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Maturity of Greenland Halibut (*Reinhardtius hippoglossoides*) in the Fjords of Northwest Greenland.

by

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Abstract

In order to clarify the spawning of Greenland halibut in the fjords of West Greenland we performed a study on the maturity of Greenland halibut in Disko Bay. Our goals were to: 1) Describe the maturity of the female fish throughout the year by looking at the oocyte development month by month. 2) If growth in maturity was observed, then locate the time of spawning. 3) To quantify the amount of fish participating in the fjord spawning. Each month up to 60 fish bigger than 70 cm were random sampled from the commercial landings. The relation between gonad and fish weight was used to set up a gonadosomatic index. The oocyte development was followed month by month by diameter analysis. Due to sampling failure from June and onwards we were not able to give a yearly maturation cycle for the oocytes of the Greenland halibut in Disko Bay. There was no clear evidence to an overall increase in gonad weight in during the four months investigated. Gonad index indicated that most ovaries were in an immature or early maturing stage. However, when looking at the oocyte development month by month a weak trend to an increase in the oocyte size was evident with respect to the diameter range of the leading cohort. If spawning is happing in Disko Bay we believe that the most likely period for any extensive spawning is the same as in the Barents Sea namely November-January. Based on the available data we were not able to quantify the proportion of Greenland halibut in Disko Bay participating in such a spawning event.

INTRODUCTION

The Greenland halibut stocks of the Northwest Atlantic are all considered to be part of the same biological stock complex. The management of the Greenland halibut in the fjords of Greenland is based on several assumptions. One is that each of the stocks in Disko Bay, Uummannaq and Upernavik fjord are separate stock units. Tagging studies have confirmed this, as it has shown that very little intermingling exists between the fjords (Boje, 1999). Another assumption is that the fish all originate from the spawning stock in Davis Strait. Spawning seems to take place in deeper waters (approximately 800-2000 meters depths) over an extended area from Davis Strait, south of 67°N (Jensen, 1935; Jørgensen, 1997; Smidt, 1969) to south of Flemish Pass off Newfoundland (Junquera and Zamarro, 1994). The timing of the spawning is not totally understood. From the Flemish Pass area Junquera *et al*. (1999) report spawning throughout the year. From the Davis Strait and off Labrador Bowering (1983) and Jørgensen (1997) suggested a pre-spawning migration of maturing Greenland halibut in the autumn and a spawning around January to March.

It is hypothesized that the fry are transported to fjords by the prevailing current systems along the west coast of Greenland, and that no extensive spawning takes place in the fjords (Riget and Boje, 1989). Thus it is believed that Greenland halibut in the fjords of Northwest Greenland once arrived in the fjord stay there throughout their life and

do not contribute to reproduction. Nursery grounds for young Greenland halibut (ages 1-3, fish less than 45 cm long) are well known in West Greenland waters, where they are most abundant between 66°-69° North at Store Hellefiske Bank to Disko and in Disko Bay, mainly at depths of about 200 m (Riget and Boje, 1989).

Even though there are several indications that the above-mentioned assumptions do hold (e.g. Riget and Boje, 1989) only few studies have been conducted to confirm if spawning and reproduction in the fjord occur. None have been conducted on a yearly basis. Jørgensen and Boje (1994) synthesized data from more than 4 000 observations on Greenland halibut gonads sampled in February/March and August from the three main inshore areas for Greenland halibut Ilulissat, Uummannaq and Upernavik in the period 1989 – 1994. Their conclusion was that spawning only takes place sporadically and to an unknown extent in both time and space. Presence of female fish with ripe, running or spent condition was observed in the study as well as in several others. For example, observations of both ripe and spent specimens in the fjords of West Greenland are described by Smidt (1969), Boje and Riget (1988) and Jørgensen and Boje (1994). This agrees well with observations made by local fishermen of running Greenland halibut in their catches (Roepstorff and Simonsen, 2002).

At present the Greenland Institute of Natural Resources carry out sampling from the commercial fishery, including a length stratified maturity assessment (visual observation) on Greenland halibut. However, the sampling is only carried out in the summer period (July or August). In all years the far majority of the female fish are in the immature stage 1 or 2[1] (Simonsen and Boje, 2001).

Objectives

In order to clarify the spawning of Greenland halibut in the fjords of West Greenland we chose Disko Bay as the area for a case study. Our goals were to:

1) Describe the maturity of the female fish throughout the year by looking at the oocyte development month by month.
2) If development in maturity was observed, then locate the time of spawning.
3) To quantify the amount of fish participating in the fjord spawning.

METHODS

Sampling locations

Sampling was conducted in co-operation with Royal Greenland's fishplant in Ilulissat and local fishermen. The plant is the main landing site for Greenland halibut in Disko Bay. Most of the landed fish are caught on 3 fishing grounds (Fig. 2.1.1), with variable exploitation throughout the year. *Area 1*, Kangia Icefjord covers exclusively a winter fishery conducted from January to April. *Area 2* constitutes a small area (10 x10 km), in the vicinity of Ilulissat where there is a winter, spring and summer fishery. In *Area 3*, Torsukkataq Icefjord in the North-eastern part of Disko Bay, a late summer-fishery is carried out in the period July to October. The fishery is a mixture of gillnets and a longlines. In 1998 54 % of the landings were taken by longline (Simonsen and Boje, 1999).

