

Master Thesis Spring 2023

Disparities in growth, condition, and contaminant exposure: A study of Atlantic cod, Norway pout, and whiting in the inner and outer Oslofjord

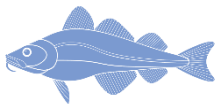
Investigating otolith data, condition indices, and exposure to pyrene: Simulating possible population-level effects of reduced growth scenarios on Atlantic cod.

Julie Marie Følstad

Marine Biology and Limnology
60 credits

Department of Biosciences
Section for Aquatic Biology and Toxicology





© Julie Marie Følstad

2023

Disparities in growth, condition, and contaminant exposure: A study of Atlantic cod, Norway pout, and whiting in the inner and outer Oslofjord

Author: Julie Marie Følstad

Department of Biosciences
Faculty of Mathematics and Natural Sciences
UNIVERSITY OF OSLO

<http://www.duo.uio.no/>

Print: Reprosentralen, University of Oslo

Abstract

The aim of this study was to quantify differences, if present, in the general condition and exposure to environmental contaminants in populations of Atlantic cod (*Gadus morhua*), whiting (*Merlangius merlangus*), and Norway pout (*Trisopterus esmarkii*) between the inner and outer Oslofjord regions. Ultimately, this study aimed to address how potential impacts on growth could translate to the population-level. The fish were sampled by bottom trawling in the inner and outer Oslofjord on four different occasions between May 2021 – February 2022. The length-weight relationship (LWR), liver somatic index (LSI), and gonad somatic index (GSI) was estimated based on gutted weight, somatic weight, fork-length, liver weight and gonad weight measurements. The LWR and LSI was compared for each species between the different sites to uncover potential disparities in condition and energy budget, while GSI was compared to determine maturity, and uncover potential disparities in reproductive investment. Contaminant exposure can have adverse effects on fish health, therefore, it was also investigated whether contaminant exposure differs between the regions using the PAH-metabolite pyrene as a proxy for general contaminant exposure. Cod otoliths were analyzed to determine age and calculate size-at-age estimates using back-calculation. Potential differences in the growth of Atlantic cod between the regions was assessed based on size-at-age estimates. The size-at-age estimates, indexes from the LWR, and size-at-maturity determined from GSI and otolith estimates, was used to adapt a population model. The population model was then used to simulate how hypothetical reduced growth scenarios could potentially affect the current Atlantic cod population in the Oslofjord. There was observed a significantly higher concentration of pyrene in inner Oslofjord, indicating that the fish in inner Oslofjord are likely to be more negatively affected by contaminants. However, there was varying results in terms of the condition indices. While there were no significant differences in the LWR for either species between the study areas, there was a significant difference in LSI for Norway pout and Atlantic cod between inner and outer Oslofjord. While Norway pout displayed higher LSI in the outer fjord, Atlantic cod displayed higher LSI in the inner fjord. Otolith analysis indicated similar growth for Atlantic cod between inner and outer Oslofjord. By adapting a population model, it was shown that reduced growth scenarios can indeed have individual-level effects, however these effects will likely be hard to detect in time-series data of abundance or biomass.

Acknowledgments

First and foremost, I would like to thank my supervisors Øystein Langangen and Ketil Hylland. Øystein, thank you for teaching me about population dynamics and coding – you make it look way easier than it is. I also want to thank you for being so thoughtful and always asking “how’s it going” whenever I come into your office. Ketil, thank you for giving me valuable field work and lab experience - and for reminding me who’s the professor whenever I mistake haddock for whiting.

A special thanks to Arild Folkvord and Julie Skadal at UiB, for teaching me about otoliths and letting me use their otolith-lab in Bergen. Big thanks to Eira and Magnus for helping me out in the lab. To the crew at Trygve Braarud, if you ever read this, thank you for the endless coffee refills and for making the data sampling such a fun experience.

Thank you to all of my Bio-friends, both old and new ones, who have made these years at KB both precious and bearable. Especially the OG’s Selma, Harald, Benjamin, Truls Nikolai, and Ragnhild for five amazing years that I’ll cherish forever (yes I did just subtract covid). Thank you to the girls in room 3525: Eli, Hanna, Tonje, Jacquelynn, and Silje for the snacks, laughs, memes, and numerous ligretto-brakes.

Thank you Truls, for being such a kind roommate and for all the pep talks during this stressful time. I also want to express my gratitude to my dear colleagues and friends Julie and Hauk, for helping me prioritize my studies. Thank you to my friends from back home, that now think I know everything related to plants and animals. Thank you to mamma og pappa for always cheering me on and for teaching me to not give up when things get hard. To my siblings, Karl Morten and Jenni, thank you for being so understanding about the fact that I almost haven’t been home the last year. Lastly, thank you to my boyfriend, Nikolai, for being the most patient, supportive, and steady guy there is. We finally did it!

Julie Marie Følstad

Kristine Bonnevis hus, June 2023.

Abbreviations

ANOVA – Analysis of variance

Annulus – The annular growth increment of the otolith.

B-C – Back-calculation

BCF – Back calculation formula

EQS – Environmental quality standards

GSI – Gonad somatic index

LSI – Liver somatic index

LWR – Length – Weight relationship

PAH – Polycyclic aromatic hydrocarbons

PCBs – Polychlorinated biphenyls

PFOS – Perfluorooctanesulfonic acid

SPH – Scale-proportional Hypothesis

UiO – University of Oslo

UiB – University of Bergen

Table of Contents

ABSTRACT	3
ACKNOWLEDGMENTS	4
ABBREVIATIONS	5
CHAPTER 1: INTRODUCTION	8
1.1 A FJORD UNDER ANTHROPOGENIC PRESSURE	8
1.2 THE STUDY.....	9
1.2.1 <i>Species of interest</i>	9
1.2.2 <i>Population-relevant parameters</i>	10
1.2.3 <i>Contaminant exposure</i>	11
1.2.4 <i>Modelling population dynamics</i>	12
1.3 OBJECTIVES AND RESEARCH QUESTIONS.....	13
1.4 AN OVERVIEW OF THE STUDY	14
CHAPTER 2: MATERIALS AND METHODS	15
2.1 DATA COLLECTION.....	15
2.2 SAMPLING PROCEDURE	17
2.3 BIOMARKERS.....	19
2.3.1 <i>Gonad Somatic Index (GSI)</i>	19
2.3.2 <i>Liver Somatic Index (LSI)</i>	19
2.3.3 <i>Linear length-weight regression</i>	19
2.4 OTOLITH ANALYSIS: DETERMINING AGE AND GROWTH.	20
2.4.1 <i>Preparing the otoliths</i>	20
2.4.2 <i>Weighing and sectioning the otoliths</i>	20
2.4.3 <i>Measuring the otoliths using ImageJ</i>	21
2.4.4 <i>Back-calculation of growth</i>	22
2.5 POLYCYCLIC AROMATIC HYDROCARBON (PAH) METABOLITES IN BILE.....	23
2.5.1 <i>Making the pyrene standards and diluting bile samples</i>	23
2.5.2 <i>Preparation of the microtiter plate</i>	23
2.5.3 <i>Measuring fluorescence</i>	23
2.5.4 <i>Cleaning procedure of the microtiter plate</i>	24
2.6 DATA ANALYSIS.....	24
2.6.1 <i>PAH-metabolites</i>	24
2.6.2 <i>Condition factors</i>	25
2.6.3 <i>Otoliths</i>	25
2.6.4 <i>Modelling population dynamics</i>	25
2.6.5 <i>Adapting the model</i> :.....	27
2.6.6 <i>Reduced growth scenarios</i>	27
2.7 <i>Additional tools</i>	28
CHAPTER 3: RESULTS	29
3.1 PAH EXPOSURE	29
3.2 CONDITION FACTORS	31
3.2.1 <i>Length-weight relationship (LWR)</i>	31
3.2.2 <i>LSI</i>	33
3.2.3 <i>GSI</i>	35
3.3 OTOLITH READINGS	37
3.3.1 <i>Size-at-age</i>	37
3.3.2 <i>Age-distribution</i>	38
3.3.3 <i>Age-at-maturity</i>	38

3.4 MODELLING POPULATION DYNAMICS	39
CHAPTER 4: DISCUSSION	42
R1: IS CONTAMINANT EXPOSURE DIFFERENT FOR FISH IN THE INNER AND OUTER OSLOFJORD?	42
R2: DOES THE CONDITION IN FISH DIFFER BETWEEN THE INNER AND OUTER OSLOFJORD?.....	43
R3: ARE THERE DIFFERENCES IN GROWTH BETWEEN THE INNER AND OUTER FJORD?	44
R4: HOW WOULD EFFECTS ON GROWTH TRANSLATE TO THE POPULATION LEVEL?	46
4.5 METHODS	47
4.6 FUTURE OUTLOOK.....	49
CHAPTER 5: CONCLUSIONS.....	50
REFERENCES	51
APPENDIX A	57
APPENDIX B.....	58
APPENDIX C.....	59
APPENDIX D	61

Chapter 1: Introduction

1.1 A fjord under anthropogenic pressure

For centuries, coastal areas have had to withstand immense anthropogenic influence (Lotze et al., 2006). The Oslofjord is no exception. Historical overfishing, coastal degradation, and pollution have all contributed to cumulative stressors that have caused ecological distress in the fjord (Miljødirektoratet, 2022). Over the past decades, there has been a notable shift in the species distribution in the fjord, with a concerning decline in Atlantic cod (*Gadus morhua*) populations in recent years (Espeland & Knutsen, 2019). The fish community in the inner Oslofjord used to be dominated by bottom-dwelling species with a strong presence of small cod fish such as fourbeard rockling (*Enchelyopus cimbrius*), Norway pout (*Trisopterus esmarkii*), and poor cod (*Trisopterus minutus*), but is now dominated by whiting (*Merlangius merlangus*) (Staalstrøm et al., 2021).

Due to its topography, the inner Oslofjord is particularly vulnerable to human impacts (Arvnes et al., 2019). This is because of a shallow sill of about 20 meters that restricts water exchange between the inner and outer fjord (Gade, 1973). The largest basin in the inner Oslofjord, Vestfjorden, undergoes an annual deep-water exchange with the outer Oslofjord (Stigebrandt et al., 2002). This makes the inner fjord susceptible to the accumulation of contaminants, nutrient accumulation, and oxygen depletion (NIVA, 2019). Inner Oslofjord is surrounded by a densely populated area of about 1 million people (Staalstrøm, 2020). Urbanization contributes to various sources of contaminants, such as runoff from urban areas, sewage, and stormwater (Grung et al., 2021; Ruus et al., 2019). In addition, industry, agriculture, and maritime traffic, particularly at Norway's largest cargo and passenger port, Oslo Port, also contribute to the contamination of the fjord (Arvnes et al., 2019; Hylland, 2006; Oslo Havn KF, 2019). On average, Oslo Port receives about 50 to 70 ships per week, which further exacerbates the cumulative impact of urbanization on the fjord (Oslo Havn KF).

Pollution has been a long-lasting issue for the fjord, which has been thoroughly examined in terms of environmental conditions. Despite the implementation of various restorative measures, the monitoring program "Contaminants in coastal waters" (MILKYS) has revealed that sediment concentrations of several contaminants, including the heavy metals mercury (Hg), lead (Pb), and nickel (Ni), as well as the persistent organic pollutants PFOS and PCB,

continue to surpass the Environmental Quality Standards (EQS) (Arvnes et al., 2019). Studies have also suggested that pollution can have negative effects on Atlantic cod population dynamics in the coastal waters of Norway, including the Oslofjord (Ono et al., 2019). Others have also found that fish sampled at sites of higher contamination had smaller size-at-age according to otoliths in relation to body size (Bose et al., 2018), which is something that this study will delve further into.

1.2 The study

This study aims to investigate potential differences in contaminant exposure, condition, and growth between the inner and outer Oslofjord regions by studying three different fish species: Atlantic cod (*Gadus morhua*), whiting (*Merlangius merlangus*), and Norway pout (*Trisopterus esmarkii*), before upscaling potential effects of reduced growth scenarios to the population level for Atlantic cod specifically (Carroll et al., 2018; Langangen et al., 2023; Ohlberger & Langangen, 2015). All three species are commercially and recreationally important (Staalstrøm et al., 2021), and were chosen based on ecological and economic relevance. Understanding their life history traits and ecology is essential for effective conservation and management.

1.2.1 Species of interest

Atlantic cod is an important commercial fish species. However, it is also under heavy exploitation. Cod can be described as an omnivorous predator with a broad diet (Daan, 1973). There are two genetically distinct ecotypes found to coexist in the Oslofjord, a “fjord” ecotype and a “North Sea” ecotype, i.e. coastal cod and North Sea cod (Knutsen et al., 2018). With the coastal cod typically being the dominating ecotype in the fjords. The North Sea ecotype is typically larger compared to the coastal ecotype (Knutsen et al., 2018). Coastal cod reaches maturity earlier around age two to three (Olsen et al., 2004), and North Sea cod has been found to reach maturity later, between age two to six (Cook et al., 1999).

In recent years, whiting has dominated the fish community in the inner Oslofjord (Staalstrøm et al., 2021). Whiting subsists mainly on crustaceans and small fish (Hislop et al., 1991). Whiting in the North Sea has been found to mature around the age of two to three (Hislop & Hall, 1974). However, it still remains unclear whether the whiting spawns in the inner Oslofjord (Staalstrøm et al., 2021).

Norway pout has a short longevity, with a maximum reported age of five years (Nigrelli, 1959). Norway pout is of high commercial importance as it is widely used for fishmeal (Moen & Svensen, 2020). It is also a key prey species for other commercially important predator species such as cod, whiting, saithe (*Pollachius virens*), and haddock (*Melanogrammus aeglefinus*) (Bigné et al., 2019). Norway pout is a fast-growing species, subsisting on planktonic crustaceans, and typically reach maturity between one to two years of age (Cohen, 1990).

1.2.2 Population-relevant parameters

Population-relevant parameters, including length and weight measurements, size-at-age estimates, and condition indices, are relevant parameters to investigate growth and metabolic performance (Rose, 2019). These parameters will thus be utilized to assess potential differences in the condition of the species of interest, between the inner and outer Oslofjord.

The relationship between length and weight (LWR) can be a useful indicator of the growth pattern of the population (Rose, 2019; Yaragina & Marshall, 2000). When using gutted weight, it isolates the energy stored in muscle (Sherwood et al., 2007). The LWR can thus contribute to uncover energy deficits or surpluses in the energy budget. The Liver Somatic Index (LSI) also gives insight into the energy reserves of the fish, as it reflects the lipid content in the liver (Yaragina & Marshall, 2000). Both field and laboratory experiments have shown both increased and decreased LSI values in fish exposed to PAHs and other contaminants (van der Oost et al., 2003). The liver plays a vital role in detoxifying xenobiotics, and an increase in metabolic activity has been linked to liver enlargement (Bernet et al., 2000; Samanta et al., 2018). When fish are exposed to contaminants it may also disturb energy allocation, as ingested energy is allocated towards detoxification and repair mechanisms rather than maintenance (Pi et al., 2016). For females, LSI can also vary significantly depending on reproductive status since the liver plays an essential role in vitellogenesis (Dahle et al., 2003).

The maturity of a fish and how much energy it invests in reproduction can be indicated by the gonad somatic index (GSI). Throughout the reproductive cycle, the GSI will vary, following gonadal maturation and reaching its peak during the spawning season (Rizzo & Bazzoli, 2020).

A relevant approach is back-calculation (B-C) which is utilized to estimate a fish's length in the past by using its current body length and the corresponding dimensions of growth marks found in calcified structures like otoliths (Francis, 1990). This estimated length is termed back-

calculated length (BCL). While physiological parameters provide a rough estimate of the fish's current overall health, growth analysis from otolith reading provides valuable time series data on individual fish growth and earlier size-at-age estimates. (Hoie et al., 2008). B-C is an alternative to the mark-recapture method in fish growth studies, as it can be applied to all captured individuals in the stock, not just marked-recaptured individuals, providing an advantage for field-based studies (Folkvord & Mosegaard, 2002).

Otoliths, which are located in the inner ear of the fish, grow throughout the life in proportion to body length (Hoie et al., 2008). Otoliths are valuable sources of information about a fish's age, environmental conditions, growth rate, reproduction, and changes in diet (Midway, 2014). Among teleost fishes, there are three pairs of otoliths: *sagitta*, *astericus*, and *lapillus*. For this study, the largest otolith, *sagitta*, was used. Otoliths grow differently throughout the year, with denser growth during the winter when the metabolic rate is low and less dense growth in the summer when the metabolic rate is high. This results in opaque "winter zones" and translucent "summer zones" (Midway, 2014).

It has been observed that the relationship between somatic growth and otolith growth can fluctuate due to changes in temperature (Folkvord et al., 2004). In some species, otolith growth has been found to increase in warmer temperatures, resulting in higher growth rates compared to cooler temperatures. Otolith growth is, in other words, a complex process dependent on a combination of temperature and metabolism (Fablet et al., 2011).

1.2.3 Contaminant exposure.

In order to investigate whether there are differences in contamination between the study areas, this study will use the PAH-metabolite pyrene as a proxy for general contaminant exposure. Polycyclic aromatic hydrocarbons (PAHs) entering the environment are typically derived from oil or combustion-related processes, such as soot particles (Lima et al., 2005). PAHs have been reported to cause many adverse effects, including suppressing the immune system (Reynaud & Deschaux, 2006), developmental toxicity (Incardona et al., 2006; Rhodes et al., 2005), genotoxicity, metabolic effects, reducing growth, reproductive suppression, and endocrine disruption (Sparling, 2016). Exposure to PAH has also been shown to cause developmental abnormalities, such as malformations in embryos and larvae in haddock (Sørhus et al., 2015).

When fish are exposed to PAHs via water or diet (Grung et al., 2009), most PAHs are converted into more hydrophilic compounds that are excreted via bile (Sparling, 2016). That is the reason concentration of PAH-metabolites in bile indicates *recent* exposure to PAH (van der Oost et al., 2003). PAHs can provide a general idea of contaminant exposure as they are widely found in the environment due to anthropogenic activity. To account for potential temporal differences in PAH exposure, sampling was done on four separate occasions throughout the year (May 2021 – Feb. 2022).

1.2.4 Modelling population dynamics

Ultimately, this study will simulate possible population-level effects from reduced growth scenarios (Spromberg & Birge, 2005). Modeling population dynamics can be used in situations where experiments are difficult, in order to evaluate conservation efforts and management strategies in ecological communities, such as the Oslofjord (Carroll et al., 2018; Langangen et al., 2023). This study aims, through a scenario approach, to quantify the potential effects of different perturbations in growth by adapting a population model to field observations. Incorporating field-based observations enables the possibility of simulating the population dynamics of the fish populations in the fjord under different conditions. The scenario approach enables investigation of how possible widespread reduced growth scenarios could possibly affect the population.

1.3 Objectives and research questions

The objective of this study was to quantify differences, if present, in the general condition and exposure to environmental contaminants in populations of Atlantic cod, whiting, and Norway pout between the inner and outer Oslofjord regions. Ultimately, this study aimed to address how potential impacts on growth can translate to the population-level. Concentrations of PAH-metabolite pyrene was quantified as proxies for general contaminant exposure, and condition factors LSI and LWR as an estimate of health plus GSI as an indicator for maturity and reproductive investment. In addition, otolith data was used to back-calculate growth and assess whether growth is different between the two areas. Growth estimates were used in a population dynamics model to determine potential effects of reduction in growth.

