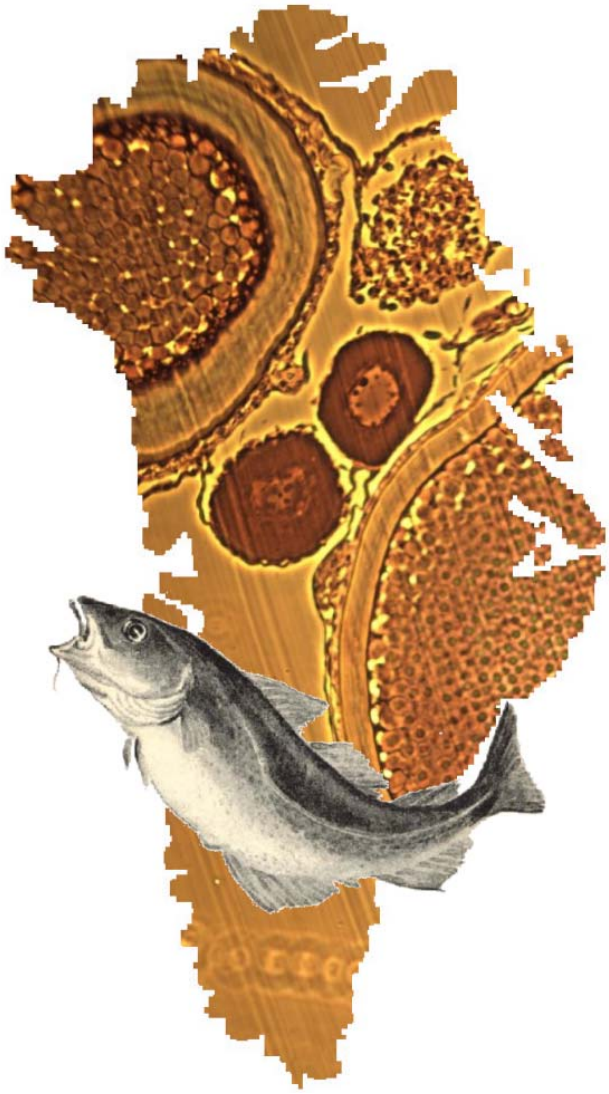
Maturity Determination and Spawning Characteristics for North Atlantic Cod (*Gadus morhua*) in the Nuuk Fjord System

Specialerapport af Kirstine Haidarz Olesen



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AARHUS UNIVERSITET

PINNGORTITALERIFFIK
GRØNLANDS NATURINSTITUT

Maturity Determination and Spawning Characteristics for North Atlantic Cod (*Gadus morhua*) in the Nuuk Fjord System

Et Specialeprojekt

af

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FORORD

Dette speciale er afslutningen på et års studie omkring den reproduktive modenhed og gydning hos Atlanterhavs torsk (*Gadus morhua*) indenskærs i Vestgrønland. Det er udarbejdet i perioden fra januar 2008 til april 2009. Et sideløbende specialeprojekt, ud fra nogle af de samme data, er udført af biologistuderende Suna Schleiss Thomsen. Hendes speciale omhandler den reproduktive investering af energi sammenholdt med fekunditeten hos Atlanterhavstorsken indenskærs i Vestgrønland. Vi indsamlede data i Grønland fra januar til maj 2008, hvorefter indsamlingen resten af året blev varetaget af Grønlands Naturinstitut.

Det var en uforglemmelig oplevelse at arbejde i Grønland med den fantastisk smukke natur, men også det at skulle takle de ekstreme vejrforhold var spændende. Det har været yderst lærerigt og personudviklende i Grønland, da man får chance for at arbejde selvstændigt og bliver givet ansvar når man arbejder på Naturinstituttet.

På afdelingen for Marin Økologi ved Århus universitet hjalp Suna med at dehydrere og indstøbe gonadevæv til de histologiske undersøgelser, hvorefter jeg hen over sommeren fik dem skåret ud og analyseret under mikroskop. Ligeledes hjalp jeg Suna med at lave energi målinger, som hun har brugt til hendes analyser. Derudover har vi diskuteret data og kommet med input til hinandens områder.

Dette specialeprojekt er en del af et større EU-projekt, der skal kortlægge og dokumenterer den indenskærs gydning hos Atlanterhavstorsk i Vestgrønland fra Diskobugten til Sydgrønland. Det overordnede projekt udføres af Grønlands Naturinstitut der vil fortsætte med at indsamle data. Det fremtidige perspektiv for resultatet af dette specialeprojekt er publicering af en artikel og en modenhedsmanual.

Jeg har bevidst valgt at bruge mit mellemnavn *Haidarz* som reference til artiklen og manualen. Artikel og manual (appendiks) er skrevet på engelsk, da disse senere skal publiceres. Introduktionen har jeg valgt at skrive på dansk, for også at formidle emnet og termerne på modersmålet.

Projektet er fondet af Hjemmestyrets EU-fiskerimidler.

Tak til…

En stor tak til Suna Schleiss Thomsen for godt samarbejde og mange uforglemmelige oplevelser specielt i Grønland. Hendes rolige og glade sind, samt store overskud har jeg sat stor pris på. Tak til Grønlands Naturinstitut, specielt Fisk og Rejer afdelingen, for at få mig til at føle mig yderst velkommen og hjemme på instituttet. En speciel tak til Anja Retzel og Holger Hovgård for vejledning og hjælp i felten, men også tak til Rasmus Nygård, Susanne Hvass, Sofie Ruth Jerimiassen, Lars Heilmann, Flemming Heinrich og Morten Kristensen for stor hjælp med feltarbejdet. Også en stor tak til Thomas Juul-Pedersen fra Naturinstituttet (Center for Marinøkololgi og Klimaeffekter), for at være en rigtig god nabo og ven i Grønland, men også for at supplere med hydrografisk og klimatisk viden. Min vejleder Kurt Thomas Jensen for altid at have tid til mine spørgsmål. Og tak til laborant Susanne Vase Petersen for stor hjælp i laboratoriet. En kæmpe tak til ph.d. studerende Rasmus Berg Hedeholm, for stor hjælp og rådgivning igennem hele forløbet, samt for kommentarer og forslag til opgaven. En helt speciel og varm tak til Jonna Tomkiewicz og Rikke Hagstrøm Bucholtz fra DTU Aqua, for vejledning og hjælp til analyse af histologien, begge har brugt flere timer på at gennemgå mine bestemmelser med mig. Derudover har de gennemlæst min artikel og manual og kommet med forslag, kommentarer og rettelser. En tak skal også gives til Pigekammeret, hvor humøret altid er højt, hvilket har givet et rigtig dejligt arbejdsmiljø. En ekstra stor tak skal gives til Mette Schledermann Søndergaard, der er tværfaglig specialestuderende i Kunsthistorie og Biologi, og som har været en hel enorm støtte og medsammensvoren, de mange timer vi har tilbragt sammen på kontoret. Sidst men ikke mindst skal min mand Kim Haidarz Olesen også have en tak, for at give mig plads og støtte til at gennemføre min uddannelse, og så skal han have tak for at designe forsiden til denne opgave.

Opsummering

Dette speciale omhandler overordnet Atlanterhavstorsken (*Gadus morhua*) i Grønland, og mere specifikt de faktorer, der har betydning for den reproduktive udvikling og gydning hos den indenskærs population i Vestgrønland. Data er indsamlet i Nuuk Fjorden fra december 2007 til november 2008 med primært 1 – 3 ugers interval. Fokus har været at udarbejde en modenhedsmanual, der skal kunne benyttes af forskere, til at bestemme den reproduktive modenhed hos de Grønlandske torsk. En sådan manual er ikke tidligere blevet udviklet for denne population. Ud fra dette er faktorer som gydetidspunkt, gydelængde og alder, samt gonadeudviklingen blevet estimeret, hvilket er resulteret i et artikeludkast.

Gydetidspunktet blev bestemt til primært at være i maj og juni, men hannerne startede før og sluttede senere end hunnerne. Derudover var hannerne både yngre og mindre end hunnerne når 50 % af dem var gydende, men hannerne investerede også mindre energi i reproduktion og behøvede dermed ikke et nært så højt energiniveau som hunnerne. Dette er i overensstemmelse med, hvad andre har observeret hos populationer af Atlanterhavstorsken, i andre områder af Nord Atlanten.

Specialet bliver indledt med en introduktion til Atlanterhavstorskens biologi og dens historiske forekomst ved Grønland, med fokus på den indenskærs population. Den hidtidige viden om gydningen indenskærs bliver gennemgået, samt torskens gydestrategi og fekunditet. Torskens udvikling og gydning bliver påvirket af temperaturforhold, hvorfor torskens respons på klimaforandringer også belyses.

Summary

This thesis deals with the Atlantic cod (*Gadus morhua*) in Greenland, and more specific, with the factors affecting the reproductive development and spawning of the inshore population in West Greenland. Data is gathered in the Nuuk Fjord from December 2007 to November 2008 with approximately 1 – 3 weeks between samplings. Focus has been on the preparation of a maturity manual, for scientist to determine the reproductive maturity of Greenlandic Atlantic cod. Such a manual has not previously been developed for this population. In this relation factors such as spawning time, spawning length and age, along with the gonadal development have been estimated and have resulted in an article draft.

The spawning time was determined to be primarily in May and June, but the males initiated before and terminated later than the females. In addition the males were both younger and smaller than the females as they reached the 50 % of maturity, but the males invested less energy in reproduction and were therefore not dependant of as high an energy level. This is in agreement with observations from other studies of Atlantic cod populations elsewhere in the North Atlantic

The thesis is opened with an introduction to the biology and historic occurrence of the Atlantic cod in Greenland, with emphasis on the inshore population. The knowledge of the inshore spawning up to now is reviewed along with the spawning strategy and fecundity of the cod. The ontogenetic development and the spawning of cod are influenced by temperature conditions for which reason the response towards climate change is also looked at.

Indholdsfortegnelse

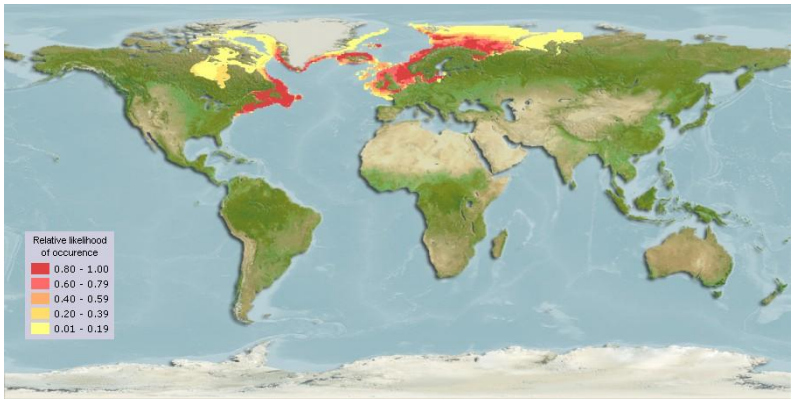
Introduktion

1 Lidt omkring torsk

1.1 Den Nordatlantiske torsk

Den Nordatlantiske torsk tilhører den slægt af fisk, der kaldes *Gadus* fra familien Gadidae. Andre slægter inden for samme familie er kuller (*Melanogrammus*), hvilling (*Merlangius*) og sej (*Pollachius*). Der findes tre arter inden for slægten *Gadus*; Stillehavstorsken (*G. macrocephalus*), Atlanterhavstorsken (*G. morhua*) og Grønlandstorsken (*G. ogac*). Atlanterhavstorsken er en yndet spisefisk og er derfor kommercielt vigtig, og har derved stor økonomisk og samfundsmæssig betydning. Økologisk spiller den dog også en stor rolle, da den er en opportunistisk generalist, hvor føden variere bredt inden for fisk og invertebrater, både pelagiske og benthiske (Link & Garrison, 2002). Den indgår altså i et samspil med flere trofiske niveauer, da den også selv er byttedyr for fx sæler.

Dens udbredelsesområde i Atlanten er fra Nordvest (35˚– 79˚N og 95˚– 42˚V) til Nordøst (36˚– 90˚N og 42˚V – 69˚Ø), fra Kap Hatteras til Ungavabugten langs den Nordamerikanske østkyst, Grønlands øst- og vestkyst, omkring Island og langs Europas vestkyster fra Biscayabugten til Barentshavet, inklusiv Østersøen og området omkring Bjørneøen (Figur 1) (Froese & Pauly, 2009 og Holm *et al.*, 1991). Den findes i forskellige habitater, fra kystlinjen og ned til kontinentalsokkelen, samt i fjorde. De største populationer i Nordatlanten er den norsk-arktiske i Barentshavet samt den Islandske. Generelt er populationen dog faldende, og den er blevet rødlistet som sårbar (Froese & Pauly, 2009). I flere områder, såsom Nordsøen, Østersøen, Barentshavet, Newfoundland og ved Grønland, er bestanden stort set kollapset. ICES (*International Council for the Exploration of the* Sea) og NAFO (*Northwest Atlantic Fishery Organization*) rådgiver hvert eneste år, på videnskabelig basis, omkring et bæredygtigt fiskeri, der vil sikre torskens overlevelse, og dermed at fiskerne også i fremtiden kan udnytte denne ressource. Disse anbefalinger bliver dog sjældent fuldstændig efterlevet, og dermed er udsigterne for en opgang i torskebestanden usandsynlig.

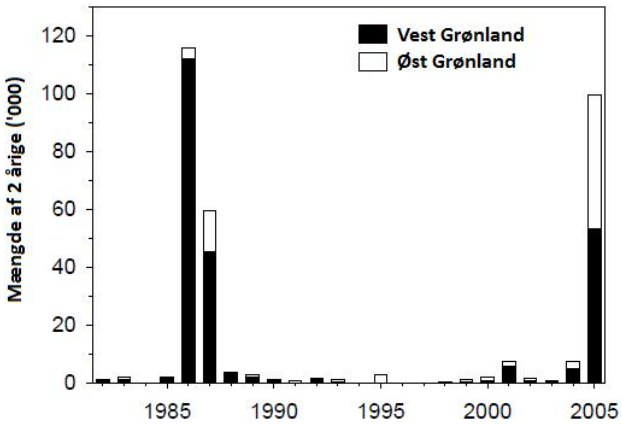


Figur 1
Atlanterhavs-torskens udbredelse og relative forekomst (revideret fra Froese & Pauly, 2009).