[1] According to Riget and Boje (1989)

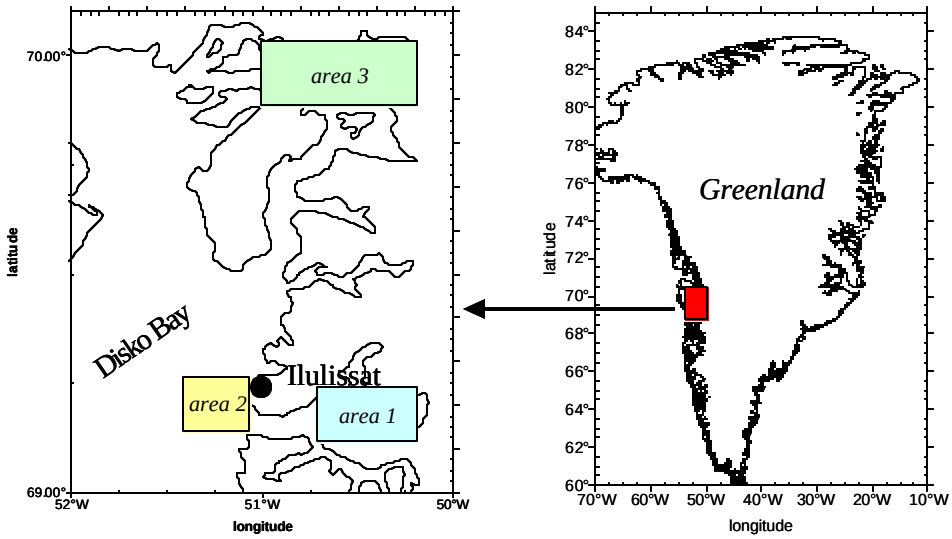

Figure 2.1.1. Map illustrating sampling locations. Area 1: Kangia Icefjord; Area 2: Ilulissat fishing ground; Area 3: Torsukkataq Icefjord.

Sampling

Sampling was originally planned to go on for a year with one monthly sampling. The plan was that in the first week in each month, up to 60 fish bigger than 70 cm, were to be sampled at random from the total landings, not discriminating between the 3 fishing areas. Unfortunately the plan including monthly sampling throughout the year was not realised. During the summer months the Greenland halibut were generally small and below the size limit of 70 cm set for sampling. In addition the late summer and autumn fishery were generally poor. Thus no sampling was carried out later than May.

A total of 298 Greenland halibut female gonads were sampled in the period February to May. The sampling took place in 1998 but for March sampling was in addition also performed in 1999, (Table 2.2.1).

Table 2.2.1. Sampling scheme. Number of samples from each month

	Month				
Year	February	March	April	May	Grand Total
1998	45	100	69	17	231
1999		67			67
Grand Total	45	167	69	17	298

Each fish was length measured (total length) and weighed. The gonads were removed, frozen and shipped to the laboratory. In the laboratory samples were defrosted, weighed and transferred to a 3.6% formaldehyde solution. The weight of the defrosted gonads was converted to fresh weight using a factor of 1.4 (Fig. 2.2.1). The samples were stored on formaldehyde for a period of 1-2 month in order to harden the oocytes. As we were focusing on mature gonads (mature 2 and above) only gonads that were heavier than 40 g were used. This included 95% of all gonads in the mature stage 2 and 100% of the gonads in more mature stages, (Fig. 2.2.2).

In the laboratory we followed the procedure described in Tuene *et al.* (2001). From each ovary a sub-sample from the middle part of the right lobe of the gonad was taken for diameter analysis. The oocytes were first separated by pipette, using fast in/out movements through pipette tips of appropriate size. The oocytes were distributed in physiological saline water on a petri-disc and measured. The oocytes were lighted to appear bright on a dark background. A Polaroid DMC 1a digital camera was mounted on a Wild MS5 microscope, and the pictures were analysed using the program NIH-image on a Power Macintosh G3.