The following research questions will be investigated in this study:

1. R1: Is contaminant exposure different for fish in the inner and outer Oslofjord?
2. R2: Does the condition in fish differ between the inner and outer Oslofjord?
3. R3: Are there differences in growth between the inner and outer Oslofjord?
4. R4: Will effects on growth translate to modelled changes at the population level?

Which will be as the following and corresponding hypothesis:

1. H1: Fish in inner Oslofjord are more exposed to contaminants than outer Oslofjord.
2. H2: Fish in inner Oslofjord have lower condition than outer Oslofjord.
3. H3: Fish in inner Oslofjord have lower growth than outer Oslofjord.
4. H4: Reduced growth can translate to modelled changes at the population-level.

1.4 An overview of the study

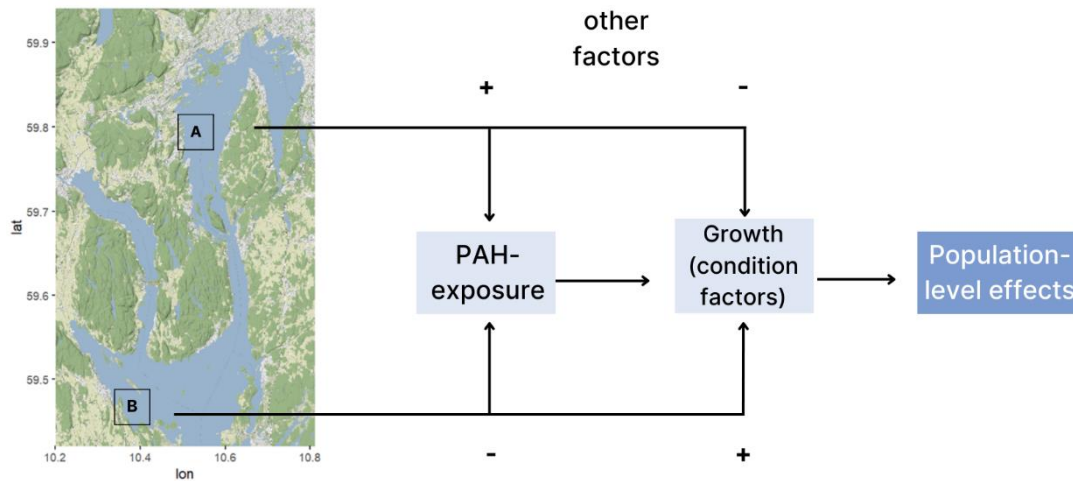


Figure 1: An overview of the main elements of this study. Firstly, data sampling in area A and B: area A is the sampling site in the inner Oslofjord, while area B is the sampling site in the outer Oslofjord. Secondly, assessing PAH-exposure by analyzing the PAH-metabolite pyrene in bile, then estimating growth based on size-at-age estimates from otoliths. Condition factors (LWR, LSI, GSI) were investigated to assess health and reproductive investment. Otolith and GSI data was also used to determine age-at-maturity. Other relevant factors will be discussed. Finally, all these measurements were upscaled to a population level to assess and simulate whether there could be any population level effects from reduced growth scenarios.

After sampling in the two sites (A = inner fjord, B = outer fjord), PAH-exposure, condition, growth, and population-level effects will be assessed. Since the inner fjord is more exposed to anthropogenic impacts, it is expected that the PAH exposure is greater in site A (+) than in site B (-). As pollution can have adverse effects, it is hypothesized that growth and condition factors could possibly be lower in site A (-) than in site B (+). This study did not address other factors, such as diet, temperature, genetics, oxygen concentration, etc., but they are included in the discussion. Finally, assessing if the effects are observable at the population level. Growth and condition factors were upscaled to the population level by using the field data to adapt a population dynamics model similar to Langangen et. al., 2023, before simulating potential population-level effects of reduced growth scenarios.

Chapter 2: Materials and Methods

2.1 Data collection

Atlantic cod, whiting, and Norway pout were collected by bottom trawling with the research vessel Trygve Braarud in the inner and Outer Oslofjord on four separate occasions: May 2021, September 2021, October 2021, and February 2022. Each cruise lasted two to three days, depending on the vessel's availability. Our sampling site in the inner Oslofjord was Midtmeie (square A, Fig. 2), while our reference site in the outer Oslofjord was Holmestrandfjorden (square B, Fig. 2).

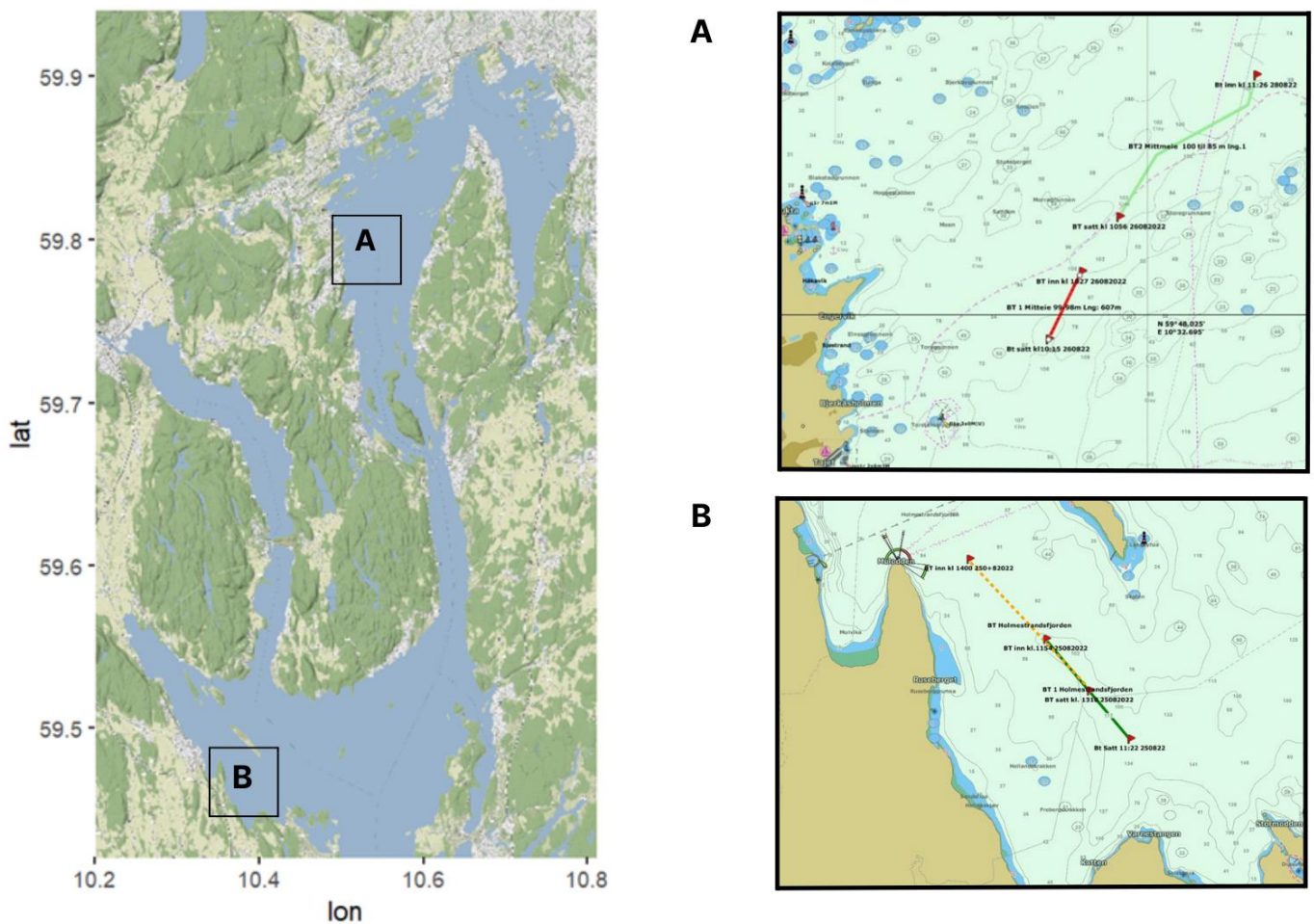


Figure 2: The map on the left shows an overview of the inner and outer Oslofjord, where squares indicate the two sampling sites shown on the right side. The zoomed-in maps on the right indicate the trawling tracks. Square A in the inner Oslofjord, Midtmeie, where the trawling track is illustrated in a long, green, and short red line. Square B shows the trawling track outer Oslofjord, indicated by the green line. The orange line shows the remaining part of the track that could have been trawled but was omitted in order to maximize the condition of the fish.

At each location, one or more trawls were performed (see Appendix A), each lasting 30-40 minutes with a speed of 1.5 knots. Trawling depth ranged between 130- and 90-meter intervals in the outer fjord. For the inner fjord, the trawling depth ranged between 110- and 90-meter intervals.

The dimension of the trawl was twenty meters in width and six meters in height. Sonars on both the boat and the trawl enabled the captain to monitor the catch.



Figure 3: Images show the research vessel Trygve Braarud on the left-hand side, and the process of retrieving the trawl on the right-hand side.

2.2 Sampling procedure

After sorting and counting the catch and discarding the bycatch, all of the captured Atlantic cod was sampled and between 10 to 20 individuals of whiting and Norway pout, depending on the catch and available time. The sampled individuals were selected randomly.



Figure 4: To the left, you can see co-supervisor Ketil Hylland counting and sorting some Whiting. To the right, you can see the tub where we kept the cod for additional blood sampling for other projects, accompanied by a thornback ray we got as a bycatch, which was later released.

The fish were euthanized by a hard blow to the head before measuring the fork length (cm) with an accuracy of (\pm) 0.5 cm and weight (g) with an accuracy of (\pm) 0.5 g. Noticeable irregularities, such as parasites, unusual pigmentation, or appearance, were noted before and during dissection.



Figure 5: Atlantic Cod ready for fork length measurements and dissection.

The abdomen was opened by dissecting from the pelvic fins and down below the anus. Bile was extracted from the gall bladder using a single-use 1-mL syringe (HSW Soft-Ject®) and a 0.5x25mm needle (HSW Fine-Ject®). Extracting the bile first was prioritized due to the risk of the gall bladder bursting during dissection and contamination from other tissues. The entire gall bladder often had to be removed for smaller fish (particularly Norway pout) due to the difficulty of extracting it. The bile sample was transferred to an Eppendorf tube and stored in a -20°C freezer. The sample was protected from light until analysis.

Then liver and gonads were excised and weighed (g). Gonads were also used to determine gender (M – male, F – female). After discarding the remaining intestines, the carcass was weighed (gutted weight). Lastly, extracting the sagittal otoliths by cutting open the skull, revealing the sagittal otoliths on the left and right-hand side of the brain. Both otoliths were extracted and stored in Eppendorf tubes at room temperature until further analysis.

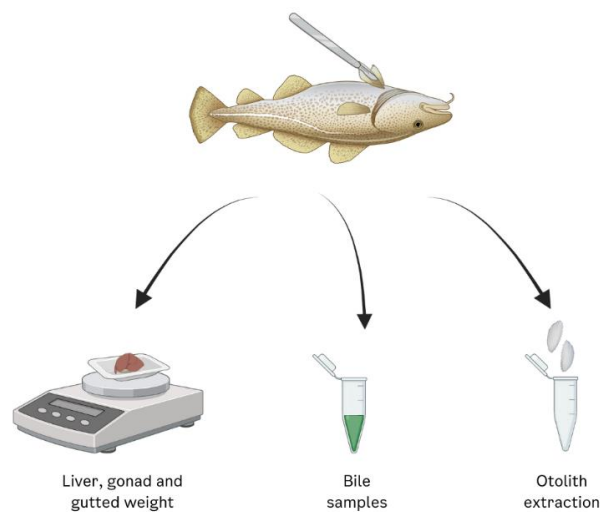


Figure 6: Illustration of the sampling procedure during dissection. This illustration was made using biorender.com and canva.com.

2.3 Biomarkers

2.3.1 Gonad Somatic Index (GSI)

GSI, the ratio between gonad and somatic body weight, indicates the fish's reproductive maturity by showing whether its juvenile, mature, or maturing. Threshold values for mature/developing individuals are set to > 1% for males and > 2.5% for females. Individuals with a GSI below the respective values are juvenile. GSI was calculated using the following formula:

$$GSI = \frac{\text{gonad weight}(g)}{\text{somatic weight}(g)} \times 100$$

Equation 1

2.3.2 Liver Somatic Index (LSI)

LSI is the ratio between liver weight and somatic body weight, and indicates amount of energy stored (lipids) in the liver. LSI was calculated using the following formula:

$$LSI = \frac{\text{liver weight}(g)}{\text{somatic weight}(g)} \times 100$$

Equation 2

2.3.3 Linear length-weight regression

The relationship between length and weight (LWR) can give an indication of growth patterns and the condition ontogeny within a fish population (Rose, 2019). This relationship was calculated by performing a linear regression on log transformed length and gutted weight measurements in Rstudio. Gutted weight, i.e. exclusion of gonads, liver and stomach contents, isolates energy stored in body musculature (Sherwood et al., 2007).

LWR can be described like this:

$$W = aL^b$$

Equation 3

And in its logarithmic form like this:

$$\log W = \log a + b \log L$$

Equation 4

Where W is weight, a is a constant, L is length and b is the allometric scaling coefficient (Froese, 2006).

2.4 Otolith analysis: determining age and growth.

The University of Bergen kindly lent its Otolith-Laboratory to conduct the otolith analysis for two weeks, one in March 2022 and the other in October 2022.

The initial plan was to analyze the otoliths of all three species: Atlantic cod, whiting, and Norway pout. However, whiting otoliths proved tricky to analyze. It took much trial and error before it was decided to discard the whiting otoliths. Cod otoliths were prioritized for analysis.

2.4.1 Preparing the otoliths.

Otoliths were stored in airtight Eppendorf tubes after capture, and the remaining debris was wiped off before leaving them in a drying cabinet overnight. When the otoliths were dry, they were placed on a dark surface under a Leica N125 microscope to look for signs of damage. The otoliths were placed in pairs: side by side (left and right) during the comparison.

2.4.2 Weighing and sectioning the otoliths.

The left otolith was used as a default for further analysis. When there were signs of damage or abnormality, the right otolith was used instead. The left otolith was weighed (mg) before being embedded in epoxy resin and sectioned in the transverse plane (Fig. 7).

The epoxy was made using a 9:5 ratio of lamination (*NM Laminering 275 A*) and hardener (*NM Härdare 275 B*) and set to dry overnight. From the hardened epoxy blocks, 450 μm thick sections were cut through the core area of the sagittae using two low-speed diamond wafering saw blades. Sections were then glued to microscope slides and polished using high to low-grit sandpaper. Images were taken with a Nikon DS F12 camera before and after polishing in transmitted white light. The following default resolution settings were used: fast (focus) 2560 x 1920 Fine and quality (capture) 2560 x 1920 Fine – 8bit. Calibration images for each magnification were taken using a Leica calibration ruler. Whenever the core of the

sagittae was missed, or sections were of poor quality, one or more sections were cut from the same otolith.

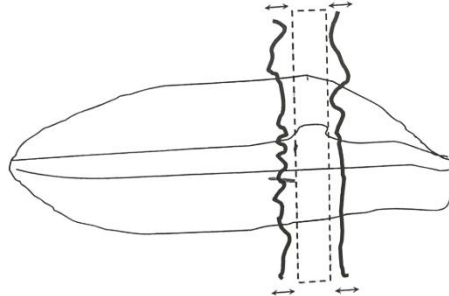


Figure 7: Schematic representation of where the otoliths were sectioned, which was at the dotted lines. The solid scribbled lines represent the part of the otolith that is lost during sectioning.

2.4.3. Measuring the otoliths using ImageJ

The correct scale (pixels/ μm) was set using corresponding calibration images. This step was repeated with the corresponding calibration image whenever another magnification was applied.

The core, i.e. the focus-point, was marked using the multi-point tool function in ImageJ. It was essential to mark the core first since all other measurements would be relative to this point. The annular growth increment (annulus) was marked at the translucent zones' outer edge along the otolith's dorsal radius (Fig. 8, left). It was decided to use the dorsal radius (Fig. 8, right) of the otolith for analysis. XY coordinates for each point were retrieved from each marking using the 'measure' function in ImageJ. Measurements were exported into Excel for back-calculation of growth.

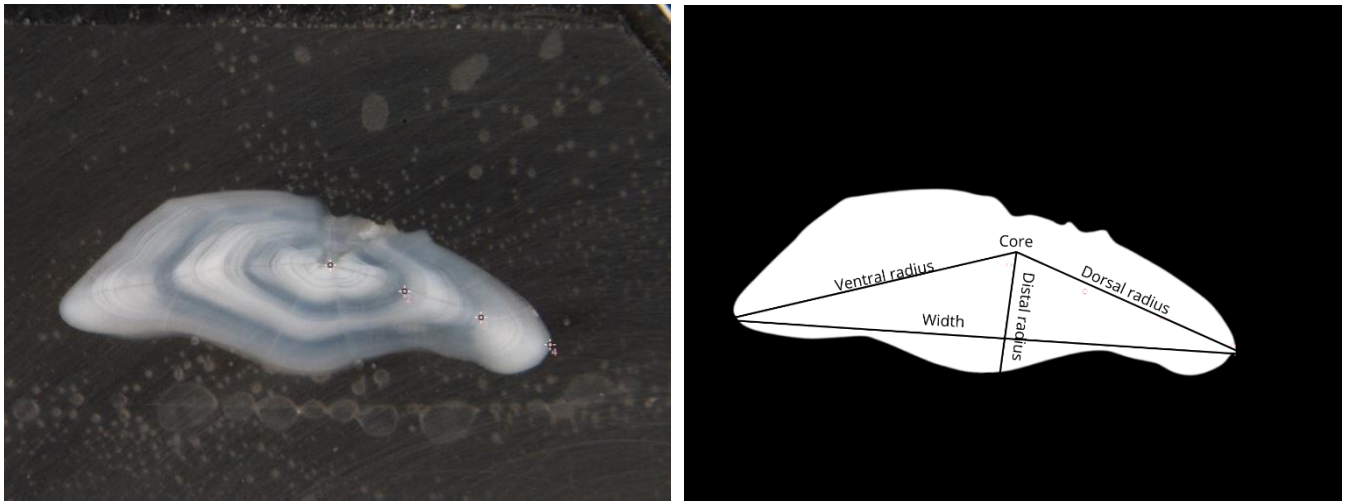


Figure 8: Example of how the points were marked along the dorsal radius of the otolith to the left: the otolith in the example was estimated to be two years old. To the right there is a schematic representation of the different radiuses on the otolith. The illustration to the right was made in canva.com.

2.4.4. Back-calculation of growth

The distance between each annulus was calculated based on the XY coordinates retrieved from the measurements in ImageJ. Then, the back-calculation of growth was done based on the scale-proportional hypothesis (SPH) developed by Whitney and Carlander (1956). The SPH suggests that otolith growth is proportional to the overall development of the fish, which leads to the following back-calculation formula (BCF):

$$f(L_i) = (S_i/S_c)fL_c$$

Equation 5

Fish length (L_i) at age (i) is based on length at capture (L_c), otolith size at capture (S_c), and otolith size at age (S_i) (R. Francis, 1990).

