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Histological Assessment of Sexual Maturity of the Flemish Cap Cod in 1995

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Abstract

In this paper the proportion of females with postovulatory follicles in their ovaries in July 1995 is used to determine the length- and age-at-maturity of cod (*Gadus morhua*) in the year 1995. The proportion of females in the cortical alveoli stage is then used to estimate the same parameters for 1996. This histological method allows to accurately detect the proportion of both adult and primiparous females in the population at the very beginning of the gonad development, as it happens in the summer, and few months after spawning, when it is very difficult to make a visual diagnosis of the sexual maturity stage. Length and age at 50% maturity obtained from the curves plotting the proportion of cortical alveoli were 39.07 cm and 3.1 years, respectively. The same parameters obtained from postovulatory follicle frequencies were 42.65 cm and 3.9 years, respectively. During the period from 1990 to 1995, a decrease in both length- and age-at-maturity was observed for cod on Flemish Cap. It seems likely that in those recent years the age-at-maturity of female cod was fixed around age 3. The possibility that these features could be a density-dependent response is discussed.

Key words: age-at-maturity, cod, cortical alveoli, Flemish Cap, length-at-maturity, postovulatory follicles

Introduction

Since 1990 an histological method has been used to determine the length and age of maturity of the Flemish Cap female cod (*Gadus morhua*) in summer, when the EU–research survey is carried out. It consists of using the cod ovaries that are in the cortical alveoli stage as an index of the onset of the oocyte development for the next breeding season, whereas the presence of postovulatory follicles in the ovaries show the females that already spawned (Zamarro *et al.*, MS 1993). The cortical alveoli are the first structures to appear during the phase of oocyte growth (Wallace and Selman, 1981), and thus are the first sign of the start of the female ripening before the beginning of the vitellogenesis. The postovulatory follicles are the structures left in the ovary after oocyte ovulation, once its maturation is finished. It is indicative of a previous spawning, but the use of this structure to determine the proportion of females that have spawned requires the knowledge of its duration in the ovary.

The spawning season of the Flemish Cap cod is known to be short and the earliest in relation to other areas of the Northwest Atlantic (Myers *et al.*, 1993), around March. Thus, this method of analysis of ovary structures allows to accurately detect the proportion of both adult and primiparous females at the very beginning of the gonad development, as it happens in summer, and into a few months after spawning, when it is very difficult to make a visual diagnosis of the sexual maturity stage (Morrison, 1990). In this paper we use the proportion of females with postovulatory follicles in 1995 (July) to determine the length- and age-at-maturity in this year, and the proportion of females in the cortical alveoli stage to estimate the same parameter for the forthcoming year of 1996.

Materials and Methods

A total of 201 cod ovaries were sampled during the summer survey in Flemish Cap (Vazquez, MS 1996). Total length and weight were recorded for each individual and otoliths removed for age determination. Gonads were immediately fixed in 4% buffered formalin (Hunter, 1985). Pieces of gonads 0.5 cm in thickness, were embedded in paraffin based on conventional histological processing, and 5μ sections stained with Harris' hematoxyline and eosine-floxine.

Different stages were identified in the sections according to the classification of West (1990) and Morrison (1990). Immature females were identified on the basis that 100% of the oocytes were in the circumnuclear ring stage. Conversely the mature females had oocytes in cortical alveoli (CA), vitellogenesis, postovulatory follicles (POF) or degenerating (atresic) oocytes. Primiparous females were identified on the basis that they have no postspawning structures such as POF or atresic oocytes. The recent postspawning stage was identified by the presence of postovulatory follicles and atresic oocytes, without any oocyte in the cortical alveoli or showing any further stage of development.

The proportion of mature female by size and age was adjusted to a logistic equation as described by Ashton (1972):

$$\hat{P} = \frac{e^{\alpha + \beta L}}{1 + e^{\alpha + \beta L}} \quad (1)$$

and the logit transformation:

$$\ln \frac{\hat{P}}{1 - \hat{P}} = \alpha + \beta L \quad (2)$$

where

$\hat{P}$ = predicted mature proportion,

α and β = the coefficients of the logistic equation, and

L = the length (or age).

The BMDP LR program (Dixon *et al.*, 1990) was used to calculate the predicted values and the coefficients. Thus, the age and age-at-maturity could be estimated as the minus ratio of coefficients $(-\alpha/\beta)$ by substituting $\hat{P} = 0.5$ in equation 2.

Two maturity curves were generated: one using CA as the index of the onset of ripening for 1996, and another one using the presence of POF as a guide for spawned females in summer 1995.

Results

Tables 1 and 2 show the number of mature and immature females sampled by length and age, respectively. Of the females sampled, 57% were either primiparous or multiparous adult females, while 43% were immature. The frequencies of females with cortical alveoli, postovulatory follicles, vitellogenic oocytes and in postspawning stage by length intervals and age are presented in Tables 3 and 4, respectively. All the ovaries of adult females sampled had oocytes in the CA stage. There were no females in recent postspawning stage, since oocytes in CA were already present in all the ovaries with POF or atresic oocytes in July 1995.