1.2 Den Nordatlantiske torsk ved Grønland

1.2.1 Udbredelse

Torskens udbredelse ved Grønland er primært mellem 59˚–70˚N i farvandene vest for, og mellem 59˚–66˚N øst for Grønland, men udbredelsen kan dog variere som effekt af klimaet, hvor varmere perioder udvider den nordlige udbredelse (ICES, 2005) (se også afsnit 1.5). Den samlede torskepopulation ved Grønland inddeles almindeligvis i tre bestande, defineret ud fra deres respektive gydeområder (Hovgård & Wieland, 2008). Den bestand der er i fokus i dette studie, og som vil blive gennemgået mere uddybende i de efterfølgende afsnit, er den indenskærs torsk, der gyder i de vestgrønlandske fjorde. Ud over denne er der en udenskærs bestand, der gyder i farvandene ud for Grønlands øst- og vestkyst. Den sidste bestand opholder sig langs de grønlandske sydvest og sydøst kyster, men har islandsk oprindelse. Via mærkeforsøg har man erfaret, at denne bestand med tiden migrerer tilbage til Island for at gyde (Storr-Paulsen *et al.*, 2004). Larverne bliver transporteret med Irmingerstrømmen fra Sydvestisland til Grønland, og til tider kan mængden af tilførte larver være signifikant og danne grundlag for store årgange i Grønland, hvilket fx var tilfældet i 1973 og især i 1984 (Hovgård, 1993). Der er endnu en rimelig stor årgang fra 2003 (Figur 2), og det tyder på at den ligeledes er blevet tilført fra Island, da den primært kun observeres i sydvest og sydøst Grønland (Hovgård & Wieland, 2008).

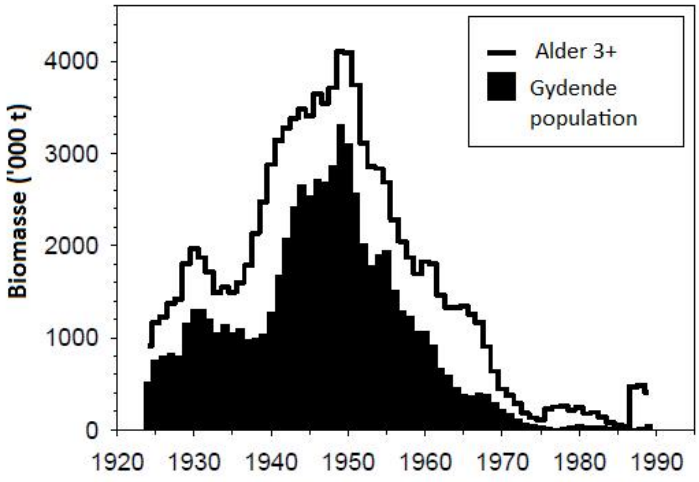


Figur 2
Rekruttering af 2 årige atlantiske torsk i øst- og vestgrønlandske udenskærs farvande i årene 1982 til 2005 (Revideret fra Hovgård & Wieland, 2008).

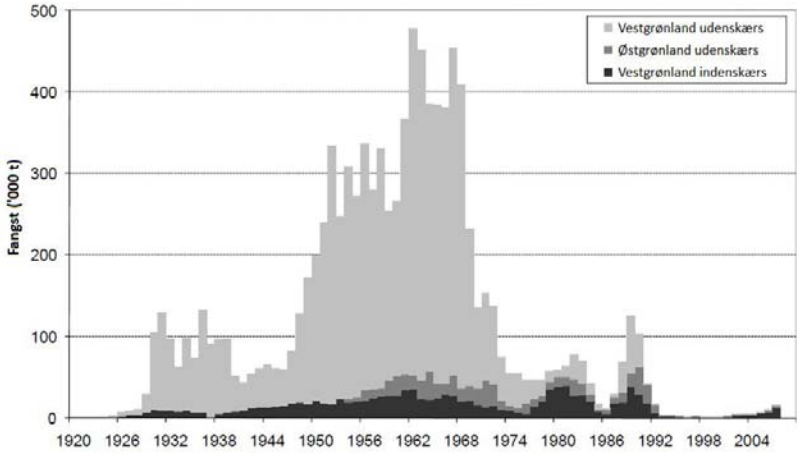
1.2.2 Status – historisk og i dag

Der findes historiske optegnelser omkring forekomsten af torsk ved Grønland, helt tilbage til det 17. århundrede; og disse indikere, at forekomsten har været episodisk og har varieret som følge af ændringer i havmiljøet (Jensen & Hansen, 1931 og Hovgård & Wieland, 2008). I begyndelsen af det 20. århundrede opstod der en stor bæredygtig population i Vestgrønland, hvilket var sammenfaldene med en generel varmeperiode og en markant stigning i hav temperaturen, der gav populationen gode vækst og overlevelsesbetingelser (Hansen, 1949; Buch *et al.*, 1994 og Drinkwater, 2006). De første observationer af populationen var ved sydvest Grønland, og man formoder derfor, at den i larvestadiet var blevet ført med Irmingerstrømmen fra Island (Hovgård & Wieland, 2008). Denne opgang i torskebestanden blev startskuddet for det kommercielle torskefiskeri, der så småt kom i gang i 1911, dog kun indenskærs og kun af grønlandske fiskere med hånd- og langline (Hansen, 1949 og Storr-Paulsen *et al.*, 2004). I 1920'erne gjorde det internationale fiskeri sit indtog i de grønlandske farvande, hvormed størstedelen af fiskeriet flyttede til udenskærs fangstområder, og den indenskærs fangst blev mindre vigtig (Storr-Paulsen *et al.*, 2004). Torskepopulationen voksede stødt i Vestgrønland frem til 1950, hvor den toppede med en biomasse på 4.1 millioner t (Figur 3). Herefter faldt biomassen dog hurtigt, hvilket kan tilskrives et overdrevent fiskeritryk (Figur 4) (Hovgård & Wieland, 2008), koblet med at mængden af gydende torsk faldt (Figur 3), som følge af et fald i temperaturen (Drinkwater, 2006). Disse faktorer kombineret har resulteret i en høj mortalitetsrate for de gydende fisk, samtidig med en lav rekruttering og lav overlevelsesrate for torskelarver, hvormed bestanden ikke længere var bæredygtig.

På trods af dette fortsatte det høje fiskeritryk i 1960'erne, hvor det toppede med en årlig fangst på mellem 350.000 – 500.000 t (Storr-Paulsen *et al.*, 2004). I 1990'erne kollapsede bestanden totalt, og der har stort set ikke været noget torskefiskeri førend for ca. 4 år siden i 2005 (Figur 4), hvor der igen er tegn på nye rekrutter (Hovgård & Wieland, 2008).



Figur 3
Estimering af biomassen for 3+ årige og gydende Atlanterhavs torsk i Vestgrønland ud fra Virtuel Populations Analyser (VPA). (Revideret fra Hovgård & Wieland, 2008).



Figur 4
Fangsttal for henholdsvis Øst- og Vestgrønland udenskærs og for Vestgrønland indenskærs fra 1920 til 2007. Søjlerne er stablet. (Revideret fra ICES, 2008b).

Mens det udenskærs fiskeri var på sit højeste, udgjorde fiskeriet indenskærs mindre end 10 % af den samlede fangst (Hovgård, 1993). Generelt har det indenskærs fiskeri på Atlanterhavstorsk, defineret som den fangst der landes af grønlandske kommercielle fiskere i kyst- og fjordområder, fulgt den overordnede fangst trend. Frem til 1942 holdt den årlige indenskærs fangst sig under 10.000 t,

men steg så gradvist herefter, og fra 50'erne til 80'erne fluktuerede den stort set mellem 20.000 – 40.000 t årligt. I 1990'erne under kollapset var fangsten helt nede under 400 t om året, hvilket er historisk lavt (Hovgård & Wieland, 2008). På trods af dette udgør den indenskærs fangst i dag mere end 90 % af det samlede torskefiskeri ved Grønland (Storr-Paulsen *et al.*, 2004). I rådgivning behandles de tre bestande (den indenskærs, den udenskærs og den islandske) under et, da det ikke er muligt at opmåle bestandskomponenterne separat. Derfor er den præcise indenskærs bestandsstørrelse og udnyttelsesrate ukendt. I 2006 var de samlede fangster for både uden- og indenskærs torsk i grønlandske farvande på 10.000 t. I 2007 var fangsten på 17.000 t, hvoraf 14.000 blev taget i Vestgrønland. På trods af fremgangen er rådgivningen fra ICES, at der ikke fiskes direkte på torskebestanden i 2009, for at give gydebestanden de bedste forudsætninger for en genopbygning (ICES, 2008b). ICES anbefaler yderligere, at der bør udarbejdes en flerårig forvaltningsplan for fiskeriet ved Grønland, der skal sikre at det ikke ekspanderer, før der er registreret en betragtelig stigning i biomassen, og i rekrutteringen til bestanden.

Koblingen mellem de forskellige populationer er endnu ikke helt klart for forskerne. Det vides derfor ikke, i hvilken grad de stærke islandske årgange har betydning for den indenskærs biomasse, eller om denne kun består af rekrutter fra lokal gydning (Storr-Paulsen *et al.*, 2004). Taget i betragtning, at det indenskærs fiskeri følger det udenskærs, specielt i de år med mange islandske torsk (fx 1989), kunne det tyde på, at der er en vis grad af udenskærs fisk i fjordene og langs kysterne. Man har dog siden midten af 1920'erne lavet fangst-genfangst forsøg, hvor man har mærket torsk og genudsat dem samme sted, som de blev fanget. Ud fra genfangst af mærkede torsk, har man kunnet kortlægge populationernes overordnede migrationsmønstre. Forsøgene indikerer, at den indenskærs population forbliver i fjordene og langs kysterne (Hansen, 1949). Ydermere ser det også ud til, at der kun er en meget begrænset blanding af populationer imellem fjordene (Storr-Paulsen *et al.*, 2004). Det vil sige at de primært vokser op i samme fjord, som de er blevet gydt. Kort sagt ser det ud til, at udenskærs torsk svømmer ind i fjordene, men at de ikke gyder der. De indenskærs torsk forbliver i den fjord de er gydt hele deres liv. Det er derfor ikke sandsynligt at de to populationer bliver blandet, da de gyder hver for sig, men der mangler dog genetisk understøttelse for denne påstand (Storr-Paulsen *et al.*, 2004).

1.3 Sammenligning af udenskærs og indenskærs forhold i Vestgrønland

1.3.1 Hydrografi

Dette afsnit er baseret på beskrivelser af de hydrografiske forhold i Vestgrønland af Smidt (1979). Der er stor forskel på de hydrografiske forhold udenskærs og indenskærs i Sydvestgrønland. De inderste fjorde er ikke nær så påvirket af den variation i de hydrografisk forhold, der forekommer udenskærs. Klimaet og sæsonvariationen i Sydvestgrønland er stærkt påvirket af to store havstrømme.

- Irmingerstrømmen, der er en sidegren til Den Nordatlantiske strøm, og som medbringer varmt og højsalint vand.
- Den Østgrønlandske Strøm, der medbringer koldt polarvand, med en lav saltholdighed, som følge af store mængder havis.

De to havstrømme mødes ved Kap Farvel og flyder op langs vestkysten, hvor de efterhånden bliver opblandet. Det lavsaline polarvand dominerer dog i de øverste vandlag. Den relative indflydelse fra havstrømmene varierer dog fra år til år og imellem sæsoner, hvilket bevirker variationen. Noget af det varme højsaline irmingervand strømmer som bundvand et stykke ind i fjordene. Dette opvejes dog af det lavsaline smeltevand, der tilføres fjordene fra floder og gletscherer. Generelt er temperaturen gennem vandsøjlen stabil inderst i fjordene, hvor havstrømmene ikke når ind; overfladen bliver dog påvirket af lufttemperaturen. Saliniteten ved bunden i fjordene er ligeledes stabil, men i overfladen svinger den i takt med sæsonerne, og er markant lavere om sommeren, når isen smelter.

Hydrografien har stor betydning for torskens udvikling og overlevelse. Blandt andet afhænger tidspunktet for hvornår æggene klækkes, af temperaturen i det vandlag, de flyder i (se afsnit 1.4.3 - *Fekunditet*). Larvernes evne til at overleve, samt primærproduktion og sekundærproduktion, bliver også påvirket af hydrografien (Smidt, 1979). Primær- og sekundærproduktionen udgør de første led i fødekæden, og har dermed stor indflydelse på de højere trofiske niveauer, inklusivt torsken. Udenskærs bliver der i løbet af vinteren og det tidlige forår tilført næring til overfladen, som følge af en vertikal opblanding af vandsøjlen. Dette medfører en god

basis for en høj primærproduktion. Indenskærs er der stort set ikke nogen opblanding af vandsøjlen, og derfor er primærproduktionen generelt meget lav.

1.3.2 Økologi

Som nævnt i afsnit 1.1.1 er torsken en generalist med hensyn til fødevalg, men omvendt har faktorer, som fiskens størrelse, samt sæson og område, betydning for selve fødeudbuddet (Link & Garrison, 2002 og Smith, 2007). Generelt er der et klart ontogensisk skift i fødevalg, fra små pelagiske krebsdyr og invertebrater, mens torsken selv lever pelagisk, til større benthiske invertebrater og fisk, når torsken senere i sin udvikling skifter til benthisk fouragering; det er primært torsk > 50 cm, der spiser fisk (Link & Garrison, 2002).

Hvis der er mangel på føde, benytter torsken lipider oplageret i leveren, som energireserve (Lambert & Dutil, 1997). Derudover bruger huntorsken også denne energikilde i forbindelse med modning af oocyter (Kjesbu *et al.*, 1991) Det er derfor vigtigt for torsken med en meget lipidholdig føde. Lodden (*Mallotus villosus*) har et højt lipidindhold, og der er en positiv korrelation mellem torskens leverindex (forholdet mellem levervægt og renset vægt) og mængden af lodder i et område (Rose & O'Driscoll, 2002). Lodden udgør mere end 57 % af den relative fødevægt for Atlanterhavstorsken i Vestgrønland (Rasmus Nielsen & Andersen, 2001). Friis-Rødel & Kanneworrf (2002) har lavet en opsummering på lodden i de grønlandske farvande, ud fra publiceret og upubliceret litteratur. Den er vidt udbredt i fjordene i Vestgrønland, men findes også udenskærs fra kysten til vest for bankerne. Den gyder inderst i fjordene mellem april og juli, afhængig af nordlig eller sydlig udbredelse. Allerede fra december begynder migrationen ind i fjordene, og i den forbindelse kan man også opleve en større tæthed af torsk.