Age was determined for all individuals by counting the number of annuli in the pictures of the sectioned otoliths. Back-calculation of growth and size-at-age two was estimated using the BCF above. Estimates for size-at-age was obtained for all individuals above the age of two, which was approximately 52 individuals. The motivation behind excluding individuals below the age of two was to reduce potential variation due to early-life variability.

2.5 Polycyclic aromatic hydrocarbon (PAH) metabolites in bile

2.5.1 Making the pyrene standards and diluting bile samples.

Pyrene (10.28 mg) was dissolved in 500 μl of the solution (50:50 H_2O and MeOH) and 500 μl methanol, giving an initial concentration of 10.28 mg/mL. Due to the high pyrene concentration, the standard had to be diluted extensively to match the pyrene concentration of the samples. A 5000x (0.002056 mg/ml) diluted standard was made using the dilution buffer before making a final dilution series of 4.112×10^{-11} mg/ml, 2.056×10^{-11} mg/ml, and 1.028×10^{-11} mg/ml, referred to as STD3, STD2, and STD1.

Bile was taken out of the freezer to thaw for around 15 minutes. Meanwhile, preparing the solution from 50:50 MeOH and distilled H_2O to dilute the bile samples 2000x through a dilution series (20x + 100x dilution).

2.5.2 Preparation of the microtiter plate

A transparent 96-well quartz plate was used for all measurements. 200 μl of the 1:2000 diluted samples were pipetted in quadruplicate into wells. In addition to diluted samples, 200 μl quadruplicates of blanks (solution) and the three standards (STD1, STD2, STD3) were added.

In each individual analysis in the fluorometer, the microtiter plate contained up to 20 samples, one blank, and three different standards (Fig. 9).

2.5.3 Measuring fluorescence

Fluorescence was quantified at excitation/emission wavelengths for pyrene metabolites using a BioTek Synergy MX plate reader, i.e., 341/383 nm. The data was then exported into Excel for further analysis.

The fluorescence at the excitation/emission wavelengths for 2- and 3-ring PAHs: 290/335 nm and for 3-OH-benzo[a]pyrene: 379/425 nm was also measured. Since no standards were used for either PAH metabolite, the focus will be pyrene. The following settings were used for all measurements with the plate reader: slit – 13.5 nm, optics position: top, sensitivity: auto.

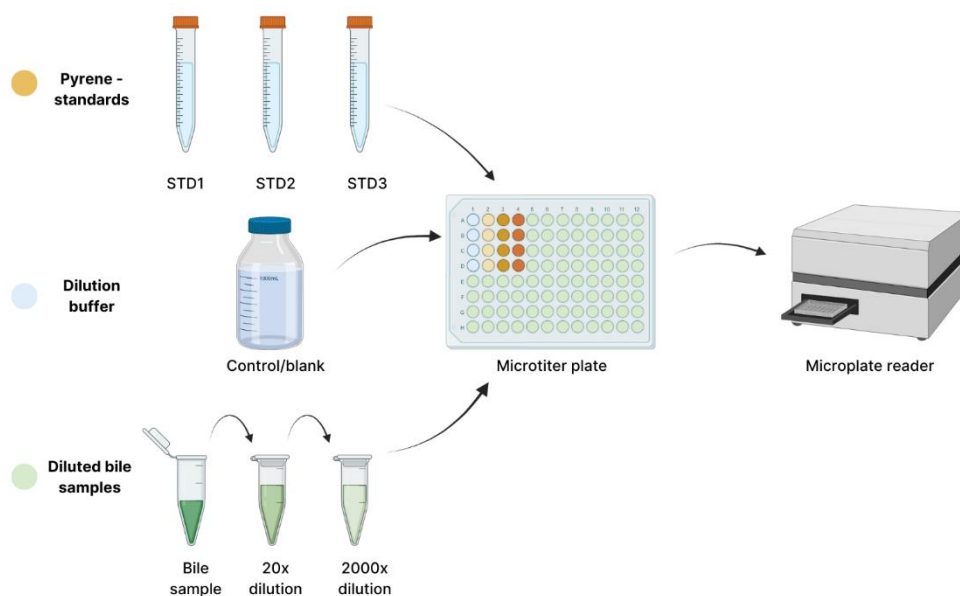


Figure 9: Illustration of the set-up of the microtiter plate. Quadruplicates of the control (MeOH + H₂O) were added to the first row (blue dots), followed by the diluted standards (orange dots). All the green dots were quadruplicates of the diluted bile samples. The dilution series is illustrated in the lower left corner of the picture. This illustration was made using biorender.com and canva.com.

2.5.4 Cleaning procedure of the microtiter plate

After measuring fluorescence, the microtiter plate was emptied and rinsed thoroughly with distilled water and dilution buffer. This step was repeated approximately six times before placing the plate upside down on a paper towel to dry off.

2.6 Data analysis

All data analysis in this study was performed in RStudio (version 4.1.1, 2021-08-10). Parts of the code to the plots in this study were generated by chat.openai.com, and modified.

2.6.1 PAH-metabolites

The potential difference in pyrene concentration between locations was assessed by a two-way ANOVA with location and month as explanatory variables. Assumptions of normality was checked both visually (histogram and Q-Q plot) and by performing a Shapiro-Wilk normality test (Shapiro & Wilk, 1965). To test for the homogeneity of variances, a Levene's test was performed (Levene, 1952). If the data did not fulfill the assumptions of homogeneity or normal distribution, it was log₁₀-transformed and retested. Difference in metabolite-concentration between species did not meet the assumptions of homogenous variance, thus a non-

parametric Kruskal-Wallis test on non-transformed data was used instead (Kruskal & Wallis, 1952).

2.6.2 Condition factors

The relationship between length and weight was assessed by performing a linear regression, using the “lm ()” - function in R to fit the model. The model used log(length) as the predictor variable and log(weight) as the response variable. Regression lines were plotted using for each species at each site, along with scatter plots of the data, using the package “ggplot”. Model diagnostics were then checked to assess goodness of fit.

LSI and GSI was assessed by performing individual two-way ANOVAs for all three species, where LSI and GSI were response variables and location was the explanatory variable. Assumptions of normal distribution and homogeneous variance was assessed using the same method as described in section 2.6.1. If assumptions were met, a two-way ANOVA was performed. If data did not meet the assumptions of ANOVA, a non-parametric Kruskal-Wallis test was performed.

2.6.3 Otoliths

To assess whether there was a difference in growth between the inner and outer fjord, the estimated size-at-age-two was evaluated using a student t-test. Data was checked for variance homogeneity by performing a Levene's test.

2.6.4 Modelling population dynamics

In order to simulate the potential impact of decreased growth on the population level, a population dynamics model similar to the model used by (Langangen et al., 2023) was utilized. Langangen et. al applied the model to 40 distinct fish species to evaluate early life survival following mass mortality events. The present study will adapt the species-specific model for Atlantic cod to our collected field data and simulate how potential changes in growth can impact the current cod population in the Oslofjord. To do so, the model was modified to the collected data by analyzing otolith data and adjusting mortality rates and growth parameters, originally from Fish Base.

The initial model from Langangen et al., 2023 is built on empirical data from two sources: stock-recruitment data from the original R. Myers database (Myers et al., 1995) and biological parameters, including growth, natural mortality, and age-at-maturity, from Fish Base (Froese

& Pauly, 2000). The data used for the model is based on Atlantic cod in the Skagerrak region (Myers et al., 1995). Firstly, the model uses a linearized Ricker model (Ricker, 1954) to describe the stock-recruitment relationship, i.e. the relationship between recruits and spawners:

$$\log\left(\frac{R_t}{S_t}\right) = \beta_0 + \beta_1 S_t + \epsilon_t$$

Equation 6

R_t and S_t represent recruit- and spawner biomass, respectively. β_0 and β_1 are constants which represent density-independent and density-dependent effects. Additionally, the script includes an error term, ϵ_t , to account for autocorrelation and for the residuals from the fit used to generate stochastic population dynamics between R_t and S_t . Furthermore, the model calculates the abundance at age by adding an age-structured model which accounts for growth and mortality:

$$N_{a,t} = \begin{cases} R_t & \text{for } a = r \\ N_{a-1,t-1} \exp(-(M + F)) & \text{for } a > r \end{cases}$$

Equation 7

$N_{a,t}$ represents the abundance at age a in year t . R_t (number of recruits in year t) obtained from the stock-recruitment model. M and F represent natural and fishing mortality, respectively, and r represents age at recruitment, which was set to 1. Finally, the model generates a value for spawning stock biomass over time (S_t), calculated as:

$$S_t = \sum_{a=a_{mat}}^{max-age} W_a \cdot N_{a,t}$$

Equation 8

Max-age is the number of year classes included in the model, which was calculated based on the Hoenig relationship which is expressed as $t_{max} = \exp((1.46 - \ln(M))/1.01)$ (Hoenig, 1983). W_a represents weight at age, assumed to follow a von Bertalanffy growth curve and $N_{a,t}$ is

abundance at age a , obtained from the age-structured model (Eq. 7). The von Bertalanffy growth curve is expressed as:

$$W_a = W_\infty(1 - \exp(-k(a - t_0)))^b$$

Equation 9

W_∞ is the maximum weight, k is the growth rate, and t_0 scales the size at age zero, which was set to zero. b is the power coefficient in the length-weight relationship, which will be adapted to field-data.

2.6.5 Adapting the model:

This study will simulate the population's total biomass over 100 years. Years with large trends in the dynamics were removed due to initial conditions, implementing a stable start in the population dynamics. Age-at-maturity was determined from otolith and GSI data. The growth parameter was k adapted to fit the size-at-age data. The b and a values (Eq. 9) was obtained from the calculated LWR.

2.6.6 Reduced growth scenarios

Reduced growth scenarios were simulated by altering the growth parameter k (Eq. 9). The growth was reduced 5%, 10%, and 15% in separate growth scenarios. These particular values were chosen based on the maximum observable differences in size-at-age-two between the inner and outer Oslofjord. The reduction in growth was simulated in one, five, and ten-year scenarios – where the respective reduction in growth was applied to every year throughout the scenario. Each scenario was simulated approximately ten times. The model was first run for the same amount of times before being adapted to field data to account for potential differences. After the simulated reduced growth scenarios, the population dynamics were investigated and compared to the unperturbed population dynamics curve. The decrease in population biomass was calculated for each scenario, with the total impact being defined as

the relative difference between the perturbed and the unperturbed biomass at the highest measured biomass ten years post-impact. The adapted model can be found in Appendix D.

2.7 Additional tools

Grammarly.com and chat.openai.com was used to elevate the language in some parts of this thesis. Canva.com and Biorender.com was used to create and modify the visuals.

Chapter 3: Results

3.1 PAH exposure

A two-way ANOVA on log-transformed data was performed to assess the difference in exposure to pyrene between the two sites (Fig. 10). The inner fjord measured a significantly higher concentration of pyrene than the outer fjord ($p = 0.01$).

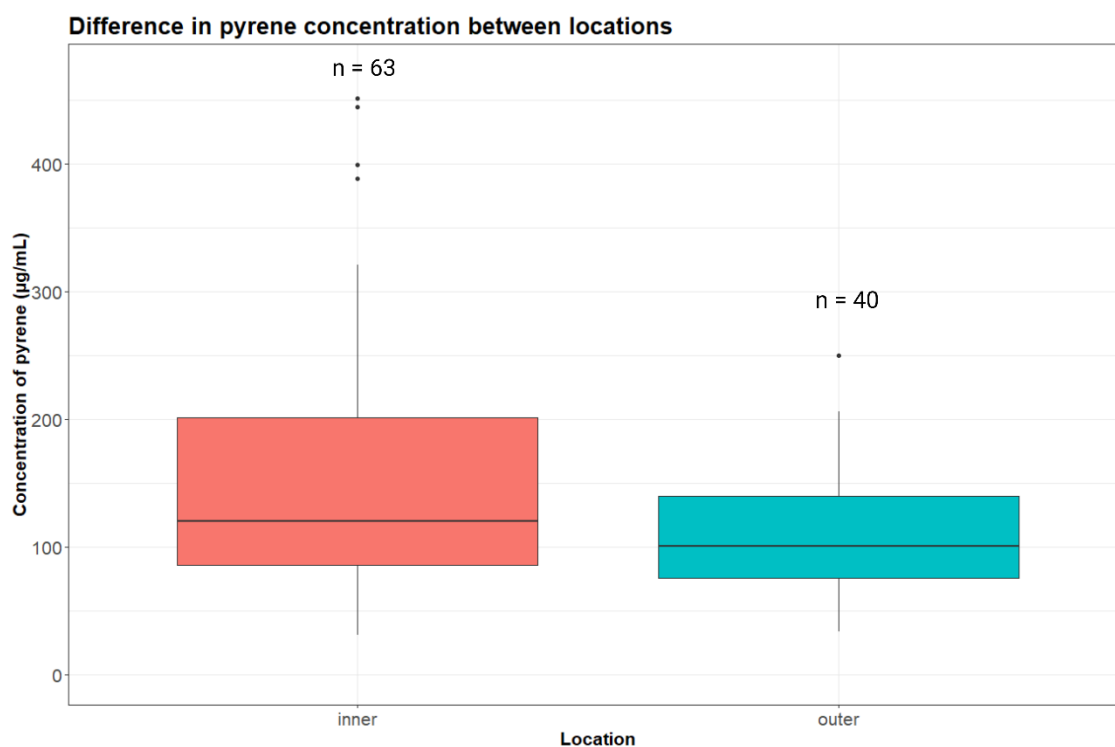


Figure 10: Measured difference in pyrene concentration between the inner and outer Oslofjord. The boxes represent the first and third quantiles, while the whiskers represent the maximum and minimum values. The thick, horizontal, and black lines represent median values. Dots represent outliers. N = sample size.

Pyrene exposure was also compared between months in the inner fjord (Fig. 11). A two-way ANOVA on log-transformed data showed no significant difference between months ($p = 0.12$). Pyrene exposure between months in the outer fjord was not statistically tested due to lack of bile samples in September, and low sample size in October and February (Appendix B, Fig.1)

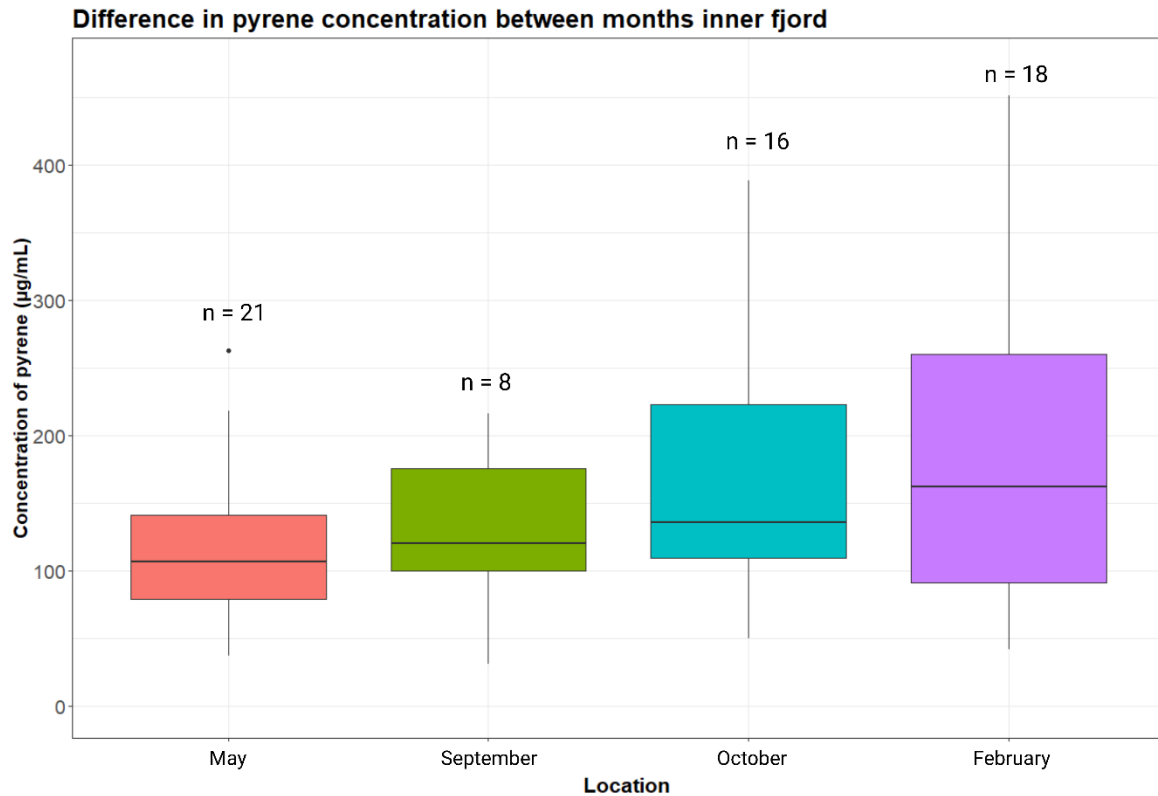


Figure 11: Pyrene concentration in different months in the inner Oslofjord. The boxes represent the first and third quartiles, while the whiskers represent the maximum and minimum values. The thick, horizontal, and black lines represent median values. The dots represent outliers. N = sample size.

Due to large differences in sample sizes for the species in outer fjord, difference in pyrene-exposure between species was not assessed for this site (Appendix B, Fig. 2). For the same reason, Norway pout was not included in the statistical testing for difference in pyrene-exposure between species in the inner fjord. A non-parametric Kruskal-Wallis test was used to compare the pyrene exposure between Atlantic cod and whiting in the inner fjord (Fig. 12). The Kruskal-Wallis test on non-transformed data showed no significant difference in pyrene exposure ($p = 0.8$).

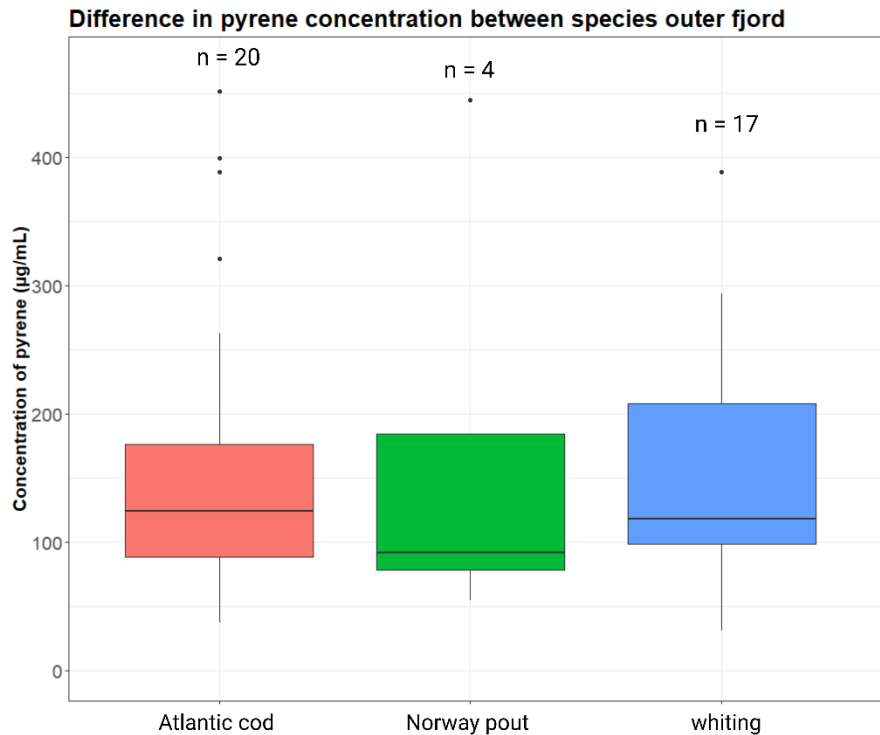


Figure 12: Pyrene concentration in Atlantic cod, Norway pout and whiting. The boxes represent the first and third quantiles, while the whiskers represent the maximum and minimum values. Median values are represented by the thick, horizontal, black lines. The dots represent the outliers. N = sample size.