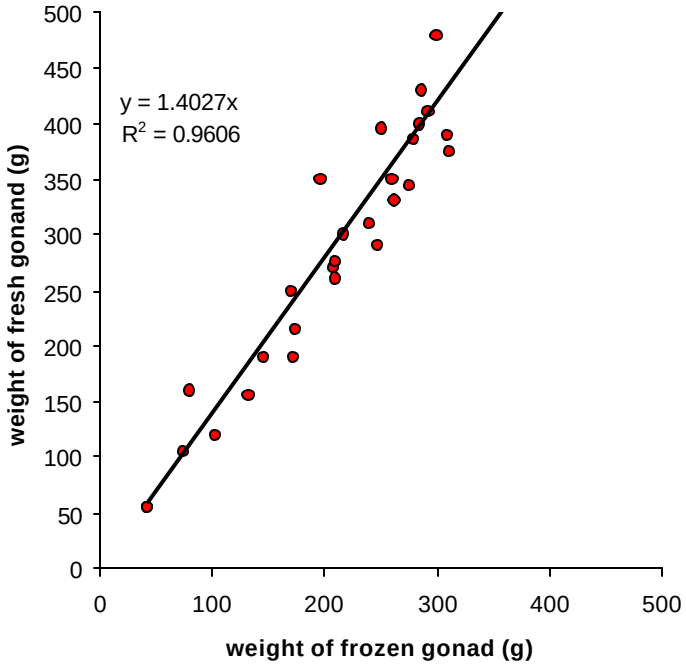

Figure 2.2.1. Comparisons of fresh and frozen weight of Greenland halibut ovaries (A. C. Gundersen, Unpublished data). Fresh weight was measured from 50 Greenland halibut gonads on the board research vessel. There were frozen, transported to the lab, defrosted and re-weighted.

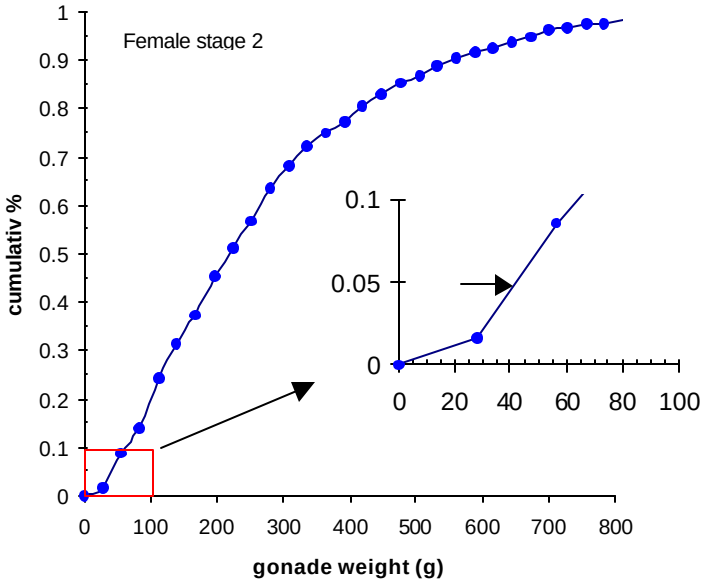

Figure 2.2.2. Gonad weight of 724 Greenland halibut gonads in GI stage 2 sampled from the NE Atlantic in 1995, 96, 97, 99 and 2000. The cumulative frequency showed that 95% of gonads in mature stage 2 weighed 40 g or more.

For each individual, the first 80 oocytes were used in the data analyses. The oocytes were counted and measured by semi-automatic procedure in image processing system (NIH Image). A minimum oocyte diameter of 300 µ and a roundness factor (A) of 1.2 were set as limits in the procedure in order to eliminate data recording of non-oocyte objects. The minimum and maximum diameter of each oocyte were measured automatically, using image analysis. Only the average oocyte diameter was used in the data analyses.

(A) $$Roundness = \frac{circumference^2}{4 \bullet p \bullet area}$$

A gonadosomatic index (GSI; B) was defined as the relation between the gonad weight and the weight of the fish:

(B) $$GSI(\%) = \frac{\text{gonad weight}}{\text{fish weight}} \bullet 100$$

RESULTS

Weight of gonads and GSI

The size of the ovaries varied considerably, ranging from 5 – 408 g. A skewed distribution was however seen, with the majority of the gonads below 100 g causing a low mean of 49 g.

The gonads from March in 1998 and 99 were not different (z-Test: Two Sample for Means, P=0.81) and were thus pooled. There was no trend to an overall increase in gonad weight during the four months investigated (Fig. 3.1.1). GSI for all months were, except for one individual, relatively low, between 0.1 and 2%, but with an increasing trend with fish size (length). The exception was the single fish from May with a GSI on 6.9 %. This was a significant higher GSI than other samples. Excluding this individual we did not find any significant difference between the sampling months February to May (ANOVA, p=0.07).

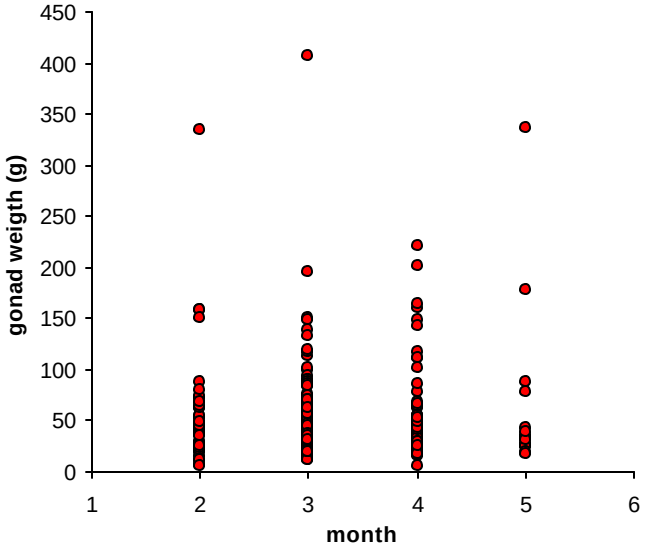

Figure 3.1.1. Gonad Weight (fresh weight corrected) of all gonads sampled monthly from February (2) to May (5).