I et tidligere specialeprojekt om Atlanterhavstorskens fødeøkologi (Mikkelsen & Svendsen, 2008), blev konditionsfaktoren (forholdet mellem torskens vægt og dens længde i tredje potens) udregnet for udenskærs torsk i Vestgrønland. De fandt at konditionen i efteråret 2006 gennemsnitligt lå på 0,91. Ifølge Lambert & Dutil (1997) findes de højeste værdier for leverindeks og konditionsfaktor i sensommeren og om efteråret, mens de laveste værdier forekommer i gydeperioden. Til sammenligning med den udenskærs kondition viser resultaterne fra dette specialeprojekt, at den indenskærs konditionsfaktor fra december 2007 til juni 2008, hvilket inkluderer

gydeperioden, var > 0,98 for hunner og > 0,96 for hanner (Haidarz *et al.*, in prep.). Det tyder dermed på, at den indenskærs bestand er i bedre kondition end den udenskærs, hvilket også indikeres af tabel 1, der viser det totale maveindhold for Atlanterhavstorsk i Vestgrønland, henholdsvis ved kysten og inde i fjordene. Det ses at torsk inde i fjordene generelt spiser mere end de der lever uden for fjordene.

Tabel 1 Totale maveindhold for Atlanterhavstorsk i Vestgrønland (i fjorde og ved kysten) i forhold til længdegruppe (cm) (Revideret fra Rasmus Nielsen & Andersen, 2001).

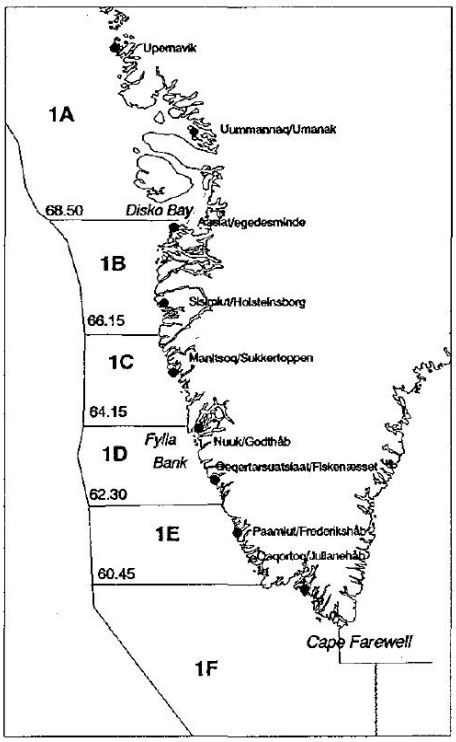
Område	30–34	35–39	40–44	45–49	50–54	55–59	60–69	70–79	80–89	>90	Middel
Kyst	…	20.53	23.38	27.61	34.23	50.98	84.90	3.70	184.60	…	53.74
Fjord	…	54.79	182.20	56.21	52.60	65.17	69.71	26.69	81.14	107.20	73.56

… Ingen observationer

1.4 Gydningen indenskærs

1.4.1 Områder hvor torsken gyder

Der foregår gydning i flere af de grønlandske fjorde på vestkysten. I 1949 konkluderede Hansen (1949), efter at have undersøgt forekomsten af torskeæg i fjordene på vestkysten, fra Umanak Fjord (70˚N; NAFO inddeling 1A) til Nanortalik syd for Qaqortoq (60˚N; NAFO inddeling 1F) (Figur 5), at gydningen primært finder sted mellem 67 – 64˚N (fra Sisimiut til Nuuk; NAFO inddelinger 1B – 1D). Mest intens var gydningen i Nuuk Fjorden, især i bunden af Kapisillit fjordarmen ved bygden Kapisillit. Ud fra nyere data estimerede Hovgård & Wieland (2008) dog, at gydningen nu primært foregår mellem 66 – 62˚N, men stadig at den er størst i Nuuk Fjorden og primært i Kapisillit. Grønlænderne har længe vidst, at torsken gyder i Kapisillit og Hansen (1949) fortæller, at de lokale om foråret tørrede store mængder af modne torskegonader på klipperne.



Figur 5
Kort der viser den sydvestlige del af Grønland inklusiv NAFO område inddelinger, samt navne på byer og bygder (grønlandsk/ dansk) (Fra Buch *et al.*, 1994).

1.4.2 Tidspunkt for gydning

Den måned, hvor Hansen (1949) observerede flest æg i fjordene var i maj. I Nuuk Fjorden fik han fx ud af 22 ægtræk indsamlet 28.112 æg i maj. I juni og juli var mængden af æg allerede faldet drastisk, og ud af 18 ægtræk i begge måneder, fik han kun henholdsvist 1.509 og 23 æg. Der er desværre ingen oplysninger fra april. Ud fra undersøgelser af modenhed på gonaderne fra torsk, fanget mellem den 21. – 30. maj 1935 i Nuuk Fjorden, kunne Hansen (1949) se, at størstedelen af hunnerne var udgydte og han konkluderede, at gydningen var ved at være slut. I 1936 viste lignende undersøgelser til gengæld, at gydningen var på sit højeste mellem den 26. maj og den 5. juni. Generelt om gydeperioden i fjordene på vestkysten siger Hansen, at den i milde år angiveligt starter allerede i slutningen af marts og vare end til en gang i juni. Det varierer imellem fjorde, hvilke betingelser der er mest optimale for gydning og udvikling af æg, hvilket de to følgende eksempler fra henholdsvis Kapisillit og Ikertoq Fjord (syd for Sisimiut) viser (Hansen, 1949). I Kapisillit gyder torsken på en dybde mellem 25 – 50 m og ved temperaturer mellem 1 – 4˚C. I 1936, hvor der blev fundet mange æg i overfladen, lå temperaturen mellem 5 – 6˚C. I Ikertoq Fjord gyder

torsken mellem 100–200m ved temperaturer omkring 1˚C og overfladetemperaturen lå i måleperioden 24. maj – 5. juni 1934 på ca. 1 – 3˚C.

I 1979 viste en undersøgelse foretaget af Smidt (1979) fra 1954 – 1964, at gydningen foregik mellem februar og juni. Den største koncentration af æg forekom primært i april, men også i maj og juni i nogle år. Der findes ingen recente dokumentationer omkring gydetidspunktet for indenskærs torsk i Vestgrønland (Storr-Paulsen *et al.*, 2004). Dette specialeprojekt har derfor blandt andet haft det formål, at fastlægge denne periode. For at sammenligne den nuværende gydeperiode med de tidligere undersøgelser (Hansen, 1949 og Smidt, 1979) (Tabel 2), vil det efterfølgende således være fra resultaterne af dette specialeprojekt (Haidarz *et al.*, in prep.). Der blev indsamlet torsk i fjordene ved henholdsvis Qaqortoq (60˚N; NAFO 1F), Paamiut (62˚N; NAFO 1E) og Nuuk (64˚N; NAFO 1D). Desværre blev der ved Qaqortoq og Paamiut kun indsamlet i februar og marts, og der blev ingen af stederne fanget nogen gydende fisk. Derfor er resultaterne kun baseret på undersøgelser foretaget i Nuuk Fjorden. Estimaterne for gydetidspunktet er baseret på tidspunktet for tilstedeværelsen af gydende torsk, og på ændringer i deres energiniveau i leveren. Der blev observeret gydende torsk af begge køn fra april til juni, med flest modne fisk i maj og juni. Det var også fra maj, at den største ændring i energiniveau blev observeret (energien bliver allokeret fra lever til gonade). Der var forskel på gydestrategien mellem kønnene; blandt andet på hvor tidligt og hvor længe de var gydemodne. Hannerne blev både modne tidligere og forblev med at være det i længere tid end hunnerne.

Tabel 2 opsummering af gydetidspunkt for *Gadus morhua* indenskærs i Vestgrønland. Se detaljer i teksten.

Kilde	Årstal for undersøgelse	Primære gydetidspunkt	Gydeperiode
Hansen, 1949	Før 1949	Maj	Marts-juni
Smidt, 1979	1954-1964	April	Februar-juni
Haidarz *et al.*, in prep.	2008	Maj-juni	April-juni

1.4.3 Gydestrategi

Som tilpasning til det miljø de lever i, udvikler fisk en gydestrategi, der giver de bedste betingelser for deres afkom. Murua & Saborido-Rey (2003) har opsummeret

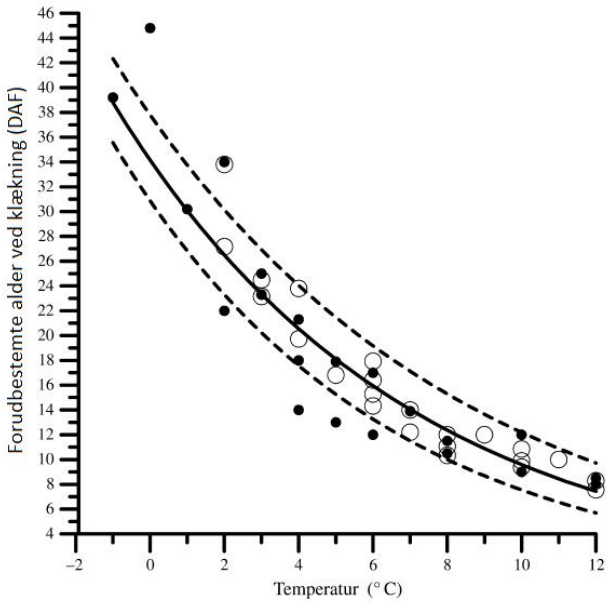
de forskellige strategier, som hunfisk anvender i Nordatlanten. De fleste af de fisk, der lever i kolde eller tempererede egne, inklusiv Atlanterhavstorsken, er såkaldte portionsgyderer, hvor en mængde oocyter gydes løbende i et antal portioner over en periode. Antallet af portioner og periodens længde kan dog variere mellem arter, men generelt kan strategien betragtes som en mulighed for at gyde over en længere periode, og dermed øge sandsynligheden for afkommets overlevelse. Alternativet er at være totalgyder, hvor alle oocyterne modnes og frigives af én omgang. Oocyterne hos Atlanterhavstorsk modnes gruppe-synkront, hvilket ifølge Murua og Saborido-Rey (2003) betyder, at mindst to populationer af oocyter er til stede i ovariet på ethvert givent tidspunkt. De to populationer består henholdsvis af store relativt homogene oocyter, der vil blive gydt den følgende sæson, og en population af små mere heterogene oocyter, der udvikler sig til de oocyter, som vil blive gydt i senere sæsoner. Den gruppe-synkrone udvikling ses generelt hos fisk med en relativ kort gydeperiode, og hvor dannelsen af æggehvide primært afhænger af kropsressourcer, hvilket gælder for mange marine arter, der lever i kolde områder. Andre strategier, for udvikling af oocyter, er henholdsvis synkron, hvor alle oocyterne udvikler sig ens i samme tempo, og asynkron, hvor alle udviklingsstadier forekommer på samme tid uden nogen dominans. Når Atlanterhavstorsken bliver reproduktionsmoden, vil den gyde hvert år, med mindre den springer en gydning over, eller gonaden degenererer. Dette kaldes iteropar og gælder for langt de fleste fisk, samt for de fleste dyr og planter. Ligesom alle andre fisk der gyder er Atlanterhavstorsken ovipar, hvor embryoet udvikler sig udenfor ovariet.

Fekunditet

Fekunditeten hos Atlanterhavstorsk er determineret, hvilket betyder at den samlede mængde af vitellogenesiske oocyter, er bestemt forud for gydningen, og der bliver ikke produceret flere, i stedet falder antallet ved hver gydning. Modsætningen til determineret fekunditet er indeterminered fekunditet, hvor antallet af vitellogenesiske oocyter ikke er forudbestem, og dermed udvikler de sig løbende hen over gydeperioden (Murua & Saborido-Rey, 2003). Holm *et al.* (1991) har samlet forskningsresultater omkring torskens gydning i opdrætsøjemed, men drager paralleller til de naturlige forhold. Det følgende er ud fra deres undersøgelser. I naturen gyder huntorsken omkring 15 gange på en sæson med 2 – 3 dages

mellemrum. Der er forskel på mængden, størrelsen og vitaliteten af oocyter, alt efter hvornår portionen bliver gydt, men også om fisken er førstegangsgydende, samt fiskens længde har betydning. En førstegangsgydende har oftest lavere fekunditet og oocyterne er mindre end hos fisk, der har gydt i tidligere sæsoner. Jo større fisken er jo højere er dens fekunditet, både i forhold til længde, hvilket Thomsen *et al.* (in prep.) også har vist, men også i forhold til vægt. Generelt er de første portioner, der bliver gydt, mindre og kvaliteten af oocyterne kan være dårligere end i senere portioner, hvilket medfører at mange går til grunde. Den bedste kvalitet af oocyter fås typisk omkring femte gydeportion (Støttrup, 2002). Holm *et al.* (1991) har gjort observationer ved Norge, der viser at kysttorsk (inklusiv indenskærs torsk) har en væsentlig højere fekunditet end skrei (norske udenskærs torsk). Dette kan muligvis forklares ved, at kysttorsken vandre mindre og derfor kan investere mere energi i oocyt produktion.

For at opsummere, så er huntorskens gydestrategi altså iteropar, med determineret fekunditet, gruppe-synkron oocyt udvikling og gydning i portioner. Andre fisk med den samme strategi er fx hellefisk (*Reinhardtius hippoglossoides*), sej (*Pollachius virens*) og rødspætte (*Hippoglossoides platessoides*) (Murua & Saborido-Rey, 2003). Selve befrugtningen foregår ved at han- og huntorsken svømmer med bugen mod hinanden, mens oocyter og spermatozoer sprøjtes ud og bliver blandet sammen (Holm *et al.*, 1991). Når æggene er befrugtet, flyder de op i de øverste vandmasser og opholder sig på den dybde, hvor saliniteten holder dem neutralt flydende (Mackenzie *et al.*, 2000). Temperaturen har stor betydning for, hvornår æggene klækkes (Holm *et al.*, 1991), hvilket Geffen *et al.* (2006) illustrere i en generel model, der viser antal dage fra befrugtning (DAF) til ægget klækkes i forhold til temperatur (Figur 6).



