight and implicitly heat, decreasing the ocean's albedo. This causes the Arctic to warm, melting more ice and further decrease the ocean's albedo, and reinforcing the loop. This positive feedback loop is thought to locally contribute up to 1°C per decade to surface warming in winter (Holland & Bitz, 2003). Arctic sea ice thickness in September may fall below 0.5 m by the end of this century, although internal variability gives this date an uncertainty of 10-20 years (Labe, Magnusdottir & Stern, 2018).

One of the biggest physical effects of temperature increase is on the circulation of the Arctic Ocean, which affects global ocean circulation and stratification (Greene & Pershing, 2007). The

Fram Strait (FIGURE 1) is key for the exchange of water masses between the Arctic and North Atlantic Oceans, as it is the only deep passage (Quadfasel, Gascard & Koltermann, 1987). It is characterised by two main water bodies with distinct hydrographic regimes. The relatively warm, salty West Spitsbergen Current is the main inflow, and the colder, fresher East Greenland Current the main outflow (FIGURE 1). The West Spitsbergen Current carries Atlantic water northwards, and the East Greenland current carries water and ice from the Atlantic southwards (Blachowiak-Samolyk et al., 2007; Maslowski et al., 2004). These processes affect the stocks of nutrients and organisms in the Fram Strait (Gluchowska et al., 2017).

In summer months, the ice-covered East Greenland current has a low standing stock of phytoplankton dominated by flagellates, whereas chain-forming diatoms dominate further East (Gradinger & Baumann, 1991). The mixing of the two water bodies and their rapidly changing physiochemical properties affects the stocks of nutrients and organisms in the Fram Strait: the freshening, warming waters are increasing stratified, reducing nutrient cycling and allowing different organisms to thrive (Gluchowska et al., 2017; Basedow et al., 2018). Additionally, the western Fram Strait is thought the be experiencing 'Atlantification', the increasing influence of Atlantic water in the Arctic, causing a decline in the lateral and vertical extent of polar water (Karpouzoglou et al., 2022).

Arctic warming is likely to simulate an increase in nutrient availability. Nutrient upwelling from the deep occurs in areas where the ice edge has retreated, facilitating an increase in production of phytoplankton (Carmack & Chapman, 2003). The change to nutrient supply means that the demand for nutrients also changes, allowing distinct types of phytoplankton to thrive. However, light is as much a controlling factor as nutrient availability in the winter months, and temperature controls metabolic rates. The intersection of these demands create DIFFFERNT SPECIES!!! MOVE ABOVE

Increased temperatures directly affect metabolic rates of the organisms within the Arctic Ocean in keeping with metabolic theory of ecology (Regaudie-De-Gioux & Duarte, 2012), and impacts the interactions between ice algae, phytoplankton, zooplankton, and their larger predators (Smetacek & Nicol, 2005). The effects of temperature increase are wide ranging and often interact with other factors – principally light and nutrient availability. However, biological responses to increased temperature can be classfied into three main mechanisms (Walther et al., 2002):

1. Reduction in body size
2. Changes to biogeographic distributions
3. Changes to phenology

Increasing temperatures are causing decreasing body, cell, and species size distributions across the planet (Peter & Sommer, 2012). A reduction in body size is supported by both theoretical and

observational evidence (Daufresne, Lengfellner & Sommer, 2009). The theory is governed by the Temperature Size Rule, which predicts that body size at maturity will be smaller when growth is under warmer conditions, because maturation is accelerated more strongly by warming than somatic growth (Atkinson, 1994). The mechanism behind the reduction in body size is still uncertain, and there seems to be many factors at play including repsiration mode and genome size (Verberk et al., 2021 and references within). There are experimental excpetions to the rule (Rüger & Sommer, 2012), but this particular study excludes the indirect effects that temperature change can have and so real-world applications are limited. The Temperature Size Rule is more likely a combination of the direct effects that the environment has on metabolic rate, and an indirect effect of behaviour and altered grazing pressure (Winder & Sommer, 2012).

The changing physical and chemical landscape is expected to change the composition, distribution, timing and magnitude of primary production (Castellani et al., 2022a; Lewis, Van Dijken & Arrigo, 2020; Neukermans, Oziel & Babin, 2018; Li et al., 2009; Rüger & Sommer, 2012; Kahru et al., 2011). The physio-chemical is linked to primary production primarily by indirect changes to water stratification and related differences in the availability of nutrients and light, resulting in an environment which is suitable for some species more than others, or changes to grazing pressure by heterotrophs. In this way, changes to the biogeographic distribution and the phenology of primary production are inextricably linked. These variations have resulted in a widespread advance in the timing of the phytoplankton spring bloom; one study found earlier blooms were detected in 11 % of the areas of the Arctic, and in these areas, the bloom had moved forward 50 days (Kahru et al., 2011). A decadal dataset from between 2003 -2016 found blooms have moved 20 days earlier (Hunter-Cevera et al., 2016). As a result, the timing of the peak suitability for the dominant genus of Arctic copepods (*Calanus)* (Blachowiak-Samolyk et al., 2007) is also shifting to earlier in the season (Freer, Daase & Tarling, 2021).

The Arctic spring bloom consists of both an ice algae bloom that begins growing under the ice before it melts, and a pelagic phytoplankton bloom. However, earlier ice melt would mean a reduced habitat for the ice algae. Picoplankton (< 2 μm diameter) can outcompete larger taxa (i.e. algae) because their large surface area to volume ratio allows efficient acquisition of nutrients and photons (Li et al., 2009). The reduction in ice cover and resultant increase in light to the Arctic changes phenology and bloom timing (Arrigo, van Dijken & Pabi, 2008; Aberle et al., 2012), potentially causing a mismatch in trophic interactions between the phytoplankton and the consumers (Søreide et al., 2010). Research to date has identified that both the abundance and composition of phytoplankton communities is changing (Arrigo, van Dijken & Pabi, 2008; Tremblay & Gagnon, 2009) but the mechanisms with which this affects the higher trophic levels are still poorly understood.

A changing zooplankton community composition affects downward carbon transport and so carbon sequestration (Brun et al., 2019). Hence, it is important to understand the controls on that zooplankton community. As the effects of climate change are most pronounced at the poles, it is logical to study these effects here first. Plankton communities are ideal indicator species for the effects of climate change. They are short lived, leading to tight coupling between their dynamics and the climate; and they are free-floating, meaning they respond strongly to changes in the environment.

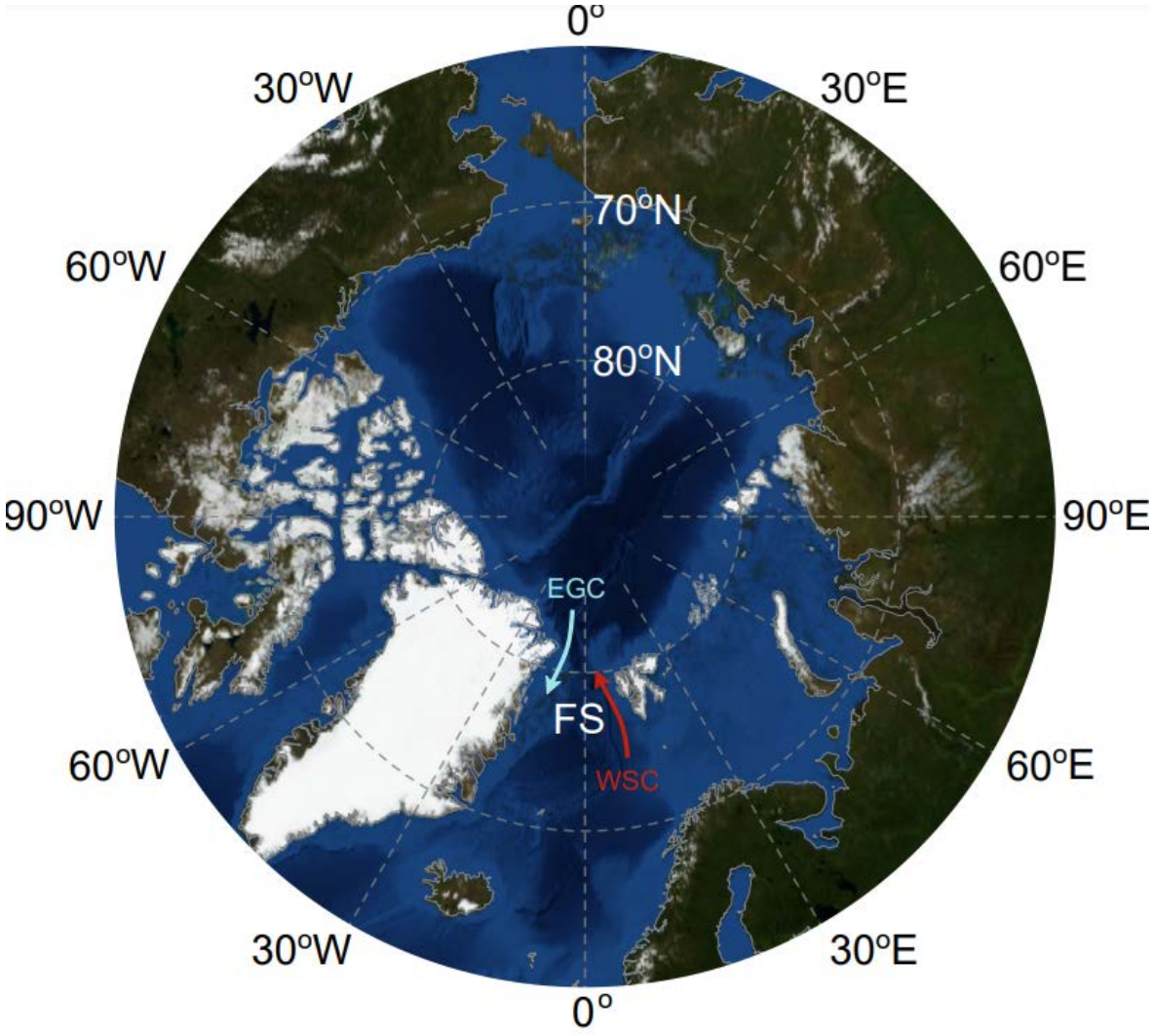


FIGURE 1 | The Fram Strait (FS) and surrounding areas. Svalbard is to the east of the Strait, and Greenland to the west. EGC and the blue arrow represent the East Greenland Current and its direction; WSC and the red arrow represent the West Spitsbergen Current. Water depth is shown as a gradient with deeper blues representing deeper water.

1.2 The importance of copepods

Copepods are among the most numerous multicellular animals in the world and are found in nearly every freshwater and marine environment. They are a diverse group of small crustaceans with over 13,000 known species (Boxshall & Defaye, 2008). CCopepods dominate Arctic biomass (Falk-Petersen et al., 2009; Ashjian et al., 2003; Auel & Hagen, 2002; Blachowiak-Samolyk et al., 2007; Cleary et al., 2017; Nöthig et al., 2015). In Kongsfjorden, Svalbard, 70-92% of zooplankton in July 1996-2002 were in the subclass Copepoda (Hop et al., 2006), in line with global estimates of 70 – 90 % (Turner, 2004). Their dominance partially explains their importance in the food web; at the most basic level, they are an important grazer of phytoplankton and prey for higher trophic levels, including the larvae of commercially important fish (Sakshaug, 2004). Copepods ingest carbohydrates and proteins in ice algae and phytoplankton and synthesize and store them as wax esters, which have long been known as an efficient energy store vital to higher trophic levels (Gatten & Sargent, 1973).

Copepods, especially the dominant taxa Calanus - the large-sized key Arctic grazer - play an important role in the global carbon cycle too (FIGURE 2). They ingest significant quantities of microplankton and egest dense faecal pellets. The carbon in these faecal pellets can sink and be remineralised at depth, sequestering carbon from the atmosphere to the deep ocean for long timescales (Fowler & Knauer, 1986; Jónasdóttir et al., 2015). The faecal pellets produced form part of the 'biological carbon pump' (Ducklow et al., 2001) – the biological control of vertical carbon transport (Cavan et al., 2017; Steinberg & Landry, 2017; Turner, 2015a).

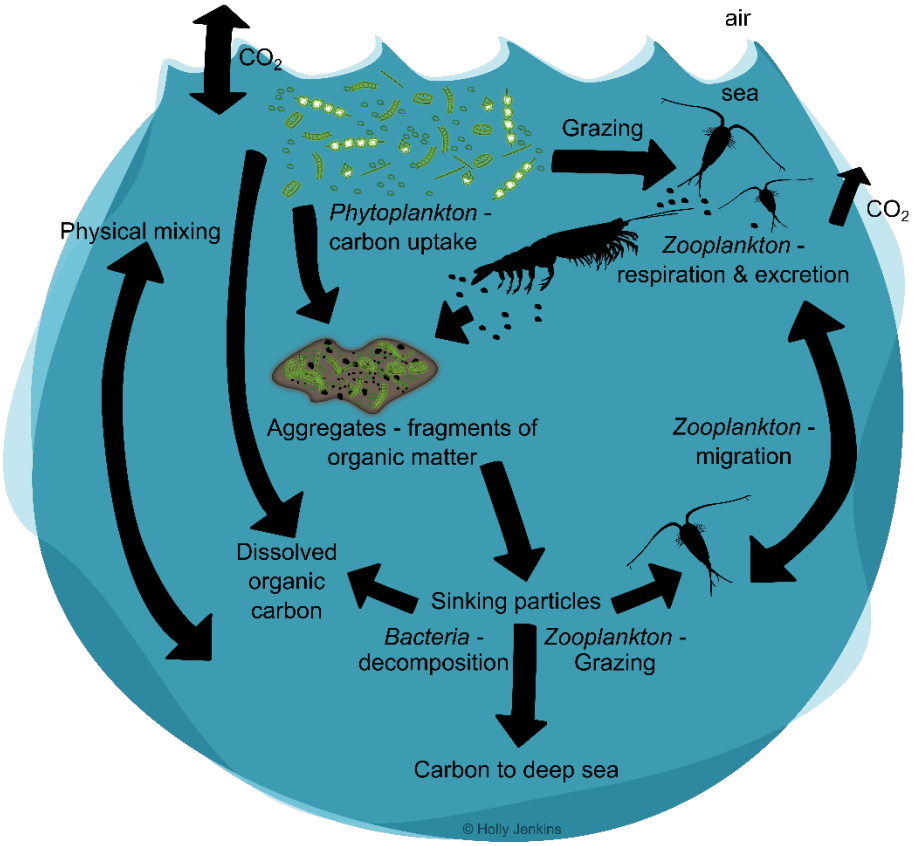


FIGURE 2 | A diagram showing the biological carbon pump. The pump sequesters carbon from the atmosphere into the surface ocean. The amount of carbon exported to depth depends on the strength of the pump.

Copepods significantly affect global biogeochemical cycles in ways beyond their egestion. *Calanus* facilitates the movement of carbon-rich lipid-based compounds into the oceans interior through daily vertical migrations (Darnis et al., 2017). Diel vertical migration is thought to balance the need to feed in the euphotic zone, where food is most abundant, with the need to reduce the risk of being eaten in waters where they themselves are visible (Berge et al., 2014) (although there are alternative explanations, some detailed by Lampert (Lampert, 1989)). When at depth, *Calanus* respires, excretes and egests material, translocating carbon, nitrogen and lipid below the mixed layer (Falk-Petersen et al., 2009; Jónasdóttir et al., 2015).

High latitude copepods have a life cycle that is adapted to the 24-hour winter darkness and 24-hour sunlight in summer, so it includes another important migration. *Calanus* migrate to depth in Autumn and enter a hibernation state called diapause where development is arrested, feeding ceases, and metabolism is lowered (Hirche, 1996a; Wilson et al., 2016; Maps, Record & Pershing, 2014). Diapause allows the copepods to survive the low winter temperature and food availability and to avoid predation, providing enough lipid is accumulated and stored by the copepods in the spring and summer phytoplankton blooms (Lee, Hagen & Kattner, 2006a; Schmid, Maps & Fortier,

2018). Diapause results in the transport of lipid to below the permanent thermocline (Jónasdóttir et al., 2015), enhanceing the efficiency of the7oilogical carbon pump, yet future climate change is likely to affect copepod lipid accumulation (Pond et al., 2012), with knock-on effects on diapause duration, timing and overall (Wilson et al., 2016).

There are three species of *Calanus* that inhabit the Arctic: *C. hyperboreus, C. glacialis, and C. finmarchicus*. The species *C. finmarchicus* traditionally inhabits the Atlantic, but recent Arctic warming has expanded its niche. *C. finmarchicus* typically feeds on the pelagic plankton bloom, while its congeners inhabit the seasonal ice-covered seas and the Arctic Ocean and rely on ice algae (Conover, 1988). All three have distinct phenology (population-level timing of life cycle events) linked to the seasonality in the food environment (Svensen et al., 2019). For example, the largest of the three, *C. hyperboreus* reproduces in winter, prior to the productive season (Falk-Petersen et al., 2007), *C. glacialis* reproduce prior to and during the ice algae bloom (Søreide et al., 2010) and *C. finmarchicus* has its main reproductive period during the open water spring bloom (Hirche, 1996b) while also continuing later in the year. As *Calanus* success and behaviour are so intrinsically linked to global biogeochemical cycles and food webs, a solid understanding of their physiology is essential to predict future changes: *Calanus* migration, ingestion, and export are relevant to the entire planet.

1.3 The physiology of calanoid copepods

Understanding the success of *Calanus* demands an understanding of the main physiological processes: ingestion (through grazing), production (including growth and egg production; often growth is assumed to be zero when the copepods are adults as they undergo no further moults, although there are some exceptions (Hirst & McKinnon, 2001)), respiration, egestion and excretion. In essence, this is what enters the copepod and what leaves, at rates that are limited by compounds within the copepods. The controls on these processes are poorly understood. Previously, it was thought that copepod success was governed by the quantity of food they had access to (Huntley & Boyd, 1984; Hirst & Bunker, 2003), but food quantity does not fully explain the variability in egg production rates . Now it is recognised that food ‘quality’ has a role (Noyon & William Froneman, 2013; Jónasdóttir, Fields & Pantoja, 1995; Anderson & Pond, 2000). Studies indicate that certain compounds are essential for copepod production and must be derived from their prey, as they cannot synthesize them in sufficient quantities otherwise (Jónasdóttir, Visser & Jespersen, 2009; Jónasdóttir, Fields & Pantoja, 1995; Kleppel & Hazzard, 2000). These compounds include polyunsaturated fatty acids (PUFAs). The long chain omega-3 fatty acids eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are two of these PUFAs, and are produced in high densities only by marine algae (Søreide et al., 2010). How rich a food source is in essential PUFAs,

or the DHA:EPA ratio, indicates its 'quality' (Vehmaa et al., 2011),These are key PUFAs as they play a role in the reproduction and growth of all marine organisms (Ackman, 1989). Being essential, consumers – copepods included – cannot synthesize EPA and DHA efficiently and must obtain them from their food. That said, recent evidence has found that *C. finmarchicus* feeding on an EPA-deplete dinoflagellate appeared to synthesize sufficient amounts of EPA from precursors to support growth (Bell et al., 2007). The lipid composition of many copepod species have been quantified (Kattner & Krause, 1987, 1989; Sargent & Falk-Petersen, 1988; Albers, Kattner & Hagen, 1996), wherePUFA levels correlated with limiting copepod egg production (Chen, Liu & Chen, 2012; Broglio et al., 2003; Pond et al., 1996; Jónasdóttir et al., 2002; Anderson & Pond, 2000; Jónasdóttir, Fields & Pantoja, 1995) and egg hatching (Jónasdóttir et al., 2005). Another study found PUFAs did not limit production when assuming a high utilisation efficiency of EPA (Mayor et al., 2009a). Other physiological roles of these compounds have only been researched more recently (Pond & Tarling, 2011; Pond, Tarling & Mayor, 2014).

Prey type can indicate its quality. Focusing on prey type avoids additional analysis of quantifying prey lipids.. Many studies correlate ciliate biomass with egg production (Jónasdóttir et al., 2005). Despite the fact that diatoms are considered to be higher in quality than dinoflagellates, egg production rates in some calanoid copepods are higher in copepods fed dinoflagellates or a mixture of dinoflagellates and diatoms (Vehmaa et al., 2011), suggesting it is not EPA or DHA alone that control production. It is necessary to find out what compound or compounds are limiting the Arctic dominating *Calanus* genus.

Zooplankton communities in the Fram Strait remained unquantified until 2003 when their structure was first comprehensively documented. It was found that, numerically, herbivores dominated in May, and copepod nauplii and one species of calanoid copepod, *Calanus finmarchicus,* were the most important herbivores in autumn (Blachowiak-Samolyk et al., 2007). There are contrasting hydrographic regimes across the Strait; the ice-covered East Greenland current has a low standing stock dominated by flagellates, whereas chain diatoms dominate further East (Gradinger & Baumann, 1991). However, there is little recent work, and this gap in knowledge means we are missing key insights into future ecological change. Phytoplankton community composition in the Fram Strait is expected to shift from diatom dominated to flagellate dominated in the summer (Li et al., 2009), although there is experimental evidence that this shift is not universal (Rüger & Sommer, 2012). What effect will this have on grazing by copepods? Some models predict that blooms of the eurythermal algae *Phaeocystis* spp. will benefit copepods in the area, due to the vast amounts of nutritious detritus that precede and follow a bloom (Weisse et al., 1994). However, there are reports of larger copepods having a low

ingestion rate when in a *Phaeocystis* spp. bloom, and other models agree with that (Saiz et al., 2013; Vernet et al., 2017).

1.4 Metabolic budgets

Calculating metabolic budgets for copepods allows the assessment of what limited their production (FIGURE 3). Metabolism, feeding, growth, and egg production of copepods in the Fram Strait have been previously examined (Hirche et al., 1991). Much is predicted and inferred about the life history of many copepods (Ji et al., 2012; Jager et al., 2017; Varpe, 2012). However, a mechanistic understanding of the physiology of copepods is needed to understand how they will be affected by future climate change. Different species have different life cycles (Hansen et al., 2012), and so a shift in the dominant species will cause a shift in overwintering patterns, and affect the biological carbon and lipid pumps. The poleward migration of *C. finmarchicus*, a typically temperate species with a shorter diapause than the endemic *C. hyperboreus* is just one example of this (Chust et al., 2014).

As discussed, copepod production is important to both the food web and the global carbon cycle. To understand production, a carbon budget must be created, balancing the inputs and the outputs. This is particularly relevant in a rapidly warming world – many physiological processes are dependent on temperature (Gillooly et al., 2001), meaning the budget may become mismatched in the future, or the increased metabolic costs may be balanced by increased ingestion (Anderson et al., 2017). Yet ingestion and respiration have different thermal responses so the changes are difficult to predict. A stoichiometric approach to modelling can ascertain the limiting nutrient or micronutrients.

Stoichiometry uses assumptions from the law of conservation of matter, the law of definite proportion, and Liebig's law of the minimum combined to determine the reactions between multiple elements (for detail see Sterner & Elser (Sterner & Elser, 2002)). When applied to biology, it can be used to investigate the constraints acting on biological processes such as production. The nutrient in lowest supply, relative to demand, is assumed to limit the process and can set the uptake rate of the most abundant. This is visualised in the famous Liebig's barrel (FIGURE 4).

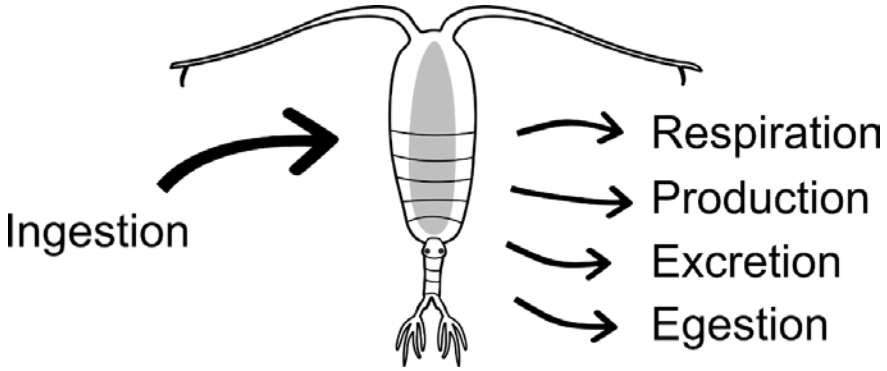


FIGURE 3 | A diagram representation of a simple metabolic budget for a copepod, highlighting the key inputs and outputs.

Phytoplankton typically reflect the nutrient ratios of their environment, which can vary widely (Moore et al., 2013; Mitra & Flynn, 2005; Finkel et al., 2010). This is because carbon fixation and nutrient assimilation are separate processes and only loosely coupled (Van De Waal et al., 2010). Contrarily, consumers take up carbon and nutrients simultaneously, so their carbon:nutrient ingestion matches their prey. In addition to carbon, consumers require more complicated nutrients in the form of nitrogen, phosphorus, vitamins, and a variety of micronutrients such as PUFAs (Hessen, 1992). Consumers, including copepods, are partially constrained in their chemical composition, as they have homeostatic requirements (Villar-argaiz, Medina-Sánchez & Carrillo, 2002). This highlights the need for a more detailed, mechanistic understanding of an organism's physiology, the stoichiometric constraints on these processes, and how that is driven by the changing environment.

To examine what limits the growth of a consumer organism using stoichiometry, the ratios of the elements in the biomass produced need to be quantified, and how efficiently the organism can handle each one. After this, increases and decreases in the diet can be investigated to determine what will affect growth and what is the limiting element. Quantifying a full budget of physiological processes is complex, but ingestion is the first step in doing so, especially in communities where top-down control dominates (Anderson et al., 2013). Arctic *Calanus* ingest prey through grazing, at a rate and selectivity that seem to relate to prey environment (Djeghri et al., 2018; Mayor et al., 2006; Kiørboe, 2011).

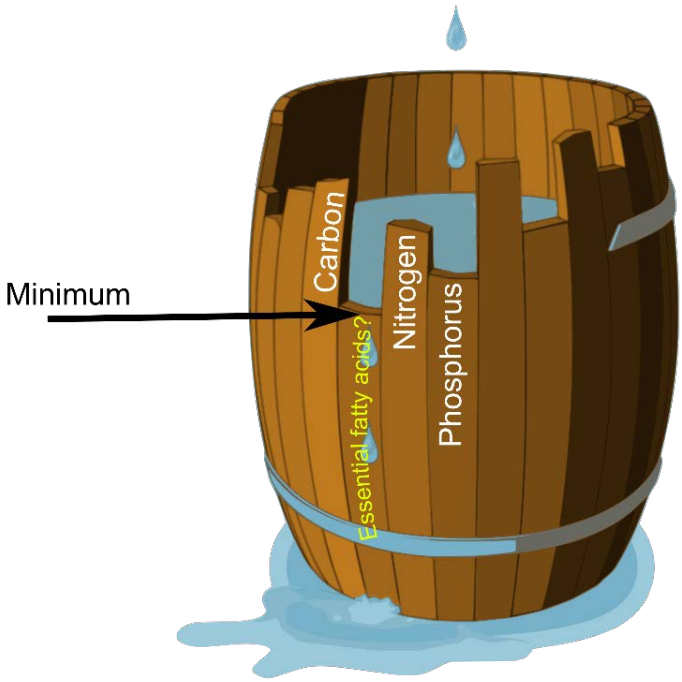


FIGURE 4 |- A diagram representing Liebig's law of the minimum shown by a barrel of water. The barrel can never be filled above the level of the shortest stave - the rate of the process can never be faster than the uptake rate of the element in shortest supply. In the case of limitation on copepod production, production is controlled by the component in the shortest supply, relative to demand.

Absorption efficiencies are the proportion of ingested prey that is absorbed through the gut wall, and utilisation efficiency is their turnover rate. Both are needed to create stoichiometric budgets. They both determine the availability of nutrients to meet the requirements for growth and metabolism. Absorption efficiencies are usually assumed to be 70% in ecosystem models (Steinberg & Landry, 2017), backed up by some laboratory studies of that magnitude (Madin & Purcell, 1992). There are some contrasting studies, finding efficiencies varying between <10% and >95% (Besiktepe & Dam, 2002), or varying between compounds (Mayor et al., 2011). Absorption efficiencies may be related to food quantity and quality, but this is yet to be fully tested (Anderson et al., 2017). Until recently, utilisation efficiencies were assumed to be high for limiting nutrients, but it has been shown that turnover rates of some PUFAs are higher than others (Mayor et al., 2015). Previously, Arctic copepods have had a carbon absorption efficiency of 37-49%, but this is when ingesting algal monocultures (Thor et al., 2007).

A metabolic carbon budget necessitates the quantification of production (assumed to be equal to growth in a mature female copepod) in addition to ingestion, so the gross growth efficiencies (GGE) can be calculated. GGEs are straightforward methods to assess energy flow through a system. In copepods, this is egg production as a fraction of intake, with an expected range of 0.2 – 0.3 (Straile, 1997).

1.5 Research questions

The overarching aim of this thesis was to investigate how grazing and production by *Calanus finmarchicus* are affected by the quantity and quality (i.e. composition) of the plankton assemblage in the Fram Strait. Spring and summer were compared to provide variation in the quantity and quality of the natural plankton assemblage. The relationship between ingestion and production was investigated using a metabolic carbon budget to elucidate the limiting factors.

Null hypothesis 1: Grazing and production by *Calanus finmarchicus* in spring do not correlate with any measure of food quantity or quality.

Alternative hypothesis 1: Grazing and production by *Calanus finmarchicus* in spring positively correlate with a measure of either quantity; quality; or both quality and quantity.

Null hypothesis 2: Grazing and production by *Calanus finmarchicus* in summer do not correlate with any measure of food quantity or composition.

Alternative hypothesis 2: Grazing and production by *Calanus finmarchicus* in summer positively correlate with a measure of either quantity; quality; or both quality and quantity.

Null hypothesis 3: Potential future changes to available food will affect neither grazing and production by *Calanus finmarchicus*.

Alternative hypothesis 3: Potential future changes to available food will have an effect on either grazing; production; or both grazing and production by *Calanus finmarchicus*.

At the moment, understanding of the mechanism that controls production in *C. finmarchicus* and the effects of different prey quality and quantity on the copepods is incomplete, with limited data in the field (see Table 2 (Mitra et al., 2014)). It is likely a balance between the two that limits production, but this needs to be ascertained in the rapidly changing Arctic.

This research will provide observations across a natural plankton assemblage to better understand the effects of dietary nutrient supply on consumer metabolism within the context of stoichiometric theory.

Chapter 2 Grazing, egg production and carbon budgets for *Calanus finmarchicus* across the Fram Strait

This paper has been published in Frontiers in Marine Science

Jenkins, H.E., Atherden, F., Cook, K.B., Anderson, T.R., Thornton, B., Mitchell, E., Jacob, E. & Mayor, D.J. (2022) Grazing, egg production and carbon budgets for Calanus finmarchicus across the Fram Strait. *Frontiers in Marine Science*. 9, 1–17. doi:10.3389/fmars.2022.981461.

HJ, KC, TA and DM contributed to the conception and design of the study. HJ conducted experiments, microscopy analysis, budget analysis and statistical analysis. BT performed the elemental analyses. EM determined plankton cell sizes. EJ analysed gonad maturity. HJ wrote the drafts of the manuscript and created the figures. All authors read and approved the submitted version.

2.1 Abstract

Calanoid copepods comprise around 90% of Arctic zooplankton biomass and are fundamental to the ecological and biogeochemical functioning of high-latitude pelagic ecosystems. They accumulate lipid reserves during the productive months and represent an energy-rich food source for higher trophic levels. Rapidly changing climate in the Arctic may alter the quantity and composition of the food environment for one of the key copepod species, *Calanus finmarchicus,* with as yet unquantified effects on its production. Here we present rates of feeding and egg production in female *C. finmarchicus* exposed to the range of feeding conditions encountered across the Fram strait in May/June 2018. Carbon (C) budgets were constructed and used to examine the relationship between feeding and growth (= egg production) in these animals. C-specific ingestion rates (mean ± standard deviation) were highly variable, ranging from 0.015 ± 0.004 to 0.645 ± 0.017 day^{-1} (mean = 0.295 ± 0.223 day^{-1}), and were positively correlated with food availability. C-specific egg production rates ranged from 0.00 to 0.049 day^{-1} (mean = 0.012 ± 0.011) and were not correlated with either food availability or ingestion rate. Calculated gross growth efficiencies (GGE: growth/ingestion) were low, 0.12 ± 0.13 (range = 0.01 to 0.39). The assembled C budgets indicate that the average fraction of ingested food that was surplus to the requirements for egg production, respiration and losses to faecal pellets was 0.17 ± 0.42. We suggest that this excess occurred, at least in part, because many of the incubated females were still undergoing the energetically (C-) expensive process of gonad

maturation at the time of sampling, an assertion that is supported by the relatively high C:N (nitrogen) ratios of the incubated females, the typically low egg production rates, and gonad maturation status. Ontogenetic development may thus explain the large variability seen in the relationship between egg production and ingestion. The apparently excessive ingestion rates may additionally indicate that recently moulted females must acquire additional N via ingestion to complete the maturation process and begin spawning. Our results highlight the need for improved fundamental understanding of the physiology of high-latitude copepods and its response to environmental change.

2.2 Introduction

Copepods are among the most numerous multicellular animals in the world, dominating zooplankton biomass in the Arctic (Mauchline et al., 1998; Nöthig et al., 2015). In the Fram Strait, 70-92 % of zooplankton biomass is in the subclass Copepoda (Hop et al., 2006) where the biomass is generally dominated by three key species in the genus *Calanus* (Hop et al., 2006; Blachowiak-Samolyk et al., 2007). These animals are important grazers of phytoplankton and represent high-quality prey for higher trophic levels (Gatten & Sargent, 1973) including the larvae of commercially important fish (Sakshaug, 2004). *Calanus* spp. also play an important role in ocean biogeochemistry due to their dense faecal pellets, their daily vertical migrations, and their ontogenetic migration to depth to over winter, all of which lead to sequestration of carbon in the deep ocean (Jónasdóttir et al., 2015).

The Arctic Ocean (FIGURE 5) is experiencing rapid, human-led change (Thomas et al., 2022). It is warming at three times the global mean rate (Dai et al., 2019; AMAP, 2021) leading to a cycle of sea ice loss, decreasing ocean albedo, increasing poleward ocean heat transport and increasing polar cloud cover (Holland & Bitz, 2003). The Fram Strait is both the main inflow and outflow gateway between the Arctic and the Atlantic and so has variable physicochemical conditions across its width. The relatively warm, salty West Spitsbergen Current (WSC) is the main inflow, and the colder, fresher East Greenland Current (EGC) the main outflow (FIGURE 5). The ice-covered East Greenland current has a low standing stock of phytoplankton dominated by flagellates, whereas chain-forming diatoms dominate further East (Gradinger & Baumann, 1991). The mixing of the two water bodies and their rapidly changing physiochemical properties affects the stocks of nutrients and organisms in the Fram Strait: the freshening, warming waters are increasing stratified, reducing nutrient cycling and allowing different organisms to thrive (Gluchowska et al., 2017; Basedow et al., 2018). Additionally, the western Fram Strait is thought the be experiencing ‘Atlantification’, the increasing

influence of Atlantic water in the Arctic (Karpouzoglou et al., 2022). For one key species in the *Calanus* genus – *Calanus finmarchicus* – recent Atlantification seems to have allowed a range expansion, with them now completing their life cycles further north in the Arctic (Tarling et al., 2022b). The changing physical and chemical ocean environment is expected to change the composition, distribution, timing and magnitude of primary production (Li et al., 2009; Kahru et al., 2011; Yool, Popova & Coward, 2015; Neukermans, Oziel & Babin, 2018; Lewis, Van Dijken & Arrigo, 2020) – the food on which copepods rely. With a different food environment comes the potential for changes to the productivity of *Calanus* spp., and in turn their population success. Understanding how *Calanus* spp. will respond to changing food environment is essential for predicting how the ecological and biogeochemical functioning of Arctic pelagic ecosystems will change in the future.

Egg production in *Calanus* is often positively correlated with temperature (Pasternak et al., 2013) and also typically increases with food availability (Hirche & Bohrer, 1987; Runge, 1984a; Hirst & Bunker, 2003; Mayor et al., 2009b). Indeed, the effects of temperature and food availability on copepod reproduction likely interact because, as poikilotherms, their physiological rates increase with temperature. Ingestion rates may therefore increase with warming, providing the animals with more food to fuel increased reproductive rates, but only when sufficient resources are available (Anderson, Hessen & Mayor, 2021). When food is scarce, reproductive demands cannot be met by ingested food alone, and may instead be met from maternal biomass (Smith, 1990; Niehoff, 2004; Mayor et al., 2009a). This is termed capital breeding, as opposed to income breeding, where reproductive demands are met by ingested food only. Without capital resources, when food concentrations are not saturating, egg production, which is considered to be equivalent to growth in adult females (Poulet et al., 1995), may therefore decline with warming because of the higher metabolic costs associated with higher temperatures (Anderson, Hessen & Mayor, 2021).

The relationship between reproduction and ingestion in *Calanus* is further complicated by prey selection and how associated feeding behaviour influences the degree to which the available food is ingested. There is evidence for and against selective feeding by calanoid copepods, both dependent and independent of food availability (Kleppel, 1993; Koski & Wexels Riser, 2006). For example, there are numerous examples of dietary selection by food type: for large conic ciliates (Mayor et al., 2006; Leiknes et al., 2014), for diatoms (Kiørboe, Saiz & Viitasalo, 1996; Nejstgaard et al., 2008; Kiørboe, 2011; Peter & Sommer, 2012; Ray et al., 2016a, 2016b), directly by nutritional content (Cowles, Olson & Chisholm, 1988; Carroll et al., 2019), by size (Hansen, Tande & Berggreen, 1990; Meyer et al., 2002), by toxicity (Teegarden et al., 2008), motility or chemical cues. In contrast, other studies have suggested that *Calanus* shows little or no prey selectivity (Castellani et al., 2008; Mayor et al.,

2009a; Djeghri et al., 2018). Prey preference is rooted in achieving nutritional balance - copepods that ingest food which does not meet their stoichiometric demands can face decreased growth, egg production and hatching success (Jónasdóttir et al., 2002). Diatoms are thought to be key in the diet of *Calanus* (Irigoien et al., 2002; Kohlbach et al., 2021), positively correlating with both ingestion and production. Understanding patterns of prey selection by Arctic *Calanus* is a fundamental precursor to determining how the changing food environment will impact their ability to obtain the necessary resources to reproduce.

Our aim was to investigate the relationship between reproductive output and the food environment in female *Calanus finmarchicus* (Gunnerus, 1770) across a range of food environments in the Fram Strait in May - June 2018. We conducted a series of experiments in which rates of ingestion, prey selection, and egg production were measured for replicate groups of animals and determined the elemental content of the experimental animals. Metabolic budgets, which compare C intake to that lost via egg production, respiration, and egestion of faecal matter, are used to examine how the reproductive physiology of *C. finmarchicus* varies in response to the local food environment.

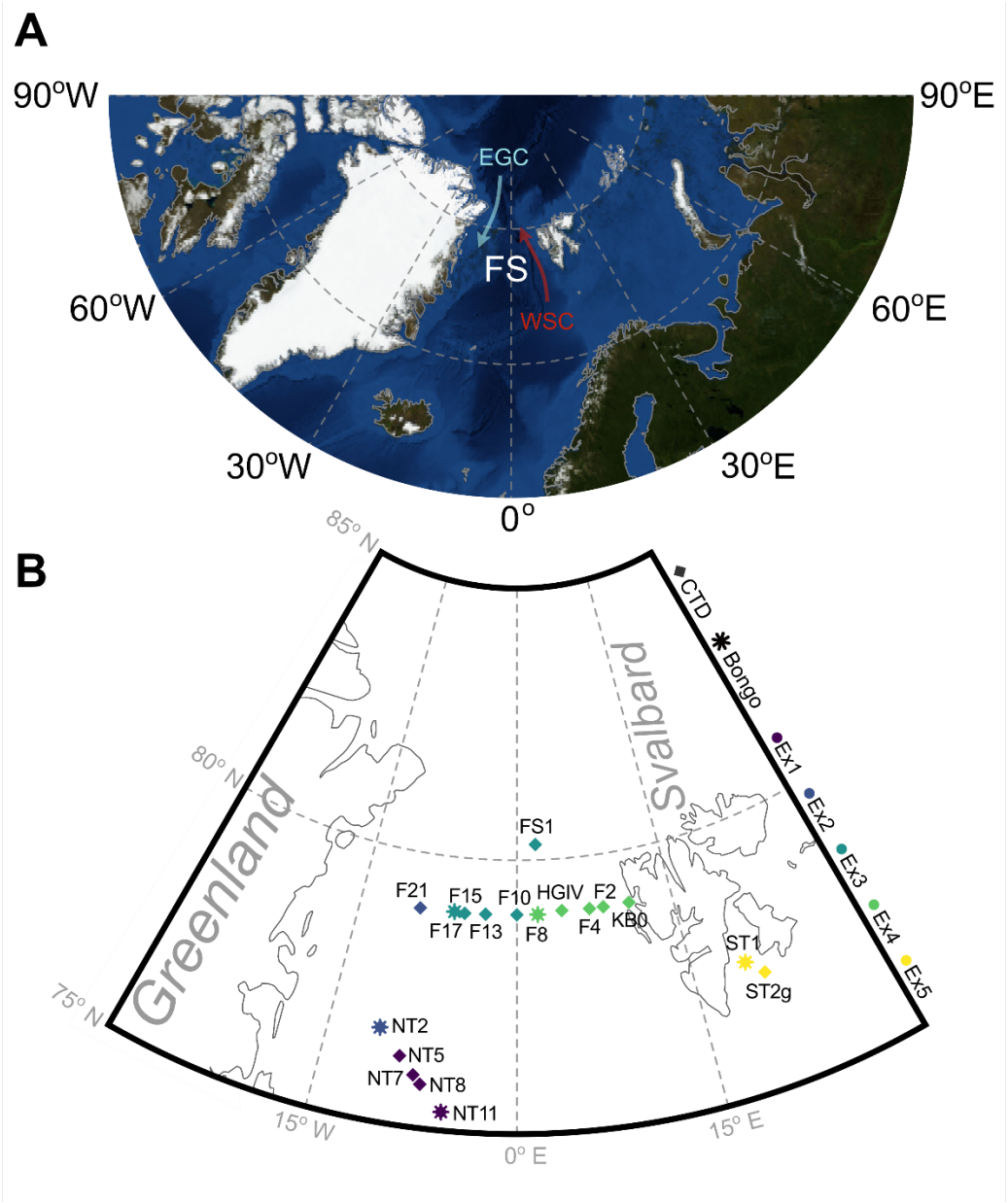


FIGURE 5 |The location of the Fram Strait (FS) in the Arctic Ocean (A) and the stations that were sampled on cruise JR17005 in May-June 2018 (B). The Arctic Ocean map is coloured bathymetrically showing the deep channel of the Fram Strait, where most water enters and exits the Arctic Ocean. Sequential long-term grazing experiments (1-5) are marked by Ex1-Ex5. Bongo and CTD show the locations where animals for experimental incubations and the natural plankton assemblage were collected, respectively.

2.3 Methods

2.3.1 Experimental procedure

Ingestion and egg production rates of female *Calanus finmarchicus* were measured simultaneously at 18 stations across the Fram Strait in May-June 2018 (FIGURE 5; TABLE 1; RRS James Clark Ross cruise JR17005). The natural plankton assemblage was collected daily from the chlorophyll maximum via 20 L Niskin bottles. Two 200 mL water samples were collected at each station and preserved with 1 % acidified Lugol's iodine for subsequent microplankton analysis. Copepods were collected using a motion-compensated bongo net fitted with a 200 µm mesh hauled vertically from 200 m and subsequently transferred into buckets containing surface seawater. Female *C. finmarchicus* were picked using a dissection microscope (Wilde M5) and swan necked forceps under gentle illumination. The identities of the animals collected at each station were verified using molecular analysis of the 16S rDNA barcode (Lindeque et al., 2022). All experimental work was conducted in a temperature-controlled room at 1.6 ± 1.1 °C.

Ex	Day	Station	Latitude (°N)	Longitude (°E)	Date	Temp (°C)	Depth (m)	Data
Ex1	1	NT11	75.3356	-5.46428	16/05/18	1.32	20	E B
Ex1	2	NT8	75.79556	-7.21797	17/05/18	1.28	20	M P
Ex1	3	NT7	75.94908	-7.81496	18/05/18	-0.74	6*	M P
Ex1	4	NT5	76.25775	-9.028673	19/05/18	-0.75	20	M E B
Ex2†	1	NT2	76.71327	-10.90499	20/05/18	-1.78	10	M B
Ex2†	3	F21	78.98491	-9.2813	23/05/18	-1.67	10	M B
Ex3	1	F17	78.99929	-5.98215	25/05/18	-1.52	22	M I E B G
Ex3	2	F15	78.98609	-4.99978	26/05/18	-1.50	32	M I E G
Ex3	3	F13	78.99685	-2.99575	27/05/18	-0.59	15	M I E G
Ex3	4	F10	78.99993	-0.00006	28/05/18	-0.58	8	M I E G
Ex3	5	FS1	80.28328	2.00005	29/05/18	-0.97	10	M I E B G
Ex4	1	F8	79.00002	2.00024	30/05/18	0.56	9	M I E B G
Ex4	2	HGIV	79.04837	4.33207	31/05/18	4.07	23	M I E G
Ex4	3	F4	79.03329	6.99998	01/06/18	4.31	12	M I E G
Ex4	4	F2	79.0333	8.33323	02/06/18	3.14	10	M I E G
Ex4	5	KB0	79.03509	10.84316	03/06/18	-0.66	20	M I E B G
Ex5	1	ST1	77.41672	19.50015	05/06/18	-0.53	23	M I E B
Ex5	2	ST2g	77.12498	20.74961	06/06/18	-0.83	17	M E B

TABLE 1 | Locations of the stations sampled on research cruise JR17005 in May and June 2018. Temp. is the water column temperature at the chlorophyll-a maximum. Depth is the depth of water sampled using the CTD, chosen to be at the chlorophyll maximum. Data

shows what was measured at that station: M = microplankton analysis; I = copepod ingestion; E = egg production; B = copepod biomass; G = gonad maturation stage. For *C. finmarchicus* abundance and body condition, please see Tarling et al. 2022. Measurements are associated with the experiment start time throughout. * denotes underway water sampling due to ice making CTD sampling impossible. † denotes disruption to sampling due to transit and heavy ice.

Ingestion and egg production rates were determined simultaneously using a series of sequential 24-hour particle removal experiments, as previously described (Mayor et al., 2009a). These were planned to last for a total of 5 days to allow the robust measurement of the change in biomass over that period, but experiments 1, 2 and 5 were curtailed due to adverse weather conditions and logistics. At the outset of each experiment, groups of 10 healthy and active female *C. finmarchicus* were transferred into replicate (n = 6) 2.2 L glass bottles containing natural seawater from the chlorophyll maximum and incubated for 24 hours on a plankton wheel at ~1 rpm (see **Error! Reference source not found.** in Appendix A). Three additional control bottles were incubated without the addition of copepods to account for microplankton growth during the incubations. Microplankton samples (100 mL) were collected from the control and grazed bottles at the start and end of each 24-hour incubation period and preserved with 1 % acidified Lugol's iodine. The remaining water from the grazed bottles was gently passed through a 63 µm mesh sieve to collect and enumerate any eggs produced by the experimental females during the incubation. Any eggs found in the microplankton samples were added to the respective sample's egg total. Nauplii were excluded as they were unlikely to have hatched from experimental eggs – hatching of *Calanus* eggs at these temperatures takes around five days (Corkett, McLaren & Sevigny, 1986). This procedure was repeated for up to 5 consecutive days, with experimental females being gently transferred into fresh seawater every day via a wide-bore (10 mm internal diameter) plastic dip tube. Replicate groups (n = 6) of 5 copepods were frozen in tin cups at -80 °C at the start and end of each experiment to determine any changes in the C and nitrogen (N) content of their biomass over the duration of the experiment. Elemental analysis of the freeze-dried experimental animals was conducted using a Flash EA 1112 Series Elemental Analyser (Thermo Fisher). The gonad maturation stage (GS) of ≥10 females from each station where the experimental animals were collected was determined following the description of Niehoff & Runge (Niehoff & Runge, 2003).

2.3.2 Microplankton analysis

Samples were gently agitated for one minute before being transferred to 25 mL Utermöhl sedimentation chambers and left to settle for 48 hours (Lund, Kipling & Le Cren, 1958). Cells were identified and enumerated with a Brunel SP95I inverted microscope at × 250 and × 400 magnification for cells >2 µm and small flagellates, respectively (Båmstedt et al., 2000; Mayor et al., 2006). Small and large diatom categories refer to centric cells with diameters <20 µm (e.g. *Chaetoceros* spp.) and ≥20 µm (e.g. *Thalassiosira* spp.), respectively. Cell dimensions were measured for each genus present using an ocular micrometer and their volumes were calculated by applying appropriate geometric formulae as is common practice (Hillebrand et al., 1999; Menden-Deuer & Lessard, 2000; Mayor et al., 2009b). Measurements of representative cells were repeated until the cell volumes for each group were normally distributed. Cell volumes were converted to carbon biomass using published conversion factors specific to the cell type (Menden-Deuer & Lessard, 2000; Malzahn & Boersma, 2012). Chlorophyll-*a* (CAS 479-61-8) was measured with an in-situ Chelsea Aqua 3 Fluorometer.

2.3.3 Carbon budgets

Metabolic C budgets were constructed for copepods according to (Equation 1):

$$I = E + R + W + \Omega \qquad (1)$$

where the terms, all expressed as biomass-specific rates per day, are ingestion (I), egg production (E), respiration (R), production of faecal matter (W) and a balancing term (Ω). The balancing term captures processes not specified in the simple budget. C-specific ingestion rates (day^{-1}) were estimated using the mean C content of the females within each experiment and the ingestion rate per experimental bottle.

Ingestion rates of individual animals (I_I; µmol C ind^{-1} day^{-1}) were calculated using established equations (Frost, 1972) and converted to biomass-specific rates (I; day^{-1}) as described above. Egg production (E_I; µmol C ind^{-1} day^{-1}) was measured and converted to C units assuming 20.9 nmol C egg^{-1} (Mayor et al., 2009a); Mayor, unpublished data). Gross growth efficiency (GGE) was then calculated as E/I, i.e., egg production as a fraction of intake. The respiration rate of individual animals was estimated from the globally-used equations of Ikeda et al. (Ikeda et al., 2001) (Equation 2):

$$\ln O_2 \text{ consumption rate } (\mu l\ O_2\ ind^{-1}\ hr^{-1}) = 1.640 + 0.843 \times \ln B + 0.068 \times T \qquad (2)$$

where B is body weight (mg N ind.$^{-1}$) and T is temperature ($^{\circ}$C) of the laboratory in which our experiments were conducted.

Values were converted to C units, R_I, (µg CO_2-C ind^{-1} hr^{-1}) by assuming a respiratory quotient (RQ) of 0.97 and multiplying by 12/22.4 (Ikeda et al., 2000). Conversion of R_I to the corresponding specific rate, R, was as described above. Production of faecal matter (W), i.e. the fraction of ingested food that does not pass across the gut wall, was calculated as I × 1- absorption efficiency (AE), using an assumed AE of 0.47 (Mayor et al., 2011). Excess (surplus) C, Ω, was calculated by difference using Equation 1. The sensitivity of the budget to the assumed values for RQ and AE was examined by changing these values to 0.7 (Mayzaud, 1976) and 0.74 (Anderson et al., 2017), respectively.

2.3.4 Statistical and computational analysis

Prey preference was examined by comparing the abundance of a cell type in the food environment relative to the abundance of that cell type ingested. Parametric statistical tests were used (ANOVA and Pearson's). Results were considered significant at the 0.05 level. When assumptions of normality, linearity and variance homogeneity were not maintained, non-parametric tests were used (Spearman's rho, ρ). Averages shown are mean average followed by standard deviation. Statistical analysis and data visualisation were done using the R programming environment (R Core Team, 2022) using the packages ggplot2 (Wickham, 2016) and viridis (Garnier, 2018).

2.4 Results

2.4.1 Dominant water masses

Water temperatures at stations NT7, NT2, F21, F17, F15, F13 and F10 were all < 0.0 °C (TABLE 1) showing the water body was the southerly moving East Greenland Current. At F8 there was a sharp increase in temperature showing the front between the East Greenland Current and the West Spitsbergen Current, where temperatures ranged from 0.58 to 4.31 °C. Station KB0, near the mouth of Kongsfjorden, and stations ST1 and ST2g in Storfjorden, also had temperatures < 0.0 °C due to their proximity to coastal meltwater runoff. Across all the stations, the average temperature at the chlorophyll-a maximum was 0.14 ± 1.9 °C (TABLE 1).

2.4.2 Microplankton

The composition and biomass of the microplankton varied considerably across the Fram Strait (FIGURE 6; Supplementary Table 1). Total biomass ranged from 18.0 to 187.0 µmol C L^{-1}, averaging 61.6 ± 46.6 µmol C L^{-1}. Stations in the area of East Greenland, NT2, F21, F17 and F15, were generally below this average, with 18.0, 21.0, 69.5, and 30.0 µmol carbon L^{-1}, respectively. Biomass was high at the two stations in the south of the study area, NT8 and NT5, where the water column contained 109.3 µmol C L^{-1} and 187.0 µmol C L^{-1}, respectively. Biomass was also high at station F13, where it reached 143.0 µmol C L^{-1}.

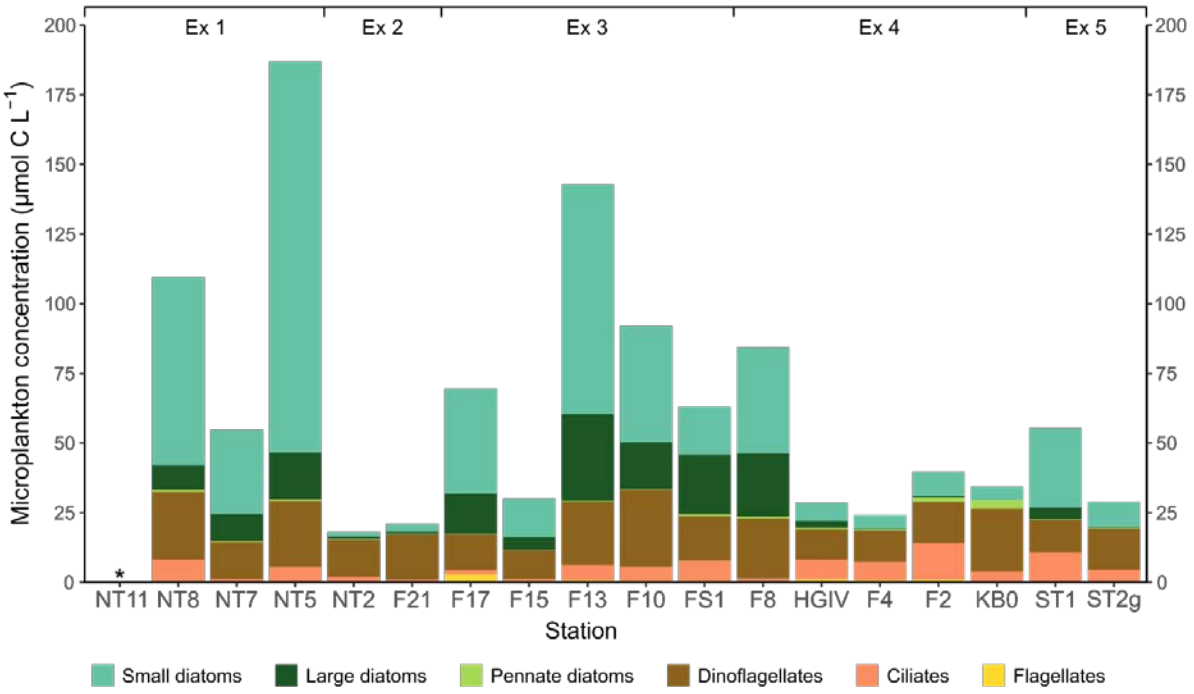


FIGURE 6 | The microplankton food environment for *Calanus finmarchicus* across the Fram Strait, calculated from inverted microscopy of water sampled at the chlorophyll maximum at each station. * indicates where the parameter was not measured.