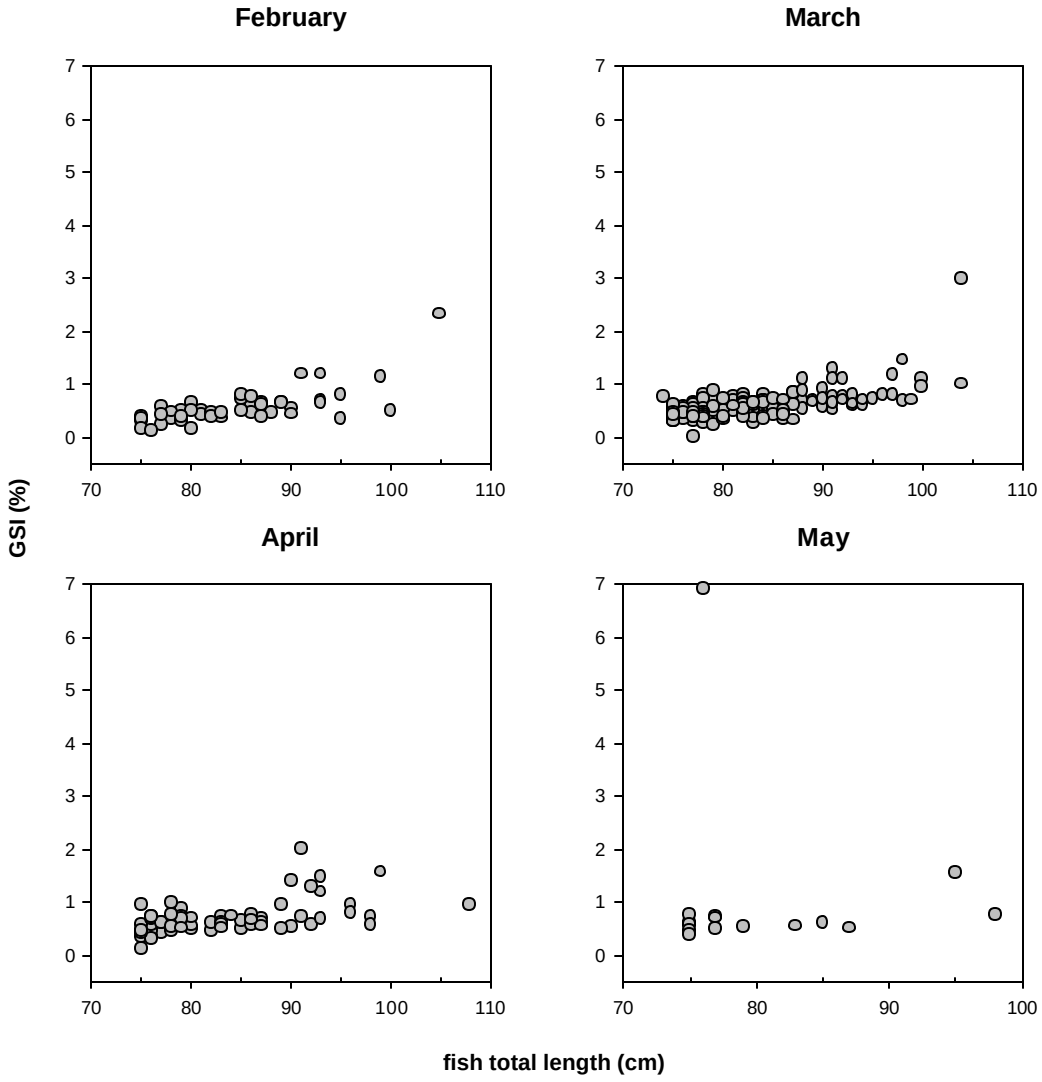

Figure 3.1.2. Gonad index (GSI) of Greenland halibut females with respect to month of sampling.

Oocyte development

In many of the smaller gonads it was difficult to separate the oocytes without damaging them. If *Zona radiata* broke the oocyte could not be used. If 80 accepted oocytes were not obtained in the first sub-sample a new sub-sample was taken and the new oocyte measurement were added. For most of the immature gonads, it was difficult to obtain enough good oocytes for analyses, and they were therefore discarded. Table 3.2.1 gives an overview of the oocyte measurements.

Table 3.2.1. Oocyte measurements for Greenland halibut ovaries collected in Disko area during February – May.

Month	Minimum (µ)	Maximum (µ)	Peak 1 (µ)	Peak 2 (µ)
February	300	1 533	400 – 600	900 – 1 000
March 98 /99	300	1 280 / 1 310	400 – 600	800 – 1 100
April	300	1 814	400 – 600	1 000 – 1 200
May	300	2 015	400 – 800	1 900 – 2 000

In February 45 fish was sampled. 9 fish met the set constrains on the oocytes size, shape and number. Most gonads had oocytes in the size range 300 to 1 400 µ with a maximum of 1 553 µ. (Fig. 3.2.1) The distribution was multi-modal, often with two peaks. In the bimodal distribution the first peak was around 400-600 µ and the second around 900-1000 µ.