3.2 Condition factors

3.2.1 Length-weight relationship (LWR)

Mean, median, and range measurements from gutted weight and length for all three species are presented in Table 1 and 2. The measurements are based upon 51 Atlantic cod, 54 whiting, and 63 Norway pout sampled from the inner Oslofjord, and 42 Atlantic cod, 56 whiting, and 41 Norway pout sampled from the outer Oslofjord. A more detailed table from the sampling can be found in Appendix A.

Table 1: Summary of the gutted weight measurements (g) (median, mean and range) in all three species in inner fjord (I) and outer fjord (O). Decimals rounded to nearest tenth.

Species (site)	Min (g)	1st Qu. (g)	Median (g)	Mean (g)	3 rd Qu. (g)	Max (g)
Cod (I)	96.8	231.1	296.0	334.3	452.9	745.5
Cod (O)	95.6	250.9	325.3	505.3	525.3	2600.0
Whiting (I)	50.3	117.8	151.1	171.7	221.9	398.0
Whiting (O)	25.2	82.5	119.9	136.7	172.8	346.0
Norway p. (I)	26.1	46.5	58.4	62.4	72.2	127.0
Norway p. (O)	13.0	26.70	31.3	31.0	34.4	49.5

Table 2: Summary of the fork length measurements (cm) (median, mean and range) in all three species at site A (inner fjord) and site B (outer fjord). Decimals rounded to nearest tenth.

Species (site)	Min (cm)	1st Qu. (cm)	Median (cm)	Mean (cm)	3 rd Qu. (cm)	Max (cm)
Cod (I)	22.5	30.5	33.5	33.7	37.3	46.0
Cod (O)	23.9	32.1	35.5	37.3	40.0	67.0
Whiting (I)	20.5	26.0	28.5	29.0	32.0	38.0
Whiting (O)	16.0	23.0	26.0	26.3	29.4	37.0
Norway p. (I)	16.0	19.5	20.5	20.8	22.0	26.0
Norway p. (O)	13.7	16.0	17.0	17.0	18.0	19.5

The fork length and gutted weight measurements were utilized to construct a length-weight regression of the relationship between log length and log gutted weight (Fig. 13).

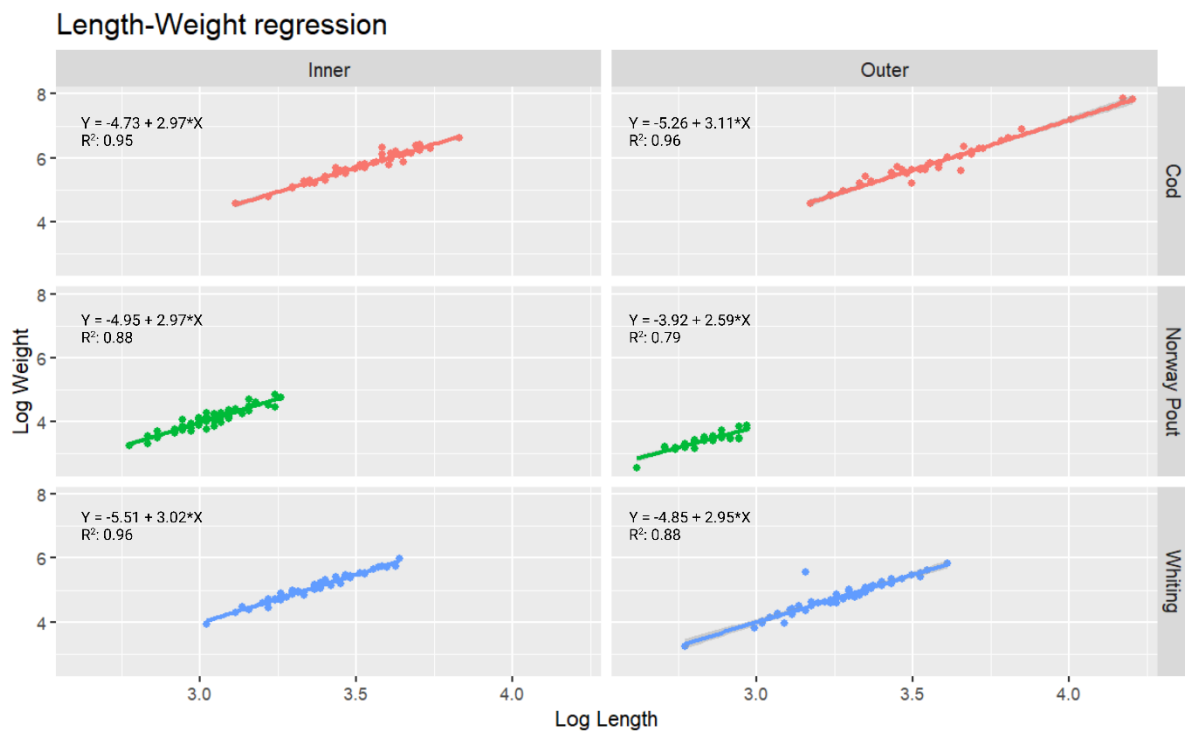


Figure 13: Regression of the relationship between log gutted weight and log length for Atlantic cod, Norway pout, and whiting in the inner and outer Oslofjord. Data for both genders in all four months are combined.

There were no significant differences in LWR of Atlantic cod ($p = 0.32$), Norway pout ($p = 0.13$) or whiting ($p = 0.66$) between the sites. Model diagnostics were checked by a visual inspection, and there were no serious issues.

3.2.2 LSI

A two-way ANOVA on log-transformed data revealed a significant difference in LSI for cod between inner and outer Oslofjord ($p = 0.01$), where the ones in the inner fjord had a higher LSI than those in the outer fjord (Fig. 14).

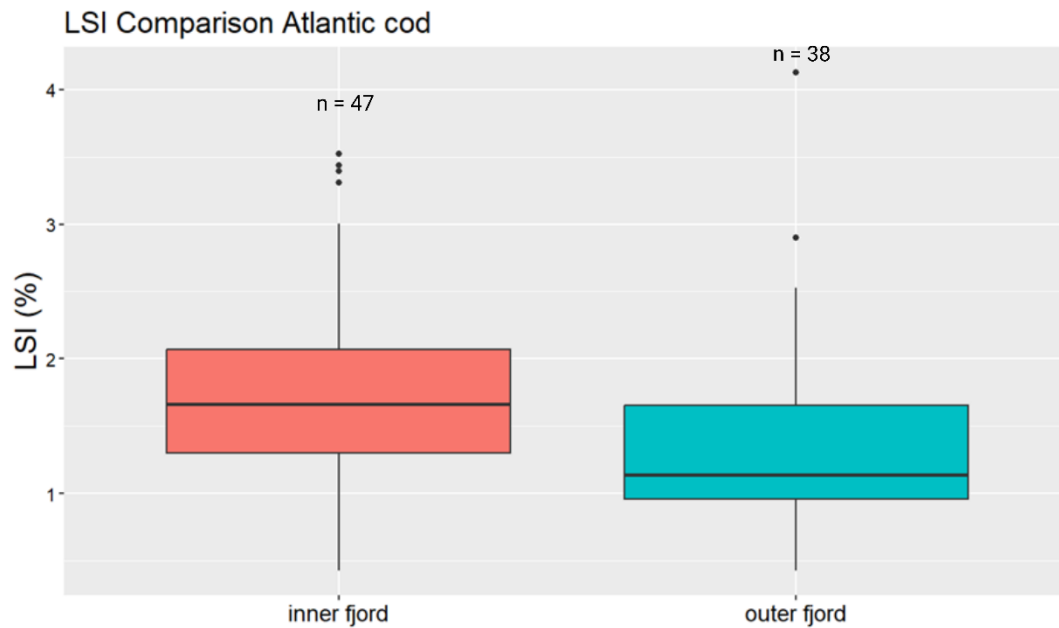


Figure 14: Observed LSI in Atlantic Cod between May 2021, September 2021, October 2021, and February 2022. The boxes represent the first and third quantiles, while the whiskers represent the maximum and minimum values. Median values are represented by the thick, horizontal, black lines. Dots represent outliers. N = sample size.

A two-way ANOVA on non-transformed data also revealed a significant difference in LSI for Norway pout ($p = 0.002$), where outer fjord had a higher LSI compared to the inner fjord (Fig 15).

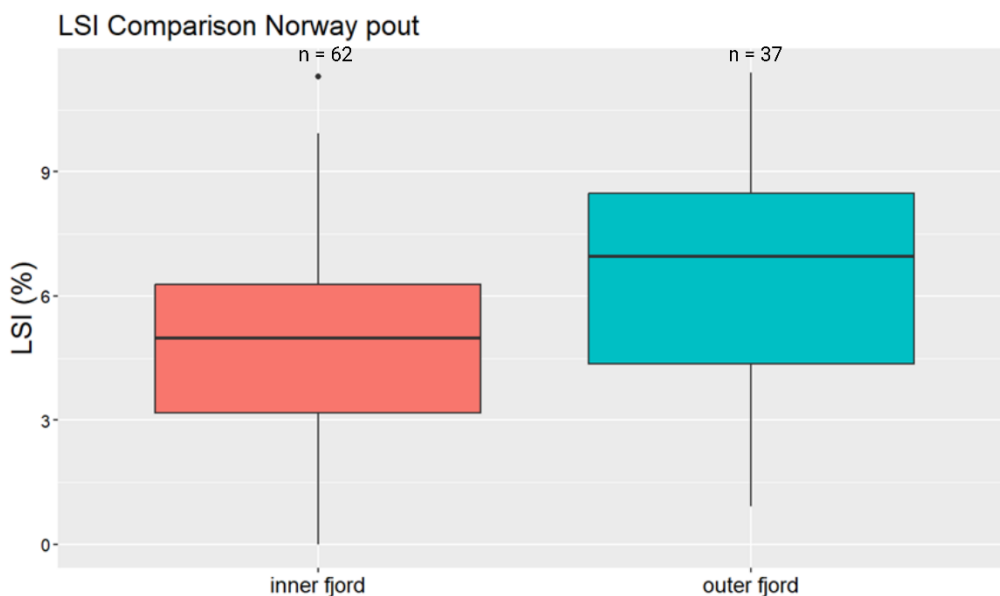


Figure 15: Observed LSI in Norway pout between May 2021, September 2021, October 2021, and February 2022. The boxes represent the first and third quantiles, while the whiskers represent the maximum and minimum values. Median values are represented by the thick, horizontal, black lines. Dots represent outliers. N = sample size.

There was no significant difference in LSI between whiting in the outer and inner Oslofjord according to a two-way ANOVA on non-transformed data ($p = 0.6$, Fig. 16).



Figure 16: Observed LSI in whiting between May 2021, September 2021, October 2021, and February 2022. The boxes represent the first and third quantiles, while the whiskers represent the maximum and minimum values. Median values are represented by the thick, horizontal, black lines. Dots represent outliers. N = sample size.

3.2.3 GSI

In February, there was a significantly higher GSI for female Norway pout in the outer fjord (Two-way ANOVA on non-transformed data, $p = 0.0008$). There was no significant difference between female whiting between the sites (Two-way ANOVA on log-transformed data, $p = 0.8$). Difference between males was not tested for neither whiting nor Norway pout due to the low sample size (Fig 17).

No statistical tests were performed on the difference in GSI for Atlantic cod between sites due to the large difference in sample size. Mature cod were almost exclusively sampled in February for both study areas.

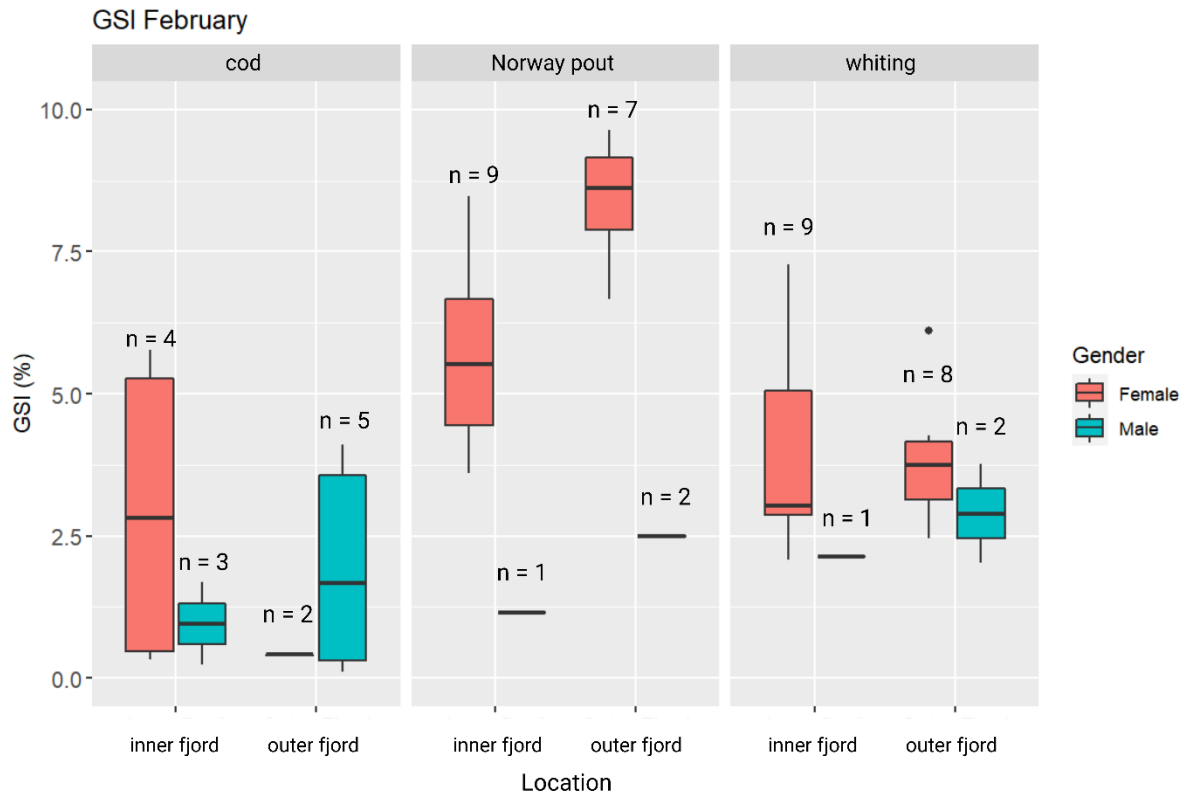


Figure 17: GSI for Atlantic cod, Norway pout and whiting in February 2022 in the inner and outer Oslofjord. N = sample size. The boxes represent the first and third quantiles, while the whiskers represent the maximum and minimum values. Median values are represented by the thick, horizontal, black lines. N = sample size.

Norway pout and whiting also spawns further into the spring, and thus it was of interest to analyze May as well (Fig. 18). It was not performed any statistical tests between female Norway pout in May due to the low sample size in the outer fjord. The Kruskal-Wallis test revealed no significant differences for female whiting between the inner and outer fjord ($p = 0.1$).

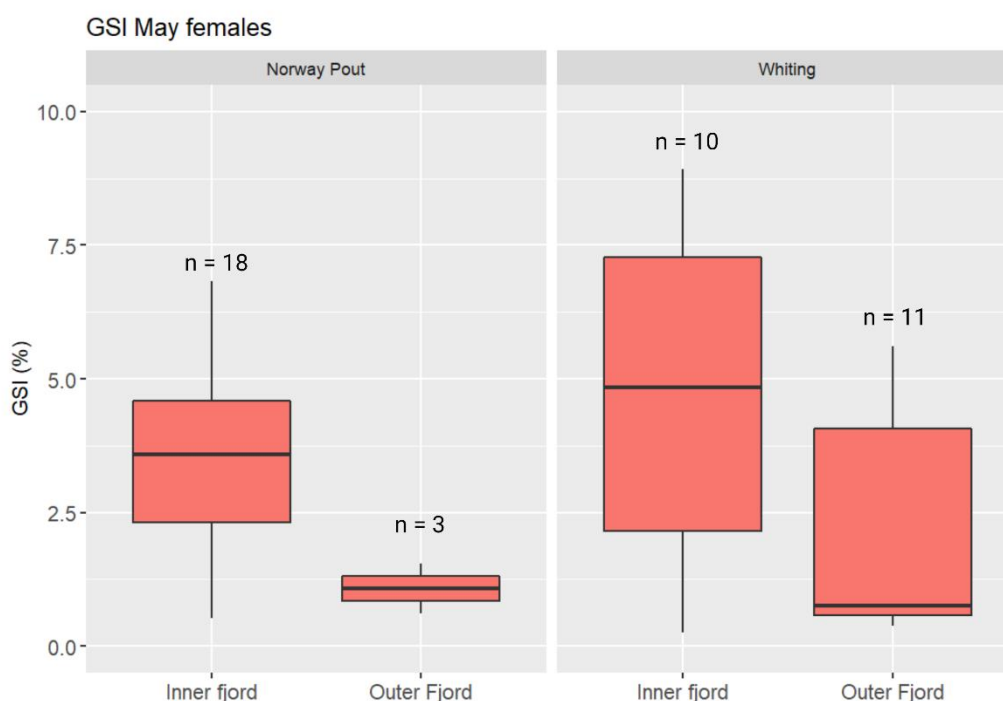


Figure 18: GSI in Norway pout and whiting females in May 2021 in the inner and outer Oslofjord. N = sample size. The boxes represent the first and third quartiles, while the whiskers represent the maximum and minimum values. Median values are represented by the thick, horizontal, black lines.

3.3 Otolith readings

3.3.1 Size-at-age

Size-at-age was obtained from back-calculation of growth based on cod otolith readings (Table 4).

Table 4: Size-at-age estimates based on back-calculation of otolith readings (median, mean and range). N = sample size.

inner fjord							
Age (years)	Min (cm)	1 st Qu.	Median (cm)	Mean (cm)	3 rd Qu.	Max (cm)	n
1	10.36	12.63	15.63	14.87	16.37	19.83	25
2	16.63	22.31	23.36	23.76	23.76	24.70	25
3	23.82	27.27	28.50	29.28	32.53	34.08	16
4	29.32	33.59	33.70	33.84	33.72	38.85	5
5	36.50	38.39	40.27	42.15	42.15	44.04	2
outer fjord							
Age (years)	Min (cm)	1 st Qu.	Median (cm)	Mean (cm)	3 rd Qu.	Max (cm)	n
1	10.78	12.31	14.26	14.26	15.88	18.97	27
2	15.94	19.62	23.03	23.11	25.42	36.56	27
3	21.22	27.73	29.90	29.45	31.16	36.52	20
4	31.64	34.38	36.19	37.18	39.28	45.11	7
5	47.00	47.00	47.00	47.00	47.00	47.00	1

A t-test revealed no significant differences in size-at-age-two between the regions, indicating equal growth of Atlantic cod between inner and outer Oslofjord.