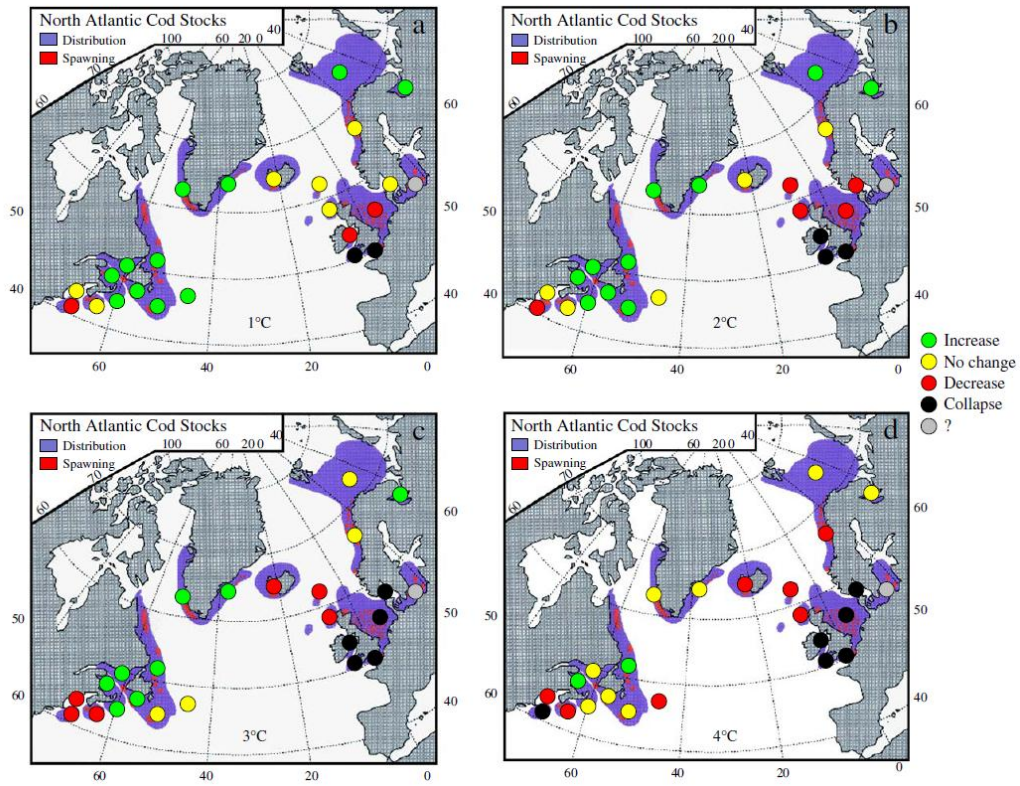
Figur 6
Det forudsete antal dage fra befrugtning (DAF) til 50 % æg er klækket, i forhold til temperatur, for Atlanterhavs torsk (lukkede cirkler for N.V. Atlanten og åbne cirkler for N.Ø.) Stiplede linje er 95 % CI. (Revideret fra Geffen *et al.*, 2006).

Temperaturen i de øvre vandmasser, i den inderste del af Nuuk Fjorden, lå i midten af maj 2008 på ca. 2˚C (T. Juul-Pedersen, Grønlands Naturinstitut, pers. komm.). Ud fra figur 6 vil det sige, at der går omkring 27 dage fra et æg bliver befrugtet, til det klækkes i dette område.

1.5 Torskens respons på klimaforandring

Det er efterhånden gået op for alle, at klimaet er ved at ændre sig, som følge af en unaturlig høj mængde drivhusgasser i atmosfæren, udledt af menneskelige aktiviteter og interesser. Man taler om en global opvarmning, men det er ikke ensbetydende med, at det til ethvert tidspunkt bliver varmere. Sæsonvariationerne vil i nogle områder blive mere ekstreme, mens de i andre vil blive mere udlignet. Generelt vil temperaturen dog stige, set over alle sæsoner samlet. Der er utrolig mange faktorer, der har indflydelse på og konsekvens for hinanden med hensyn til klimaet, hvilket også er grunden til, at forskerne ikke blot kan komme med det præcise fremtidige klimascenario. Drinkwater (2005) og ICES (2008a) har opsummeret, hvilke konsekvenser klimaforandringerne kan have for Atlanterhavstorsken. Klimamodeller indikerer, at den største temperaturstigning vil ske i de arktiske og subarktiske områder, herunder Grønland. Set over hele Nordatlanten vil en opvarmning af hav temperaturen forskyde torskens udbredelse

længere nordpå, hvilket man så i Vestgrønland under varmeperioden i starten af det forrige århundrede (jf. afsnit 1.2.2). Ved Island bredte torskepopulationen sig samtidig fra det sydlige område til det nordlige. I 1960'erne, hvor det igen blev koldere, trak begge bestande sig igen sydpå. Umiddelbart har det ikke nogen negativ, men nærmere en positiv effekt for den grønlandske torskepopulation, at hav temperaturen stiger. Figur 7a-d viser fire scenarier for torsken, hvis temperaturen stiger med 1-4˚C.



Figur 7 Forventet ændring i forekomsten af Atlanterhavstorsk populationer ved temperaturstigninger nær havbunden på (a) 1˚C, (b) 2˚C, (c) 3˚C og (d) 4˚C over det nuværende niveau. Violet farve viser udbredelsesområde; rød farve indikere gydeområde; grønne prikker indikere en forøgelse af bestanden; gule prikker indikere at der ikke er nogen forandring; røde prikker indikere et fald i bestanden; sorte prikker indikere at bestanden vil kollapse. (Revideret fra Drinkwater, 2005).

Ved 1-3˚C stigning vil populationerne ved Grønland blive større (Figur 7a-c) og ved en stigning på 4˚C vil der ikke være nogen forandring (Figur 7d). Dette skyldes, at Grønland på nuværende tidspunkt allerede udgør det nordligste af torskens udbredelsesområde. Knapt så positivt ser det ud for populationerne i torskens sydligste udbredelsesområder, blandt andet Den Engelske Kanal og Det Keltisk hav, hvor selv en stigning på 1˚C nok vil medføre at populationerne i disse områder

kollapser totalt (Figur 7a). Vandtemperaturen vil simpelthen blive for varm, hvilket øger mortaliteten for æg og larver, og de adulte torsk stopper med at æde.

En temperaturstigning har dog ikke kun indflydelse på torskens udbredelsesområde, også vækstraten, den alder hvor de bliver modne, samt gydetidspunktet, bliver ligeledes påvirket. 90 % af torskens vækst kan forklares ud fra havbundstemperaturen, hvor en varmere temperatur giver en højere vækstrate. Samtidig med at væksten stiger, vil alderen for hvornår de bliver gydemodne falde med ca. 1 år for hver 2˚C temperaturstigning. Disse konsekvenser er allerede blevet dokumenteret i flere områder, blandt andet i Grønland. Ved højere temperaturer udvikler gonaderne sig hurtigere, hvilket betyder at fiskene hurtigere bliver klar til at gyde, og dermed vil gydesæsonen starte tidligere. Problemerne med disse ændringer er, at hele det økologiske samspil, som torsken har udviklet sig til at være tilpasset, ikke nødvendigvis ændre sig i parallel forstand, og dermed kan der opstå en negativ effekt for torskens udvikling. En temperaturstigning vil få indflydelse på mængden af nedbør samt vindforhold, hvilket vil påvirke hydrografien. Ændringer i hydrografien har blandt andet konsekvenser for primærproduktionen, som har indflydelse på resten af fødekæden (jf. afsnit 1.3.1). Netop på grund af alle de komponenter, der findes i det økologiske samspil, er det svært at spå om torskens helt nøjagtige respons på klimaforandringerne. Ikke mindst derfor, bør der tages ekstra meget hensyn til torskebestanden, og fangstregulativer bør følges, så der ikke udøves uopretteligt skade på bestandene. Det er yderligere vigtigt, at alle de økologiske interaktioner torsken indgår i belyses, både udenskærs og indenskærs, for at få det mest nuancerede og præcise billede.

2 Efterskrift

2.1 Manualen - Layout og anvendelse

Dette speciale indeholder blandt andet en modenhedsmanual, der kan bruges til at identificer den reproduktive status af indsamlede torsk. Jeg har valgt et layout, der viser både de makroskopiske og de histologiske karakteristika for et givent modenhedsstadie. Dette har jeg valgt på baggrund af, at de to måder at vurdere modenhed, kan supplere hinanden. Histologien kan give en forståelse for gonadens

makroskopiske udseende og omvendt. Ved hanner er der fx en stor forskel på gonadestørrelsen, fra den er umoden til den bliver modnende. Dette skyldes at kønscellerne deler sig adskillige gange via mitose, hvilket giver en lineær forøgelse af celler og en tilsvarende forøgelse i gonadestørrelsen. Omvendt kan de makroskopiske kendetegn også bruges til at forstå det histologiske udseende, fx kan det være svært at skelne imellem de tre gydende modenhedsstadier hos hunner (start-, hoved- og afsluttende gydning). Her er det den relative mængde af oocyter der definere modenhedsstadiet, hvilket bedst vurderes ud fra det makroskopiske og det histologiske udseende kombineret.

At kombinere den histologiske og den makroskopiske modenhedsbestemmelse er inspireret af Morrison (1990), der dog viser flere eksempler på begge, samt mere detaljeret eksempler. Layoutet, med kun ét stadie pr side og få billeder, er inspireret af Tomkiewicz *et al.* (2002), der dog kun viser de makroskopiske karakteristika. Manualen fra Tomkiewicz *et al.* (2002) er en feltmanual, der skal bruges når man står med torsken og skal vurdere dens modenhed, hvorimod Morrison mere er til, at give en forståelse for torskens reproduktive udvikling. Formålet med denne manual er, at den skal bruges i felten i Grønland, på samme plan som Tomkiewicz *et al.* (2002), der bruges til Østersøen.

2.2 Forbedringer

Der er grundlag for enkelte forbedringer og overvejelser i forbindelse med udførelsen af dette projekt. Til indsamling af torsk blev der brugt lang- og håndline med madding på kroge. Dette er ikke optimalt da det er bevist, at især huntorsk stort set ikke spiser i gydeperioden (Fordham & Trippel, 1999), hvorfor de modne torsk sandsynligvis bider sjældnere på krogen. Dette kan resultere i en skæv modenhedsfordeling med overrepræsentering af ikke gydende individer. Alternativt kan man i stedet bruge net, her er det dog vigtigt, at bruge små maskestørrelser, så der ikke bliver en skæv størrelsesfordeling af fisk. For at bestemme gydeperioden for fisk, er det mere præcist at bruge forekomsten af æg i vandoverfladen, da både haner og hunner kan være i et gydende stadie i nogen tid, uden egentligt at gyde.

2.3 Perspektivering

I det følgende artikeludkast vil resultaterne af de oparbejdede data, indsamlet i Grønland, blive præsenteret. Resultaterne tegner et billede af de tidsmæssige tendenser omkring torskens gydning, såsom tidspunkt, alder og længde for gydningen. De opnåede resultater benyttes i modenhedsmanualen, som en hjælp til at bestemme det reproduktive modenhedsstadie hos torsk. Undersøgelserne kan derudover bruges i forbindelse med den kortlægning og dokumentering af gydningen indenskærs i Vestgrønland, som Grønlands Naturinstitut netop er i gang med. Også med hensyn til torskens fødesøgningsadfærd, respons på klimaændringer og gydeadfærd kan disse undersøgelser bruges som grundlag for videre forskning.

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Maturity Determination and Spawning Characteristics for North Atlantic Cod (*Gadus morhua*) in the Nuuk Fjord System

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The time of spawning together with the age and length at spawning were estimated for male and female North Atlantic cod (*Gadus morhua*) from the fjord systems in the Nuuk area in West Greenland in 2007 - 2008. Peak spawning for both males and females was observed in May and June. The length at which 50 % of the females spawn is 50 cm and for the males, which reach the reproductive age earlier than the females, the length at 50 % maturity is 40 cm. The age at 50 % maturity is between 4 – 5 years for females and between 3 – 4 years for the males. The females had a higher mean hepatosomatic index (*HSI*) throughout the entire sampling period, but they also had a larger decrease in liver energy during the spawning period than the males. This indicates that the females allocate a larger amount of energy to reproduction compared to the males. The gonadosomatic index (*GSI*) for the females peaked in May and decreased hereafter during the spawning period. For the males the peak in *GSI* was already in January where after it decreased in two stages; the first from January to March and the second from May and during the rest of the spawning period. The males start spawning early to ensure fertilization of potentially early spawned oocytes. The primary spawning ground in the Nuuk Fjord system is Kapisillit but spawning specimens both males and females were also caught in Ameralik and Buksefjorden.

Key words: Gadus morhua; maturity staging; reproduction; spawning characteristics; West Greenland inshore

Introduction

It is generally known that the North Atlantic cod (*Gadus morhua*) spawn in several of the fjords in West Greenland. However the reproductive cycle of the inshore cod population has never been documented. Knowledge about the reproductive status and the spawning characteristics of a fish population is important in management studies, e.g. spawning probabilities (Tomkiewicz *et al.*, 2003), minimum size at capture to allow for reproduction and timing of reproduction (Morrison, 1990). Today about 90 % of the total cod fishery in Greenland waters are taken inshore at West Greenland (Storr-Paulsen *et al.*, 2004). This dispersion in catches is a consequence of the collapse of the offshore cod population since the 1990s (Hovgård & Wieland, 2008). To ensure a sustainable utilization of inshore cod stocks a proper management should be based on knowledge of spawning characteristics.

For standardization, maturity scales have been developed dividing the reproductive development of the cod into reproductive phases (ICES, 2008). These, however, vary between countries. A four-scale division is used by the countries surrounding the North Sea; Norway has a six-scale division; England a five-scale division and Sweden uses Maier's eight-stage scale (ICES, 2008). The superior common features, for the divisions, are the reproductive phases *Immature*, *Maturing*, *Spawning* and *Spent*. After the ICES workshop 2007 on Maturity Staging of Cod, Whiting and Saithe (WKMSCWHS), a common six-stage maturity scale was recommended. This maturity scale in addition includes *Resting/Skip-of-Spawning* and *Abnormal*. Tomkiewicz *et al.* (2003) has developed a ten-stage maturity scale based on Baltic cod and revised from Maier's eight-stage scale from 1908. The ten-stage scale uses the same superior phases as the ICES six-stage scale, but subdivides them into even more specific developmental steps (see specifications in *Materials and methods*).