Samples from March were obtained both in 1998 and 1999. 15 gonads in March 1998 and 9 in 1999 were selected for oocyte diameter analyses (Fig. 3.2.2). The majority of the oocytes were within the same size range as in February, but the maximum diameter on 1 280 and 1310 µ was somewhat smaller. Only few fish in March 1998 followed the bimodal distribution of oocytes while this distribution was more pronounced for the March 1999 samples. The upper distribution in these had a modal class around 800 to 1 100 µ.

Nine gonads from Greenland halibut caught in April were accepted for analysis. Again the oocytes followed the same distribution and size range as observed in earlier months (Fig. 3.2.3). However, a single fish did have oocytes up to 1 814 µ.

In May six gonads were analysed. Five gonads had most oocytes in the size range 300 to 1400 µ while one gonad (fish no. 18) had oocytes up to 2015 µ (Fig. 3.2.4). The distribution of the oocytes in this specimen was clearly different from any of the other gonads sampled. The bimodal distribution contained a second batch with a modal class between 1 900-2 000 µ.

Looking at the oocyte development month by month, a weak trend to an increase in oocyte size of the leading cohort was evident (Peak 2). Mean oocyte diameter with respect to month did not reveal any trend (Fig. 3.2.5).

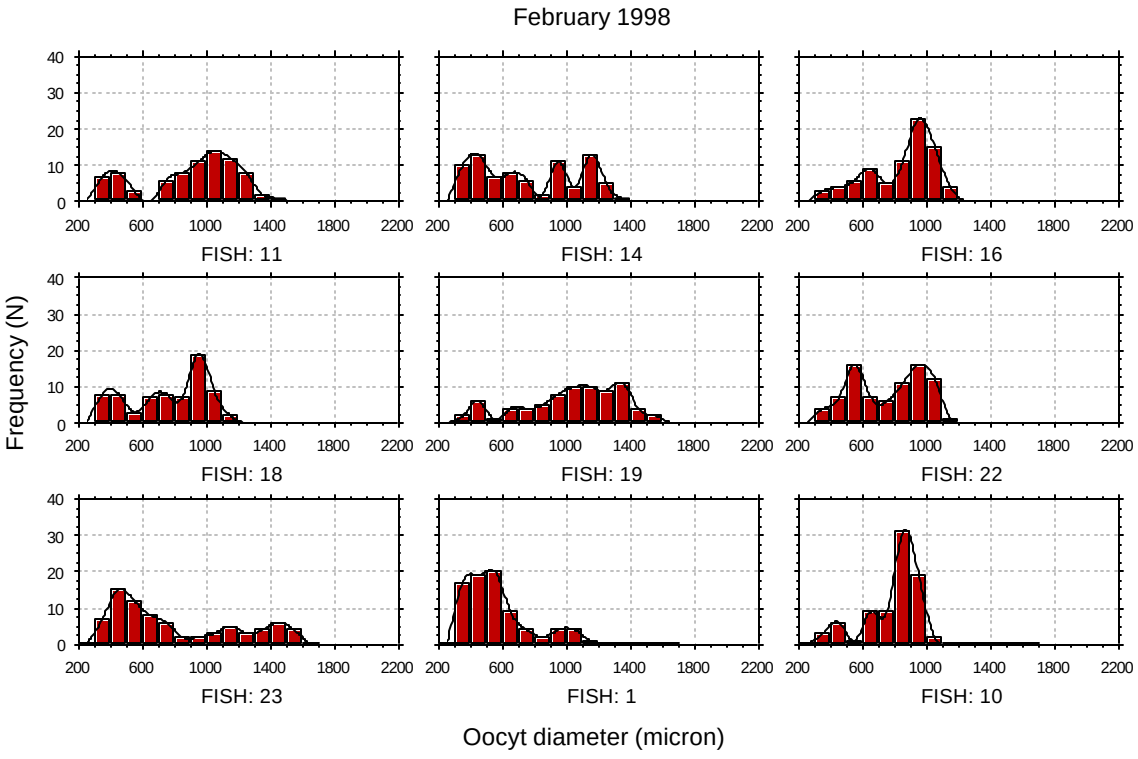

Figure 3.2.1. Oocyte diameter frequencies obtained form individuals sampled in February 1998.