A total of 94 of the sampled females had spawned in 1995, as indicated by the presence of POF and atresic oocytes in the ovary. The proportion of females with this kind of postspawning structures increased with length and age, and became 100% in females larger than 57 cm and age 5+ years. As occurred in 1994, the number of females with oocytes in the vitellogenic stage was very low in the summer (only 3 individuals in the 1995 survey).

The maturity curves obtained by length and by age are shown in Fig. 1 and 2, respectively. Each include two curves, one corresponding to the spawners in 1995, using the frequencies of POF to identify mature females and the other corresponding to the next year spawners, identified by the presence of CA. Length and age at 50% maturity obtained from the CA curves were 39.07 cm and 3.2 years, respectively (Table 5). The same parameter obtained from POF frequencies were 42.65 cm and 3.9 year, respectively. Like in 1994, there was a difference of less than one year between the two estimates of the age-at-maturity, which is an unusual situation compared with the results of previous years (Table 5). During the period from 1990 to 1995, a decrease in both the age- and length-at-maturity was observed.

Discussion

The age-at-maturity in 1995 obtained using POF was 3.9 years, the same as in 1994 (Saborido-Rey and Junquera, MS 1995). The same parameter obtained from CA, that is the age-at-maturity for the next year spawners, showed a slight reduction from 3.4 years in 1994 to 3.2 years in 1995. Conversely, the decreasing trend in the length-at-maturity already noted in 1994, continued in 1995 and it was very pronounced, when compared with 1994 when a reduction of 9 and 8 cm were observed in the estimates based on POF and CA, respectively. The fact of this great reduction in the length-at-maturity while the age-at-maturity remained almost invariant was not attributable to a change in the ageing criteria. It is suggested that this could indicate that the growth parameters of the stock are changing as well as the reproductive ones.

It is remarkable that the maturity curve in 1995 based on POF has a knife-edge shape. Almost all the females became mature between the ages 3 (0% spawned in July 1995) and 4 (80% spawned). At age 5, 100% of the females were mature. The information obtained in the summer 1995 gives further support to the fact already pointed out in 1994 (Saborido-Rey and Junquera, MS 1995), that it is likely that in recent years the age-at-maturity of female cod has been decreasing to around age 3. This age apparently determines a biological limit for the onset of the maturation process in this species, as the curve shows sudden occurrences of mature females without the occurrence of any

TABLE 1. Number of adult and immature females by length (cm) sampled on Flemish Cap in summer 1995.

Length	Immature	Mature	Total
<27	40	0	4
27–30	15	0	15
31–33	35	0	35
34–36	13	0	13
37–39	12	3	15
40–42	7	8	15
43–45	0	17	17
46–48	0	15	15
49–51	0	27	27
52–54	0	23	23
55–57	0	9	9
58–60	0	6	6
61–63	0	3	3
64–66	0	1	1
70–72	0	1	1
>73	0	2	2
Total	86	115	201

TABLE 2. Number of adult and immature females cod by age sampled on Flemish Cap in summer 1995.

Age	Immature	Mature	Total
2	2	0	2
3	63	0	63
4	19	4	23
5	2	93	95
6	0	15	15
7	0	1	1
9	0	1	1
Total	86	115	201

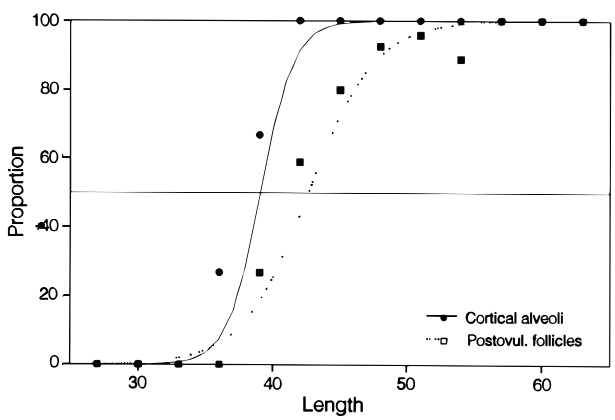

Fig. 1. Proportion of mature females by length (cm) obtained using the frequency of cortical alveoli and of postovulatory follicles in July 1995.