3.3.2 Age-distribution

Determined age from the otolith readings of cod are presented in table 5.

Table 5: Age distribution from the age-determined cod otoliths.

Age	Inner fjord	Outer fjord
0	3	0
1	2	2
2	9	7
3	11	13
4	5	6
5	2	1

3.3.3 Age-at-maturity

Otolith data was used to determine the age-at-maturity parameter for the population dynamics model by comparing age estimates based on otoliths to GSI values. Since otolith data was only obtained for cod, these are the only size-at-age estimates that was compared to GSI data (Table 6).

Table 6: Size-at-age estimates and maturity from GSI data for cod in February. NA = indeterminate.

inner fjord			
Age (years)	Gender	Maturity	Length
NA	M	Mature	33.0cm
3	F	Mature	33.0cm
3	F	Juvenile	38.5cm
2	F	Mature	40.5cm
>2	F	Juvenile	29.0cm
>2	M	Juvenile	22.0cm
outer fjord			
Age (years)	Gender	Maturity	Length
2	M	Mature	23.9cm
NA	M	Mature	32.4cm
4	F	Juvenile	47.0cm
3	M	Mature	40.0cm
2	M	Juvenile	35.0cm
4	F	Juvenile	41.0cm
4	M	Juvenile	45.0cm

3.4 Modelling population dynamics

Median size-at-age estimates (Table 4) from the otolith readings were plotted together with a von Bertalanffy growth curve. The growth curve was adapted visually to fit the data by adjusting the growth parameter k (Fig. 19).

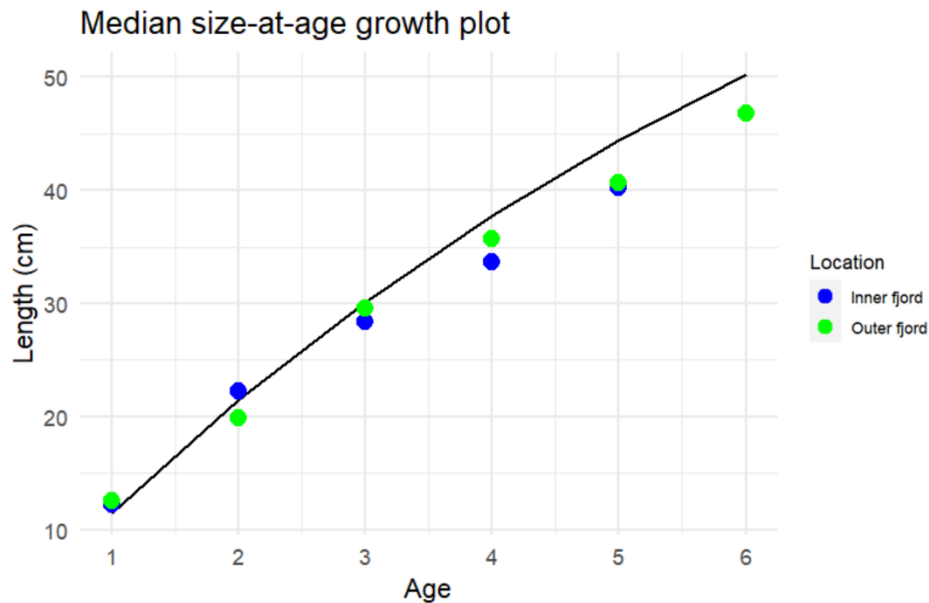


Figure 19: Projected growth of the Oslofjord cod population, assumed to follow a von Bertalanffy growth curve: $W_a = W_\infty (1 - \exp(-k(a - t_0)))^b$. The growth curve was visually adapted to median size-at-age data estimates from otolith readings, illustrated as green (outer fjord) and blue (inner fjord) dots.

Estimates of reduced population biomass impacts from 5%, 10%, and 15% reduction in growth was simulated in scenarios: lasting for one, five, and ten years (Fig. 20).

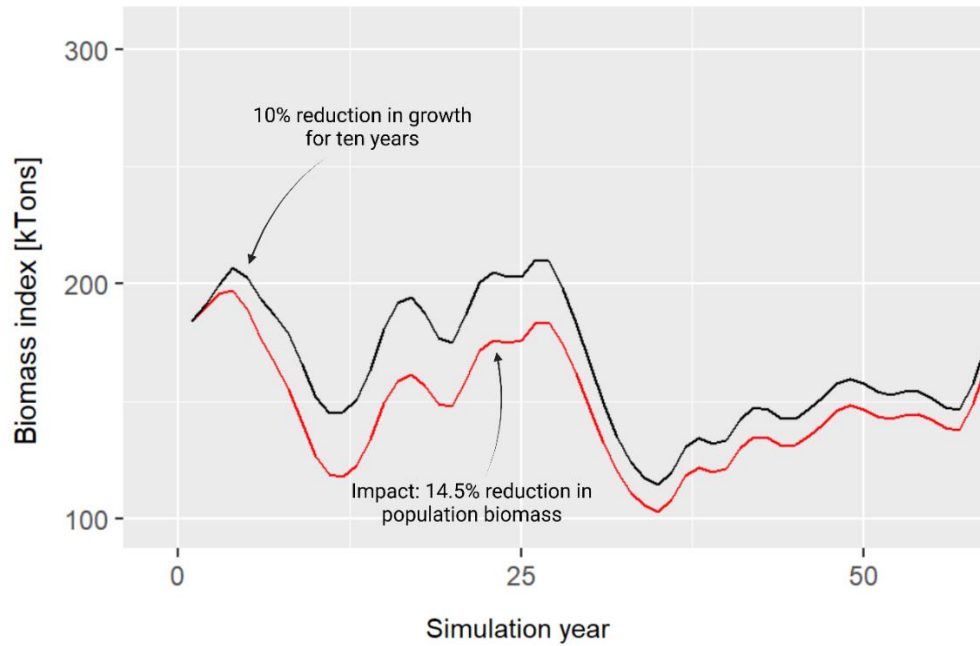


Figure 20: The population-level impact of a scenario of 10% reduction in growth lasting ten years. The decrease in population biomass was calculated as the relative difference between the perturbed biomass (red line) and the unperturbed biomass (black line) at the highest measured biomass ten years post-impact. In this example, the impact resulted in a 14.5% reduction in population biomass.

Each simulation was run approximately ten times, recording the impact for all simulations which was defined as the relative difference between the perturbed biomass and the unperturbed biomass at the highest measured biomass ten years post-impact. The impacts for each simulation, i.e. the reduction in population biomass, was fitted into impact distributions (Fig. 21).

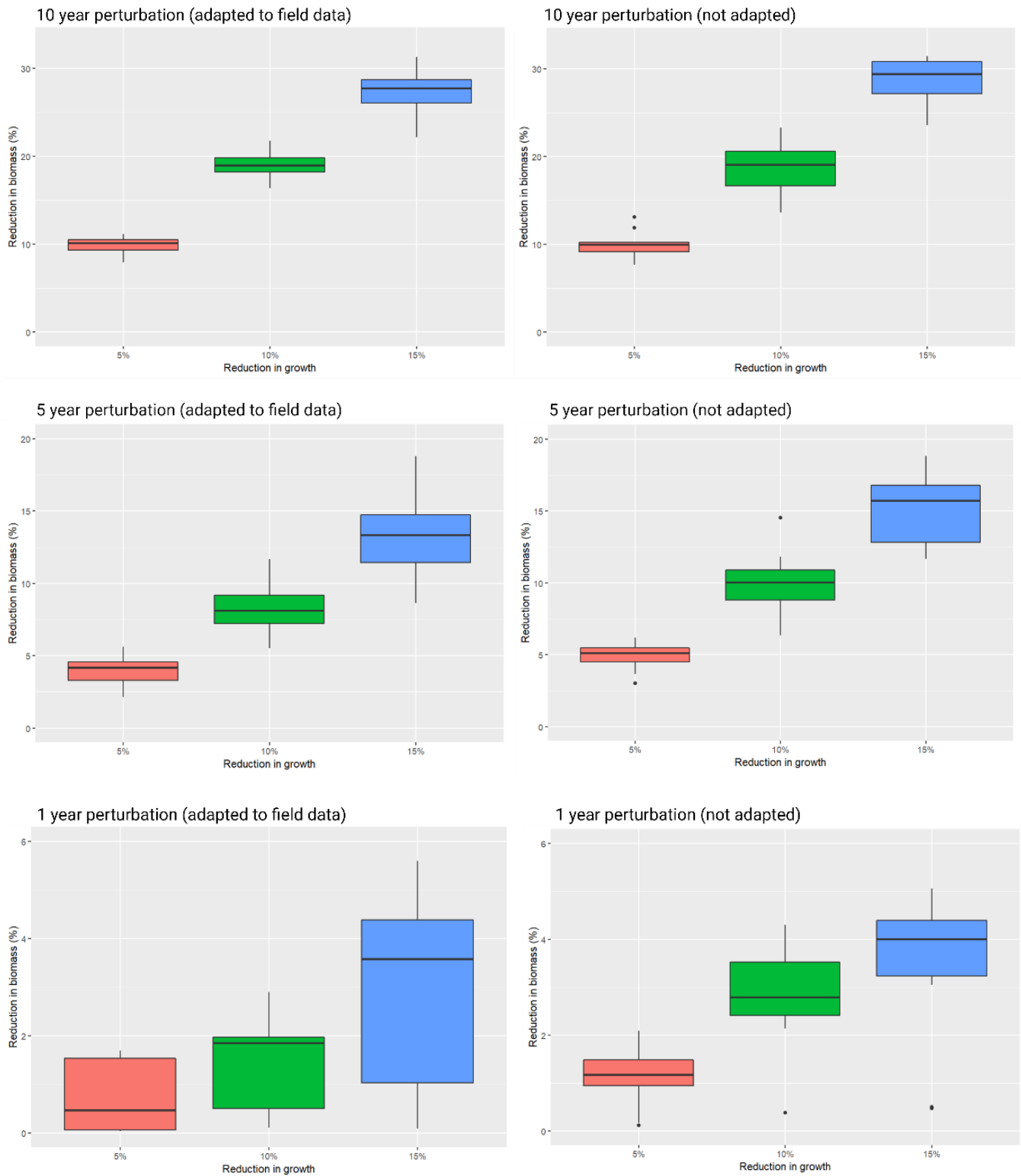


Figure 21: The impact distribution for scenarios with 5%, 10%, and 15% reduction in growth, lasting one, five, and ten years shown as boxplots. The boxplots shown on the left-hand panel are adapted to field-based data, while the ones on the right panel are not adapted.

Chapter 4: Discussion

The results and findings of this study will be interpreted and discussed in relation to each research question stated in Chapter 1 (section 1.3).

R1: Is contaminant exposure different for fish in the inner and outer Oslofjord?

Obtained concentrations of the PAH-metabolite pyrene from the bile samples revealed significantly higher concentrations in the inner fjord compared to the outer fjord (Fig. 10). This result is consistent with previous studies, which have also found significantly elevated concentrations of several PAH-metabolites in the inner Oslofjord compared to the outer Oslofjord (Imrik, 2010; Kristensen, 2022; Lundsør & Sundeng, 2018). PAH-metabolites in bile are indicators of recent exposure to PAH (van der Oost et al., 2003), and are considered as sensitive and reliable estimates (Beyer et al., 1997).

This study did not detect significant differences in pyrene concentration between months (Fig. 11) or between whiting and cod in the inner Oslofjord (Fig. 12), indicating that both species were recently exposed to similar concentrations of PAH close to capture. It should not be excluded that there could potentially be a difference in PAH-exposure between Norway pout and the other two species, i.e. whiting and cod, as this was not tested for. Since fish can be exposed to PAH via diet (Grung et al., 2009), the differences in diet between Norway pout and the two other species in this study could possibly affect PAH exposure (Froese & Pauly, 2000; Hislop et al., 1991).

PAH pollution usually involves a complex mixture of PAHs (Beyer et al., 2010). As this study only tests for pyrene, it does not account for other PAH-metabolites that could likely be present as well, according to previous studies in the fjord (Lundsør & Sundeng, 2018). Exposure to PAHs can have adverse effects on the biology of fish (Incardona et al., 2006; Reynaud & Deschaux, 2006; Rhodes et al., 2005; Sparling, 2016; Sørhus et al., 2015). This could, in turn, affect the condition factors as they are strongly affected by surpluses and deficits in the energy budget of the fish (Rose, 2019). Fish in the inner fjord are thus likely to be more affected by the adverse impacts of PAH exposure, e.g. reduced growth, endocrine suppression or metabolic effects (Donald, 2016), than fish in the outer fjord.

R2: Does the condition in fish differ between the inner and outer Oslofjord?

The obtained regression model for the length-weight relationship displayed no significant differences between the inner and outer fjord for any of the three species, indicating that the relationship between length and weight is similar between the two locations (Fig. 13). The allometric scaling coefficient (b) was around three for both cod and whiting, which indicates isometric growth (Froese, 2006). There is somewhat of a difference in the allometric scaling coefficient for Norway pout between the regions. The slight deviation from isometric growth for Norway pout in outer Oslofjord could possibly implicate that the weight is not increasing as fast as the length. A plausible factor is differences in diet, as it could potentially affect the LWR, since it can strongly influence the energy budget of the fish (Sherwood et al., 2007). However, this study did not analyze stomach contents to assess potential differences in diet between the locations. The LWR can also vary throughout the year, considering that there were only a few samples of Norway pout from the outer fjord in May. Compared to the other months, this could possibly be the cause of the variation.

There was a significant difference in LSI for both cod (Fig. 14) and Norway pout (Fig. 15) between the inner and outer Oslofjord. Whiting, on the other hand, displayed no significant difference in LSI between the sites, indicating similar condition (Fig. 16). While cod measured a significantly higher LSI for cod in the *inner* fjord, Norway pout measured a significantly higher LSI in the *outer* fjord. LSI reflects lipids stored in the liver (Yaragina & Marshall, 2000), and differences in LSI could be attributed to various factors. Increased LSI values have been linked to contaminant exposure, as more energy is allocated towards detoxification (Bernet et al., 2000; van der Oost et al., 2003), which could be possible as there was observed higher levels of contaminants in the inner fjord. However, as this trend was only observed for cod, there are likely other plausible reasons for the difference in LSI.

A lipid rich diet has been shown to increase lipid content and liver weight (Nanton et al., 2001), thus the differences in LSI might also indicate a more lipid rich diet at the respective sites with higher LSI values. In cod, a shrimp-based diet has been shown to decrease the LSI compared to a fish-based diet (Sherwood et al., 2007). Both body and liver weight is known to vary throughout the reproductive cycle (Dahle et al., 2003), which may also have affected the observed difference in LSI for both Norway pout and cod.

Norway pout measured a significantly higher GSI in the outer fjord in February (Fig. 17). This could indicate that Norway pout in inner Oslofjord invests more energy into spawning, which coincide with the elevated LSI observed in this area. There was not observed any significant differences for whiting. Both whiting and Norway pout in the inner fjord displayed a high GSI in May (Fig. 18). However, this study cannot conclude that there is a certain difference in GSI for Norway pout between the sites in this month due to the low sample size in the outer fjord. For whiting, on the other hand, the sample size was similar in the inner and outer fjord, which provides a good fundament for comparison between the sites in May; which revealed no significant difference.

Mature individuals of cod were almost exclusively sampled in February; it follows that this also was the month with the highest GSI. It is important to note that the sample size for cod in February was small, with only seven cod captured at each site (Appendix A). As there were not sampled any mature females in the outer fjord and only two mature females in the inner fjord this month, statistical comparisons between the sites were not considered meaningful and thus not performed. The sampling might not have been performed in the best months to determine the GSI for cod, as the onset of vitellogenesis is during autumn equinox (Kjesbu et al., 2010), and ends with spawning. Cod is also a batch spawner and spawns in batches every two to three days during the spawning period and lasts for weeks (Kjesbu, 1994); this enables the cod to spawn much more eggs than its size suggests (Fordham & Trippel, 1999). These mechanisms could, however, affect the GSI measurements in this study, making them less certain.

R3: Are there differences in growth between the inner and outer fjord?

Age at capture was determined from otolith readings (table 5). The oldest registered age was five years in both study areas. For both areas, the vast majority of the sampled cod were at the age of three and below, according to the otolith readings. This concluded an average age of 2.9 in the outer fjord ($n = 29$) and 2.6 in the inner fjord ($n = 32$) based on the age-determined otoliths. There was captured few one and zero-year-old individuals typically inhabits more shallow waters (Gjørøseter & Danielssen, 1990). The age distribution indicates that the Oslofjord cod population contains few old individuals. A strong bias towards younger individuals is not unique for the Oslofjord population, as this is a widespread phenomenon in most Atlantic cod stocks; consequently the recruitment in the population largely rely on first-

time spawners (Caddy & Agnew, 2004; Hutchings & Myers, 1993; Ottersen et al., 2006). Older and more fecund female fish can produce larger and more eggs that grow into larvae which grow faster and are more resilient towards starvation (Hixon et al., 2014). A possible shift in age structure could have occurred as a result of decades of overfishing (Barnett et al., 2017; Cook et al., 1997), this includes both commercial and recreational fishing (Kleiven et al., 2016). Shifts in age structure is a common response to overfishing, since it selectively removes the larger and older fish from the population (Ohlberger et al., 2022). This can negatively affect the productivity of the population since older fish have more reproductive importance (Rose, 2019).

Obtained size-at-age estimates from the otolith analysis showed no significant differences in size-at-age two between the inner and outer Oslofjord (table 4). There was large individual variation in the size-at-age estimates, which can likely be affected by individual and year-to-year variability. Nevertheless, size-at-age comparisons are useful indicators of changes in growth patterns over time within populations (Rose, 2019). Earlier investigations, from more than 70 years ago, of cod otoliths in the Oslofjord measured mean length for age one, two, three, and four year-olds to be 20, 33, 41 and 49 cm, respectively (Otterbech, 1954). The mean of the back-calculated lengths in this study to the corresponding age were 15, 24, 29, and 34 cm in the inner Oslofjord, and 14, 29, 33, and 37 cm in outer Oslofjord, which is consistently smaller than the mean length measurements from the Otterbechs study. The difference in growth between the present study and Otterbech is also much larger than the scenarios of 15% reduction in growth (Fig. 21). It should be noted that Otterbech compared the length at capture for individuals within the same age group, which were sampled during both spring and fall. In contrast, size-at-age estimates in the present study compares the individuals at a specific point in time, i.e. their birthday. Nevertheless, this could potentially indicate that the length of Atlantic cod in the Oslofjord has decreased over time. Size-at-age has been found to decrease in response to overfishing, which has been a long-lasting problem in the Oslofjord, due to selective removal of larger individuals (Neuheimer & Taggart, 2010). Overfishing has also been associated with selection against earlier maturation, as late-maturation increases the risk of being fished before spawning (Heino & Godø, 2002).

Another factor which could potentially affect the size-at-age is temperature (Rogers et al., 2011; Suthers & Sundby, 1996). Warmer summer temperatures have been shown to limit

juvenile growth, while warm spring temperatures have been linked to increased juvenile growth (Rogers et al., 2011). Smaller size-at-age could also be caused by the temperature-size rule. As the average body size of ectotherms have been found to decrease in response to increasing temperatures (Bauon et al., 2014; Ohlberger, 2013). This study did not conduct control measurements for temperature. However, measurements from the Institute of Marine Research have shown clear indications of increasing temperatures throughout the water column in the Oslofjord the past thirty years (Arvnes et al., 2019).