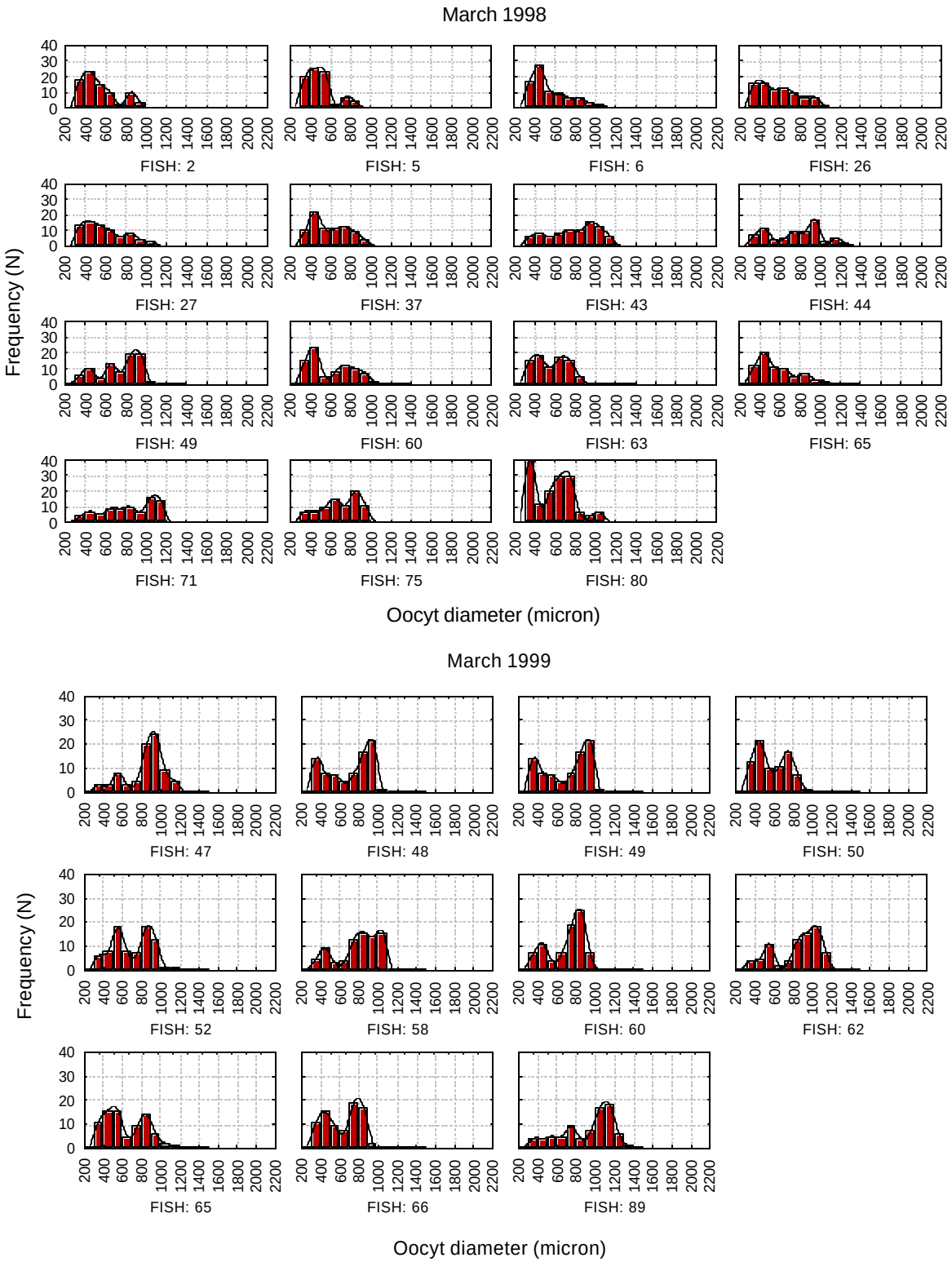

Figure 3.2.2. Oocyte diameter frequencies obtained from ovaries sampled in 1998 and 1999.