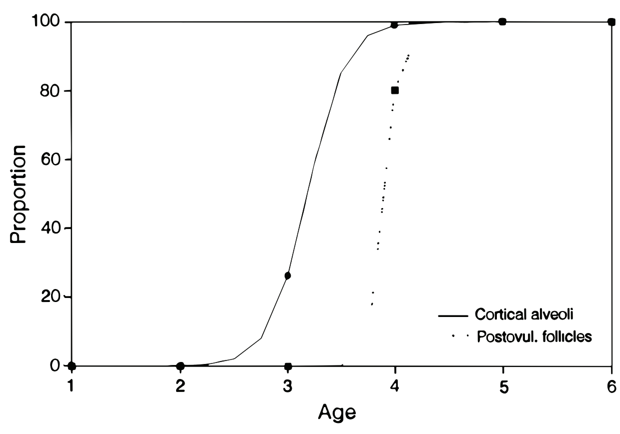

Fig. 2. Proportion of mature females by age obtained using the frequency of cortical alveoli and of postovulatory follicles in July 1995.

TABLE 3. Number of females with ovaries in the cortical alveoli stage (CA), postovulatory follicles (POF), vitellogenic oocytes (OV) and postspawning (PS) stage by length (cm) sampled in summer 1995.

Length	AC	POF	OV	PS	TOTAL
<27	0	0	0	0	4
27–30	0	0	0	0	15
31–33	0	0	0	0	35
34–36	0	0	0	0	13
37–39	3	0	0	0	15
40–42	8	4	0	0	15
43–45	17	10	1	0	17
46–48	15	12	0	0	15
49–51	27	25	0	0	27
52–54	23	22	1	0	23
55–57	9	8	1	0	9
58–60	6	6	0	0	6
61–63	3	3	0	0	3
64–66	1	1	0	0	1
70–72	1	1	0	0	1
>72	2	2	9	9	2
TOTAL	115	94	3	0	201

TABLE 4. Number of females with ovaries in the cortical alveoli stage (CA), postovulatory follicles (POF), vitellogenic oocytes (OV) and postspawning (PS) stage by age sampled in summer 1995.

Age	AC	POF	OV	PS	TOTAL
1	0	0	0	0	2
2	0	0	0	0	63
3	4	0	0	0	23
4	93	76	3	0	95
5	15	15	0	0	15
6	1	1	0	0	1
7	1	1	0	0	1
9	1	1	0	0	1
TOTAL	115	94	3	0	1

TABLE 5. Age and length at 50% maturity of female cod obtained from the maturity curves based on postovulatory follicles (POF) and cortical alveoli (CA) frequencies, from July 1990 to July 1995. (From Zamarro *et al.*, MS 1993; González and Larrañeta, MS 1994 and Saborido-Rey and Junquera, MS 1995.)

	Age		Length (cm)	
Year	POF	AC	POF	AC
1990	5.6	3.8	61	?
1992	–	–	–	–
1993	5	4	64	50
1994	3.9	3.4	51.2	47.8
1995	3.9	3.2	42.6	39.1

mature females at earlier ages. Similarly the knife-edge shows that from the 4+ age group on, virtually all the females are already mature.

From the maturity curves obtained using POF it can be concluded that in the period 1990 to 1995, the age-at-maturity decreased from about age 5 to about age 4 (3.9 years). A similar reduction was obtained using CA. Although the proportion of ovaries with CA was a useful tool to predict a maturity curve one year in advance, as it was observed in 1994 (Saborido-Rey and Junquera, MS 1995), the use of this kind of structure has the problem of the interannual variations in the date of mass spawning of the stock, that can lead to incongruent results from year to year (less than one year between both estimates, with POF and with CA).

The presence of CA in the cod ovaries is seasonal (Zamarro *et al.*, MS 1993). They appear soon after the spawning (probably in less than two months), and so its frequency would be underestimated in the summer samples whenever the spawning shifts from March toward the summer. Taking into account that the small cod are supposed to spawn later (Morrison, 1990), an overestimation of both the age- and length-at-maturity could be systematically expected when the CA are used to generate maturity curves in the Flemish Cap area, especially considering that the stock is at the moment mainly composed of small fish (Vazquez, MS 1995; MS 1996). In 1995, the absence of females in recent postspawning conditions indicate that the spawning occurred not as late as in 1994, when a high proportion of females were in postspawning stage in July (Saborido-Rey and Junquera, MS 1995). However, the spawning season still seems delayed compared with the period 1990–92, when a high proportion of the females were already in the vitellogenesis stage in July (Zamarro *et al.*, MS 1993, González and Larrañeta, MS 1994), while only negligible proportions were found both in 1994 and 1995 in the same month.

The possible effects of the decrease in the cod stock abundance, especially of the largest individuals, in Flemish Cap on the reduction of the age-at-maturity during the period analyzed, has already been pointed (Saborido-Rey and Junquera, MS 1995). The 1995 summer research survey in this area indicated that the cod biomass had noticeably reduced compared with 1994. The 1991 year-class, which was the most important one in those previous years, had virtually disappeared and the year-classes of 1993 and 1994 were very weak. Consequently, if the reduction in the age-at-maturity as observed in this study is a density-dependent response, this feature can be expected to be maintained into the next few years.

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