The reproductive strategy for female North Atlantic cod is iteroparous with determinate fecundity, and group-synchronous batch spawning. Spermatogenesis is more asynchronous often with primary- and secondary spermatocytes (SC1 and SC2),

spermatids (ST) and spermatozoa (SZ) all present at the same time, although the development amongst the cells within individual cysts always is synchronous (Rideout & Burton, 2000 and Dahle *et al.*, 2003). Female cod spawn approximately 15 times in one spawning season with 2 – 3 days separation between batches (Holm *et al.*, 1991). Accordingly the mean length of the spawning period for individual females is about one to three months. The males spawn continuously to ensure fertilization of all ovulated oocytes (ICES, 2008).

There is some but not much variation in the time of spawning and the age of maturity between the different spawning areas of the northern North Atlantic cod. In West Greenland the inshore spawning of North Atlantic cod populations is documented for several fjords and coastal areas predominately between 62˚N and 66˚N (Hovgård & Wieland, 2008). The most intense spawning is observed in small, shallow-water fjord branches (Hovgård, 1993 and Smidt, 1979) where ambient temperatures range between 0.5 – 4.0˚C (Hansen, 1949). Besides in the Nuuk area Storr-Paulsen *et al.* (2004) also identifies a major spawning ground in the Sisimiut area in southern part of NAFO division 1B, and also sporadic spawning in the Disko Bay (69˚N). Table 1 gives an overview of the spawning periods (see also figure 1) along with age and length at 50 % spawning, for different areas in the North Atlantic including this study (grayish row). In general the North Atlantic cod spawn in the spring (Brander, 1993), but the East Atlantic populations starts slightly earlier than the West Atlantic populations. This coincides with spring arriving earlier in the East Atlantic due to the warm North Atlantic Current. Most cod eggs develop between 2 and 7˚C, but exceptions occur at both cooler and warmer temperatures (ICES, 2005). The age of maturity has more or less decreased in most areas during the last decades (ICES, 2005 – *and ref. therein*), and in general the age at fifty percent maturity is approximately five to eight years old with the exception of the North Sea North Atlantic cod which is mature already after its second year.

Table 1 Spawning period, main spawning and age and length at 50 % maturity for different areas of the North Atlantic. The grayish row is based on this study.

Geographic site of cod population	In-/Offshore	Spawning period	Peak spawning	Age and length of 50 % maturity
Eastern Baltic	Brackish	March to September	June & July	
Western Baltic	Brackish	January to May	February & April	
Southern part of North Sea	Off	January to April	Late January to February	2 - 3 yr & 50 – 75 cm
Northern part of North Sea	Off		February & March	
Norwegian coast (Arcto-)	Off	February to May	Late March early April	Changing
North Gulf of St. Lawrence	Off	April to June	May	5.4 yr & 46.8 cm
Iceland	Off	March to May	April	♀ 6.6 yr & 75.6 cm ♂ 5.8 yr & 67.2 cm
East Greenland	Off	March to July	April	8 yr
West Greenland	Off	March to June	Mid April	6 – 7 yr (in 1960-1983)^
West Greenland	In	February to July	March & April	5 – 6 yr (in 1930s)*
West Greenland	In	March/April to July	May & June	♀ 4 – 5 yr & 50 cm ♂ 3 – 4 yr & 40 cm

Information based on tables from ICES (2005)
* Hansen (1949)
^Wieland & Hovgård (2002)

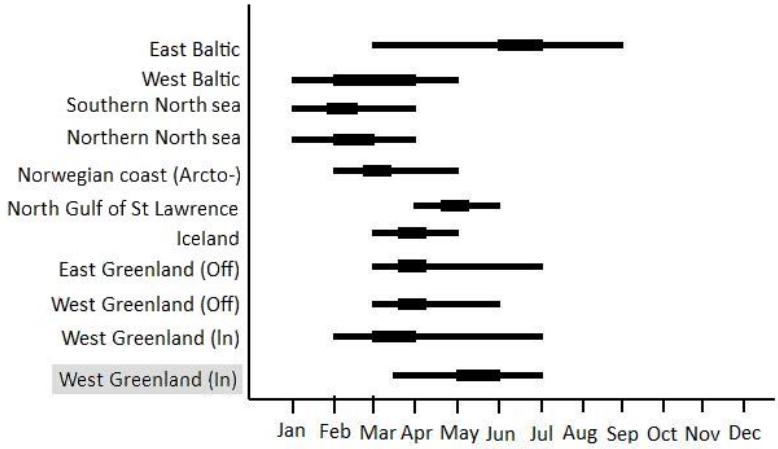

Figure 1 Length of spawning periods, including the main periods (thick lines), of different North Atlantic cod populations, according to information given in table 1.

This study has resulted in a maturity manual for staging the reproductive status and identification of the reproductive development for both male and female cod, specifically for the inshore North Atlantic cod in West Greenland (appendix). In this relation, the gametogenesis together with the developmental stages and spawning phenology have been studied. This work can be used as basis for further studies of the inshore spawning in West Greenland, e.g. the extension and distribution of the inshore spawning together with factors affecting the reproductive development e.g. excessive fishery and climate changes.

Materials and methods

Sampling of cod

A total of 1112 cod (529 females and 583 males) were sampled in collaboration with Greenland Institute of Natural Resources from December 2007 to November 2008 (Table 2); most intensively from February to June 2008 with a maximum of three weeks between samplings. The sampling was conducted in different inlets of the Nuuk Fjord system (Figure 2) using longlines with 90 hooks and jigs. The longlines were set at depths of 70 to 100 meters for two to three hours using chumps of squid as bait.

Table 2 Sampling information – Location of sampling sites can be seen in figure 2.

Date	Place	Field code	NAFO ICES	Pos. N deg.	Pos. N min.	Pos. W deg.	Pos. W min.	Sample size$_T$	L_T span cm	W_T span kg	Mean *HSI*
14-12-2007	Kapisillit	JG031	1C	64	24	50	14	64	35-82	0.42-5.30	6.00
28-01-2008	Kapisillit	JG031	1C					49	37-84	0.50-6.70	5.42
05-02-2008	Qurqut	JE029	1D	64	13	50	57	61	39-71	0.57-4.00	4.57
21-02-2008	Qurqut	JE029	1D	64	18	50	57	43	34-69	0.33-4.12	3.99
11-03-2008	Qurqut	JE029	1D	64	13	50	57	64	32-57	0.23-1.99	4.68
26-03-2008	Qurqut	JE029	1D	64	13	50	59	68	33-64	0.32-2.79	5.51
01-04-2008	Amitsuarssut	JH030	1C	64	30	50	36	31	34-57	0.38-1.88	5.92
01-04-2008	Kapisillit	JG031	1C	64	26	50	18	107	29-75	0.22-3.62	4.56
15-04-2008	Qurqut	JE029	1D	64	13	50	57	75	33-77	0.32-5.96	6.56
23-04-2008	Ameralik	JF031	1C	64	19	50	26	50	31-74	0.26-3.92	4.50
24-04-2008	Qugssuk	JK028	1C	64	48	51	7	2	44-49	0.96	5.26
24-04-2008	Terte	JG026	1C	64	26	51	34	8	36-60	0.36-2.20	4.63
24-04-2008	Qarusuk	JF027	1C	64	21	51	23	30	31-49	0.24-1.26	5.89
30-04-2008	Kapisillit	JG031	1C	64	26	50	23	29	52-67	1.38-3.02	6.27
08-05-2008	Kapisillit	JG031	1C					63	32-81	0.34-5.84	5.79
17-05-2008	Ameralik	JF031	1C					39	40-104	0.76-13.7	5.11
22-05-2008	Kapisillit	JG031	1C					38	38-100	0.54-10.2	6.98
25-05-2008	Ameralik	JF031	1C					22	59-107	2.02-13.1	5.23
10-06-2008	Amitsuarssut	JH030	1C					25	34-50	0.46-1.40	6.08
12-06-2008	Buksefjorden							20	59-74	2.02-4.48	2.96
23-06-2008	Qorqut	JE029						23	33-50	0.29-1.15	4.14
24-06-2008								46	28-57	0.17-2.02	5.08
26-06-2008								33	37-59	0.40-1.77	4.66
11-07-2008	Ameralik	JF031	1C					2	54-58	1.36-1.65	5.26
18-07-2008	Kobbefjorden							29	35-47	0.38-1.00	3.46
05-11-2008	Kobbefjorden							91	36-62	0.10-2.40	4.57
Total								**1112**	**28-107**	**0.10-13.7**	**5.11**

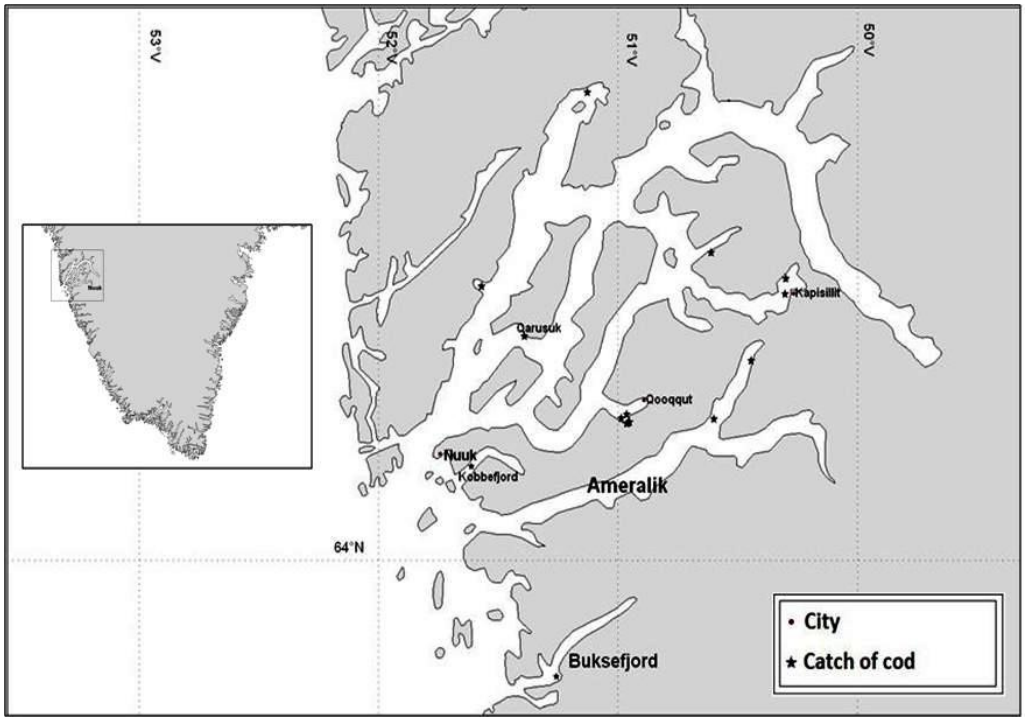

Figure 2 The Nuuk Fjord system, with sampling sites.

Data collection

For each specimen the following was recorded: Sex; total length (L_T, cm) to the nearest cm below; total weight (W_T, kg); gutted weight (W_{CL}, kg); weight of the gonad (W_{GO}, kg); weight of liver (W_{LI}, kg) and the stage of maturity, using the ten-stage classification scale (Tomkiewicz *et al.*, 2002 and 2003) (appendix). All weights were measured to the nearest 0.1 g. The saggital otoliths were removed for age determination. Data from August to October have not yet been processed. For histological verification of the macroscopic classifications, a tissue section from gonads randomly sub sampled (approximately 5 individuals per 10 cm length group per sex) was preserved. This was conducted between December 2007 and June 2008. The subsamples were selected and handled according to the procedure described below.

Sample preparation

A photograph of the whole gonad as well as a close up, and with an *id* tag of the fish, was taken of all the specimens, males and females, selected for histological sampling. Gonad tissue was preserved in histoformaldehyde. All of smaller gonads were

preserved, whereas a transverse slice approximately 2 cm wide was taken of larger gonads. In female cod there is no significant difference between the left and the right lobe or between the anterior, middle and posterior part of the gonad (Holdway & Beamish, 1985), subsamples were therefore taken randomly. For male cod it is important to dissect a sample from both the proximal and the distal side of the testis (Figure 3) and carefully keep record of the orientation to be able to analyze the spatial difference in the development (description of gamete development below).

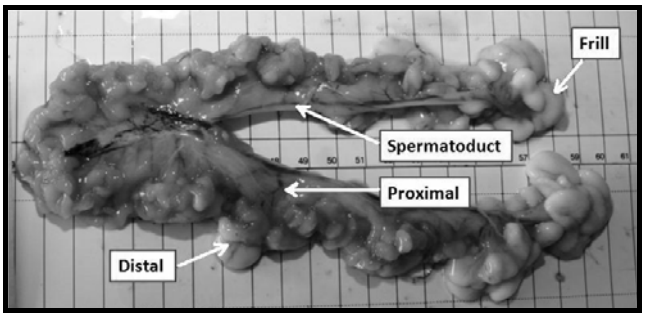

Figure 3 A male reproductive organ with the features described in the text.

Prior to analysis the samples were dehydrated in a series of increasing ethanol concentrations and imbedded in a polymerization solution. The hardened samples were sliced in 5 µm sections on a R. Jung Heidelberg microtome and mounted. Sections were stained with Mayers haematoxylin and 1 % aqueous Eosine Y. Pictures of the sections were taken with a microscope using an Infinity X Digi cam and the image program DeltaPix Viewer LE version 1.14.5 at various magnifications (x2.5, x3.2, x4.0, x10, x20 or x40).

Histological analysis

Histological sections from 32 ovaries were analyzed according to the ten-stage classification scale developed by Tomkiewicz *et al.* (2003). In addition, the studies of oocyte development by Morrison (1990) and ICES (2008) were used. For staging the reproductive status of female cod, the most developed oocytes, based on the appearance and organization of cellular structures, are used as stage indicators together with the size of the oocyte. For male cod there is no histological study of the reproductive development classified by the ten-stage scale; however the immature, maturing and spawning stages in the histological maturity scale recommended by

ICES (2008) can be divided into sub-phases providing a ten-stage classification scale for males. Samples from 17 testes were analyzed. Testes often hold germ cells of various developmental stages at the same time (Rideout & Burton, 2000), and the relative abundance of these can be used as a criterion in staging the maturity (ICES, 2008). Not all samples collected for histological analysis were sliced and analyzed; due to time limitation only a proportion assumed to represent all stages was analyzed. A schematic display of the ten stages and their relative occurrence in the reproductive cycle is shown in figure 4.