Small chain-forming diatoms, and to a lesser extent large diatoms and dinoflagellates, were the dominant microplankton type at the majority of stations sampled (FIGURE 6). The stations close to Svalbard and the West Spitsbergen Current (HGIV, F4, F2, and KB0) had a lower proportion of both small and large diatoms. The westward stations (NT2, F21, F17, and F15) had a lower proportion of ciliates than those eastward (F13, F10, FS1, HGIV, F4, F2, with the exception of F8 and KB0). The small and large diatom peaks in the mid stations (F13, F10, FS1, F8) suggest a diatom bloom. The microplankton communities along the southerly transect (NT8, NT7 and NT5) also denote a bloom of small chain-forming diatoms. Flagellates were numerous, with colonies of *Phaeocystis* spp. found at many of the stations sampled, but they did not contribute significantly to the community biomass.

2.4.3 Elemental composition of *Calanus finmarchicus*

The average C and N contents of the experimental animals were variable (FIGURE 7; Supplementary Table 2). The average C content of *C. finmarchicus* females was 15.0 ± 2.9 µmol female^{-1}, ranging from 11.2 to 22.9 µmol female^{-1}, and the average N content was 2.2 ± 0.4 µmol female^{-1}, ranging from 1.5 to 3.0 µmol female^{-1}. The molar C:N ratio of the females averaged 6.8 ± 1.0 and ranged from 5.6 to 8.7. There was more variability in the ratio between individual animals than there was between average values at the start and the end of the experiments. The C and N content of the experimental females did not vary significantly between the different the five experiments ($F_{(4, 20)} \leq 2.318$, $p \geq 0.09$ in both cases) or change between the start and end of the incubations ($F_{(1, 20)} \leq 0.093$, $p \geq 0.76$) (Supplementary Table 2).

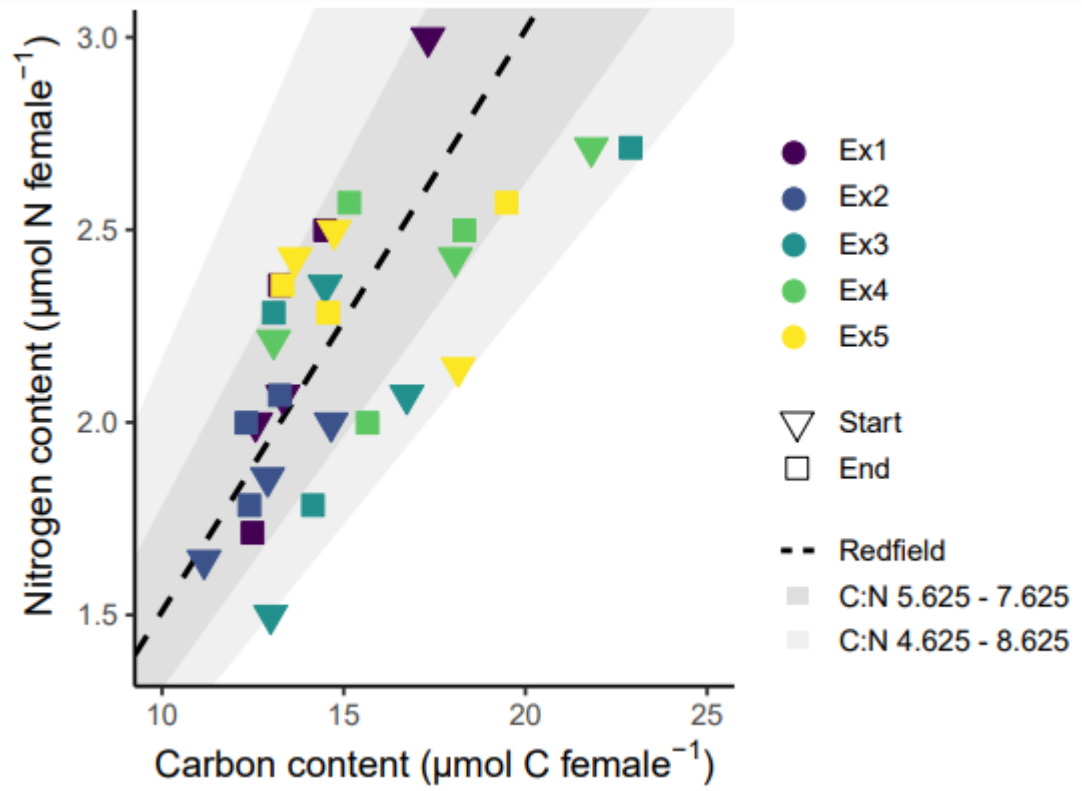


FIGURE 7 | The elemental content of female *Calanus finmarchicus* across experiments 1-5 (Ex1-5), before and after experimentation. Start indicates representative adults sampled at the beginning of each experiment, and end represents the composition of the same cohort of adults used in the experiments. 'Redfield' is the Redfield ratio (Redfield, 1958) found between carbon and nitrogen in phytoplankton (6.625 by atoms).

2.4.4 Ingestion and egg production

Total daily ingestion rates ranged from 0.3 to 12.3 µmol C female^{-1} day^{-1} and averaged 4.7 ± 3.6 µmol C female^{-1} day^{-1} (FIGURE 8; Supplementary Table 1). The station with the highest average ingestion was F10 (10.1 ± 0.3 µmol C female^{-1} day^{-1}) and the station with the lowest was KB0 (0.3 ± 0.1 µmol C female^{-1} day^{-1}). C-specific ingestion rates ranged from 0.015 ± 0.004 to 0.645 ± 0.017 day^{-1} and averaged 0.295 ± 0.223 day^{-1}.

Egg production in the grazing experiments ranged from 0.0 to 36.7 eggs female^{-1} day^{-1}, and averaged 8.9 ± 8.1 eggs female^{-1} day^{-1} across all experiments (FIGURE 8). This corresponds to C-specific egg production rates ranging from 0.00 to 0.049 day^{-1}, and an average of 0.012 ± 0.011 day^{-1}. Egg production rates did not correlate with female C content, N content, or the C:N ratio of the experimental animals (Supplementary Figure 2). At the stations where the experimental animals were collected, between 5-50% of the females were ≤GS3, and many of those within GS4 were developing relatively small clutches of eggs (= GS4B and GS4C) (TABLE 2).

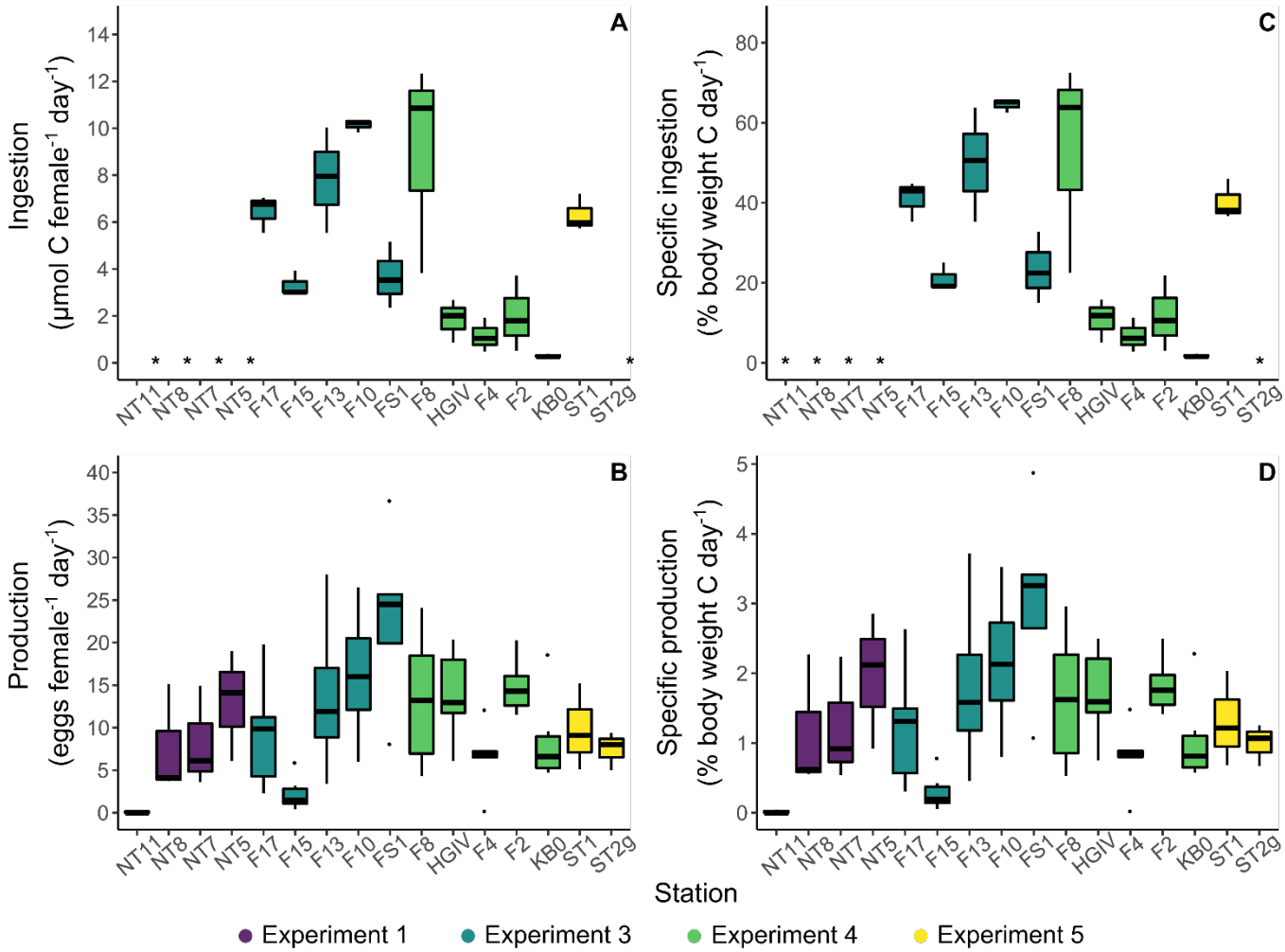


FIGURE 8 | Total ingestion (A), specific ingestion (B), total egg production (C), and specific egg production (D) by *Calanus finmarchicus* across the Fram Strait. The mid-line represents the median, the box the upper and lower quartiles, whiskers the range, and black points the outliers. Specific ingestion and production were converted from a fraction to a percentage for these figures. * indicates where the parameter was not measured.

Station	n	GS1	GS2	GS3	GS4C	GS4B	GS4A
F17	20	5	0	10	45	30	10
F15	20	0	0	35	25	25	15
F13	20	0	0	35	45	20	0
F10	20	0	0	20	25	40	15
FS1	20	0	0	25	30	45	0
F8	20	0	0	35	30	20	15
HGIV	10	0	10	40	40	10	0
F4	20	0	5	25	20	35	15
F2	10	0	0	30	40	30	0
KB0	20	0	0	5	35	40	20

TABLE 2 | Gonad maturation stages of female *C. finmarchicus*. N shows sample size. GS shows the percentage of females in gonad maturation stages 1-4a as per Niehoff & Runge 2003.

Prey was ingested in approximately similar proportions to that which was available. The proportions of all diatom types that were ingested correlated positively, albeit weakly, with the proportions available in the prey field (small diatoms: $\rho = 0.31$; large diatoms: $\rho = 0.52$; pennate diatoms: $\rho = 0.72$, $p < 0.05$ in all cases). The proportion of ciliates ingested also correlated with the proportion available ($\rho = 0.48$, $p = 0.004$). By contrast, there was a negative relationship between available and ingested dinoflagellates ($\rho = -0.56$, $p < 0.001$) and no significant correlation between the proportions of flagellates available and those ingested (FIGURE 9).

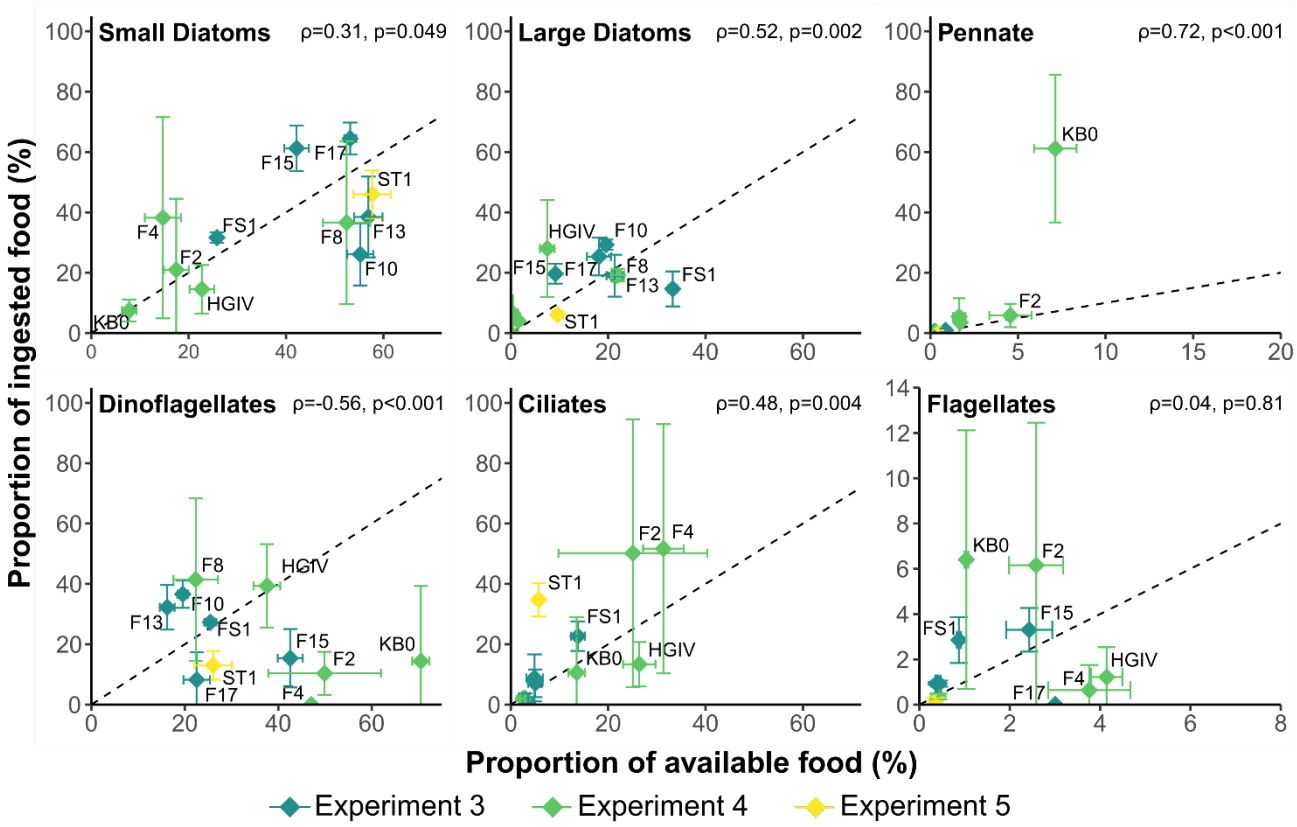


FIGURE 9 | The contribution of food types to the diet of *Calanus finmarchicus* against the contribution of the food types to the available food environment. The dotted line represents the 1:1 line where ingestion is proportional to available food, i.e. non-selective feeding. Above this, a greater proportion is ingested than is available, so the food is selected for, and below, the food is selected against. Note the variable scales on the x- and y-axes.

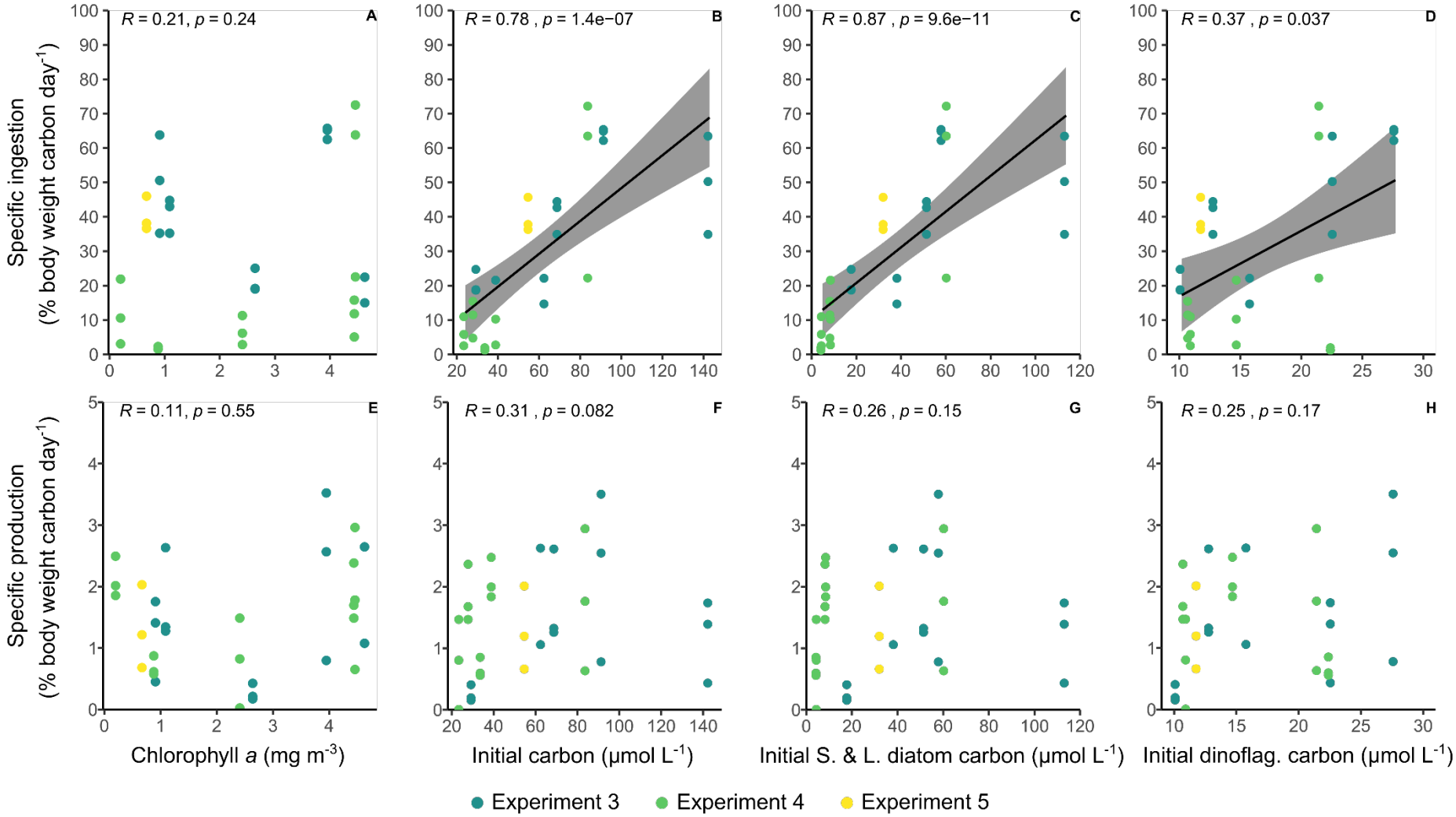

A
R = 0.21, p = 0.24
B
R = 0.78 , p = 1.4e−07
C
R = 0.87 , p = 9.6e−11
D
R = 0.37 , p = 0.037
E
R = 0.11, p = 0.55
F
R = 0.31 , p = 0.082
G
R = 0.26 , p = 0.15
H
R = 0.25 , p = 0.17
Specific ingestion (% body weight carbon day-1)
Specific production (% body weight carbon day-1)
Chlorophyll a (mg m-3)
Initial carbon (µmol L-1)
Initial S. & L. diatom carbon (µmol L-1)
Initial dinoflag. carbon (µmol L-1)
Experiment 3
Experiment 4
Experiment 5

FIGURE 10 | The relationships between the quantity of available food and ingestion (A, B, C & D) and egg production rate (E, F & G, H). The quantity of available food is estimated from using the chlorophyll a concentration as a proxy (A & E), and calculated using inverted microscopy for all cell types (B & F), for small (S) and large (L) diatoms (C & G), and for dinoflagellates (D & H). Shaded areas show 95 % confidence intervals. *R* is Spearman's rho.

The specific rate of ingestion increased significantly with the concentration of microplankton carbon (FIGURE 10B). This relationship appeared to be driven largely by the concentrations of small and large diatoms (FIGURE 10C), and was not reflected in the chlorophyll concentrations (FIGURE 10A).Specific egg production, in contrast to ingestion, showed no relationship with any measure of food availability (FIGURE 10E-H; all $p > 0.05$). Similar relationships were seen between total rates of ingestion and production and measures of food availability (Supplementary Figure 3). Specific rates of egg production were generally higher when specific ingestion rates were higher, but the correlation between these variables was not significant when examined across all stations ($\rho = 0.21$, $p > 0.2$; Supplementary Figure 4). There was a significant positive relationship between the specific egg production rate measured here and the and the proportion of females that were spawning (Cook et al., this issue; $\rho = 0.39$, $p = 0.027$; Supplementary Figure 5). There was no relationship between ingestion or egg production and environmental temperature before the experiment began ($r = -0.2$, $p > 0.1$ and $r = 0.2$, $p > 0.1$, respectively).

2.4.5 Carbon budgets

The daily metabolic C budgets for the experimental animals are shown in Table 3. The calculated gross growth efficiencies of the animals ranged from 0.01 to 0.39, averaging 0.12 ± 0.13 (TABLE 3). Ingestion was consistently higher than the total C needed for egg production, respiration, and egestion combined. In all but one station, *C. finmarchicus* had an excess of C. The surplus was on average 1.6 ± 1.6 µmol C individual^{-1} day^{-1} or 19.2 µg C individual^{-1} day^{-1}. As a proportion of intake, this corresponds to 0.17 ± 0.42 (TABLE 3), which is more than needed for egg production and only slightly less than used for respiration. Stations F8, F10 and F13 had the greatest amounts of excess, where the fractions of C intake (C individual^{-1} day^{-1}) were 0.40, 0.40 and 0.41, respectively. By contrast station KB0 had a C-deficit of 1.04 as a fraction of C intake individual^{-1} day^{-1}.

Increasing the AE to 0.74 had the greatest effect on the budget of all the non-measured terms, with higher efficiencies adding to the C surplus (TABLE 3). Changing the assumed metabolic substrate by adjusting the RQ had only a small effect on the surplus. Assuming complete lipid metabolism, i.e. RQ = 0.7 (Ikeda et al., 2000), the surplus would again be increased (TABLE 3).

Ex	Day	Station	Ingestion (I)	Production (E)	GGE	Production	Respiration	Carbon surplus (Ω)		
			(µmol C fem^{-1} day^{-1})	(µmol C fem^{-1} day^{-1})		(Proportion of C intake fem^{-1} day^{-1})	(Proportion of C intake fem^{-1} day^{-1})	(Proportion of C intake fem^{-1} day^{-1})		
3	1	F17	6.45	0.28	0.04	0.04	0.05	0.38	(0.39)	[0.65]
3	2	F15	3.31	0.04	0.01	0.01	0.09	0.36	(0.39)	[0.63]
3	3	F13	7.84	0.19	0.02	0.02	0.04	0.41	(0.42)	[0.68]
3	4	F10	10.14	0.36	0.04	0.04	0.03	0.40	(0.41)	[0.67]
3	5	FS1	2.94	0.29	0.09	0.10	0.10	0.27	(0.29)	[0.54]
3 (average)			6.14 ± 3.05	0.23 ± 0.12	0.04 ± 0.03	0.04 ± 0.03	0.06 ± 0.03	0.36 ± 0.06	(0.38 ± 0.05)	[0.63 ± 0.06]
4	1	F8	9.01	0.31	0.06	0.03	0.04	0.40	(0.41)	[0.67]
4	2	HGIV	1.85	0.32	0.20	0.17	0.18	0.11	(0.17)	[0.38]
4	3	F4	1.15	0.13	0.18	0.12	0.30	0.06	(0.14)	[0.33]
4	4	F2	2.01	0.36	0.31	0.18	0.17	0.12	(0.17)	[0.39]
4	5	KB0	0.30	0.12	0.39	0.39	1.12	-1.04	(-0.73)	[-0.77]
4 (average)			2.87 ± 3.5	0.25 ± 0.11	0.23 ± 0.13	0.18 ± 0.13	0.36 ± 0.44	-0.07 ± 0.56	(0.03 ± 0.44)	[0.20 ± 0.56]
5	1	ST1	6.30	0.20	0.03	0.03	0.05	0.38	(0.40)	[0.65]
All (average)			4.66 ± 3.4	0.24 ± 0.11	0.12 ± 0.13	0.1 ± 0.11	0.20 ± 0.32	0.17 ± 0.42	(0.22 ± 0.33)	[0.44 ± 0.42]

TABLE 3 | Daily metabolic carbon (C) budgets for *Calanus finmarchicus*, showing measured ingestion and production. As there was no somatic growth, production is assumed to equal egg production only (Poulet et al., 1995). Budgets are calculated as Ingestion (I) = Egg production I + Respiration I + Egestion (W) + C surplus (Ω). Ex = experiment, Fem = female, GGE = Gross Growth Efficiency, the ratio of biomass production to ingestion. Respiration was estimated using nitrogen biomass-specific equations (Ikeda et al., 2001) and a respiratory quotient (RQ) of 0.97. The

budgets were calculated assuming that egestion is I × (1- absorption efficiency), where absorption efficiency = 0.47 (Mayor et al., 2011) and therefore egestion is 0.53 as a proportion of C intake fem^{-1} day^{-1}. The C content of the animals was 15.72 µmol C ind^{-1} for Ex3, 17.01 µmol C ind^{-1} for Ex4, and 15.65 µmol C ind^{-1} for Ex5. The nitrogen (N) content of the animals was 2.12 µmol N ind^{-1} for Ex3, 2.40 µmol N ind^{-1} for Ex4, and 2.38 µmol N ind^{-1} for Ex5. The C surplus was also calculated using a respiratory quotient of 0.7 (shown in "( )") and with an absorption efficiency of 0.74 (shown in "[ ]"). The mean average is shown ± standard deviation.

2.5 Discussion

Our study quantified feeding and reproduction in female *Calanus finmarchicus* and examined how these varied in response to the food environment across the Fram Strait in May-June 2018. We show that ingestion rates typically increased as the total amount of microplankton food available increased. By contrast, egg production showed no obvious relationship with food availability. The incubated animals mostly displayed low gross growth efficiencies (GGE <0.20). This suggests that a large fraction of the ingested C was used for physiological processes other than egg production.

2.5.1 Ingestion

The stations sampled across the Fram Strait were typically characterised by elevated microplankton concentrations, and thus feeding conditions were almost always favourable. This was particularly evident in Experiments 1 (NT8, NT7, NT5) and 3 (F17, F15, F13, F10, FS1), where concentrations were > 50 µmol C L^{-1} on all but one day. Food concentrations > 42 µmol C L^{-1} have been found to be saturating for *C. finmarchicus* (Båmstedt, Nejstgaard & Solberg, 1999). At stations NT5 and F13 microplankton C concentrations were ≥ 142 µmol L^{-1}, and many of the other stations sampled (NT8, NT7, NT5, F17, F13, F10, FS1, F8, KB0, ST1) had food concentrations > 42 µmol C L^{-1} (500 µg C L^{-1}). Diatoms were abundant at the southerly stations sampled during Experiment 1 (NT8, NT7, NT5), and at the stations in Experiment 3 (F17, F15, F13, F10, FS1). In contrast, much lower diatom concentrations were encountered at the stations sampled during Experiment 2 (NT2, F21), and at several of those sampled during Experiment 4 (HGIV, F4, F2, and KB0).

C. finmarchicus typically consumed prey in proportion to their availability in the plankton (FIGURE 9). This pattern of intake is similar to what has been found in other areas, such as North Atlantic (Mayor et al., 2006; Castellani et al., 2008; Mayor et al., 2009a) and the English Channel (Djeghri et al., 2018), and supports the understanding that *C. finmarchicus* are less selective than other calanoid copepods (Teegarden et al., 2008). Diatoms dominated the diet of *C. finmarchicus* at most stations examined, as is often reported (Irigoien et al., 2002; Søreide et al., 2008; Cleary et al., 2017; Kohlbach et al., 2021). This intake simply reflects the predominance of diatoms in the microplankton, rather than these cells being positively selected for. There was some evidence for positive selection towards ciliates at stations F2, F4 and ST1, where ciliate biomass was high and diatom biomass, particularly that of large diatoms, was low. It seems that the copepods actively selected for ciliates at these stations in order to compensate for the reduction in diatom biomass, as has been observed previously (Mayor et al., 2006). By contrast, the proportion of dinoflagellates in the ingested ration

was negatively correlated with their availability in the microplankton (FIGURE 9) suggesting that there were increasingly selected against. A range of dinoflagellates are known to be capable of producing toxins that reduce food absorption, egg production rates and egg hatching success in *C. finmarchicus* (Roncalli et al., 2016) and the apparent avoidance of dinoflagellates may indicate the presence of one or more toxin-producing species. However, the absence of a negative relationship between ingested dinoflagellate C and egg production (FIGURE 10H) suggests that any potential negative effect of consuming dinoflagellates was insufficient to cause a noticeable effect.

Ingestion by *C. finmarchicus* was on average 4.7 ± 3.6 µmol C female^{-1} day^{-1}, well within the previously reported range for this species when feeding on natural plankton assemblages (0.04 – 7.33 µmol C female^{-1} day^{-1} (Jónasdóttir et al., 2008; Mayor et al., 2009a)). C-specific ingestion rates were generally close to 0.3 day^{-1}, which also fits well within the published range (Gamble, 1978; Ohman & Runge, 1994; Nejstgaard, Gismervik & Solberg, 1997), matching with similar ingestion rates found during bloom periods in the Norwegian Sea (Irigoien et al., 1998). The maximum value for C-specific ingestion, 0.645 ± 0.017 day^{-1}, was high, but again, consistent with the 0.80 day^{-1} previously reported for *C. finmarchicus* when feeding during periods of elevated food availability (Smith & Lane, 1988). Indeed, it is probable that the elevated ingestion rates reported herein were because of the high concentration of food available, as suggested by the strong positive correlations between ingestion and both the total concentration of microplankton and that of diatoms (FIGURE 10b and 10c); the weakly significant correlation between ingestion and dinoflagellate concentrations likely reflects the collinearity between dinoflagellate- and diatom C, rather than a causal relationship.

Interestingly, there was no relationship between chlorophyll *a* concentration, a common proxy for food availability, and ingestion (FIGURE 10a). This could be because the chlorophyll *a* data were just a snapshot of the sampling location and therefore not representative of the food available to the incubated copepods. This suggestion is supported by the lack of a relationship between available food and chlorophyll *a* (add details of correlation ($p > 0.05$). This disparity is also potentially attributable to the high abundances of picoplankton (≤2 µm cell diameter) that have been reported to occur in the Fram Strait, in particular the *Micromonas* genus of prasinophytes (Bachy et al., 2022), that were not enumerated in our study.

2.5.2 Egg production

The reproductive strategy of *C. finmarchicus* can vary in response to food supply, likely a necessity of its one-year life cycle (Falk-Petersen et al., 2009). Maximum egg production rates occur during the

spring bloom, but the timing of this differs between areas and is controlled by hydrography, light conditions and climate (Niehoff, 2004). Egg production is subject to prior gonad maturation and oocyte development.

The measured egg production rates were within the range 0.3 – 36.7 eggs female^{-1} day^{-1} previously observed for *C. finmarchicus* in the Arctic (Hirche, 1990; Hirche & Kosobokova, 2007; Møller et al., 2016). C-specific egg production rates were within the range of 0.00 to 0.049 day^{-1}, in good agreement with the published range for *C. finmarchicus* (Hirche, Meyer & Niehoff, 1997; Mayor et al., 2006; Møller et al., 2016; Jónasdóttir et al., 2022). The observed rates did not correlate with any measure of ingested food quantity or prey type. This either suggests that the link between recent feeding history and egg production rate is weak, or that the rate at which eggs are produced is not directly limited by the available food. However, many studies have found a strong link between food quantity and egg production rate (Marshall & Orr, 1958; Hirche, 1990; Harris et al., 2000a; Ohman & Runge, 1994; Hirche, Meyer & Niehoff, 1997; Hirst & Bunker, 2003; Jónasdóttir et al., 2022). Two of these studies were conducted in the Arctic, but the animals were either kept under identical feeding conditions for some time before experimentation (Hirche, Meyer & Niehoff, 1997), or their eggs were only counted after 48 hours (Hirche, 1990), rather than 24 hours used herein, to allow for a longer spawning interval. A time 'lag' of > 24 hours between ingestion and the production of eggs could potentially explain the absence of a relationships between egg production rates and both food availability and ingestion rates; *C. finmarchicus* has previously been observed to display a spawning interval of > 24 hours in the Arctic at 0°C (Hirche, 1990). However, egg production did not correlate with the amount of food ingested during the preceding day ($p \geq 0.88$ in all cases). Furthermore, independent egg production experiments conducted in parallel to those presented herein revealed that, on average, 66 % (ranging from 0-94 %) of the 20 individual females incubated at each station produced eggs within the first 24 hours (Cook et al., submitted), indicating that the spawning interval was generally < 24 hours throughout the period of our study.

The disconnect between ingestion and egg production seen in our experiments could, alternatively, indicate that the animals were using maternal reserves, rather than, or in addition to, the ingested food to produce eggs. *C. finmarchicus* has previously been observed to adopt a capital breeding reproductive mode in the North Atlantic when feeding conditions are poor, losing significant quantities of maternal biomass C and N to fuel continued egg production (Niehoff, 2004; Mayor et al., 2006, 2009a). The C and N contents of the experimental animals in the present study did not, however, change significantly throughout the experiments (FIGURE 7, Supplementary Figure 2), indicating that they were not using biomass reserves to fuel reproduction. Indeed, the metabolic

budgets show that the copepods ingested C in excess of that required for egg production and other physiological processes (discussed below). Regardless of the underlying mechanism, the lack of 38a relationship between the observed rates of ingestion and reproductive output indicates that accurately predicting how *C. finmarchicus* will respond to projected changes in their food environment is complex and likely requires information beyond simple metrics of food concentration.

2.5.3 Carbon budgets

Carbon budgets were constructed for each experiment by combining measured rates of C intake and egg production rates with empirically-estimated rates of respiration and faecal pellet production. In all but one instance (Experiment 4, day 5; TABLE 3), C intake could not be fully accounted for by respiration and the production of eggs and faecal matter. Indeed, across all experiments, the average fraction of ingested C that was in excess to requirements was 0.17 ± 0.42.

The fractions of intake allocated to respiration and egestion were approximately 0.20 and 0.531, respectively. Changing the respiratory quotient towards lipid-fuelled metabolism (RQ = 0.7) only reduced the estimated C required for respiration by a small amount, causing the apparent excess of C to increase slightly (TABLE 3). The chosen value of AE, 0.47, was selected because it relates specifically to *C. finmarchicus* feeding on diatoms (Mayor et al., 2011), the main prey item in our experiments. Increasing AE to 0.74, which is commonly assumed when modelling marine copepods (Anderson et al., 2017, 2020; Anderson, Hessen & Mayor, 2021), decreases the fraction of ingested C released as faecal matter to 0.26, further increasing the excess of C in the metabolic budget. We therefore suggest that our estimated C excesses are conservative.

Calculated GGEs were low, usually well below the expected range of 0.2 – 0.3 observed for copepods (Straile, 1997), meaning that egg production accounted for a relatively small fraction of the consumed food. Cannibalism of eggs could potentially explain the relatively low GGEs (Bonnet, Titelman & Harris, 2004) and the C surpluses. However, previous work using the same experimental design as used here concluded that the effects of cannibalism in these experiments were negligible (Mayor et al., 2006, 2009a). Furthermore, if the apparent C excesses were simply caused by egg cannibalism, the actual EPRs would have been 89.1 ± 76.8 (with a maximum EPR of 212.8) – towards or beyond the upper end of field-reported values (Hirche, 1990; Hirche, Meyer & Niehoff, 1997; Niehoff et al., 1999; Richardson et al., 1999; Swalethorp et al., 2011; Møller et al., 2016), and well in excess of the rates determined in parallel experiments with individual females that were excluded

from their eggs to prevent cannibalism (24.1 ± 14.4 eggs female^{-1} day^{-1}; Cook et al., submitted). This suggests that egg cannibalism cannot explain the apparent C excesses.

Like many polar copepods, all of those in the genus *Calanus* are well-known for their ability to produce and store lipids (Lee, Hagen & Kattner, 2006b). It is therefore possible that the excess C reflects lipid biosynthesis and accumulation by these animals. However, this seems unlikely, given that a) the experiments were conducted in May at the very start of the growth season, b) *C. finmarchicus* typically undertake a 1-year life cycle (Falk-Petersen et al., 2009), and thus females are not thought to re-enter diapause, and, most importantly, c) the C content of the experimental animals did not increase significantly over the course of the experiments (FIGURE 7; Supplementary Figure 2). We therefore propose that the observed metabolic C surpluses may, at least in part, be explained by the significant energetic costs associated with gonad maturation. In *C. finmarchicus*, this process generally starts in stage V copepodites (CVs) several months before the animals emerge from their over-wintering period and progresses in newly moulted females during the following spring (Tande, 1982; Hirche, 1996b; Jónasdóttir, 1999; Niehoff et al., 2002; Niehoff, 2007). The rates at which newly moulted females produce eggs increases from zero to maximal rates over 15 days at 5°C (Rey et al., 1999), and likely takes longer at colder temperatures (Melle & Skjoldal, 1998). The observed gonad maturation stages (TABLE 2) further supports the suggestion that a proportion of the experimental females were still in the process of developing their ovaries.

Gonad maturation in *C. finmarchicus* is known to require large amounts of energy, ~ 5.8 µmol C individual^{-1} (Rey-Rassat et al., 2002). At times these animals are able to provide the resources for gonad maturation and/or egg production from their own biomass (Irigoien et al., 1998; Niehoff, 2004; Mayor et al., 2006, 2009a). Females that have just undergone gonad maturation therefore often exhibit depleted lipid reserves (Sargent & Falk-Petersen, 1988; Rey-Rassat et al., 2002; Anderson et al., 2022) with biomass C:N ratios declining as low as ~5 by atoms when spawning begins (Tande, 1982; Mayor et al., 2009b). However, the lack of a clear decline in biomass C and N content over the duration of our experiments (FIGURE 7) suggests that our experimental animals were not meeting the costs of maturation from internal reserves, and were instead acquiring them via ingestion. There are multiple observations of recently moulted females needing to feed prior to completing maturation and commencing egg production. For example, in the lower St. Lawrence Estuary, the final stages of oocyte maturation in *C. finmarchicus* females does not begin until feeding conditions become favourable in June (Plourde & Runge, 1993), and in the Norwegian Sea, <50 % of female *C. finmarchicus* are mature during the pre-bloom period (March through April), after which the population undergoes rapid maturation as the bloom develops through May (Niehoff et al.,

1999). Indeed, recent work suggests that the final step in terminating diapause in *C. finmarchicus* may also be dependent upon the presence of food (Hatlebakk et al., 2022).

The average C:N ratio of our experimental females was 6.8 (ranging between 5.6 – 8.7) by atoms, which is consistent with the understanding that many of them were likely still in the process of reaching maturation (Supplementary Figure 2). Indeed, only 66 % of the females incubated in parallel egg production experiments produced eggs (Cook et al., submitted), suggesting that the remaining third of the population were still undergoing gonad maturation. In addition to explaining the fate of the excess C, the process of gonad maturation occurring in some, but not all of our experimental females would also explain why the observed egg production rates were not correlated with ingestion (Section 4.3), and why the proportion of females that were spawning was positively correlated with egg production rate (Supplementary Figure 5; Cook et al., submitted). In turn, this suggests that our ability to estimate egg production rates in *C. finmarchicus* and many other high-latitude copepods may be improved by considering their level of maturity, whether by means of morphological investigation or by the development of a metabolic proxy for the level of gonad maturation.

Our budgets focused on C, but that is not to say that the animals were necessarily requiring this element only. Indeed, the experimental animals contained visible quantities of lipid (Supplementary Figure 6), confirmed by their average biomass C:N (6.8 by atoms), which was well above that of an actively spawning female and suggests that they still had C available. Producing mature ovaries and the resulting eggs from stored lipids only, which are largely devoid of N, seems unlikely, particularly as the C:N ratio of *C. finmarchicus* eggs ranges between 4-7 by atoms (Ohman & Runge, 1994; Runge & Plourde, 1996; Mayor et al., 2009b; Swalethorp et al., 2011) and hence contain a substantial amount of N. We therefore suggest that, in addition to helping meet the energetic costs of maturation, the apparently excessive rates of ingestion prior to reproduction were also required to provide the animals with the amino acids and proteins required to finish producing and maturing their ovaries. We still know relatively little about N-based physiology in *C. finmarchicus* and if, how, or where they are able to store compounds that bear this element (Mayor et al., 2022). This lack of fundamental understanding hinders our ability to mechanistically represent important aspects of their life histories in ecosystem- and biogeochemical models and predict how they will change in the future (Anderson et al., 2022).

2.6 Conclusion

We have shown that female *C. finmarchicus* are able to take advantage of the abundant feeding conditions encountered during May in the Fram Strait, in part due to their flexible and diverse diet. Egg production did not correlate with food availability or ingestion. Metabolic budgets for our experimental females showed that the ingested food was typically more than that required to produce the observed numbers of eggs and estimated rates of respiration and faecal pellet production. The generally low egg production rates and the relatively high biomass C:N values suggest that a sizeable fraction of the incubated females were reproductively immature, and were using the excess food to meet the energetically-expensive process of gonad maturation and as a source of N-bearing compounds that are required to produce ovary tissues and eggs. This suggestion is supported by the observed gonad maturation status of the sampled female populations. Our study highlights the need to consider ontogenetic development when examining the relationship between ingestion and production in copepods. Developing mechanistic models to reliably predict how the ecological and biogeochemical roles of *C. finmarchicus* and other high-latitude copepods will respond to climate-driven changes in their food environment requires improved understanding of both the C- and N-based physiologies of these important animals, particularly during the gonad maturation phase.

Chapter 3 Post-bloom grazing, egg production and carbon budgets for Calanus finmarchicus in the Fram Strait

This paper is in preparation for submission to the Journal Plankton Research

Jenkins, H.E., Cook, K.B., Anderson, T.R., Atherden, F., Jacob, E., Thornton, B. & Mayor, D.J. (in prep.) Post-bloom grazing, egg production and carbon budgets for *Calanus finmarchicus* in the Fram Strait.

HJ and DM contributed to the conception and design of the study. HJ conducted experiments, microscopy analysis, budget analysis and statistical analysis. BT performed the elemental analyses. EJ analysed gonad maturity. HJ wrote the drafts of the manuscript and created the figures. DJM revised the draft. All authors read and approved this draft with some final changes to make in response to feedback from TA.

3.1 Abstract

Calanoid copepods typically dominate Arctic zooplankton communities and are vital to the ecological and biogeochemical functioning of this region. Rapid climate change at high latitudes is altering the quantity and composition of the food available to copepods, potentially shifting towards flagellate-dominated communities and reduced levels of primary production by the end of the 21st century. To better understand how these changes may affect the future growth and reproductive success of copepods we quantified rates of feeding and egg production in females of a key species of calanoid copepod, *Calanus finmarchicus,* at stations across the Fram Strait under post-bloom conditions in August where the microplankton biomass was dominated by flagellates and ciliates. Carbon-specific rates of ingestion and egg production were both low, averaging 0.0040 ± 0.0048 day^{-1} (mean ± SD) and 0.0024 ± 0.0034 day^{-1}, respectively, indicated that feeding conditions were poor. A positive linear relationship between ingestion and food availability confirmed that the animals were food limited throughout the study region. The resulting unrealistically high GGE (0.64 ± 1.12) showed metabolic requirements were higher than ingestion. As such, egg production was not related to food availability or ingestion, indicating that another C source was fuelling production. This suggests that the copepods were drawing upon internal biomass reserves to sustain their reproductive output and/or consuming the eggs of conspecifics. Estimated metabolic requirements of the experimental females exceeded the measured ingestion rates at all of the stations investigated, with a mean carbon deficit of 0.37 ± 0.06 µmol copepod^{-1} day^{-1}. This further supports that the idea that the animals were food limited at the time of sampling and suggests that the projected decline in primary production and a shift towards increased dominance of flagellates in the Arctic may have 42negative consequences for the success of copepods like *C. finmarchicus* that typically feed on larger cells.

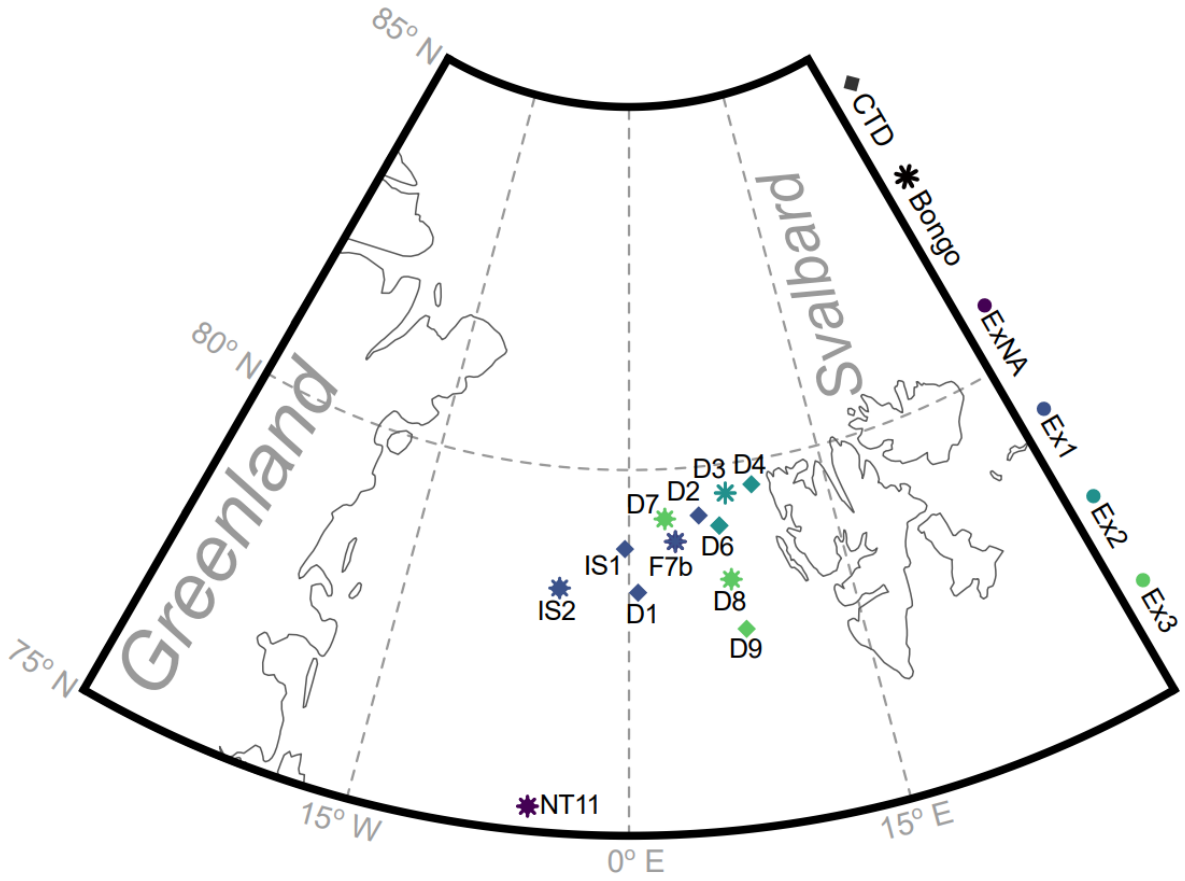


FIGURE 11 | The locations of sampling stations in August 2019. Sequential long-term grazing experiments (Ex1-Ex3) are defined by colour, in addition to station NT11 which fell outside the grazing experiments. Bongo and CTD show the locations where animals for experimental incubations and the natural plankton assemblage were collected, respectively. Sampling station D6 marks D6, D6b and D6c.

3.2 Introduction

Copepods of the genus *Calanus* dominate zooplankton biomass in the Arctic (Smith & Schnack-Schiel, 1990; Ashjian et al., 2003; Falk-Petersen et al., 2009). They occupy an important position in the Arctic food web, converting their microplankton prey into high-quality biomass for higher trophic levels such as herring, capelin, juvenile cod (Gatten & Sargent, 1973; Sakshaug, 2004) and even baleen whales (Moore et al., 2019). Copepods play an equally important role in the biogeochemical cycling of both macro- and micro- nutrients.

Human activity is causing the Arctic Ocean to change at a much greater pace than at lower latitudes (Dai et al., 2019; Thomas et al., 2022). Poleward atmospheric moisture transport (Hofsteenge et al., 2022) and enhanced oceanic heat transport (Tsubouchi et al., 2021) have reduced sea ice area and

thickness. Seasonal changes to sea ice are linked inextricably to the timing, composition, distribution and magnitude of the phytoplankton blooms on which *Calanus* spp. rely. Primary production in the Arctic Ocean increased by 57% between 1998 and 2018 (Lewis, Van Dijken & Arrigo, 2020) owing to the longer growing season and an expanded area of open water. However, continued warming and the associated stratification in the Arctic is expected to restrict the supply of nutrients to surface waters and thereby limit future phytoplankton productivity (Tremblay & Gagnon, 2009; Farmer et al., 2021; Noh et al., 2023). Smaller phytoplankton are expected to thrive in the new, warmer conditions as they outcompete larger phytoplankton (Li et al., 2009), but may not be as nutritionally beneficial as the larger diatoms that are typically associated with contemporary Arctic spring blooms (Søreide et al., 2010).

The current phenology of Arctic phytoplankton is coupled to the timing of the sea-ice retreat, with the majority of growth beginning in the marginal ice zone in spring when the ice retreats, solar elevation increases, and meltwater strengthens haline-based stratification (Kahru et al., 2011; Perrette et al., 2011). This leads to an annual bloom of phytoplankton dominated by centric and pennate diatoms, followed by, in areas that are influenced by Atlantic waters and therefore weaker stratification, a secondary bloom of smaller, motile microplankton dominated by flagellates (Oziel et al. 2020;). The succession between the blooms is driven by availability of N and timing varies across the Arctic. Flagellates may rely on mixotrophy (e.g. osmotrophic or phagotrophic processes) which allow them to outcompete phototrophic phytoplankton (Li et al., 2009). The size spectra of the plankton community is important to plankton dynamics on both population and community levels (Falkowski, 1998) and determines the mass transfer to higher trophic levels (Thingstad & Cuevas, 2010).In the last two decades, the reduction of sea ice cover in the Arctic has allowed populations of the traditionally North Atlantic and sub-Arctic *Calanus finmarchicus* to dominate over the 'true' Arctic species, *C. glacialis* and *C. hyperboreus* (Møller & Nielsen, 2020). Now, *C. finmarchicus* is reproductively viable in parts of the Arctic (Tarling et al., 2022b). These changes have largely been attributed to Arctic warming and shifting thermal niches, although the underlying mechanisms remain unclear (Aarflot et al., 2018). *C. finmarchicus* females are thought to optimise their reproductive success by timing their spawning activity to coincide with good feeding conditions in spring, allowing their progeny to rapidly develop and sequester sufficient lipid reserves to enable diapause towards the end of summer (Irigoien, 2004; Anderson et al., 2022). Egg production in *C. finmarchicus* typically increases with food availability (Runge, 1984a; Hirche & Bohrer, 1987; Hirst & Bunker, 2003; Mayor et al., 2009b), but can also occur at low rates even in the absence of food throughout much of the year (e.g. Hirche, Meyer & Niehoff, 1997; Niehoff, 2004, 2007). Indeed, their reproductive effort is not completely reliant upon the immediate food environment; the energy-

intensive process of gonad maturation is thought to be fuelled by lipids sequestered as juvenile copepodites during the preceding spring (Rey-Rassat et al., 2002; Jenkins et al., 2022) and egg production may be supported through the use of maternal biomass when food concentrations are low (Niehoff, 2004; Mayor et al., 2006, 2009a). Exactly how the changing food environment is affecting the ability of *C. finmarchicus* to reproduce in the Arctic remains poorly understood.

The Fram Strait is the only deep gateway to the central Arctic Ocean, characterised by warm, saline Atlantic Water in the Eastern Fram Strait being transported north and forming the largest oceanic heat transport into the Arctic Basin (Basedow et al., 2018). Concurrently cold, fresher Arctic Water is transported Southwards in the west. In Autumn, the East Greenland Polar Front separates these two regimes (Paquette et al., 1985), while eddy driven circulation of Atlantic Water modifies the movement of heat north (Wekerle et al., 2017). Currently, phytoplankton in the Atlantic water are larger than those in Arctic Water (Trudnowska et al., 2016) and the initial Arctic Water bloom is weeks earlier than the Atlantic bloom (Lampe, Nöthig & Schartau, 2021), likely due to the under-ice blooms (Arrigo et al., 2012; Mayot et al., 2018). There has been a long-term gradual increase in phytoplankton biomass reported in the Atlantic Water of the Fram Strait (Nöthig et al., 2015) but not the Arctic Water (Nöthig et al., 2020). In 2005 – 2007 there was a shift in the microplankton structure of the Atlantic water towards smaller flagellates and nanoflagellates, linked to a warm water anomaly (Nöthig et al., 2015). The differing conditions between the two regimes mean that in July, there can simultaneously be a pre-bloom community in the ice-covered western regime and a post-bloom community in the ice-free eastern regime (Fadeev et al., 2018). By August, nutrients often become limiting in both regimes and smaller cells dominate (Lampe, Nöthig & Schartau, 2021). Coupling the high variability with the effects of global heating, *Calanus* need plasticity to cope with this environmental variability.