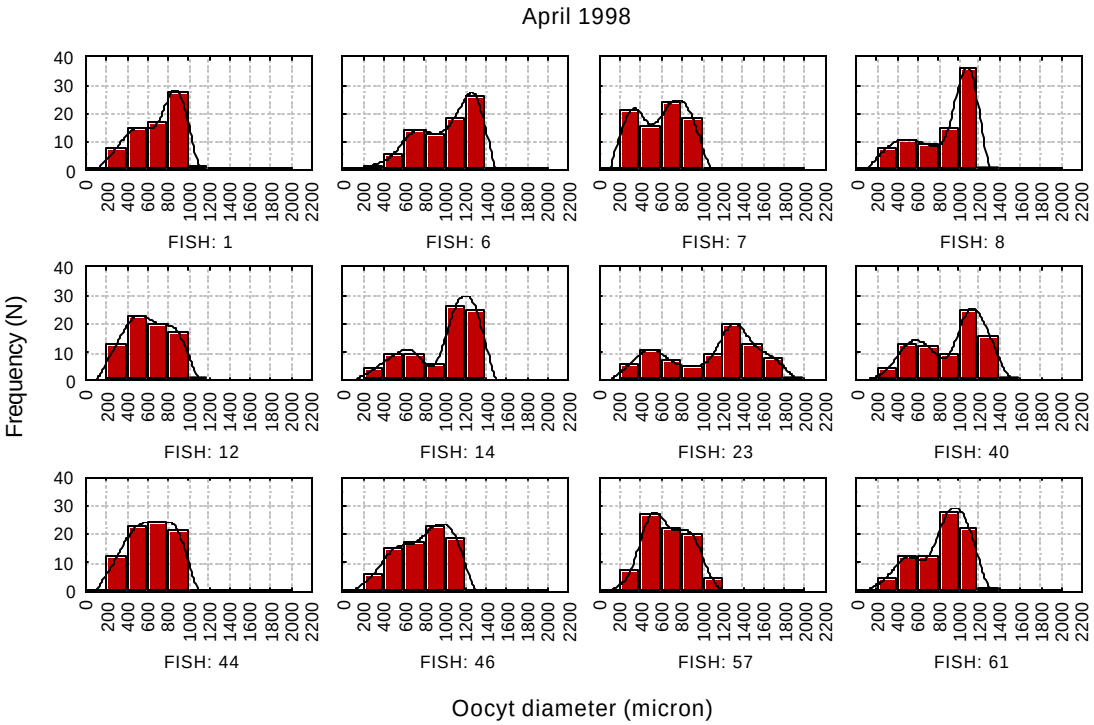

Figure 3.2.3. Oocyte diameter frequency obtained from ovaries collected in April 1998

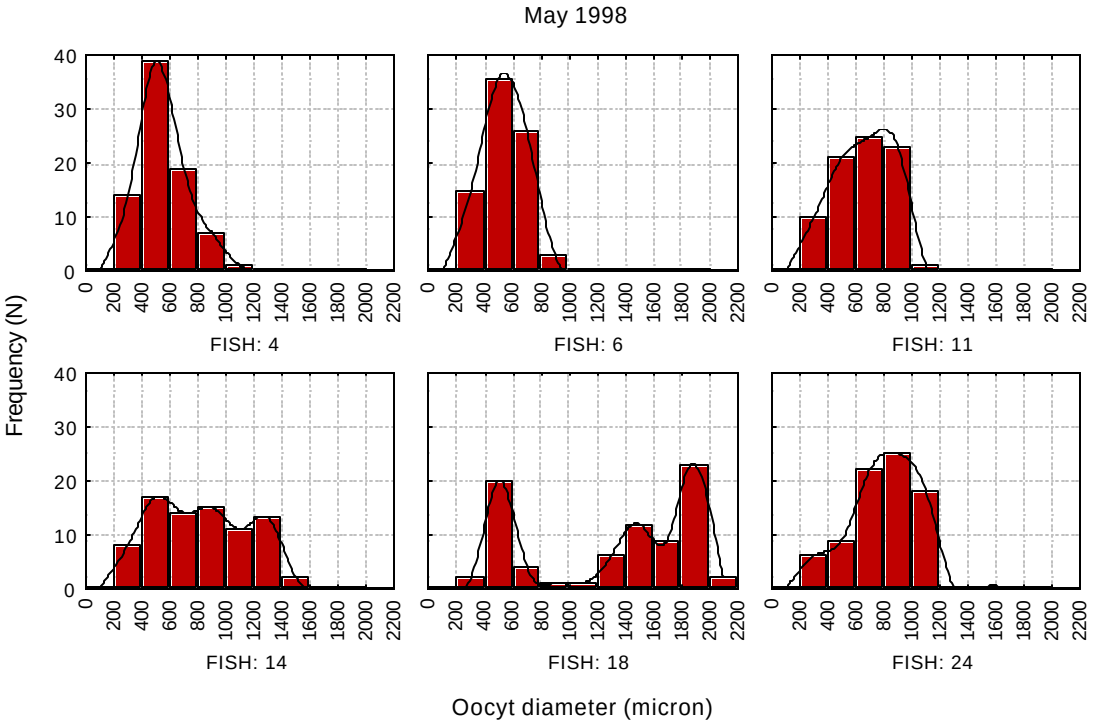

Figure 3.2.4. Oocyte diameter frequency obtained from ovaries collected in May 1998.

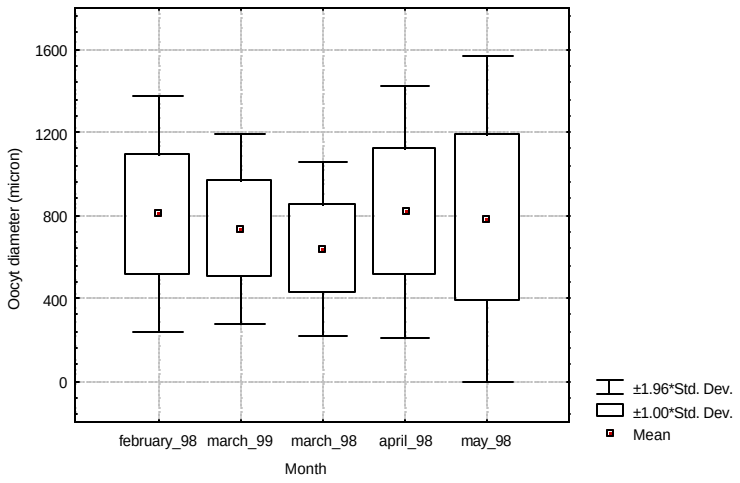

Figure 3.2.5. Mean oocyte diameter for each sampling month shown with std. dev.