GSI was also compared to otolith data to determine age at maturity (table 6). Since it was a low sample size in February, with four mature individuals that was determined based on otoliths. Two of them were two years old, while the other two were three years old. Both estimates coincide with what has been found previously (Olsen et al., 2004). As these individuals were sampled in February, the two-year-old was on the verge of turning three since the annulus stops forming after winter. It was thus decided to round the age-at-maturity up to three for the population dynamics model.

There were some individuals who stood out from the rest in the outer Oslofjord as they were larger in size relative to their age and was not mature, according to the GSI. Three of the sampled individuals were above the age of four and had a length that ranged between 40-47cm. These individuals could be North Sea cod, but in order to accurately determine this a DNA analysis should be performed. Genetic differences are important factors in determining growth. It has been found that the size-at-age for the North Sea ecotype was considerably larger and could have faster individual growth than the fjord ecotype (Knutsen et al., 2018).

R4: How would effects on growth translate to the population level?

The median size-at-age estimates showed no significant differences in the growth of Atlantic cod between inner and outer Oslofjord (Fig. 19). The model was adapted by adjusting the growth rate parameter k from the original model until the visual fit was obtained (Section 2.6.5). The model could have been adapted with more formal methods c.f. (Nater et al., 2018), but a visual approach was used for simplicity due to time constraints.

Reduction in growth could potentially be caused by several factors, with the most important external drivers of variation in growth rate being temperature and food availability (Olsen et al., 2011; Suthers & Sundby, 1996). Additionally, factors such as contaminant exposure and

low oxygen concentration, have also been shown to affect growth rates (Chabot & Dutil, 1999; Sparling, 2016). All of these potential factors being highly relevant for the Oslofjord, with regards to both the current state of the fjord and its cod population. This study aimed to reflect how much or little growth can be reduced before noticing population-level effects through the simulated reduced growth scenarios. There seems to be some effect of reduction in growth at the population level in this study, particularly for the ten-year perturbation which had the estimated impact of up to 30% reduction in population biomass (Fig. 21). However, these perturbations can be hard to detect in a biomass time series, but easy to see based on the individual level. Perturbations with 10% and 15% reduced growth scenario for the ten-year impact scenario showed great negative effects on the estimated population biomass (Fig. 21). This is because the weight, i.e. the allometric scaling coefficient, is raised to the power of three (parameter b , Eq. 9).

The five-year scenario showed some effect for the 10% and 15% reduced growth scenarios as well, with an impact of up to 10% and 15% reduction in population biomass, respectively. For the one-year scenario, there was a low effect for all three reductions in growth, with the maximum impact on population biomass being around 5%. Thus, these estimates indicate that there needs to be above 10% reduction for five years in order to have an effect.

The model in the present study assumes equal growth for all individuals, and does not account for individual heterogeneity. The obtained size-at-age estimates showed great variation within age groups (Table 4), which is something that could have implications on population growth. For further improvements of the study, one could apply a model which allows for differences in individual growth (Vindenes & Langangen, 2015).

4.5 Methods

The catch varied between each cruise which resulted in differences in sample size particularly between months, therefore precaution regarding the representativeness should be taken when comparing and interpreting some of the results. All data was collected by bottom trawling with the same gear, at similar depths (>90m), and trawling speed, i.e., catchability was about the same for each cruise. The duration of each trawl varied to some extent (± 10 minutes), as it was prioritized to preserve the condition of the fish. One trawl was performed at each cruise, with the exception of the outer fjord in May 2021, when two trawls were performed. This was because no Norway pout was caught in the initial trawl. An additional

trawling was therefore performed. Even though this introduced sampling bias, particularly since the second trawl also yielded more cod and whiting which is consequently overestimated for this month, it was prioritized in order to ultimately secure enough samples for meaningful comparisons between the inner and outer fjord.

The amount of sampled Norway pout and whiting also varied between cruises due to time constraints (usually between 10-20 individuals, Appendix A). All captured cod were consistently sampled from each trawl at each site. This was done, again, in order to secure enough samples for meaningful comparisons between the regions, as the cod population has been declining the last decades (Espeland & Knutsen, 2019). The catch of cod was highly variable compared to Norway pout and whiting. The variability in sample size across months could reduce the statistical power of the comparisons, and also cause increased variability which might not be representative for the population.

Some of the fish were not dissected right away, which had implications for bile extraction as the gall bladder was typically empty the following day. This could also have had implications on the weight measurements, due to the loss of fluids. This issue was a trade-off between securing as much samples as possible and time constraints. Extracting bile from Norway pout was particularly difficult due to the small size of the gall bladder. Often, the entire gall bladder was extracted due to the high difficulty of extracting bile. Additionally, the majority of the bile was gone in Norway pout as sampling occurred right after breakfast hours. Consequently, the comparison in pyrene metabolites was primarily based on samples from whiting and cod.

Initially, this study intended to include otolith readings for all three species. During the course of the analysis, the whiting otoliths turned out to be challenging to reliably determine, something other studies have experienced as well (Ross & Hüsey, 2013). Due to the lack of experience with otolith readings and the risk of unreliable or inaccurate estimations, the whiting otoliths were hence discarded. Following the challenges with the whiting otoliths, efforts had to be prioritized. Otolith reading is a time consuming technique, and the decision was made to prioritize cod otoliths for this study. However, it is important to note that the exclusion of Norway pout and whiting otoliths does not diminish the possible insights these

otoliths could have provided in terms of assessing potential differences in growth between the regions.

The technique of back-calculation is largely based on subjectivity (Buckmeier, 2002). To ensure consistency, all otoliths were determined two times. Prior to the analysis it was established that the translucent zone needed to be complete before being defined as a full year. Inspection of the images pre-analysis revealed the completion of the translucent zone to occur somewhere between February and May. All annuli were marked at the edge of the completed translucent zone. Some otoliths had unclear or diffuse age rings, which could contribute to potential bias in the back-calculation and overestimation of the age, as it could look like there were more annuli than it was. For older fish, it is also easy to underestimate the age of the fish if the core of the otolith is missed. However, the determined otoliths were quite young, so this is likely not an issue for this analysis. This analysis did not include individuals below the age of two, nor did it include samples of too low quality as they could not be estimated with sufficient accuracy. The reason why individuals below the age of two was not included was to reduce potential variation due to early-life variability.

Other than the limitations discussed throughout this chapter, a major limitation of this study is that there was no control data on other factors which may also affect growth and condition, e.g. temperature, oxygen saturation, salinity and diet.

4.6 Future outlook

First of all, the inclusion of time-series data for future research could provide valuable insights into this subject. Time-series data could increase the robustness and reliability of the data, as it enables long-term analysis of patterns and potential changes within the population. It would be particularly interesting to focus more on Norway pout, as there was observed some interesting indications of differences between the regions. Additionally, analysis of stomach contents, e.g. by using stable isotopes, could possibly provide further knowledge about whether diet could be the cause of these differences.

Chapter 5: Conclusions

R1: Contaminant exposure did indeed differ significantly between the inner and outer Oslofjord. This study provides evidence that the inner fjord was more exposed to the PAH-metabolite pyrene, which served as a proxy for general contaminant exposure.

R2: There was varying results in terms of comparing the condition indices between the sites. While there were no significant differences in the Length-weight relationships between the sites, there were significant differences in the LSI for Atlantic cod and Norway pout between the sites: which could indicate better condition for cod in the inner Oslofjord, and better condition for Norway pout in the outer fjord.

R3: There was no significant differences in growth between the inner and outer Oslofjord, indicating that growth is similar between the sites.

R4: Reduced growth scenarios can indeed have some population-level effects, however these effects are hard to identify in time-series data.

References

- Arvnes, M. P., Albretsen, J., Naustvoll, L., Falkenhaus, T., Heiberg, S. E., Bjørge, A., Eikrem, W., Walday, M., Gitmark, J. K., Borgersen, G., Ruus, A., Green, N., & Hansen, S. A. (2019). *Kunnskapsstatus Oslofjorden* [Oppdragsrapport](1036). <https://www.miljodirektoratet.no/globalassets/publikasjoner/m1556/m1556.pdf>
- Barnett, L. A. K., Branch, T. A., Ranasinghe, R. A., & Essington, T. E. (2017). Old-Growth Fishes Become Scarce under Fishing. *Curr Biol*, 27(18). <https://doi.org/10.1016/j.cub.2017.07.069>
- Bauon, A., Needle, C., Rijnsdorp, A. D., & Marshall, C. T. (2014). Warming temperatures and smaller body sizes: synchronous changes in growth of North Sea fishes. *Global change biology*, 20(4), 1023-1031.
- Bernet, D., Schmidt-Posthaus, H., Wahli, T., & Burkhardt-Holm, P. (2000). Effects of wastewater on fish health: An integrated approach to biomarker responses in brown trout (*Salmo trutta* L.). *Journal of Aquatic Ecosystem Stress and Recovery*, 8(2), 143-151. <https://doi.org/10.1023/a:1011481632510>
- Beyer, J., Jonsson, G., Porte, C., Krahn, M. M., & Ariese, F. (2010). Analytical methods for determining metabolites of polycyclic aromatic hydrocarbon (PAH) pollutants in fish bile: A review. *Environ Toxicol Pharmacol*, 30(3), 224-244. <https://doi.org/10.1016/j.etap.2010.08.004>
- Beyer, J., Sandvik, M., Skåre, J. U., Egaas, E., Hylland, K., Waagbø, R., & Goksøyr, A. (1997). Time- and dose-dependent biomarker responses in flounder (*Platichthys flesus* L.) exposed to benzo[a]pyrene, 2,3,3',4,4', 5-hexachlorobiphenyl (PCB-156) and cadmium. *Biomarkers*, 2(1), 35-44. <https://doi.org/10.1080/135475097231959>
- Bigné, M., Nielsen, J. R., & Bastardie, F. (2019). Opening of the Norway pout box: will it change the ecological impacts of the North Sea Norway pout fishery? *ICES journal of marine science*, 76(1), 136-152. <https://doi.org/10.1093/icesjms/fsy121>
- Bose, A. P. H., McCallum, E. S., Raymond, K., Marentette, J. R., & Balshine, S. (2018). Growth and otolith morphology vary with alternative reproductive tactics and contaminant exposure in the round goby *Neogobius melanostomus*. *J Fish Biol*, 93(4), 674-684. <https://doi.org/10.1111/jfb.13756>
- Buckmeier, D. L. (2002). Assessment of reader accuracy and recommendations to reduce subjectivity in age estimation. *Fisheries (Bethesda)*, 27(11), 10-14. [https://doi.org/https://doi.org/10.1577/1548-8446\(2002\)027<0010:AORAAR>2.0.CO;2](https://doi.org/https://doi.org/10.1577/1548-8446(2002)027<0010:AORAAR>2.0.CO;2)
- Caddy, J. F., & Agnew, D. J. (2004). An overview of recent global experience with recovery plans for depleted marine resources and suggested guidelines for recovery planning. *Reviews in fish biology and fisheries*, 14(1), 43-112. <https://doi.org/10.1007/s11160-004-3770-2>
- Carroll, J., Vikebø, F., Howell, D., Broch, O. J., Nepstad, R., Augustine, S., Skeie, G. M., Bast, R., & Juselius, J. (2018). Assessing impacts of simulated oil spills on the Northeast Arctic cod fishery. *Mar Pollut Bull*, 126, 63-73. <https://doi.org/10.1016/j.marpolbul.2017.10.069>
- Chabot, D., & Dutil, J. D. (1999). Reduced growth of Atlantic cod in non-lethal hypoxic conditions. *Journal of fish biology*, 55(3), 472-491. <https://doi.org/10.1006/jfbi.1999.1005>
- Cohen, D. M., Inada, T., Iwamoto, T., & Scialabba, N. (1990). FAO species catalogue. In Vol. 10. *Gadiform fishes of the world (Order Gadiformes). An annotated and illustrated catalogue of cods, hakes, grenadiers and other gadiform fishes known to date*. Rome: FAO.
- Cook, R. M., Kunzlik, P. A., Hislop, J. R. G., & Poulding, D. (1999). Models of growth and maturity for North Sea cod. *Journal of Northwest Atlantic fishery science*, 25, 91-99. <https://doi.org/10.2960/J.v25.a8>
- Cook, R. M., Sinclair, A., & Stefánsson, G. (1997). Potential collapse of North Sea cod stocks. *Nature (London)*, 385(6616), 521-522. <https://doi.org/10.1038/385521a0>
- Dahle, R., Taranger, G. L., Karlsen, Ø., Kjesbu, O. S., & Norberg, B. (2003). Gonadal development and associated changes in liver size and sexual steroids during the reproductive cycle of captive male and female Atlantic cod (*Gadus morhua* L.). *Comp Biochem Physiol A Mol Integr Physiol*, 136(3), 641-653. [https://doi.org/10.1016/S1095-6433\(03\)00215-0](https://doi.org/10.1016/S1095-6433(03)00215-0)

- Donald, W. S. (2016). Chapter 7 - Polycyclic Aromatic Hydrocarbons. In (pp. 193-223). Elsevier Inc. <https://doi.org/10.1016/B978-0-12-801947-4.00007-X>
- Daan, N. (1973). A quantitative analysis of the food intake of North Sea cod, *Gadus Morhua*. *Netherlands journal of sea research*, 6(4), 479-517. [https://doi.org/10.1016/0077-7579\(73\)90002-1](https://doi.org/10.1016/0077-7579(73)90002-1)
- Espeland, S. H., & Knutsen, H. (2019). *Rapport fra høstundersøkelsene med strandnot i indre og ytre Oslofjord 2018*. <http://hdl.handle.net/11250/2598641>
- Fablet, R., Pecquerie, L., de Pontual, H., Høie, H., Millner, R., Mosegaard, H., & Kooijman, S. A. L. M. (2011). Shedding light on fish otolith biomineralization using a bioenergetic approach. *PLoS One*, 6(11). <https://doi.org/10.1371/journal.pone.0027055>
- Folkvord, A., Johannessen, A., & Moksness, E. (2004). Temperature-dependent otolith growth in Norwegian spring-spawning herring (*Clupea harengus* L.) larvae. *Sarsia*, 89(5), 297-310. <https://doi.org/10.1080/00364820410002532>
- Folkvord, A., & Mosegaard, H. (2002). Chapter V - Some uses of individual age data. A. Age and growth analysis. In H. d. P. J. Panfili, H. Troadec, P. J. Wright. (Ed.), *Manual of fish sclerochronology* (pp. 146-166). Ifremer-IRD Co-edition, Brest, France.
- Fordham, B. S. E., & Trippel, E. A. (1999). Feeding behaviour of cod (*Gadus morhua*) in relation to spawning. *Journal of applied ichthyology*, 15(1), 1-9. <https://doi.org/10.1046/j.1439-0426.1999.00098.x>
- Francis, R. I. C. C. (1990). Back-calculation of fish length: a critical review. *Journal of fish biology*, 36(6), 883-902. <https://doi.org/10.1111/j.1095-8649.1990.tb05636.x>
- Froese, R. (2006). Cube law, condition factor and weight-length relationships: history, meta-analysis and recommendations. *Journal of applied ichthyology*, 22(4), 241-253. <https://doi.org/10.1111/j.1439-0426.2006.00805.x>
- Froese, R., & Pauly, D. (2000). *Fishbase 2000: Concepts, designs & data source*.
- Gade, H. G. (1973). Deep Water Exchanges in a Sill Fjord: A Stochastic Process. *Journal of physical oceanography*, 3(2), 213-219. [https://doi.org/https://doi.org/10.1175/1520-0485\(1973\)003%3C0213:DWEIAS%3E2.0.CO;2](https://doi.org/https://doi.org/10.1175/1520-0485(1973)003%3C0213:DWEIAS%3E2.0.CO;2)
- Gjøsaeter, J., & Danielssen, D. S. (1990). *Recruitment of cod (Gadus morhua), whiting (Merlangius merlangus) and pollack (Pollachius pollachius) in the Risør area on the Norwegian Skagerrak coast 1945 to 1985*. In: *Flødevigen rapportserie no 1, 1990* (0333-2594). <http://hdl.handle.net/11250/115166>
- Grung, M., Holth, T. F., Jacobsen, M. R., & Hylland, K. (2009). Polycyclic Aromatic Hydrocarbon (PAH) Metabolites in Atlantic Cod Exposed via Water or Diet to a Synthetic Produced Water. *J Toxicol Environ Health A*, 72(3-4), 254-265. <https://doi.org/10.1080/15287390802539210>
- Grung, M., Jartun, M., Bæk, K., Ruus, A., Rundberget, T., Allan, I., Beylich, B., Vogelsang, C., Schlabach, M., Hanssen, L., Borgå, K., & Helberg, M. (2021). Environmental Contaminants in an Urban Fjord, 2020. In: *Norsk institutt for vannforskning*.
- Heino, M., & Godø, O. R. (2002). Fisheries-Induced Selection Pressures in the Context of Sustainable Fisheries. *Bulletin of marine science*, 70(2), 639-656.
- Hislop, J. R. G., & Hall, W. B. (1974). The fecundity of whiting, *Merlangius merlangus* (L.) in the North Sea, the Minch and at Iceland. *ICES journal of marine science*, 36(1), 42-49. <https://doi.org/10.1093/icesjms/36.1.42>
- Hislop, J. R. G., Robb, A. P., Bell, M. A., & Armstrong, D. W. (1991). The diet and food consumption of whiting (*Merlangius merlangus*) in the North Sea. *ICES journal of marine science*, 48(2), 139-156. <https://doi.org/10.1093/icesjms/48.2.139>
- Hixon, M. A., Johnson, D. W., & Sogard, S. M. (2014). BOFFFFs: On the importance of conserving old-growth age structure in fishery populations. *ICES journal of marine science*, 71(8), 2171-2185. <https://doi.org/10.1093/icesjms/fst200>
- Hoenig, J. M. (1983). Empirical use of longevity data to estimate mortality rates. *Fishery Bulletin*, 82.
- Hoie, H., Geffen, A. J., Heegaard, E., Skadal, J., Folkvord, A., & Li, L. (2008). Back-calculation of previous fish size using individually tagged and marked Atlantic cod (*Gadus morhua*). *Journal*