Our aim here was to determine the metabolic carbon (C) budgets for *C. finmarchicus* across the Fram Strait in August 2019, in a comparable manner to our previous work in May/June 2018 (Jenkins et al., 2022), to examine how they respond to conditions that may be analogous to those that become prevalent in the future Arctic. We achieved this by conducting experiments to simultaneously determine rates of ingestion, prey selection, and egg production at stations in both Atlantic- and Arctic-dominated waters within the Fram Strait.

3.3 Methods

3.3.1 Experimental procedure

Ingestion and egg production rates (EPR) of female *Calanus finmarchicus* were measured simultaneously at 11 stations across the Fram Strait in August 2019 (FIGURE 11; TABLE 4; RRS James Clark Ross cruise JR18007) using a series of sequential 24-hour particle removal experiments (Mayor et al., 2009a). Ingestion rates (µmol C copepod^{-1} day^{-1}) were calculated using the established equations of Frost (1972) and converted to biomass specific rates using the mean C content of females and the mean ingestion rate of the experimental mesocosm. Further details of the procedures are described elsewhere (Jenkins et al., 2022). In brief, experimental animals were collected using a motion-compensated bongo net (200 µm) hauled vertically from 200 m and water containing the natural microplankton community was collected from the chlorophyll maximum via 20 L Niskin bottles. All experimental work was conducted in a temperature-controlled room at 3.1 ± 2.0 °C. Experiments were planned to last for five consecutive days to allow for the measurement of biomass change over the course of the experiments, however some were shorter: an experiment could not be started at the first station (NT11) because there were no mature female *C. finmarchicus* present, and experiment 3 was curtailed due to changes in the cruise plan. The carbon (C) and nitrogen (N) contents of the experimental animals were determined on individuals collected immediately before and after each experiment. Copepod species identities were verified via molecular analysis of animals collected at each station (Cook et al., Submitted). The gonad maturation stage (GS) of ≥10 females from each station was determined following established procedures, as the proportion of GS4 (i.e. most mature) *C. finmarchicus* females in a preserved sample as an index of the proportion of spawning females in a population (detailed in Appendix B Supplementary methodology) (Niehoff & Runge, 2003).

3.3.2 Microplankton analysis

Lugols-preserved microplankton samples (200 mL) from the grazing experiments were analysed using a FlowCam® 8400 (Yokogawa Fluid Imaging Technologies LLC) to enumerate and identify the particles (Poulton, 2016). This semi-automated technique combines digital imaging, flow cytometry, and microscopy. The FlowCam was focused using non-fluorescent latex beads. Prior to analysis, microplankton samples were concentrated by a factor of 3-4 via settling for 24 hours followed by gently siphoning the supernatant until 50mL of the sample remained. Samples were then gently agitated for one minute before a 5mL subsample was inserted into the FlowCam. Details of the

FlowCam technical specification, including setup, image sorting and measurement outputs are presented in Supplementary Table 4.

A classification algorithm within the programme Visual Spreadsheet v4.3.55 separated the images into 26 categories (classes). The algorithm was trained by creating image libraries for every class, each consisting of ≥1000 images from across all samples. Manual verification was needed for approximately 60% of images to confirm particle identity and remove non-target particles. Classes were combined or eliminated to form the six presented here, where small and large diatom categories refer to centric cells with diameters <20 µm (e.g. *Chaetoceros* spp.) and ≥20 µm (e.g. *Thalassiosira* spp.) respectively. Cell volumes were calculated using area-based diameter and then converted to carbon biomass using published conversion factors specific to the cell type (Menden-Deuer & Lessard, 2000; Malzahn & Boersma, 2012; Hillebrand et al., 1999).

3.3.3 Metabolic carbon budgets

Metabolic carbon budgets were assembled using daily ingestion rates (µmol C copepod^{-1} day^{-1}) and daily EPR (µmol C copepod^{-1} day^{-1}) which was converted from number of eggs to C units assuming 20.9 nmol C egg^{-1} (Mayor et al., 2009a). Egestion rates were estimated by assuming that C is assimilated with 74% efficiency (Anderson et al., 2017, 2020; Anderson, Hessen & Mayor, 2021). Oxygen consumption rates were calculated using the laboratory temperature and body N contents (Ikeda et al., 2001) and converted into C units by assuming a respiratory quotient (RQ) of 0.97. The sensitivity of the budget to the assumed RQ value was examined by changing this to 0.7 (Mayzaud, 1976).

3.3.4 Statistical and computational analysis

Biomass change over the experiments was examined by comparing the C and N contents of the copepods before and after experimentation. Prey preference was examined by comparing the abundance of a cell type in the food environment relative to the abundance of that cell type ingested. Parametric statistical tests were used for these comparisons (T-test, ANOVA and Pearson's), unless assumptions of normality, linearity and variance homogeneity were not maintained, in which case non-parametric tests were used (Spearman's rho, ρ). Results were considered significant below the 0.05 level. Mean averages shown followed by standard deviation. Statistical analysis and data visualisation were done using the R programming environment (R Core Team, 2022) using the packages ggplot2 (Wickham, 2016), purrr (Henry & Wickham, 2020), ggpubr (Kassambara, 2020), and viridis (Garnier, 2018).

Ex	Day	Station	Latitude (°N)	Longitude (°E)	Date	Temp (°C)	Depth (m)	Data
NA	1	NT11	75.336	-5.464	08/08/2019	4.4	40	M
Ex1	1	F7b	79.000	3.334	12/08/2019	7.5	18	M I E B G
Ex1	2	IS1	78.915	-0.288	14/08/2019	6.0	24	M I E G
Ex1	3	IS2	78.364	-4.644	15/08/2019	2.5	31	M I E G
Ex1	4	D1	78.317	0.616	16/08/2019	6.0	20	M I E G
Ex1	5	D2	79.333	5.167	17/08/2019	6.7	15	M I E B G
Ex2	1	D3	79.600	7.333	18/08/2019	7.0	10	M I E B G
Ex2	2	D4	79.667	9.400	19/08/2019	6.1	14	M I E G
Ex2	3	D6	79.167	6.600	20/08/2019	6.6	20	M I E G
Ex2	4	D6b	79.167	6.600	21/08/2019	6.7	6*	M I E
Ex2	5	D6c	79.167	6.600	22/08/2019	7.0	20	M I E B
Ex3	1	D7	79.317	2.649	23/08/2019	6.0	9	M I E B G
Ex3	2	D8	78.417	7.000	24/08/2019	6.5	15	M G
Ex3	3	D9	77.717	7.583	25/08/2019	7.4	27	M B G

TABLE 4 | Locations of the stations sampled on research cruise JR18007 in August 2019. Temp. is the water column temperature at the chlorophyll-*a* maximum. Depth is the depth of water sampled using the CTD, chosen to be at the chlorophyll maximum. Data shows what was measured at that station: M = microplankton analysis; I = *C. finmarchicus* ingestion; P = *C. finmarchicus* egg production; B = *C. finmarchicus* biomass; G = *C. finmarchicus* gonad maturation stage. For *C. finmarchicus* abundance and body condition, see Tarling et al., 2022a. Measurements are associated with the experiment start time throughout. * denotes underway water sampling due to CTD sampling being logistically impossible.

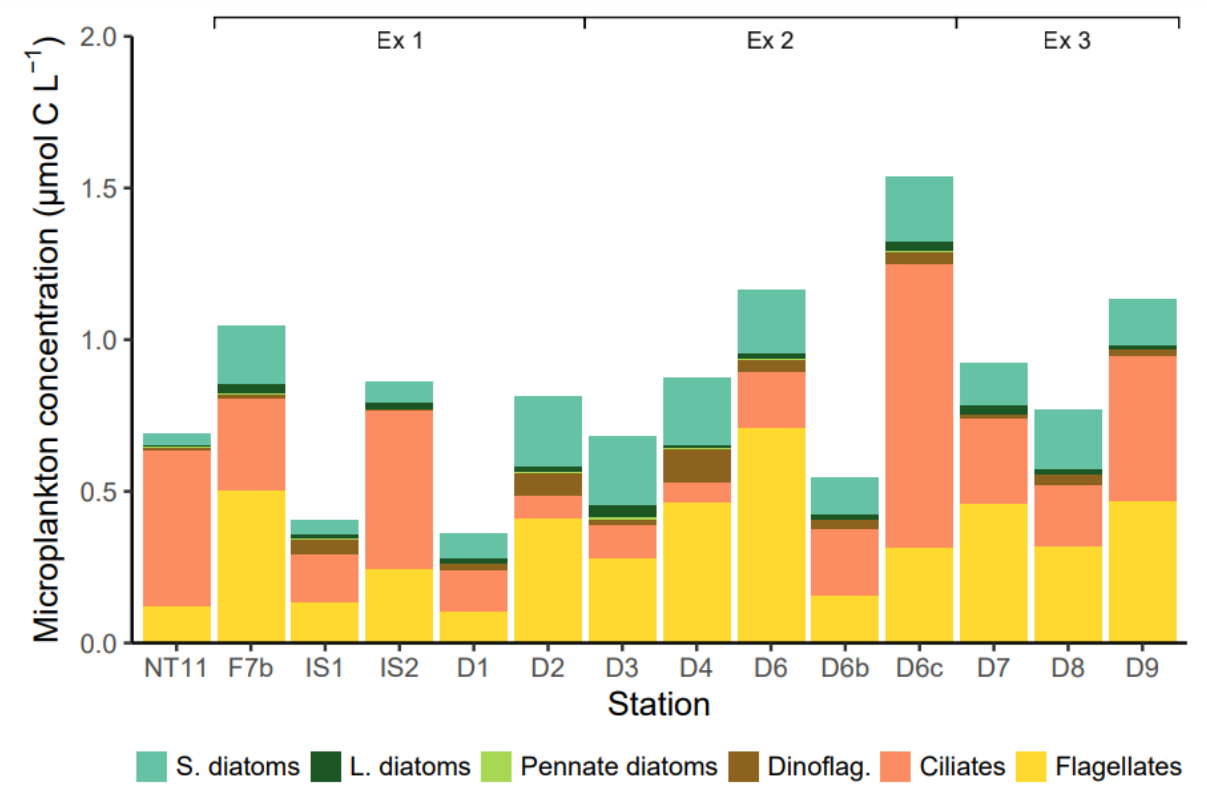


FIGURE 12 | The microplankton food environment available to *Calanus finmarchicus* across the Fram Strait in 2019, from water sampled at the chlorophyll maximum. S. diatoms refers to small centric diatoms (<20 µm); L. diatoms refers to large centric diatoms (≥20 µm); Dinoflag. Refers to dinoflagellates. Experiments 1-3 (Ex 1-3) are marked at the top.

3.4 Results

3.4.1 Microplankton

Microplankton biomass remained low across the sampled stations (FIGURE 12). Total biomass ranged from 0.36 to 1.54 µmol C L^{-1}, averaging 0.84 ± 0.33 µmol C L^{-1}. Biomass was highest at stations D6c and D6, with 1.54 and 1.17 µmol C L^{-1} respectively. These stations were located close to the ones with the lowest biomass, D1 and IS1, where microplankton biomass was 0.36 and 0.41 µmol C L^{-1}, respectively. Microplankton biomass also varied in time, with values at the same location (D6, D6b and D6c) ranging between 1.54 and 0.55 µmol C L^{-1}.

Ciliates and flagellates were the dominant microplankton types at most of the sampled stations (FIGURE 12), whereas large centric- and pennate diatoms were scarce throughout. Small centric diatom biomass was generally low but increased at the stations near the north of Svalbard, reaching 0.23 µmol C L^{-1} at D2 and D3 as opposed to 0.04 µmol C L^{-1} at NT11 and 0.05 and 0.07 µmol C L^{-1} at

the ice stations IS1 and IS2, respectively. Flagellate biomass remained high, ranging from an average of 0.10 µmol C L^{-1} at D1 to 0.71 µmol C L^{-1} at D6, as did ciliate biomass, which ranged from an average of 0.07 µmol C L^{-1} at D4 to 0.93 µmol C L^{-1} at D6c (Supplementary Table 5).

3.4.2 Elemental composition of female *Calanus finmarchicus* (CVI)

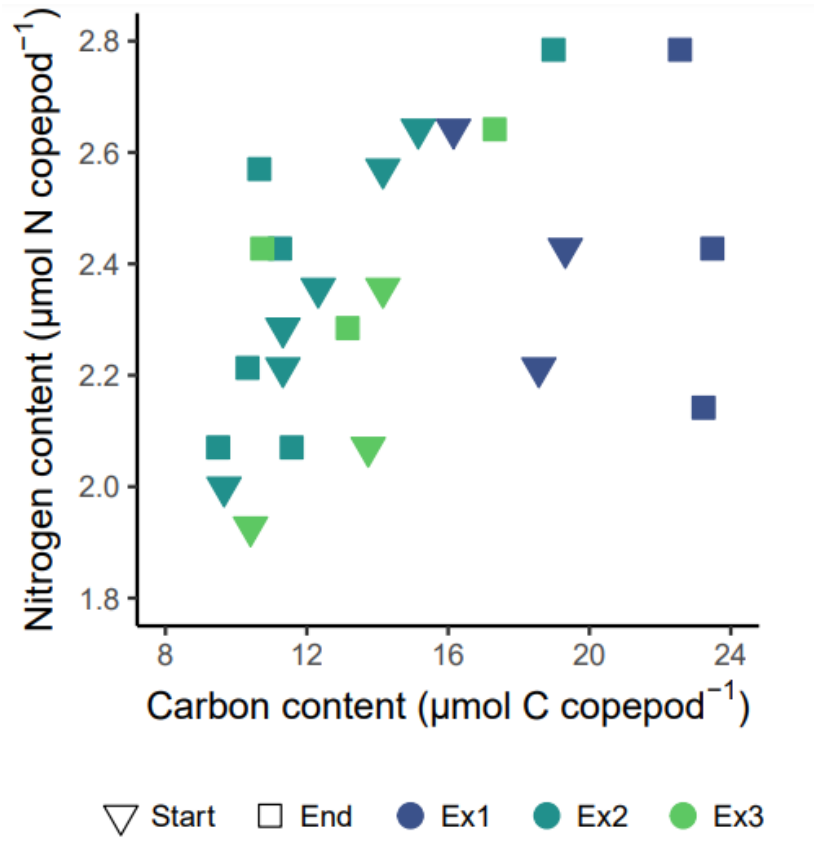


FIGURE 13 | The elemental content of female *Calanus finmarchicus* (CVI) across experiments 1-3 (Ex1-3) in the Fram Strait in summer 2019, before and after experimentation. Start indicates representative adults sampled at the beginning of each experiment, and end indicates the composition of the same cohort of adults used in the experiments.

The mean C content of the experimental female *C. finmarchicus* was 14.5 ± 4.4 µmol C copepod^{-1}, ranging from 9.5 to 23.5 µmol C copepod^{-1}, and the mean N content was 2.4 ± 0.2 µmol N copepod^{-1}, ranging from 1.9 to 2.8 µmol N copepod^{-1} (FIGURE 13; Supplementary Table 6). Biomass concentrations of C and N did not differ between the start and the end of the experiments ($F_{(1,18)} \leq 1.74$, $p \geq 0.37$ in both cases; Supplementary Figure 2), nor was there a significant interaction between the effects of time and station ($F_{(2,18)} \leq 2.23$, $p \geq 0.14$). However, the C contents of the females did vary significantly between the three experiments ($F_{(2,21)} = 19.53$, $p < 0.01$), with higher C

content in experiment 1, although the N contents did not ($F_{(2,21)}$ = 0.58, p ≥ 0.05). The C:N ratio varied from 3.6 – 9.3, averaging 5.3. There were no significant differences between then ratios at the start and end of the experiments (p > 0.05 in all cases).

3.4.3 Ingestion and egg production

Total daily ingestion rates ranged from <0.01 to 0.26 µmol C copepod^{-1} day^{-1}, and across all stations, averaged 0.06 ± 0.06 µmol C copepod^{-1} day^{-1} (FIGURE 14; Supplementary Table 5). The station with the highest mean ingestion was D6c at 0.12 ± 0.13 µmol C copepod^{-1} day^{-1}, whereas the lowest was D1 at <0.01 ± 0.00 µmol C copepod^{-1} day^{-1}. C-specific ingestion rates ranged from <0.0001 to 0.0214 day^{-1} and averaged 0.0040 ± 0.0048 day^{-1}.

Egg production rates (EPR) ranged from 0.0 to 6.3 eggs copepod^{-1} day^{-1} and averaged 1.3 ± 1.7 eggs copepod^{-1} day^{-1} across all experiments (FIGURE 14). This corresponds to C-specific EPR ranging from 0.000 to 0.013 day^{-1}, and a mean average of 0.0024 ± 0.0034 day^{-1}. Egg production rates did not correlate with female C content (ρ=-0.11, p=0.57), N content (ρ=-0.21, p=0.3), or their C:N ratio (ρ=-0.24, p=0.17). At the stations where the experimental animals were collected, between 0-90% of the females were ≤GS3, and many of those within GS4 were developing relatively small clutches of eggs (= GS4B and GS4C) (TABLE 5). The mean proportion of spawning females across stations was 70 ± 24 %. Gonad maturation was highest for the individuals in experiment 1, despite EPR being higher in experiments 2 and 3.

The C-specific rate of ingestion did not vary significantly with the concentration of chlorophyll-*a* (FIGURE 15A). However, ingestion increased significantly with total microplankton carbon (FIGURE 15B), likely driven by concentrations of centric diatoms (p=0.04; FIGURE 15C). Ingestion did not vary significantly with flagellate concentration (FIGURE 15D), nor ciliate or dinoflagellate concentrations (Supplementary Table 7). In contrast to this, C-specific rates of egg production were not related to any measure of food availability (FIGURE 15E-H; Supplementary Table 7). The correlation coefficients of the relationships between food availability and C-specific rates of ingestion and production were similar to those between food availability and the absolute rates of ingestion and production (Supplementary Figure 8), except for a weak negative relationship between absolute production and dinoflagellate concentration (ρ=-0.35, p=0.046). There was no relationship between C-specific rates of ingestion and production (ρ=0.07, p=0.68; Supplementary Figure 9), even with time lags of 24, 48, and 72 hours (p>0.30 in all cases). Similarly, the relationship between specific production and the proportion of females with mature gonads was not significant (Supplementary Figure 10), nor was the relationship between specific production and temperature (p=0.07).

The ingested diet was typically dominated by flagellates and ciliates, and to a lesser extent by small centric diatoms (FIGURE 16). The percentage of ciliates in the diet was greater than their availability in the plankton at all stations excluding D6b. All other cells constituted a relatively minor component of the diet and were generally ingested in similar proportions to their availability in the plankton; the percentages of dinoflagellates and pennate diatoms in the diet were positively correlated with their availability in the plankton (dinoflagellates: $\rho=0.9$; pennate diatoms: $\rho=0.9$; $p<0.01$ in both cases).

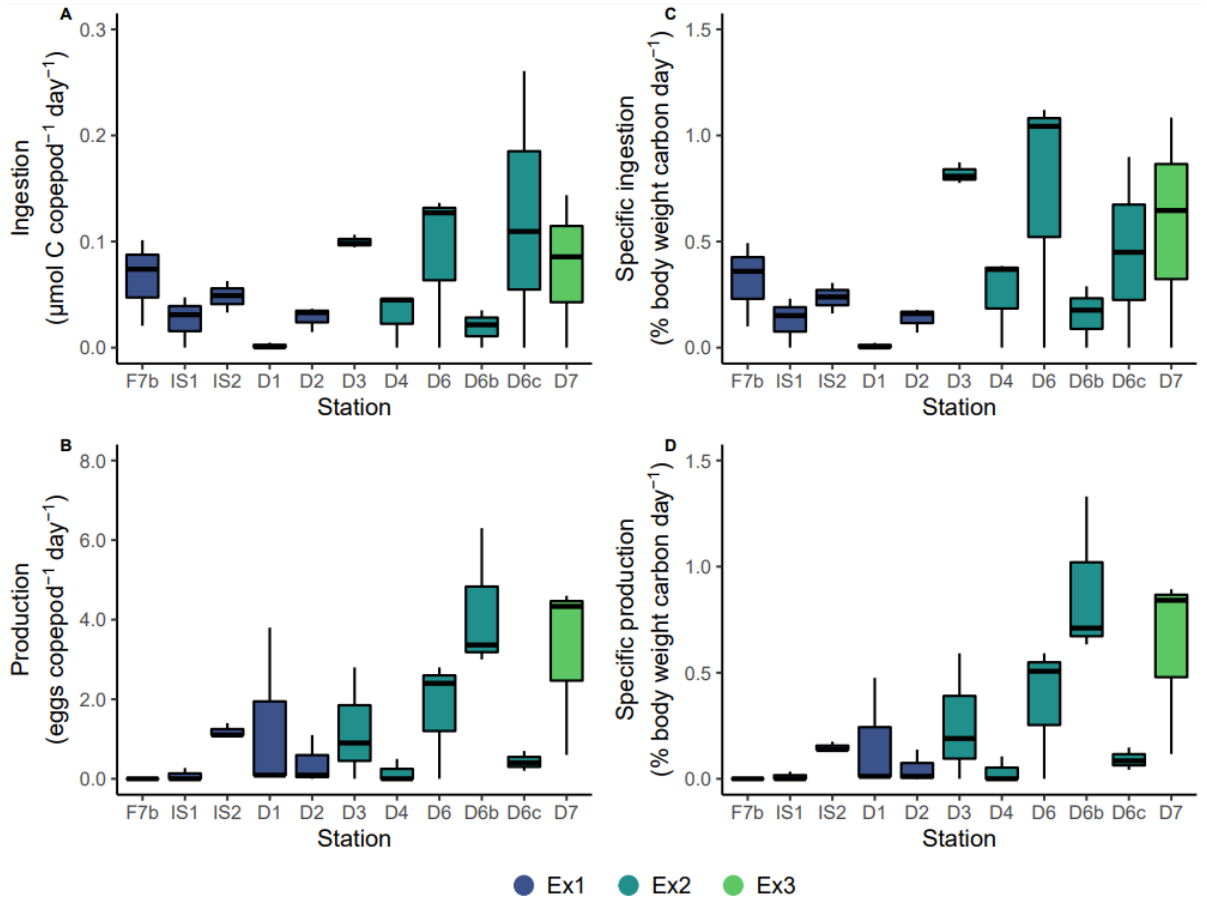


FIGURE 14 | Total ingestion (A), specific ingestion (B), total egg production (C), and specific egg production (D) by *Calanus finmarchicus* females across experiments 1-3 (Ex1-3) in the Fram Strait in summer 2019. The mid-line represents the median, the box the upper and lower quartiles, whiskers the range, and black points the outliers. Specific ingestion and production were converted from a fraction to a percentage for these figures.

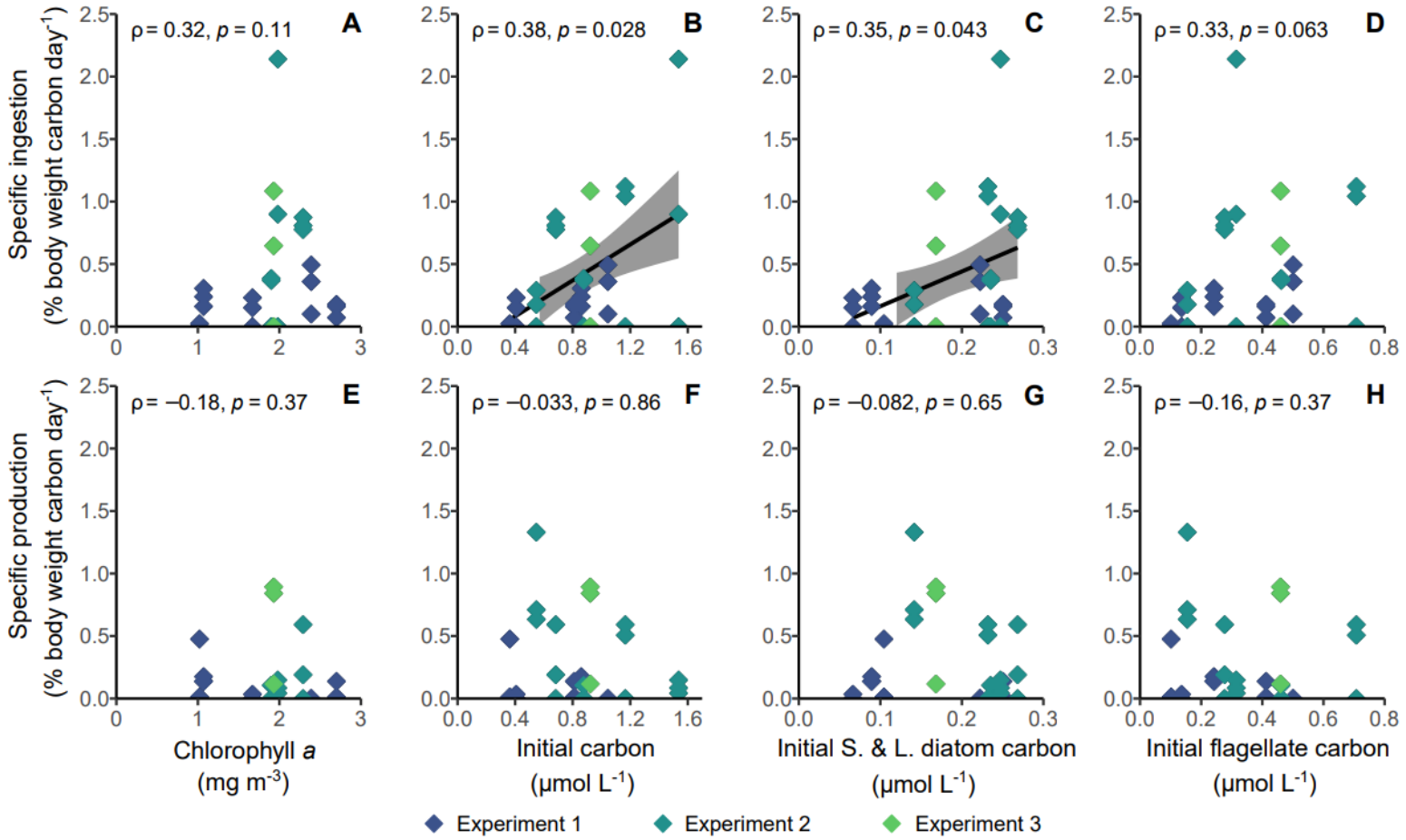


FIGURE 15 | The relationships between the quantity of available food and ingestion (A, B, C & D) and egg production rate (E, F & G, H) of *C. finmarchicus* in the Fram Strait in summer 2019. The quantity of available food is estimated from using the chlorophyll a concentration as a proxy (A & E) and calculated through flow imaging microscopy for all cell types (B & F), for small (S) and large (L) centric diatoms (C & G), and for flagellates (D & H). Shaded areas show 95 % confidence intervals. The spearman's rank correlation coefficient is shown as ρ and p-value by p.

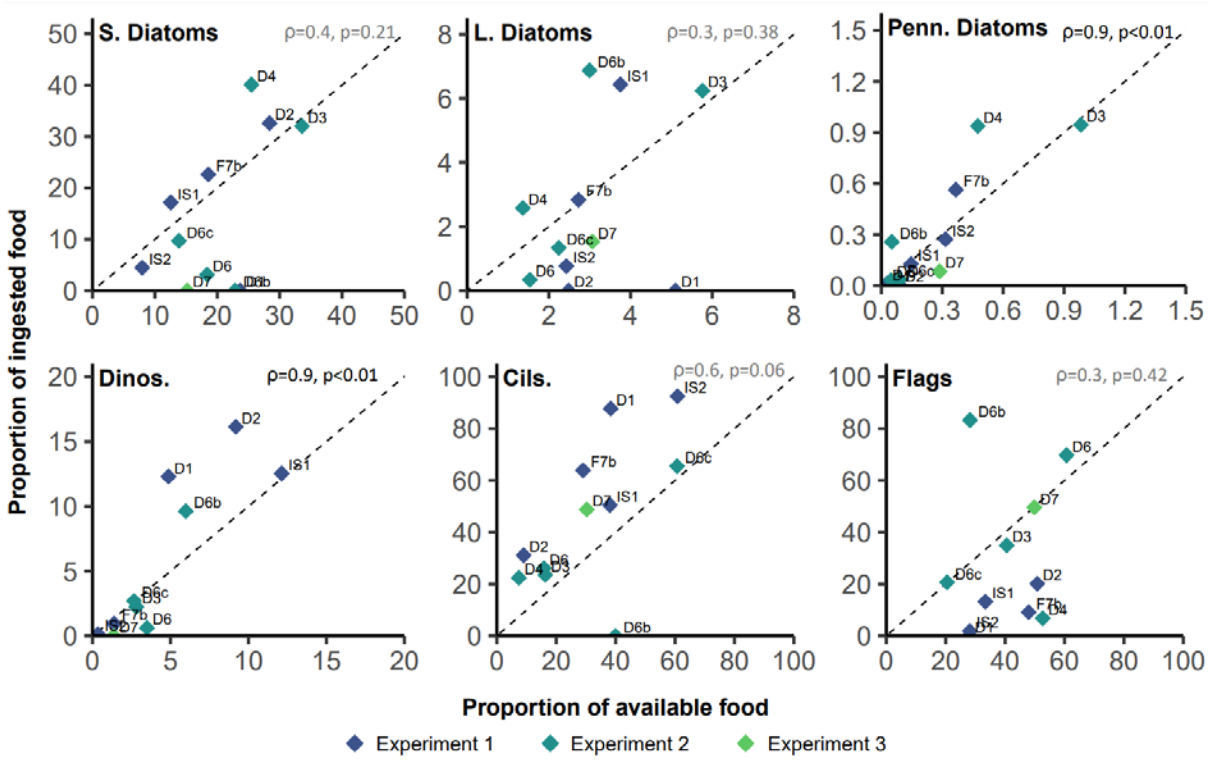


FIGURE 16 | The contribution of food types to the diet of *C. finmarchicus* against the contribution of the food types to the available food environment, in the Fram Strait in summer 2019. The dotted line represents the 1:1 line where ingestion is proportional to available food, i.e. non-selective feeding. Above this, a greater proportion is ingested than is available, so the food is selected for, and below, the food is selected against. The spearman's rank correlation coefficient is shown as ρ and p-value by p. Significant relationships are in black font, insignificant are in grey font. Note the variable scales on the x and y-axes.

3.4.4 Metabolic carbon budgets

The daily metabolic C budgets for the experimental animals are shown in TABLE 6. The calculated gross growth efficiencies (GGE) of the animals were highly variable, ranging from 0.00 to 4.61, and averaging 0.64 ± 1.12, largely attributable to the wide range of ingestion rates (TABLE 6). A GGE over 1 indicates a source of additional carbon. Ingestion was consistently much lower than the C needed for metabolism alone, accounting for only 15 ± 16 % of respiratory demand. When including ingestion and production, the C deficit became even more pronounced. The mean deficit was 0.37 ± 0.06 µmol C copepod^{-1} day^{-1} (TABLE 6), which is very similar to the amount of C needed for respiration (mean = 0.38 ± 0.01 µmol copepod^{-1} day^{-1}), indicating that respiration is the dominant factor. The largest C deficits were at stations D1 and D6b, where they were 0.39 ± 0.00 and 0.47 ± 0.04 µmol C copepod^{-1} day^{-1}, respectively (TABLE 6). Changing the assumed metabolic substrate to

lipid metabolism by adjusting the RQ to 0.7 (Ikeda et al., 2000) reduced the deficit to a mean average of 0.26 ± 0.06 µmol copepod^{-1} day^{-1} (TABLE 6).

Station	Ex	n	GS1	GS2	GS3	GS4C	GS4B	GS4A
F7b	2	10	0	0	60	30	10	0
IS1	2	10	0	0	0	40	30	30
IS2	2	9	11	0	0	22	33	33
D1	2	10	0	0	60	30	10	0
D2	2	10	0	0	10	10	50	30
D3	3	10	0	0	30	20	20	30
D4	3	10	10	10	70	0	10	0
D6	3	10	0	0	30	40	10	20
D7	4	10	0	0	30	40	20	10
D8	4	12	8	8	25	50	0	8
D9	4	10	0	10	30	40	20	0

TABLE 5 | Gonad maturation stages of *C. finmarchicus*. N denotes sample size. Ex denotes experiment number. GS shows the percentage of females in gonad maturation stages 1-4a as per Niehoff & Runge, 2003.

Ex	Day	Station	Ingestion (I)	Production I	GGE	Respiration I	Egestion (W)	Carbon required	
			(µmol C cop-1 day-1)	(µmol C cop^{-1} day-1)		(µmol C cop^{-1} day-1)	(µmol C cop^{-1} day-1)	(µmol C cop^{-1} day-1)	
1	1	F7b	0.07 ± 0.04	NIL	NA	0.38	0.02 ± 0.01	0.34 ± 0.03	[0.23 ± 0.03]
1	2	IS1	0.03 ± 0.02	NIL	0.11 ± 0.16	0.38	0.01 ± 0.01	0.37 ± 0.02	[0.26 ± 0.02]
1	3	IS2	0.05 ± 0.01	0.03 ± 0.00	0.71 ± 0.34	0.38	0.01 ± 0.00	0.38 ± 0.02	[0.27 ± 0.02]
1	4	D1	<0.01 ± 0.00	NIL	NA	0.38	<0.01 ± 0.00	0.39 ± 0.00	[0.28 ± 0.00]
1	5	D2	0.03 ± 0.01	0.01 ± 0.02	0.34 ± 0.46	0.38	0.01 ± 0.00	0.37 ± 0.02	[0.27 ± 0.02]
1 (average)			0.04 ± 0.03	0.01 ± 0.01	0.32 ± 0.37	0.38 ± 0.00	0.01 ± 0.01	0.37 ± 0.02	[0.26 ± 0.02]
2	1	D3	0.10 ± 0.01	0.03 ± 0.44	0.33 ± 0.37	0.37	0.03 ± 0.00	0.33 ± 0.04	[0.23 ± 0.04]
2	2	D4	0.03 ± 0.03	0.00 ± 0.09	0.14 ± 0.20	0.37	0.01 ± 0.01	0.35 ± 0.02	[0.25 ± 0.02]
2	3	D6	0.09 ± 0.08	0.04 ± 0.47	0.26 ± 0.37	0.37	0.02 ± 0.02	0.35 ± 0.08	[0.25 ± 0.08]
2	4	D6b	0.02 ± 0.02	0.11 ± 0.56	4.10 ± 0.72	0.37	0.00 ± 0.00	0.47 ± 0.04	[0.36 ± 0.04]
2	5	D6c	0.12 ± 0.13	0.01 ± 0.08	0.04 ± 0.01	0.37	0.03 ± 0.03	0.29 ± 0.10	[0.19 ± 0.10]
2 (average)			0.07 ± 0.07	0.04 ± 0.05	0.92 ± 1.61	0.37 ± 0.00	0.02 ± 0.02	0.36 ± 0.08	[0.26 ± 0.08]
3	1	D7	0.08 ± 0.07	0.08 ± 0.06	1.08 ± 0.43	0.36	0.02 ± 0.02	0.39 ± 0.03	[0.29 ± 0.03]
All (average)			0.06 ± 0.06	0.03 ± 0.04	0.64 ± 1.12	0.38 ± 0.01	0.01 ± 0.02	0.37 ± 0.06	[0.26 ± 0.06]

TABLE 6 | Daily metabolic carbon (C) budgets for mature female *Calanus finmarchicus* (57ccount^{-1} day^{-1}), showing measured ingestion and production. Showing averages per bottle (every row), per experiment (average rows), and across all (All average row). Budgets are calculated as Ingestion (I) = Egg production (E) + Respiration (R) + Egestion (W) + C surplus (Ω). Production is assumed to equal egg production only due to lack of somatic growth (Poulet et al., 1995). Respiration was estimated using nitrogen biomass-specific equations (Ikeda et al., 2001) and a respiratory quotient (RQ) of 0.97. The budgets were calculated assuming that egestion is I × (1- absorption efficiency), where absorption efficiency = 0.74 (Anderson et al., 2017, 2020; Anderson, Hessen & Mayor, 2021). The C content of the animals was 20.55 µmol C copepod^{-1} for Ex1, 12.18 µmol C copepod^{-1} for Ex2, and 13.25 µmol C copepod^{-1} for Ex3. The nitrogen (N) content of the animals was 2.44 µmol N copepod^{-1} for Ex1, 2.35 µmol N copepod^{-1} for Ex2, and 2.28 µmol N copepod^{-1} for Ex3. The C surplus was also calculated using a respiratory quotient of 0.7 (shown in "[]"). Ex, experiment; Cop, copepod; GGE, Gross Growth Efficiency, the ratio of biomass production to ingestion. The mean average is shown ± standard deviation.

3.5 Discussion

This study quantified rates of feeding and egg production in female *Calanus finmarchicus* at stations across the Fram Strait under post-bloom conditions and examined how these varied in response to the food environment. The positive relationship between ingestion and food availability suggests that the animals were food-limited throughout the study period. By contrast, the lack of relationship between the rate of egg production and any measure of food quantity indicates that their reproductive output is likely controlled by factors other than simple metrics of food availability. Measured rates of ingestion were not sufficient to meet the estimated metabolic requirements and/or the eff production of the experimental females at all the stations investigated. This further supports the idea that they were food-limited., and that the animals were relying on additional source of C – likely internal lipid biomass or egg cannibalism.

3.5.1 Ingestion

Microplankton concentrations encountered throughout the sampled stations in the Fram Strait in August ranged between 0.36 to 1.54 µmol C L^{-1} and were in agreement with concentrations determined using traditional inverted microscopy techniques (Cook et al., submitted; Supplementary Figure 11). The observed values were at least an order of magnitude lower than the concentrations observed in May/June (range = 18.0 to 187.0 µmol C L^{-1}; Jenkins et al., 2022). This is consistent with the understanding that all of the sampled stations were in a 'post-bloom' condition, based on the low microplankton biomass and nutrient concentrations encountered (Tarling et al., 2022a). Microplankton concentrations were also variable in time; sequential sampling over three days at a single location (stations D6, D6b, D6c) produced variable results (FIGURE 12). High variability in microplankton concentrations in the Fram Strait between July and August has previously been attributed to eddy currents and proximity to the ice shelf (Hirche et al., 1991; Tarling et al., 2022a). During these months, upwelling nutrients caused by isopycnal doming and mesoscale eddies control phytoplankton distribution in the Fram Strait, with contrasting regimes on either side of the Strait (Schourup-Kristensen et al., 2021).

The composition of the microplankton community was also very different to that observed in the Fram Strait in May/June, switching from being a diatom-dominated system (Jenkins et al., 2022) to one that is dominated by flagellates and microzooplankton (this study). A similar compositional shift towards the end of the productive cycle has been reported previously (Hegseth & Sundfjord, 2008; Tremblay et al., 2009; Ardyna et al., 2017; Ardyna & Arrigo, 2020), with small flagellates (Bauerfeind et al., 2009; Engel et al., 2019), haptophytes and nanoflagellates (Nöthig et al., 2015),

Synechococcus spp. (Paulsen et al., 2016), and *Micromonas* spp. (Kilias et al., 2014) dominating the Fram Strait and other areas of the Arctic during this period. This shift has been attributed to a low availability of nitrate-nitrogen (N), the supply of which is exhausted earlier in the year, enabling the small, motile organisms to outcompete the larger diatoms (Robinson, 2017). As anthropogenic warming causes sea-ice melt to begin earlier in the year, there is a longer growing season for phytoplankton and greater open sunlit area, increasing primary production (Arrigo, van Dijken & Pabi, 2008). However, increased stratification decreases the supply of N (Tremblay & Gagnon, 2009; Farmer et al., 2021; Noh et al., 2023), meaning that smaller cells thrive and outcompete the larger ones (Li et al., 2009). The seasonal transition from diatoms to ciliates and flagellates may become exaggerated in the future as the climate continues to warm (Li et al., 2009).

C. finmarchicus has previously been reported to consume prey in approximate proportion to their availability in the plankton (Mayor et al., 2006; Castellani et al., 2008; Mayor et al., 2009a; Djeghri et al., 2018; Jenkins et al., 2022), potentially being less selective than other calanoid copepods (Teegarden et al., 2008). The present study also provides general support for this understanding, with the relative abundances of many of the enumerated cell groups in the diet being in approximate balance with their availability in the water at the start of the experiments (Figure 6). A possible exception to this generalist feeding behaviour was the positive selection for ciliates, as their contribution to the diet exceeded their availability at all but one of the stations examined. Ciliates have previously been noted as an important component in the diets of copepods (Calbet & Saiz, 2005; Saiz & Calbet, 2011), including *C. finmarchicus* in the North Atlantic (Mayor et al., 2006; Castellani et al., 2008). The apparent preference for ciliates may reflect their higher nutritional value compared to other prey and/or the increased ability of *C. finmarchicus* to detect these motile cells.

The positive relationship between food availability and ingestion rates suggests that the animals were food-limited through the study, most likely because the encountered concentrations of microplankton were well below the concentration at which feeding saturates for *C. finmarchicus* (42 µmol C L^{-1}; Båmstedt, Nejstgaard & Solberg, 1999). The post-bloom ingestion rates for *C. finmarchicus* presented herein (0.06 ± 0.06 µmol C $copepod^{-1}$ day^{-1}, ranging from 0 to 0.26 µmol C $copepod^{-1}$ day^{-1} at 3.1 ± 2.0 °C) agree with measurements made under similar conditions in other areas of the Atlantic, e.g. 0.43 µmol C $copepod^{-1}$ day^{-1} in June to July in the Gulf of St Lawrence at ~7°C (Ohman & Runge, 1994); and ranging between 0.18 – 0.67 µmol C $copepod^{-1}$ day^{-1} in July to August in the Northeast Atlantic at 4.1 – 11.4 °C (Mayor et al., 2006). The reported ingestion rates are much lower than those determined under bloom or near-bloom conditions during May in the Fram Strait, which averaged 4.7 ± 3.6 µmol C $copepod^{-1}$ day^{-1} and ranged from 0.3 to 12.3 µmol C

copepod^{-1} day^{-1} (Jenkins et al., 2022). C-specific ingestion rates ranged from <0.0001 to 0.0214 day^{-1} and averaged 0.0040 ± 0.0048 day^{-1}, which is also lower than reported elsewhere: 0.023 day^{-1} in June to July in the Gulf of St Lawrence at ~7°C (Ohman & Runge, 1994); 0.025 – 0.047 day^{-1} in July to August in the North Atlantic at 4.1 – 11.4 °C (Mayor et al., 2006). However, this is likely attributable, at least in part, to the larger C-biomass of the females in the present study (14.2 µmol C copepod^{-1}) compared to the smaller sized animals incubated elsewhere (average = 9.79 µmol C copepod^{-1}; Mayor et al. 2006). The generally low ingestion rates may also be partially attributed to the small size of flagellates, the dominant group encountered in the microplankton communities at many of the sampled stations (FIGURE 12). There is evidence to suggest that *C. finmarchicus* can clear small cells at high rates (Levinsen et al., 2000; Mayor et al., 2006, 2009a). However, it is widely accepted that clearance rates increase with prey sizes above ~10 µm equivalent spherical diameter (which would include the smallest ciliates and dinoflagellates found in this study), whereas the smallest cells are barely perceived or retained (Traboni, Calbet & Saiz, 2020). This preference creates a dome-shaped relationship between feeding rates and prey size that is well-established for other calanoid copepods (e.g. Berggreen, Hansen & Kiørboe, 1988; Traboni, Calbet & Saiz, 2020). Coupled with the overall scarcity of prey, the relationship between food size and ingestion rates likely contributed to the low ingestion rates recorded here.

3.5.2 Egg production

The observed egg production rates (EPRs) were variable (range: 0.0 to 6.4 eggs copepod^{-1} day^{-1}; mean: 1.3 ± 1.7 eggs copepod^{-1} day^{-1}) and at the lower end of the range previously observed for *C. finmarchicus* in the Arctic (0.3 – 36.7 eggs copepod^{-1} day^{-1}; Hirche, 1990; Hirche & Kosobokova, 2007; Møller et al., 2016). These results are consistent with the post-bloom conditions encountered; individual EPRs in *C. finmarchicus* reach their maximum during the spring phytoplankton bloom when food supply is highest and then decrease thereafter (Niehoff et al., 1999). The observed EPRs were lower than those observed in a suite of parallel experiments where individual copepods were incubated without food in mesh-bottomed egg production chambers for 24 hrs (16.1 ± 24.8 eggs copepod^{-1} day^{-1}; Cook et al., Submitted), although these two datasets are not directly comparable because the rates presented herein were derived from groups of females that were in the presence of food rather than starved, and sequentially incubated for a period of up to 5-days. The C-specific EPRs (mean = 0.0024 ± 0.0034 day^{-1}; range: 0.000 to 0.013 day^{-1}) were also within the published range, albeit towards the lower end (Supplementary Table 3). Egg production in *C. finmarchicus* varies throughout the year, often in response to food availability (Runge, 1984b; Hirche & Bohrer, 1987; Hirst & Bunker, 2003; Mayor et al., 2009b), temperature (Pasternak et al., 2013), and gonad maturity (Niehoff, 2004; Jenkins et

al., 2022). However, EPRs did not correlate with any measure of ingested food quantity or prey type (FIGURE 15E-H), temperature ($p = 0.07$), nor did they relate to the amount of food Ingested during the preceding day ($p \geq 0.79$ in all cases). These results suggest that the absence of a relationship between ingestion and egg production was not due to a lag between food being ingested and it being packaged into eggs. We reported similar results from identical experiments conducted in the Fram Strait during May and attributed the lack of a relationship between ingestion and egg production to variability in gonad maturity (Jenkins et al., 2022). This may also be the case here, as the variability in gonad maturity is similar (70 ± 24 % mature in here compared to 72 ± 13 % mature in May)(TABLE 5). It is possible that the lack of available food had prevented the immature females from undergoing the final stages of gonad maturation, as these processes are known to be food dependent (Niehoff et al., 2002). However, the proportion of mature females did not correlate with EPR (Supplementary Figure 10), meaning another factor than gonad maturity was confounding the relationship between ingestion and production. It therefore seems likely that the mature incubated females were sustaining egg production via the consumption of prey items not determined herein, including the eggs of conspecifics, and/or by catabolising their own biomass (explored in Section 3.5.3, below). Exactly why a component of the *C. finmarchicus* population remained reproductively active throughout August, when much of the population was already in or entering diapause (Tarling et al., 2022a, 2022b) remains unclear as their progeny are unlikely to find sufficient food to sustain their development and survival through the winter months (Anderson et al., 2022). Nevertheless, multiple lines of evidence demonstrate that a fraction of the *C. finmarchicus* population, possibly those that have not acquired sufficient lipid to overwinter, remain active in the surface and continue to feed (Pasternak et al., 2001; Berge et al., 2015; Hobbs et al., 2020).

3.5.3 Carbon budgets

Carbon budgets were constructed for each experiment by combining measured rates of C intake and egg production with empirically estimated rates of respiration and faecal pellet production. In all instances, ingested C was insufficient to cover the amount required for respiration (TABLE 6). Changing the respiratory quotient from carbohydrate- to lipid-fuelled metabolism (i.e., changing RQ from 0.97 to 0.7), consistent with the idea that the females were consuming their remaining lipid reserves, reduced the metabolic costs of respiration by 0.10 µmol copepod^{-1} day^{-1} and therefore reduced the shortfall substantially. However, metabolic deficits remained in all cases. This result contrasts with our observations made at similar locations in May, where in all but one instance, female *C. finmarchicus* ingested excess C relative to their metabolic requirements (Jenkins et al., 2022). Nevertheless, this is not the first study to indicate that these copepods face

periods where ingested food falls below the requirements for respiration and egg production (Mayor et al., 2006, 2009a), and other studies have shown that *C. finmarchicus* can continue to produce eggs when food is scarce or even absent (e.g. Hirche, Meyer & Niehoff, 1997; Niehoff, 2004, 2007).

It is possible that the metabolic deficits arose because we underestimated rates of food intake. Ingestion was estimated using well-established particle removal experiments (Harris et al., 2000b) in which the prey cells were enumerated using FlowCam technology (Poulton, 2016). The resulting microplankton biomass estimates agreed well with those determined using traditional inverted microscopy (Cook et al., Submitted; Supplementary Figure 11), providing confidence that the FlowCam had not underestimated the biomass of cells available, or missed groups of cells that made a substantial contribution to the diet. An alternative, and perhaps more parsimonious explanation is that the missing metabolic substrates were derived from the biomass of the experimental females. Similar, previous experiments in the North Atlantic have shown that *C. finmarchicus* can derive >80% of their C requirements from their own biomass when feeding conditions are unfavourable (Mayor et al., 2009a). Changes in the C and N contents of the population of incubated females before and after the experiments were not statistically significant, although this is perhaps not surprising given the large amount of individual variability in these (FIGURE 13). *Calanus* spp. are known to consume their own eggs at times (Bonnet, Titelman & Harris, 2004) and therefore a further, non-mutually exclusive explanation is that egg cannibalism contributed additional metabolic substrates to the experimental animals. The incubated groups of females were not excluded from their eggs and were therefore potentially able to consume any eggs spawned within each 24-hr incubation period. Using the assumed value of 20.9 nmol C egg^{-1} (Mayor et al., 2009a), each female would have needed to ingest approximately 18 eggs day^{-1} in order to meet the 0.37 µmol C deficit, or 13 eggs day^{-1} if assuming RQ = 0.7. These appear to be plausible egg consumption rates and approximate the mean EPR observed in parallel experiments in which the females were excluded from their eggs (16.1 ± 24.8 eggs $copepod^{-1}$ day^{-1}; Cook et al., Submitted). Unfortunately, it is not possible to conclusively identify the source(s) of the substrates required to balance the metabolic budgets from the data presented herein. Regardless, our study adds to the growing body of evidence to indicate that a fraction of the *C. finmarchicus* population remains active in surface waters after the majority of the population has descended into the deep ocean to overwinter (Berge et al., 2015; Hobbs et al., 2020). Given that the encountered food concentrations and the resulting ingestion rates were too low for these animals to achieve metabolic balance, it seems improbable that their progeny would be able to find sufficient food to survive and develop during the subsequent months. However, extending the period over which reproductively active *C. finmarchicus* remain in surface waters

increases the ability of their population to exploit ephemeral pulses of food availability, and the very presence of 'late-spawners' suggests that selection pressure against this trait is not sufficiently strong to remove it from the population. We suggest that reproductive flexibility is one of the characteristics that helps *C. finmarchicus* exploit new or changing environments, and likely contributes to the penetration of this species at increasingly high latitudes as the climate changes. Nevertheless, if the conditions encountered throughout the study period are representative of those that will occur in the future Arctic, the resulting low rates of ingestion and egg production suggest that copepods like *C. finmarchicus* may struggle to persist in this food-limited environment. More work is required to better understand the likely outcome of climate change in Arctic ecosystems, including how patterns of primary and secondary production will respond to both the direct and indirect effects of increased temperature.

Chapter 4 Characterising the plankton assemblage across the Fram Strait: a methodological comparison

4.1 Abstract

The composition of the pico- and micro- plankton environment of the Arctic has crucial implications for the production and survival of key zooplankton species, and in turn for biogeochemical cycling. Calanoid copepods dominate biomass and rely on the microplankton to support growth, lipid storage, and to complete their life cycle. Rapid changes to the Arctic are altering the plankton composition, with a trend from large diatoms to more motile, smaller organisms. This study compares the taxonomic composition of plankton in the Fram Strait in two seasons analysed via different methodologies: through metabarcoding of the 18S nuclear small subunit ribosomal RNA, and through both traditional morphological microscopic identification and more modern high-throughput flow imaging identification. The final metabarcoding dataset consisted of 872,101 barcoding reads representing 212 different species. The metabarcoding analysis demonstrated that despite the diversity, the seston was dominated by *Calanus* spp. (36.2 % of the reads in Spring 2018 27.8 % in Summer 2019) and *Prorocentrum micans* (13.2 % of the reads in Spring 2018 and 8.0 % in Summer 2019). The plankton assemblage had a larger proportion of small diatoms in Spring 2018 than in Summer 2019 (7.2 ± 6.6 % and 2.1 ± 1.6 % respectively), and a smaller proportion of flagellates (0.6 ± 1.0 % and 6.6 ± 5.2 % respectively). There were potentially harmful diatom and flagellate species in Summer 2019, including *Pseudo-nitzschia australis* and *Aureococcus anophagefferens* that were not present in Spring 2018. Each method had distinct strengths, resulting in differences in the taxonomic composition measured via each method. The traditional microscopy and metabarcoding taxonomic compositions were more similar than the flow imaging and metabarcoding taxonomic compositions. Our results highlight that researchers should choose their method carefully depending on the size of the target organisms and the resource available. This study also also supports a potential shift to a plankton assemblage dominated by the smaller taxa in the future, with a higher prevalence of harmful species, while highlighting the need for long term monitoring.

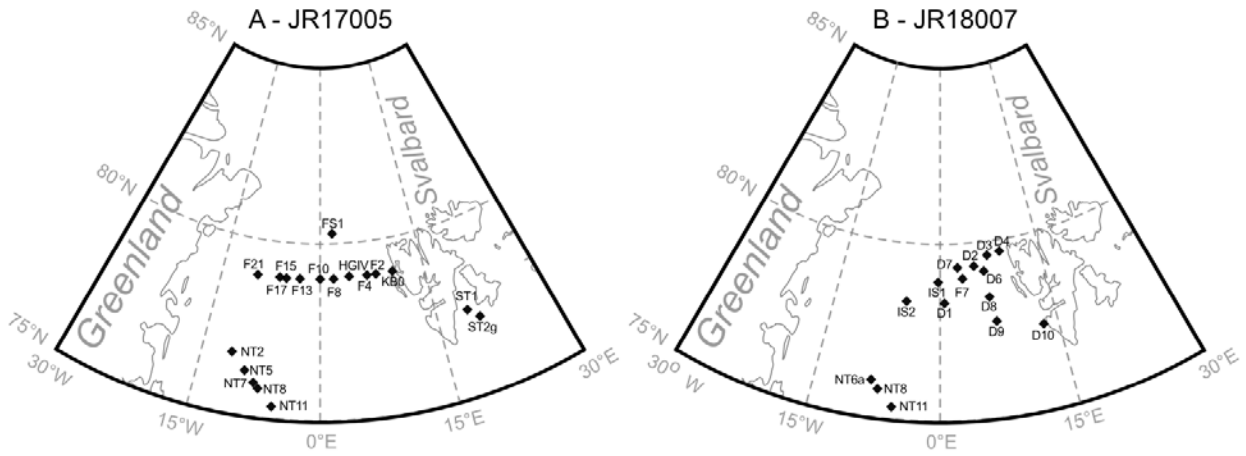


FIGURE 17 | The locations of stations where sampling for metabarcoding analysis was undertaken, in May – June 2018 and August 2019 on research cruises JR17005 and JR18007 respectively. Sampling station D6 marks D6, D6b and D6c.