DISCUSSIO N

Length range of the mother fish investigated was 70-110cm. This corresponds to the part of the stock that normally is considered as mature compared to other Greenland halibut stocks (Albert *et al*., 2001; Bowering, 1983; Gundersen, Rønneberg and Boje, 2001b; Jørgensen and Boje, 1994). One should therefore expect that the females investigated were reproductively active. The range of the gonadosomatic indices (GSI) from the investigated period was in general below 2%. During a monthly-based study on the spawning grounds of Greenland halibut in the Barents Sea (Gundersen, Kjesbu and Albert, 2001a) GSI's at this level were found in the months after spawning. In these waters there is a peak spawning in November – January (Albert *et al*., 2001) and in general GSI's were below 6% until May-June, when GSI started to increase until the next spawning. GSI below 6% were found between ovaries in maturity stage 1 (immature), 2 (early maturing), 3 (maturing) and 6 (spent) (Gundersen *et al*., 2001a).

Comparing our results to those findings, it is evident that spawning is not about to happen in the near future in the investigated areas. However, the female with a GSI of 6.9% found in May, indicate that a maturing process had begun for some fish in the population. This agrees well with the yearly maturing cycle observed in the Barents Sea (Gundersen *et al*., 2001a).

Diameter distributions indicated oocytes in an early vitellogenic phase. Looking at the oocyte development month by month, a weak trend to an increase in oocyte size was evident with respect to the diameter range of the leading cohort. In February the leading cohort peak was in the range 900 - 1 000 µ, whereas the same peak had increased to a diameter range of about 1800 – 2 000 µ in May. There is a marked increase from April to May with respect to diameter range of the leading cohort. However, our data was not unambiguous. While oocytes from all fish in February had a relatively well-defined and uniform size distribution of the leading cohort, the classification of the leading cohort was vaguer when looking at the samples from April and May. In April, we found that 10 of the 12 fish sampled had oocytes that were not different from the February samples. In May it was 4 out of 6. The remaining fish from April and May had cohorts of oocytes that clearly had increased in size since February.

Oocytes diameters measured in February and March correspond to what Gundersen *et al*. (2001a) found for the months January – April in the Barents Sea. From this, we suggest that there is a spawning in the area prior to the time of our sampling, or even overlapping with the first samples. Furthermore, in April / May a small proportion of the stock had ripening gonads with oocytes in the same size range as seen in August in the Barents Sea. Using this to forecast a spawning season in Disko Bay, the period September to November is suggested. However, a visual assessment of maturity carried out in August since late 1980'ies have found very few mature fish just one to three months prior to this suggested spawning period (unpublished results by Simonsen; Jørgensen and Boje, 1994). Thus, if spawning is happing in Disko Bay we believe that the most likely period for any extensive spawning is the same as in the Barents Sea namely November-January.

Several studies have reported Greenland halibut in spawning condition throughout the year, independent of the time of the peak spawning in the area (Morgan and Bowering, 1997). Even though it often has been few fish, it shows that Greenland halibut have complicated and not fully understood reproductive biology. Our observation of a fish with a relatively high GSI in May could be such an "outlier" that matures outside the normal maturation cycle. On the other hand this GSI level fits very well in with observations from May in the Barents Sea (Gundersen *et al*., 2001a).

Sampling gear may affect the results in a maturity study. Especially long-line catches can introduce bias, as late maturing females not are attracted to food (Engås and Løkkeborg, 1994). As mentioned earlier 54% of the landed Greenland halibut in this study was caught on longlines. Our samples were taken randomly, without information on gear used, and we were thus not able to test for gear influence.

CONCLUSIONS

Due to sampling failure from June and onwards we were not able to give a yearly maturation cycle for the oocytes of the Greenland halibut in Disko Bay.

There was no clear evidence to an overall increase in gonad weight in during the four months investigated. Gonad index indicated that most ovaries were in an immature or early maturing stage. However, when looking at the oocyte development month by month a weak trend to an increase in the oocyte size was evident with respect to the diameter range of the leading cohort

If spawning is happing in Disko Bay we believe that the most likely period for any extensive spawning is the same as in the Barents Sea namely November-January. Based on the available data we were not able to quantify the proportion of Greenland halibut in Disko Bay participating in such a spawning event..

We believe that further studies are needed in order to clarify the spawning and sexual maturation of the Greenland halibut in the inshore stock components in West Greenland. A suited method would be a gillnet survey (using mesh-size 200 – 240 mm) conducted monthly in the period August to January. Spawning frequency, oocyte development, atresia, and the actual number of eggs produced are important in that respect. Identifications of separate spawning entities contributes to better understanding of general reproductive capacity, and gives further indications on where nursery grounds for the juveniles may be located. Due to the wide distribution of Greenland halibut in the Atlantic it is important to link collected information to abiotic factors, to be able to explain possible differences in reproductive strategy within the species.

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