- canadien des sciences halieutiques et aquatiques*, 65(11), 2496-2508.
<https://doi.org/10.1139/F08-157>
- Hutchings, J. A., & Myers, R. A. (1993). Effect of Age on the Seasonality of Maturation and Spawning of Atlantic Cod, *Gadus morhua*, in the Northwest Atlantic. *Canadian journal of fisheries and aquatic sciences*, 50(11), 2468–2474. <https://doi.org/10.1139/f93-271>
- Hylland, K. (2006). Polycyclic Aromatic Hydrocarbon (PAH) Ecotoxicology in Marine Ecosystems. *J Toxicol Environ Health A*, 69(1-2), 109-123. <https://doi.org/10.1080/15287390500259327>
- Imrik, E. C. R. (2010). *Effekter av miljøgifter på torsk fra Indre Oslofjord*. Master Thesis, University of Oslo.
- Incardona, J. P., Day, H. L., Collier, T. K., & Scholz, N. L. (2006). Developmental toxicity of 4-ring polycyclic aromatic hydrocarbons in zebrafish is differentially dependent on AH receptor isoforms and hepatic cytochrome P4501A metabolism. *Toxicol Appl Pharmacol*, 217(3), 308-321. <https://doi.org/10.1016/j.taap.2006.09.018>
- Kjesbu, O. S. (1994). Time of start of spawning in Atlantic cod (*Gadus morhua*) females in relation to vitellogenic oocyte diameter, temperature, fish length and condition. *Journal of fish biology*, 45(5), 719-735. <https://doi.org/10.1111/j.1095-8649.1994.tb00939.x>
- Kjesbu, O. S., Righton, D., Kruger-Johnsen, M., Thorsen, A., Michalsen, K., Fonn, M., & Witthames, P. R. (2010). Thermal dynamics of ovarian maturation in Atlantic cod (*Gadus morhua*). *Journal canadien des sciences halieutiques et aquatiques*, 67(4), 605-625.
<https://doi.org/10.1139/F10-011>
- Kleiven, A. R., Fernandez-Chacon, A., Nordahl, J.-H., Moland, E., Espeland, S. H., Knutsen, H., & Olsen, E. M. (2016). Harvest pressure on coastal atlantic cod (*Gadus morhua*) from recreational fishing relative to commercial fishing assessed from tag-recovery data. *PLoS One*, 11(3).
<https://doi.org/10.1371/journal.pone.0149595>
- Knutsen, H., Jorde, P. E., Hutchings, J. A., Hemmer-Hansen, J., Grønkjær, P., Jørgensen, K. M., André, C., Sodeland, M., Albretsen, J., & Olsen, E. M. (2018). Stable coexistence of genetically divergent Atlantic cod ecotypes at multiple spatial scales. *Evolutionary Applications*, 11(9), 1527-1539. <https://doi.org/10.1111/eva.12640>
- Kristensen, S. H. (2022). *Species differences in sensitivity to environmental pollutants in Gadidae in the Oslofjord*. Master Thesis, University of Oslo.
- Kruskal, W. H., & Wallis, W. A. (1952). Use of Ranks in One-Criterion Variance Analysis. *Journal of the American Statistical Association*, 47(260), 583-621.
<https://doi.org/10.1080/01621459.1952.10483441>
- Langangen, Ø., Ohlberger, J., Stige, L. C., Patin, R., Buttay, L., Stenseth, N. C., Ono, K., & Durant, J. M. (2023). Effects of early life mass mortality events on fish populations. *Fish and fisheries (Oxford, England)*, 24(1), 176-186. <https://doi.org/10.1111/faf.12718>
- Levene, H. (1952). On the Power Function of Tests of Randomness Based on Runs up and Down. *The Annals of mathematical statistics*, 23(1), 34-56. <https://doi.org/10.1214/aoms/1177729484>
- Lima, A. L. C., Farrington, J. W., & Reddy, C. M. (2005). Combustion-Derived Polycyclic Aromatic Hydrocarbons in the Environment-A Review. *Environmental forensics*, 6(2), 109-131.
<https://doi.org/10.1080/15275920590952739>
- Lotze, H. K., Lenihan, H. S., Bourque, B. J., Bradbury, R. H., Cooke, R. G., Kay, M. C., Kidwell, S. M., Kirby, M. X., Peterson, C. H., & Jackson, J. B. C. (2006). Depletion, Degradation, and Recovery Potential of Estuaries and Coastal Seas. *Science (American Association for the Advancement of Science)*, 312(5781), 1806-1809. <https://doi.org/10.1126/science.1128035>
- Lundsør, E., J. K. Dolven, G. L. Haugestøl, P. Bechmann, G. R. Salomonsen, K., & Sundeng, H. G., O. al-Khayat, T. Kornstad. (2018). *Overvåkning av Indre Oslofjord 2017*.
- Midway, S. (2014, 12.04.2023). Through the ears of a fish.
<https://thefisheriesblog.com/2014/01/13/through-the-ears-of-a-fish/>
- Miljødirektoratet. (2022, 21.11.2022). *Oslofjorden: Tiltaksplan for bedre miljøtilstand*. Retrieved 09.04.2023 from <https://www.miljodirektoratet.no/ansvarsomrader/vann-hav-og-kyst/vann-hav-kyst-forvaltning/oslofjorden/>

- Moen, F. E., & Svensen, E. (2020). *Dyreliv i havet : norsk marin fauna* (7. utgave. ed.). Kolofon.
- Myers, R. A., Bridson, J., & Barrowman, N. J. (1995). Summary of world-wide Spawner and recruitment data. *Canadian technical report of fisheries and Aquatic sciences*, 2020, 1-327.
- Nanton, D. A., Lall, S. P., & McNiven, M. A. (2001). Effects of dietary lipid level on liver and muscle lipid deposition in juvenile haddock, *Melanogrammus aeglefinus* L: Lipid utilization by haddock. *Aquaculture research*, 32, 225-234. <https://doi.org/10.1046/j.1355-557x.2001.00019.x>
- Nater, C. R., Rustadbakken, A., Ergon, T., Langangen, Ø., Moe, S. J., Vindenes, Y., Vøllestad, L. A., & Aass, P. (2018). Individual heterogeneity and early life conditions shape growth in a freshwater top predator. *Ecology*, 99(5), 1011-1017. <https://doi.org/10.1002/ecy.2178>
- Neuheimer, A. B., & Taggart, C. T. (2010). Can changes in length-at-age and maturation timing in Scotian Shelf haddock (*Melanogrammus aeglefinus*) be explained by fishing? *Journal canadien des sciences halieutiques et aquatiques*, 67(5), 854-865. <https://doi.org/10.1139/F10-025>
- Nigrelli, R. F. (1959). Longevity of fishes in captivity, with special reference to those kept in the New York Aquarium. In G. E. W. Wolstenholme, O'Connor, M. (Ed.), *Ciba Foundation Colloquium on Ageing: The life span of animals* (Vol. 5, pp. 212-230). Churchill.
- NIVA. (2019). *Dypvannsfornyelse i Indre Oslofjord*. NIVA. Retrieved 04.04.2023 from <https://www.niva.no/nyheter/dypvannsfornyelse-i-indre-oslofjord>
- Ohlberger, J. (2013). Climate warming and ectotherm body size — from individual physiology to community ecology. *Functional ecology*, 27(4), 991-1001. <https://doi.org/10.1111/1365-2435.12098>
- Ohlberger, J., & Langangen, Ø. (2015). Population resilience to catastrophic mortality events during early life stages. *Ecol Appl*, 25(5), 1348-1356. <https://doi.org/10.1890/14-1534.1>
- Ohlberger, J., Langangen, Ø., & Stige, L. C. (2022). Age structure affects population productivity in an exploited fish species. *Ecol Appl*, 32(5). <https://doi.org/10.1002/eap.2614>
- Olsen, E. M., Knutsen, H., Gjøsæter, J., Jorde, P. E., Knutsen, J. A., & Stenseth, N. C. (2004). Life-history variation among local populations of Atlantic cod from the Norwegian Skagerrak coast: atlantic cod life-history variation. *Journal of fish biology*, 64(6), 1725-1730. <https://doi.org/10.1111/j.0022-1112.2004.00402.x>
- Olsen, E. M., Ottersen, G., Llope, M., Chan, K.-S., Beaugrand, G., & Stenseth, N. C. (2011). Spawning stock and recruitment in North Sea cod shaped by food and climate. *Proc. R. Soc. B*, 278(1705), 504-510. <https://doi.org/10.1098/rspb.2010.1465>
- Ono, K., Knutsen, H., Olsen, E. M., Ruus, A., Hjermann, D. Ø., & Stenseth, N. C. (2019). Possible adverse impact of contaminants on Atlantic cod population dynamics in coastal ecosystems. *Proc Biol Sci*, 286(1908), 1-9. <https://doi.org/10.1098/rspb.2019.1167>
- Oslo Havn KF. *Oslo Havn*. Retrieved 09.04.2023 from <https://www.oslohavn.no/no/meny/om-oslo-havn/om-oslo-havn-kf/>
- Oslo Havn KF. (2019). *Strategiplan for Oslo Havn KF 2019 - 2034*. <https://www.oslohavn.no/globalassets/oslo-havn/dokumenter/sentrale-planer/3153-rapport-strategiplan-2019-pdf.pdf>
- Otterbech, F. (1954). *Undersøkelser over torsken i Oslofjorden*. <http://hdl.handle.net/11250/114566>
- Ottersen, G., Hjermann, D. A. G. Ø., & Stenseth, N. C. (2006). Changes in spawning stock structure strengthen the link between climate and recruitment in a heavily fished cod (*Gadus morhua*) stock. *Fisheries oceanography*, 15(3), 230-243. <https://doi.org/10.1111/j.1365-2419.2006.00404.x>
- Pi, J., Li, X., Zhang, T., & Li, D. (2016). Effects of Acute Exposure to Sublethal Waterborne Cadmium on Energy Homeostasis in Silver Carp (*Hypophthalmichthys molitrix*). *Bull Environ Contam Toxicol*, 97(4), 497-503. <https://doi.org/10.1007/s00128-016-1896-2>
- Reynaud, S., & Deschaux, P. (2006). The effects of polycyclic aromatic hydrocarbons on the immune system of fish: A review. *Aquat Toxicol*, 77(2), 229-238. <https://doi.org/10.1016/j.aquatox.2005.10.018>

- Rhodes, S., Farwell, A., Mark Hewitt, L., MacKinnon, M., & George Dixon, D. (2005). The effects of dimethylated and alkylated polycyclic aromatic hydrocarbons on the embryonic development of the Japanese medaka. *Ecotoxicol Environ Saf*, 60(3), 247-258.
<https://doi.org/10.1016/j.ecoenv.2004.08.002>
- Ricker, W. E. (1954). Stock and recruitment. *Journal of the Fisheries Research Board of Canada*, 11(5), 559–623. <https://doi.org/https://doi.org/10.1139/f54-039>
- Rizzo, E., & Bazzoli, N. (2020). Biological Indices. In E. C. U. Bernardo Baldisserotto & J. E. P. Cyrino (Eds.), *Biology and Physiology of Freshwater Neotropical Fish*. Academic Press Inc.
<https://doi.org/https://doi.org/10.1016/C2017-0-03766-7>
- Rogers, L. A., Stige, L. C., Olsen, E. M., Knutsen, H., Chan, K.-S., & Stenseth, N. C. (2011). Climate and population density drive changes in cod body size throughout a century on the Norwegian coast. *Proc Natl Acad Sci U S A*, 108(5), 1961-1966.
<https://doi.org/10.1073/pnas.1010314108>
- Rose, G. A. (2019). What Happens to the Food Cod Eat: Individual Growth. In *Atlantic cod : the bio-ecology of the fish* (First edition. ed., pp. 237-246). Wiley Blackwell.
- Ross, S. D., & Hüsey, K. (2013). A reliable method for ageing of whiting (*Merlangius merlangus*) for use in stock assessment and management. *J. Appl. Ichthyol*, 29(4), 825-832.
<https://doi.org/10.1111/jai.12204>
- Ruus, A., Bæk, K., Rundberget, T., Allan, I., Beylich, B., Vogelsang, C., Schlabach, M. N., Götsch, A. N., Borgå, K. U., & Helberg, M. U. (2019). *Environmental Contaminants in an Urban Fjord, 2019*. Report M1131 from the Norwegian Environment Agency.).
<https://www.miljodirektoratet.no/globalassets/publikasjoner/m1766/m1766.pdf>
- Samanta, P., Im, H., Na, J., & Jung, J. (2018). Ecological risk assessment of a contaminated stream using multi-level integrated biomarker response in *Carassius auratus*. *Environ Pollut*, 233, 429-438. <https://doi.org/10.1016/j.envpol.2017.10.061>
- Shapiro, S. S., & Wilk, M. B. (1965). An Analysis of Variance Test for Normality (Complete Samples). *Biometrika*, 52(3/4), 591. <https://doi.org/10.2307/2333709>
- Sherwood, G. D., Rideout, R. M., Fudge, S. B., & Rose, G. A. (2007). Influence of diet on growth, condition and reproductive capacity in Newfoundland and Labrador cod (*Gadus morhua*): Insights from stable carbon isotopes ($\delta^{13}C$). *Deep-sea research. Part 2. Topical studies in oceanography*, 54(23), 2794-2809. <https://doi.org/10.1016/j.dsr2.2007.08.007>
- Sparling, D. W. (2016). Polycyclic Aromatic Hydrocarbons. In *Ecotoxicology Essentials : Environmental Contaminants and Their Biological Effects on Animals and Plants* (pp. 193-223).
<https://doi.org/10.1016/B978-0-12-801947-4.00007-X>
- Spromberg, J. A., & Birge, W. J. (2005). Modeling the effects of chronic toxicity on fish populations: The influence of life-history strategies. *Environmental Toxicology and Chemistry*, 24(6), 1532-1540. <https://doi.org/10.1897/04-160.1>
- Stigebrandt, A., Magnusson, J., & Magnusson, J. (2002). *Utredning av konsekvenser for vannutskiftningen i indre Oslofjord ved utvidelse av skipsleden over Drøbakerskelen*. NIVA.
<http://hdl.handle.net/11250/211615>
- Staalstrøm, A. (2020). *Modellering av miljøtilstanden i indre Oslofjord sett i lys av utslippstillatelser og befolkningsutvikling*. NIVA. <https://hdl.handle.net/11250/2653243>
- Staalstrøm, A., Engesmo, A., Andersen, G. S., Gran, S., Borgersen, G., Moy, S. R., Valestrand, L., Brooks, S., Hylland, K., & Holth, T. F. (2021). *Undersøkelse av hydrografiske og biologiske forhold i Indre Oslofjord Årsrapport 2020* (978-82-577-7386-1).
- Suthers, I. M., & Sundby, S. (1996). Role of the midnight sun: Comparative growth of pelagic juvenile cod (*Gadus morhua*) from the Arcto-Norwegian and a Nova Scotia stock. *ICES journal of marine science*, 53(5), 827-836. <https://doi.org/10.1006/jmsc.1996.0104>
- Sørhus, E., Edvardsen, R. B., Karlsen, Ø., Nordtug, T., van der Meeren, T., Thorsen, A., Harman, C., Jentoft, S., & Meier, S. (2015). Unexpected Interaction with Dispersed Crude Oil Droplets Drives Severe Toxicity in Atlantic Haddock Embryos.
<https://doi.org/https://doi.org/10.1371/journal.pone.0124376>

- van der Oost, R., Beyer, J., & Vermeulen, N. P. E. (2003). Fish bioaccumulation and biomarkers in environmental risk assessment: a review. *Environ Toxicol Pharmacol*, 13(2), 57-149. [https://doi.org/10.1016/S1382-6689\(02\)00126-6](https://doi.org/10.1016/S1382-6689(02)00126-6)
- Vindenes, Y., & Langangen, Ø. (2015). Individual heterogeneity in life histories and eco-evolutionary dynamics. *Ecol Lett*, 18(5), 417-432. <https://doi.org/10.1111/ele.12421>
- Yaragina, N. A., & Marshall, C. T. (2000). Trophic influences on interannual and seasonal variation in the liver condition index of Northeast Arctic cod (*Gadus morhua*). *ICES journal of marine science*, 57(1), 42-55. <https://doi.org/10.1006/jmsc.1999.0493>

Appendix A

Table 1: Number of sampled individuals from each species at location and date sorted by gender: Females (F), Males (M), or Indeterminate (NA).

Date & location	Atlantic Cod				Whiting				Norway Pout			
	F	M	NA	TOT	F	M	NA	TOT	F	M	NA	TOT
05.05.2021 outer fjord	4	1	0	5	6	5	0	11	0	0	0	0
06.05.2021 outer fjord	9	11	0	20	7	1	0	8	3	3	0	6
11.05.2021 inner fjord	11	13	1	25	10	6	1	17	18	3	3	24
31.08.2021 outer fjord	2	2	0	4	5	9	1	15	1	2	12	15
01.09.2021 inner fjord	5	3	0	8	5	9	0	14	7	8	0	15
14.10.2021 inner fjord	7	4	0	11	13	2	0	15	6	4	4	14
15.10.2021 outer fjord	4	2	0	6	7	3	0	10	8	1	1	10
08.02.2022 outer fjord	2	5	0	7	8	2	0	10	7	2	1	10
09.02.2022 inner fjord	4	3	0	7	9	1	0	10	9	1	0	10
Total IF	25	25	1	51	34	19	1	54	38	17	8	63
Total OF	23	19	0	42	36	19	1	56	21	7	13	41

Appendix B

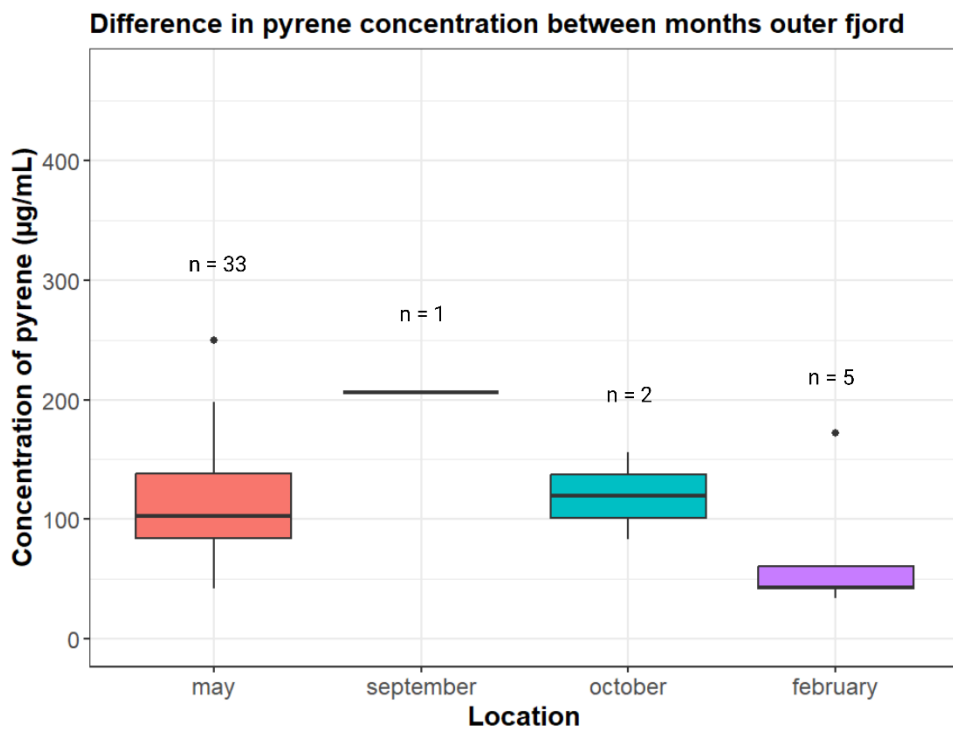


Figure 1: measured difference in pyrene concentration between months in outer Oslofjord. The boxes represent the first and third quantiles, while the whiskers represent the maximum and minimum values. The thick, horizontal, and black lines represent median values. N = sample size.