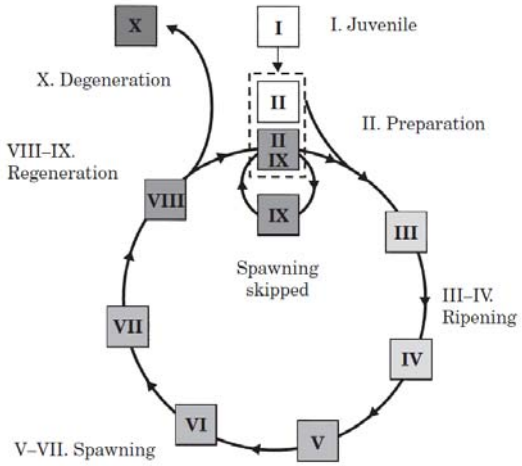

Figure 4 Schematic presentation of the reproductive cycle showing the ten maturity stages and combining them into six reproductive phases: Juvenile (**I**), Preparation (**II**), Ripening (**III** = oocyte recruitment/early spermatogenesis and **IV** = late vitellogenesis/late spermatogenesis), Spawning (**V** = initiation, **VI** = main period and **VII** = cessation), Regeneration (**VIII** = spent and **IX** = resting/spawning omission) and Degeneration (**X**). (Revised from Tomkiewicz *et al.*, 2003)

Description to identification of gamete development

Oogenesis

The ovaries consist of two nearly identical lobes, in which the reproductive tissue forms ovigerous folds that extend from the ovary wall, creating a lumen running longitudinally in the center of the lobe. Oogonia are all formed by mitosis from the primordial germ cells in the vicinity of the luminal epithelium (Kjesbu & Kryvi, 1989). The following description of oogenesis is based on descriptions by ICES (2008), Morrison (1990), Kjesbu & Kryvi (1989) and Tomkiewicz *et al.* (2003). A histological display of the cell stages is given in the appendix.

In the juvenile fish, oogonia transform into oocytes with densely staining cytoplasm and a large central nucleus with few peripheral nucleoli, this is the

commencement of oogenesis and is called the perinuclear stage (PN) (stage I). In their first growth phase the oocyte increase slightly in size and by the end of the phase a light staining circumnuclear ring forms around the nucleus and is therefore termed the circumnuclear ring stage (CNR) (stage II). The second growth phase is the initiation of oocyte maturation and is identified by the displacement of the CNR towards the periphery, where it gradually disintegrates and is replaced by the appearance of spherical and transparent cortical alveoli (CA) (stage III). The CA seal up the oocyte after it has been fertilized preventing double fertilization. Vitellogenesis (VT) starts in the second growth phase and is characterized by the appearance of yolk granules initially only in the periphery but as development progresses they fill out and expand the entire cytoplasmic area (stage IV). Accordingly the oocytes increase in size resulting in the entire ovary increasing in size as well. When the oocyte approaches final maturation the nucleus becomes irregular and migrates towards the micropyle. Simultaneously a hydration process of coalesced yolk granules results in the oocytes becoming hydrated (HYD) (stage V). These tend to lose their round shape during fixation for histological analysis. At ovulation oocytes are released into the lumen while the ruptured follicles, called the postovulatory follicles (POF), remain in the ovary (stage VI and VII). While the POFs are resorbed relatively fast, the VTs and HYDs that do not complete maturation becomes atretic (AT) and can be a disturbance to the tissue if they are numerous, rendering the ovary abnormal (stage X). In line with the HYDs being ovulated, the ovary shrinks which can be observed macroscopically, but the formation of new generations of oocytes has already started and an appearance of PNs is seen (stage VIII and IX).

Spermatogenesis

Like the female reproductive organ, the testes consist of two nearly identical lobes as well, but unlike the ovary the testes are composed of several frills (Morrison, 1990), arranged around one central collecting duct, or spermatoduct, per testis side (Figure 3). In each individual frill spermatogenesis occurs with the least developed germ cells distal in the periphery of the frill and the most advanced germ cells proximal in the

vicinity of the spermatoduct. A branching system of small efferent ducts in each frill passes into the proximal part and fuse with the collecting duct (Almeida *et al.* 2008 and Morrison, 1990). Spermatogenesis is not as well studied as oogenesis (Morrison, 1990; Rideout & Burton, 2000; Dahle *et al.*, 2003 and ICES, 2008). The following description of spermatogenesis is based on the studies by ICES (2008 – *and ref. therein*) and Morrison (1990); histological displays can be viewed in the appendix.

The early juvenile stage (stage I) is characterized by spermatogonia (SG) with an elliptical appearance and a lightly staining cytoplasm; they are primarily located in the distal part of the frills. Eventually as development progresses they lodge into cysts, and through mitotic division primary spermatocytes (SC1) that are smaller, circular and more deeply stained, appear (stage II). Due to the mitotic divisions the number of germ cells within the cysts increases and as a consequence the cysts expand accordingly. The SC1s divide further into even smaller and even more numerous secondary spermatocytes (SC2), that again divides into spermatids (ST) (stage III). This linear increase in number of germ cells can be observed macroscopically in that a remarkable expansion of the testis occurs. The next developmental step is the formation of flagella, turning the STs into flagellated spermatozoa (SZ). The number of SZ increases especially in the proximal part of the frills. Simultaneously the cysts and interstitial walls disappear, forming long tubules filled with SZ that are aligned with their flagella lying alongside each other and the heads facing the same direction (stage IV). When the SZ become ripe they are located in the efferent ducts and the spermatoducts which can be observed macroscopically (stage V – VII). As the sperm is released the tissue and the walls of the spent testes increase in thickness (Stage VII – IX), due to the contraction of the testes. There might be atretic spermatozoa without flagella left in the ducts but they will be resorbed. Mass atresia however will disturb the tissue and render the testes abnormal (stage X). In the distal part of the testes earlier developmental stages of germ cells can be found throughout the spermatogenesis.

For separation of stages containing maturing-, mature and ripe spermatozoa a set of criteria were applied in this study. Maturing SZ, primarily found in stage IV, are bound in cysts lying alongside each other, with their heads facing the interstitial tissue and their flagella aligned. Mature SZ, primarily found in stage IV and V, are no longer bound in cysts but lie strayed and disorderly in tubules. The ripe SZ, which are

primarily found in stage VI and VII, are distinguished from the mature ones by the density of them in the tubules. The ripe SZ are the ones being spawned (ICES, 2008), and therefore the density decreases in line with them being released. Table 3 gives an overview of the relative occurrence of the different cell stages in the male histology samples.

Table 3 Relative occurrences of the germ cells found in the various maturity stages in the male histology samples, subjectively determined. There are no histology data for stage II, III and X.

		Maturity stage of males									
		I	**II**	**III**	**IV**	**V**	**VI**	**VII**	**VIII**	**IX**	**X**
Cell type	**SG**	+++			++	¤	¤	++	++	+++	
	1. SC				++	+	¤	¤	¤	+	
	2. SC				++	+	¤	¤	¤	+	
	ST				++	+	+	+	+	+	
	SZ- maturing				+++	++	+	+	+	+	
	SZ- ripe				+	+++	+++	++	+	¤	
	AT							¤	¤	¤	

+++ abundant; **++** common; **+** scarce; **¤** potential.

Data analysis

To estimate the age and length of the cod that could be assumed to have spawned (stages III – VI) in the spawning season of 2008, a spawning probability function (*pS*) for age and length was calculated using individuals caught in the prespawning period (December 2007 – April 2008). Stage VII is not included in the assumed spawners in *pS*, as it is the stage of spawning cessation and is not found in the prespawning period.

$$pS = \frac{n \text{ stage III–VI}}{n \text{ stage I–X}} * 100$$

For *pS* according to length a logistic regression model was fitted to data. The hepatosomatic index (*HSI*) and the condition factor (*K*) were calculated for stage III – VII for both sexes to determine the allocation of energy throughout the year in all the ripening and spawning stages.

$$HSI = \frac{W_{LI}}{W_{CL}} * 100 \qquad K = \frac{W_T}{L_T^3} * 100$$

The gonadosomatic index (*GSI*) was calculated for all stages to help determine the gonadal development.

$$GSI = \frac{W_{GO}}{W_{CL}} * 100$$

SPSS version 17.0 Explore analysis was used for descriptive statistics and means were calculated for *HSI*, *K* and *GSI*. To analyze if there is a difference in length according to age between the sexes the following tests was used; *Kruskall-Wallis* to test for any effect between age and length, *Mann-Whitney* to test for any difference between the sexes and for any difference between the single ages.

RESULTS

Stage determination

For females the 32 ovaries selected for histological analysis were macroscopically determined as shown in table 4; 13 ovaries (40.6 %) were reclassified after histological analysis with all but one (a stage II reclassified as IX) entering a contiguous stage. Stage I and III had the highest occurrence of errors in the macroscopic classifications with more than half being reclassified. Stage VI and IX were all correctly determined macroscopically and stage II had a relatively low percentage of error (20 %). December and March had an error of ≥ 50 %, but December is based on only one specimen. The month with the lowest occurrence of misclassifications is April (33 % error).

The sample size for histological verification of males was 17 testes and 8 of them (47 %) were reclassified (Table 5). All samples macroscopically determined as stage II and III were reclassified histological as spent stages (VIII or IX). The stages I, IV, V and VII were all correctly determined macroscopically. No samples were staged as IX macroscopically, but three samples were reclassified as a stage IX. Just as for

the females the month with the highest error in stage determination of males was March (67%) and the month with the lowest was April (43 %).

Table 4 Macroscopic vs. histological determination of maturity in ovaries and error distribution according to month.

		Maturity determined histological												Error distribution (%)						
		I	II	III	IV	V	VI	VII	VIII	IX	X	Total	Error (%)	Dec*	Jan	Feb	Mar	Apr	May	Jun
Maturity determined macroscopic	I	**0**	4									4	100							
	II		**4**							1		5	20			33	50		7	
	III		1	**1**	1							3	67			0				
	IV				**1**	1						2	50	100		11				
	V				1	**2**	1					4	50					0	7	
	VI						**3**					3	0					33	0	
	VII							**1**	1			2	50						0	
	VIII								**2**	2		4	50						7	
	IX									**5**		5	0						21	0
	X										**0**	0	0							
	Total	0	9	1	3	3	4	1	3	8	0	32		100		44	50	33	42	0

* 2007

Table 5 Macroscopic vs. histological determination of maturity in testes and error distribution according to month.

		Maturity determined histological												Error distribution (%)						
		I	II	III	IV	V	VI	VII	VIII	IX	X	Total	Error (%)	Dec*	Jan	Feb	Mar	Apr	May	Jun
Maturity determined macroscopic	I	**1**										1	0				0			
	II		**0**						1	1		2	100				33	14		
	III			**0**					2			2	100				33		17	
	IV				**1**							1	0	0						
	V					**2**						2	0					0		
	VI				1	1	**2**					4	50					29		
	VII							**3**				3	0						0	
	VIII								**0**	2		2	100						33	
	IX									**0**		0	0							
	X										**0**	0	0							
	Total	1	0	0	2	3	2	3	3	3	0	17		0			67	43	50	

* 2007

Temporal occurrence of reproductive stages

The majority of cod was caught from February to June with the highest total catch in April, 127 females (24.0 %) and 205 males (35.2 %). There is a clear progression temporally in observations of the different stages, especially for the ripening, spawning and spent stages (III – IX) (Table 6). Stage III has a sporadic appearance in only a few months but stage IV is found in every month from December to May for both sexes. Stage V and VI for females are first seen from April but they are not dominant until May with 20.5 % and 21.7 % respectively. By the end of May there are no more occurrences of females in stage V and only one female stage VI was caught in June. For males however the occurrence of stage V is observed as early as December although not numerously; the peak of appearances is from February to April. In comparison stage VI for males is slightly staggered and is primarily observed

from April to June. Stage VII and VIII for females are only seen in May and June, stage IX is additionally seen in July and November. Males in stage VII were observed in February, May, June and November. Stage VIII and IX for males occurred from March to July and in November but with missing occurrences in April (VIII) and July (IX). Data from August to October is missing but there were empirical observations of females in stage VII from July and of males in stage VII from August, September and some in October. The juvenile stages (I and II) are found more or less throughout the sampling period with the exception of a few months for both sexes.

Table 6 Percent of sampled maturity stages for male and female cod per month. Estimations are in percent of a particular stage from a particular month. Stages validated histological are corrected if they were wrongly classified macroscopic. Data from August to October 2008 is missing.

		Sampled gonads (%)																					
		Dec.*		Jan.		Feb.		Mar.		Apr.		May		Jun.		Jul.		Aug. – Oct.	Nov.		Tot. (n)		Tot. (%)	
		♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	♀	♂	-	♀	♂	♀	♂	♀	♂
Maturity Stage	I	2.9	13.3	4.2	12.0	13.7	17.0	30.5	28.8	33.9	22.9	9.6	13.9	64,1	47,8	93.3	87.5	-			**142**	**142**	26,8	24,4
	II	47.1	3.3	58.3		43.1	3.8	54.2	8.2	31.5	9.8	10.8	6.3	1,3				-	44.8	3.0	**160**	**35**	30,2	6,0
	III	38.2	33.3	33.3		7.8				9.4	0.5							-		3.0	**37**	**12**	7,0	2,1
	IV	11.8	46.7	4.2	72.0	35.3	11.3	15.3	21.9	16.5	3.4	6.0	2.5	1,3				-			**59**	**63**	11,2	10,8
	V		3.3		16.0		64.2		32.9	7.9	35.6	20.5	1.3					-			**27**	**137**	5,1	23,5
	VI						1.9		4.1	0.8	27.3	21.7	51.9	1,3	2,9			-			**20**	**103**	3,8	17,7
	VII						1.9					18.1	13.9	1,3	11,6			-		30.3	**16**	**30**	3,0	5,1
	VIII								2.7			7.2	2.5	1,3	13,0		12.5	-		3.0	**7**	**16**	1,3	2,7
	IX								1.4		0.5	6.0	6.3	29,5	24,6	6.7		-	55.2	60.6	**61**	**44**	11,5	7,5
	X												1.3					-			**0**	**1**	0,0	0,2
	Tot. (n)	**34**	**30**	**24**	**25**	**51**	**53**	**59**	**73**	**127**	**205**	**83**	**79**	**78**	**69**	**15**	**16**	**-**	**58**	**33**	**529**	**583**	100	100
	Tot. (%)	6.4	5.1	4.5	4.3	9.1	9.9	11.2	12.5	24.0	35.2	15.7	13.6	14.7	11.8	2.8	2.7	-	11.0	5.7	100	100		

*2007.