4.2 Introduction

The Arctic Ocean is experiencing rapid, human-led change (Thomas et al., 2022), warming at nearly four times the global mean rate (Rantanen et al., 2022). This temperature increase causes a range of hydrographical changes, with the reduction of sea ice cover and glacier retreat resulting in an increase in freshwater discharge and a change in the overall circulation, including strengthening stratification (Haine et al., 2015; Haine & Martin, 2017; Farmer et al., 2021). The reduction in sea-ice extent has increased open-water habitat and growing season for phytoplankton (Arrigo, van Dijken & Pabi, 2008; Kahru et al., 2011), while simultaneously the strengthened stratification reduces the upward supply of nutrients (Ardyna et al., 2014). The changing physical and chemical ocean environment is expected to impact the composition, distribution, timing and magnitude of primary production (Li et al., 2009; Kahru et al., 2011; Yool, Popova & Coward, 2015; Neukermans, Oziel & Babin, 2018; Lewis, Van Dijken & Arrigo, 2020). Traditionally in the Arctic, there has been a spring bloom of diatoms and the subsequent sedimentation of organic matter (Lovejoy et al., 2002). Physical and chemical oceanic changes may mean a phenology shift for plankton, with a second late phytoplankton bloom becoming more prevalent (Ardyna et al., 2014; fig. 5 in Ardyna & Arrigo, 2020). In addition to diatoms, ciliates and dinoflagellates are important unicellular grazers and channelling primary production to the higher trophic levels, particularly in the late summer (Verity et al., 2002). With a warming and freshening ocean, it is predicted that conditions will be more favourable to picoplankton and so there will be a shift away from the larger phytoplankton cells towards a system dominated by ciliates, dinoflagellates, and flagellates (Li et al., 2009; Morán et al., 2010; Comeau et al., 2011; Henson et al., 2021).

Changes to the pico- and micro- plankton affect the nutritional content of the food ingested by calanoid copepods – generally the dominant zooplankton group in the Fram Strait (Hop et al., 2006; Blachowiak-Samolyk et al., 2007) and a key taxa in the transfer of energy to higher trophic levels (Gatten & Sargent, 1973) including amphipods (Dischereit et al., 2022) and juvenile fish. Microplankton taxa vary in their nutritional composition in terms of carbon, fatty acids, and macronutrient composition (Ho et al., 2003; Jónasdóttir, 2019 and references within), and so will differ in meeting the metabolic needs of the copepods – needs that will increase with temperature (Gillooly et al., 2001). Arctic *Calanus* spp. tend to feed unselectively, with their ingestion reflecting the prey composition around them (Jenkins et al., 2022,in prep.). Therefore, understanding the microplankton composition, and how it is affected by the changing Arctic, is necessary to understand what controls production in copepods, to understand the trophic interactions, and to parameterise ocean biogeochemical models.

However, a challenge in measuring the composition of plankton communities is the identification and quantification of samples in a time-efficient manner. Traditionally this has been done by inverted microscopy of settled seston samples, but this is labour- and expertise- intensive. More recently, high-throughput techniques have become available, including imaging flow cytometers like the Flow Cytometer and Microscope (FlowCam) (Poulton, 2016). This allows rapid enumeration of particles and uses an algorithm to automatically classify them based on morphology. An alternative approach is deoxyribonucleic acid (DNA) metabarcoding, where DNA sequencing approaches can be applied to a mixed plankton sample to identify the taxa present (De Vargas et al., 2015; Sunagawa et al., 2015; Gran-Stadniczeñko et al., 2019). There is no standard methodology for analysis of microplankton, yet there is a need to compile long-term datasets from different studies to understand the full picture of change in the Arctic Ocean. The efficacy of the more recent approaches has not yet been assessed for the Arctic microplankton assemblage in comparison to traditional microscopy. There is a need for a direct comparison of methods with the rapidly increasing prevalence of studies basing their findings on metabarcoding (e.g. Egge et al., 2021; Cerfonteyn et al., 2023).

The aim of this study was to understand how the composition of the Arctic nano and microplankton can be reliably measured. We characterised the seston of the Fram Strait in May – June 2018 and August 2019 using molecular methods. We then consolidated seston analysis by inverted microscopy (Jenkins et al., 2022) and by flow imaging (Jenkins et al.,in prep.) and directly compared it to the molecular methods to assess the reliability of each method.

4.3 Methods

4.3.1 Experimental procedure

The plankton assemblage was sampled at 32 stations across the Fram Strait during May – June 2018 and August 2019 (FIGURE 17; RRS James Clark Ross cruises JR17005 and JR18007). To collect samples for metabarcoding analysis, the natural plankton assemblage was collected daily from the chlorophyll maximum via 20 L Niskin bottles and filtered under a gentle vacuum onto pre-combusted glass-fibre filters (Whatman GF/F; 0.7µm nominal pore size) and transferred into a 5-ml cryotube, which was stored at −80 °C until extraction. To collect samples for microplankton analysis, two 200 mL water samples were collected at each station across both cruises and immediately preserved with 1 % acidified Lugol's iodine for subsequent analysis.

4.3.2 Microplankton analysis

Lugols-preserved microplankton samples (200 mL) were analysed to enumerate and identify the particles present. In spring 2018 (JR17005), particles were analysed using inverted microscopy at x 250 and x 40 magnification for cells >2 µm and small flagellates, respectively. Cell dimensions were measured as detailed in Chapter 2. In summer 2019 (JR18007), particles were analysed using a FlowCam® 8400 (Yokogawa Fluid Imaging Technologies LLC) as detailed in Chapter 3. Cell volumes for the methods were calculated geometric formulae and using area-based diameter respectively, then converted to carbon biomass using published conversion factors specific to the cell type (Menden-Deuer & Lessard, 2000; Malzahn & Boersma, 2012; Hillebrand et al., 1999).

4.3.3 Molecular analysis

Primers (SSU_F04 and SSU_R22), designed by Fonseca et al., 2010, were chosen for amplicon generation (GTACACACCGCCCGTC and TGATCCTTCTGCAGGTTCACCTAC). These primers target a homologous region of the 18S nuclear small subunit (nSSU) ribosomal RNA (rRNA) gene and flank a region that is highly divergent (V1-V2). The 18S nSSU rRNA gene V4 variable region PCR primers SSU_F04 and SSU_R22 with barcode on the forward primer were used in a 30-35 PCR using the HotStarTaq Plus Master Mix Kit (Qiagen, USA) under the following conditions: 94°C for 3 minutes, followed by 30-35 cycles of 94°C for 30 seconds, 53°C for 40 seconds and 72°C for 1 minute, after which a final elongation step at 72°C for 5 minutes was performed. After amplification, PCR products were checked in 2% agarose gel to determine the success of amplification and the relative intensity of bands. Multiple samples were pooled together in equal proportions based on their molecular weight and DNA concentrations. Pooled

samples were purified using calibrated Ampure XP beads, Then the pooled and purified PCR product was used to prepare the Illumina DNA library. Sequencing was performed at MR DNA (www.mrdnalab.com, Shallowater, TX, USA) on a MiSeq following the manufacturer's guidelines. Sequence data were processed using MR DNA analysis pipeline (MR DNA, Shallowater, TX, USA). In summary, sequences were joined, sequences <150bp removed, and sequences with ambiguous base calls removed. Sequences are quality filtered using a maximum expected error threshold of 1.0 and dereplicated. The dereplicated or unique sequences are denoised; unique sequences identified with sequencing or PCR point errors are removed, followed by chimera removal, thereby providing a denoised sequence or zOTU.

4.3.4 Sequence data processing

The 454 sequencing reads were processed using the Qiime (Quantitative Insights into Microbial Ecology, v1.3.0) pipeline (Caporaso et al., 2010). The data were processed using default settings for all parameters, namely an operational taxonomic unit (I) threshold of 0.97, 0 primer mismatches, 0 ambiguous bases, a maximum length of homopolymer run of 6, and 200 nucleotides as a minimum sequence length. The samples were not multiplexed so the barcode area of the mapping file was left blank, and the split libraries script was altered accordingly. The data were then de-noised using the de-noiser wrapper within Qiime to remove the sequence errors characteristic of 454 sequencing machines (for review see Gaspar and Thomas 2013). Chimeras were identified using ChimeraSlayer and rejected from the dataset before construction of the OTU (Operational Taxonomic Units) table. The OTUs were assigned using UCLUST (Edgar, 2010), a de novo OTU picker within Qiime. A representative set of sequences was then generated, and these sequences were assigned taxonomy (at the level of 95% homology) using the BLASTN search of the NCBI non-redundant dataset.

4.3.5 Methodological comparison

Metabarcoding data were compared to samples taken in parallel for inverted microscopy (CHAPTER 2) and flow imaging analysis (CHAPTER 3), the methods for which are details in the cited literature. Previous methodological comparisons between microscopy and metabarcoding have shown that the relative abundances of the 18S rRNA amplicon at the group level best fitted the microscopy carbon biomass metric (Andersson et al., 2023), so this was used for comparisons.

Potential toxicity of the species identified was collated from an Arctic-specific list of harmful species (Poulin et al., 2011) and a keyword search of the literature, where one or more papers stating the species caused harm was classed as 'potentially harmful'.

4.3.6 Statistical and computational analysis

Statistical analysis and data visualisation were undertaken using the R programming environment (R Core Team, 2022) using the packages ggplot2 (Wickham, 2016), dplyr (Wickham et al., 2022), purrr (Henry & Wickham, 2020), ggpubr (Kassambara, 2020), viridis (Garnier, 2018), and plotly (Sievert, 2020).

Cruise	Station	Date	Latitude (°N)	Longitude (°E)	Temp (°C)	Depth (m)
JR17005	NT11	16/05/2018	75.336	-5.464	1.5	20
	NT8	17/05/2018	75.796	-7.218	1.3	20
	NT7	18/05/2018	75.949	-7.815	-0.7	6*
	NT5	19/05/2018	76.258	-9.029	-0.8	20
	NT2	20/05/2018	76.713	-10.905	-1.8	10
	F21	23/05/2018	78.985	-9.281	-1.7	10
	F17	25/05/2018	78.999	-5.982	-1.5	22
	F15	26/05/2018	78.986	-5.000	-1.5	32
	F13	27/05/2018	78.997	-2.996	-0.6	15
	F10	28/05/2018	79.000	0.000	-0.6	8
	FS1	29/05/2018	80.283	2.000	-1	10
	F8	30/05/2018	79.000	2.000	0.6	9
	HGIV	31/05/2018	79.048	4.332	4.1	23
	F4	01/06/2018	79.033	7.000	4.3	12
	F2	02/06/2018	79.033	8.333	3.1	10
	KB0	03/06/2018	79.035	10.843	-0.7	20
	ST1	05/06/2018	77.417	19.500	-0.5	23
	ST2g	06/06/2018	77.125	20.750	-0.8	17
JR18007	NT11	08/08/2019	75.336	-5.464	4.4	40
	NT8	10/08/2019	75.796	-7.219	4.8	34
	NT6A	11/08/2019	76.039	-8.095	5.1	31
	F7	12/08/2019	79.000	3.334	7.5	18
	IS1	14/08/2019	78.915	-0.288	6.0	24
	IS2	15/08/2019	78.364	-4.644	2.5	31
	D1	16/08/2019	78.317	0.616	6.0	20
	D2	17/08/2019	79.333	5.167	6.7	15
	D3	18/08/2019	79.600	7.333	7.0	10
	D4	19/08/2019	79.667	9.400	6.1	14
	D6	20/08/2019	79.167	6.600	6.6	20
	D6b	21/08/2019	79.167	6.600	6.7	6*
	D6c	22/08/2019	79.167	6.600	7.0	20
	D7	23/08/2019	79.317	2.649	6.0	9
	D8	24/08/2019	78.417	7.000	6.5	15
	D9	25/08/2019	77.717	7.583	7.4	27
	D10	26/08/2019	77.467	13.494	7.2	31

TABLE 7 | Locations of the stations sampled on research cruises JR17005 in May-June 2018 and JR18007 in August 2019. Temp. is the water column temperature at the chlorophyll-*a* maximum. Depth is the depth of water sampled using the CTD, chosen to be at the chlorophyll maximum. * denotes underway water sampling due to CTD sampling being logistically impossible.

FIGURE 18 | The relative composition of the seston across all stations in the Fram Strait during Spring 2018 on cruise JR17005 (A) and during Summer 2019 on cruise JR18007 (B). The width of each bar represents the frequency of occurrence. The outer ring shows the species while the centre shows their functional group. Genus names are not capitalised to save space. S. diatoms denotes small centric diatoms < 20 µm; L. diatoms denotes large centric diatoms ≥ 20 µm; and dinos denotes dinoflagellates. To view these figures with their interactive labels, download them at bit.ly/JR17005-ses and bit.ly/JR18007-ses respectively or view the QR codes above.

4.4 Results

4.4.1 Water temperature and water masses

The temperatures at the chlorophyll maxima in summer 2019 were consistently higher those in spring 2018 at 6.1 ± 1.3 °C and 0.1 ± 1.9 °C, respectively. There were two distinct temperature regimes Stations in Spring 2018 which reflected the colder East Greenland Current and the warmer West Spitsbergen Current; stations NT7, NT2, F21, F17, F15, F13 and F10 were < 0.0 °C at the chlorophyll maxima whereas the easterly stations HGIV, F4, and F2 were 3 – 4 °C (Jenkins et al., 2022). In summer 2019 there were less stations in the west due to the presence of ice sheets preventing sampling. However, the only westerly station showed the lowest temperature at 2.5 °C, suggesting a similar water mass as experienced in that area in spring 2018. The southern stations in summer 2019 (NT11, NT8 and NT6A) had lower temperatures despite being further south (4.4, 4.8 and 5.1 °C respectively) also indicating the influence of the southerly moving East Greenland Current. All northerly stations in 2019, other than IS2, had temperatures ≥ 6.0 °C.

4.4.2 Sequencing summary

The metabarcoding process produced 1,116,505 paired-end raw reads for the seston across cruises JR17005 and JR18007 in the Fram Strait. After all quality filtering steps, the final metabarcoding dataset consisted of 872,101 barcoding reads, clustered into 549 denoised OTUs at the 97% homology and representing 212 different species (131 eukaryotes and 81 prokaryotes). Each unique barcoding read was repeated an average of 5.3 times through the dataset, ranging from 1 to 119,794 times.

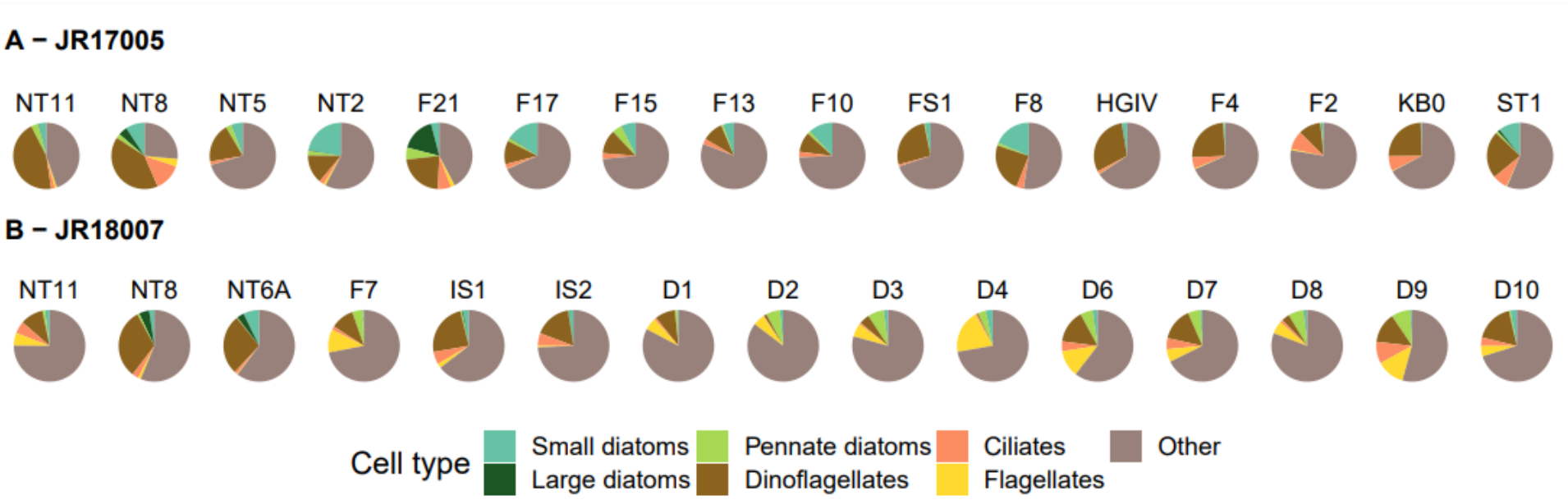


FIGURE 19 | The proportional composition of the seston in the Fram Strait during Spring 2018 on research cruise JR17005 from metabarcoding analysis (A) and in Summer 2019 on research cruise JR18007 (B). The seston is classified into cell types based on their role in the diet of a *Calanus* spp. in the Arctic.

4.4.3 Metabarcoding analysis of the microplankton

The plankton communities across both spring 2018 and summer 2019 were diverse yet both dominated by a small number of key taxa (FIGURE 18). The seston at the stations across both seasons were dominated by the functional groups calanoid copepods, bacteria and thecate dinoflagellates. However, in spring 2018, calanoid copepods made up 34.0 ± 22.2 % of the seston, bacteria 18.2 ± 10.8 %, and thecate dinoflagellates 17.4 ± 10.1 % while in summer 2019, bacteria made up 37.8 ± 11.1 %, calanoid copepods made up 27.6 ± 17.5 % and thecate dinoflagellates made up 10.5 ± 7.5 % (Supplementary Figure 12). When looking only at groups that are of interest to grazing *Calanus finmarchicus*, there remained considerable variation in the seston composition between across time and space (FIGURE 19). Across both seasons, dinoflagellates were the largest group with the seston comprising of 21.7 ± 10.5 % and 13.4 ± 9.2 % dinoflagellates, respectively, although in summer 2019 that proportion was much lower. Small diatoms were the second most abundant group in spring 2018, comprising 7.2 ± 6.6 % of the seston while in summer 2019 only making up 2.1 ± 1.6 %. The second most abundance group in Summer 2019 was flagellates, comprising 6.6 ± 5.2 % of the seston, whereas this group was the smallest in Spring 2018 at 0.6 ± 1.0 %.

The southern stations in spring 2018, NT11 and NT8, had high proportions of dinoflagellates – 44.8 % and 40.6 % respectively. While the same was not observed at NT11 in summer 2019 (only 10.4 % dinoflagellates), there were still high proportions at NT8 (31.6 %) and at the nearby station NT6A (27.2 %). The stations to the east of the Fram Strait in spring 2018, NT2, F17, F8 and ST1 had high proportions of small centric diatoms – 22.2 %, 15.8 %, 18.5 % and 10.6 %, respectively – while station F21 had a high proportion of large centric diatoms – 16.9 % and a relatively high proportion of pennate diatoms (5.8 %) when compared to the other stations across that season. The proportion of flagellates in the seston in spring 2018 was largest at NT8, yet still only 3.7 %. In summer 2019, there were high proportions of flagellates at stations D4, D9, D6, and F7 (19.6 %, 12.7 %, 12.1 %, and 10.5 % respectively).

The most common taxa overall across both cruises were *Calanus* spp. and *Prorocentrum micans* (a thecate dinoflagellate), comprising 36.2 % and 13.2 % of the OTUs in spring 2018, respectively, while in Summer 2019 these two contributed 27.8 % and 8.0 % of OTUs, respectively. *Phaeocystis* sp. was also abundant across both cruises, making up 4.8 % and 2.1 % in spring 2018 and summer, 2019 respectively. The other dominant species were different between the seasons, with spring 2018 having a high relative abundance of *Gymnodinium aureolum* (3.7 % of the spring 2018 seston whereas only comprising 1.3 % of the summer 2019 seston), *Micromonas pusilla* (3.6 % in spring 2018 compared to 0.7 % in summer 2019), and *Thalassiosira concaviuscula* (3.1 % in spring 2018 compared to 0.8 % in summer 2019.

Summer 2019 had high relative abundances of *Pseudoalteromonas* spp. (5.1 % in summer 2019 compared to 1.1 % in spring 2018), *Pseudo-nitzschia australis* (3.6 % in summer 2019 compared to 0.1 % in spring 2018), and *Fucophilus fucoidanolyticus* (3.1 % in summer 2019 compared to 0.1 % in spring 2018). The sea-ice diatoms *Navicula* spp. were found at all stations except NT11 in spring 2018, whereas they were only found at station IS2 in summer 2019.

Most of the species identified were not ones that are not groups of interest for grazing by *Calanus* (see 'Other' category in Supplementary Figure 13 and FIGURE 19). The food environment in spring 2018 consisted of 20 % dinoflagellates and 8 % small diatoms, whereas these categories were less prominent in summer 2019, consisting of 12 % and 2 % respectively (Supplementary Figure 13).

4.4.4 Method comparison

Comparable samples of the seston in the Fram Strait in May 2018 and June 2019 were analysed through both microscopy (inverted microscopy in Spring 2018 and flow imaging microscopy in Summer 2019) and through metabarcoding. The results of the microplankton analyses are published elsewhere – spring 2018 (Jenkins et al., 2022) and summer 2019 (Jenkins et al., in prep.).

The microscopy analysis showed that the composition and biomass of the microplankton varied considerably across the Fram Strait in both spring 2018 and summer 2019. In summer 2018, the microscopy analysis found that small chain-forming diatoms, and to a lesser extent large diatoms and dinoflagellates, were the dominant microplankton type at most stations sampled, while the stations close to Svalbard and the West Spitsbergen Current (HGIV, F4, F2, and KB0) had a lower proportion of both small and large diatoms. The westward stations (NT2, F21, F17, and F15) had a lower proportion of ciliates than those eastward (F13, F10, FS1, HGIV, F4, F2, except for F8 and KB0) (Jenkins et al., 2022). Overall, the metabarcoding results also reflected these patterns (FIGURE 20). There are lower diatom proportions at HGIV, F4, F2, and KB0, and higher diatom proportions at F13, F10, and F8 (although FS1 does not match) as discussed above. The westward stations again had a lower proportion of ciliates than those eastward (except for FS1, and F8 as with microscopy). The station with the highest proportion of ciliates was F2 for both microscopy and metabarcoding. However, the microscopy analysis found higher proportions of small diatoms at all stations except NT2 and FS1, and higher proportions of large diatoms at all but FS1. In contrast, microscopy found lower concentrations of pennate diatoms in microscopy than metabarcoding in all but HGIV, F4, F2, and KB0. When comparing the correlations between the methods for each cell group, there were no significant relationships ($p > 0.05$ in all cases; Supplementary Figure 14) other than in the pennate diatoms where there was a negative correlation

between the proportion detected in metabarcoding and microscopy rather than the expected 1:1 relationship ($\rho = -0.71$, $p < 0.01$).

In summer 2019, the microscopy analysis found that ciliates and flagellates were the dominant microplankton types at most of the sampled stations, whereas large centric- and pennate diatoms were scarce throughout. Small centric diatom biomass was generally low but increased at the stations near the north of Svalbard (D2 and D3) as opposed to NT11 and the ice stations IS1 and IS2 (Jenkins et al., in prep.). Overall, the metabarcoding data showed similar patterns to the microscopy data, with flagellates dominating most stations. However, the differences between the proportions were greater than in spring 2018. The proportions of ciliates were much greater in microscopy than with metabarcoding at every station, and conversely the proportions of dinoflagellates and pennate diatoms were lower in microscopy than metabarcoding at all stations other than D2 and D4 for dinoflagellates and IS2 for pennate diatoms. When comparing the correlations between the methods for each cell group, there were no significant relationships ($p > 0.05$ in all cases; Supplementary Figure 15) other than in the ciliates, where there was a positive relationship between the proportion detected by metabarcoding and by microscopy ($\rho = 0.83$, $p < 0.01$).

There was less agreement between the flow imaging microscopy data and the metabarcoding data than the microscopy data and metabarcoding data, due to the influence of pennate diatoms, dinoflagellates, and ciliates and the relationship was not significant ($\rho = 0.17$, $p = 0.15$). Despite this, the main conclusion that the proportions of diatoms were higher in spring 2018 whereas the proportion of flagellates were higher in summer 2019 was maintained across datasets.

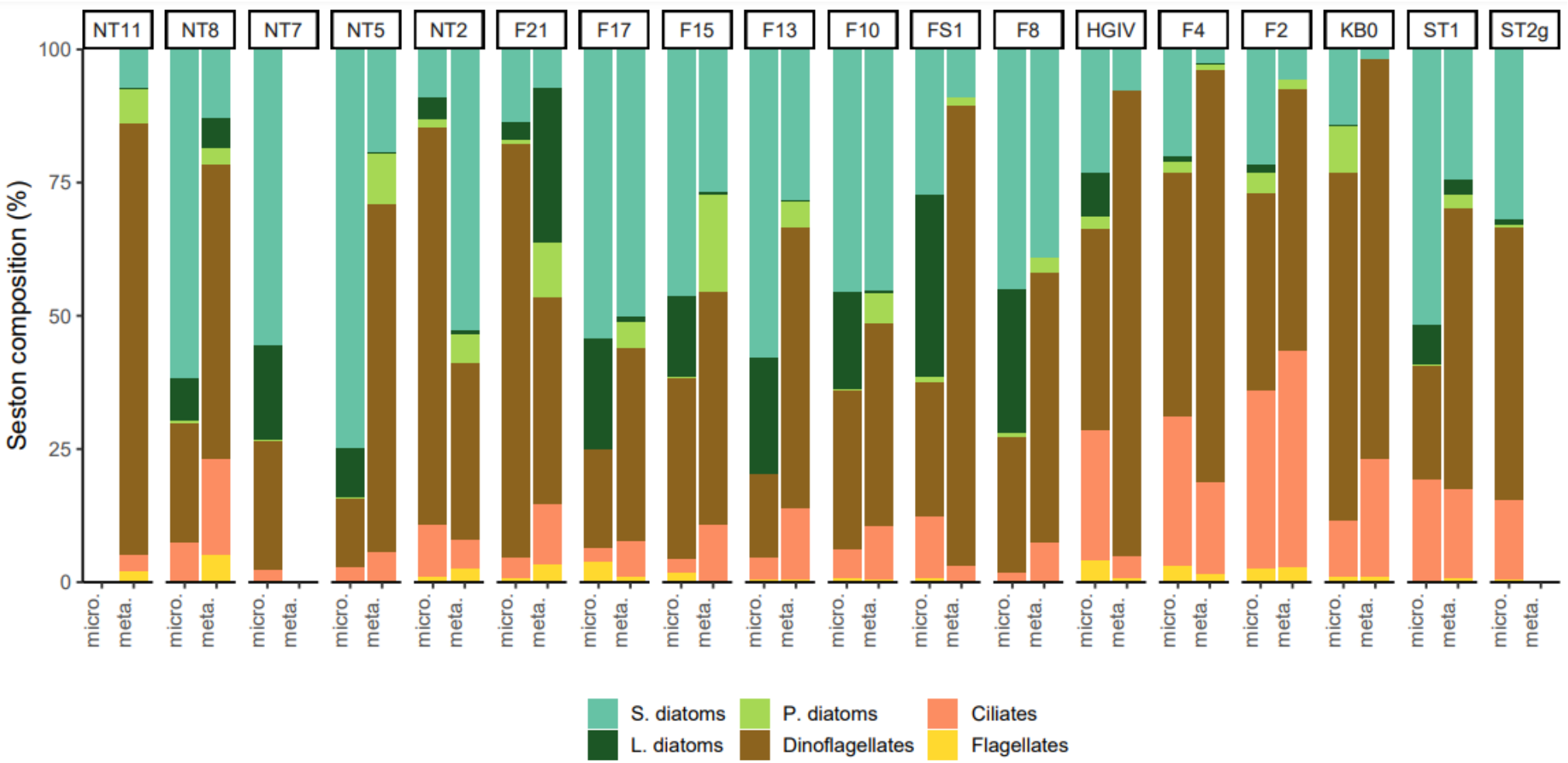


FIGURE 20 | The microplankton food environment for *C. finmarchicus* across the Fram Strait in May and June 2018, comparing methods of analysis. Micro. denotes analysis using inverted microscopy. Meta. denotes metabarcoding analysis of the 18S nuclear small subunit ribosomal RNA. Small and large centric diatoms are denoted by S. and L. diatoms, while pennate diatoms are denoted as P. diatoms.

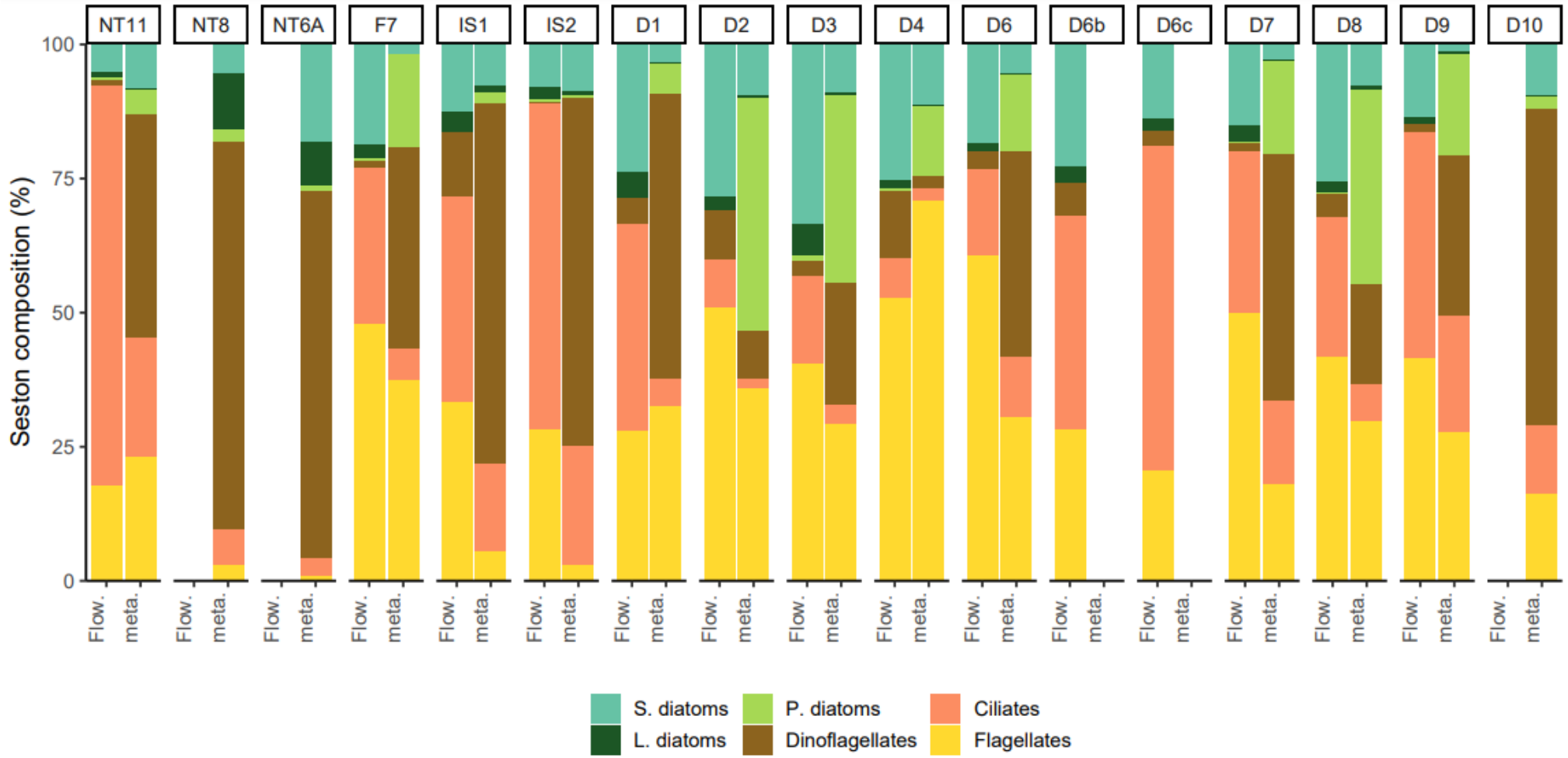


FIGURE 21 | The microplankton food environment for *C. finmarchicus* across the Fram Strait in August 2019, comparing methods of analysis. Flow. Denotes analysis using Flow Imaging microscopy via a FlowCam. Meta. denotes metabarcoding analysis of the 18S nuclear small subunit ribosomal RNA. Small and large centric diatoms are denoted by S. and L. diatoms, while pennate diatoms are denoted as P. diatoms.

4.4.5 Toxic and harmful microplankton

A total of 27 toxic or potentially harmful species (known as harmful from here on out) were found across the stations sampled. The majority of the seston was comprised of species that are non-toxic; 81.0 % in spring 2018 and 79.0 % in summer 2019 (FIGURE 22). On average the seston in spring 2018 comprised of 18.6 ± 9.6 % toxic dinoflagellates, ranging from 5.9 % at F2 to 42.1 % at NT11.The seston in summer 2019 has a slightly smaller average percentage of toxic dinoflagellates at 12.6 ± 8.8 %, ranging from 0.6 % at D4 to 31.3 % at NT8. Interestingly, there were only very small proportions of toxic diatoms, toxic eukaryotes and toxic flagellates at all the stations in spring 2018 (each < 0.25 %), whereas in summer 2019 there were 4.3 ± 2.7 % toxic diatoms, ranging from 0.1 % at IS2 to 8.6 % at D9 and 3.6 ± 3.7 % toxic flagellates, ranging from 0.1 % at NT6a to 13.5 % at D4. Of the toxic dinoflagellates, *Karenia mikimotoi* was found through both spring 2018 and summer 2019 and is a known toxin producer (Gill & Harris, 1987; Hansen, 1995; Poulin et al., 2011). The harmful diatoms found included *Pseudo-nitzschia australis* and *Skeletonema* sp. Of the harmful flagellates, *Aureococcus anophagefferens* and *Pelagomonas calceolata* were found only in summer 2019, both of which create brown tide events possibly fuelled by urea (Gobler, Lonsdale & Boyer, 2005; Guérin et al., 2022).

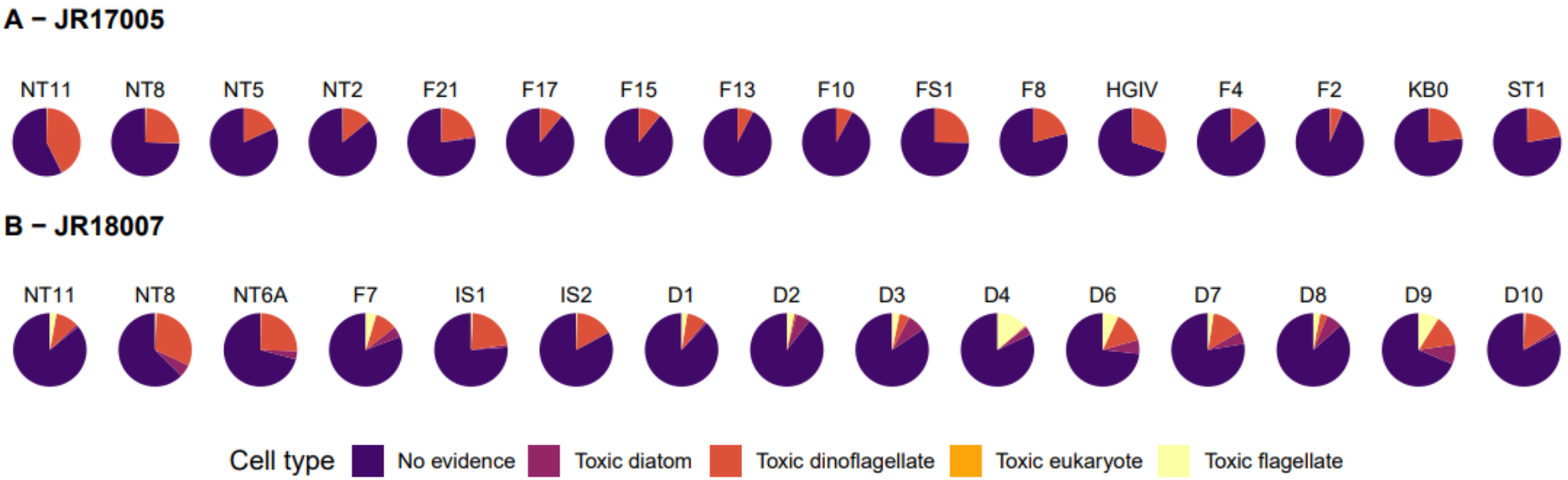


FIGURE 22 | The proportional composition of the seston in the Fram Strait during Spring 2018 on research cruise JR17005 (A) and in Summer 2019 on research cruise JR18007 (B). The seston is classified by its potential toxicity, in this case including harmful organisms with the toxic, based on the literature.

4.5 Discussion

4.5.1 The dominant taxa across the plankton assemblage

The metabarcoding data showed the genus *Calanus* dominated the seston across most stations, despite there only being three species within this group (bacteria was also a dominant category but that contained 53 species). *Calanus* spp. were present at all stations other than NT8. The presence of the genetic matter could indicate any stage of their lifecycle, from eggs to copepodites to adults, as well as potentially faecal pellets, moults and exuviae. Zooplankton biomass in the Fram Strait is 70-92 % in the subclass Copepoda (Hop et al., 2006), and this analysis confirms this – the only other zooplankton genera that were identified were *Edwardsia*, *Hiatella*, *Macoma*, *Bolinopsis*, *Martensia*, *Resomia*, *Halomonhystera, Codonellopsis*, *Cymatocylis*, and *Xystonella*, all with relatively low frequencies. This highlights the importance of these copepods in the Arctic ecosystem.

Prorocentrum micans was the other main dominant species present and had high relative abundance in both spring 2018 (13.2 %) and summer 2019 (8.0 %). *P. micans* is a thecate dinoflagellate, meaning it is covered with cellulose plates, and is a high quality food for copepods due to higher carbon to volume ratio than similar sized athecate dinoflagellates (Menden-Deuer & Lessard, 2000). There is evidence that *P. micans* supports normal rates of egg production and hatching success in *C. finmarchicus* (Starr, Runge & Therriault, 1999), even with a monospecific diet. The small decrease in abundance between spring 2018 and summer 2019 may be due to natural variation and patchiness.

4.5.2 Seasonal and temporal differences in the seston composition

At 6.1 ± 1.3 °C, the average temperature at the chlorophyll maxima in summer 2019 was 6 °C warmer than the that of spring 2018, at 0.1 ± 1.9 °C. One key difference between the seston in spring 2018 and summer 2019 was the much larger proportion of flagellates in the latter, rising from 0.6 ± 1.0 % of the seston in spring to 6.6 ± 5.2 % of the seston in summer. Future warming scenarios for the Arctic suggest a change to a picoplankton-based system (Li et al., 2009). The mechanism behind this is thought to be that ice retreat allows wind-driven upwelling of deep nutrient-rich waters and a longer growing season, but Arctic phytoplankton remain limited by nitrogen supply. The smaller picoplankton have a larger surface-area: volume ratio which increases the efficiency of nutrient uptake, and have hydrodynamic resistance to sinking, so they outcompete the larger phytoplankton (Li et al., 2009). This is an established concept and is

corroborated by decreasing trends in the abundances of large phytoplankton and increasing picoplankton (e.g. *Micromonas* sp. and haptophytes) during a warm anomaly in the Fram Strait in 2005-2007 (Onda et al., 2020). The relative proportion of flagellates increased significantly with temperature in summer 2019. The seston composition of summer 2019 holds some parallels to these results and could be viewed as the simulation of the seston composition in the spring of the future.

Another key difference was the presence of the sea-ice diatom *Navicula* spp. in all stations except the most southerly in spring 2018, while these species were absent from all except IS2 in summer 2019. This taxa is recognised as a sympagic one (Poulin et al., 2011) and its presence is a reflection of the proximity of the ice shelf. Station IS2 in summer 2019 was 'Ice Station 2' where sampling occurred next to ice (although this was also the case in IS1). It has been suggested that while *C.* glacialis utilise ice algae, *C. finmarchicus* rely solely on phytoplankton (Leu et al., 2011), but as feeding during these periods in the Fram Strait was mostly unselective (Jenkins et al., 2022), it follows that *C. finmarchicus* also ingests ice algae.

The change towards a smaller size-spectra can mean more steps to the food web. Using the metabolic theory of ecology, the ratio of microzooplankton herbivory to phytoplankton growth will change in a warming environment due to the different temperature dependencies of autotrophic and heterotrophic organisms, and therefore warming may enhance phytoplankton losses to microzooplankton herbivory (Chen et al., 2012). For example, with the higher proportion of ciliates found in summer 2019, it is likely that larger ciliates are feeding on smaller ones and there is an overall increase in microzooplankton grazing instead of the traditional food chain of nutrient to phytoplankton to copepod. For every extra loop added to the food web, the efficiency of nutrient transfer to the higher trophic levels is reduced, and more dissolved organic molecules are released to become available for heterotrophic bacterial consumption (Azam et al., 1983; Lampe, Nöthig & Schartau, 2021). The bacterial proportion of the seston in summer 2019 (37.8 ± 11.1 %) was almost double that of spring 2018 (18.2 ± 10.8 %), suggesting high amounts of dissolved organic matter, with the possibility of more microzooplankton grazing and a more active microbial loop. This research adds to the body of literature which suggests a more active picoplankton size fraction in the future Arctic (Nöthig et al., 2015; Fadeev et al., 2018; Szeligowska et al., 2020; Lampe, Nöthig & Schartau, 2021).

4.5.3 Harmful algal blooms in the future Arctic Ocean

Harmful algal blooms (HABs) occur when toxic or ecosystem-disruptive algal species reach a threshold abundance. This threshold abundance is the point at which it is deemed the bloom be

damaging – the concept of a HAB is a societal one rather than a specific definition. The warming of the oceans seems to be intensifying HABs worldwide (Glibert et al., 2014; Gobler et al., 2017). Some of those classified here are more dangerous than others, for example *Karenia mikimotoi* has caused mass mortality of fish, shellfish, and almost all marine organisms (Li et al., 2019), while *Scrippsiella* spp. forms blooms that are non-toxic but high density and so may cause oxygen depletion (Liu, Zhang & Wang, 2022). Dense blooms of any of these species are likely to at least reduce the feeding of juvenile fish (Esenkulova et al., 2022), so an increase in HABs in the Arctic would have negative impacts on fisheries. Approximately 15 % of the world's marine fishes are caught in the Arctic and Subarctic (Zeller et al., 2016), so further research needs to be done to understand HABs in these areas.

There were harmful diatoms (particularly *Pseudo-nitzschia australis*) and flagellates (*Aureococcus anophagefferens*) found in summer 2019 and not in spring 2018, which could suggest this a pattern of plankton succession for warming waters. However, this is purely speculation as this study is limited by the short duration of sampling. Long term studies scaling the full seasonal cycle are needed to do anything more than speculate about changes in blooming and toxin-producing species. That said, it has been previously reported that typical Arctic seasonal succession comprises of a spring bloom dominated by diatoms followed by dinoflagellates in summer, with an increase in potentially toxic taxa (Bruhn et al., 2021).

4.5.4 Methodology consensus and divergence

Overall, the composition of the seston measured by inverted microscopy correlated as expected with the composition measured by metabarcoding. There were some key differences, particularly in the proportions of diatoms and the dinoflagellates (Supplementary Figure 14) – mainly that the proportion of large diatoms was lower in the metabarcoding analysis than the microscopy, and the proportions of pennate diatoms and dinoflagellates were greater. Smaller diatoms could be wrongly classified as flagellates at suboptimal magnifications under an inverted microscope, if the sample had been agitated too roughly and diatom chains had been broken down. However, this would increase the proportion of flagellates in the samples which would still not match the metabarcoding data – it is the dinoflagellate category where the difference lies. Additionally, it is the large diatom category where the difference is more pronounced. It is unlikely that large and distinct dinoflagellates were wrongly counted as large diatoms in the microscopy data. It is more likely that the number of diatoms counted in each methodology were similar, but due to the proportional representation, the high proportion of dinoflagellates in the metabarcoding data skewed the proportion of diatoms.

There were higher proportions of dinoflagellates in the metabarcoding analysis of both spring 2018 and summer 2019. This was unexpected, as there is evidence that dinoflagellates are sometimes not detected by generalist primers or even when primers are specifically aimed at them (Sildever et al., 2021), so this shows the primers were suitable. Metabarcoding detects the genetic material of dinoflagellates independent of their life stage, whereas imaging will only identify their trophic form. Approximately 13–16% of living dinoflagellates produce a dormant "resting cyst", though their pelagic distribution is not well understood (Head, 1996; Marret et al., 2020). Therefore, it is possible that the metabarcoding dinoflagellate proportion is higher due to the detection of these cysts. Some of these cysts (e.g. *Islandinium* spp.) are important for reconstruction of the paleoclimate due to their modern and ancient presence in the Arctic (Head, Harland & Matthiessen, 2001), so their detection has benefits for tangential studies.

A greater number of dinoflagellates being detected would explain the lesser proportion of diatoms in spring 2018, but the proportion of diatom categories in summer 2019 were similar across both flow imaging and metabarcoding, when the diatom categories were taken together. It seems that discrepancies here come from the type of diatom rather than the category in general. If diatoms classified as pennate in summer 2019 were classified as small diatoms, the flow imaging and metabarcoding results would have been more in line. However, this error is unlikely, even when considering the point of view of the particles for imaging, as some pennate diatoms would have been too large to have been classified as small centric diatoms. It is more likely that a greater proportion of pennate diatoms were found with metabarcoding in summer 2019 instead. It is possible that pennate diatoms move through the flow imaging cell at a faster rate than other cells due to their shape, and therefore could be less likely to be photographed with the sufficient edge ratio, however pennate diatoms have been detected at high concentrations in other image-based studies (e.g. Sosik & Olson, 2007).

In contrast, there were less ciliates identified in the metabarcoding dataset than the flow imaging one. This is possibly due to the overestimation of size and therefore volume of the ciliate category due to morphology and a low edge gradient. An alternative explanation is that there are undeveloped references assignments in the database used for taxonomic alignment here – ciliates have been highlighted as a group that needs extra research (Rajter & Dunthorn, 2021).

It is likely that the metabarcoding dataset is not the complete picture of the diversity of the samples. The lack of replicate samples and lack of replication of unique reads suggests some rare and low-abundance species are missing, and others might be overrepresented. This could potentially have a large effect on the data because relative abundance is estimated directly from the number of DNA sequences assigned. No metrics of biodiversity have been calculated using

this dataset as the low sampling depth suggests that the true alpha diversity is not well represented in these samples. More replicates would likely have increased the measured alpha diversity and given a higher sampling depth (Alberdi et al., 2018). However, due to the patchy nature of plankton in the Fram Strait (Jenkins et al., 2022,in prep.), it is likely that many replicates would be needed to increase the reliability of the data by a meaningful amount.

4.5.5 Advantages and disadvantages of the methods

4.5.6 Metabarcoding

Metabarcoding is, by definition, is a high-throughput sequencing method, and the high-throughput itself means that a large amount of data can be obtained with a relatively small amount of labour. Metabarcoding can detect plankton taxa that are difficult to distinguish by morphology – e.g. those of small sizes, eggs, larvae and cysts – and so give more accurate estimations of proportional abundance (MacNeil et al., 2021; Pierella Karlusich et al., 2022; Wang et al., 2022). However, it cannot distinguish between these life stages themselves, and each has a distinct ecological niche, so it is not always preferable to have these stages count towards that group's abundance. When functional traits, metabolic pathways, and ecological interactions are of interest, metatranscriptomics is a more relevant method (Stewart, 2013; Zhang et al., 2019).

Metabarcoding can provide information on the genetic diversity and population structure within the plankton (Pierella Karlusich et al., 2022). However, the methods used here are only semi-qualitative and are so limited to relative abundances. The carbon and other nutrient or macronutrient biomass of the plankton is often the focus of the research as it can constrain and improve biogeochemical models (Lombard et al., 2019), be used to approximate carbon sequestration (Henson et al., 2022), and help examine potential changes in a warming future. When quantitative measurements are needed, metabarcoding must be combined with an addition of artificial communities of known concentrations to serve as internal standards (Santoferrara, 2019).

The results of a metabarcoding analysis depend greatly on the database to which sequences are compared. In the past, database incompleteness was a great problem that meant, for example, that there was only 24 % taxonomic assignment success (Abad et al., 2016). Steady progress is being made in adding to these databases, and in this study, 60.7% were successfully taxonomically assigned. This figure could have been higher if region-specific databases were used (Questel et al., 2021), but there is less coverage in the Arctic Ocean than other oceans. The results of the analysis also depends on the marker and the primers, with some being more effective for certain types of plankton (Esenkulova et al., 2020). The primers chosen for this study have been effective in

revealing diverse marine metazoan communities, exhibited pronounced homology across meiofaunal phyla, and also flanked a highly divergent region of the 18S nuclear small subunit (Fonseca et al., 2010). However, metabarcoding with a combination of markers (i.e., 16S, 18S-diatom, 18S-dinoflagellate, and 28S) has proven to be a better technique for phytoplankton community structure analysis (Esenkulova et al., 2020).

The most important problem with the metabarcoding methodology for detecting all plankton species is the largely varying 18S rRNA gene copy numbers among marine protists, ranging from tens to thousands (Martin et al., 2022). This variability can lead to a rapid decline in concordance between the read number and the organismal abundance (Gong & Marchetti, 2019). There have been efforts to correct for this effect – if the gene copy number can be estimated and it can be assumed that one read is equal to one gene copy number, the median gene copy number per plankton group can correct the relative abundances (Martin et al., 2022). However, this method has so far only improved estimates of dinoflagellate and ciliate abundances and not those for the diatoms, possibly due to large biovolume plasticity in the diatoms causes a larger variety in gene copy number (Martin et al., 2022).

4.5.7 Inverted microscopy

Inverted microscopy is extremely labour-intensive and requires a higher level of expertise with an increasingly detailed taxonomic level; classification of functional groups needs only basic expertise, whereas genus level needs a great deal of experience, and it is often not possible to identify to species level regardless of expertise. However, information about sizes, life stage and morphology can be collected. When the size and count are combined, accurate estimations of the carbon contained within the plankton can be made (e.g. Jenkins et al., 2022). Other than the labour-intensive nature of inverted microscopy analysis, there are also preservation artifacts due to the addition of acidified Lugol's iodine or equivalent preservative. Lugol's iodine has been found to change cell abundances, especially in the nanoplankton size range through the formation of aggregates, to reduce cell sizes, and to not efficiently preserve certain species (Zarauz & Irigoien, 2008). However, with a mixed natural assemblage and low concentration of Lugol's iodine, these effects are small (Menden-Deuer, Lessard & Satterberg, 2001). Overall, microscopy data better reflects real phytoplankton abundance and community structure than metabarcoding does (Wang et al., 2022).

4.5.8 Flow imaging microscopy

Flow imaging microscopy using the FlowCam® 8400 (Yokogawa Fluid Imaging Technologies LLC) allowed a large number of particles to be imaged, counted and sized very quickly. However, there was a significant amount of processing required to taxonomically assign these particles and diversity in the morphology within taxonomic groups in the training dataset can reduce the efficiency of the algorithm, necessitating manual verification for approximately 60% of images to confirm particle identity and remove non-target particles (Jenkins et al.,in prep..). In this case, labour costs are almost as high as with traditional microscopy. Different machine learning algorithms, custom-built for purpose, can improve identification success, for example the accuracy of the FlowCytobot is 88 % for phytoplankton at genus level (Sosik & Olson, 2007).

The FlowCam 8400 is designed for particles sized from 2 µm to 1000 µm. However, the images towards the smaller end of that range are often blurred and so the taxonomic assignment and cell volume calculations are less accurate. For plankton with cilia and appendages, accurate measurement of the cell's dimensions become more difficult and the VisualSpreadsheet software does not accurately calculate the volume. Instead, taxonomic groups within the software must be of similar sizes and a sample must be measured manually to calculate an average volume for further estimates of carbon content.

Flow imaging microscopy has the same limitations as traditional microscopy in terms of preservation artifacts, although the speed of processing can allow swift analysis and reduce the amount of time in which preservation artifacts come about.

4.6 Conclusion

Overall, this study agrees with previous work in that metabarcoding provides a better insight into the diversity of an ecosystem while microscopy provides more accurate quantitative results regarding abundance and biomass (Santi et al., 2021; Pierella Karlusich et al., 2022). The seasonal patterns in relative read abundance of major phytoplankton groups was well in accordance with microscopical carbon in spring 2018 as in another study (Gran-Stadniczeñko et al., 2019), although not so well in summer 2019. Flow imaging microscopy did not correlate with the metabarcoding data, likely due to incorrect assignment of inert particles as small diatoms and pennate diatoms as large diatoms (Supplementary Figure 10), both because of the imaging process. As artificial intelligence improves at an exponential rate, it is likely that the taxonomic assignment of images will improve too. Coupling this with better specifications in the camera that take the images, and it can be expected that in future flow imaging will be preferable to manual microscopic methods.

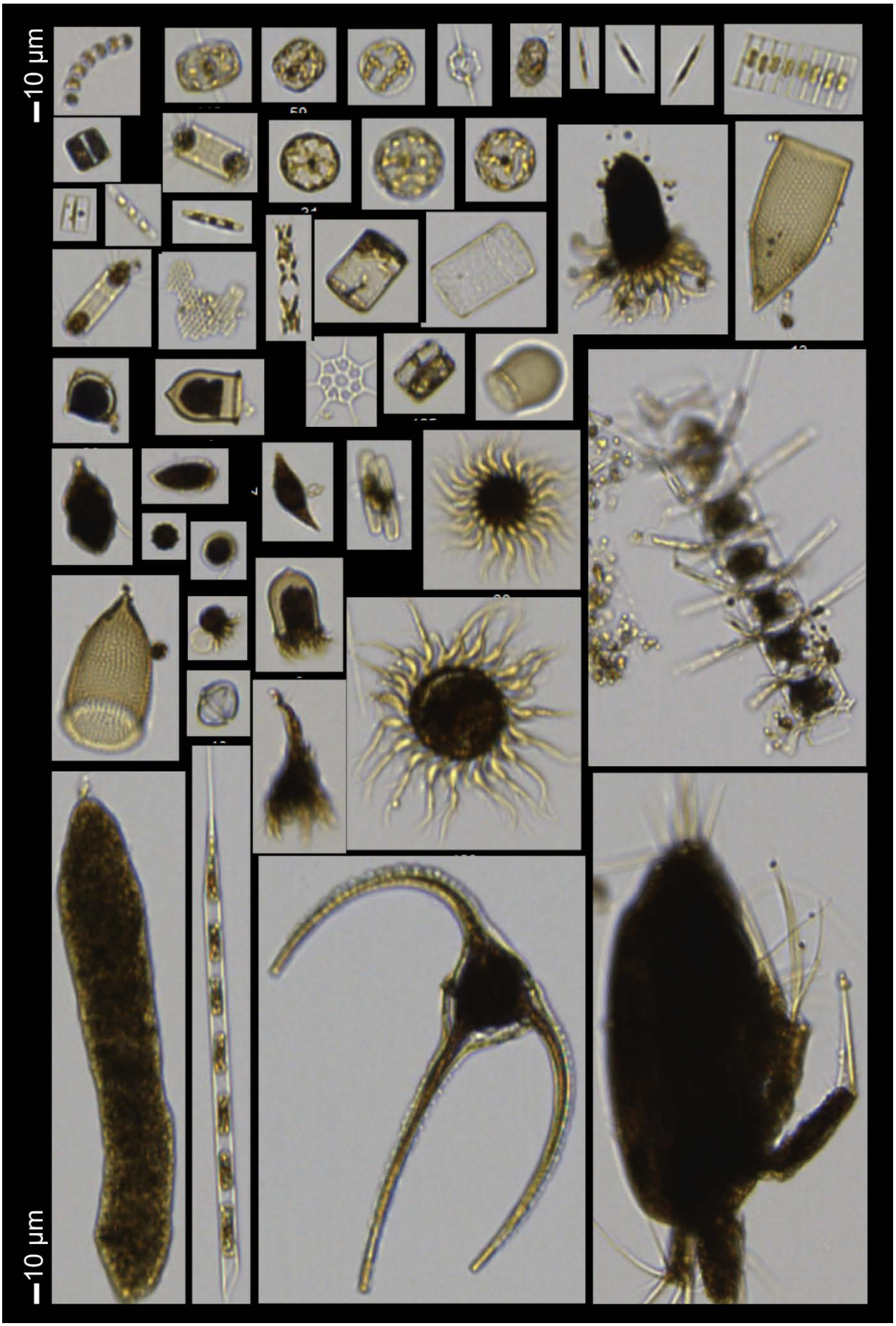


FIGURE 23 | Images taken by the FlowCam of the microplankton of the Fram Strait in spring 2018 and summer 2019.