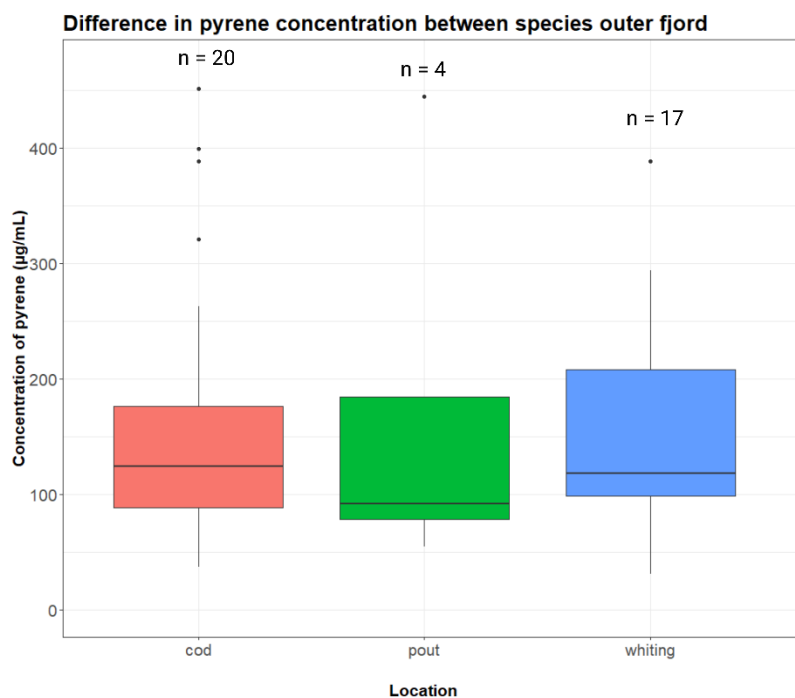


Figure 2: measured difference in pyrene concentration between species in outer Oslofjord. Pout is "Norway pout". The boxes represent the first and third quantiles, while the whiskers represent the maximum and minimum values. The thick, horizontal, and black lines represent median values. N = sample size.

Appendix C

ID	Location	Season	Gender	Age	length_c	length_c (um)	weight_c	otolith_lengt	ann_1	ann_2	ann_3	ann_4
TI009	inner	may	M	2	27,0	270000	183,0	3,27	2,04	2,93	NA	NA
TY005	outer	may	M	3	25,5	255000	136,0	2,80	1,29	1,75	2,33	NA
TY022	outer	may	F	2	29,0	290000	222,3	3,03	1,86	2,73	NA	NA
TY020	outer	may	F	3	33,0	330000	313,2	3,27	1,11	2,55	3,03	NA
TI101	inner	september	F	2	32,0	320000	287,5	3,14	1,59	2,66	NA	NA
TY205	outer	october	F	2	38,7	387000	338,6	3,02	1,48	2,40	NA	NA
TI208	inner	october	F	2	28,0	280000	207,0	3,08	1,77	2,59	NA	NA
TY204	outer	october	F	2	36,0	360000	358,8	2,72	1,08	2,27	NA	NA
TI302	inner	february	F	2	33,0	330000	344,0	3,06	1,55	2,29	NA	NA
TI303	inner	february	F	2	38,5	385000	437,0	3,05	1,17	2,26	NA	NA
TY305	outer	february	M	2	35,0	350000	384,6	2,89	1,26	2,18	NA	NA
TY003	outer	may	F	3	34,8	348000	349,0	2,70	0,94	1,66	2,3	NA
TY006	outer	may	M	2	44,0	440000	796,0	2,78	1,10	2,31	NA	NA
TY015	outer	may	F	3	40,0	400000	497,8	2,94	0,96	1,39	2,32	NA
TY301	outer	february	M	2	23,9	239000	113,1	2,52	1,17	2,12	NA	NA
TI205	inner	october	F	3	30,0	300000	225,0	2,49	0,86	1,38	2,08	NA
TY004	outer	may	F	4	55,0	550000	1480,0	3,78	1,26	1,95	2,51	3,10
TY019	outer	may	F	3	28,0	280000	204,7	2,39	0,92	1,44	2,07	NA
TI013	inner	may	M	5	46,0	460000	825,0	3,28	1,09	1,44	2,19	2,77
TI015	inner	may	F	3	30,0	300000	253,8	2,56	1,04	1,66	2,34	NA
TI019	inner	may	M	3	28,5	285000	209,0	3,13	1,25	1,89	2,89	NA
TI020	inner	may	M	3	37,0	370000	515,0	2,91	1,24	2,01	2,68	NA
TI025	inner	may	M	3	35,0	350000	385,0	2,62	1,17	1,73	2,51	NA
TI105	inner	september	F	2	37,5	375000	575,0	3,05	1,25	2,57	NA	NA
TY203	outer	october	M	3	33	330000	312	2,73	1,18	1,64	2,30	NA
TY304	outer	february	M	3	40,0	400000	563,6	2,81	1,11	1,52	2,18	NA
TI201	inner	october	M	3	37	370000	437	2,95	0,99	1,79	2,60	NA
TI021	inner	may	F	3	35,5	355000	390,0	2,49	1,11	1,62	2,28	NA
TI025	inner	may	M	3	35,0	350000	385,0	2,62	1,17	1,73	2,51	NA
TI210	inner	october	F	2	33,5	335000	344,4	2,44	0,85	1,79	NA	NA
TI103	inner	september	F	4	38,5	385000	500,0	2,66	1,37	1,67	1,94	2,33
TI203	inner	october	F	4	40,5	405000	645,5	2,92	1,15	1,64	2,04	2,43
TY008	outer	may	M	3	36	360000	238	5,01	1,92	3,06	4,39	NA
TY012	outer	may	M	5	44,0	440000	744,5	3,21	0,95	1,68	2,15	2,64
TI023	inner	may	M	5	42	420000	642	2,98	0,91	1,37	1,69	2,08
TY018	outer	may	M	3	38,0	380000	301,8	2,95	0,97	1,87	2,37	NA
TI304	inner	february	F	2	40,5	405000	600	3,39	1,37	2,63	NA	NA
TY010	outer	may	M	3	37,0	370000	460,0	2,55	1,10	1,73	2,3	NA
TY303	outer	february	F	4	47	470000	1000,7	3,63	1,00	1,50	2,28	2,72
TY023	outer	may	M	3	31,5	315000	331,4	2,92	1,42	1,84	2,55	NA
TY021	outer	may	M	4	36	360000	378	2,78	0,87	1,40	2,27	2,59
TY307	outer	february	M	4	45,0	450000	910,0	3,19	1,07	1,71	2,43	2,96
TI106	inner	september	F	3	32	320000	463	3,03	1,61	2,03	2,54	NA
TI203	inner	october	F	4	40,5	405000	645,5	2,93	1,23	1,69	2,08	2,43
TY009	outer	may	M	4	41,5	415000	523,2	3,18	1,39	1,86	2,31	2,82
TI211	inner	october	M	3	31,5	315000	294,5	2,82	0,96	2,19	2,57	NA
TY306	outer	february	F	4	41,0	410000	600,5	3,59	1,29	1,63	2,07	2,77
TI207	inner	october	M	2	34	340000	376,8	2,86	1,45	2,49	NA	NA
TY014	outer	may	F	3	34,0	340000	306,6	2,44	0,82	1,73	2,2	NA
TY203	outer	october	M	3	33	330000	312	2,79	1,06	1,52	2,22	NA
TI107	inner	september	F	3	32,0	320000	233	2,94	1,16	2,05	2,60	NA
TY206	outer	october	F	2	28,5	285000	255,9	2,66	1,51	2,29	NA	NA

ann_5	ann_6	ann_7	TL_01_cm	TL_02_cm	TL_03_cm	TL_04_cm	TL_05_cm	TL_06_cm	TL_07_cm	Last_incr	Last_growth
NA	NA	NA	16,8	24,2	NA	NA	NA	NA	NA	24,2	2,8
NA	NA	NA	11,7	15,9	21,2	NA	NA	NA	NA	21,2	4,3
NA	NA	NA	17,8	26,1	NA	NA	NA	NA	NA	26,1	2,9
NA	NA	NA	11,2	25,7	30,6	NA	NA	NA	NA	30,6	2,4
NA	NA	NA	16,2	27,1	NA	NA	NA	NA	NA	27,1	4,9
NA	NA	NA	19,0	30,8	NA	NA	NA	NA	NA	30,8	7,9
NA	NA	NA	16,1	23,5	NA	NA	NA	NA	NA	23,5	4,5
NA	NA	NA	14,3	30,0	NA	NA	NA	NA	NA	30,0	6,0
NA	NA	NA	16,7	24,7	NA	NA	NA	NA	NA	24,7	8,3
NA	NA	NA	14,8	28,5	NA	NA	NA	NA	NA	28,5	10,0
NA	NA	NA	15,3	26,4	NA	NA	NA	NA	NA	26,4	8,6
NA	NA	NA	12,1	21,4	29,6	NA	NA	NA	NA	29,6	5,2
NA	NA	NA	17,4	36,6	NA	NA	NA	NA	NA	36,6	7,4
NA	NA	NA	13,1	18,9	31,6	NA	NA	NA	NA	31,6	8,4
NA	NA	NA	11,1	20,1	NA	NA	NA	NA	NA	20,1	3,8
NA	NA	NA	10,4	16,6	25,1	NA	NA	NA	NA	25,1	4,9
NA	NA	NA	18,3	28,4	36,5	45,1	NA	NA	NA	45,1	9,9
NA	NA	NA	10,8	16,9	24,3	NA	NA	NA	NA	24,3	3,7
3,14	NA	NA	15,3	20,2	30,7	38,8	44,0	NA	NA	44,0	2,0
NA	NA	NA	12,2	19,5	27,4	NA	NA	NA	NA	27,4	2,6
NA	NA	NA	11,4	17,2	26,3	NA	NA	NA	NA	26,3	2,2
NA	NA	NA	15,8	25,6	34,1	NA	NA	NA	NA	34,1	2,9
NA	NA	NA	15,6	23,1	33,5	NA	NA	NA	NA	33,5	1,5
NA	NA	NA	15,4	31,6	NA	NA	NA	NA	NA	31,6	5,9
NA	NA	NA	14,3	19,8	27,8	NA	NA	NA	NA	27,8	5,2
NA	NA	NA	15,8	21,6	31,0	NA	NA	NA	NA	31,0	9,0
NA	NA	NA	12,4	22,5	32,6	NA	NA	NA	NA	32,6	4,4
NA	NA	NA	15,8	23,1	32,5	NA	NA	NA	NA	32,5	3,0
NA	NA	NA	15,6	23,1	33,5	NA	NA	NA	NA	33,5	1,5
NA	NA	NA	11,7	24,6	NA	NA	NA	NA	NA	24,6	8,9
NA	NA	NA	19,8	24,2	28,1	33,7	NA	NA	NA	33,7	4,8
NA	NA	NA	16,0	22,7	28,3	33,7	NA	NA	NA	33,7	6,8
NA	NA	NA	13,8	22,0	31,5	NA	NA	NA	NA	31,5	4,5
2,97	NA	NA	13,0	23,0	29,5	36,2	40,7	NA	NA	40,7	3,3
2,59	NA	NA	12,8	19,3	23,8	29,3	36,5	NA	NA	36,5	5,5
NA	NA	NA	12,5	24,1	30,5	NA	NA	NA	NA	30,5	7,5
NA	NA	NA	16,4	31,4	NA	NA	NA	NA	NA	31,4	9,1
NA	NA	NA	16,0	25,1	33,4	NA	NA	NA	NA	33,4	3,6
NA	NA	NA	12,9	19,4	29,5	35,2	NA	NA	NA	35,2	11,8
NA	NA	NA	15,3	19,8	27,5	NA	NA	NA	NA	27,5	4,0
NA	NA	NA	11,3	18,1	29,4	33,5	NA	NA	NA	33,5	2,5
NA	NA	NA	15,1	24,1	34,3	41,8	NA	NA	NA	41,8	3,2
NA	NA	NA	17,0	21,4	26,8	NA	NA	NA	NA	26,8	5,2
NA	NA	NA	17,0	23,4	28,8	33,6	NA	NA	NA	33,6	6,9
NA	NA	NA	18,1	24,3	30,1	36,8	NA	NA	NA	36,8	4,7
NA	NA	NA	10,7	24,5	28,7	NA	NA	NA	NA	28,7	2,8
NA	NA	NA	14,7	18,6	23,6	31,6	NA	NA	NA	31,6	9,4
NA	NA	NA	17,2	29,6	NA	NA	NA	NA	NA	29,6	4,4
NA	NA	NA	11,4	24,1	30,7	NA	NA	NA	NA	30,7	3,3
NA	NA	NA	12,5	18,0	26,3	NA	NA	NA	NA	26,3	6,7
NA	NA	NA	12,6	22,3	28,3	NA	NA	NA	NA	28,3	3,7
NA	NA	NA	16,2	24,5	NA	NA	NA	NA	NA	24,5	4,0

Appendix D

#some general info

```
name='Cod'  
st.name='CODSKAG'  
species='Gadus morhua'  
recage=1 #recruitment age  
SSB.conv=1. #convert to tonnes
```

#Get recruitment params from JAGS runs

```
param_file=paste('./',name,'_recparams.csv',sep='')  
recres_file=paste('./',name,'_recres.csv',sep='')  
param=read.csv(param_file,header=T,sep=',')  
recres=as.numeric(read.csv(recres_file,header=T,sep=',')$x)
```

#adapting to field-data

```
matu=3 #age-at-maturity  
mortality=0.55 #natural mortality (from fishbase)  
Loo=90 #maximum length  
kk=0.20.68 #growth parameter adjusted to size-at-age  
t0=0 #scaling size at age zero  
bb=3.11 #convert to weight. Power coefficient from LWR calculations.  
aa=0.00587 #Convert to weight. Obtained from LWR calculation.  
Winf=aaLoo^bb  
  
redkk = 0.98 #reduced growth
```

#————GGPLOT————

```
library("ggplot2") age <- 1:6  
  
inner <- c(12.19, 22.24, 28.41, 33.65, 40.27, 46.81) #median size-at-age estimates  
inner fjord  
  
outer <- c(12.54, 19.82, 29.52, 35.70, 40.71, 46.81) #median size-at-age estimates  
outer fjord  
  
data <- data.frame(age = age, growth = Loo * (1 - exp(-kk * (age - t0))),  
perturbed_growth = Loo * (1 - exp(-kk * redkk * (age - t0))), inner = inner, outer =  
outer)  
  
ggplot(data, aes(x = age)) + geom_line(aes(y = growth), color = "black", linetype =  
"solid") + geom_point(aes(y = inner), color = "blue", size = 3) +  
geom_point(aes(y = outer), color = "green", size = 3) +  
labs(x = "Age", y = "Length (cm)") +  
ggtitle("Median size-at-age growth plot") +
```

```

theme_minimal() +
scale_x_continuous(breaks = age, labels = age)

#create dataframe with the defined values above
par=data.frame(name=name,

                st.name=st.name,
                species=species,
                beta0=param$beta0,
                beta1=param$beta1,
                phi=param$phi,
                res0=param$res0,
                sig=param$sig,
                recage=recage,
                matage=matu,
                mort=mortality,
                winf=winf,
                k=kk,
                t0=t0,
                b=bb
                )

popy=popdyn(par$beta0, par$beta1 , par$phi, par$res0, par$sig, recres, par$recage,
par$matage, par$mort, 0.25, par$Winf, par$k, par$t0, par$b,name)

popdyn_year<-function(beta0, beta1 , phi, res0, sig, recres, recage, matage, mort,
fmort, Winf, kk, t0, bb,name){

##### POPDYN #####

nYsim<-200 # total number of years simulated (incl 100 years discarded)

maxage=min(c(exp((1.46-log(mort))/1.01)*2 ,30)) #Hoenig Fisheries Bulletin 1983 vol
82 no1

waa<-Winf(1-exp(-kk(1:maxage-t0)))^bb
n0=1000

nadd=array(dim=c(50,nYsim)) #abundance at age over time
ntot=array(dim=nYsim) #total biomass starting at age of recruitment

for(j in recage:50) nadd[j,1]=n0exp(-mortj) #Start with an age distribution ntot[1]=n0
ntot1=n0

ssb=sum(nadd[matage:dim(nadd)[1],1]*c(waa[matage:length(waa)],replicate(dim(nad
d)[1]-length(waa),waa[length(waa)])))/1000

ssbt=array(NA,dim=nYsim)

```

```

ssbt[1]=ssb
res.uc=sample(recres,nYsim,replace=T)

res=array(NA,dim=nYsim)
res[1]=res0

## run with random rec
for(i in 2:nYsim){

  ssb=ifelse(i>recage, sum(nadd[matage:dim(nadd)[1],i-
recage]c(waa[matage:length(waa)],replicate(dim(nadd)[1]-
length(waa),waa[length(waa)])))/1000,ntot1) #tonnes (number of individuals in
thousands, weight in grams, [N*WAA]=[kg])

  ssbt[i-recage]=ssb res[i]=phires[i-1]+res.uc[i]
  nadd[recage,i]=ssbexp(beta0)exp(beta1ssb+res[i]-sig^2/2)
  for(j in (recage+1):50) {

    nadd[j,i]=nadd[j-1,i-1]exp(-mort-fmort)

  }

  ntot[i]=sum(nadd[recage:dim(nadd)[1],i]*c(waa[recage:length(waa)],replicate(dim(nad
d)[1]-length(waa),max(waa))))/1000.

}

##### GROWTH REDUCTION SCENARIOS #####

#growth reduction

red= 0.95 #5% reduction = 0.95, 10% reduction = 0.90, 15% reduction = 0.85

kk2=kk*red

waa2<-Winf(1-exp(-kk2*(1:maxage-t0)))^bb

ress=res
nadds=array(dim=c(50,nYsim)) #abundance at age over time
ntots=array(dim=nYsim) #total biomass starting at age of recruitment

ntots[1]=ntot1
for(j in recage:50) nadds[j,1]=n0exp(-mortj) #Start with an age distribution
ssb=sum(nadds[matage:dim(nadds)[1],1]*c(waa[matage:length(waa)],replicate(dim(n
add)[1]-length(waa),waa[length(waa)])))/1000

for(i in 2:nYsim){

  ssb=ifelse(i>recage, sum(nadds[matage:dim(nadds)[1],i-
recage]c(waa[matage:length(waa)],replicate(dim(nadds)[1]-

```

```

length(waa),waa[length(waa)]))/1000,ntot1)#tonnes (number of individuals in
thousands, weight in grams, [N*WAA]=[kg])

if(i > 200 & i<205){ssb=ifelse(i>recage, sum(nadds[matage:dim(nadds)[1],i-
recage]c(waa2[matage:length(waa2)],replicate(dim(nadds)[1]-
length(waa2),waa2[length(waa2)])))/1000,ntot1)} #tonnes (number of
individuals in thousands, weight in grams, [N*WAA]=[kg])

nadds[recage,i]=ssbexp(beta0)exp(beta1ssb+ress[i]-sig^2/2)
#if(i > 200 & i<205) for five years sim
#if(i > 200 & i<210) for ten years sim
#if(i == 202) for one year sim
for(j in (recage+1):50) {

    nadds[j,i]=nadds[j-1,i-1]exp(-mort-fmort)
}

ntots[i]=sum(nadds[recage:dim(nadds)[1],i]c(waa[recage:length(waa)],replicat
e(dim(nadds)[1]-length(waa),max(waa))))/1000.

if(i>200 & i<201)
{ntots[i]=sum(nadds[recage:dim(nadds)[1],i]c(waa2[recage:length(waa2)],repli
cate(dim(nadds)[1]-length(waa2),max(waa2))))/1000.}

}

```