The time when the spawning stages (V, VI and to some extend VII) are observed is not the same for male and female cod (Table 6); the females have a narrow period of three months (April – June) where spawning specimens are seen. But as mentioned females in stage VII were observed until July indicating that some spawning still goes on at this time although to a lesser extent. Regarding the males, some of them reach the stage of spawning as early as December and they are observed until June; they are succeeded by stage VII which is seen as late as November. In the field, however, it was not until late march that freely running milt was observed.

Age and length at maturity

The age distribution of all the sampled cod is shown in figure 5A. The age span is from two to eleven years but only one cod at age two, nine, ten and eleven respectively was caught among both males and females. By far the most dominant age group is

the four year olds (year class 2004) for both males and females representing more than half of the total sample (58 % and 57 % respectively). When focusing only on ripening specimens (III and IV), possible first time spawners, the youngest individuals are in their third year although not many (Figure 5B). The majority is once again the four year olds for both sexes, females representing 48 % and males 65 %.

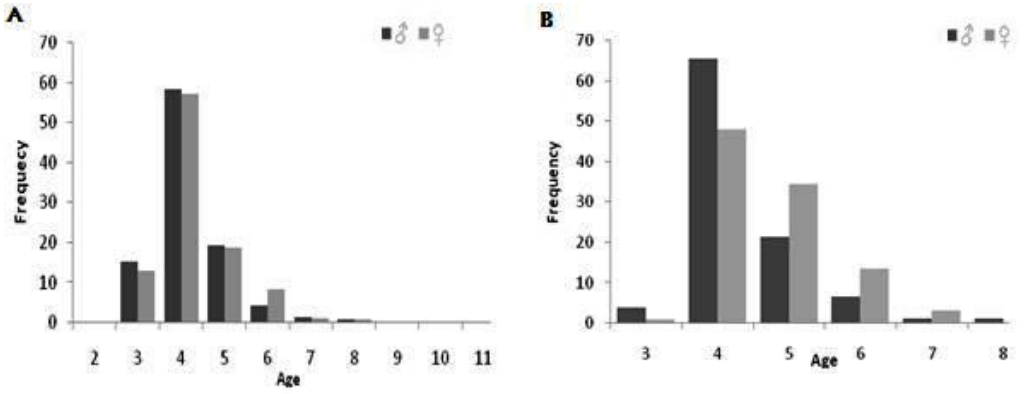

Figure 5 Age distribution of female and male cod. **A** is the age distribution of the whole sample. **B** is the age distribution of only ripening individuals (stage III and IV).

Combining both sexes there is a significant effect of age on length (*Kruskall-Wallis*, χ^2_9 = 806.737, p < 0.0005). When the difference in mean length between sexes for all ages is tested, there is also a significant difference with females in general being larger than the males (*Mann-Whitney*, z = 2.161, p = 0.031), however, when the difference between the sexes for each age is tested, there is no difference in mean value (*Mann-Whitney*, z = 0.125 – 1.902, p > 0.05) except at age six (*Mann-Whitney*, z = 3.172, p < 0.05) (Figure 6). Figure 6 showing mean length according to age, implies a difference in length at age ten and eleven but both ages are only represented by two fish, one male and one female, and can therefore not be tested. Both sexes have a mean length of about 43 cm at the age of four. The spawning probability (*pS*) in terms of length (based on logistic regression) and age is generally lower for the females than for the males (Figure 7A-B) indicating that the females are larger ($pS_{L\text{-}50}$ = 50 cm, p ≤ 0.0005) and older ($pS_{A\text{-}50}$ = 4 – 5 years) than the males ($pS_{L\text{-}50}$ = 40 cm, p ≤ 0.0005 and $pS_{A\text{-}50}$ = 3 – 4 years) when they start preparing to spawn.

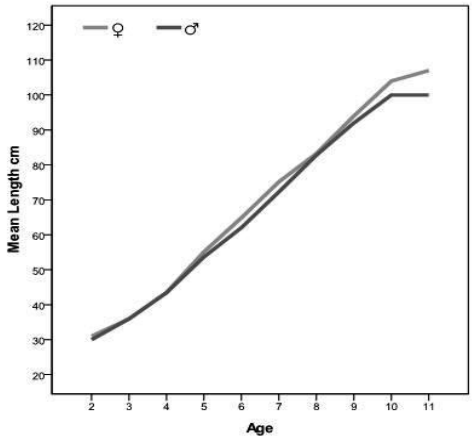

Figure 6 Mean length according to age for males and females.

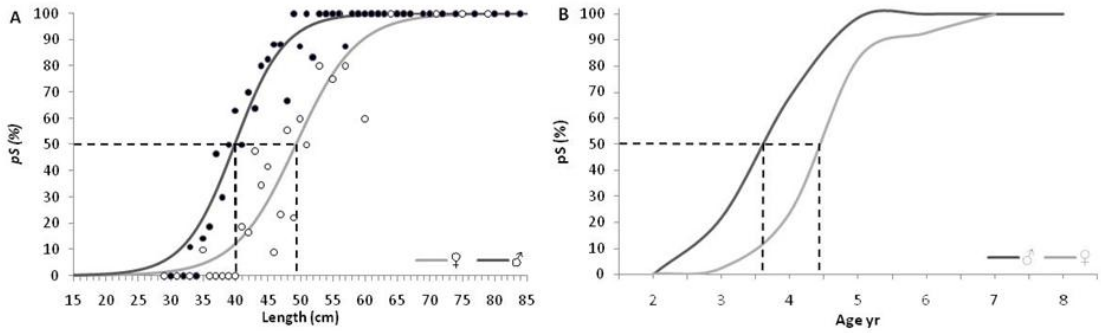

Figure 7 Spawning probability for males and females in maturing and spawning stages (III - VI) in the prespawning period (Dec. 2007 - Apr. 2008). **A** is *pS* according to length, with lines based on logistic regression ($p \leq 0.0005$). **B** is *pS* according to age.

All the males are in the ripening or spawning stage (III-VI) when they have reached the length of 60 cm and age between five and six, in comparison the females first reach a total spawning population from 75 cm and the age of seven. Table 7 presents the ranges for the weight of gonad, clean fish weight and total fish length according to the reproductive stages. The gonads of both males and females are small when the fish is juvenile but as the fish reaches the reproductive population the gonad enlarges as it becomes mature and ready for spawning and subsequently it decreases in weight as the fish ceases to spawn and becomes spent. The length and weight ranges are similar for cod in the maturing, spawning and spent stages (III – IX) as they will go through all the stages every year unless they skip spawning or the gonad becomes abnormal.

Table 7 Range of length (L_T), weight of gonad (W_{GO}) and clean weight (W_{CL}) according to stage of maturity for female and male cod respectively. See appendix.

Stage of maturity		W_{GO} (g) ♀	W_{GO} (g) ♂	W_{CL} (g) ♀	W_{CL} (g) ♂	L_T (cm) ♀	L_T (cm) ♂
	I	< 12	< 3	200 – 1710	200 – 1500	28 – 58	30 – 56
	II	< 12	< 3	340 – 2780	320 – 1280	34 – 69	34 – 52
	III	12 – 159	3 – 88	> 400	> 360	> 41	> 36
	IV	12 – 328	14 – 66				
	V	139 – 1180	14 – 1200				
	VI	163 – 2260	14 – 619				
	VII	19 – 219	4 – 244				
	VIII	6 – 306	1 – 35				
	IX	2 – 87	< 26				
	X	-	-				

Annual fluctuations in energy

From the mean hepatosomatic index (*HSI*) for males and females in the maturing and spawning stages (III – VII) (Figure 8A), it is apparent that the males have a relatively lower amount of energy stored in the liver ($HSI_{\cdot}$ = 3.9 – 5.4) than the females (*HSI* = 4.6 – 7.7). Additionally, the variation in mean liver energy content for the males is not as great ($HSI_{var.}$ = 1.5) as for the females ($HSI_{var.}$ = 3.1). The amount of energy stored in the liver more or less follows the same yearly trend for both sexes; the mean *HSI* decreases from January to February for males (5.4 – 3.9, $HSI_{var.}$ = 1.5) and from January to March for the females (7.7 – 5.4, $HSI_{var.}$ = 2.3), and then after a small increase yet another decrease is seen from April to June for both sexes (HSI_{males} = 4.7 – 3.9, $HSI_{var.}$ = 0.8) and ($HSI_{females}$ = 6.6 – 4.6, $HSI_{var.}$ = 2.0) with the greatest decrease from May to June. The oscillations in mean liver energy content are greater for the females than for the males. There is no data from July to November for the females and from July to October for the males in the stages III – VII.

There is not much variation in the condition factor (*K*) for either female ($K_{\cdot}$ = 0.98 – 1.09) or male ($K_{\cdot}$ = 0.96 – 1.07) maturing and spawning cod (Figure 8B). The mean value for males is somewhat constant from December to May (between 1.02 and 1.06) but from May to June a small decrease is apparent (1.05 – 0.96). Data for males is missing from July to October but in November mean *K* has returned to winter and spring values. For females the variation in mean *K* through winter and spring lies between 0.99 (February) and 1.09 (May) with a relatively large decrease from January to February (1.08 – 0.99). Just as for the males a decrease in condition

factor is seen from May to June (1.09 – 0.98) for the females. There is no data of maturing and spawning females from July to November.

The mean *GSI* (Figure 9A) for both sexes increases from the time when they enter the maturing stages (III and IV). For males the curve flattens as they reach the spawning stages (V and VI). The females, however, keep increasing in mean *GSI* until they enter the main spawning stage (VI). After stage VI *GSI* decreases in both sexes as the gonads become spent (VIII and IX) although it does not descent to the values of the juvenile stages (I and II) (stage $II_{females}$ = 0.56 and stage $IX_{females}$ = 1.14, stage II_{males} = 0.17 and stage IX_{males} = 0.22). The single male found in stage X had a mean *GSI* = 0.27.

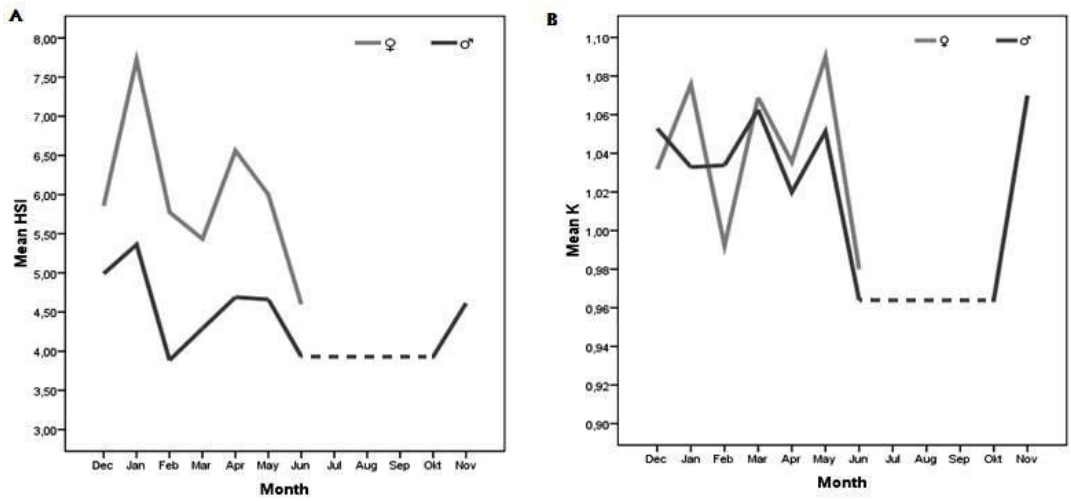

Figure 8 Mean hepatosomatic index **A** and mean condition **B** by month for maturing and spawning males and females (stages III-VII). Note December is from 2007 and data is missing from July to October 2008. There are no occurrences of females in November. Outliers, more than 1.5 box length from the first and third interquartile, are removed.

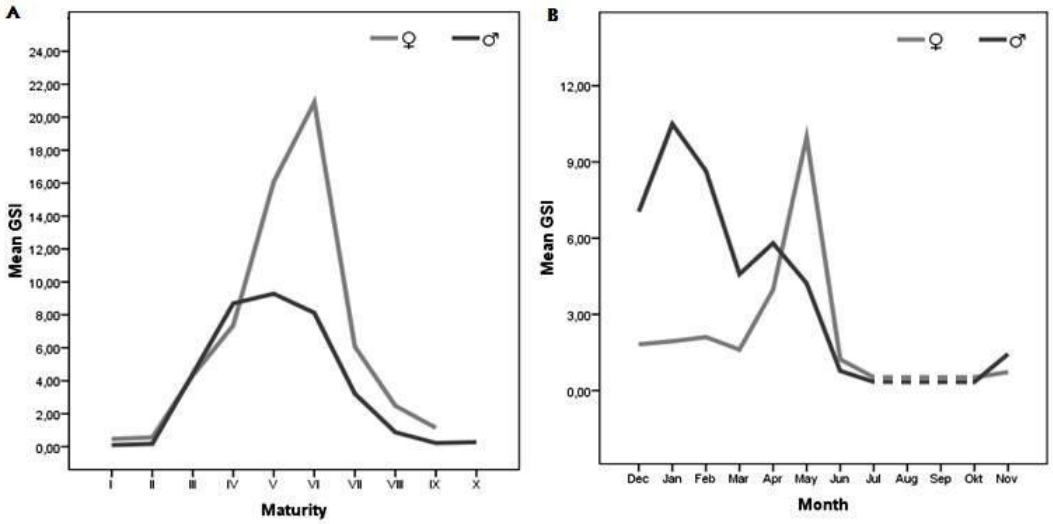

Figure 9 Mean gonadosomatic index according to maturity stage **A** and month **B** for male and female cod. Note December is 2007 and data is missing from August to October 2008. Outliers, more than 1.5 box length from the first and third interquartile, are removed.

When the gonadosomatic index is viewed according to the yearly variation, it is apparent that males and females do not follow the same trend (Figure 9B); the *GSI* for female cod peak in May but the male *GSI* peak already in January. Females have a rather stable mean *GSI* during winter and early spring (between 1.6 and 2.1) but from March to May it increases rapidly (1.6 – 10.0). From May to July a rapid and great decrease is observed (10.0 – 0.5). The male mean *GSI* decreases in two steps; firstly, a decrease is observed from January to March (10.5 – 4.6), and a second decrease is observed from April to July (5.8 – 0.3).