Chapter 5 Final discussion

5.1 Changes to primary production

The waters of the Fram Strait increased in temperature by 0.73 °C per decade during 1980 – 2016 (Goszczko, Ingvaldsen & Onarheim, 2018). General ecological theory suggests warming of the oceans will directly or indirectly lead to a shift to smaller sized cells through decreased individual size and/or replacement of the larger species with smaller ones (Daufresne, Lengfellner & Sommer, 2009). There is a wealth of evidence to support this; experimental (Coello-Camba et al., 2014), observational (e.g. Li et al., 2009; Morán et al., 2010), modelled (e.g. Carozza, Bianchi & Galbraith, 2019; Henson et al., 2021), and even fossilised (Finkel et al., 2005). The data in Chapter 4 follow this trend with a higher proportion of small flagellates in the warmer sampling period. However, these results reflect seasonal changes rather than climate change. How much does the seasonal plankton succession in the Arctic resemble future warming? Here, I examine whether the parallels between late summer plankton and the expected future plankton assemblages extend further than temperature, and so assess the validity of this assumption.

The Arctic is no longer a region dominated by thick multi-year ice, but is facing a regime controlled by thinner, fragmented, and more dynamic first year ice (e.g. Aksenov et al., 2021). This transition to first year ice can increase light transmission to the surface waters by 200 % (Castellani et al., 2022b), reducing ice thickness and potentially increasing the growing season for ice algae inhabiting the water beneath. Currently, blooms of ice algae begin by February to mid-March (Bluhm et al., 2011), but models predict an increased likelihood of blooms happening earlier in the season in future (Post et al., 2013; Tedesco, Vichi & Scoccimarro, 2019; Lannuzel et al., 2020). This shift in the timing of the blooms is restricted by solar angle and thus the shift may not be able to remain synchronised with the ice in future (Leu et al., 2011). For this reason, the reduction in ice could mean a reduction in ice algae (Barber et al., 2015). The ice algae may 'seed' diatoms which help to stimulate the pelagic phytoplankton bloom (Haecky, Jonsson & Andersson, 1998; Benkort et al., 2020), an effect which can be seen by the ice algae species present at all Arctic spring stations, and in one of the two ice stations in summer in Chapter 4. The full effect of the reduction of the sympagic system on the pelagic system is still unclear. It is logical, however, that earlier pelagic blooms of the Arctic in future, due to reduced ice cover, could shift the plankton community succession forward by some months and create summer-like conditions in spring.

The future of the Arctic Ocean is likely to be a largely ice-free Arctic Ocean during summer, in view of the 2 °C warming threshold (Notz & Stroeve, 2018), which we are predicted to reach mid-century (Diffenbaugh & Barnes, 2023). The change to a more Atlantic-like, ice-free dynamic may increase nutrient availability and the duration of seasonal drawdown of nutrients in Arctic shelf regions (Sakshaug & Slagstad, 1992; Reigstad et al., 2002; Tremblay et al., 2008), but the extent to which this increased nutrient availability and longer drawdown periods will lead to increases in primary production will depend on changes in upper ocean mixing and stratification (Henley et al., 2020). An increase in stratification across the Arctic is ongoing (Sallée et al., 2021) and this is expected to intensify (Kwiatkowski et al., 2020), but the extent of this has thus far proved variable. The Fram Strait has contrasting dynamics: the Atlantic-water dominated West Spitsbergen Current has a stratified water column and a shallow nitracline, resulting in reduced nitrogen limitation. To the contrary, the Polar-water dominated EGC has greater nitrogen limitation due to stronger stratification inhibiting nutrient resupply from deeper water, and low lateral nitrate supply from central Arctic waters (Tuerena et al., 2021). The loss of winter sea ice, the resultant increase in wind-driven mixing (Lewis, Van Dijken & Arrigo, 2020), and continued atmospheric warming has the potential to inhibit deep winter mixing and limit primary production in the eastern Fram Strait in the future (Tuerena et al., 2021). A nitrogen-limited plankton assemblage is favourable to smaller phytoplankton with a larger surface area to volume ratio, and also more favourable to more motile microzooplankton (Tremblay & Gagnon, 2009; Ardyna et al., 2011; Mousing, Richardson & Ellegaard, 2018). In contrast, diatoms have a high affinity for nitrate uptake (Glibert et al., 2016) and silicate. The prevalence of smaller species in summer 2019 in Chapter 3 & Chapter 4 suggests a nitrogen-limited environment, which is confirmed by parallel measurements of the nutrient conditions in the Fram Strait during the same period (Tuerena et al., 2022). There is additional evidence that nitrogen co-limits primary production with iron and silicate (Krause et al., 2019; Krisch et al., 2020), with silicate concentrations decreasing below 1 mmol in the summer months, limiting diatom abundance. This effect is seen clearly in the low abundance of diatoms presented in Chapter 4. Here, the presence of *Phaeocystis* sp. across both spring and summer suggests that nutrient limitation could have begun in June.

Changes to the nutrient supply, light conditions and temperature alter the composition of the plankton assemblage, both in terms of species composition and the composition of individual organisms. Autotrophic organisms take up nutrients and carbon separately, as carbon dioxide fixation and nutrient uptake are only loosely coupled, resulting in flexible stoichiometry between the two. While the C:N ratio of phytoplankton is variable under nutrient limiting conditions, when not limiting, it loosely follows the Redfield ratio of 106:16:1 C:N:P (Redfield, 1934; Finkel et al., 2010). However, in a changing Arctic, increasing stratification of the ocean promotes a greater

C:N ratio in the phytoplankton (Díez, Van Nieuwerburgh & Snoeijs, 2013; Clark, Flynn & Fabian, 2014; Eberlein et al., 2016; Tanioka & Matsumoto, 2020) through reduced nutrient supply to the surface waters (Bopp et al., 2001; Steinacher et al., 2010). Dinoflagellates are hetero- or mixotrophic and thus do not follow the Redfield ratio, but have a higher C:N ratio of 90:12 (Carnicer, Irwin & Finkel, 2022). Further, smaller-sized and thecate taxa are even higher in C:N than larger-size and athecate taxa (Carnicer, Irwin & Finkel, 2022). In general, diatoms contain higher proportions of carbohydrate than dinoflagellates or flagellates (Jónasdóttir, 1994). Flagellate C:N is very varied depending on species and growth phase. Different types of microplankton also contain differing levels of micronutrients. Diatoms contain more of the omega fatty acid eicosapentaenoic acid (EPA) while flagellates and dinoflagellates contain more docosahexaenoic acid (DHA) (Jónasdóttir, 1994). Both the ratio of N:C and the ratio of DHA:EPA can be used as approximations of food quality for these organisms, and justify the collection of data of functional groups as a proxy for quality.

Changes to the plankton assemblage can also increase the concentrations of some toxic diatoms, for example *Pseudo-nitzschia* spp., many types of which produce the potent neurotoxin, domoic acid (Price et al., 2021; Campbell et al., 2021). The research in Chapter 4 shows the appearance of *Pseudo-nitzschia australis*, another species known to produce domoic acid in varying quantities (Hubbard et al., 2023). Domoic acid is transferred through trophic levels via planktivorous fish through to marine mammals and seabirds. Accumulation in an organism can lead to domoic acid poisoning in wildlife. Moreover, consumption of seafood contaminated with domoic acid once led to Amnesic Shellfish Poisoning in humans, causing vomiting, memory loss, coma and death (Bates et al., 1989). The data in Chapter 4 are a snapshot, which does not capture natural or interannual variability, so the presence of *P. australis* could be caused by a rare factor that has not been accounted for. However, it could suggest that the future Arctic conditions will be more favourable to this toxic diatom. In addition, *Pseudo-nitzschia* spp. can interact with *Calanus* copepodites. When *Pseudo-nitzchia seriata* comes into close proximity to *Calanus* spp., its toxicity may increase by up to 3300% (Harðardóttir et al., 2015). This interaction between *Pseudo-nitzschia* spp. and *Calanus* spp. is potentially worrying given the high abundance of the latter. Other potentially harmful algal blooms are expanding northwards with the increase in temperature (e.g. *Alexandrium tamarense* (Natsuike et al., 2013)). These toxic and harmful species pose a risk to coastal communities who rely on the Arctic coastlines to harvest food. They also pose a significant risk to the developing fisheries in the Arctic Oceans – ones that are already facing competition to exploit the newly ice-free areas (Mendenhall et al., 2020).

Understanding the effects of climate change on microplankton can be both complex (meaning with many components) and complicated (meaning many relationships between these

components), because the effects can be attributed both to the direct effects of temperature on the organisms but also to indirect effects mediated by grazing and nutrient supply – for example, the effect of temperature lessens under nutrient limitation (Sommer et al., 2017 and references within). As another example, the interaction of temperature and salinity changes may have a synergistic effect on reducing the diversity of Arctic diatoms due to their effect on density, and therefore stratification (Sugie et al., 2020). The interacting variables necessitate careful study, and experiments to separate them are often too simplistic to have real-world applications. For example, one of the only published studies which did not find a relationship between plankton size and temperature was aiming to elucidate the impact of temperature alone, and thus used monoculture conditions without the impact of nutrient cycling or changes to grazing pressure by zooplankton (Rüger & Sommer, 2012). However, when we do understand the microplankton, the abundance and diversity of plankton can be used as a multivariate predictor of zooplankton composition and function, as well as giving insight on nutrient conditions and even ocean-wide carbon sequestration.

5.2 Changes to secondary production

Copepods of the genus *Calanus* are vital to the functioning of the Arctic, both in terms of the biogeochemical functioning of the ocean and in terms of their role in the transfer of nutrients to higher trophic levels. The grazing and production rates presented in Chapter 2 & Chapter 3 of this thesis are vital measurements essential to all other calculations on these two topics.

As discussed in Chapter 1, there has been a long debate on whether the quantity of the food environment limits production or whether the quality does. As the C:N ratio of the phytoplankton increases, a quantity limited organism would maintain the same production, whereas a quality limited organism would reduce production. Thus, this question is vital to the function of the Arctic Ocean. It has been suggested that the fatty acid composition (particularly the DHA:EPA ratio) could limit copepod production (Jónasdóttir & Kiørboe, 1996; Arendt et al., 2005; Vehmaa et al., 2011). These fatty acids are essential for copepods to grow and reproduce, and have to be derived from prey, because copepods are unable to synthesize them *de novo* in sufficient quantities (Jónasdóttir, Fields & Pantoja, 1995; Jónasdóttir, Visser & Jespersen, 2009; Kleppel & Hazzard, 2000). However, it seems that limitation by these fatty acids mostly occurs in experimental conditions with monospecific diets (Anderson & Pond, 2000).

Chapter 2& Chapter 3 directly relate the microplankton prey to the production of *C. finmarchicus* and provide experimental data to find the limiting factor. Simultaneously, new state-of-the-art ecological stoichiometry models were developed (Anderson et al., 2017, 2020; Anderson, Hessen

& Mayor, 2021). These models now represent metabolism (hence growth efficiencies), and have explicit terms for biomass turnover, basal metabolism, and specific dynamic action. These advanced models have provided convincing evidence that copepods are limited by C (i.e. the quantity) when consuming food at Redfield C:N (6.625) (Anderson, Hessen & Mayor, 2021). Chapter 2 shows conditions where food was plentiful and diverse, so likely at Redfield C:N, yet still there was no correlation between ingestion and production. Here, the copepods were still undergoing the energetically expensive process of gonad maturation at the time of sampling, an assertion that is supported by the relatively high C:N ratios of the incubated females, the typically low egg production rates, and gonad maturation status. This indicates that there is cause to include an explicit gonad maturation term in future stoichiometric models. In Chapter 3, again there was no significant correlation between ingestion and production. Here, however, the food environment for the copepods was not at Redfield: the phytoplankton biomass was low and nutrient limited, while hetero- and mix-trophic microplankton dominated the plankton assemblage. Therefore, it is possible the copepods were nutrient limited, although as they were also limited by quantity, nutrient limitation would be masked. The copepods were using capital breeding strategies, drawing on their internal reserves to fuel production. In these copepods, the spawned eggs were unlikely to gain the reserves to survive and ultimately to diapause before the polar night, and as such they were nonviable. For summer 2019, this indicates that the sampled copepods were not within the important section of the overall population. However, if these conditions were viewed as analogous to predicted spring conditions in the future, the low ingestion and low production would be a cause of concern for the survival of *C. finmarchicus* populations.

C. finmarchicus populations directly affect the biogeochemical functioning of the Arctic Ocean through their metabolic processes (e.g. respiration (Kaiser et al., 2022)), active migrations, production of faecal pellets (Turner, 2015b) and through their sinking carcasses (Glud et al., 2015). Additionally, *C. finmarchicus* populations indirectly affect the biogeochemical functioning of the Arctic Ocean through the grazing pressures they exert on primary production, leakage of dissolved organic carbon from their faecal pellets (Møller, Thor & Nielsen, 2003), and through particle fragmentation (Mayor et al., 2014). Seasonal migrations to depths below the deep convection layer (diapause) by *Calanus* spp. are also a significant element of the biological pump (Visser, Grønning & Jónasdóttir, 2017).

Diapause in *C. finmarchicus* lasts for over 100 days (Tarling et al., 2022a) at the temperatures encountered in the Fram Strait, and functions to allow the copepods to ensure survival during periods of food shortage – the Arctic winter. During diapause, metabolism is lowered (Saumweber & Durbin, 2006; Maps, Record & Pershing, 2014), development is arrested, and feeding stops

(Hirche, 1996a; Wilson, Heath & Speirs, 2016). The cycle, shown in FIGURE 24, is initiated by adults from the overwintering stock spawning eggs during the spring bloom (Niehoff et al., 1999; Madsen et al., 2008). The eggs that successfully hatch develop through six naupliar stages and six copepodite stages, the latter of which is the fully mature adult (Marshall & Orr, 1955). The timing of this cycle is dependent on many interrelated factors, including temperature, microplankton prey and lipid stores (Hygum et al., 2000).The most important of these factors differs at different stages. For example, there is evidence that early gonad maturation is fuelled by internal reserves (Sargent & Falk-Petersen, 1988; Hirche, 1996b; Rey-Rassat et al., 2002), while the final stage is food dependent (Niehoff et al., 2002). Diapause usually occurs in the late copepodite stages (Bandara et al., 2021). The initiation, regulation and timing of diapause are poorly understood and subject to debate (Baumgartner & Tarrant, 2017), but thought to be reliant on lipid accumulation surpassing a threshold (Rey-Rassat et al., 2002) and an endogenous clock mechanism (Häfker et al., 2018). The timing of diapause is vital to the functioning of the Arctic ecosystem, as the copepods must survive the polar night and return to the surface water alongside the first ice algae (for *C. glacialis*) or pelagic phytoplankton bloom. Potential shifts in composition and timing of these blooms are likely to cause a mismatch between primary production and associated copepod grazing – critical events between interacting species that have been finely tuned over evolutionary time scales (Kharouba & Wolkovich, 2020) – thus compromising the life cycle of the copepods (Søreide et al., 2010). In addition to the changing phytoplankton phenology, climate change may affect the duration, depth, and timing of diapause for *Calanus* (Wilson et al., 2016). But diapause is also vital to sequester carbon at depth – *C. finmarchicus* sequesters an estimated 0.3 Mt C year^{-1} in the Fram Strait alone via the biological lipid pump which, unlike with the flux of detrital material, transports carbon directly to the deep ocean with low loss rates (Jónasdóttir et al., 2015). The relative importance of diapausing copepods to carbon sequestration is made even more important by their deep residency and location in areas of deep-water formation, which may change with a northwards shift (Pinti et al., 2023).

Seston biomass available to *C. finmarchicus* as presented in Chapter 2 was high (61.6 ± 46.6 µmol C L^{-1}), so it can be assumed that feeding would support a successful diapause. In contrast, in Chapter 3, seston biomass was low (0.84 ± 0.33 µmol C L^{-1}), and the carbon budget showed a deficit, implying the *C. finmarchicus* used stored lipids to sustain their reproductive output. Parallel research at two stations of the same research cruise, NT11 and D1, found that lipid reserve levels were sufficient for many individuals to survive the overwintering period and reproduce the following spring (Tarling et al., 2022a). Tarling et al. reported C contents of

23.3 ± 4.7 and 23.1 ± 5.9 µmol C copepod^{-1} at NT11 and D1 respectively (Tarling et al., 2022a), which is comparable to those measured here for Chapter 2: 20.6 ± 3.0 at NT11 (Jenkins et al., in

prep.). Therefore, it could be generally assumed that the experimental animals herein had similar lipid stores and would also be likely to survive diapause. However, it is likely that the animals left at the surface were at the end of their life cycle rather than about to diapause again, as evidenced by the egg production rates, high abundance of mature females at depth, and the fact that they were in carbon deficit in feeding conditions that were unlikely to change.

There is evidence that *C. finmarchicus* can now complete its life cycle in the Arctic (Svensen et al., 2019; Freer, Daase & Tarling, 2021; Tarling et al., 2022b). Before anthropogenic climate change, the Arctic Ocean and parts of the Fram Strait exhibited Arctic- type cold, stratified and ice-covered features. Now, conditions in places resemble a more boreal Atlantic- type warm, well-mixed open-water system – conditions more suited to *C. finmarchicus* (Tarling et al., 2022b). At present, *C. finmarchicus* is likely able to complete its life cycle in the warmer part of the Fram Strait – in waters of the West Spitsbergen current that are ~ 2 °C warmer than those of the East Greenland Current (station D2 in Chapter 3 and Chapter 4, named S3 in Tarling et al. 2022b). As warming continues, the area in which *C. finmarchicus* can successfully complete its life cycle will grow larger. It may even dominate the populations of the traditional Arctic species *C. glacialis* and *C. hyperboreus*. There is not yet consensus on the effect that the replacement of Arctic with Atlantic *Calanus* will have on the higher trophic levels. A shift to smaller species typically means the transfer of energy becomes less efficient. The lipid content of the female *Calanus* spp. in Disko Bay was 34 % less when the area was dominated by *C. finmarchicus* compared to *C. glacialis* (Møller & Nielsen, 2020). However, in other areas there is considerable overlap between the sizes of the two copepods, and the faster development times (Møller et al., 2012) coupled with faster population turnover (Renaud et al., 2018) may compensate for the inefficiency. However, these factors do not compensate for the role of *Calanus* spp. in the food web, as there is evidence that *C. finmarchicus* is too small to replace *C. glacialis* as prey of the Little Auk (Steen et al., 2007). There is no evidence of direct competition between the two *Calanus* spp. in areas of co-existence (Hop et al., 2019; Møller & Nielsen, 2020), so their functional niches cannot be identical. More research needs to be done to understand where these niches overlap and how well *C. finmarchicus* may be able to replace *C. glacialis*.

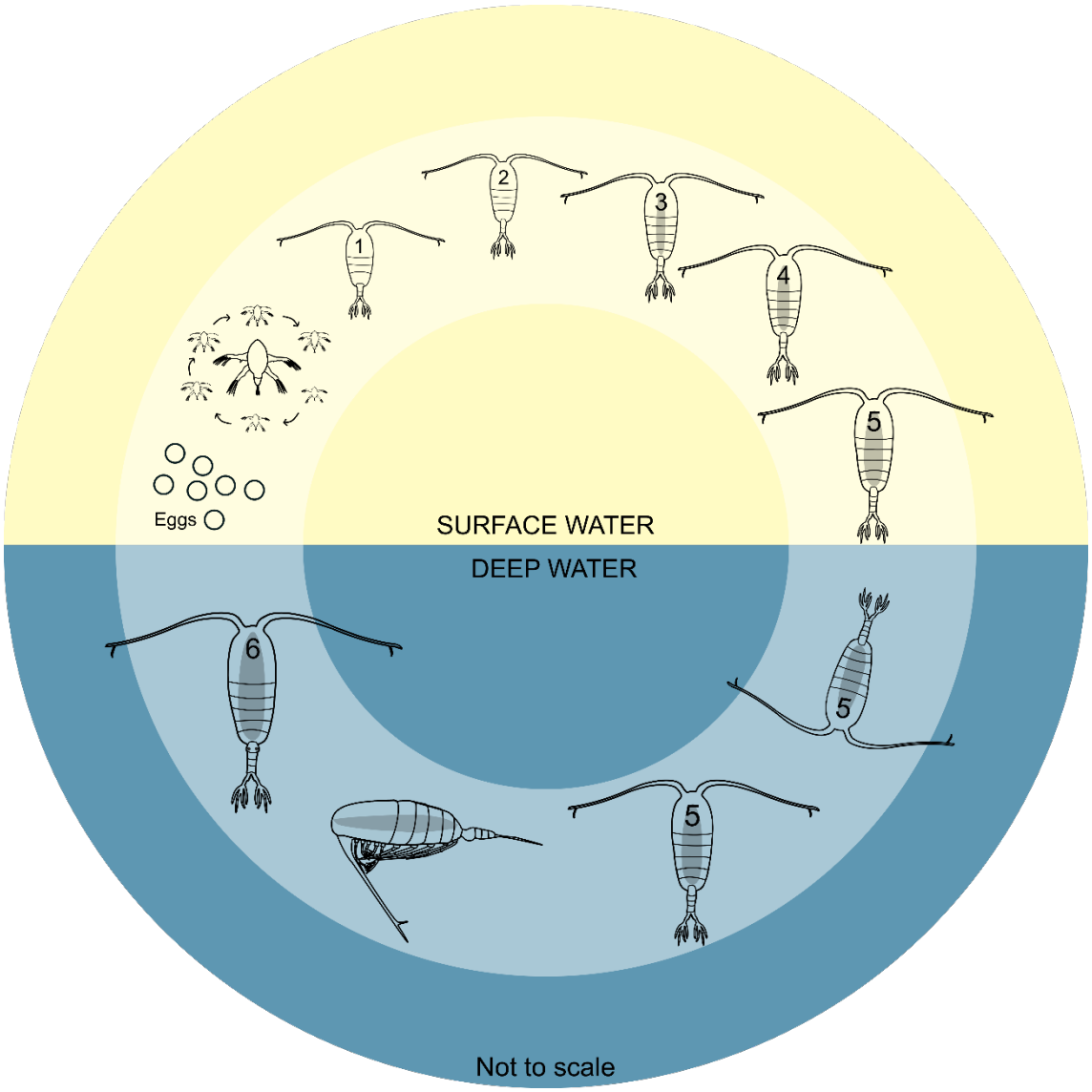


FIGURE 24 | The first year in the life history of *Calanus finmarchicus*, showing eggs, the six naupliar stages, the five juvenile copepodite stages and the mature adult. The grey interior of the animals represents the lipid sac. The last stage of the life cycle (where the mature adult continues to feed and reproduce until death) is not shown.

5.3 Methodology for plankton analysis and research limitations

The advantages and disadvantages of some of the current methods for analysing microplankton were discussed in Chapter 4. Relating the microplankton to grazers was an essential part of this research. To directly measure grazing of the natural plankton assemblage by *Calanus finmarchicus*, prey disappearance was measured after considering algal growth by the use of a control (detailed in Chapter 2). A plankton wheel kept the sampled natural plankton assemblage in suspension, rotating at one revolution per minute. This is a well-established and widely used

method (e.g. Corner, Head & Kilvington, 1972; Bell et al., 2007; Mayor et al., 2009a, 2009b; Morata & Søreide, 2015; Cole et al., 2019). The main concerns with the method are bottle effects (Kiørboe, Møhlenberg & Riisgård, 1985) and animal manipulation (Nőges, 1992). To reduce potential bottle effects, ten copepods were assigned to one 2.2 L bottle using published clearance rates to confirm the volume was large enough (Mayor, 2005). At this concentration, excretion is unlikely to bias the experiments. There is some limited evidence that copepods can be stressed and cause simultaneous defecation, impacting the level of particulate carbon in the bottle (Nőges, 1992). While these data are not copepod specific, as much care was taken as possible to avoid unnecessary manipulation. When beginning a new 24-hour experiment, a new bottle was prepared in advance containing the natural plankton assemblage at the same temperature as the existing bottle. A 5 mm internal diameter dip tube was used to move the animals and the water around them, blocking one end to create a vacuum and meaning transfer time was ~ 5 seconds. The death rate of experimental animals across both spring 2018 and summer 2019 was 0.56 %, with a total of two animals from one station expiring after the first day of the experiment. It was noted that the animals were in poor condition before the experiment began.

Sequential egg production experiments allowed a robust measurement of biomass change over the course of the experiment, to better assess if they copepods were using internal reserves. Well-established experiments to quantify the time it takes for copepods to 'package' ingested material into eggs suggests there is interspecific variability, but the longest period for incorporation of any copepod was four days (Tester & Turner, 1990). Thus, the five-day experiments are sufficient.

Grazing rates presented in this thesis are likely conservative estimates due to the possibility of microzooplankton grazing in the control bottles (Nejstgaard, Naustvoll & Sazhin, 2001). The control bottles are essential to calculate normal algal growth, but the grazing pressure of the experimental *C. finmarchicus* on the microzooplankton is missing from the control bottles. This is particularly relevant in conditions where ciliates were selected for (stations F2, F4 and ST1 in spring 2018), because it widens the gap between microzooplankton grazing in the control and in the experimental bottles. Any form of pre-filtering the bottles would have removed large chains of diatoms and colonies of flagellates and diverged from the natural plankton assembly. The same concept stopped eggs from being removed for the experiment, which made egg cannibalism a concern. However, parallel experiments in spring 2018 showed similar egg production rates. Alternative methods have been suggested to avoid these problems (Nejstgaard, Naustvoll & Sazhin, 2001), for example by measuring faecal pellet production, but this method is less effective with the natural plankton assemblage.

In terms of other indirect methods of interest, it may be possible to use colour pigmentation to assess fitness in *C. finmarchicus* and *C. glacialis*. Red pigmentation allowed the two species to be distinguished in samples taken on the same research expeditions herein (JR18007) (Lindeque et al., 2022). However, more than a species-specific process, it seems to be an indication of fitness. The copepods each contain more of this pigment when they are in their preferred water mass (Trudnowska et al., 2020) – Atlantic and Arctic respectively – meaning that they are better able to bioconvert Astaxanthin to carotenoids (Vilgrain et al., 2023 and references within).

The biggest challenge facing the analysis of plankton is the need to improve the machine learning algorithms that should expedite the high throughput flow imaging techniques. The process presented in Chapter 3 & Chapter 4 using the FlowCam was almost as labour intensive as inverted microscopy, with each sample taking 2-4 hours to manually verify and to remove non-target particles. VisualSpreadsheet Classifier was trained using an assortment of ≥ 1000 images for each class taken from across all samples. Interspecific variation in sizes between species and the functional groups used across the sampling stations could have introduced error to the training database and contributed to some of the inaccurate classifications. Future training sets should be based on size more than any other morphological factor. Compounding the difficulty was that many of the target particles were at the lower end of the size capabilities for the FlowCam, and so many of the images were blurred and the edge detection was inaccurate. While the cells > 20 µm in diameter were easy for the algorithm to classify, smaller cells often had to be manually sorted. The large range in the particle size did not seem to help here, with aggregates forming around the large particles and forming one image. The diatom chains that were not broken up by the process through the FlowCell were also photographed as one organism, and the 'Particles per chain' property did not function. With the capabilities of artificial intelligence increasing exponentially, it is doubtless that a more efficient algorithm could be made specifically for Arctic plankton. The skills of data scientists and software engineers need to be integrated with taxonomists and ecologists to hone these high-throughput methods and enable the fast, automated sampling they promise.

Looking to the future, metabarcoding may become the standard for plankton analysis. As reference databases expand and the technology becomes less expensive, its popularity can only grow. When the links are better understood between the base of the food web and the higher trophic levels, metabarcoding analysis of protists can act as bioindicators for environmental impact assessments, rather than the expensive and labour intensive macrofaunal-based surveys that are common currently (Stoeck et al., 2018). Future sampling equipment and laboratories will likely have in-situ semi-autonomous imaging and omics sensors deployed onto platforms or submersibles, coupled to sensors collecting physical and chemical measurements (Pierella

Karlusich et al., 2022). This will provide vast amounts of data and long-term series that will allow us to explore the main variables controlling the plankton, and interactions between the plankton simultaneously. What an exciting time for science!

Finally, speculating on the future of methods for scientists, I think it will be increasingly necessary to collaborate not just with data scientists and software engineers, but with disciplines adjacent to our own. More efficient cross-discipline collaboration will prevent vital science from being buried as the scientific literature grows exponentially, with an average doubling period of 15 years (Fortunato et al., 2018). The recent MoSAiC expedition exemplified this by using a "science question based" approach, resulting in cross-cutting and interdisciplinary groups of scientists who are working on specific topics (Alfred Wegener Institute, 2022). Better collaboration can help to make science more inclusive. This is something that is drastically needed in the UK polar science community which has < 3 % representation of Black, Indigenous and People of Colour (Frater, 2021). While the aims of every research project are different, a common aim is to share what we learn. How widespread this sharing will be depends on the research question and its applications. The other common aim is for what we share to be understood, and hence the way we share our knowledge needs to develop just as our other methods do. The breadth of collaboration needs to be balanced against information overwhelm, so nobody should lose their key area of expertise. However, in my opinion, to become better scientists, collaboration and knowledge sharing are key.

The research in this thesis does not need to be shared widely with all other ocean scientists or every member of the public, but the measurements made here can constrain global biogeochemical models, which in turn can make useful predictions of carbon sequestration and the impacts of climate change on our world – and it is these that should be shared widely.

Appendix A Supplementary materials for Chapter 2

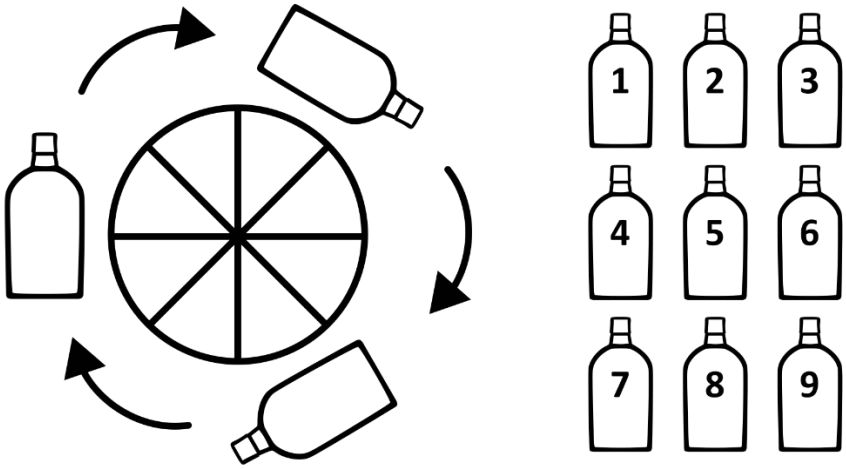
Supplementary methodology

Particle removal experiments were used to determine grazing and production simultaneously, as detailed in section 2.3.1. Natural seawater was sampled from the chlorophyll maximum every 24 hours, using silicon tubing to rinse and fill the nine experimental 2.2L bottles. They were filled a little at a time to ensure homogeneity. Zooplankton visible to the naked eye were removed. The seawater was not pre-screened, however, as this would remove or break down the prey necessary for the experimental *C. finmarchicus* to feed on and would mean the plankton assemblage did not mimic the true environment.

Female *C. finmarchicus* were added to the experimental bottles, 10 to each 2.2 L bottle. No copepods were added to the control bottles. All were transferred to a plankton wheel to ensure all plankton remained in suspension (Supplementary Figure 1). After the 24-hour period, copepods were transferred to new bottles containing the next sampled seawater.

Supplementary Figure 1 | Schematic diagram (A) and photograph (B) of the plankton wheel used to analyse grazing and production in the Fram Strait in spring 2018. To each of the experiment bottles (bottles 1- 6), 10 female *C. finmarchicus* were added. To the control bottles, no copepods were added.

(A)



(B)

Supplementary Table 1 | The composition and biomass of the microplankton across the Fram Strait (initial carbon), and the clearance rate and ingestion rate of the *Calanus finmarchicus* females. Water samples were taken at the chlorophyll maximum using 20L Niskin bottles in May – June 2018. Clearance rates and ingestion rates were calculated by particle removal experiments and grazing equations (Frost, 1972). Ind represents individual females. Mean averages are shown ± standard deviation.

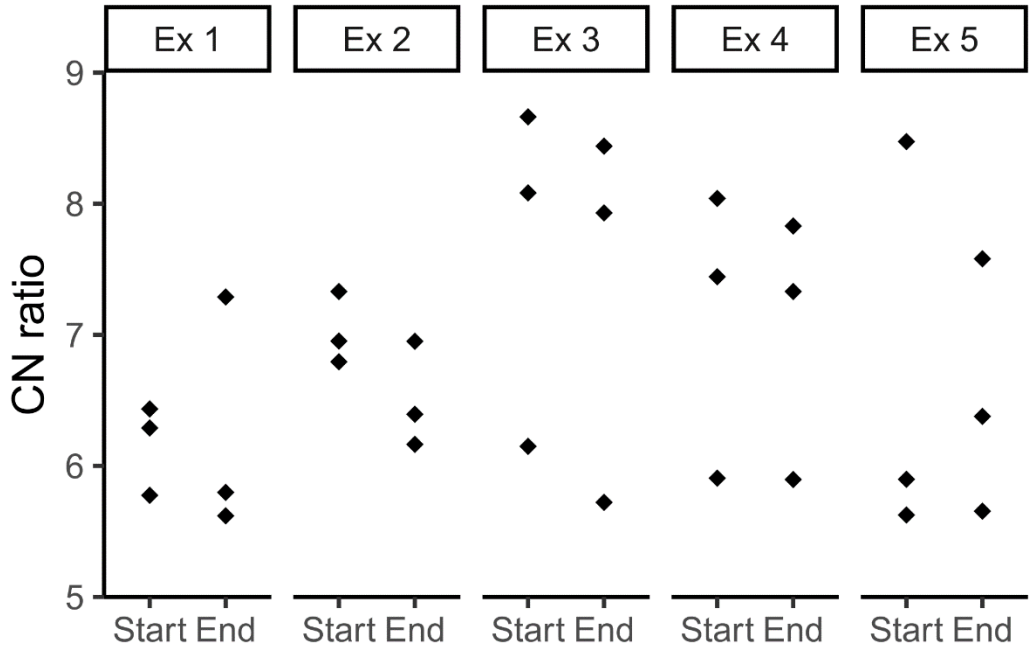
Ex	Day	Station	Type	Initial carbon	Clearance rate	Ingestion rate
				(µmol C L^{-1})	(mL ind^{-1} day^{-1})	(µmol C ind^{-1} day^{-1})
			Small diatoms	37.8 ± 10.2	474.65 ± 63.01	4.1 ± 0.2
			Large diatoms	14.3 ± 8.9	382.33 ± 150.52	1.6 ± 0.4
			Pennate diatoms	0.2 ± 0.1	14.33 ± 24.81	0.0 ± 0.0
3	1	F17	Dinoflagellates	12.9 ± 6.6	118.38 ± 150.89	0.6 ± 0.7
			Ciliates	1.7 ± 0.4	110 ± 95.57	0.1 ± 0.1
			Flagellates	2.7 ± 2.2	0.00 ± 0.00	0.0 ± 0.0
			Total	69.5 ± 2.6		6.5 ± 0.8
			Small diatoms	13.8 ± 1.2	468.96 ± 185.20	2.0 ± 0.3
			Large diatoms	4.6 ± 1.4	660.46 ± 131.64	0.6 ± 0.0
			Pennate diatoms	0.1 ± 0.0	13.79 ± 23.89	0.0 ± 0.0
3	2	F15	Dinoflagellates	10.2 ± 2.5	120.39 ± 103.95	0.5 ± 0.4
			Ciliates	0.7 ± 0.0	31.47 ± 18.17	0.0 ± 0.0
			Flagellates	0.5 ± 0.1	214.14 ± 94.18	0.1 ± 0.0
			Total	30.0 ± 2.1		3.3 ± 0.5
			Small diatoms	82.7 ± 15.3	273.02 ± 105.76	3.0 ± 1.5
			Large diatoms	31.0 ± 1.9	87.40 ± 59.32	1.5 ± 0.7
			Pennate diatoms	0.3 ± 0.1	134.07 ± 122.95	0.0 ± 0.0
3	3	F13	Dinoflagellates	22.7 ± 10.4	247.75 ± 141.00	2.6 ± 1.2
			Ciliates	5.6 ± 1.1	199.73 ± 176.88	0.6 ± 0.5
			Flagellates	0.7 ± 0.1	130.53 ± 67.86	0.1 ± 0.0
			Total	143.0 ± 8.0		7.8 ± 2.3
			Small diatoms	41.8 ± 20.8	296.20 ± 7.38	2.7 ± 1.1
3	4	F10	Large diatoms	16.8 ± 0.7	257.63 ± 11.51	3.0 ± 0.1
			Pennate diatoms	0.2 ± 0.0	32.86 ± 29.53	0.0 ± 0.0

Ex	Day	Station	Type	Initial carbon	Clearance rate	Ingestion rate
				(µmol C L^{-1})	(mL ind^{-1} day^{-1})	(µmol C ind^{-1} day^{-1})
			Dinoflagellates	27.7 ± 7.1	371.18 ± 44.98	3.7 ± 0.4
			Ciliates	5.0 ± 1.0	230.79 ± 165.97	0.7 ± 0.5
			Flagellates	0.5 ± 0.1	222.42 ± 36.93	0.1 ± 0.0
			Total	92.1 ± 29.7		10.1 ± 0.3
			Small diatoms	17.3 ± 3.2	355.63 ± 103.79	1.2 ± 0.4
			Large diatoms	21.5 ± 3.5	59.24 ± 41.64	0.6 ± 0.4
			Pennate diatoms	0.6 ± 0.4	80.61 ± 78.34	0.0 ± 0.0
3	5	FS1	Dinoflagellates	15.9 ± 0.3	118.33 ± 55.82	1.0 ± 0.4
			Ciliates	7.2 ± 2.4	264.88 ± 137.14	0.8 ± 0.2
			Flagellates	0.6 ± 0.0	179.27 ± 9.81	0.1 ± 0.0
			Total	63.2 ± 2.5		3.7 ± 1.4
			Small diatoms	38.1 ± 14.4	306.80 ± 422.81	4.1 ± 3.5
			Large diatoms	22.8 ± 2.1	205.47 ± 178.49	1.8 ± 0.9
			Pennate diatoms	0.4 ± 0.1	143.37 ± 83.42	0.0 ± 0.0
4	1	F8	Dinoflagellates	21.6 ± 2.3	322.60 ± 42.10	2.9 ± 0.2
			Ciliates	1.2 ± 0.3	311.88 ± 28.34	0.2 ± 0.1
			Flagellates	0.3 ± 0.1	117.12 ± 101.52	0.0 ± 0.0
			Total	84.4 ± 14.9		9.0 ± 4.5
			Small diatoms	6.6 ± 0.1	27.31 ± 285.25	0.3 ± 0.3
			Large diatoms	2.3 ± 2.2	483.41 ± 200.19	0.4 ± 0.1
			Pennate diatoms	0.7 ± 0.3	138.50 ± 66.39	0.1 ± 0.0
4	2	HGIV	Dinoflagellates	10.8 ± 0.0	157.82 ± 117.78	0.8 ± 0.5
			Ciliates	6.9 ± 1.8	161.75 ± 207.06	0.3 ± 0.3
			Flagellates	1.2 ± 0.5	29.29 ± 35.95	0.0 ± 0.0
			Total	28.6 ± 0.3		1.9 ± 0.9
			Small diatoms	4.9 ± 0.9	378.30 ± 459.08	0.4 ± 0.3
			Large diatoms	0.2 ± 0.3	147.76 ± 139.54	0.0 ± 0.0
			Pennate diatoms	0.5 ± 0.0	135.93 ± 107.15	0.0 ± 0.0
4	3	F4	Dinoflagellates	11.1 ± 0.1	0.00 ± 0.00	0.0 ± 0.0
			Ciliates	6.7 ± 3.4	108.69 ± 153.18	0.7 ± 0.6
			Flagellates	0.8 ± 0.3	9.24 ± 16.01	0.0 ± 0.0
			Total	24.1 ± 1.8		1.2 ± 0.7

Ex	Day	Station	Type	Initial carbon	Clearance rate	Ingestion rate
				(µmol C L^{-1})	(mL ind^{-1} day^{-1})	(µmol C ind^{-1} day^{-1})
4	4	F2	Small diatoms	8.6 ± 3.0	82.75 ± 206.74	0.3 ± 0.2
			Large diatoms	0.5 ± 0.4	551.60 ± 51.50	0.1 ± 0.0
			Pennate diatoms	1.5 ± 0.5	56.03 ± 23.91	0.1 ± 0.0
			Dinoflagellates	14.8 ± 0.6	21.23 ± 36.77	0.2 ± 0.3
			Ciliates	13.2 ± 4.5	233.06 ± 167.89	1.3 ± 1.3
			Flagellates	1.0 ± 0.6	94.89 ± 66.41	0.1 ± 0.0
			Total	39.6 ± 0.2		2.0 ± 1.6
4	5	KB0	Small diatoms	4.9 ± 1.6	0.00 ± 169.60	0.0 ± 0.0
			Large diatoms	0.0 ± 0.0	0.00 ± 0.00	0.0 ± 0.0
			Pennate diatoms	3.0 ± 1.0	68.77 ± 22.89	0.2 ± 0.1
			Dinoflagellates	22.5 ± 7.7	0.00 ± 0.00	0.1 ± 0.1
			Ciliates	3.5 ± 1.8	19.52 ± 33.81	0.0 ± 0.0
			Flagellates	0.4 ± 0.1	58.09 ± 50.65	0.0 ± 0.0
			Total	34.4 ± 12.0		0.3 ± 0.1
5	1	ST1	Small diatoms	28.6 ± 2.1	486.53 ± 172.45	3.0 ± 0.9
			Large diatoms	4.1 ± 2.9	402.92 ± 170.92	0.4 ± 0.1
			Pennate diatoms	0.2 ± 0.0	3.46 ± 6.00	0.0 ± 0.0
			Dinoflagellates	11.9 ± 3.0	115.33 ± 112.47	0.8 ± 0.3
			Ciliates	10.5 ± 13.9	2171.55 ± 139.54	2.2 ± 0.1
			Flagellates	0.1 ± 0.0	77.28 ± 67.88	0.0 ± 0.0
			Total	55.4 ± 16.2		6.3 ± 0.8

Appendix A

Supplementary Figure 2 | The change in ratio of carbon (C) to nitrogen (N) in *C. finmarchicus* individuals in the Fram Strait in spring 2018. The C and N content of the experimental females did not vary significantly between the start and end of the incubations. Ex represents experiment.

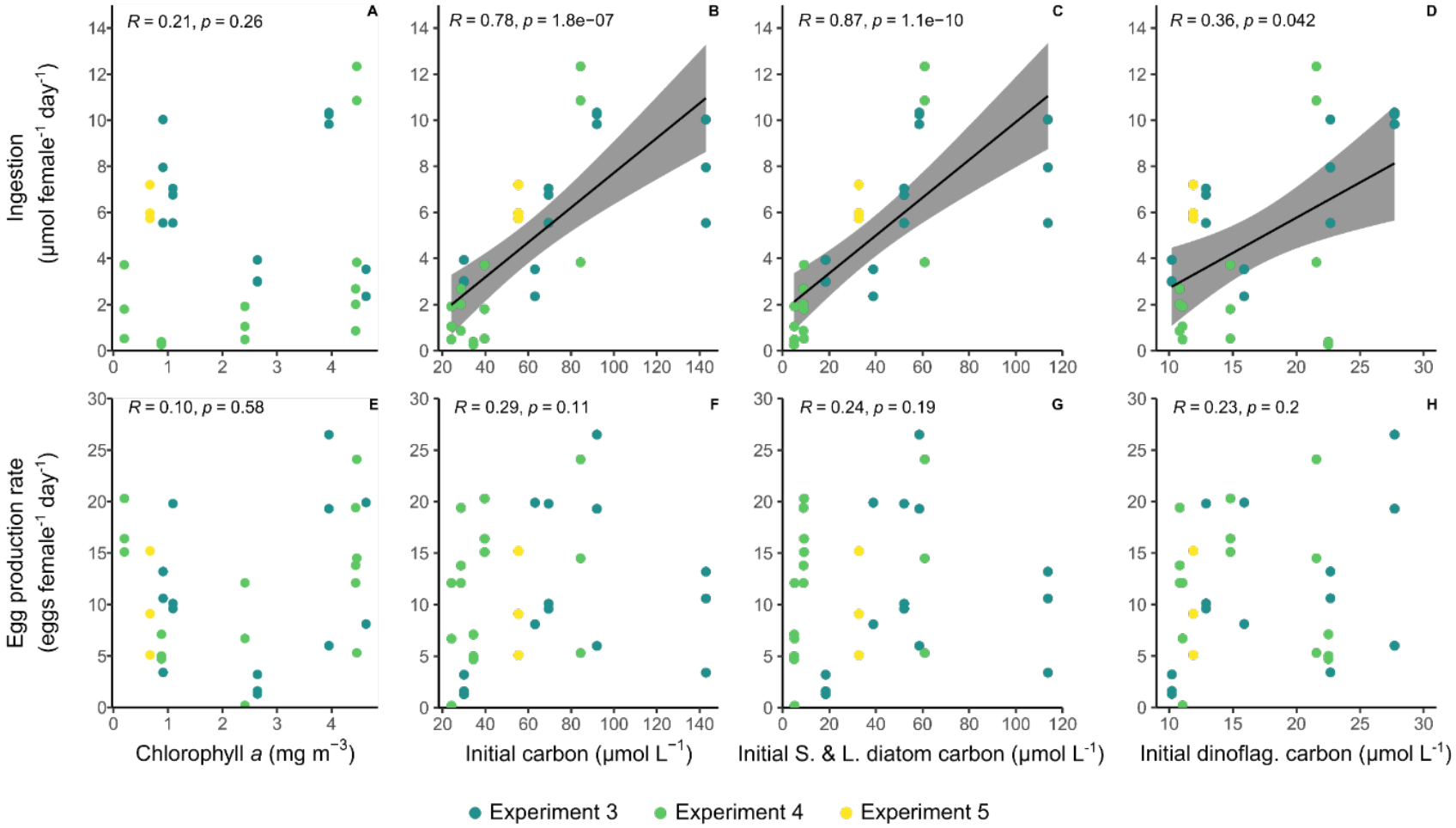


Supplementary Table 2 | The carbon (C) and nitrogen (N) content of the experimental female *C. finmarchicus* caught in the Fram Strait in May to June 2018. Start denotes animals that were preserved immediately after catch and identification. End denotes animals that were caught at the same time as those in Start, but then underwent bottle incubations. Ind represents individual female.

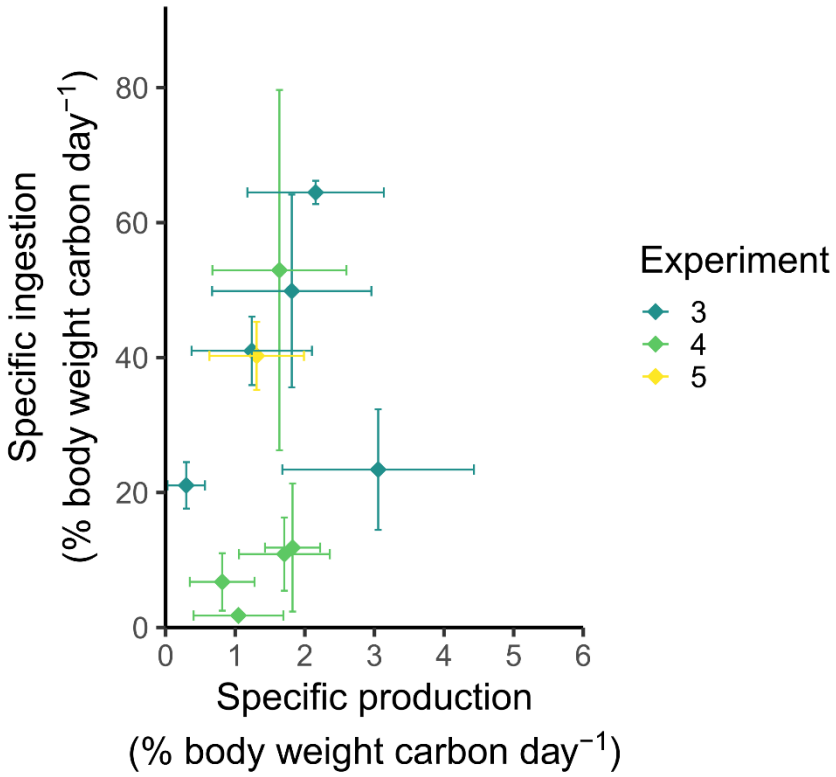
Ex	Length	Timepoint	C content	N content	C:N
	(days)		(µmol C ind^{-1})	(µmol N ind^{-1})	
1	4	Start	12.57	2.00	6.29
			13.32	2.07	6.43
			17.32	3.00	5.78
		End	13.24	2.36	5.62
			14.49	2.50	5.80
			12.49	1.71	7.29
2	4	Start	14.65	2.00	7.33
			11.16	1.64	6.79
			12.91	1.86	6.95
		End	12.32	2.00	6.16
			13.24	2.07	6.39
			12.41	1.78	6.95
3	5	Start	16.74	2.07	8.08
			14.49	2.36	6.15
			12.99	1.50	8.66
		End	14.15	1.78	7.93
			22.90	2.71	8.44
			13.07	2.28	5.72
4	5	Start	13.07	2.21	5.91
			21.81	2.71	8.04
			18.07	2.43	7.44
		End	15.65	2.00	7.83
			15.15	2.57	5.90
			18.32	2.50	7.33
5	2	Start	18.15	2.14	8.47
			13.65	2.43	5.63
			14.74	2.50	5.90
		End	14.57	2.28	6.38
			19.48	2.57	7.58
			13.32	2.36	5.65

Appendix A

Supplementary Figure 3 | The relationships between the quantity of available food and the ingestion (A, B, C & D) and egg production rate (E, F & G, H) of *C. finmarchicus* in bottle-incubation experiments in May and June 2018. The quantity of available food is estimated from using the chlorophyll a concentration as a proxy (A & E), and calculated using inverted microscopy for all cell types (B & F), for small (S) and large (L) diatoms (C & G), and for dinoflagellates (D & H). Shaded areas show 95 % confidence intervals. R is spearman's ρ.