Spawning grounds

Mature and spawning females (stage V-VII) were observed in Ameralik, Buksefjorden, Kapisillit and a single female of stage V was caught in Qurqut. Mature males were observed in the same inlets as the females but in contrast they were abundant in Qurqut. A few mature males were also caught in Qarusuk. In Kobbefjord only mature males in spawning cessation (stage VII) were observed. All other sampling sites (Figure 2) only yielded juvenile, maturing or spent stages. The samplings between the inlets are not consistent in either sampling time or sampling size and can therefore not be compared to one another.

Discussion

Evaluation of the reproductive divisions

In this study a division of the reproductive status was divided into the ten-stage scale developed by Tomkiewicz *et al.* (2003). This division gives a more detailed and differentiated picture of the entire reproductive development throughout the year. However this division can also be somewhat of an obstacle that is time consuming and possibly a cause of more errors in accurate stage determination. The process where the reproductive maturity stages are determined is primarily conducted on research vessels, where a part of the crew might be relatively untrained in this. The reproductive development is still very gradual, and the differences between the adjacent stages are sometimes vague, both factors could cause a possible high rate of error or a time consuming thorough investigation of the gonad. From this study and the study by Tomkiewicz *et al.* (2003), after histological verification of the macroscopic determinations, it is apparent that most errors lie within the superior phase divisions (i.e. a stage III proven to be a stage IV but both are in the maturing phase) at least for the females. Consequently, it should be evaluated, whether the best approach is to use the division into phases, which is more effective and still represents the superior reproductive stages, or the ten-stage division where all the details of the reproductive development are apparent. In this study the objective is partially to identify the temporally reproductive development and therefore the ten-stage scale was most adequate.

One aspect that none of the maturity scales applied in the North Atlantic considers, is the fact that because the females are batch spawners they are only occasionally in a running condition during the spawning stage, which is also the case for other gadoid batch spawners (ICES, 2008). This means that females switch between maturing and spawning throughout the entire spawning period. It has no effect on the estimation of spawning probability but it can have some impact on the spawning area and the time of spawning peak (ICES, 2008). As neither spawning area nor time of peak spawning is given as a precisely fix point in this study, but rather as an encirclement of an area and a period, it is assessed that the inaccuracy has no effect on the conclusions made.

Time of spawning

According to the time when spawning stages (V and VI) were first observed, the 2008 spawning for inshore North Atlantic female cod can be concluded to have started in late April. However main spawning does not occur until May and June, indicated by the great loss of both liver- (*HSI*) and somatic (*K*) energy from May. Energy is instead being invested in reproduction (Mello & Rose, 2005 and Bromley, 2003), which is also shown by Thomsen *et al.* (in prep.) In addition the gonadosomatic index (*GSI*) for females reaches its highest at this time, indicating that they are mature for spawning (Dahle *et al.*, 2003). Mature males of stage V were observed already from December 2007, but the main spawning stage (VI) was not seen until February and milt release was not observed until March. In general, male cod finish gonad development and produce their sperm before a reduction in feeding sets in during winter, saving their energy and giving them a better chance for survival. They postpone their spawning until conditions are optimal, e.g. when the temperature is right and the females become ready for reproduction as well (Rideout & Burton, 2000). This can explain why even though the mean *GSI* for male cod peak at maturity stage V the highest mean value in relation to month is already seen in January prior to spawning. By the time of spawning in May and June the mean value has dropped to about half. The time of observed milt release in the field is consistent with the observed drop in *GSI* in March. Between April and June and especially between May and June yet another drop in *GSI* occurs indicating an upgrading of spawning activity in this period. As in males, females can also postpone their spawning and remain in the ripening or maturing state, until conditions are optimal (Rideout *et al.*, 2000). Even a drop of 1˚C during vitellogenesis can delay spawning with 8 – 10 days (Kjesbu, 1994). The temperature has a significant influence of egg survival and hatching and for larval development as well (Holm *et al.*, 1991 and Geffen *et al.*, 2006).

Energy allocation

Both the condition factor and the hepatosomatic index for male cod decrease from May, just as for the females, although the drop in *HSI* is not as pronounced for the males, which was also observed by Karlsen *et al*. (1995). The minor decrease in *HSI* for the males indicates that less energy is being allocated to reproduction compared to the females (Jørgensen & Fiksen, 2006). Studies by Karlsen *et al.* (1995) on Atlantic

cod and by Bromley (2003) on sole (*Solea solea*) revealed that the condition factor is slightly lower for males than for females, which was also observed in this study, indicating that males invest less energy on somatic growth as well. In addition males generally feed less than females and therefore store less energy (Fordham & Trippel, 1999). In analyzing the liver index and somatic index only maturing and spawning cod were used, as the presence of nonspawners only contribute to the stock's total liver weight without allocating any energy for reproduction, and will therefore be the source of much variation (Jørgensen & Fiksen, 2006). In addition to the energy being allocated from storage in the liver to reproduction, both male and female cod reduce their feeding activity during spawning, as a result of a decrease in appetite (Fordham & Trippel, 1999; Kjesbu *et al.*, 1991 and Lambert & Dutil, 2000). This could also be a part of the explanation for the decrease in *HSI* seen from May to June in this study. During winter when the amount of prey items is scarce, the cod suffers starvation (Schwalme & Choinard, 1999) which can explain the drop in *HSI* from January to February. After February the energy level increases again until the initiation of spawning. This increase coincides with the arrival of the capelin (*Mallotus villosus*) which migrates into the fjords from December to spawn in the late spring and early summer (Friis-Rødel & Kanneworff, 2002).

Cessation of spawning

The main spawning period where both males and females spawn is May and June, with an initiating period from March for the males and from April for the females. Then follows a cessation period in July for the females, and apparently the males have some degree of spawning until fall. According to Wieland & Storr-Paulsen (ICES, 2005) the Atlantic cod living in West Greenland inshore areas spawn from February to July but with a mean in March/April. Jensen & Hansen (1931) however state that the majority of Greenlandic cod finish their spawning in May and only some continue into June.

It is difficult to set a month for cessation of spawning in this study, as data from June and July are scarce and is missing from August until October. However, neither males nor females in the main spawning stage (VI) were observed in July, indicating that not much spawning takes place at this time. Nevertheless stage VII, which is defined as the spawning cessation stage (Tomkiewicz *et al.*, 2002 and 2003),

was found in November for the males, and unhandled samples of stage VII were collected from July to October as well. The unhandled samplings also include observations of females in stage VII in July.

Age at maturation

Some individuals of both sexes are maturing at age three, but the majority is not ready until they are approximately four years old. The age at which fifty percent of the maturing and spawning population is likely to spawn, is between three and four for the males and between four and five for the females. This is early compared to the observations made by Hansen (1949) from the 1930s; here fifty percent maturity was reached by age five to six for Atlantic cod in the Nuuk and Sisimiut areas. But Hansen also discovered that during the 1930s the younger year classes matured at an earlier age than the older ones. He assigned this tendency to changes in hydrographic conditions, where the water temperature had warmed; the warmer the water the younger and smaller the cod will mature. Jørgensen (1990) found a reduction of 2.5 years from 1923 to 1976 in median age at maturity for Northeast Arctic cod. He suggests that it could be a compensatory response to a reduced stock size and that higher fishing pressure on late maturing individuals that are larger than on early maturing individuals may have contributed to the changes as well, displacing evolution towards maturity at younger age. The entire male stock in the current study is spawning between five and six years old, and the females are all spawning when they reach the age of seven. No recent information on age at maturity for inshore populations in West Greenland exists (ICES, 2005).

Size at maturation

The size classes that can be expected to represent the spawning stock are 36 cm and above for males and 41 cm and above for females for the entire sampling. However when focusing on only the maturing and spawning specimens in the prespawning period, the spawning probability for the length group where fifty percent can be assumed to reproduce is 40 cm for the males and approximately 50 cm for the females. It is more reliable to use the spawning probability function rather than a maturity ogive when estimating $Length_{50}$ and Age_{50}; the former takes into account that not all the mature fish are included in the spawning stock (i.e. stage VII-X) (ICES,

2008). Spawning probability also avoids the errors that occur due to uncertainty in differentiating between stage II and stage IX, which can appear very similar both macroscopically and histological. By only including individuals caught in the prespawning period, the representatives will be all the individuals that would have spawned in the following spawning period had they not been caught (Tomkiewicz *et al.*, 2002 and 2003). There is no point in giving length- and weight ranges of fish above stage III as these individuals henceforward yearly will go through all the stages of maturing, spawning and spent, unless they skip spawning or become abnormal (Figure 4).

It is apparent that male cod mature earlier in their development than females, as the males are both smaller and younger when they are ready to reproduce; this relation was also observed by others (Morrison, 1990; Marteinsdottir & Begg, 2002; ICES, 2005 and Jørgensen, 1990). An explanation as to why females enter the spawning stock a season later in their development than males can be that females invest more energy in maturation. The egg represents a massive cytoplasmic investment (Wootton, 1998) and therefore the females have less energy, for maintaining physiological fitness for survival when they enter vitellogenesis.

Spawning ground

Spawning cod of both sexes were found in Ameralik, Buksefjorden, Kapisillit and Qurqut; in Qarusuk and Kobbefjord only spawning males were observed. However, samples were also taken from other inlets (Figure 2) but no spawning specimens were caught. Kapisillit is the most important spawning ground in the Nuuk fjord system (Storr-Paulsen *et al.*, 2004 and Hansen, 1949). Unfortunately due to ice cover it was not possible to sample in the winter and early spring of 2008 at the Kapisillit spawning ground. Not until late spring did the ice melt and as a consequence there are no continuous data from this site. Instead Qurqut, which is also known to be a spawning ground (Smidt, 1979) and was ice free during winter and early spring, was used as the main sampling station during that period. However, no more than one spawning female was caught in the inlet; this is due to the fact that the last sampling in Qurqut was conducted in the middle of April, and mature females were only observed until ultimo April. Both ripening females and spawning males were observed in Qurqut though, indicating that spawning probably does occur there.

Histological stage determination

The analysis of the reproductive development of oocytes was rather simple and followed the descriptions from other studies (Morrison, 1990; Tomkiewicz *et al.*, 2003 and ICES, 2008). However, the mean *GSI*, especially for the main spawning stage (VI), is lower in this study compared to the study by Tomkiewicz *et al.* (2003) on Baltic cod (mean GSI = 20.9 for Greenlandic and about 30.0 for Baltic). The developmental phases of spermatogenesis are more difficult to differentiate, as the transitions are gradual and all the germ cell types can be present throughout the entire reproductive cycle. This was often the case in this study. Further it can be a challenge to distinguish the individual germ cells (e.g. SC1, SC2 and ST) from one another; a helping guideline to this challenge is provided by Rideout & Burton (2000), in form of a table with the various germ cell diameters of cod from Newfoundland. However, observations indicate that the germ cells in this study were generally larger than the table indicates. Rideout & Burton (2000) and Dahle *et al.* (2003) observed the presence of primary- and secondary spermatocytes (SC1 and SC2), spermatids (ST) and spermatozoa (SZ) at the same time, but in this study also spermatogonia (SG) were almost always present at the distal end of the testes. Due to the spatial difference in development of the germ cells in male testes it is very important to have histological samples of both the distal and proximal side, to be able to determine the accurate stages. In comparison the phases of the oocyte development are all discrete, except in the spawning stages (V – VII) where the relative abundance of vitellogenic oocytes (VT), hydrated oocytes (HYD) and postovulatory follicles (POF) determines the accurate stage. When looking at histological samples it is a good idea to also look at a picture of the actual gonad, as this will provide a combination of clues, and thereby the best foundation for the accurate stage classification (see appendix). One example is when one is wavering between a stage V, VI or VII in females, and the relative abundance of VT, HYD and POF is difficult to determine from the histological sections. The macroscopic picture can provide the conclusive evidence because the HYDs can be seen with the naked eye, and thereby the relative abundance of these is clearer. Another example, from the males, is when one is unsure whether it is a stage IV or V as they can be rather similar in terms of histology.

Macroscopically in stage V a viscous white fluid of milt is apparent in the collecting duct which has not yet been formed in stage IV.

The greatest challenge in stage determination, in both sexes, is distinguishing between stage II and stage IX as they appear very similar both macroscopic and histological. In a correspondence between members of the ICES WKMSCWHS workshop 2007, Tomkiewicz and Bucholtz specify it as follows: Outside the spawning season, where the ovaries shrink and have no visible development, repeat spawners (fish that have spawned before) in resting condition (between spawnings) and large juvenile in preparation cannot be distinguished from each other with certainty, and stage II and IX will therefore overlap (Figure 4). In the prespawning and spawning season, if a fish is identified as a repeat spawner but is not maturing, it should be classified as a stage IX *skip-of-spawning*. Fortunately, as mentioned earlier an error in determination of stage II or IX has no effect on estimating the spawning probability. To distinguish between spawning and non spawning specimens, some studies have used the wall thickness of the gonad tissue (Burton *et al.*, 1997 and Holdway & Beamish, 1985). The wall thickness of a gonad from a fish that have spawned should be thicker than in a fish which has not yet spawned. This criterion, however, is to be used with caution, as wall thickness is not uniform throughout the gonads (Morrison, 1990 and Rideout & Burton, 2000) and therefore a standard sampling position should be applied. The method is not used in this study as the histological samplings were not taken from a standard position. To distinguish more precisely between the individual germ cells, e.g. maturing-, mature- and ripe SZ, but also SC1 and SC2 an electron microscope should be used (Morrison, 1990) but this technique is very time consuming. Several studies on cod have shown that histological analysis is an effective methodology to determine sexual maturity (Saborido-Rey & Junquera, 1998 and Tomkiewicz *et al.*, 2003).

The occurrence of atretic oocytes (AT) was very low in this study, only a few VTs and HYDs that had not completed maturation and had become atretic were observed in a few females. According to Kjesbu *et al.* (1991) the intensity of ATs increase with the proportion of oocytes spawned. So the highest occurrence of ATs would be at the end of the spawning season, and as there are no histological samplings from that period in this study, the presence of ATs is limited.

In conclusion

The results presented in this study are based on only one annual cycle. There can be some variation in time of spawning between years and also variation in age and length of spawning on a scale of several years. It is requisite to conduct the samplings continuously in the future to be able to assess a general tendency. This will be governed by the Greenland Institute of Natural Resources in continuity of this study. Winter and spring 2008 was designated as extremely cold compared to previous decades in Greenland and is therefore not representative of a typical year. The consequence of the low temperatures was extensive ice cover inshore as well as offshore. And another consequence is likely to be a delay of spawning. This can only be verified with continuously yearly sampling and studying.

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