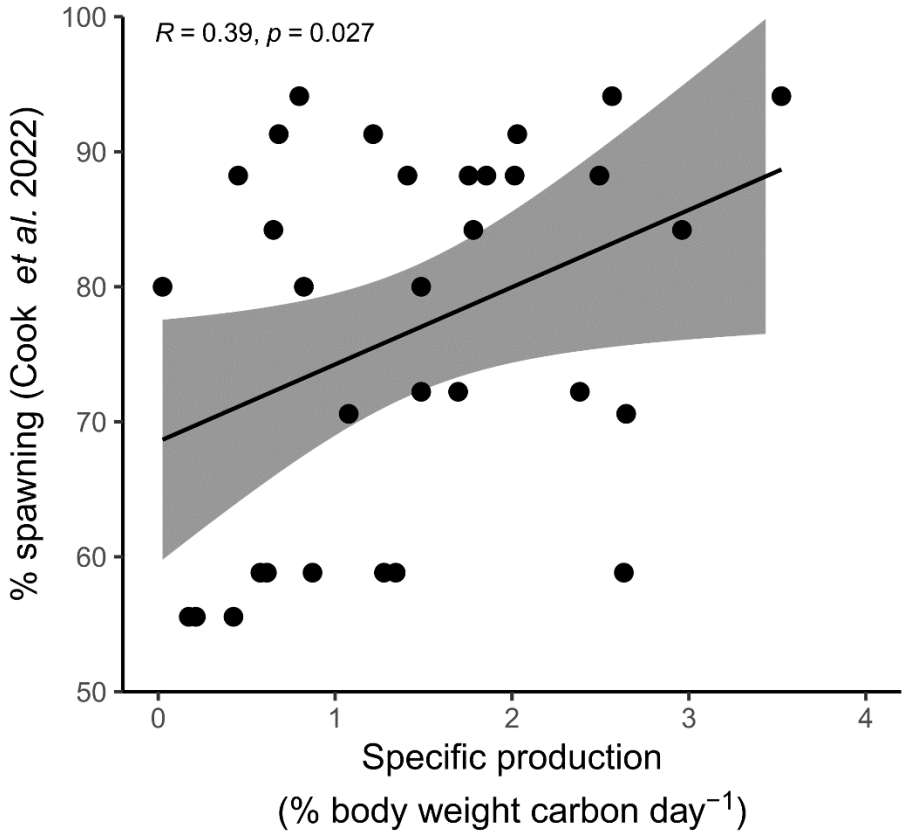


Supplementary Figure 4 | Specific ingestion and specific production by *C. finmarchicus* females in bottle incubation experiments in May to June 2018. The relationship between specific

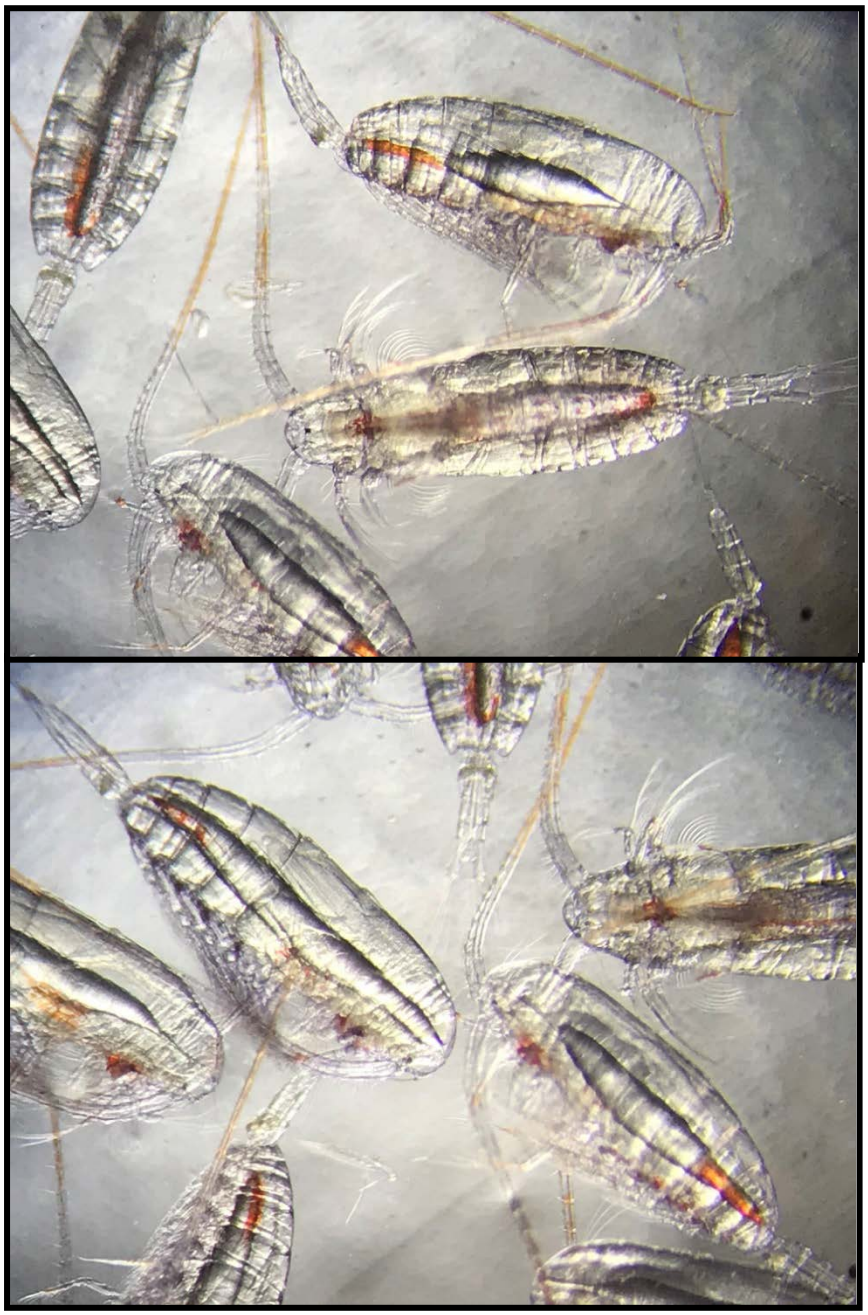


ingestion and specific production is insignificant ($\rho = 0.21$, $p = 0.24$).

Supplementary Figure 5 | The significant positive correlation between specific egg production and the percentage of *C. finmarchicus* females that were spawning in experiments run alongside the experiments detailed here in the Fram Strait in spring 2018.



Supplementary Figure 6 | Photographs taken of experimental female *C. finmarchicus* on 21/5/18, showing the lipid sac and gonopore but no obvious ovaries or eggs.



Appendix A

Supplementary Table 3 | Egg production rates (EPR) of *Calanus finmarchicus* in the Arctic. The Arctic Ocean here excludes the Norwegian Sea. Temp. is the ocean temperature at surface/animal sampling depth. False-bottom includes any container that allows eggs to pass but not adults, including those with mesh bottoms and those with narrow elongated ends. Literature search using terms: "Calanus finmarchicus" & "egg production rate" & Arctic giving 624 results.

Area	Year	Temp.	Season	Exp. type	EPR	Error	EPR	Error	Notes	Reference
		°C			(eggs female^{-1} day^{-1})	SD (* = SE)	(% body carbon day^{-1})	SD (* = SE)		
North Svalbard	1987	-1 to -1.5	Summer	False-bottom incubations	0.3	NA	NA	NA	Eggs from only one station	(Hirche & Mumm, 1992)
East Greenland Shelf	1988	0	Summer	False-bottom incubations	19.9	5.7	4.5	1.3	Mean	(Hirche, 1990)
East Greenland Shelf	1988	0	Summer	False-bottom incubations	34	NA	NA	NA	Maximum	(Hirche, 1990)
Central Greenland Sea	1989	0 to 2	Summer	False-bottom incubations	4.3	NA	NA	NA		(Hirche & Kosobokova, 2007)

Area	Year	Temp.	Season	Exp. type	EPR	Error	EPR	Error	Notes	Reference
		°C			(eggs female^{-1} day^{-1})	SD (* = SE)	(% body carbon day^{-1})	SD (* = SE)		
Barents Sea	1989	-1 to -0.5	Spring	False-bottom incubations	0	0	NA	NA	Prebloom – all had immature gonads	(Hirche & Kattner, 1993)
Disko Bay	2005	0	Spring	Bottle incubations	4	0.7*	NA	NA	Average	(Madsen et al., 2008)
Disko Bay	2005	0	Spring	Bottle incubations	44	7*	NA	NA	Maximum	(Madsen et al., 2008)
Disko Bay	2008	-1.3	Early spring	Bottle incubations	0.03		0.01		Pre-bloom	(Swalethorp et al., 2011)
Disko Bay	2008	1	Spring	Bottle incubations	9.7		1.7		Bloom	(Swalethorp et al., 2011)
Disko Bay	2008	1	Summer	Bottle incubations	7.0		1.0		Post-bloom	(Swalethorp et al., 2011)
Barents Sea	2010	0, 2.5, 5, 7.5, 10	Spring	False-bottom incubation	6.3 – 14.5		NA	NA		(Pasternak et al., 2013)
Disko Bay	2012	0	Spring	Bottle incubations	36		0.05		Maximum	(Møller et al., 2016)

Appendix B Supplementary material for Chapter 3

Supplementary methodology

The gonad maturation stage (GS) of ≥10 females from each station throughout JR18007 were determined following procedures established by Niehoff & Runge, 2003. The stage of development was recorded for representative samples of animals to determine the proportion of mature *C. finmarchicus* females as an index of the proportion of spawning females in the population. There are four stages of development (GS1-4), and three divisions within GS4, as detailed in Table 1 of Niehoff & Runge, 2003. In summary, GS1-3 are characterised by undeveloped oocytes. GS4 represents mature females carrying stage oocytes in their mature form or oocytes with no nucleus present, indicating they are the next clutch. The proportion of GS4 females in a preserved sample is an index of the proportion of spawning females in a population.

- GS1: anterior and posterior diverticula (two diverticula extend anteriorly into the head region and posteriorly along each side of the thorax to the genital pore on the first urosomal segment) are empty and the diverticula walls are visible as thin lines.
- GS2: small developing oocytes are present (without a follicle cell layer or lipid and yolk droplets), either transparent or lightly opaque, in one row in both the anterior and posterior diverticula.
- GS3: multiple layers of small developing oocytes are present in both anterior and posterior diverticula. Oocytes may be visible as lightly coloured, may have prominent follicle cell layer, and be <90 µm in diameter.
- GS4: medium to dark brown oocytes are present and are >90 µm in diameter, forming the most ventral later in the gonads. Developing oocytes are also present, located dorsally.
    - GS4a: Developed oocytes are densely packed in multiple layers, pouches are prominent.
    - GS4b: Developed oocytes form one solid row, oocytes touching but less densely packed than GS4a; pouches in posterior diverticula are absent or rudimentary.
    - GS4c: Developed oocytes are loosely packed and posterior pouches are always absent.

More detailed understanding of the development stage can be made by histological section, but to understand the proportion of the population that are spawning, this examination under a stereomicroscope was sufficient.

Appendix B

Supplementary Table 4 | The technical specifications, setup details, image sorting details and measurement outputs for the FlowCam® 8400 used to analyse microplankton communities across the Fram Strait in August 2019.

FlowCam technical specifications	
FlowCam model number	8400
FlowCam unit serial number	10313
Camera colour/monochrome	Colour
Fluidics	5.0 mL C80 syringe pump
Software details	Visual Spreadsheet v4.3.55
Any additional upgrades to the machine or optional accessories	N/A
FlowCam setup details	
Flow cell sizes and types used, and objectives used for each flow cell	FC100FV flow cell with a 10x objective
Image acquisition mode	Auto-image at 45 frames per second
Flow rate	0.300 mL min^{-1}
Sample volume analysed	100x 5 mL samples, totalling 12,719,855 particles imaged and analysed
Full context settings	Please contact author for full context settings
Image sorting details	
Image sorting method	Combination of automated and manual
Software used, with version details	Visual Spreadsheet v4.3.55 with Classifer
Image library description and sizes	There were 26 image libraries created with over 1000 of representative images in each, taken from across all samples. The libraries were based on morphological and functional groupings

Particle property selections	Please contact author for full particle property settings
Evaluation of the accuracy of auto-classifications	The number of particles manually placed into classes was roughly counted in three samples and compared to the total number of particles, reaching an efficiency of ~40%
Measurement outputs	
Measurement type(s) used	Volume ABD
Justification for this choice	Based on similar previous studies
Validation of measurement outputs	This was compared to microscopy

Appendix B

Supplementary Table 5 | The composition and biomass of the microplankton across the Fram Strait (initial carbon), and the clearance rate and ingestion rate of the Calanus finmarchicus females. Water samples were taken at the chlorophyll maximum using 20L Niskin bottles in August 2019. Clearance rates and ingestion rates were calculated by particle removal experiments and grazing equations (Frost, 1972). Ind represents individual females. Mean averages are shown ± standard deviation.

Ex	Day	Station	Type	Initial carbon	Clearance rate	Ingestion rate
				(µmol C L^{-1})	(mL ind^{-1} day^{-1})	(µmol C ind^{-1} day^{-1})
1	1	NT11	Small diatoms	0.04 ± 0.03	60.68 ± 65.14	0.00 ± 0.00
			Large diatoms	0.01 ± 0.01	0.00 ± 447.87	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	472.85 ± 194.47	0.00 ± 0.00
			Dinoflagellates	0.01 ± 0.01	625.33 ± 803.35	0.00 ± 0.00
			Ciliates	0.51 ± 0.59	0.00 ± 61.67	0.00 ± 0.00
			Flagellates	0.12 ± 0.11	0.00 ± 176.52	0.01 ± 0.01
			Total	0.73 ± 0.80		0.01 ± 0.02
2	1	F7b	Small diatoms	0.19 ± 0.06	111.31 ± 141.09	0.01 ± 0.01
			Large diatoms	0.03 ± 0.00	76.29 ± 41.57	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	503.73 ± 150.92	0.00 ± 0.00
			Dinoflagellates	0.01 ± 0.00	244.49 ± 258.51	0.00 ± 0.00
			Ciliates	0.30 ± 0.11	409.97 ± 287.94	0.04 ± 0.02
			Flagellates	0.50 ± 0.03	0.00 ± 431.31	0.01 ± 0.01
			Total	1.04 ± 0.20		0.07 ± 0.04
2	2	IS1	Small diatoms	0.05 ± 0.02	129.53 ± 1.00	0.01 ± 0.00
			Large diatoms	0.02 ± 0.01	64.03 ± 17.40	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	539.03 ± 193.04	0.00 ± 0.00
			Dinoflagellates	0.05 ± 0.03	314.12 ± 222.93	0.00 ± 0.00
			Ciliates	0.15 ± 0.13	442.4 ± 411.49	0.02 ± 0.01
			Flagellates	0.14 ± 0.07	95.34 ± 103.99	0.01 ± 0.00

Ex	Day	Station	Type	Initial carbon	Clearance rate	Ingestion rate
				(µmol C L^{-1})	(mL ind^{-1} day^{-1})	(µmol C ind^{-1} day^{-1})
			Total	0.41 ± 0.02		0.03 ± 0.02
			Small diatoms	0.07 ± 0.00	0.00 ± 122.10	0.00 ± 0.00
			Large diatoms	0.02 ± 0.00	0.00 ± 80.04	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	0.00 ± 89.37	0.00 ± 0.00
2	3	IS2	Dinoflagellates	0.00 ± 0.00	0.00 ± 506.05	0.00 ± 0.00
			Ciliates	0.52 ± 0.10	135.96 ± 54.58	0.04 ± 0.02
			Flagellates	0.24 ± 0.10	0.00 ± 297.81	0.00 ± 0.00
			Total	0.86 ± 0.04		0.05 ± 0.01
			Small diatoms	0.09 ± 0.00	0.00 ± 83.35	0.00 ± 0.00
			Large diatoms	0.02 ± 0.00	0.00 ± 7.04	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	0.00 ± 174.99	0.00 ± 0.00
2	4	D1	Dinoflagellates	0.02 ± 0.00	0.00 ± 151.48	0.00 ± 0.00
			Ciliates	0.14 ± 0.15	46.79 ± 0.00	0.00 ± 0.00
			Flagellates	0.10 ± 0.05	0.00 ± 216.87	0.00 ± 0.00
			Total	0.36 ± 0.10		0.00 ± 0.00
			Small diatoms	0.23 ± 0.03	0.00 ± 230.85	0.01 ± 0.02
			Large diatoms	0.02 ± 0.01	0.00 ± 49.96	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	0.00 ± 211.87	0.00 ± 0.00
2	5	D2	Dinoflagellates	0.07 ± 0.06	337.89 ± 580.86	0.00 ± 0.00
			Ciliates	0.10 ± 0.03	332.4 ± 179.66	0.01 ± 0.00
			Flagellates	0.41 ± 0.33	0.00 ± 351.16	0.01 ± 0.01
			Total	0.81 ± 0.24		0.03 ± 0.01
			Small diatoms	0.23 ± 0.01	163.23 ± 25.58	0.03 ± 0.00
3	1	D3	Large diatoms	0.04 ± 0.00	217.76 ± 96.10	0.01 ± 0.00
			Pennate diatoms	0.01 ± 0.00	184.64 ± 32.49	0.00 ± 0.00

Ex	Day	Station	Type	Initial carbon	Clearance rate	Ingestion rate
				(µmol C L^{-1})	(mL ind^{-1} day^{-1})	(µmol C ind^{-1} day^{-1})
			Dinoflagellates	0.02 ± 0.01	35.91 ± 611.35	0.00 ± 0.00
			Ciliates	0.11 ± 0.02	730.49 ± 314.78	0.02 ± 0.00
			Flagellates	0.28 ± 0.18	313.56 ± 54.80	0.03 ± 0.00
			Total	0.68 ± 0.22		0.10 ± 0.01
			Small diatoms	0.22 ± 0.02	136.26 ± 101.64	0.02 ± 0.01
			Large diatoms	0.01 ± 0.00	0.00 ± 378.00	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	0.00 ± 148.78	0.00 ± 0.00
3	2	D4	Dinoflagellates	0.11 ± 0.03	0.00 ± 244.68	0.01 ± 0.00
			Ciliates	0.07 ± 0.02	354.76 ± 165.58	0.01 ± 0.00
			Flagellates	0.46 ± 0.25	0.00 ± 113.32	0.00 ± 0.00
			Total	0.88 ± 0.25		0.03 ± 0.03
			Small diatoms	0.21 ± 0.03	0.00 ± 126.67	0.00 ± 0.01
			Large diatoms	0.02 ± 0.00	18.89 ± 67.83	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	439.75 ± 40.88	0.00 ± 0.00
3	3	D6	Dinoflagellates	0.04 ± 0.02	0.00 ± 422.60	0.00 ± 0.00
			Ciliates	0.19 ± 0.01	551.15 ± 48.96	0.03 ± 0.00
			Flagellates	0.71 ± 0.7	233.72 ± 263.74	0.09 ± 0.02
			Total	1.17 ± 0.63		0.09 ± 0.08
			Small diatoms	0.13 ± 0.02	0.00 ± 2.41	0.00 ± 0.00
			Large diatoms	0.02 ± 0.01	284.47 ± 397.53	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	499.96 ± 107.77	0.00 ± 0.00
3	4	D6b	Dinoflagellates	0.03 ± 0.02	143.97 ± 775.15	0.00 ± 0.00
			Ciliates	0.22 ± 0.23	0.00 ± 281.49	0.00 ± 0.00
			Flagellates	0.15 ± 0.04	187.02 ± 102.37	0.02 ± 0.00
			Total	0.55 ± 0.30		0.02 ± 0.02

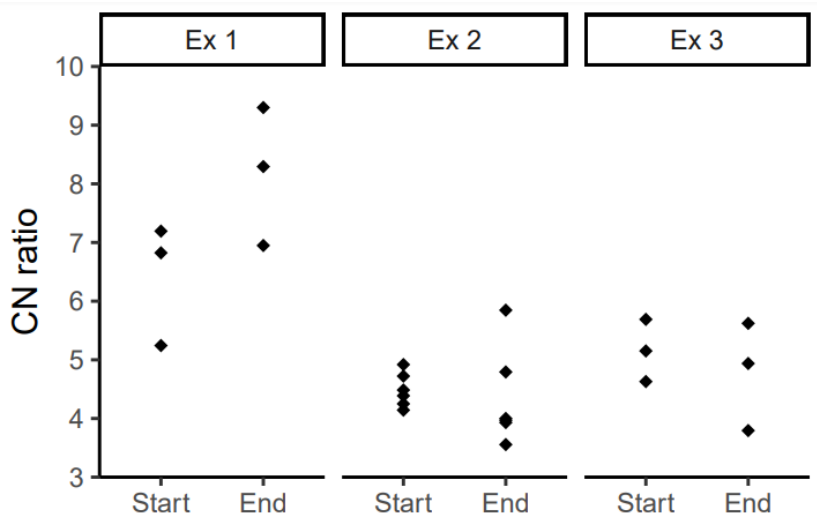
Ex	Day	Station	Type	Initial carbon	Clearance rate	Ingestion rate
				(µmol C L^{-1})	(mL ind^{-1} day^{-1})	(µmol C ind^{-1} day^{-1})
3	5	D6c	Small diatoms	0.21 ± 0.05	77.22 ± 391.27	0.02 ± 0.03
			Large diatoms	0.03 ± 0.01	43.96 ± 326.58	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	0.00 ± 466.95	0.00 ± 0.00
			Dinoflagellates	0.04 ± 0.05	318.95 ± 545.26	0.00 ± 0.00
			Ciliates	0.93 ± 0.29	983.95 ± 374.48	0.14 ± 0.06
			Flagellates	0.31 ± 0.30	476.99 ± 931.04	0.04 ± 0.05
			Total	1.54 ± 0.36		0.12 ± 0.13
4	1	D7	Small diatoms	0.14 ± 0.00	0.00 ± 103.90	0.00 ± 0.00
			Large diatoms	0.03 ± 0.01	328.02 ± 64.12	0.00 ± 0.00
			Pennate diatoms	0.00 ± 0.00	564.27 ± 101.57	0.00 ± 0.00
			Dinoflagellates	0.01 ± 0.01	0.00 ± 36.42	0.00 ± 0.00
			Ciliates	0.33 ± 0.22	720.14 ± 622.48	0.06 ± 0.02
			Flagellates	0.46 ± 0.42	641.27 ± 211.64	0.06 ± 0.02
			Total	0.92 ± 0.71		0.08 ± 0.07
4	2	D8	Small diatoms	0.20 ± 0.03	NA	NA
			Large diatoms	0.01 ± 0.00	NA	NA
			Pennate diatoms	0.00 ± 0.00	NA	NA
			Dinoflagellates	0.03 ± 0.00	NA	NA
			Ciliates	0.20 ± 0.07	NA	NA
			Flagellates	0.32 ± 0.13	NA	NA
			Total	0.97 ± 0.25	NA	NA
4	3	D9	Small diatoms	0.15 ± 0.01	NA	NA
			Large diatoms	0.01 ± 0.00	NA	NA
			Pennate diatoms	0.00 ± 0.00	NA	NA
			Dinoflagellates	0.02 ± 0.00	NA	NA

Ex	Day	Station	Type	Initial carbon	Clearance rate	Ingestion rate
				(µmol C L^{-1})	(mL ind^{-1} day^{-1})	(µmol C ind^{-1} day^{-1})
			Ciliates	0.48 ± 0.35	NA	NA
			Flagellates	0.47 ± 0.16	NA	NA
			Total	1.33 ± 0.55	NA	NA

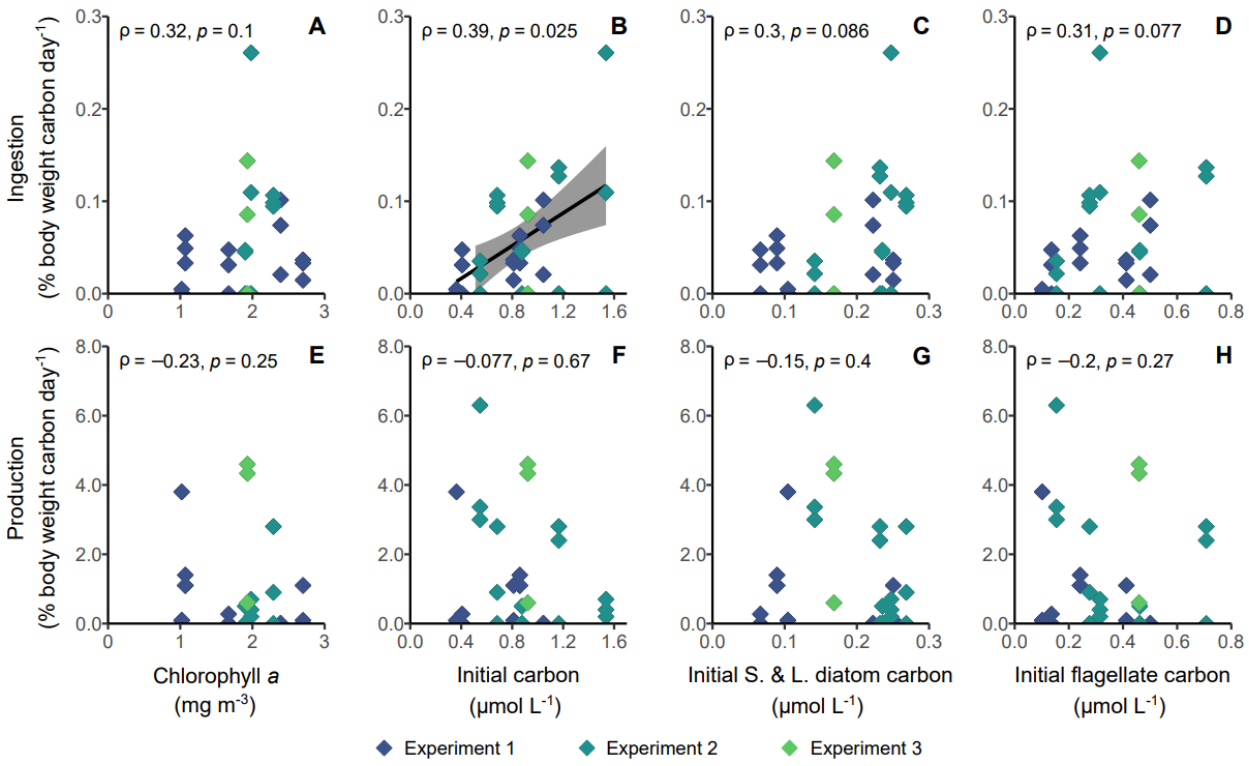
Supplementary Table 6 | The carbon (C) and nitrogen (N) content of the experimental female *C. finmarchicus* caught in the Fram Strait in August 2019. Start denotes animals that were preserved immediately after catch and identification. End denotes animals that were caught at the same time as those in Start, but then underwent bottle incubations. Cop represents *C. finmarchicus* female copepods.

Ex	Length	Timepoint	C content	N content	C:N
	(days)		(µmol C cop^{-1})	(µmol N cop^{-1})	
1	5	Start	19.32	2.43	6.82
			18.57	2.21	7.19
			16.15	2.64	5.24
		End	23.48	2.43	8.29
			22.56	2.78	6.95
			23.23	2.14	9.30
2	5	Start	11.32	2.21	4.39
			12.32	2.36	4.48
			9.66	2.00	4.14
			11.32	2.28	4.25
			14.15	2.57	4.72
			15.15	2.64	4.92
2	5	End	11.57	2.07	4.79
			18.98	2.78	5.85
			11.24	2.43	3.97
			10.66	2.57	3.56
			9.49	2.07	3.93
			10.32	2.21	4.00
3	3	Start	13.74	2.07	5.69
			10.41	1.93	4.63
			14.15	2.36	5.15
		End	10.74	2.43	3.79
			17.32	2.64	5.62
			13.15	2.28	4.94

Supplementary Figure 7 |The change in ratio of carbon (C) to nitrogen (N) in *C. finmarchicus* individuals in the Fram Strait in summer 2019, throughout grazing experiments. The C and N content of the experimental females did not vary significantly between the start and end of the incubations. Ex represents experiment.



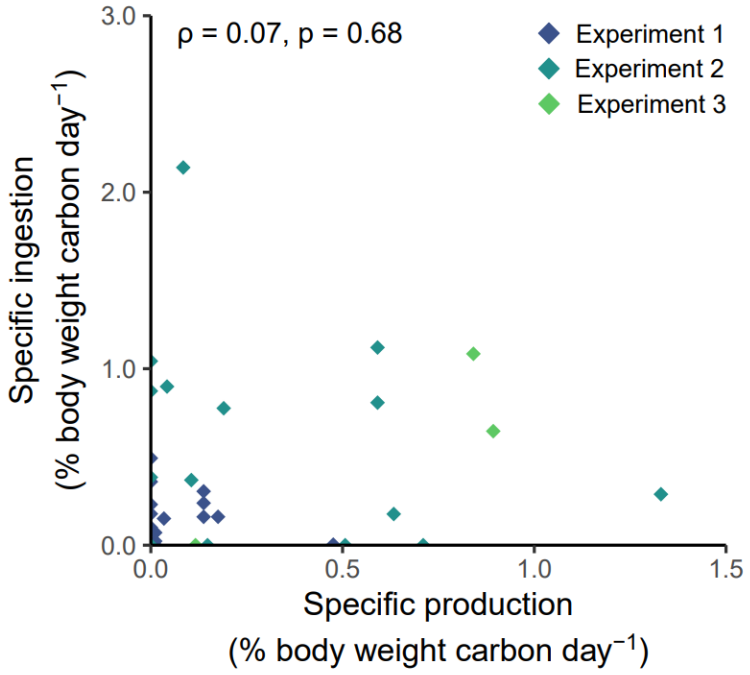
Supplementary Figure 8 | The relationships between the quantity of available food and the ingestion (A, B, C & D) and egg production rate (E, F & G, H) of *C. finmarchicus* in bottle-incubation experiments in the Fram Strait in August 2019. The quantity of available food is estimated from using the chlorophyll a concentration as a proxy (A & E) and calculated using flow imaging microscopy for all cell types (B & F), for small (S) and large (L) centric diatoms (C & G), and for flagellates (D & H). Shaded areas show 95 % confidence intervals. R is spearman's rank correlation coefficient.



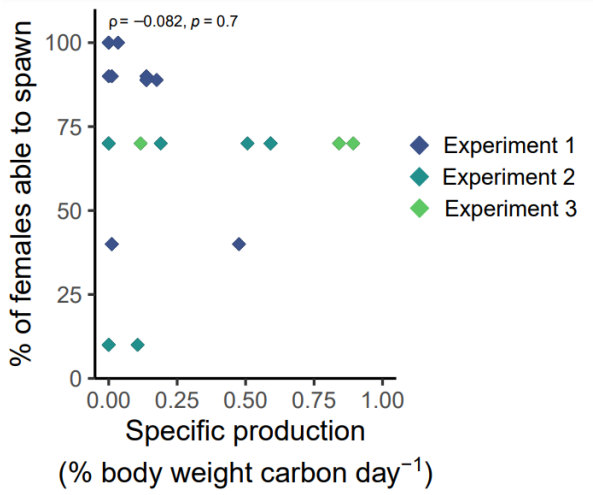
Supplementary Table 7 | The strength of the relationships between the quantities of available food types and the ingestion and egg production rate of *C. finmarchicus* in bottle-incubation experiments in the Fram Strait in August 2019. The spearman's rank correlation coefficient p-values are shown, with significant relationships in bold.

	Ingestion	Production	Specific ingestion	Specific production
Small diatoms	0.18	0.5	0.095	0.5
Large diatoms	**0.014**	0.5	**0.035**	0.62
Centric diatoms	0.086	0.4	**0.043**	0.65
Dinoflagellates	0.38	**0.046**	0.6	0.069
Ciliates	0.077	0.27	0.42	0.18
Flagellates	0.077	0.27	0.063	0.37

Supplementary Figure 9 |Specific ingestion and specific production by *C. finmarchicus* females in bottle incubation experiments in the Fram Strait in August 2019. The relationship between specific ingestion and specific production is insignificant (spearman's rank coefficient = 0.07, p = 0.68)



Supplementary Figure 10 | The specific egg production and the percentage of female *C. finmarchicus* that were spawning in experiments run alongside the experiments detailed here, in the Fram Strait in summer 2019.



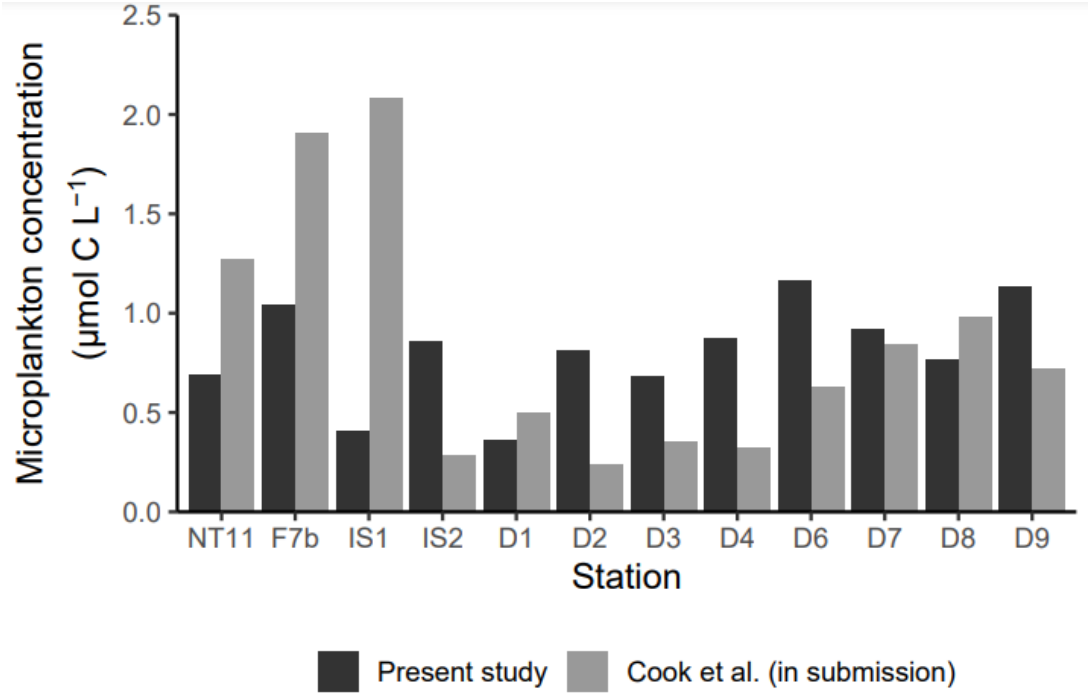
Supplementary Table 8 |Daily metabolic carbon (C) budgets for mature female *Calanus finmarchicus* (copepood^{-1} day^{-1}), showing measured ingestion and production. Showing averages per bottle (every row), per experiment (average rows), and across all (All average row). Budgets are calculated as Ingestion (I) = Egg production (E) + Respiration (R) + Egestion (W) + C surplus (Ω). Production is assumed to equal egg production only due to lack of somatic growth (Poulet et al., 1995). Respiration was estimated using nitrogen biomass-specific equations (Ikeda et al., 2001) and a respiratory quotient (RQ) of 0.97. The budgets were calculated assuming that egestion is I × (1- absorption efficiency), where absorption efficiency = 0.74 (Anderson et al., 2017, 2020; Anderson, Hessen & Mayor, 2021). The C content of the animals was 20.55 µmol C copepod^{-1} for Ex1, 12.18 µmol C copepod^{-1} for Ex2, and 13.25 µmol C copepod^{-1} for Ex3. The nitrogen (N) content of the animals was 2.44 µmol N copepod^{-1} for Ex1, 2.35 µmol N copepod^{-1} for Ex2, and 2.28 µmol N copepod^{-1} for Ex3. The C surplus was also calculated using a respiratory quotient of 0.7 (shown in "[]"). Ex, experiment; Cop, copepod; GGE, Gross Growth Efficiency, the ratio of biomass production to ingestion. The mean average is shown ± standard deviation

Appendix B

Ex	Day	Station	Ingestion (I)	Production (E)	GGE	Production (E)	Respiration (R)	Carbon required	
			(µmol C cop-1 day-1)	(µmol C cop-1 day-1)		(% C intake cop^{-1} day^{-1})	(% C intake cop^{-1} day^{-1})	(% C intake cop^{-1} day^{-1})	
1	1	F7b	0.07 ± 0.04	NIL	NA	0.00 ± 0.00	9.22 ± 8.21	8.48 ± 8.21	[5.91 ± 5.93]
1	2	IS1	0.03 ± 0.02	NIL	0.11 ± 0.16	0.11 ± 0.16	10.25 ± 3.02	9.62 ± 3.18	[6.77 ± 2.34]
1	3	IS2	0.05 ± 0.01	0.03 ± 0	0.71 ± 0.34	0.71 ± 0.34	8.53 ± 2.81	8.49 ± 3.14	[6.12 ± 2.36]
1	4	D1	0.00 ± 0.00	NIL	NA	0.5 ± 0	81.38 ± 0	81.14 ± 0	[58.48 ± 0.00]
1	5	D2	0.03 ± 0.01	0.01 ± 0.02	0.34 ± 0.46	0.34 ± 0.46	16.15 ± 8.8	15.75 ± 8.69	[11.25 ± 6.24]
1 (average)			0.04 ± 0.03	0.04 ± 0.03	0.01 ± 0.01	0.32 ± 0.37	0.32 ± 0.37	16.96 ± 21.21	[16.55 ± 21.27]
2	1	D3	0.1 ± 0.01	0.03 ± 0.04	0.33 ± 0.37	0.33 ± 0.37	3.74 ± 0.22	3.33 ± 0.52	[2.29 ± 0.47]
2	2	D4	0.03 ± 0.03	<0.01 ± 0.01	0.14 ± 0.20	0.14 ± 0.20	8.12 ± 0.24	7.52 ± 0.44	[5.26 ± 0.37]
2	3	D6	0.09 ± 0.08	0.04 ± 0.04	0.26 ± 0.37	0.26 ± 0.37	2.83 ± 0.14	2.35 ± 0.23	[1.57 ± 0.27]
2	4	D6b	0.02 ± 0.02	0.11 ± 0.05	4.10 ± 0.72	4.10 ± 0.72	13.95 ± 4.75	17.3 ± 4.03	[13.42 ± 2.71]
2	5	D6c	0.12 ± 0.13	0.01 ± 0.01	0.04 ± 0.01	0.04 ± 0.01	2.42 ± 1.40	1.72 ± 1.40	[1.05 ± 1.01]
2 (average)			0.07 ± 0.04	0.04 ± 0.05	0.92 ± 1.61	0.92 ± 1.61	5.99 ± 4.70	6.16 ± 6.03	[4.32 ± 3.39]

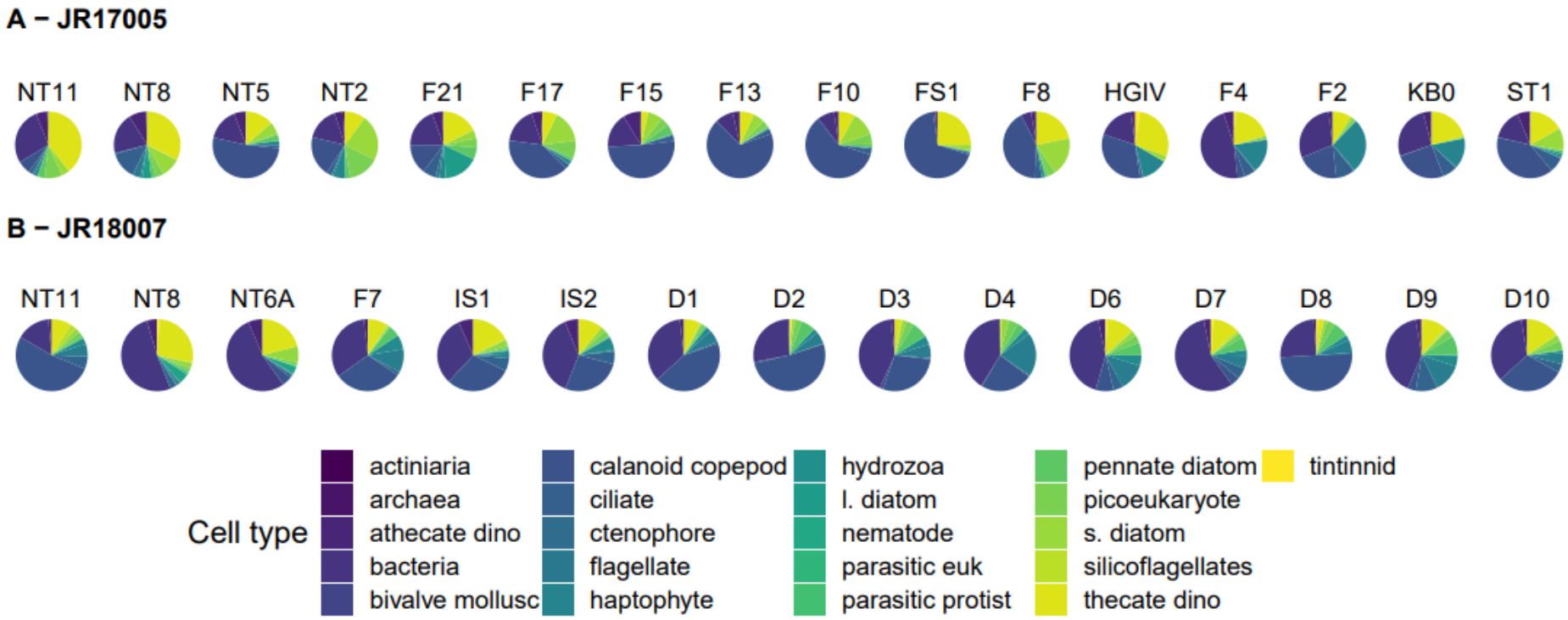
Ex	Day	Station	Ingestion (I)	Production (E)	GGE	Production (E)	Respiration (R)	Carbon required	
			(µmol C cop-1 day-1)	(µmol C cop-1 day-1)		(% C intake cop^{-1} day^{-1})	(% C intake cop^{-1} day^{-1})	(% C intake cop^{-1} day^{-1})	
3	1	D7	0.08 ± 0.07	0.08 ± 0.06	1.08 ± 0.43	1.08 ± 0.43	3.39 ± 1.21	3.73 ± 1.64	[2.79 ± 1.31]
All	(average)		0.06 ± 0.06	0.03 ± 0.04	0.64 ± 1.12	0.64 ± 1.12	11.05 ± 15.80	10.95 ± 15.91	[7.97 ± 11.40]

Supplementary Figure 11 | Microplankton biomass across stations in the Fram Strait in summer 2019. Samples were taken at different times, approximately 12 hours apart, and analysed by FlowCam® (present study) and by microscopy (Cook et al.).

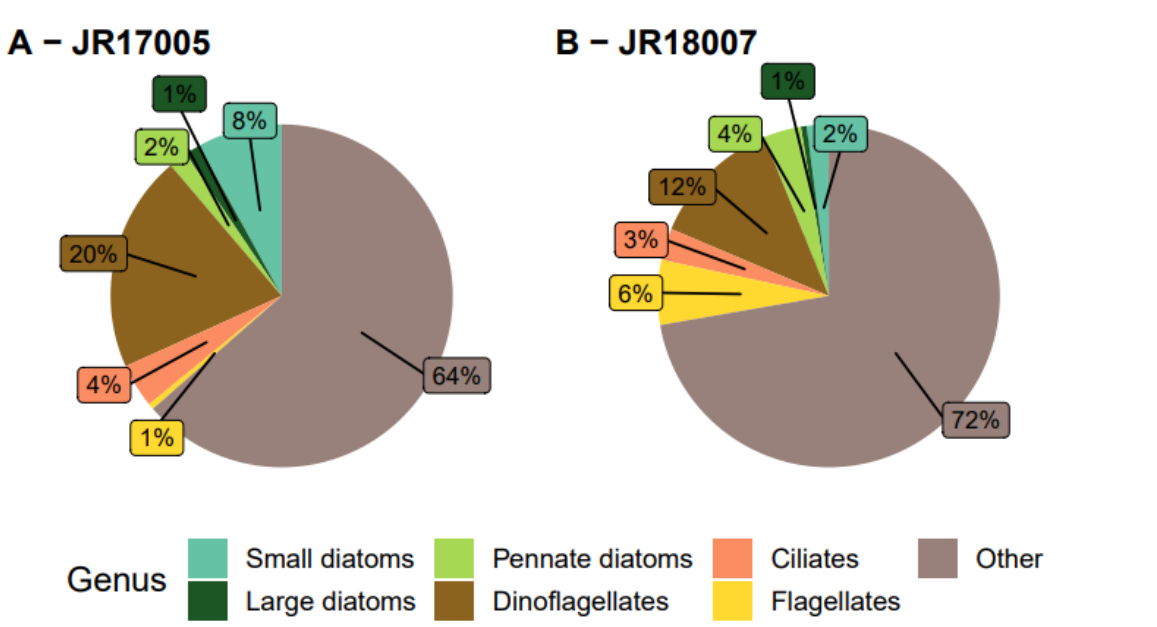


Appendix C Supplementary materials for Chapter 5

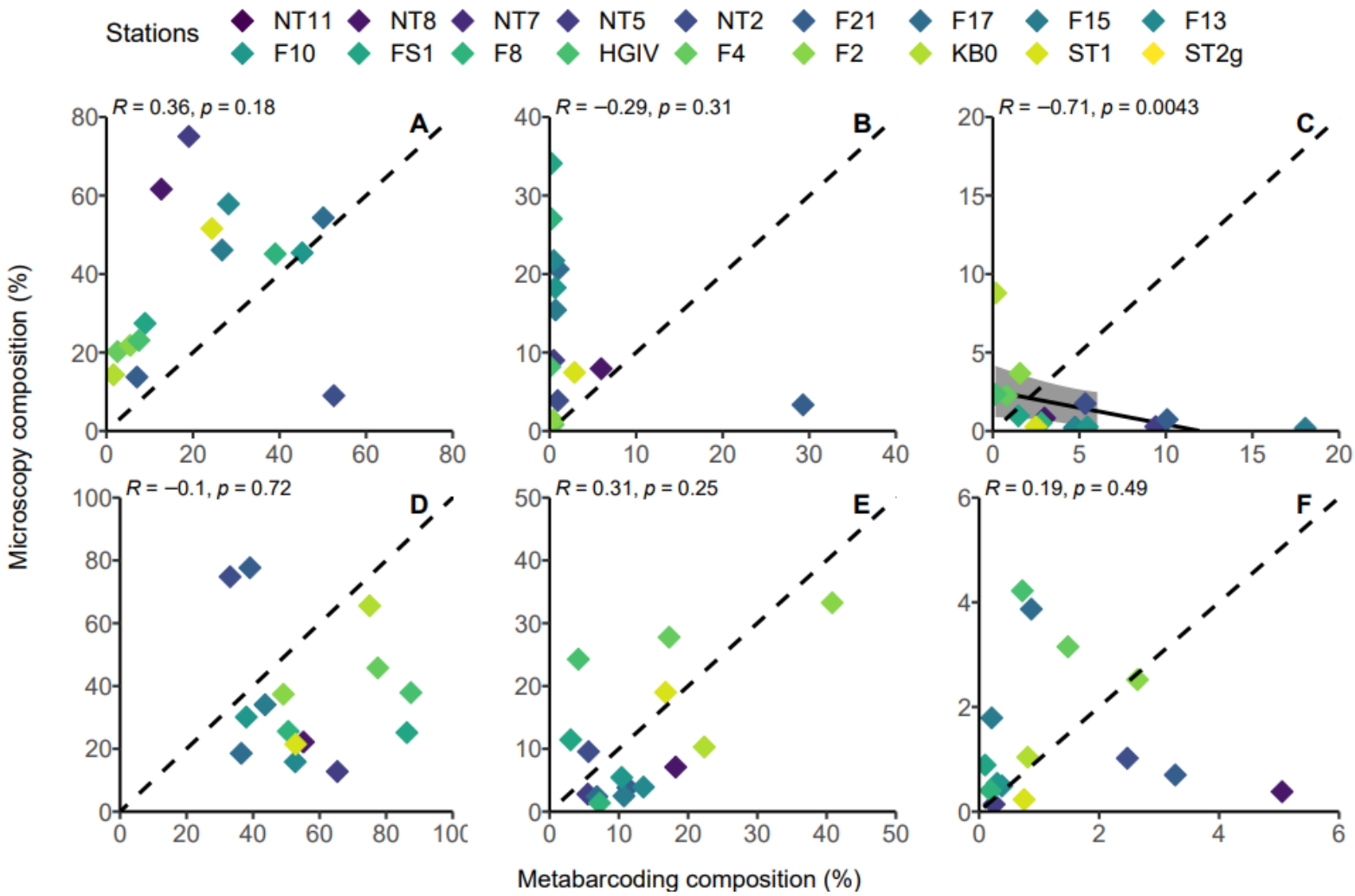
Supplementary Figure 12 | The relative composition of the seston across all stations in the Fram Strait during Spring 2018 on cruise JR17005 (A) and during Summer 2019 on cruise JR18007 (B), showing cell types divided into functional groups.



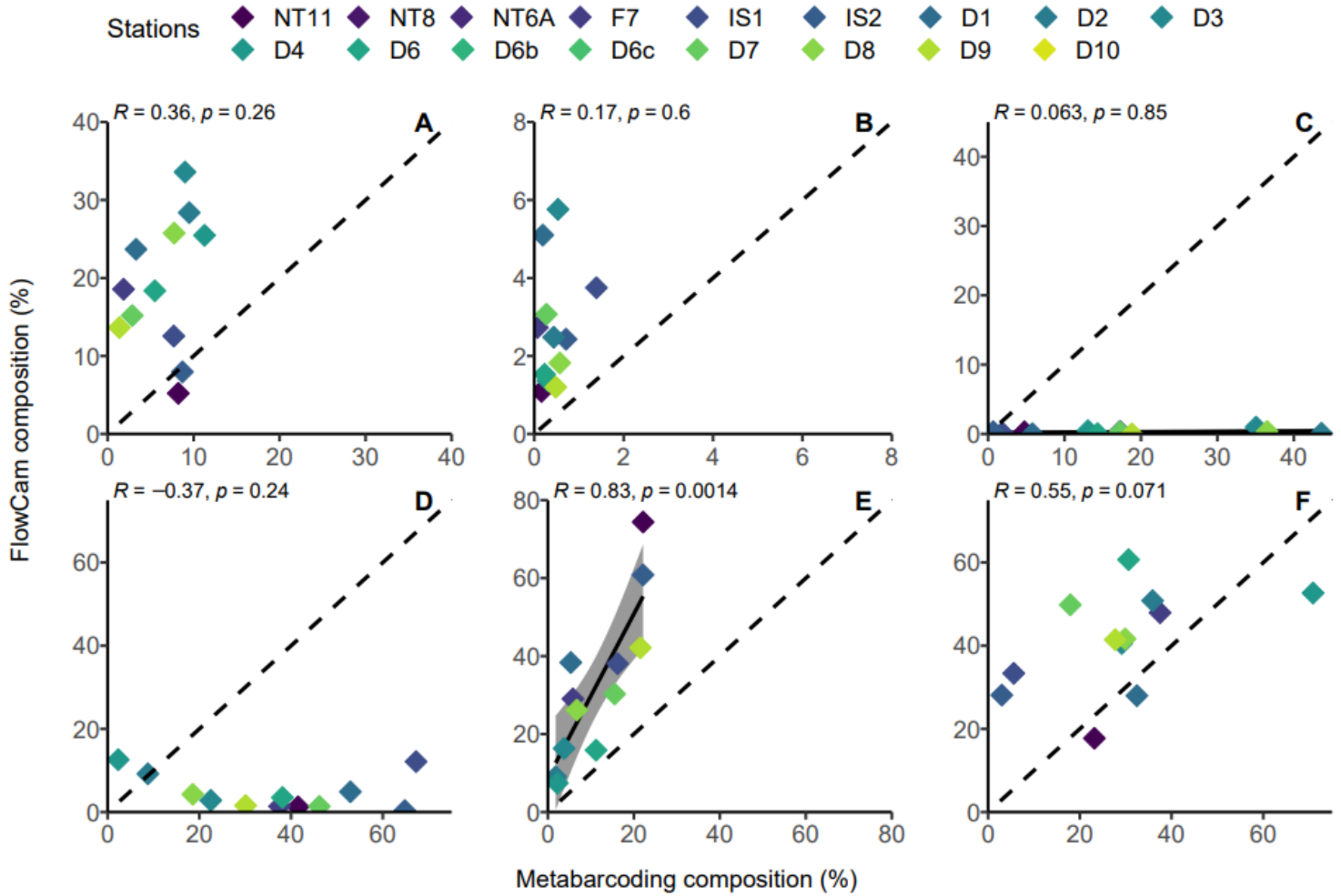
Supplementary Figure 13 | The proportional composition of the seston in the Fram Strait in spring 2018 (A) and summer 2019 (B) from metabarcoding analysis. The seston is classified into cell types based on their role in the diet of *C. finmarchicus* in the Arctic.



Supplementary Figure 14 | The relative seston composition of the seston in the Fram Strait in spring 2018 compared across different methods: inverted microscopy and metabarcoding for small diatoms (A), large diatoms (B), pennate diatoms (C), dinoflagellates (D), ciliates (E), and flagellates (F). R is Spearman's rho.



Supplementary Figure 15 | The relative seston composition of the seston in the Fram Strait in summer 2019 compared across different methods: flow imaging microscopy and metabarcoding for small diatoms (A), large diatoms (B), pennate diatoms (C), dinoflagellates (D), ciliates (E), and flagellates (F). R is Spearman's rho



Bibliography

Aarflot, J.M., Skjoldal, H.R., Dalpadado, P. & Skern-Mauritzen, M. (2018) Contribution of Calanus species to the mesozooplankton biomass in the Barents Sea. *ICES Journal of Marine Science*. 75 (7), 2342–2354. doi:10.1093/icesjms/fsx221.

Abad, D., Albaina, A., Aguirre, M., Laza-Martínez, A., Uriarte, I., Iriarte, A., Villate, F. & Estonba, A. (2016) Is metabarcoding suitable for estuarine plankton monitoring? A comparative study with microscopy. *Marine Biology*. 163 (7), 1–13. doi:10.1007/s00227-016-2920-0.

Aberle, N., Bauer, B., Lewandowska, A., Gaedke, U. & Sommer, U. (2012) Warming induces shifts in microzooplankton phenology and reduces time-lags between phytoplankton and protozoan production. *Marine Biology*. 159 (11), 2441–2453. doi:10.1007/s00227-012-1947-0.

Ackman, R.G. (1989) *Marine Biogenic Lipids, Fats, and Oils*. Boca Raton, Florida, Florida, CRC Press.

Aksenov, Y.K., Rynders, S., Feltham, D.L., Popova, E.E., Yool, A. & Heorton, H. (2021) Safer operations in changing ice-covered seas: approaches and perspectives. *Nature (in press)*. doi:https://doi.org/10.1007/978-3-030-80439-8.

Alberdi, A., Aizpurua, O., Gilbert, M.T.P. & Bohmann, K. (2018) Scrutinizing key steps for reliable metabarcoding of environmental samples. *Methods in Ecology and Evolution*. 9 (1), 134–147. doi:10.1111/2041-210X.12849.

Albers, C.S., Kattner, G. & Hagen, W. (1996) The compositions of wax esters, triacylglycerols and phospholipids in Arctic and Antarctic copepods: Evidence of energetic adaptations. *Marine Chemistry*. 55 (3–4), 347–358. doi:10.1016/S0304-4203(96)00059-X.

Alfred Wegener Institute (2022) *MOSAiC Expedition*. 2022. https://mosaic-expedition.org/ [Accessed: 28 August 2023].

AMAP (2021) *Arctic Climate Change Update 2021: Key Trends and Impacts. Summary for Policy-makers.* https://www.amap.no/documents/download/6759/inline.

Anderson, T.R., Hessen, D.O., Boersma, M., Urabe, J. & Mayor, D.J. (2017) Will Invertebrates Require Increasingly Carbon-Rich Food in a Warming World? *The American Naturalist*. 190 (6), 000–000. doi:10.1086/694122.

Anderson, T.R., Hessen, D.O., Gentleman, W.C., Yool, A. & Mayor, D.J. (2022) Quantifying the roles of food intake and stored lipid for growth and development throughout the life cycle of a high-latitude copepod, and consequences for ocean carbon sequestration. *Frontiers in Marine Science*.

9, 928209.

Anderson, T.R., Hessen, D.O. & Mayor, D.J. (2021) Is the growth of marine copepods limited by food quantity or quality? *Limnology and Oceanography Letters*. 6 (3), 127–133. doi:10.1002/lol2.10184.

Anderson, T.R., Hessen, D.O., Mitra, A., Mayor, D.J. & Yool, A. (2013) Sensitivity of secondary production and export flux to choice of trophic transfer formulation in marine ecosystem models. *Journal of Marine Systems*. 125, 41–53. doi:10.1016/j.jmarsys.2012.09.008.

Anderson, T.R. & Pond, D.W. (2000) Stoichiometric theory extended to micronutrients: comparison of the roles of essential fatty acids, carbon, and nitrogen in the nutrition of marine copepods. *Limnology and Oceanography*. 45 (5), 1162–1167. doi:10.4319/lo.2000.45.5.1162.

Anderson, T.R., Raubenheimer, D., Hessen, D.O., Jensen, K., Gentleman, W.C. & Mayor, D.J. (2020) Geometric Stoichiometry: Unifying Concepts of Animal Nutrition to Understand How Protein-Rich Diets Can Be "Too Much of a Good Thing". *Frontiers in Ecology and Evolution*. 8 (July), 1–12. doi:10.3389/fevo.2020.00196.

Andersson, A., Zhao, L., Brugel, S., Figueroa, D. & Huseby, S. (2023) Metabarcoding vs Microscopy: Comparison of Methods To Monitor Phytoplankton Communities. *ACS ES and T Water*. doi:10.1021/acsestwater.3c00176.

Ardyna, M. & Arrigo, K.R. (2020) Phytoplankton dynamics in a changing Arctic Ocean. *Nature Climate Change*. 10 (10), 892–903. doi:10.1038/s41558-020-0905-y.

Ardyna, M., Babin, M., Devred, E., Forest, A., Gosselin, M., Raimbault, P. & Tremblay, J.-É. (2017) Shelf-basin gradients shape ecological phytoplankton niches and community composition in the coastal Arctic Ocean (Beaufort Sea). *Limnology and Oceanography*. 62 (5), 2113–2132. doi:10.1002/lno.10554.

Ardyna, M., Babin, M., Gosselin, M., Devred, E., Rainville, L. & Tremblay, J.-É. (2014) Recent Arctic Ocean sea ice loss triggers novel fall phytoplankton blooms. *Geophysical Research Letters*. 41, 6207–6212. doi:10.1002/2014GL061047.

Ardyna, M., Gosselin, M., Michel, C., Poulin, M. & Tremblay, J.É. (2011) Environmental forcing of phytoplankton community structure and function in the Canadian High arctic: Contrasting oligotrophic and eutrophic regions. *Marine Ecology Progress Series*. 442, 37–57. doi:10.3354/meps09378.

Arendt, K.E., Jónasdóttir, S.H., Hansen, P.J. & Gärtner, S. (2005) Effects of dietary fatty acids on

the reproductive success of the calanoid copepod Temora longicornis. *Marine Biology*. 146 (3), 513–530. doi:10.1007/s00227-004-1457-9.

Arrigo, K.R., van Dijken, G. & Pabi, S. (2008) Impact of a shrinking Arctic ice cover on marine primary production. *Geophysical Research Letters*. 35 (19), 1–6. doi:10.1029/2008GL035028.

Arrigo, K.R., Perovich, D.K., Pickart, R.S., Brown, Z.W., Van Dijken, G.L., et al. (2012) Massive Phytoplankton Blooms Under Arctic Sea Ice. *Science*. 336 (6087), 1408. doi:10.1007/BF02390423.

Ashjian, C.J., Campbell, R.G., Welch, H.E., Butler, M. & Van Keuren, D. (2003) Annual cycle in abundance, distribution, and size in relation to hydrography of important copepod species in the western Arctic Ocean. *Deep Sea Research Part I: Oceanographic Research Papers*. 50 (10–11), 1235–1261. doi:10.1016/S0967-0637(03)00129-8.

Atherden, F. (2023) *The Metabolome of Calanus finmarchicus at High Latitudes*. University of Southampton.

Atkinson, D. (1994) Temperature and organism size-A biological law for ectotherms? Advances in Ecological Research 25: 1. *Res.* 25.

Auel, H. & Hagen, W. (2002) Mesozooplankton community structure, abundance and biomass in the central Arctic Ocean. *Marine Biology*. 140 (5), 1013–1021. doi:10.1007/s00227-001-0775-4.

Azam, F., Fenchel, T., Field, J.G., Gray, J.S., Meyer-Reil, L.A. & Thingstad, F. (1983) The Ecological Role of Water-Column Microbes in the Sea. *Marine Ecology Progress Series*. 10, 257–263. doi:10.3354/meps010257.

Bachy, C., Sudek, L., Choi, C.J., Eckmann, C.A., Nöthig, E.-M., Metfies, K. & Worden, A.Z. (2022) Phytoplankton Surveys in the Arctic Fram Strait Demonstrate the Tiny Eukaryotic Alga Micromonas and Other Picoprasinophytes Contribute to Deep Sea Export. *Microorganisms*. 10 (5). doi:10.3390/microorganisms10050961.

Båmstedt, U., Gifford, D.J., Irigoien, X., Atkinson, A. & Roman, M.R. (2000) Chapter 8 - Feeding. In: *ICES Zooplankton Methodology Manual*. pp. 297–399. doi:10.1016/B978-012327645-2/50009-8.

Båmstedt, U., Nejstgaard, J.C. & Solberg, P.T. (1999) Utilisation of small-sized food algae by Calanus finmarchicus (Copepoda, Calanoida) and the significance of feeding history. *Sarsia*. 84 (1), 19–38. doi:10.1080/00364827.1999.10420449.

Bandara, K., Varpe, Ø., Maps, F., Ji, R., Eiane, K. & Tverberg, V. (2021) Timing of Calanus finmarchicus diapause in stochastic environments. *Ecological Modelling*. 460 (September).

doi:10.1016/j.ecolmodel.2021.109739.

Barber, D.G., Hop, H., Mundy, C.J., Else, B., Dmitrenko, I.A., Tremblay, J.-É., Ehn, J.K., Assmy, P., Daase, M., Candlish, L.M. & Rysgaard, S. (2015) Selected physical, biological and biogeochemical implications of a rapidly changing Arctic Marginal Ice Zone. *Progress in Oceanography*. 139, 122–150. doi:10.1016/j.pocean.2015.09.003.

Basedow, S.L., Sundfjord, A., von Appen, W.-J., Halvorsen, E., Kwasniewski, S. & Reigstad, M. (2018) Seasonal Variation in Transport of Zooplankton Into the Arctic Basin Through the Atlantic Gateway, Fram Strait. *Frontiers in Marine Science*. 5 (June), 1–22. doi:10.3389/fmars.2018.00194.

Bates, S.S., Bird, C.J., Freitas, A.S.W. de, Foxall, R., Gilgan, M., Hanic, L.A., Johnson, G.R., McCulloch, A.W., Odense, P., Pocklington, R., Quilliam, M.A., Sim, P.G., Smith, J.C., Rao, D.V.S., Todd, E.C.D., Walter, J.A. & Wright, J.L.C. (1989) Pennate Diatom Nitzschia pungens as the Primary Source of Domoic Acid, a Toxin in Shellfish from Eastern Prince Edward Island, Canada. *Canadian Journal of Fisheries and Aquatic Sciences*. 46 (7), 1203–1215. doi:10.1139/f89-156.

Bauerfeind, E., Nöthig, E.-M., Beszczynska, A., Fahl, K., Kaleschke, L., Kreker, K., Klages, M., Soltwedel, T., Lorenzen, C. & Wegner, J. (2009) Particle sedimentation patterns in the eastern Fram Strait during 2000-2005: Results from the Arctic long-term observatory HAUSGARTEN. *Deep-Sea Research Part I: Oceanographic Research Papers*. 56 (9), 1471–1487. doi:10.1016/j.dsr.2009.04.011.

Baumgartner, M.F. & Tarrant, A.M. (2017) The Physiology and Ecology of Diapause in Marine Copepods. *Annual Review of Marine Science*. 9 (1), 387–411. doi:10.1146/annurev-marine-010816-060505.

Bell, M. V., Dick, J.R., Anderson, T.R. & Pond, D.W. (2007) Application of liposome and stable isotope tracer techniques to study polyunsaturated fatty acid biosynthesis in marine zooplankton. *Journal of Plankton Research*. 29 (5), 417–422. doi:10.1093/plankt/fbm025.

Benkort, D., Daewel, U., Heath, M. & Schrum, C. (2020) On the Role of Biogeochemical Coupling Between Sympagic and Pelagic Ecosystem Compartments for Primary and Secondary Production in the Barents Sea. *Frontiers in Environmental Science*. 8, 217. doi:10.3389/FENVS.2020.548013/BIBTEX.

Berge, J., Cottier, F.R., Varpe, Ø., Renaud, P.E., Falk-Petersen, S., Kwaśniewski, S., Griffiths, C., Søreide, J.E., Johnsen, G., Aubert, A.B., Bjærke, O., Hovinen, J., Jung-Madsen, S., Tveit, M. & Majaneva, S. (2014) Arctic complexity: A case study on diel vertical migration of zooplankton. *Journal of Plankton Research*. 36 (5), 1279–1297. doi:10.1093/plankt/fbu059.

Berge, J., Daase, M., Renaud, P.E., Ambrose, W.G., Darnis, G., et al. (2015) Unexpected levels of biological activity during the polar night offer new perspectives on a warming arctic. *Current Biology*. 25 (19), 2555–2561. doi:10.1016/j.cub.2015.08.024.

Berggreen, U., Hansen, B.W. & Kiørboe, T. (1988) Food size spectra, ingestion and growth of the copepod Acartia tonsa during development: Implications for determination of copepod production. *Marine Biology*. 99 (3), 341–352. doi:10.1007/BF02112126.

Besiktepe, S. & Dam, H.G. (2002) Coupling of ingestion and defecation as a function of diet in the calanoid copepod Acartia tonsa. *Marine Ecology Progress Series*. 229 (September 2014), 151–164. doi:10.3354/meps229151.

Blachowiak-Samolyk, K., Kwasniewski, S., Dmoch, K., Hop, H. & Falk-Petersen, S. (2007) Trophic structure of zooplankton in the Fram Strait in spring and autumn 2003. *Deep-Sea Research Part II: Topical Studies in Oceanography*. 54 (23–26), 2716–2728. doi:10.1016/j.dsr2.2007.08.004.

Bluhm, B.A., Gebruk, A. V., Gradinger, R., Hopcroft, R.R., Huettmann, F. & Kosobokova, K.N. (2011) Arctic marine biodiversity: An update of species richness and examples of biodiversity change. *Oceanography*. 24 (3), 232–248. http://dx.doi.org/10.5670/oceanog.2011.80.

Bochdansky, A.B., Deibel, D. & Rivkin, R.B. (1999) Absorption efficiencies and biochemical fractionation of assimilated compounds in the cold water appendicularian Oikopleura vanhoeffeni. *Limnology and Oceanography*. 44 (2), 415–424. doi:10.4319/lo.1999.44.2.0415.

Bonnet, D., Titelman, J. & Harris, R.P. (2004) Calanus the cannibal. *Journal of Plankton Research*. 26 (8), 937–948. doi:10.1093/plankt/fbh087.

Bopp, L., Monfray, P., Aumont, O., Dufresne, J.L., Le Treut, H., Madec, G., Terray, L. & Orr, J.C. (2001) Potential impact of climate change on marine export production. *Global Biogeochemical Cycles*. 15 (1), 81–99. doi:10.1029/1999GB001256.

Boxshall, G.A. & Defaye, D. (2008) Global diversity of copepods (Crustacea: Copepoda) in freshwater. *Hydrobiologia*. 595 (1), 195–207. doi:10.1007/s10750-007-9014-4.

Broglio, E., Jónasdóttir, S.H., Calbet, A., Jakobsen, H.H. & Saiz, E. (2003) Effect of heterotrophic versus autotrophic food on feeding and reproduction of the calanoid copepod Acartia tonsa: Relationship with prey fatty acid composition. *Aquatic Microbial Ecology*. 31 (3), 267–278. doi:10.3354/ame031267.

Bruhn, C.S., Wohlrab, S., Krock, B., Lundholm, N. & John, U. (2021) Seasonal plankton succession is in accordance with phycotoxin occurrence in Disko Bay, West Greenland. *Harmful Algae*. 103

(January), 101978. doi:10.1016/j.hal.2021.101978.

Brun, P., Stamieszkin, K., Visser, A.W., Licandro, P., Payne, M.R. & Kiørboe, T. (2019) Climate change has altered zooplankton-fuelled carbon export in the North Atlantic. *Nature Ecology & Evolution*. 3 (3), 416–423. doi:10.1038/s41559-018-0780-3.

Calbet, A. & Saiz, E. (2005) The ciliate-copepod link in marine ecosystems. *Aquatic Microbial Ecology*. 38 (2), 157–167. http://www.researchgate.net/publication/230660217_The_ciliate-copepod_link_in_marine_ecosystems/file/5046351ac501e1b0be.pdf%0Ahttp://www.int-res.com/articles/ame2004/38/a038p157.pdf%0Apapers2://publication/uuid/A856D2A0-BE63-453B-9487-C56B6B5C3E2A.

Caldeira, K. & Wickett, M.E. (2003) Oceanography: Anthropogenic carbon and ocean pH. *Nature*. 425 (6956), 365–365. doi:10.1038/425365a.

Campbell, K., Lange, B.A., Landy, J.C., Katlein, C., Nicolaus, M., Anhaus, P., Matero, I., Gradinger, R.R., Charette, J., Duerksen, S., Tremblay, P.O.G., Rysgaard, S., Tranter, M., Haas, C. & Michel, C. (2021) Widespread net heterotrophy in High Arctic first-year and multi-year sea ice. *Elementa Science of the Anthropocene*. (in review.

Caporaso, J.G., Kuczynski, J., Stombaugh, J., Bittinger, K., Bushman, F.D., et al. (2010) QIIME allows analysis of high-throughput community sequencing data. *Nature Methods*. 7 (5), 1–12. doi:10.1038/nmeth.f.303.

Carmack, E.C. & Chapman, D.C. (2003) Wind-driven shelf/basin exchange on an Arctic shelf: The joint roles of ice cover extent and shelf-break bathymetry. *Geophysical Research Letters*. 30 (14). doi:10.1029/2003GL017526.

Carnicer, O., Irwin, A.J. & Finkel, Z. V. (2022) Traits influence dinoflagellate C:N:P. *European Journal of Phycology*. 57 (2), 154–165. doi:10.1080/09670262.2021.1914860.

Carozza, D.A., Bianchi, D. & Galbraith, E.D. (2019) Metabolic impacts of climate change on marine ecosystems: Implications for fish communities and fisheries. *Global Ecology and Biogeography*. 28 (2), 158–169. doi:10.1111/geb.12832.

Carroll, E.L., Gallego, R., Sewell, M.A., Zeldis, J., Ranjard, L., Ross, H.A., Tooman, L.K., O'Rorke, R., Newcomb, R.D. & Constantine, R. (2019) Multi-locus DNA metabarcoding of zooplankton communities and scat reveal trophic interactions of a generalist predator. *Scientific Reports*. 9 (1), 1–14. doi:10.1038/s41598-018-36478-x.

Castellani, C., Irigoien, X., Mayor, D.J., Harris, R.P. & Wilson, D. (2008) Feeding of Calanus

finmarchicus and Oithona similis on the microplankton assemblage in the Irminger Sea, North Atlantic. *Journal of Plankton Research*. 30 (10), 1095–1116. doi:10.1093/plankt/fbn074.

Castellani, G., Veyssière, G., Karcher, M., Stroeve, J., Banas, N.S., Bouman, A.H., Brierley, S.A., Connan, S., Cottier, F., Große, F., Hobbs, L., Katlein, C., Light, B., McKee, D., Orkney, A., Proud, R. & Schourup-Kristensen, V. (2022a) Shine a light: Under-ice light and its ecological implications in a changing Arctic Ocean. *Ambio*. 51 (2), 307–317. doi:10.1007/s13280-021-01662-3.

Castellani, G., Veyssière, G., Karcher, M., Stroeve, J., Banas, N.S., Bouman, H.A., Brierley, A.S., Connan, S., Cottier, F., Große, F., Hobbs, L., Katlein, C., Light, B., McKee, D., Orkney, A., Proud, R. & Schourup-Kristensen, V. (2022b) Shine a light: Under-ice light and its ecological implications in a changing Arctic Ocean. *Ambio*. 51 (2), 307–317. doi:10.1007/s13280-021-01662-3.

Cavan, E.L., Henson, S.A., Belcher, A. & Sanders, R. (2017) Role of zooplankton in determining the efficiency of the biological carbon pump. *Biogeosciences*. 14 (1), 177–186. doi:10.5194/bg-14-177-2017.

Cerfonteyn, M., Groben, R., Vaulot, D., Guðmundsson, K., Vannier, P., Pérez-Hernández, M.D. & Marteinsson, V.Þ. (2023) The distribution and diversity of eukaryotic phytoplankton in the Icelandic marine environment. *Scientific Reports*. 13 (1), 1–15. doi:10.1038/s41598-023-35537-2.

Chen, B., Landry, M.R., Huang, B. & Liu, H. (2012) Does warming enhance the effect of microzooplankton grazing on marine phytoplankton in the ocean? *Limnology and Oceanography*. 57 (2), 519–526. doi:10.4319/lo.2012.57.2.0519.

Chen, M., Liu, H. & Chen, B. (2012) Effects of dietary essential fatty acids on reproduction rates of a subtropical calanoid copepod, Acartia erythraea. *Marine Ecology Progress Series*. 455, 95–110. doi:10.3354/meps09685.

Chust, G., Castellani, C., Licandro, P., Ibaibarriaga, L., Sagarminaga, Y. & Irigoien, X. (2014) Are Calanus spp. shifting poleward in the North Atlantic? A habitat modelling approach. *ICES Journal of Marine Science*. 71 (2), 241–253. doi:10.1093/icesjms/fst147.

Clark, D.R., Flynn, K.J. & Fabian, H. (2014) Variation in elemental stoichiometry of the marine diatom Thalassiosira weissflogii (Bacillariophyceae) in response to combined nutrient stress and changes in carbonate chemistry. *Journal of Phycology*. 50 (4), 640–651. doi:10.1111/jpy.12208.

Cleary, A.C., Søreide, J.E., Freese, D., Niehoff, B. & Gabrielsen, T.M. (2017) Feeding by Calanus glacialis in a high arctic fjord: Potential seasonal importance of alternative prey. *ICES Journal of Marine Science*. 74 (7), 1937–1946. doi:10.1093/icesjms/fsx106.

Coello-Camba, A., Agustí, S., Holding, J., Arrieta, J.M. & Duarte, C.M. (2014) Interactive effect of temperature and CO2 increase in Arctic phytoplankton. *Frontiers in Marine Science*. 1 (OCT), 1–10. doi:10.3389/fmars.2014.00049.

Cole, M., Coppock, R., Lindeque, P.K., Altin, D., Reed, S., Pond, D.W., Sørensen, L., Galloway, T.S. & Booth, A.M. (2019) Effects of Nylon Microplastic on Feeding, Lipid Accumulation, and Moulting in a Coldwater Copepod. *Environmental Science and Technology*. 53 (12), 7075–7082. doi:10.1021/acs.est.9b01853.

Comeau, A.M., Li, W.K.W., Tremblay, J.-É., Carmack, E.C. & Lovejoy, C. (2011) Arctic ocean microbial community structure before and after the 2007 record sea ice minimum. *PLoS ONE*. 6 (11). doi:10.1371/journal.pone.0027492.

Conover, R.J. (1988) Comparative life histories in the genera Calanus and Neocalanus in high latitudes of the northern hemisphere. *Hydrobiologia*. 167–168 (1), 127–142. doi:10.1007/BF00026299.

Corkett, C.J., McLaren, I.A. & Sevigny, J.-M. (1986) The rearing of the marine calanoid copepods Calanus finmarchicus (Gunnerus), C. glacialis Jaschnov and C. hyperboreus Kroyer with comment on the equiproportional rule. *Syllogeus*. 58 (May 2016), 539–546.

Corner, E.D.S., Head, R.N. & Kilvington, C.C. (1972) On the nutrition and metabolism of zooplankton. VIII. the grazing of Biddulphia cells by Calanus helgolandicus. *Journal of the Marine Biological Association of the United Kingdom*. 52 (4), 847–861. doi:10.1017/S0025315400040595.

Cowles, T.J., Olson, R.J. & Chisholm, S.W. (1988) Food selection by copepods: discrimination on the basis of food quality. *Marine Biology*. 100 (1), 41–49. doi:10.1007/BF00392953.

Dai, A., Luo, D., Song, M. & Liu, J. (2019) Arctic amplification is caused by sea-ice loss under increasing CO2. *Nature Communications*. 10 (1), 1–13. doi:10.1038/s41467-018-07954-9.

Darnis, G., Hobbs, L., Geoffroy, M., Grenvald, J.C., Renaud, P.E., Berge, J., Cottier, F.R., Kristiansen, S., Daase, M., Søreide, J.E., Wold, A., Morata, N. & Gabrielsen, T.M. (2017) From polar night to midnight sun : Diel vertical migration , metabolism and biogeochemical role of zooplankton in a high Arctic fjord. *Limnology and Oceanography*. 62, 1586–1605. doi:10.1002/lno.10519.

Daufresne, M., Lengfellner, K. & Sommer, U. (2009) Global warming benefits the small in aquatic ecosystems. *Proceedings of the National Academy of Sciences*. 106 (3), 12788–12793.

Degen, R., Aune, M., Bluhm, B.A., Cassidy, C., Kędra, M., Kraan, C., Vandepitte, L., Włodarska-Kowalczuk, M., Zhulay, I., Albano, P.G., Bremner, J., Grebmeier, J.M., Link, H., Morata, N.,

Nordström, M.C., Shojaei, M.G., Sutton, L. & Zuschin, M. (2018) Trait-based approaches in rapidly changing ecosystems: A roadmap to the future polar oceans. *Ecological Indicators*. 91 (April), 722–736. doi:10.1016/j.ecolind.2018.04.050.

Díez, B., Van Nieuwerburgh, L. & Snoeijs, P. (2013) Water nutrient stoichiometry modifies the nutritional quality of phytoplankton and somatic growth of crustacean mesozooplankton. *Marine Ecology Progress Series*. 489, 93–105. doi:10.3354/meps10438.

Diffenbaugh, N.S. & Barnes, E.A. (2023) Data-driven predictions of the time remaining until critical global warming thresholds are reached. *Proceedings of the National Academy of Sciences*. 120 (6), 2017. doi:10.1073/pnas.2207183120.

Dischereit, A., Wangensteen, O.S., Præbel, K., Auel, H. & Havermans, C. (2022) Using DNA Metabarcoding to Characterize the Prey Spectrum of Two Co-Occurring Themisto Amphipods in the Rapidly Changing Atlantic-Arctic Gateway Fram Strait. *Genes*. 13 (11). doi:10.3390/genes13112035.

Djeghri, N., Atkinson, A., Fileman, E.S., Harmer, R.A., Widdicombe, C.E., McEvoy, A.J., Cornwell, L. & Mayor, D.J. (2018) High prey-predator size ratios and unselective feeding in copepods: A seasonal comparison of five species with contrasting feeding modes. *Progress in Oceanography*. 165 (April), 63–74. doi:10.1016/j.pocean.2018.04.013.

Ducklow, H.W., Steinberg, D.K., William, C., Point, M.G. & Buesseler, K.O. (2001) Upper Ocean Carbon Export and the Biological Pump. *Oceanography*. 14 (4), 50–58.

Eberlein, T., Van De Waal, D.B., Brandenburg, K.M., John, U., Voss, M., Achterberg, E.P. & Rost, B. (2016) Interactive effects of ocean acidification and nitrogen limitation on two bloom-forming dinoflagellate species. *Marine Ecology Progress Series*. 543, 127–140. doi:10.3354/meps11568.

Egge, E., Elferink, S., Vaulot, D., John, U., Bratbak, G., Larsen, A. & Edvardsen, B. (2021) An 18S V4 rRNA metabarcoding dataset of protist diversity in the Atlantic inflow to the Arctic Ocean, through the year and down to 1000m depth. *Earth System Science Data*. 13 (10), 4913–4928. doi:10.5194/essd-13-4913-2021.

Engel, A., Bracher, A., Dinter, T., Endres, S., Grosse, J., Metfies, K., Peeken, I., Piontek, J., Salter, I. & Nöthig, E.-M. (2019) Inter-annual variability of organic carbon concentrations in the eastern Fram Strait during summer (2009-2017). *Frontiers in Marine Science*. 6 (MAR), 1–17. doi:10.3389/fmars.2019.00187.

Esenkulova, S., Neville, C., DiCicco, E. & Pearsall, I. (2022) Indications that algal blooms may affect

wild salmon in a similar way as farmed salmon. *Harmful Algae*. 118 (April), 102310. doi:10.1016/j.hal.2022.102310.

Esenkulova, S., Sutherland, B.J.G., Tabata, A., Haigh, N., Pearce, C.M. & Miller, K.M. (2020) Operational taxonomic unit comparing metabarcoding and morphological approaches to identify phytoplankton taxa associated with harmful algal blooms. *Facets*. 5 (1), 784–811. doi:10.1139/FACETS-2020-0025.

Fadeev, E., Salter, I., Schourup-Kristensen, V., Nöthig, E.-M., Metfies, K., Engel, A., Piontek, J., Boetius, A. & Bienhold, C. (2018) Microbial communities in the east and west Fram Strait during sea ice melting season. *Frontiers in Marine Science*. 5 (NOV), 1–21. doi:10.3389/fmars.2018.00429.

Falk-Petersen, S., Mayzaud, P., Kattner, G. & Sargent, J.R. (2009) Lipids and life strategy of Arctic Calanus. *Marine Biology Research*. 5 (1), 18–39. doi:10.1080/17451000802512267.

Falk-Petersen, S., Timofeev, S., Pavlov, V. & Sargent, J.R. (2007) Climate variability and the effect on Arctic food chains. The role of Calanus. In: J.R. Ørbæk, T. Tombre, R. Kallenborn, E. Hegseth, S. Falk-Petersen, & A.H. Hoel (eds.). *Arctic-Alpine Ecosystems and People in a Changing Environment*. Springer, Berlin. pp. 147–166.

Falkowski, P.G. (1998) Biogeochemical Controls and Feedbacks on Ocean Primary Production. *Science*. 281 (5374), 200–206. doi:10.1126/science.281.5374.200.

Farmer, J.R., Sigman, D.M., Granger, J., Underwood, O.M., Fripiat, F., Cronin, T.M., Martínez-García, A. & Haug, G.H. (2021) Arctic Ocean stratification set by sea level and freshwater inputs since the last ice age. *Nature Geoscience*. 14 (9), 684–689. doi:10.1038/s41561-021-00789-y.

Finkel, Z. V., Beardall, J., Flynn, K.J., Quigg, A., Rees, T.A. V. & Raven, J.A. (2010) Phytoplankton in a changing world: Cell size and elemental stoichiometry. *Journal of Plankton Research*. 32 (1), 119–137. doi:10.1093/plankt/fbp098.

Finkel, Z. V., Katz, M.E., Wright, J.D., Schofield, O.M.E. & Falkowski, P.G. (2005) Climatically driven macroevolutionary patterns in the size of marine diatoms over the Cenozoic. *Proceedings of the National Academy of Sciences of the United States of America*. 102 (25), 8927–8932. doi:10.1073/pnas.0409907102.

Fonseca, V.G., Carvalho, G.R., Sung, W., Johnson, H.F., Power, D.M., Neill, S.P., Packer, M., Blaxter, M.L., Lambshead, P.J.D., Thomas, W.K. & Creer, S. (2010) Second-generation environmental sequencing unmasks marine metazoan biodiversity. *Nature Communications*. 1 (7).

doi:10.1038/ncomms1095.

Fortunato, S., Bergstrom, C.T., Börner, K., Evans, J.A., Helbing, D., Milojević, S., Petersen, A.M., Radicchi, F., Sinatra, R., Uzzi, B., Vespignani, A., Waltman, L., Wang, D. & Barabási, A.L. (2018) Science of science. *Science*. 359 (6379). doi:10.1126/science.aao0185.

Fowler, S.W. & Knauer, G.A. (1986) Role of large particles in the transport of elements and organic compounds through the oceanic water column. *Progress in Oceanography*. doi:10.1016/0079-6611(86)90032-7.

Frater, D. (2021) *Diversity in UK Polar Science Initiative Race Impact Survey Report*. (February). https://www.bas.ac.uk/data/our-data/publication/diversity-in-polar-science-race-impact-survey-report/.

Freer, J.J., Daase, M. & Tarling, G.A. (2021) Modelling the biogeographic boundary shift of Calanus finmarchicus reveals drivers of Arctic Atlantification by subarctic zooplankton. *Global Change Biology*. 00, 1–12. doi:10.1111/GCB.15937.

Frost, B.W. (1972) Effect of size and concentration of food particles on the feeding behaviour of the marine planktonic copepod Calanus pacificus. *Limnology and Oceanography*. 1972 (17), 805–815. doi:10.4319/lo.1972.17.6.0805.

Gamble, J.C. (1978) Copepod grazing during a declining spring phytoplankton bloom in the Northern North Sea. *Marine Biology*. 49 (4), 303–315. doi:10.1007/BF00455025.

Garnier, S. (2018) *viridis: Default Color Maps from 'matplotlib'*. https://cran.r-project.org/package=viridis.

Gatten, R.R. & Sargent, J.R. (1973) Wax ester biosynthesis in calanoid copepods in relation to vertical migration. *Netherlands Journal of Sea Research*. 7 (7), 150–158. doi:10.2307/2389678.

Gill, C.W. & Harris, R.P. (1987) Behavioural responses of the copepods Calanus helgolandicus and Temora longicornis to dinoflagellate diets. *Journal of the Marine Biological Association of the United Kingdom*. 67 (4), 785–801.

Gillooly, J.F., Brown, J.H., West, G.B., Savage, V.M. & Charnov, E.L. (2001) Effects of size and temperature on metabolic rate. *Science*. 293 (5538), 2248–2251. doi:10.1126/science.1061967.

Glibert, P.M., Icarus Allen, J., Artioli, Y., Beusen, A., Bouwman, L., Harle, J., Holmes, R. & Holt, J. (2014) Vulnerability of coastal ecosystems to changes in harmful algal bloom distribution in response to climate change: Projections based on model analysis. *Global Change Biology*. 20 (12),

3845–3858. doi:10.1111/gcb.12662.

Glibert, P.M., Wilkerson, F.P., Dugdale, R.C., Raven, J.A., Dupont, C.L., Leavitt, P.R., Parker, A.E., Burkholder, J.M. & Kana, T.M. (2016) Pluses and minuses of ammonium and nitrate uptake and assimilation by phytoplankton and implications for productivity and community composition, with emphasis on nitrogen-enriched conditions. *Limnology and Oceanography*. 61 (1), 165–197. doi:10.1002/lno.10203.

Gluchowska, M., Dalpadado, P., Beszczynska-Möller, A., Olszewska, A., Ingvaldsen, R.B. & Kwaśniewski, S. (2017) Interannual zooplankton variability in the main pathways of the Atlantic water flow into the Arctic Ocean (Fram Strait and Barents Sea branches). *ICES Journal of Marine Science*. 74 (7), 1921–1936. doi:10.1093/icesjms/fsx033.

Glud, R.N., Grossart, H.P., Larsen, M., Tang, K.W., Arendt, K.E., Rysgaard, S., Thamdrup, B. & Nielsen, T.G. (2015) Copepod carcasses as microbial hot spots for pelagic denitrification. *Limnology and Oceanography*. 60 (6), 2026–2036. doi:10.1002/lno.10149.

Gobler, C.J., Doherty, O.M., Hattenrath-Lehmann, T.K., Griffith, A.W., Kang, Y. & Litaker, R.W. (2017) Ocean warming since 1982 has expanded the niche of toxic algal blooms in the North Atlantic and North Pacific oceans. *Proceedings of the National Academy of Sciences of the United States of America*. 114 (19), 4975–4980. doi:10.1073/pnas.1619575114.

Gobler, C.J., Lonsdale, D.J. & Boyer, G.L. (2005) A Review of the Causes, Effects, and Potential Management of Harmful Brown Tide Blooms Caused by Aureococcus anophagefferens (Hargraves et Sieburth). *Estuaries*. 28 (5), 726–749.

Gong, W. & Marchetti, A. (2019) Estimation of 18S gene copy number in marine eukaryotic plankton using a next-generation sequencing approach. *Frontiers in Marine Science*. 6 (APR), 1–5. doi:10.3389/fmars.2019.00219.

Goszczko, I., Ingvaldsen, R.B. & Onarheim, I.H. (2018) Wind-Driven Cross-Shelf Exchange—West Spitsbergen Current as a Source of Heat and Salt for the Adjacent Shelf in Arctic Winters. *Journal of Geophysical Research: Oceans*. 123 (4), 2668–2696. doi:10.1002/2017JC013553.

Gradinger, R.R. & Baumann, M.E.M. (1991) Distribution of phytoplankton communities in relation to the large-scale hydrographical regime in the Fram Strait. *Marine Biology*. 111 (2), 311–321. doi:10.1007/BF01319714.

Gran-Stadniczeñko, S., Egge, E., Hostyeva, V., Logares, R., Eikrem, W. & Edvardsen, B. (2019) Protist Diversity and Seasonal Dynamics in Skagerrak Plankton Communities as Revealed by

Metabarcoding and Microscopy. *Journal of Eukaryotic Microbiology*. 66 (3), 494–513. doi:10.1111/jeu.12700.

Greene, C.H. & Pershing, A.J. (2007) Climate Drives Sea Change. *Science*. 315 (5815), 1084–1085. doi:10.1126/science.1136495.

Guérin, N., Ciccarella, M., Flamant, E., Frémont, P., Mangenot, S., et al. (2022) Genomic adaptation of the picoeukaryote Pelagomonas calceolata to iron-poor oceans revealed by a chromosome-scale genome sequence. *Communications Biology*. 5 (1). doi:10.1038/s42003-022-03939-z.

Haecky, P., Jonsson, S. & Andersson, A. (1998) Influence of sea ice on the composition of the spring phytoplankton bloom in the northern Baltic Sea. *Polar Biology*. 20 (1), 1–8. doi:10.1007/s003000050270.

Häfker, N.S., Teschke, M., Last, K.S., Pond, D.W., Hüppe, L. & Meyer, B. (2018) Calanus finmarchicus seasonal cycle and diapause in relation to gene expression, physiology, and endogenous clocks. *Limnology and Oceanography*. 63 (6), 2815–2838. doi:10.1002/lno.11011.

Haine, T.W.N., Curry, B., Gerdes, R., Hansen, E., Karcher, M., Lee, C., Rudels, B., Spreen, G., de Steur, L., Stewart, K.D. & Woodgate, R. (2015) Arctic freshwater export: Status, mechanisms, and prospects. *Global and Planetary Change*. 125, 13–35. doi:10.1016/j.gloplacha.2014.11.013.

Haine, T.W.N. & Martin, T. (2017) The Arctic-Subarctic sea ice system is entering a seasonal regime: Implications for future Arctic amplification. *Scientific Reports*. 7 (1), 1–9. doi:10.1038/s41598-017-04573-0.

Hansen, B.W., Tande, K.S. & Berggreen, U.C. (1990) On the trophic fate of Phaeocystis pouchetii (Harlot). III. Functional responses in grazing demonstrated on juvenile stages of Calanus finmarchicus (Copepoda) fed diatoms and Phaeocystis. *Journal of Plankton Research*. 12 (6), 1173–1187. doi:10.1093/plankt/12.6.1173.

Hansen, M.O., Nielsen, T.G., Stedmon, C.A. & Munk, P. (2012) Oceanographic regime shift during 1997 in Disko Bay, Western Greenland. *Limnology and Oceanography*. 57 (2), 634–644. doi:10.4319/lo.2012.57.2.0634.

Hansen, P.J. (1995) Growth and grazing response of a ciliate feeding on the red tide dinoflagellate Gyrodinium aureolum in monoculture and in mixture with a non-toxic alga. *Marine Ecology Progress Series*. 121 (1–3), 65–72. doi:10.3354/meps121065.

Harðardóttir, S., Pančić, M., Tammilehto, A., Krock, B., Møller, E.F., Nielsen, T.G. & Lundholm, N.

(2015) Dangerous Relations in the Arctic Marine Food Web: Interactions between Toxin Producing Pseudo-nitzschia Diatoms and Calanus Copepodites. *Marine Drugs*. 13 (6), 3809–3835. doi:10.3390/md13063809.

Harris, R.P., Irigoien, X., Head, R.N., Rey, C., Hygum, B.H., Hansen, B.W., Niehoff, B., Meyer-Harms, B. & Carlotti, F. (2000a) Feeding, growth, and reproduction in the genus Calanus. In: *ICES Journal of Marine Science*. 2000 p. doi:10.1006/jmsc.2000.0959.

Harris, R.P., Wiebe, P.H., Lenz, J., Skoldal, H.R. & Huntley, M. (2000b) *ICES Zooplankton Methodology Manual*. doi:10.1016/B978-012327645-2/50007-4.

Hartmann, D.L., Tank, a. M.G.K. & Rusticucci, M. (2013) IPCC Fifth Assessment Report, Climate Change 2013: The Physical Science Basis. *IPCC*. AR5 (January 2014), 31–39. doi:10.1017/CBO9781107415324.

Hassol, S.J. (2004) *Impacts of a Warming Arctic*. Cambridge, Cambridge University Press.

Hatlebakk, M., Niehoff, B., Choquet, M., Hop, H., Wold, A., Hoarau, G. & Søreide, J.E. (2022) Seasonal Enzyme Activities of Sympatric Calanus glacialis and C . finmarchicus in the High-Arctic. *Frontiers in Marine Science*. 9, 877904. doi:10.3389/fmars.2022.877904.

Head, M. (1996) Modern dinoflagellate cysts and their biological affnities. In: J. Jansonius & D.C. McGregor (eds.). *Palynology: Principles and Applications*. American Association of Stratigraphic Palynologists Foundation. pp. 1197–1248.

Head, M.J., Harland, R. & Matthiessen, J. (2001) Cold marine indicators of the late Quaternary: The new dinoflagellate cyst genus Islandinium and related morphotypes. *Journal of Quaternary Science*. 16 (7), 621–636. doi:10.1002/jqs.657.

Hegseth, E.N. & Sundfjord, A. (2008) Intrusion and blooming of Atlantic phytoplankton species in the high Arctic. *Journal of Marine Systems*. 74 (1–2), 108–119. doi:10.1016/j.jmarsys.2007.11.011.

Henley, S.F., Porter, M., Hobbs, L., Braun, J., Guillaume-Castel, R., Venables, E.J., Dumont, E. & Cottier, F.R. (2020) Nitrate supply and uptake in the Atlantic Arctic sea ice zone: Seasonal cycle, mechanisms and drivers: Arctic shelf seasonal nitrate dynamics. *Philosophical Transactions of the Royal Society A: Mathematical, Physical and Engineering Sciences*. doi:10.1098/rsta.2019.0361.

Henry, L. & Wickham, H. (2020) *purrr: Functional Programming Tools*. https://cran.r-project.org/package=purrr.

Henson, S.A., Cael, B.B., Allen, S.R. & Dutkiewicz, S. (2021) Future phytoplankton diversity in a

changing climate. *Nature Communications*. 12 (1), 1–8. doi:10.1038/s41467-021-25699-w.

Henson, S.A., Laufkötter, C., Leung, S., Giering, S.L.C., Palevsky, H.I. & Cavan, E.L. (2022) Uncertain response of ocean biological carbon export in a changing world. *Nature Geoscience*. 15 (4), 248–254. doi:10.1038/s41561-022-00927-0.

Hessen, D.O. (1992) Nutrient Element Limitation of Zooplankton Production. *The American Naturalist*. 140 (5), 799–814.

Hillebrand, H., Dürselen, C.D., Kirschtel, D., Pollingher, U. & Zohary, T. (1999) Biovolume calculation for pelagic and benthic microalgae. *Journal of Phycology*. 35 (2), 403–424. doi:10.1046/j.1529-8817.1999.3520403.x.

Hirche, H.J. (1996a) Diapause in the marine copepod, Calanus finmarchicus - A review. *Ophelia*. 44 (October), 129–143. doi:10.1080/00785326.1995.10429843.

Hirche, H.J. (1990) Egg production of Calanus finmarchicus at low temperature. *Marine Biology*. 106 (1), 53–58. doi:10.1007/BF02114674.

Hirche, H.J. (1996b) The reproductive biology of the marine copepod, Calanus finmarchicus — A review. *Ophelia*. 44 (1–3), 111–128. doi:10.1080/00785326.1995.10429842.

Hirche, H.J., Baumann, M.E.M., Kattner, G. & Gradinger, R.R. (1991) Plankton distribution and the impact of copepod grazing on primary production in Fram Strait, Greenland Sea. *Journal of Marine Systems*. 2 (3–4), 477–494. doi:10.1016/0924-7963(91)90048-Y.

Hirche, H.J. & Bohrer, R.N. (1987) Reproduction of the Arctic copepod Calanus glacialis in Fram Strait. *Marine Biology*. 94 (1), 11–17. doi:10.1007/BF00392894.

Hirche, H.J. & Kosobokova, K. (2007) Distribution of Calanus finmarchicus in the northern North Atlantic and Arctic Ocean-Expatriation and potential colonization. *Deep-Sea Research Part II: Topical Studies in Oceanography*. 54 (23–26), 2729–2747. doi:10.1016/j.dsr2.2007.08.006.

Hirche, H.J., Meyer, U. & Niehoff, B. (1997) Egg production of Calanus finmarchicus: Effect of temperature, food and season. *Marine Biology*. 127 (4), 609–620. doi:10.1007/s002270050051.

Hirst, A.G. & Bunker, A.J. (2003) Growth of marine planktonic copepods: Global rates and patterns in relation to chlorophyll a, temperature, and body weight. *Limnology and Oceanography*. 48 (5), 1988–2010. doi:10.4238/2015.June.12.13.

Hirst, A.G. & McKinnon, A.D. (2001) Does egg production represent adult female copepod growth? A call to account for body weight changes. *Marine Ecology Progress Series*. 223, 179–199.

doi:10.3354/meps223179.

Ho, T.Y., Quigg, A., Finkel, Z. V., Milligan, A.J., Wyman, K., Falkowski, P.G. & Morel, F.M.M. (2003) The elemental composition of some marine phytoplankton. *Journal of Phycology*. 39 (6), 1145–1159. doi:10.1111/j.0022-3646.2003.03-090.x.

Hobbs, L., Banas, N.S., Cottier, F.R., Berge, J. & Daase, M. (2020) Eat or sleep: availability of winter prey explains mid-winter and early-spring activity in an Arctic Calanus population. *Frontiers in Marine Science*. 7 (September), 1–14. doi:10.3389/fmars.2020.541564.

Hofsteenge, M.G., Graversen, R.G., Rydsaa, J.H. & Rey, Z. (2022) The impact of atmospheric Rossby waves and cyclones on the Arctic sea ice variability. *Climate Dynamics*. 59 (1–2), 579–594. doi:10.1007/s00382-022-06145-z.

Holland, M.M. & Bitz, C.M. (2003) Polar amplification of climate change in coupled models. *Climate Dynamics*. 21 (3–4), 221–232. doi:10.1007/s00382-003-0332-6.

Hop, H., Assmy, P., Wold, A., Sundfjord, A., Daase, M., Tatarek, A., Wiktor Jr., J., Kristiansen, S., Fransson, A., Chierici, M. & Vihtakari, M. (2019) Pelagic Ecosystem Characteristics Across the Atlantic Water Boundary Current From Rijpfjorden, Svalbard, to the Arctic Ocean During Summer (2010-2014). *Frontiers in Marine Science*. 6 (181), 1–21. doi:10.3389/fmars.2019.00181.

Hop, H., Falk-Petersen, S., Svendsen, H., Kwasniewski, S., Pavlov, V., Pavlova, O. & Søreide, J.E. (2006) Physical and biological characteristics of the pelagic system across Fram Strait to Kongsfjorden. *Progress in Oceanography*. 71 (2–4), 182–231. doi:10.1016/j.pocean.2006.09.007.

Hubbard, K.A., Villac, M.C., Chadwick, C., DeSmidt, A.A., Flewelling, L., Granholm, A., Joseph, M., Wood, T., Fachon, E., Brosnahan, M.L., Richlen, M., Pathare, M., Stockwell, D., Lin, P., Bouchard, J.N., Pickart, R. & Anderson, D.M. (2023) *Spatiotemporal transitions in Pseudo-nitzschia species assemblages and domoic acid along the Alaska coast*. doi:10.1371/journal.pone.0282794.

Hunter-Cevera, K.R., Neubert, M., Olson, R., Solow, A., Shalapyonok, A. & Sosik, H. (2016) Physiological and ecological drivers of early spring blooms of a coastal phytoplankter. *Science*. 354 (6310), 326–329.

Huntley, M. & Boyd, C. (1984) Food-Limited Growth of Marine Zooplankton. *The American Society of Naturalists*. 124 (4), 455–478.

Hygum, B.H., Rey, C., Hansen, B.W. & Tande, K.S. (2000) Importance of food quantity to structural growth rate and neutral lipid reserves accumulated in Calanus finmarchicus. *Marine Biology*. 136 (6), 1057–1073. doi:10.1007/s002270000292.

Ikeda, T., Kanno, Y., Ozaki, K. & Shinada, A. (2001) Metabolic rates of epipelagic marine copepods as a function of body mass and temperature. *Marine Biology*. 139 (3), 587–596. doi:10.1007/s002270100608.

Ikeda, T., Torres, J.J., Hernández-León, S. & Geiger, S.P. (2000) *ICES Zooplankton Methodology Manual*. Elsevier. doi:10.1016/B978-0-12-327645-2.X5000-2.

Irigoien, X. (2004) Some ideas about the role of lipids in the life cycle of Calanus finmarchicus. *Journal of Plankton Research*. 26 (3), 259–263. doi:10.1093/plankt/fbh030.

Irigoien, X., Harris, R.P., Verheye, H.M., Joly, P., Runge, J.A., Starr, M., Pond, D.W., Campbell, R., Shreeve, R., Ward, P., Smith, A.N., Dam, H.G., Peterson, W., Tirelli, V., Koski, M., Smith, T., Harbour, D. & Davidson, R. (2002) Copepod hatching success in marine ecosystems with high diatom concentrations. *Nature*. 419, 387–389. doi:10.1038/nature01072.1.

Irigoien, X., Head, R., Klenke, U., Meyer-Harms, B., Harbour, D., Niehoff, B., Hirche, H.J. & Harris, R.P. (1998) A high frequency time series at weathership M, Norwegian sea, during the 1997 spring bloom: Feeding of adult female Calanus finmarchicus. *Marine Ecology Progress Series*. doi:10.3354/meps172127.

Jager, T., Salaberria, I., Altin, D., Nordtug, T. & Hansen, B.H. (2017) Modelling the dynamics of growth, development and lipid storage in the marine copepod Calanus finmarchicus. *Marine Biology*. 164 (1), 1–15. doi:10.1007/s00227-016-3030-8.

Jenkins, H.E., Atherden, F., Cook, K.B., Anderson, T.R., Thornton, B., Mitchell, E., Jacob, E. & Mayor, D.J. (2022) Grazing, egg production and carbon budgets for Calanus finmarchicus across the Fram Strait. *Frontiers in Marine Science*. 9, 1–17. doi:10.3389/fmars.2022.981461.

Jenkins, H.E., Cook, K.B., Anderson, T.R., Atherden, F., Jacob, E. & Mayor, D.J. (n.d.) *Post-bloom grazing, egg production and carbon budgets for Calanus finmarchicus in the Fram Strait*.

Ji, R., Ashjian, C.J., Campbell, R.G., Chen, C., Gao, G., Davis, C.S., Cowles, G.W. & Beardsley, R.C. (2012) Life history and biogeography of Calanus copepods in the Arctic Ocean: An individual-based modeling study. *Progress in Oceanography*. 96 (1), 40–56. doi:10.1016/j.pocean.2011.10.001.

Jónasdóttir, S.H. (1994) Effects of food quality on the reproductive success of Acartia tonsa and Acartia hudsonica: laboratory observations. *Marine Biology*. 121 (1), 67–81. doi:10.1007/BF00349475.

Jónasdóttir, S.H. (2019) Fatty acid profiles and production in marine phytoplankton. *Marine Drugs*.

17 (3). doi:10.3390/md17030151.

Jónasdóttir, S.H. (1999) Lipid content of Calanus finmarchicus during overwintering in the Faroe-Shetland Channel. *Fisheries Oceanography*. 8 (SUPPL. 1), 61–72. doi:10.1046/j.1365-2419.1999.00003.x.

Jónasdóttir, S.H., Fields, D. & Pantoja, S. (1995) Copepod egg production in Long Island Sound, USA, as a function of the chemical composition of seston. *Marine Ecology Progress Series*. 119 (1–3), 87–98. doi:10.3354/meps119087.

Jónasdóttir, S.H., Gudfinnsson, H.G., Gislason, A. & Astthorsson, O.S. (2002) Diet composition and quality for Calanus finmarchicus egg production and hatching success off south-west Iceland. *Marine Biology*. 140 (6), 1195–1206. doi:10.1007/s00227-002-0782-0.

Jónasdóttir, S.H. & Kiørboe, T. (1996) Copepod recruitment and food composition: Do diatoms affect hatching success? *Marine Biology*. 125 (4), 743–750. doi:10.1007/bf00349257.

Jónasdóttir, S.H., Naustvoll, L., Teglhus, F.W., Agersted, M.D., Grenwald, J.C., Melle, W. & Nielsen, T.G. (2022) Calanus finmarchicus basin scale life history traits and role in community carbon turnover during spring. *ICES Journal of Marine Science*. (February), 785–802. doi:10.1093/icesjms/fsac013.

Jónasdóttir, S.H., Nguyen, H.T., Hansen, F. & Gärtner, S. (2005) Egg production and hatching success in the calanoid copepods Calanus helgolandicus and Calanus finmarchicus in the North Sea from March to September 2001. *Journal of Plankton Research*. 27 (12), 1239–1259. doi:10.1093/plankt/fbi091.

Jónasdóttir, S.H., Richardson, K., Heath, M.R., Ingvarsdóttir, A. & Christoffersen, A. (2008) Spring production of Calanus finmarchicus at the Iceland-Scotland Ridge. *Deep-Sea Research Part I: Oceanographic Research Papers*. 55 (4), 471–489. doi:10.1016/j.dsr.2007.12.009.

Jónasdóttir, S.H., Visser, A.W. & Jespersen, C. (2009) Assessing the role of food quality in the production and hatching of Temora longicornis eggs. *Marine Ecology Progress Series*. 382, 139–150. doi:10.3354/meps07985.

Jónasdóttir, S.H., Visser, A.W., Richardson, K. & Heath, M.R. (2015) Seasonal copepod lipid pump promotes carbon sequestration in the deep North Atlantic. *Proceedings of the National Academy of Sciences*. 112 (39), 12122–12126. doi:10.1073/pnas.1512110112.

Jones, E.P. (2001) Circulation in the Arctic Ocean. *Polar Research*. 20 (2), 139–146.

Kahru, M., Brotas, V., Manzano-Sarabia, M. & Mitchell, B.G. (2011) Are phytoplankton blooms occurring earlier in the Arctic? *Global Change Biology*. 17 (4), 1733–1739. doi:10.1111/j.1365-2486.2010.02312.x.

Kaiser, P., Hagen, W., Bode-Dalby, M. & Auel, H. (2022) Tolerant but facing increased competition: Arctic zooplankton versus Atlantic invaders in a warming ocean. *Frontiers in Marine Science*. 9 (August). doi:10.3389/fmars.2022.908638.

Karpouzoglou, T., Steur, L., Smedsrud, L.H. & Sumata, H. (2022) Observed Changes in the Arctic Freshwater Outflow in Fram Strait. *Journal of Geophysical Research: Oceans*. 127 (3), 1–17. doi:10.1029/2021jc018122.

Kassambara, A. (2020) *'ggplot2' Based Publication Ready Plots [R package ggpubr version 0.4.0]*. https://cran.r-project.org/package=ggpubr.

Kattner, G. & Krause, M. (1987) Changes in lipids during the development of Calanus finmarchicus s.l. from Copepodid I to adult. *Marine Biology*. 96 (4), 511–518. doi:10.1007/BF00397968.

Kattner, G. & Krause, M. (1989) Seasonal variations of lipids (wax esters, fatty acids and alcohols) in calanoid copepods from the North Sea. *Marine Chemistry*. 26 (3), 261–275.

Kharouba, H.M. & Wolkovich, E.M. (2020) Disconnects between ecological theory and data in phenological mismatch research. *Nature Climate Change*. 10 (5), 406–415. doi:10.1038/s41558-020-0752-x.

Kilias, E.S., Nöthig, E.M., Wolf, C. & Metfies, K. (2014) Picoeukaryote plankton composition off West Spitsbergen at the entrance to the Arctic Ocean. *The Journal of eukaryotic microbiology*. 61 (6), 569–579. doi:10.1111/jeu.12134.

Kiørboe, T. (2011) How zooplankton feed: Mechanisms, traits and trade-offs. *Biological Reviews*. 86 (2), 311–339. doi:10.1111/j.1469-185X.2010.00148.x.

Kiørboe, T., Møhlenberg, F. & Riisgård, H.U. (1985) In situ feeding rates of plantonic copepods: A comparison of four methods. *Journal of Experimental Marine Biology and Ecology*. 88 (1), 67–81. doi:10.1016/0022-0981(85)90202-3.

Kiørboe, T., Saiz, E. & Viitasalo, M. (1996) Prey switching behaviour in the planktonic copepod Acartia tonsa. *Marine Ecology Progress Series*. 143, 65–75.

Kleppel, G.S. (1993) On the diets of calanoid copepods. *Marine Ecology Progress Series*. doi:10.3354/meps099183.

Kleppel, G.S. & Hazzard, S.E. (2000) Diet and egg production of the copepod Acartia tonsa in Florida Bay. II. Role of the nutritional environment. *Marine Biology*. 137, 111–121. papers2://publication/uuid/EA3A189C-78F3-493B-B78D-A0BD19AD7765.

Kohlbach, D., Schmidt, K., Hop, H., Wold, A., Al-Habahbeh, A.K., Belt, S.T., Woll, M., Graeve, M., Smik, L., Atkinson, A. & Assmy, P. (2021) Winter Carnivory and Diapause Counteract the Reliance on Ice Algae by Barents Sea Zooplankton. *Frontiers in Marine Science*. 8 (March), 1–17. doi:10.3389/fmars.2021.640050.

Koski, M. & Wexels Riser, C. (2006) Post-bloom feeding of Calanus finmarchicus copepodites: Selection for autotrophic versus heterotrophic prey. *Marine Biology Research*. 2 (2), 109–119. doi:10.1080/17451000600684367.

Krause, J.W., Schulz, I.K., Rowe, K.A., Dobbins, W., Winding, M.H.S., Sejr, M.K., Duarte, C.M. & Agustí, S. (2019) Silicic acid limitation drives bloom termination and potential carbon sequestration in an Arctic bloom. *Scientific Reports*. 9 (1), 1–11. doi:10.1038/s41598-019-44587-4.

Krisch, S., Browning, T.J., Graeve, M., Ludwichowski, K.U., Lodeiro, P., Hopwood, M.J., Roig, S., Yong, J.C., Kanzow, T. & Achterberg, E.P. (2020) The influence of Arctic Fe and Atlantic fixed N on summertime primary production in Fram Strait, North Greenland Sea. *Scientific Reports*. 10 (1), 1–13. doi:10.1038/s41598-020-72100-9.

Kwiatkowski, L., Torres, O., Bopp, L., Aumont, O., Chamberlain, M., et al. (2020) Twenty-first century ocean warming, acidification, deoxygenation, and upper-ocean nutrient and primary production decline from CMIP6 model projections. *Biogeosciences*. 17 (13), 3439–3470. doi:10.5194/bg-17-3439-2020.

Labe, Z., Magnusdottir, G. & Stern, H. (2018) Variability of Arctic sea ice thickness using PIOMAS and the CESM large ensemble. *Journal of Climate*. 31 (8), 3233–3247. doi:10.1175/JCLI-D-17-0436.1.

Lampe, V., Nöthig, E.-M. & Schartau, M. (2021) Spatio-Temporal Variations in Community Size Structure of Arctic Protist Plankton in the Fram Strait. *Frontiers in Marine Science*. 7 (January), 1–18. doi:10.3389/fmars.2020.579880.

Lampert, W. (1989) The Adaptive Significance of Diel Vertical Migration of Zooplankton. *Functional Ecology*. doi:10.2307/2389671.

Lannuzel, D., Tedesco, L., van Leeuwe, M., Campbell, K., Flores, H., et al. (2020) The future of Arctic sea-ice biogeochemistry and ice-associated ecosystems. *Nature Climate Change*. 10 (11),

983–992. doi:10.1038/s41558-020-00940-4.

Lee, R.F., Hagen, W. & Kattner, G. (2006a) Lipid storage in marine zooplankton. *Marine Ecology Progress Series*. 307 (1863), 273–306. doi:Doi 10.3354/Meps307273.

Lee, R.F., Hagen, W. & Kattner, G. (2006b) Lipid storage in marine zooplankton. *Marine Ecology Progress Series*. 307 (1863), 273–306. doi:Doi 10.3354/Meps307273.

Leiknes, Ø., Striberny, A., Tokle, N.E., Olsen, Y., Vadstein, O., Sommer, U., Egil, N., Olsen, Y., Vadstein, O. & Sommer, U. (2014) Feeding selectivity of Calanus finmarchicus in the Trondheimsfjord. *Journal of Sea Research*. 85, 292–299. doi:10.1016/j.seares.2013.05.012.

Leu, E., Søreide, J.E., Hessen, D.O., Falk-Petersen, S. & Berge, J. (2011) Consequences of changing sea-ice cover for primary and secondary producers in the European Arctic shelf seas: Timing, quantity, and quality. *Progress in Oceanography*. 90 (1–4), 18–32. doi:10.1016/j.pocean.2011.02.004.

Levinsen, H., Turner, J.T., Nielsen, T.G. & Hansen, B.W. (2000) On the trophic coupling between protists and copepods in Arctic marine ecosystems. *Marine Ecology Progress Series*. 204, 65–77. doi:10.3354/meps204065.

Lewis, K.M., Van Dijken, G.L. & Arrigo, K.R. (2020) Changes in phytoplankton concentration now drive increased Arctic Ocean primary production. *Science*. 369 (6500), 198–202. doi:10.1126/science.aay8380.

Li, W.K.W., McLaughlin, F.A., Lovejoy, C. & Carmack, E.C. (2009) Smallest Algae Thrive As the Arctic Ocean Freshens. *Science*. 326 (5952), 539–539. doi:10.1126/science.1179798.

Li, X., Yan, T., Yu, R. & Zhou, M. (2019) A review of karenia mikimotoi: Bloom events, physiology, toxicity and toxic mechanism. *Harmful Algae*. 90 (June), 101702. doi:10.1016/j.hal.2019.101702.

Lindeque, P.K., Hann, I., Parry, H.E., Cook, K.B., Lindley, A. & Mayor, D.J. (2022) Red Pigmentation Can Be Used to Reliably Distinguish Between Live Calanus finmarchicus and Calanus glacialis Females in the Fram Strait. *Frontiers in Marine Science*. 9 (June), 1–8. doi:10.3389/fmars.2022.906465.

Liu, C., Zhang, X. & Wang, X. (2022) DNA metabarcoding data reveals harmful algal-bloom species undescribed previously at the northern Antarctic Peninsula region. *Polar Biology*. 45 (9), 1495–1512. doi:10.1007/s00300-022-03084-7.

Lombard, F., Boss, E., Waite, A.M., Vogt, M., Uitz, J., et al. (2019) Globally Consistent Quantitative

Observations of Planktonic Ecosystems. *Frontiers in Marine Science*. 6 (April). doi:10.3389/fmars.2019.00196.

Lovejoy, C., Legendre, L., Martineau, M.J., Bâcle, J. & von Quillfeldt, C.H. (2002) Distribution of phytoplankton and other protists in the North Water. *Deep-Sea Research Part II: Topical Studies in Oceanography*. 49 (22–23), 5027–5047. doi:10.1016/S0967-0645(02)00176-5.

Lund, J.W.G., Kipling, C. & Le Cren, E.D. (1958) The inverted microscope method of estimating algal numbers and the statistical basis of estimations by counting. *Hydrobiologia*. 11 (2), 143–170. doi:10.1007/BF00007865.

MacNeil, L., Missan, S., Luo, J., Trappenberg, T. & LaRoche, J. (2021) Plankton classification with high - throughput submersible holographic microscopy and transfer learning. *BMC Ecology and Evolution*. 21 (123), 1–11. doi:10.1186/s12862-021-01839-0.

Madin, L.P. & Purcell, J.E. (1992) Feeding, metabolism and growth of Cyclosalpa bakeri in the subarctic Pacific. *Limnology and Oceanography*. 37 (6), 1236–1251. doi:10.4319/lo.1992.37.6.1236.

Madsen, S.J., Nielsen, T.G., Tervo, O.M. & Söderkvist, J. (2008) Importance of feeding for egg production in Calanus finmarchicus and C. glacialis during the Arctic spring. *Marine Ecology Progress Series*. 353, 177–190. doi:10.3354/meps07129.

Malzahn, A.M. & Boersma, M. (2012) Effects of poor food quality on copepod growth are dose dependent and non-reversible. *Oikos*. 121 (9), 1408–1416. doi:10.1111/j.1600-0706.2011.20186.x.

Maps, F., Record, N.R. & Pershing, A.J. (2014) A metabolic approach to dormancy in pelagic copepods helps explaining inter- and intra-specific variability in life-history strategies. *Journal of Plankton Research*. 36 (1), 18–30. doi:10.1093/plankt/fbt100.

Marret, F., Bradley, L., de Vernal, A., Hardy, W., Kim, S.Y., Mudie, P., Penaud, A., Pospelova, V., Price, A.M., Radi, T. & Rochon, A. (2020) From bi-polar to regional distribution of modern dinoflagellate cysts, an overview of their biogeography. *Marine Micropaleontology*. 159 (July 2019), 101753. doi:10.1016/j.marmicro.2019.101753.

Marshall, S.M. & Orr, A.P. (1958) On the biology of Calanus finmarchicus. X. Seasonal changes in oxygen consumption. *Journal of the Marine Biological Association of the United Kingdom*. 37 (2), 459–472. doi:10.1017/S002531540002381X.

Marshall, S.M. & Orr, A.P. (1955) *The Biology of a Marine Copepod Calanus finmarchicus*

(Gunnerus). London, Oliver & Boyd.

Martin, J.L., Santi, I., Pitta, P., John, U. & Gypens, N. (2022) Towards quantitative metabarcoding of eukaryotic plankton: an approach to improve 18S rRNA gene copy number bias. *Metabarcoding and Metagenomics*. 6 (August), 245–259. doi:10.3897/mbmg.6.85794.

Maslowski, W., Marble, D., Walczowski, W., Schauer, U., Clement, J.L. & Semtner, A.J. (2004) On climatological mass, heat, and salt transports through the Barents Sea and Fram Strait from a pan-Arctic coupled ice-ocean model simulation. *Journal of Geophysical Research: Oceans*. 109 (C3). doi:10.1029/2001JC001039.

Mauchline, J., Blaxter, J.H.S., Southward, A.J. & Tyler, P.A. (1998) The Biology of Calanoid Copepods. *Advances in Marine Biology*.

Mayor, D.J. (2005) *Nutritional regulation of egg production of Calanus finmarchicus in the North Atlantic*. University of Southampton. http://eprints.soton.ac.uk/17240/.

Mayor, D.J., Anderson, T.R., Irigoien, X. & Harris, R.P. (2006) Feeding and reproduction of Calanus finmarchicus during non-bloom conditions in the Irminger Sea. *Journal of Plankton Research*. 28 (12), 1167–1179. doi:10.1093/plankt/fbl047.

Mayor, D.J., Anderson, T.R., Pond, D.W. & Irigoien, X. (2009a) Egg production and associated losses of carbon, nitrogen, and fatty acids from maternal biomass in Calanus finmarchicus before the spring bloom. *Journal of Marine Systems*. 78 (4), 505–510. doi:10.1016/j.jmarsys.2008.12.019.

Mayor, D.J., Anderson, T.R., Pond, D.W. & Irigoien, X. (2009b) Limitation of egg production in Calanus finmarchicus in the field: A stoichiometric analysis. *Journal of Marine Systems*. 78 (4), 511–517. doi:10.1016/j.jmarsys.2008.12.020.

Mayor, D.J., Cook, K.B., Thornton, B., Atherden, F., Tarling, G.A. & Anderson, T.R. (2022) Biomass Turnover Rates in Metabolically Active and Inactive Marine Calanoid Copepods. *Frontiers in Marine Science*. 9 (June), 1–6. doi:10.3389/fmars.2022.907290.

Mayor, D.J., Cook, K.B., Thornton, B., Walsham, P., Witte, U.F.M., Zuur, A.F. & Anderson, T.R. (2011) Absorption efficiencies and basal turnover of C, N and fatty acids in a marine Calanoid copepod. *Functional Ecology*. 25 (3), 509–518. doi:10.1111/j.1365-2435.2010.01791.x.

Mayor, D.J., Sanders, R., Giering, S.L.C. & Anderson, T.R. (2014) Microbial gardening in the ocean's twilight zone: Detritivorous metazoans benefit from fragmenting, rather than ingesting, sinking detritus. *BioEssays*. 36 (12), 1132–1137. doi:10.1002/bies.201400100.

Mayor, D.J., Sommer, U., Cook, K.B. & Viant, M.R. (2015) The metabolic response of marine copepods to environmental warming and ocean acidification in the absence of food. *Scientific Reports*. 5 (1), 13690. doi:10.1038/srep13690.

Mayot, N., Matrai, P., Ellingsen, I.H., Steele, M., Johnson, K., Riser, S.C. & Swift, D. (2018) Assessing Phytoplankton Activities in the Seasonal Ice Zone of the Greenland Sea Over an Annual Cycle. *Journal of Geophysical Research: Oceans*. 123 (11), 8004–8025. doi:10.1029/2018JC014271.

Mayzaud, P. (1976) Respiration and nitrogen excretion of zooplankton. IV. The influence of starvation on the metabolism and the biochemical composition of some species. *Marine Biology*. 37 (1), 47–58. doi:10.1007/BF00386778.

Melle, W. & Skjoldal, H.R. (1998) Reproduction and development of Calanus finmarchicus, C. glacialis and C. hyperboreus in the Barents Sea. *Marine Ecology Progress Series*. 169, 211–228. doi:10.3354/meps169211.

Menden-Deuer, S. & Lessard, E.J. (2000) Carbon to volume relatinship for dinoflagellates, diatoms and other protist plankton. *Limnology and Oceanography*. 45 (3), 569–579. https://aslopubs.onlinelibrary.wiley.com/doi/pdf/10.4319/lo.2000.45.3.0569.

Menden-Deuer, S., Lessard, E.J. & Satterberg, J. (2001) Effect of preservation on dinoflagellate and diatom cell volume and consequences for carbon biomass predictions. *Marine Ecology Progress Series*. 222, 41–50. doi:10.3354/meps222041.

Mendenhall, E., Hendrix, C., Nyman, E., Roberts, P.M., Hoopes, J.R., Watson, J.R., Lam, V.W.Y. & Sumaila, U.R. (2020) Climate change increases the risk of fisheries conflict. *Marine Policy*. 117, 103954. doi:https://doi.org/10.1016/j.marpol.2020.103954.

Meyer, B., Irigoien, X., Graeve, M., Head, R. & Harris, R.P. (2002) Feeding rates and selectivity among nauplii, copepodites and adult females of Calanus finmarchicus and Calanus helgolandicus. *Helgoland Marine Research*. 56 (3), 169–176. doi:10.1007/s10152-002-0105-3.

Mitra, A., Castellani, C., Gentleman, W.C., Jónasdóttir, S.H., Flynn, K.J., Bode, A., Halsband, C., Kuhn, P., Licandro, P., Agersted, M.D., Calbet, A., Lindeque, P.K., Koppelmann, R., Møller, E.F., Gislason, A., Nielsen, T.G. & St. John, M. (2014) Bridging the gap between marine biogeochemical and fisheries sciences; configuring the zooplankton link. *Progress in Oceanography*. 129 (PB), 176–199. doi:10.1016/j.pocean.2014.04.025.

Mitra, A. & Flynn, K.J. (2005) Predator-prey interactions: Is 'ecological stoichiometry' sufficient when good food goes bad? *Journal of Plankton Research*. 27 (5), 393–399.

doi:10.1093/plankt/fbi022.

Møller, E.F., Bohr, M., Kjellerup, S., Maar, M., Møhl, M., Swalethorp, R. & Nielsen, T.G. (2016) Calanus finmarchicus egg production at its northern border. *Journal of Plankton Research*. 38 (5), 1206–1214. doi:10.1093/plankt/fbw048.

Møller, E.F., Maar, M., Jónasdóttir, S.H., Nielsen, T.G. & Tönnesson, K. (2012) The effect of changes in temperature and food on the development of calanus finmarchicus and Calanus helgolandicus populations. *Limnology and Oceanography*. 57 (1), 211–220. doi:10.4319/lo.2012.57.1.0211.

Møller, E.F. & Nielsen, T.G. (2020) Borealization of Arctic zooplankton—smaller and less fat zooplankton species in Disko Bay, Western Greenland. *Limnology and Oceanography*. 65 (6), 1175–1188. doi:10.1002/lno.11380.

Møller, E.F., Thor, P. & Nielsen, T.G. (2003) Production of DOC by Calanus finmarchicus, C. glacialis and C. hyperboreus through sloppy feeding and leakage from fecal pellets. *Marine Ecology Progress Series*. 262, 185–191. doi:10.3354/meps262185.

Moore, C.M., Mills, M.M., Arrigo, K.R., Berman-Frank, I., Bopp, L., et al. (2013) Processes and patterns of oceanic nutrient limitation. *Nature Geoscience*. 6 (9), 701–710. doi:10.1038/ngeo1765.

Moore, S.E., Haug, T., Víkingsson, G.A. & Stenson, G.B. (2019) Baleen whale ecology in arctic and subarctic seas in an era of rapid habitat alteration. *Progress in Oceanography*. 176 (May), 102118. doi:10.1016/j.pocean.2019.05.010.

Morán, X.A.G., López-Urrutia, Á., Calvo-Díaz, A. & Li, W.K.W. (2010) Increasing importance of small phytoplankton in a warmer ocean. *Global Change Biology*. 16 (3), 1137–1144. doi:10.1111/j.1365-2486.2009.01960.x.

Morata, N. & Søreide, J.E. (2015) Effect of light and food on the metabolism of the Arctic copepod Calanus glacialis. *Polar Biology*. 38 (1), 67–73. doi:10.1007/s00300-013-1417-2.

Mousing, E.A., Richardson, K. & Ellegaard, M. (2018) Global patterns in phytoplankton biomass and community size structure in relation to macronutrients in the open ocean. *Limnology and Oceanography*. 63 (3), 1298–1312. doi:10.1002/lno.10772.

Natsuike, M., Nagai, S., Matsuno, K., Saito, R., Tsukazaki, C., Yamaguchi, A. & Imai, I. (2013) Abundance and distribution of toxic Alexandrium tamarense resting cysts in the sediments of the Chukchi Sea and the eastern Bering Sea. *Harmful Algae*. 27, 52–59. doi:https://doi.org/10.1016/j.hal.2013.04.006.

Nejstgaard, J.C., Frischer, M.E., Simonelli, P., Troedsson, C., Brakel, M., Adiyaman, F., Sazhin, A.F. & Artigas, L.F. (2008) Quantitative PCR to estimate copepod feeding. *Marine Biology*. 153 (4), 565–577. doi:10.1007/s00227-007-0830-x.

Nejstgaard, J.C., Gismervik, I. & Solberg, P.T. (1997) Feeding and reproduction by Calanus finmarchicus , and microzooplankton grazing during mesocosm blooms of diatoms and the coccolithophore Emiliania huxleyi. *Marine Ecology Progress Series*. 147, 197–217.

Nejstgaard, J.C., Naustvoll, L.J. & Sazhin, A. (2001) Correcting for underestimation of microzooplankton grazing in bottle incubation experiments with mesozooplankton. *Marine Ecology Progress Series*. 221, 59–75. doi:10.3354/meps221059.

Neukermans, G., Oziel, L. & Babin, M. (2018) Increased intrusion of warming Atlantic water leads to rapid expansion of temperate phytoplankton in the Arctic. *Global Change Biology*. 24 (6), 2545–2553. doi:10.1111/gcb.14075.

Niehoff, B. (2007) Life history strategies in zooplankton communities: The significance of female gonad morphology and maturation types for the reproductive biology of marine calanoid copepods. *Progress in Oceanography*. 74 (1), 1–47. doi:10.1016/j.pocean.2006.05.005.

Niehoff, B. (2004) The effect of food limitation on gonad development and egg production of the planktonic copepod Calanus finmarchicus. *Journal of Experimental Marine Biology and Ecology*. 307 (2), 237–259. doi:10.1016/j.jembe.2004.02.006.

Niehoff, B., Klenke, U., Hirche, H.J., Irigoien, X., Head, R. & Harris, R.P. (1999) A high frequency time series at Weathership M, Norwegian Sea, during the 1997 spring bloom: The reproductive biology of Calanus finmarchicus. *Marine Ecology Progress Series*. 176 (1953), 81–92. doi:10.3354/meps176081.

Niehoff, B., Madsen, S.D., Hansen, B.W. & Nielsen, T.G. (2002) Reproductive cycles of three dominant Calanus species in Disko Bay, West Greenland. *Marine Biology*. 140 (3), 567–576. doi:10.1007/s00227-001-0731-3.

Niehoff, B. & Runge, J.A. (2003) A revised methodology for prediction of egg production Calanus finmarchicus from preserved samples. *Journal of Plankton Research*. 25 (12), 1581–1587. doi:10.1093/plankt/fbg104.

Nőges, T. (1992) Comparison of Two Methods of Zooplankton Grazing Measurements. *Internationale Revue der gesamten Hydrobiologie und Hydrographie*. 77 (4), 665–672. doi:10.1002/iroh.19920770410.

Noh, K.M., Lim, H.G., Yang, E.J. & Kug, J.S. (2023) Emergent Constraint for Future Decline in Arctic Phytoplankton Concentration. *Earth's Future*. 11 (4). doi:10.1029/2022EF003427.

Nöthig, E.-M., Bracher, A., Engel, A., Metfies, K., Niehoff, B., Peeken, I., Bauerfeind, E., Cherkasheva, A., Gäbler-Schwarz, S., Hardge, K., Kilias, E., Kraft, A., Kidane, Y.M., Lalande, C., Piontek, J., Thomisch, K. & Wurst, M. (2015) Summertime plankton ecology in Fram Strait—a compilation of long- and short-term observations. *Polar Research*. 34 (1), 23349. doi:10.3402/polar.v34.23349.

Nöthig, E.M., Ramondenc, S., Haas, A., Hehemann, L., Walter, A., Bracher, A., Lalande, C., Metfies, K., Peeken, I., Bauerfeind, E. & Boetius, A. (2020) Summertime Chlorophyll a and Particulate Organic Carbon Standing Stocks in Surface Waters of the Fram Strait and the Arctic Ocean (1991–2015). *Frontiers in Marine Science*. 7 (June), 1–15. doi:10.3389/fmars.2020.00350.

Notz, D. & Stroeve, J. (2018) The Trajectory Towards a Seasonally Ice-Free Arctic Ocean. *Current Climate Change Reports*. 4 (4), 407–416. doi:10.1007/s40641-018-0113-2.

Noyon, M. & William Froneman, P. (2013) Variability in the egg production rates of the calanoid copepod, Pseudodiaptomus hessei in a South African estuary in relation to environmental factors. *Estuarine, Coastal and Shelf Science*. 135, 306–316. doi:10.1016/j.ecss.2013.10.024.

Ohman, M.D. & Runge, J.A. (1994) Sustained fecundity when phytoplankton resources are in short supply: Omnivory by Calanus finmarchicus in the Gulf of St. Lawrence. *Limnology and Oceanography*. 39 (1), 21–36. doi:10.4319/lo.1994.39.1.0021.

Onda, D.F., Wolf, C., Metfies, K., Salter, I. & Noethig, E. (2020) Changes in exported key phytoplankton taxa related to a warm anomaly in the Fram Strait inferred from three complementary 18S rRNA gene meta-barcoding primer sets. *Authorea Preprints*. 1–21.

Paquette, R.G., Bourke, R.H., Newton, J.F. & Perdue, W.F. (1985) The East Greenland Polar Front in autumn. *Journal of Geophysical Research*. 90 (C3), 4866. doi:10.1029/jc090ic03p04866.

Pasternak, A.F., Arashkevich, E., Tande, K.S. & Falkenhaug, T. (2001) Seasonal changes in feeding, gonad development and lipid stores in Calanus finmarchicus and C. hyperboreus from Malangen, northern Norway. *Marine Biology*. 138, 1141–1152. doi:10.1007/s002270100553.

Pasternak, A.F., Arashkevich, E.G., Grothe, U., Nikishina, A.B. & Solovyev, K.A. (2013) Different effects of increased water temperature on egg production of Calanus finmarchicus and C. glacialis. *Marine Biology*. 53 (5), 547–553. doi:10.1134/S0001437013040085.

Paulsen, M.L., Doré, H., Garczarek, L., Seuthe, L., Müller, O., Sandaa, R.A., Bratbak, G. & Larsen, A.

(2016) Synechococcus in the Atlantic Gateway to the Arctic Ocean. *Frontiers in Marine Science*. 3 (OCT), 191. doi:10.3389/FMARS.2016.00191/BIBTEX.

Perrette, M., Yool, A., Quartly, G.D. & Popova, E.E. (2011) Near-ubiquity of ice-edge blooms in the Arctic. *Biogeosciences*. 8 (2), 515–524. doi:10.5194/bg-8-515-2011.

Peter, K.H. & Sommer, U. (2012) Phytoplankton Cell Size: Intra- and Interspecific Effects of Warming and Grazing. *PLoS ONE*. 7 (11). doi:10.1371/journal.pone.0049632.

Pierella Karlusich, J.J., Lombard, F., Irisson, J.O., Bowler, C. & Foster, R.A. (2022) Coupling Imaging and Omics in Plankton Surveys: State-of-the-Art, Challenges, and Future Directions. *Frontiers in Marine Science*. 9 (June), 1–13. doi:10.3389/fmars.2022.878803.

Pinti, J., Jónasdóttir, S.H., Record, N.R. & Visser, A.W. (2023) The global contribution of seasonally migrating copepods to the biological carbon pump. *Limnology and Oceanography*. (1992), 1147–1160. doi:10.1002/lno.12335.

Plourde, S. & Runge, J.A. (1993) Reproduction of the planktonic copepod Calanus finmarchicus in the lower St Lawrence Estuary: Relation to the cycle of phytoplankton production and evidence for a Calanus pump. *Marine Ecology Progress Series*. 102 (3), 217–228. doi:10.3354/meps102217.

Poloczanska, E.S., Brown, C.J., Sydeman, W.J., Kiessling, W., Schoeman, D.S., et al. (2013) Global imprint of climate change on marine life. *Nature Climate Change*. 3 (10), 919–925. doi:10.1038/nclimate1958.

Pond, D.W., Harris, R.P., Head, R. & Harbour, D. (1996) Environmental and nutritional factors determining seasonal variability in the fecundity and egg viability of Calanus helgolandicus in coastal waters off Plymouth, UK. *Marine Ecology Progress Series*. 143 (1–3), 45–63. doi:10.3354/meps143045.

Pond, D.W. & Tarling, G.A. (2011) Phase transitions of wax esters adjust buoyancy in diapausing *Calanoides acutus*. *Limnology and Oceanography*. 56 (4), 1310–1318. doi:10.4319/lo.2011.56.4.1310.

Pond, D.W., Tarling, G.A. & Mayor, D.J. (2014) Hydrostatic pressure and temperature effects on the membranes of a seasonally migrating marine copepod. *PLoS ONE*. 9 (10). doi:10.1371/journal.pone.0111043.

Pond, D.W., Tarling, G.A., Ward, P. & Mayor, D.J. (2012) Wax ester composition influences the diapause patterns in the copepod Calanoides acutus. *Deep-Sea Research II*. 59–60, 93–104. doi:10.1016/j.dsr2.2011.05.009.

Post, E., Bhatt, U.S., Bitz, C.M., Brodie, J.F., Fulton, T.L., Hebblewhite, M., Kerby, J., Kutz, S.J., Stirling, I. & Walker, D.A. (2013) Ecological consequences of sea-ice decline. *Science*. 341 (6145), 519–524. doi:10.1126/science.1235225.

Poulet, S.A., Ianora, A., Laabir, M. & Klein Breteler, W.C.M. (1995) Towards the measurement of secondary production and recruitment in copepods. *ICES Journal of Marine Science*. 52 (3–4), 359–368. doi:10.1016/1054-3139(95)80051-4.

Poulin, M., Daugbjerg, N., Gradinger, R., Ilyash, L., Ratkova, T. & von Quillfeldt, C.H. (2011) The pan-Arctic biodiversity of marine pelagic and sea-ice unicellular eukaryotes: A first-attempt assessment. *Marine Biodiversity*. 41 (1), 13–28. doi:10.1007/s12526-010-0058-8.

Poulton, N.J. (2016) FlowCam: Quantification and Classification of Phytoplankton by Imaging Flow Cytometry. In: *Methods in Molecular Biology*. Methods Mol Biol. pp. 237–247. doi:10.1007/978-1-4939-3302-0_17.

Price, E.L., Stern, R.F., Mahaffey, C., Castellani, C. & Jeffreys, R.M. (2021) Sensitivity of plankton assemblages to hydroclimate variability in the Barents Sea. *Biogeosciences (submitted)*. https://doi.org/10.5194/bg-2021-279.

Quadfasel, D., Gascard, J.-C. & Koltermann, K.-P. (1987) Large-scale oceanography in Fram Strait during the 1984 Marginal Ice Zone Experiment. *Journal of Geophysical Research*. 92 (C7), 6719–6728. doi:10.1029/JC092iC07p06719.

Questel, J.M., Hopcroft, R.R., DeHart, H.M., Smoot, C.A., Kosobokova, K.N. & Bucklin, A. (2021) Metabarcoding of zooplankton diversity within the Chukchi Borderland, Arctic Ocean: improved resolution from multi-gene markers and region-specific DNA databases. *Marine Biodiversity*. 51 (1). doi:10.1007/s12526-020-01136-x.

R Core Team (2022) R core team. *R: A language and environment for statistical computing*.

Rajter, Ľ. & Dunthorn, M. (2021) Ciliate SSU-rDNA reference alignments and trees for phylogenetic placements of metabarcoding data. *Metabarcoding and Metagenomics*. 5, 121–132. doi:10.3897/MBMG.5.69602.

Rantanen, M., Karpechko, A.Y., Lipponen, A., Nordling, K., Hyvärinen, O., Ruosteenoja, K., Vihma, T. & Laaksonen, A. (2022) The Arctic has warmed nearly four times faster than the globe since 1979. *Communications Earth and Environment*. 3 (1), 1–10. doi:10.1038/s43247-022-00498-3.

Ray, J.L., Althammer, J., Skaar, K.S., Simonelli, P., Larsen, A., Stoecker, D., Sazhin, A., Ijaz, U.Z., Quince, C., Nejstgaard, J.C., Frischer, M., Pohnert, G. & Troedsson, C. (2016a) Metabarcoding and

metabolome analyses of copepod grazing reveal feeding preference and linkage to metabolite classes in dynamic microbial plankton communities. *Molecular Ecology*. 25 (21), 5585–5602. doi:10.1111/mec.13844.

Ray, J.L., Skaar, K.S., Simonelli, P., Larsen, A., Sazhin, A., Jakobsen, H.H., Nejstgaard, J.C. & Troedsson, C. (2016b) Molecular gut content analysis demonstrates that Calanus grazing on Phaeocystis pouchetii and Skeletonema marinoi is sensitive to bloom phase but not prey density. *Marine Ecology*. 542, 63–77. doi:10.3354/meps11560.

Redfield, A.C. (1934) *On the Proportions of Organic Derivatives in Sea Water and Their Relation to the Composition of Plankton*. Liverpool, University press of Liverpool. doi:citeulike-article-id:11236440.

Redfield, A.C. (1958) The Biological Control of Chemical Factors in the Environment. *American Scientist*. 46 (3), 205–221. doi:10.5194/bg-11-1599-2014.

Regaudie-De-Gioux, A. & Duarte, C.M. (2012) Temperature dependence of planktonic metabolism in the ocean. *Global Biogeochemical Cycles*. 26 (1), 1–10. doi:10.1029/2010GB003907.

Reigstad, M., Wassmann, P., Wexels Riser, C., Øygarden, S. & Rey, F. (2002) Variations in hydrography, nutrients and chlorophyll a in the marginal ice-zone and the central Barents Sea. *Journal of Marine Systems*. doi:10.1016/S0924-7963(02)00167-7.

Renaud, P.E., Daase, M., Banas, N.S., Gabrielsen, T.M., Søreide, J.E., Varpe, Ø., Cottier, F.R., Falk-Petersen, S., Halsband, C., Vogedes, D., Heggland, K. & Berge, J. (2018) Pelagic food-webs in a changing Arctic: a trait-based perspective suggests a mode of resilience. *ICES Journal of Marine Science*. doi:10.1093/icesjms/fsy063.

Rey-Rassat, C., Irigoien, X., Harris, R.P. & Carlotti, F. (2002) Energetic cost of gonad development in Calanus finmarchicus and C. helgolandicus. *Marine Ecology Progress Series*. 238, 301–306. doi:10.3354/meps238301.

Rey, C., Carlotti, F., Tande, K.S. & Hygum, B.H. (1999) Egg and faecal pellet production of Calanus finmarchicus females from controlled mesocosms and in situ populations: influence of age and feeding history. *Marine Ecology Progress Series*. 188, 133–148. doi:10.3354/meps188133.

Richardson, K., Jónasdóttir, S.H., Hay, S.J. & Christoffersen, A. (1999) Calanus finmarchicus egg production and food availability in the Faroe-Shetland Channel and northern North Sea: October-March. *Fisheries Oceanography*. 8 (1), 153–162.

Robinson, C. (2017) Phytoplankton Biogeochemical Cycles. *Marine Plankton: A practical guide to*

ecology, methodology, and taxonomy. 1, 42–51. https://academic.oup.com/book/25513/chapter/192749457.

Roncalli, V., Turner, J.T., Kulis, D., Anderson, D.M. & Lenz, P.H. (2016) The effect of the toxic dinoflagellate Alexandrium fundyense on the fitness of the calanoid copepod Calanus finmarchicus. *Harmful Algae*. 51, 56–66. doi:10.1016/j.pediatrneurol.2015.01.016.Pathophysiology.

Rüger, T. & Sommer, U. (2012) Warming does not always benefit the small - Results from a plankton experiment. *Aquatic Botany*. 97 (1), 64–68. doi:10.1016/j.aquabot.2011.12.001.

Runge, J.A. (1984a) Egg production of the marine, planktonic copepod, Calanus pacificus Brodsky: Laboratory observations. *Journal of Experimental Marine Biology and Ecology*. 74 (1), 53–66. doi:10.1016/0022-0981(84)90037-6.

Runge, J.A. (1984b) Egg production of the marine, planktonic copepod, Calanus pacificus Brodsky: Laboratory Observations. *Journal of Experimental Marine Biology and Ecology*. 74, 53–66.

Runge, J.A. & Plourde, S. (1996) Fecundity characteristics of Calanus finmarchicus in coastal waters of eastern canada. *Ophelia*. 44 (1–3), 171–187. doi:10.1080/00785326.1995.10429846.

Saiz, E. & Calbet, A. (2011) Copepod feeding in the ocean: scaling patterns , composition of their diet and the bias of estimates due to microzooplankton grazing during incubations. *Hydrobiologia*. 666, 181–196. doi:10.1007/s10750-010-0421-6.

Saiz, E., Calbet, A., Isari, S., Antó, M., Velasco, E.M., Almeda, R., Movilla, J. & Alcaraz, M. (2013) Zooplankton distribution and feeding in the Arctic Ocean during a Phaeocystis pouchetii bloom. *Deep-Sea Research Part I: Oceanographic Research Papers*. 72, 17–33. doi:10.1016/j.dsr.2012.10.003.

Sakshaug, E. (2004) Primary and Secondary Production in the Arctic Seas. In: *The Organic Carbon Cycle in the Arctic Ocean*. pp. 57–81. doi:10.1007/978-3-642-18912-8_3.

Sakshaug, E. & Slagstad, D. (1992) Sea ice and wind: Effects on primary productivity in the barents sea. *Atmosphere - Ocean*. doi:10.1080/07055900.1992.9649456.

Sallée, J.B., Pellichero, V., Akhoudas, C., Pauthenet, E., Vignes, L., Schmidtko, S., Garabato, A.N., Sutherland, P. & Kuusela, M. (2021) Summertime increases in upper-ocean stratification and mixed-layer depth. *Nature*. 591 (7851), 592–598. doi:10.1038/s41586-021-03303-x.

Santi, I., Kasapidis, P., Karakassis, I. & Pitta, P. (2021) A comparison of DNA metabarcoding and

microscopy methodologies for the study of aquatic microbial eukaryotes. *Diversity*. 13 (5), 1–12. doi:10.3390/d13050180.

Santoferrara, L.F. (2019) Current practice in plankton metabarcoding: Optimization and error management. *Journal of Plankton Research*. 41 (5), 571–582. doi:10.1093/plankt/fbz041.

Sargent, J.R. & Falk-Petersen, S. (1988) The lipid biochemistry of calanoid copepods. *Hydrobiologia*. 167 (1), 101–114. doi:10.1007/BF00026297.

Saumweber, W.J. & Durbin, E.G. (2006) Estimating potential diapause duration in Calanus finmarchicus. *Deep-Sea Research Part II: Topical Studies in Oceanography*. 53 (23–24), 2597–2617. doi:10.1016/j.dsr2.2006.08.003.

Schmid, M.S., Maps, F. & Fortier, L. (2018) Lipid load triggers migration to diapause in Arctic Calanus copepods - Insights from underwater imaging. *Journal of Plankton Research*. 40 (3), 311–325. doi:10.1093/plankt/fby012.

Schourup-Kristensen, V., Wekerle, C., Danilov, S. & Völker, C. (2021) Seasonality of Mesoscale Phytoplankton Control in Eastern Fram Strait. *Journal of Geophysical Research: Oceans*. 126 (10), 1–24. doi:10.1029/2021JC017279.

Sievert, C. (2020) *Interactive Web-Based Data Visualization with R, plotly, and shiny*. Florida, Chapman and Hall/CRC. https://plotly-r.com.

Sildever, S., Laas, P., Kolesova, N., Lips, I., Lips, U. & Nagai, S. (2021) Plankton biodiversity and species co-occurrence based on environmental DNA – a multiple marker study. *Metabarcoding and Metagenomics*. 5, 175–197. doi:10.3897/MBMG.5.72371.

Smetacek, V. & Nicol, S. (2005) Polar ocean ecosystems in a changing world. *Nature*. 437 (7057), 362–368. doi:10.1038/nature04161.

Smith, S.L. (1990) Egg production and feeding by copepods prior to the spring bloom of phytoplankton in Fram Strait, Greenland Sea. *Marine Biology*. 106, 59–69.

Smith, S.L. & Lane, P.V.Z. (1988) Grazing of the spring diatom bloom in the New York Bight by the calanoid copepods Calanus finmarchicus, Metridia lucens and Centropages typicus. *Continental Shelf Research*. 8 (5–7), 485–509. doi:10.1016/0278-4343(88)90065-9.

Smith, S.L. & Schnack-Schiel, S.B. (1990) Polar Zooplankton. In: W. Smith (ed.). *Polar Oceanography, Part B: Chemistry, Biology, and Geology*. San Diego, CA, Academic Press. pp. 527–598.

Sommer, U., Peter, K.H., Genitsaris, S. & Moustaka-Gouni, M. (2017) Do marine phytoplankton follow Bergmann's rule sensu lato? *Biological Reviews*. 92 (2), 1011–1026. doi:10.1111/brv.12266.

Søreide, J.E., Falk-Petersen, S., Hegseth, E.N., Hop, H., Carroll, M.L., Hobson, K.A. & Blachowiak-Samolyk, K. (2008) Seasonal feeding strategies of Calanus in the high-Arctic Svalbard region. *Deep-Sea Research Part II: Topical Studies in Oceanography*. 55 (20–21), 2225–2244. doi:10.1016/j.dsr2.2008.05.024.

Søreide, J.E., Leu, E., Berge, J., Graeve, M. & Falk-Petersen, S. (2010) Timing of blooms, algal food quality and Calanus glacialis reproduction and growth in a changing Arctic. *Global Change Biology*. 16 (11), 3154–3163. doi:10.1111/j.1365-2486.2010.02175.x.

Sosik, H.M. & Olson, R.J. (2007) Automated taxonomic classification of phytoplankton sampled with imaging-in-flow cytometry. *Limnology and Oceanography: Methods*. 5 (6), 204–216. doi:10.4319/lom.2007.5.204.

Starr, M., Runge, J.A. & Therriault, J.C. (1999) Effects of diatom diets on the reproduction of the planktonic copepod Calanus finmarchicus. *Sarsia*. 84 (5–6), 379–389. doi:10.1080/00364827.1999.10807345.

Steen, H., Vogedes, D., Broms, F., Falk-Petersen, S. & Berge, J. (2007) Little auks (Alle alle) breeding in a High Arctic fjord system: Bimodal foraging strategies as a response to poor food quality? *Polar Research*. 26 (2), 118–125. doi:10.1111/j.1751-8369.2007.00022.x.

Steinacher, M., Joos, F., Frölicher, T.L., Bopp, L., Cadule, P., Cocco, V., Doney, S.C., Gehlen, M., Lindsay, K., Moore, J.K., Schneider, B. & Segschneider, J. (2010) Projected 21st century decrease in marine productivity: A multi-model analysis. *Biogeosciences*. 7 (3), 979–1005. doi:10.5194/bg-7-979-2010.

Steinberg, D.K. & Landry, M.R. (2017) Zooplankton and the Ocean Carbon Cycle. *Annual Review of Marine Science*. 9 (1), 413–444. doi:10.1146/annurev-marine-010814-015924.

Sterner, R.W. & Elser, J.J. (2002) *Ecological Stoichiometry*. Princeton University Press. doi:10.1111/j.1469-8137.2010.03214.x.

Stewart, F.J. (2013) *Preparation of microbial community cDNA for metatranscriptomic analysis in marine plankton*. 1st edition. Elsevier Inc. doi:10.1016/B978-0-12-407863-5.00010-1.

Stoeck, T., Kochems, R., Forster, D., Lejzerowicz, F. & Pawlowski, J. (2018) Metabarcoding of benthic ciliate communities shows high potential for environmental monitoring in salmon aquaculture. *Ecological Indicators*. 85 (May 2017), 153–164. doi:10.1016/j.ecolind.2017.10.041.

Straile, D. (1997) Gross growth efficiencies of protozoan and metazoan zooplankton and their dependence on food concentration, predator-prey weight ratio, and taxonomic group. *Limnology and Oceanography*. 42 (6), 1375–1385. doi:10.4319/lo.1997.42.6.1375.

Sugie, K., Fujiwara, A., Nishino, S., Kameyama, S. & Harada, N. (2020) Impacts of Temperature, CO2, and Salinity on Phytoplankton Community Composition in the Western Arctic Ocean. *Frontiers in Marine Science*. 6 (January). doi:10.3389/fmars.2019.00821.

Sunagawa, S., Coelho, L.P., Chaffron, S., Kultima, J.R., Labadie, K., et al. (2015) Structure and function of the global ocean microbiome. *Science*. 348 (6237). doi:10.1126/science.1261359.

Svensen, C., Halvorsen, E., Vernet, M., Franzè, G., Dmoch, K., Lavrentyev, P.J. & Kwasniewski, S. (2019) Zooplankton communities associated with new and regenerated primary production in the Atlantic inflow North of Svalbard. *Frontiers in Marine Science*. 6 (JUN), 1–13. doi:10.3389/fmars.2019.00293.

Swalethorp, R., Kjellerup, S., Dünweber, M., Nielsen, T.G., Møller, E.F., Rysgaard, S. & Hansen, B.W. (2011) Grazing, egg production, and biochemical evidence of differences in the life strategies of Calanus finmarchicus, C. glacialis and C. hyperboreus in Disko Bay, Western Greenland. *Marine Ecology Progress Series*. 429 (August 2014), 125–144. doi:10.3354/meps09065.

Szeligowska, M., Trudnowska, E., Boehnke, R., Dąbrowska, A.M., Wiktor, J.M., Sagan, S. & Błachowiak-Samołyk, K. (2020) Spatial Patterns of Particles and Plankton in the Warming Arctic Fjord (Isfjorden, West Spitsbergen) in Seven Consecutive Mid-Summers (2013–2019). *Frontiers in Marine Science*. 7 (July), 1–18. doi:10.3389/fmars.2020.00584.

Tande, K.S. (1982) Ecological investigations on the zooplankton community of Balsfjorden, northern Norway: generation cycles, and variations in body weight and body content of carbon and nitrogen related to overwintering and reproduction in the copepod Calanus finmarchicus. *Journal of Experimental Marine Biology and Ecology*. 62, 129–142.

Tanioka, T. & Matsumoto, K. (2020) A meta-analysis on environmental drivers of marine phytoplankton C: N: P. *Biogeosciences*. 17 (11), 2939–2954. doi:10.5194/bg-17-2939-2020.

Tarling, G.A., Belcher, A., Blackwell, M., Castellani, C., Cook, K.B., Cottier, F.R., Dewar-Fowler, V., Freer, J.J., Gerrish, L., Johnson, M.L., Last, K.S., Lindeque, P.K., Mayor, D.J., Parry, H.E., Stowasser, G. & Wootton, M. (2022a) Carbon and Lipid Contents of the Copepod Calanus finmarchicus Entering Diapause in the Fram Strait and Their Contribution to the Boreal and Arctic Lipid Pump. *Frontiers in Marine Science*. 9 (June), 1–18. doi:10.3389/fmars.2022.926462.

Tarling, G.A., Freer, J.J., Banas, N.S., Belcher, A., Blackwell, M., Castellani, C., Cook, K.B., Cottier, F.R., Daase, M., Johnson, M.L., Last, K.S., Lindeque, P.K., Mayor, D.J., Mitchell, E., Parry, H.E., Speirs, D.C., Stowasser, G. & Wootton, M. (2022b) Can a key boreal Calanus copepod species now complete its life-cycle in the Arctic? Evidence and implications for Arctic food-webs. *Ambio*. 51 (2), 333–344. doi:10.1007/s13280-021-01667-y.

Tedesco, L., Vichi, M. & Scoccimarro, E. (2019) Sea-ice algal phenology in a warmer Arctic. *Science Advances*. 5 (5). doi:10.1126/sciadv.aav4830.

Teegarden, G.J., Campbell, R.G., Anson, D.T., Ouellett, A., Westman, B.A. & Durbin, E.G. (2008) Copepod feeding response to varying Alexandrium spp. cellular toxicity and cell concentration among natural plankton samples. *Harmful Algae*. 7 (1), 33–44. doi:10.1016/j.hal.2007.05.010.

Tester, P.A. & Turner, J.T. (1990) How long does it take copepods to make eggs? *Journal of Experimental Marine Biology and Ecology*. 141 (2–3), 169–182. doi:10.1016/0022-0981(90)90222-X.

Thingstad, T.F. & Cuevas, L.A. (2010) Nutrient pathways through the microbial food web: Principles and predictability discussed, based on five different experiments. *Aquatic Microbial Ecology*. 61 (3), 249–260. doi:10.3354/ame01452.

Thomas, D.N., Arévalo-Martínez, D.L., Crocket, K.C., Große, F., Grosse, J., Schulz, K., Sühring, R. & Tessin, A. (2022) A changing Arctic Ocean. *Ambio*. 51 (2), 293–297. doi:10.1007/s13280-021-01677-w.

Thor, P., Koski, M., Tang, K.W. & Jónasdóttir, S.H. (2007) Supplemental effects of diet mixing on absorption of ingested organic carbon in the marine copepod Acartia tonsa. *Marine Ecology Progress Series*. 331, 131–138. doi:10.3354/meps331131.

Traboni, C., Calbet, A. & Saiz, E. (2020) Effects of prey trophic mode on the gross-growth efficiency of marine copepods: the case of mixoplankton. *Scientific Reports*. 10 (1), 1–14. doi:10.1038/s41598-020-69174-w.

Tremblay, G., Belzile, C., Gosselin, M., Poulin, M., Roy, S. & Tremblay, J.-É. (2009) Late summer phytoplankton distribution along a 3500 km transect in Canadian Arctic waters: Strong numerical dominance by picoeukaryotes. *Aquatic Microbial Ecology*. 54 (1), 55–70. doi:10.3354/ame01257.

Tremblay, J.-É., Anderson, L.G., Matrai, P., Coupel, P., Bélanger, S., Michel, C. & Reigstad, M. (2015) Global and regional drivers of nutrient supply, primary production and CO2 drawdown in the changing Arctic Ocean. *Progress in Oceanography*. 139, 171–196.

doi:10.1016/j.pocean.2015.08.009.

Tremblay, J.-É. & Gagnon, J. (2009) *The effects of irradiance and nutrient supply on the productivity of Arctic waters: a perspective on climate change*. Springer US. doi:10.1007/978-1-4020-9460-6_7.

Tremblay, J.-É., Simpson, K., Martin, J., Miller, L., Gratton, Y., Barber, D.G. & Price, N.M. (2008) Vertical stability and the annual dynamics of nutrients and chlorophyll fluorescence in the coastal, southeast Beaufort Sea. *Journal of Geophysical Research: Oceans*. doi:10.1029/2007JC004547.

Trudnowska, E., Balazy, K., Brown, T., Gluchowska, M. & Smolina, I. (2020) In a comfort zone and beyond — Ecological plasticity of key marine mediators. *Ecology and Evolution*. 00, 1–15. doi:10.1002/ece3.6997.

Trudnowska, E., Gluchowska, M., Beszczynska-Möller, A., Blachowiak-Samolyk, K. & Kwasniewski, S. (2016) Plankton patchiness in the Polar Front region of the west Spitsbergen Shelf. *Marine Ecology Progress Series*. 560 (November), 1–18. doi:10.3354/meps11925.

Tsubouchi, T., Våge, K., Hansen, B., Larsen, K.M.H., Østerhus, S., Johnson, C., Jónsson, S. & Valdimarsson, H. (2021) Increased ocean heat transport into the Nordic Seas and Arctic Ocean over the period 1993–2016. *Nature Climate Change*. 11 (1), 21–26. doi:10.1038/s41558-020-00941-3.

Tuerena, R.E., Hopkins, J., Buchanan, P.J., Ganeshram, R.S., Norman, L., von Appen, W., Tagliabue, A., Doncila, A., Graeve, M., Ludwichowski, K.U., Dodd, P.A., de la Vega, C., Salter, I. & Mahaffey, C. (2021) An Arctic Strait of Two Halves: The Changing Dynamics of Nutrient Uptake and Limitation Across the Fram Strait. *Global Biogeochemical Cycles*. 35 (9), 1–20. doi:10.1029/2021GB006961.

Tuerena, R.E., Mahaffey, C., Henley, S.F., de la Vega, C., Norman, L., Brand, T., Sanders, T., Debyser, M., Dähnke, K., Braun, J. & März, C. (2022) Nutrient pathways and their susceptibility to past and future change in the Eurasian Arctic Ocean. *Ambio*. 51 (2), 355–369. doi:10.1007/s13280-021-01673-0.

Turner, J.T. (2004) The Importance of Small Pelagic Planktonic Copepods and Their Role in Pelagic Marine Food Webs. *Zoological Studies*. 43 (2), 255–266.

Turner, J.T. (2015a) Zooplankton fecal pellets, marine snow, phytodetritus and the ocean's biological pump. *Progress in Oceanography*. 130, 205–248. doi:10.1016/j.pocean.2014.08.005.

Turner, J.T. (2015b) Zooplankton fecal pellets, marine snow, phytodetritus and the ocean's biological pump. *Progress in Oceanography*.130 pp.205–248. doi:10.1016/j.pocean.2014.08.005.

De Vargas, C., Audic, S., Henry, N., Decelle, J., Mahé, F., et al. (2015) Eukaryotic plankton diversity in the sunlit ocean. *Science*. 348 (6237). doi:10.1126/science.1261605.

Varpe, Ø. (2012) Fitness and phenology: Annual routines and zooplankton adaptations to seasonal cycles. *Journal of Plankton Research*. 34 (4), 267–276. doi:10.1093/plankt/fbr108.

Vehmaa, A., Larsson, P., Vidoudez, C., Pohnert, G., Reinikainen, M. & Engström-Öst, J. (2011) How will increased dinoflagellate:diatom ratios affect copepod egg production? - A case study from the Baltic Sea. *Journal of Experimental Marine Biology and Ecology*. 401 (1–2), 134–140. doi:10.1016/j.jembe.2011.01.020.

Verberk, W.C.E.P., Atkinson, D., Hoefnagel, K.N., Hirst, A.G., Horne, C.R. & Siepel, H. (2021) Shrinking body sizes in response to warming: explanations for the temperature–size rule with special emphasis on the role of oxygen. *Biological Reviews*. 96 (1), 247–268. doi:10.1111/brv.12653.

Verity, P.G., Wassmann, P., Frischer, M.E., Howard-Jones, M.H. & Allen, A.E. (2002) Grazing of phytoplankton by microzooplankton in the Barents Sea during early summer. *Journal of Marine Systems*. 38 (1–2), 109–123. doi:10.1016/S0924-7963(02)00172-0.

Vernet, M., Richardson, T.L., Metfies, K., Nöthig, E.-M. & Peeken, I. (2017) Models of Plankton Community Changes during a Warm Water Anomaly in Arctic Waters Show Altered Trophic Pathways with Minimal Changes in Carbon Export. *Frontiers in Marine Science*. 4 (May), 1–19. doi:10.3389/fmars.2017.00160.

Vilgrain, L., Maps, F., Basedow, S., Trudnowska, E., Madoui, M., Niehoff, B. & Ayata, S. (2023) Copepods' true colors: astaxanthin pigmentation as an indicator of fitness. *Ecosphere*. 14 (6), 1–25. doi:10.1002/ecs2.4489.

Villar-argaiz, M., Medina-Sánchez, J.M. & Carrillo, P. (2002) Linking Life History Strategies and Ontogeny in Crustacean Zooplankton: Implications for Homeostasis. *Ecology*. 83 (7), 1899–1914. doi:10.2307/3071773.

Visser, A.W., Grønning, J. & Jónasdóttir, S.H. (2017) Calanus hyperboreus and the lipid pump. *Limnology and Oceanography*. 62 (3), 1155–1165. doi:10.1002/lno.10492.

Van De Waal, D.B., Verschoor, A.M., Verspagen, J.M.H., Van Donk, E. & Huisman, J. (2010) Climate-driven changes in the ecological stoichiometry of aquatic ecosystems. *Frontiers in Ecology and the Environment*. 8 (3), 145–152. doi:10.1890/080178.

Walther, G.-R., Post, E., Convey, P., Menzel, A., Parmesan, C., Beebee, T.J.C., Fromentin, J.-M.,

Hoegh-Guldberg, O. & Bairlein, F. (2002) Ecological responses to recent climate change. *Nature*. 416, 389–395. doi:10.1038/416389a.

Wang, Z., Liu, L., Tang, Y., Li, A., Liu, C., Xie, C., Xiao, L. & Lu, S. (2022) Phytoplankton community and HAB species in the South China Sea detected by morphological and metabarcoding approaches. *Harmful Algae*. 118 (July), 102297. doi:10.1016/j.hal.2022.102297.

Weisse, T., Tande, K.S., Verity, P.G., Hansen, F. & Gieskes, W. (1994) The trophic significance of Phaeocystis blooms. *Journal of Marine Systems*. 5 (1), 67–79. doi:10.1016/0924-7963(94)90017-5.

Wekerle, C., Wang, Q., von Appen, W.-J., Danilov, S., Schourup-Kristensen, V. & Jung, T. (2017) Eddy-Resolving Simulation of the Atlantic Water Circulation in the Fram Strait With Focus on the Seasonal Cycle. *Journal of Geophysical Research: Oceans*. 122 (11), 8385–8405. doi:10.1002/2017JC012974.

Wickham, H. (2016) *ggplot2: Elegant Graphics for Data Analysis*. https://ggplot2.tidyverse.org.

Wickham, H., François, R., Henry, L. & Müller, K. (2022) *dplyr: A Grammar of Data Manipulation*. https://cran.r-project.org/package=dplyr.

Wilson, R.J., Banas, N.S., Heath, M.R. & Speirs, D.C. (2016) Projected impacts of 21st century climate change on diapause in Calanus finmarchicus. *Global Change Biology*. 22 (10), 3332–3340. doi:10.1111/gcb.13282.

Wilson, R.J., Heath, M.R. & Speirs, D.C. (2016) Spatial modelling of Calanus finmarchicus and Calanus helgolandicus: parameter differences explain differences in biogeography. *Frontiers in Marine Science*. 3 (September), 157. doi:10.3389/fmars.2016.00157.

Winder, M. & Sommer, U. (2012) Phytoplankton response to a changing climate. *Hydrobiologia*. 698 (1), 5–16. doi:10.1007/s10750-012-1149-2.

Yool, A., Popova, E.E. & Coward, A.C. (2015) Future change in ocean productivity: Is the Arctic the new Atlantic. *Journal of Geophysical Research: Oceans*. 120 (12), 7771–7790. doi:10.1002/2015JC011167.

Zarauz, L. & Irigoien, X. (2008) Effects of Lugol's fixation on the size structure of natural nano-microplankton samples, analyzed by means of an automatic counting method. *Journal of Plankton Research*. 30 (11), 1297–1303. doi:10.1093/plankt/fbn084.

Zeller, D., Palomares, M.L.D., Tavakolie, A., Ang, M., Belhabib, D., Cheung, W.W.L., Lam, V.W.Y., Sy, E., Tsui, G., Zylich, K. & Pauly, D. (2016) Still catching attention: Sea Around Us reconstructed

global catch data, their spatial expression and public accessibility. *Marine Policy*. 70, 145–152. doi:10.1016/j.marpol.2016.04.046.

Zhang, Y., Lin, X., Shi, X., Lin, L., Luo, H., Li, L. & Lin, S. (2019) Metatranscriptomic signatures associated with phytoplankton regime shift from diatom dominance to a dinoflagellate bloom. *Frontiers in Microbiology*. 10 (MAR), 1–13. doi:10.3389/fmicb.2019.00590.